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Mosquito Management Manual

Department of Health WA 2013



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Cover photos:

Main: Briquette treatment of a wetland in the City of Bunbury (© Ryan Janes, Department of Health WA); Inset top: Aerial larviciding in the Peel Region (© Scott Severn, City of Mandurah); Inset middle: EVS CO2 trap at Roebuck Plains, Shire of Broome (© Amber Douglas, Department of Health WA); Inset bottom: Larval dipper (© Scott Dandridge, Shire of Harvey)

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1. A FEW WORDS ABOUT RECENT CONFUSION OVER NAMING OF AEDES / OCHLEROTATUS MOSQUITOES

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In order to reflect the relatedness of individual mosquito species to one another, scientists place them into groups with other species of similar appearance (morphology) and biological characters.

Individual species are grouped with other related species into a **subgenus** (plural = subgenera), which in turn are grouped into a **genus** (plural = genera).

Relationships between mosquitoes in the *Aedes* genus were reviewed in recent years (Reinert, 1999, 2000; Reinert *et al.* 2004), resulting in the reclassification of many of them into new genera.

One of the major changes of relevance to Australia was the regrouping of most mosquito species in the *Aedes* genus into several new genera. For example, the common saltmarsh mosquito *Aedes vigilax* was renamed as *Ochlerotatus vigilax*.

This significant change to mosquito naming was reluctantly accepted by mosquito professionals throughout the world and many recent texts and journal articles reflect the revised *Aedes* mosquito names.

However, the studies and the subsequent process that led to the adopting of the new names were reconsidered during 2005. The scientific community now agrees that the work that led to the change was not sufficiently detailed, nor were its impacts on mosquito naming given adequate consideration, to justify the revised naming.

Consequently, relevant international journals (e.g. Weaver, 2005; Editor-in-chief and Subject Editors of JME, 2005) and professional associations have determined that in the majority of cases, the names of mosquitoes affected by the name changes should now revert to their former names, e.g. *Aedes vigilax*.

At the 2006 meeting of the Mosquito Control Association of Australia it was agreed to adopt this policy. Therefore, this manual reflects the current scientific opinion that names of most mosquitoes affected by the above-mentioned changes should revert to *Aedes*, unless (or until) more comprehensive and convincing studies indicate otherwise.

REFERENCES:

Reinert, J.F. (1999) *Contributions of the American Entomological Institute (Gainsville)* 31: 1-83.

Reinert, J.F. (2000) Journal of the American Mosquito Control Association 16: 175-188.

Reinert, J.F., Harbach, R.E. and Kitching, I.J. (2004) *Zoological Journal of the Linnean Society* 142: 289-368.

The Editor-in-chief and subject editors of JME (2005) Journal of Medical Entomology 42,511.

Weaver S. (2005) American Journal of Tropical Medicine and Hygiene 73,481.

2. MOSQUITO BIOLOGY AND ECOLOGY

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As far as pest problems and the transmission of disease causing pathogens are concerned, it is only the adult female mosquito that appears important since the male does not suck blood nor bite in any way. However, to understand the complexities of disease and nuisance problems, and therefore the specific methods of control that are appropriate in different situations, various aspects of the natural history of mosquitoes should be understood.

Basically, the mosquito life cycle (Figure 1) begins with an adult female laying eggs. From the egg emerges an aquatic immature stage called the larva (plural = larvae), which develops through four moults, increasing in size until at the final moult it reaches a non-feeding stage the pupa (plural = pupae). Inside the pupal skin the adult is forming as either a male or female, and the terrestrial/aerial adult stage emerges from a split in the back of the pupa. The adults feed, mate, and the female develops eggs to complete the cycle and begin the next generation. Some species of mosquito may have only one generation per year, others will have several generations during a season of favourable climatic conditions, others will continue to breed throughout the year; much depends on climatological aspects of the local environment, particularly temperature and rainfall.

Before proceeding further, some simple knowledge of the classification and naming of mosquitoes is necessary. Within the 'Animal Kingdom', the insects as we know them are grouped together as the **Class Insecta**. The group of insects known to us as mosquitoes belong to the category of insects known as the **Order Diptera**, i.e. the two-winged 'flies'. Within the Diptera, all mosquitoes belong to a **Family** group known as **Culicidae**. Within this family, mosquitoes with a range of similar characters are grouped into **Subfamilies** e.g. the **Culicinae** and the **Anophelinae**. These groupings are further subdivided into closely related sub-groups called **Genera** (single = **genus**); important genera are *Anopheles* in the subfamily Anophelinae, and *Aedes* and *Culex* in the subfamily Culicinae, although other Culicinae genera found in the southeast Australian region include *Aedeomyia*, *Coquillettidia*, *Culiseta*, *Mansonia*, *Toxorhynchites*, *Tripteroides* and *Uranotaenia*.

Mosquitoes which are of the same **species** are assigned specific names, giving them a 'double-barrelled' title (e.g. *Anopheles annulipes, Aedes notoscriptus, Culex annulirostris*). By convention, the species name is always underlined or written in italics. This 'species concept' indicates that all specimens with that name are 'identical' with each other, to the extent currently defined for the species, and are different to individuals of other species and will not interbreed with individuals of other species.

Class:	Insecta				
Order:	Diptera				
Family:	Culicidae				
Subfamily:	Anophelinae	Culicinae			
Genus:	Anopheles	Aedes	Culex	Mansonia	Coquillettidia
Species:	annulipes	aegypti	sitiens	uniformis	linealis



Figure 1: The general life cycle of a mosquito

For convenience in most scientific writings, generic names are usually abbreviated to only two letters once they have been mentioned in full; thus *Aedeomyia* is written as *Ad*, *Aedes* is abbreviated as *Ae*, *Anopheles* as *An*, *Culex* as *Cx*, *Culiseta* as *Cs*, *Mansonia* as *Ma*, *Toxorhynchites* as *Tx*, *Tripteroides* as *Tp*, and *Uranotaenia* as *Ur*.

The specific names are usually taken from the Latin or Greek roots and relate to place of collection (e.g. *Culex australicus* is a member of the worldwide *Cx pipiens* group but is indigenous to Australia), physical characteristics [e.g. *Cx annulirostris* has a distinctive band (annuli) on its proboscis (rostris)], behavioural characteristics (e.g. *Cx molestus* can be a particularly annoying pest), or named to commemorate a person (e.g. *Cx fergusoni* was named after E.W. Ferguson, an eminent public health entomologist in NSW in the early decades of this century).

The characteristics by which specimens are grouped together as belonging to the one species are often minute and may require microscopic examination and expert knowledge, but many common species have distinctive markings (obvious sometimes even to the naked eye) which, with some experience, allow identification by non-specialists with little equipment other than a x10 hand lens or magnifying glass. The identification of mosquitoes will be presented elsewhere.

As the physical appearance of different species allows them to be grouped together in a genus, so these species have biological traits which are common to members of that genus. These traits can be exhibited from the initial phase of the mosquito life cycle, and there are four general 'models' within the aquatic life cycle that can be illustrated by the genera *Aedes*, *Anopheles*, *Culex* and *Mansonia*. Species of other genera are generally variants of one of these 'models'.

THE EGG

Some genera lay their eggs in a characteristic manner (Figure 2), and for many species the eggs themselves may have distinctive surface patterns and other characteristics.

All species of the genus *Anopheles* lay their eggs (oviposit) as single units on the surface of a collection of water. The eggs are cigar shaped, black and have side floats to maintain buoyancy until hatching. These eggs are completely dependent on the presence of free water and usually hatch in about 2 days after laying (oviposition). If stranded by the water drying up or by wave action they will perish. *Anopheles* species are generally associated with permanent or semi-permanent bodies of ground water but there are some which colonise temporary pools and container habitats.

Aedes species also lay their eggs as single units but deposit them, not on a free water surface, but on a moist substrate (e.g. rock surface, moist earth, inside wall of a tree-hole or container) above the receding water level or insert them under debris, and into crevices in soil and drying mud where they will be subsequently flooded. These eggs are able to withstand desiccation and can survive long periods, until the water level rises with rain, flood or tide to inundate them, at which time they begin to hatch, often only in batches rather than all at once. *Aedes* species are generally found associated with temporary bodies of water, or those that might be persistent but have fluctuating levels.

Culex (and *Coquillettidia*) species, deposit their eggs on a free water surface with the eggs in a compact cluster, glued upright together in the form of a raft, which floats on the water surface. The eggs cannot withstand desiccation, will hatch after 2 days or so if not stranded by water movement, and are usually associated with permanent or semi-permanent bodies of water.

A characteristic feature of *Mansonia* species is that they deposit their eggs in a submerged mass attached to aquatic plants in bodies of permanent and semi-permanent water. The eggs cannot withstand desiccation and hatch within a few days.

THE LARVA

The habits and habitats of mosquito larvae are rather diverse; but essentially the larval stage of the mosquito life cycle is an aquatic animal and must have an aquatic habitat in which to complete its development to the pupal stage. The larval habitat is selected by the female when she deposits her eggs; she is able to discern physical and chemical properties of different collections of water and choose between sites available for her selection.

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Larvae can be found in many different habitats and a generalised classification constructed by Dobrotworsky for mosquitoes in Victoria is applicable for southeastern Australia in general:

1. Fresh water

- A. Ground Water
 - a. Natural habitats
 - i. lakes and swamps vegetated margins and floating vegetation
 - ii. streams vegetated margins, isolated quiet reaches, backwaters, ox-bow lakes / billabongs
 - iii. temporary and semi-permanent ground pools rain filled depressions, animal wallows, hoof prints
 - iv. rock pools sides of or in stream bed, rain filled
 - v. flooded animal burrows and crayfish tunnels
 - b. Man-made habitats
 - i. irrigation ditches and run off, overflow and tail drains
 - ii. dams usually more important after stabilisation and when vegetated
 - iii. excavations borrow pits, roadworks, wells, mining operations
 - iv. wheel ruts
- B. Container Water
 - a. Natural habitats
 - i. tree-holes
 - ii. leaf axils
 - iii. fruits and husks
 - b. Man-made habitats
 - i. domestic functionals water tanks / barrels, roof guttering, animal drinking troughs, pot-plant saucers
 - ii. discarded / hoarded rubbish tins, tyres, plastic containers, derelict car bodies

2. Polluted water

- i. septic tanks
- ii. drains, sullage pits
- iii. ground water at garbage dumps

3. Brackish water

- i. estuarine marshes / swamps
- ii. tidal reaches of river margins
- iii. rain pools / irrigation run-off in inland areas with saline soil

4. Salt water

i. pools on coastal rock platforms

Within this broad classification of larval habitats, many subtle variations occur, and many other examples could be added. All breeding sites ultimately depend on the local conditions, and such a classification as that above cannot do complete justice to the complexities met with in nature. However, most instances of mosquito breeding would be covered in the above categories.

Species usually have a preference for a particular type of breeding site, but features visible to us may not always be the characteristics selected by the gravid female looking for a place to lay her eggs. In many instances such factors as shade or sunlight; presence or absence of floating, emergent or marginal vegetation; pollution; salinity; temperature; and others (such as the texture of the substrate for *Aedes* species) may influence the female, and we can often

recognise a number of these factors as being characteristic of the typical breeding site of a particular species.

The larvae (Figure 3) hatch from the egg and grow through four instars or stages, in between which they moult their rigid outer skin in order to increase in size. They feed on microscopic organisms in the water, or on decaying vegetation and other bottom detritus, either by filtering water through their mouthbrushes of fine hairs or by grazing with specially adapted mouthbrushes; some larvae are predatory and their mouthbrushes are strongly modified to grasp prey. Some species feed habitually at the surface (e.g. Anopheles), some in the middle range below the surface (e.g. Culex), and others typically feed on the bottom of the habitat (e.g. Aedes). Larvae breathe air, from openings (spiracles) at the 'tail' end of the body, generally through a tube (siphon) which can penetrate the surface of the water. Although they will submerge when disturbed and may remain motionless on the bottom for some time, larvae need to return to the surface for air to prevent suffocation. The general exception to this behaviour occurs with larvae of Mansonia and Coquillettidia species, which attach to plant tissue below the surface of the water after hatching, using a specially adapted piercing siphon, and obtain their oxygen directly from the plant tissue; they do not voluntarily detach from the plant during their period of development and feed by filtering food particles from the surrounding water with their mouthbrushes.

The time taken for development through the larval stages is dependent on a number of environmental factors, the most important of which is temperature, although availability of food and the extent of larval crowding within the habitat are also important. During favourable summer conditions *Anopheles* species may complete larval development in 7-10 days, *Aedes* species may complete development in as little as 4-5 days, *Culex* species may require at least 7-10 days, but *Mansonia* species often spend more than 3 weeks in the larval stage. Generally speaking, water temperatures between 20°C and 25°C are favourable for most species in southeastern Australia. Low temperatures delay development, may cause cessation of growth and induce a winter 'larval hibernation' in some species. Very low temperatures can be lethal for some species although in temperate areas of Australia many species are able to survive winter periods in the larval state. Exposure to water temperatures above 40°C may be lethal for many species, although it is unusual for larvae to be exposed to such temperatures for prolonged periods in field situations in Australia.

Identification of larvae is most easily accomplished with mature larvae, i.e. the fourth instar, and microscopic examination is usually required. However there are some genus characteristics that enable partial identification in the field. Larvae of *Anopheles* species have no siphon (breathing tube) and when feeding or resting they lie flat at the surface of the habitat. *Culex* and *Aedes* species have siphons and hang suspended from the surface when obtaining oxygen; *Culex* typically have 'longer' siphons than *Aedes*, and while *Aedes* species generally may be described as grazing 'bottom feeders', *Culex* species generally feed from the surface to the bottom. The larvae of *Mansonia* and *Coquillettidia*, although not commonly collected because of their submerged attachment to aquatic vegetation, can be identified as belonging to one of these two genera by their modified siphon (see Figure 3).

THE PUPA

After the fourth larval stage completes its development it moults, and a non-feeding but mobile stage call the pupa follows (Figure 4). Within the body casing of the pupae the immature tissues are breaking down and adult tissues are forming; at this stage the sex of the individual can be discerned for the first time. The pupa breathes through a tube-like organ (trumpet) situated at the 'head' end of the comma-shaped body. The duration of the pupal stage is dependent on temperature, but is generally of the order of 2-3 days for *Anopheles, Aedes* and *Culex* species, although typically longer (6-9 days) for *Mansonia* species, which like *Coquillettidia* species attach to submerged plant tissue to obtain air. If the pupa is stranded or



Figure 3: The larvae of the genera Anopheles, Aedes, Culex, Mansonia and Coquillettidia.

otherwise separated from its aquatic habitat during this period it may be able to survive to the point when the adult may successfully emerge.

It is difficult to distinguish between pupae of *Anopheles*, *Aedes* and *Culex* species although *Anopheles* have a much expanded opening to the breathing trumpet, and *Mansonia* and *Coquillettidia* have the trumpet valves modified for piercing plants (see Figure 4).

As the adult develops within the pupal casing the pupa becomes progressively darker, rises to the surface, stretches out parallel with the water surface and becomes immobile. A split in the dorsal surface appears and the adult struggles out from the pupal shell, stretching its wings, legs and mouthparts which have been folded/coiled within the pupal casing.

THE ADULT

The adult mosquito (Figure 5) rests on the water surface for a short time after emergence from the pupal casing, to allow its wings and body to dry, before flying off to pursue the next phase of its life cycle.

In a single generation, the males of the species usually develop marginally more quickly than females, and males are usually first to emerge from the larval habitat. However, this is not always noticeable in the field where generations may overlap. Male mosquitoes, upon emergence, do not normally travel far from the breeding site. They feed on plant juices, flower and fruit nectars, and mate with the females. Males generally have a relatively short life span, and as they do not bite humans nor feed on blood from any source, they are of little importance as far as disease transmission or pest nuisance is concerned. However, they are of some significance for surveillance operations in that when male mosquitoes are detected, it usually indicates that a breeding site is relatively close nearby and breeding may be current.

Following emergence, the adult female will generally seek out a carbohydrate meal of plant juices to replenish expended energy reserves. It will then mate with a male, usually near the breeding site and often at dusk. Female mosquitoes mate only once, the sperm packet introduced by a male during the mating act serving the female to fertilise all batches of eggs she subsequently produces. For mating, male mosquitoes may form swarms, often at dusk and associated with markers such as a bush, tree stump, or bare patch of ground. The males recognise females flying close to the swarm by their wing beat frequency, team up with a female and copulate.

For development of eggs, female mosquitoes require protein and this may be provided either from nutritional reserves carried from the larval stage or from a meal of a high protein source such as blood. A few species can develop a first batch of eggs without a blood meal, but will then attack a blood source for the nutrients required for second and subsequent batches.

Females can survive on plant juices, but most of the species important as pests or disease vectors seek blood soon after mating, about 2-3 days of age, and then embark upon a life of repetitive cycles – feeding, resting, developing and laying eggs; feeding, resting, developing and laying eggs; and so on.

The preferred source of the blood meal can vary widely between mosquito species and with different situations. The source of the blood meal is epidemiologically important, as some pathogens/parasites are associated with particular vertebrate hosts. Some mosquitoes feed on snakes, frogs and even fish, and these cold blooded feeders are of little concern for humans. Generally, the potentially important species for transmission of zoonotic diseases (e.g. Murray Valley encephalitis) are those that feed on bird and/or mammal hosts and also on humans – the opportunist feeders; and for primarily human diseases (e.g. malaria, dengue) the most potentially important are those that feed preferentially on humans.



Figure 4: The pupae of Anopheles, Aedes, Culex, Mansonia and Coquillettidia.

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Figure 5: The adult mosquito, (a) external appearance and structure, (b) internal anatomy

A mosquito species can be more or less particular about its source of blood, but with some species this may vary with circumstances and with different populations of that species – it could depend on the range of blood hosts available for instance. A mosquito which generally prefers stock, even in the presence of humans, may readily feed on humans in the absence of the preferred host. A species normally feeding on birds may well seek mammalian blood when mosquito populations are high and extend beyond their normal habitat, or when humans enter the mosquito habitat while hunting/fishing or walking. In this scenario it is possible for humans to be infected with pathogens considered to be normally associated with birds/mammals in forest/swamp habitats and rarely infecting humans.

In general terms, mosquitoes are attracted to a warm blooded host by a combination of factors. Carbon dioxide, a product of respiration is an important attractant, as are various body odours, and these factors seem to be the 'long range' attractants. At closer distances temperature can be a factor and visual perception may be important at very close range.

Other circumstances of feeding behaviour can vary between mosquitoes and be characteristic of particular species. Many species feed primarily during the twilight hours of sunset and at dawn; others have peak biting periods during the deep of night, and their behaviour may be influenced by moonlight; others may be described as daytime biters, although some will only bite in shaded conditions; other species may bite virtually any time a host is available. The twilight biters are termed 'crepuscular', the night biters 'nocturnal' and the day biters 'diurnal'. Apart from the influence of light conditions, it is known that temperature and humidity also play a role in determining host-seeking behaviour; adult mosquito activity may be almost non-existent in southern Australia when temperatures are below 8°C, while high humidity and even a little rain can increase activity at favourable temperatures.

Some species will readily enter a building/dwelling/shelter to obtain a bloodmeal, while others will only bite 'out-of-doors'. Some species will rest 'indoors' before or after a bloodmeal, while others even if they bite indoors will immediately fly out to find a resting place.

After taking a blood meal, the female searches for a secluded resting spot where the meal can be digested and the ovaries can develop eggs. In the field, mosquitoes will find suitable conditions for resting, for instance, amongst dense vegetation, inside a hollow log or tree, animal burrow, rock cave. The time needed for complete egg development is dependent on mosquito species and the prevailing temperature, but the minimum is of the order of 2 days (*Anopheles* species and some *Aedes* species). *Culex* species may take at least 4 days to mature the eggs, and at lower temperatures in spring and autumn the period may involve 10 days or more. When the eggs are mature within the ovaries the female will fly from the resting site in search of a suitable larval habitat where she can deposit her egg batch – often in late afternoon or evening. She will then fly off in search of a bloodmeal in order to repeat the cycle; subsequent bloodmeals can be taken the night of oviposition if a host is nearby, otherwise a day or more may elapse before the next feed. Thus some mosquitoes may feed every 2-3 days, others no more than every 5-6 days, repeatedly throughout their life.

For some species, a single bloodmeal is not sufficient for the production of the first egg batch, and although many species will produce eggs from a single meal, laboratory studies have shown that some require a second meal. In general terms the size of the blood meal (and also its source) determines the number of eggs that can be oviposited, and although as few as 20-30 eggs may be laid by some species under some circumstances, egg batches of other species may contain up to several hundred eggs at the other extreme.

As mentioned above, the oviposition site (and thus the larval habitat) may be characteristic for a particular species. However, although we know that mosquitoes have sense organs which allow them to choose between physical and chemical features of an aquatic site, the determining features important to the mosquito may not always be apparent to us. Even so, there is a definite species association with some 'types' of habitat, and therefore with other species in a 'type' of habitat. Examples of apparently important factors (either directly or indirectly influencing choice of habitat) are stream or ground pool or container; water fresh or brackish; habitat shaded or sunlit; habitat vegetated or non-vegetated; water clear or muddy; water clean or polluted.

Beyond the typical association of *Aedes*, *Anopheles*, *Culex* and *Mansonia* described earlier in this Section, some further general comments may be made. *Tripteroides* species normally oviposit in tree holes, but will breed in artificial containers on the ground. In contrast, some *Culiseta* species habitually breed in the subterranean tunnels of land crayfish. Water temperature, salinity and physical nature of the container surface are also important with some *Aedes* species.

The longevity of adult mosquitoes is a complex but very important issue. They are subject to the attack of many predators including birds, bats, frogs and other insects; parasites and

pathogens such as worms, protozoa, viruses, bacteria and fungi; they are also at the mercy of the rigors of the environment with the vagaries of wind, rain, humidity and temperature all taking their toll on the small and fragile insect. In the laboratory some mosquitoes can be kept alive for many months under certain circumstances, while others will not survive more than a few days even under apparently favourable conditions. In the natural environment an age over one month would be considered very old, although some species are innately more long-lived than others.

Age of a female mosquito, particularly one which is of threat as a vector of disease, is a most important factor in the epidemiology of disease transmission. For a female mosquito to become a disease vector, she must first take a bloodmeal from a host infected with the parasite or pathogen and must take up 'infecting' stages with that bloodmeal. If she is susceptible and becomes infected with the pathogen, she must survive for the period of incubation required for the pathogen before she becomes infective herself and can transmit the pathogen to another host at a subsequent bloodmeal.

This period of incubation within the mosquito varies with pathogen or parasite and with temperature, but for the sake of this discussion may be proposed as approximately 10 days. If the incubation period required is of the order of 10 days, a mosquito must survive for that 10 day period after she takes her 'infecting' bloodmeal before she can possibly transmit the disease organism. During that period she may bite a number of subsequent hosts without transmitting the pathogen, but all the time exposing herself to death via the different agencies mentioned above. Mosquito age can be determined by dissections of the ovaries indicating the number of egg-laying cycles completed and, with knowledge of the period required for egg development, the calendar age can then be estimated; results from studies of some common pest mosquito species in southeastern Australia, indicates that less than 10% may survive to 10 days of age.

Dispersal is an important factor in mosquito ecology. Many mosquito species typically move only relatively small distances (sometimes no more than 50-100 metres) from their larval habitats, providing appropriate blood sources are available in the vicinity, while some move back and fourth within a range of 1-5 kilometres; other species have a definite 'migratory' behaviour component that may be facultative with large population densities or obligate as part of their normal biology. There are a few species in the latter group, *Aedes vigilax*, the coastal salt marsh species is the prime example in Australia and it is known to be able to able to disperse at least 50 km, often downwind, associated with periods of peak adult activity. With a lesser range, *Culex annulirostris* in the Murray/Murrumbidgee area has been shown to readily disperse at least 5 km, but much more than 10 km is thought to be unlikely.

In southern Australia, some mosquitoes maintain reproductive activity during all seasons of the year, albeit at low levels in winter. The majority, however, tends to spend this period of adverse conditions in a dormant state. Mosquitoes can survive periods of adverse conditions (e.g. winter, dry seasons) by various mechanisms and, because of the climatic range in southern Australia, various species may display different activity and behaviour in different areas. Some species may survive dry periods by diapause in the egg stage (diapause may be broken by eggs being flooded) e.g. *Aedes* species such as *Ae vittiger*, or larval development can be arrested during cold periods (development may resume with rise in temperature) e.g. some *Aedes* species such as *Ae alboannulatus*, and some *Culex* species such as *Cx australicus* in some areas. Other species survive winter periods in temperate climates by hibernation in the adult stage; with some species hibernation occurs without bloodfeeding, in others feeding may occur during hibernation without egg development. Hibernation is usually broken by a rise in temperature and an increase in day length such as occurs in spring.

Most mosquitoes in southeastern Australia display patterns of seasonal abundance, even though fluctuations associated with meteorological conditions may intervene from time to time. Many species are highly seasonal, with peak adult abundance in mid-summer and complete

absence in winter; others are active throughout the year. In temperate regions it is not unusual for a progression of species to occur throughout the year with different species being predominant at different times and under different conditions. The critical factors determining population abundance are those primarily associated with larval production, such as extensive aquatic habitat and optimal conditions of temperature and availability of food, but also important are terrestrial environment conditions such as temperature and humidity favouring adult survival.

It is difficult to generalise about aspects of seasonality in southern Australia because of the diversity of biophysical zones within the region, the diversity of species within the various zones and the influence of local climatic and general meteorological factors. However, to illustrate, in coastal areas of southern NSW, *Ae alboannulatus* or *Ae flavifrons* may be the most prevalent species during winter, *Ae camptorhynchus* and *An annulipes* may dominate during spring, *Ae vigilax* is usually the predominant summer species but *Ae camptorhynchus*, *Cx annulirostris*, *Cq linealis* may also be abundant, and *Cx australicus* and *Ae procax* may be the most common species in late summer/autumn. In inland riverine areas of southern NSW, *Ae sagax*, *Ae theobaldi* and *An annulipes* and *Cx australicus* may variously dominate spring collections, *Cx annulirostris* is generally the major summer species but *An annulipes* and *Cx australicus* can be common and some *Aedes* e.g. *Ae theobaldi* can occasionally be prevalent. However, such scenarios can vary markedly between years in one area dependent on local conditions.

It is well to keep in mind that our perception of which species are prevalent at whatever time of the year depends on the sampling technique. Most sampling techniques have a particular bias, and the best investigations of mosquito faunal activity are those that employ a range of techniques to sample the fauna.

The time taken for development from egg to adult varies greatly with environmental variables, particularly temperature, but an estimate of 2-3 weeks under optimum conditions should see most common species reach the adult stage. Species which normally inhabit temporary situations such as rock pools, tidal ground pools, tree holes, rain-filled depressions, and containers usually have a shorter development period than those species that use more permanent waters, and may complete development in a week or less in summer periods.

Identification of mosquitoes will be presented elsewhere but it is relevant to mention a few principles here. It is relatively simple to distinguish male mosquitoes from female mosquitoes. The male sexual organs are at the end of the abdomen and consist of a pair of pincer-like appendages that are more or less noticeable depending on the species. However, the easiest way to distinguish male mosquitoes is to examine the antennae projecting forward from either side of the head. In males of all types of mosquitoes, the antennae of the male are quite distinctive, being very bushy and much more hairy than the antennae of female mosquitoes (see Figure 6).

With regard to females, species of the subfamily Anophelinae, e.g. the genus *Anopheles*, have palps (sensory organs lying either side of the proboscis) nearly equal in length to the proboscis, whereas the palps of mosquitoes of the subfamily Culicinae, e.g. the genera *Aedes*, *Culex* and *Mansonia*, are typically much less than half the length of the proboscis. It should be noted that the palps of males in both the Anophelinae and the Culicinae are long, as in female Anophelinae, but both turn upwards at their end and the ends of Anopheline species are expanded club-like (see Figure 6).

The general life cycle stages of *Anopheles*, *Aedes* and *Culex* species are summarised in Figure 7 which shows the resting habit of *Anopheles* with its body at an acute angle to the resting surface while the *Aedes* and *Culex* tend to stand with their bodies parallel to the resting surface.



Figure 6: Anopheline and Culicine mosquitoes; male and female characters.

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Figure 7: Characteristics of *Anopheles, Aedes* and *Culex* species. (Illustration by K.S. Littig and C.J. Stojanovich)

3. PUBLIC HEALTH AND MOSQUITOES

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Mosquitoes are the most important group of bloodsucking insects that cause nuisance and transmit diseases to humans and other warm-blooded animals. Australia has a relatively numerous and diverse mosquito fauna associated with its extraordinary range of environments from the tropical north to the temperate south, from the lush rainforests through the wet and dry sclerophyll bushland to the scrub plains and deserts, and from the mountains through extensive slopelands to the plains.

The nuisance and annoyance caused by mosquitoes is not easy to translate into economic values. Large areas of coastal marshes, inland floodplains, and irrigated farmland periodically produce hordes of pestiferous mosquitoes. Potential recreation sites are rendered unsuitable, agricultural development is retarded and mosquitoes can cause significant and economically damaging reductions in beef and milk production.

However, it is as vectors of disease that mosquitoes are often of most concern; an insect that transmits a disease-causing organism from one vertebrate host to another is called a '**disease vector**'. The vector mosquito and the parasites and pathogens that it can transmit, are now recognised to have played an important role in the development and dispersal of the human race, being responsible to a greater or lesser extent for events that have changed the course of history.

Mosquitoes act as transmitters or **vectors** of pathogens or parasites by one of two methods – mechanical or biological. Mechanical transmission pertains when the pathogen has no biological association with the mosquito, and the vector simply picks up the pathogen (almost as a body contaminant) from one source and then deposits it in another location; biological transmission refers to the situation where the pathogen or parasite undergoes an obligatory period of development and/or multiplication within the vector, before being passed onto another host.

Although mechanical transmission of pathogenic organisms occurs via mosquitoes with some animal diseases, biological transmission is predominant for human parasites. A current worldwide emphasis on vector-borne disease research relates to a requirement for more intensive studies of the vector species, since understanding the life cycle and ecology of a vector will leads to a greater understanding of disease epidemiology, and is more likely to ensure success in control efforts.

The study of the role of mosquitoes as vectors began more than a century ago. In 1878 Patrick Manson announced the filarial worm now known as *Wuchereria bancrofti* underwent development in mosquitoes. This was the first real evidence that an organism causing disease in humans had an obligatory development phase in an insect – a mosquito. In the next 50 years in particular, a variety of insects, ticks, mites and other arthropods were incriminated as vectors of pathogenic organisms in both medical and veterinary science.

It is now realised that mosquitoes are vectors for three types of human pathogenic organisms: (i) the filarial 'worms' (nematodes which cause lymphatic filariasis), (ii) the plasmodia (protozoans which are the casual organisms for the malarias), and (iii) arboviruses (viruses causing such diseases as yellow fever, dengue fever and various encephalitides and arthritides; the term 'arbovirus' is derived from the 'arthropod-borne-virus': mosquitoes are insects which belong to the category of animals known as the Phylum Arthropoda, and arboviruses are viruses that are transmitted between vertebrate hosts via an arthropod acting a as an intermediate host).

Although vaccines, chemoprophylaxis, chemotherapy, insecticides and other vector control measures are becoming more sophisticated, none of the major mosquito-borne diseases of the world may be said to be under complete control. Malaria still claims millions of lives annually throughout the tropics; yellow fever is still firmly entrenched in jungle areas of Africa and South America, occasionally emerging to initiate urban outbreaks; dengue fever continues to cause suffering and death in Southeast Asia, the Caribbean and Pacific regions; viruses causing fatal encephalitis and various debilitating symptoms are still transmitted in tropical and temperate parts of the African, American, Asian and Pacific regions; and filarial worms carried by mosquitoes are responsible for urban and rural ill-health in many tropical and sub-tropical regions.

Some of these disease spread by mosquitoes are associated with animal reservoirs and are called **zoonoses** (e.g. yellow fever, the virus encephalitides, Brugian filariasis), while others involve only human reservoirs (e.g. malaria, dengue, Bancroftian filariasis). However, in all cases the crucial factor in the transmission pattern to man – the epidemiology of the disease – is the amount and type of contact between the mosquito vector and the human host. The incidence of the disease will be dependent on what may be called '**human-vector contact**'; this is a function of the habitat and behaviour of the mosquito vector, and the habitat and behaviour of the human host.

The more that the potentially infective mosquito intrudes into the human environment, or the more that humans intrude into natural environments where mosquitoes harbour pathogenic organisms, the greater the risk of transmission to an individual and a subsequent risk of initiating and urban outbreak or epidemic. Therefore, the greater knowledge and appreciation that we can have of the natural history and ecology of the mosquito and the pathogenic organisms, the better prepared we can become to avoid, combat or control a potential or actual disease situation.

Australia has had a lengthy association with mosquito research. The earliest studies in this country were mostly confined to taxonomy – the description and naming of species – a continuing process. However, specific mosquito-borne public health problems such as malaria, filariasis, dengue fever, epidemic polyarthritis, arboviral encephalitis, and human quarantine concerns; as well as animal diseases such as myxomatosis, ephemeral fever, fowl pox, and dog heartworm have provided an impetus for field and laboratory investigations of mosquito ecology and transmission dynamics. At present the mosquito-borne diseases of concern in Australia still include most of those which were historically important.

- **Malaria** is still important even though the last indigenous case on the mainland occurred in 1962 and Australia has been awarded certification of malaria eradication by the World Health Organisation. Transmission of malaria continues in nearby southeast Asia and parts of the southwestern Pacific region, and infections acquired overseas continue to be imported into Australia and local infection has been recorded in recent years on Torres Strait islands and in northern Queensland. The northern regions of this country are both vulnerable and receptive to malaria, and vigilance against re-introduction leading to re-establishment needs to be maintained.
- **Filariasis** in humans was endemic in Queensland and primarily concerned only that State, although it extended into the Northern Rivers area of New South Wales. The disease virtually disappeared by 1940. However, the dog heartworm is a mosquito-borne filaria which has increased its distribution and incidence in parts of Australia.

- **Dengue fever** was first reported in Australia in 1885 from northern Queensland, and from that time occasional epidemics occurred in northern Australia, and some extended into NSW. Until 1981, dengue had not occurred on mainland Australia since the outbreak of 1955 in Townsville, Queensland, but then many cases were recorded in northern Queensland when it was reintroduced in 1981-82. Since then, outbreaks have occurred in Queensland almost annually since the early 1990s, and there have been associated deaths. Dengue fever has re-emerged as a major concern for Australia because of continuing introductions from Asia and Pacific regions, but transmission occurs only in Queensland, particularly the northern coastal regions.
- **Epidemic polyarthritis** has been suggested to have been observed form the 1920's. The causative agents, Ross River virus and Barmah Forest virus, may be transmitted by different mosquito species in different localities and environments. Virus activity and the clinical disease are now recognised as occurring annually in Australia, and epidemic polyarthritis, a non-fatal but often debilitating infection, appears to present an increasing problem in southeastern Australia.
- Arboviral encephalitis was probably first recorded when an outbreak of encephalitis and related syndromes (called "Australian X disease" at the time) occurred in 1917 and 1918 in the far west of NSW and southern Queensland, and again in 1925 and 1926 in NSW. A further outbreak in 1951 resulted in the isolation of an arbovirus known as Murray Valley Encephalitis virus. The most recent major outbreak occurred in 1974 with a concentration in the Murray Valley region of southern NSW and northern Victoria, although all mainland states were affected. Since 1974 the only cases recorded have been in Western Australia, The Northern Territory and Queensland, with none in southeast Australia, although the related Kunjin virus, which causes similar symptoms, has been active in both northern and southern regions in the intervening years.

In Australia, therefore, there is an obvious necessity to study mosquito vectors of disease. However, the irregular, sporadic nature of disease outbreaks in this country has often hindered, interrupted, or even precluded continuing research in this field.

Mosquito-borne diseases are especially complicated communicable diseases, because they involve the vector as an additional component of the disease system. Social, behavioural, environmental and immunological factors may affect the human component, yet with vector involvement further influences impinge on the system and still more may arise if the disease is a zoonosis, involving other vertebrate animals as well as humans. Such a complex system may seem formidable, but the more complicated the system, the greater the number of 'weak-point' targets for attack – the prime reason for seeking the fullest possible understanding of the system we are trying to disrupt (i.e. control).

The risk of being infected with a mosquito-borne disease in Australia depends to some extent on local circumstances. However, diseases aside, there are many situations where mosquitoes may present an unacceptable pest problem of a seasonal, perennial or incidental nature. Large numbers of mosquitoes can disrupt agriculture, industrial and recreational activities, and a community may require control of the nuisance pests to sustain a 'quality of life'.

Thoughts of mosquito control or eradication can be raised in both disease and pest contexts, and in Australia local populations may be unfamiliar with the risks associated with mosquitoes and the potential for the control of disease or pest situations. Mosquito control, whether in response to disease or pest problems, has become recognised as a public health issue in Australia principally since the major outbreak of arboviral encephalitis in 1974 and the increasing problems with Ross River virus infection in the early 1980s.

The main thrust of research in vector-borne disease is often toward control of the disease through control of the pest or vector. Although vector control programmes in various parts of the world have often failed, or have been interrupted, for a variety of reasons such as administrative problems, insecticide resistance, or behavioural characteristics of the vector population, a detailed knowledge of the ecology and behaviour of the target species is considered a prerequisite to planning a control campaign. The value of obtaining base-line data on the vector populations if control is to be undertaken has often been stressed by scientists working with mosquito-borne diseases, and a reduction of a vector population to a certain level (rather than its eradication) can often be achieved and indeed be sufficient for control of the transmission of disease.

The problems of containing mosquito populations at tolerable levels with the various techniques developed for mosquito control have led to an appreciation of the value of developing an educated approach wherein different methodologies are integrated such that reliance on particular control operations, whether physical, chemical or biological is avoided. As a result, many of the problems inherent in long-term control are avoided.

This course is intended to impart an appreciation and understanding of mosquitoes, their potential role in disease transmission, the importance of accurate identification of species, and the various methodologies used for controlling mosquito populations. Mosquito problems are a product of the contact between humans and mosquitoes, i.e. an interaction of human habits/habitats with mosquito habit/habitats. Understanding mosquitoes, their place in the natural environment, and the strengths and weaknesses of their ecology are essential prerequisites for successful control efforts.

4. MOSQUITOES AS PESTS IN WESTERN AUSTRALIA

Adapted from a paper by **A E (TONY) WRIGHT**

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INTRODUCTION

Western Australia (WA) is a very large State with climatic conditions varying from temperate mediterranean with mostly winter rainfall, through temperate and tropical arid zones, to submonsoonal and monsoonal tropics with heavy summer rain. Each of these climatic regions has characteristic mosquito faunas, although overlaps in species distributions occur for species which are more versatile in their breeding site requirements and more tolerant of a wider range of climatic conditions.

In all areas of WA, mosquito activity periods are seasonal due to the seasonal nature of rainfall, and (in the southern half of the State) seasonal temperature factors. The amount of rainfall often varies greatly from one year to the next - how often does Broome get 430mm (17 inches) of rain in one day as it did on 30 January this year? Sometimes Broome only receives less than half of that amount in a whole year. In the arid zones rainfall is never reliable and can occur at almost any time of year. In the Pilbara for example, there can be heavy summer rainfall from cyclones in some years (like 2000), but very little in others. The Pilbara can also receive large amounts of rain in winter in some years, e.g. 1992, and very little in other years.

Even in areas like Denmark and Albany on the South Coast, rainfall can vary considerably in both amount and timing from one year to the next - so, mosquito nuisance problems throughout WA vary greatly between seasons and between years, and it is essential to understand and accept this factor when considering both mosquito control programs and public education concerning the use of personal preventive measures.

Most pest mosquitoes, and for that matter disease vector mosquitoes, in WA breed in a wide variety of natural situations and cause pest or nuisance problems when people either live too close to these situations, or visit them for work or recreational purposes. This paper reviews the natural mosquito breeding situations and the dominant species that occur there for each climatic zone in WA.

DOMESTIC MOSQUITOES

In addition to mosquitoes that breed in natural situations two species of mosquitoes breed commonly throughout WA in domestic (i.e. artificial) situations. These two species are *Culex quinquefasciatus* and *Aedes notoscriptus*.

Culex quinquefasciatus breeds in a wide variety of domestic situations in both clean water and water which contains varying degrees of organic pollution. It breeds most heavily in polluted water, such as septic tanks which are not properly sealed and/or the vent pipe is not screened or cowled, poorly maintained sewage lagoons, and street gullies (especially silt traps). It also breeds heavily in organically polluted earth drains and ditches such as those sometimes found adjacent to stock yards and the like. *Culex quinquefasciatus* also breeds in containers holding clean water, such as fish ponds, blocked roof gutters, rainwater tanks, bird baths, old tyres, pot plant drip trays, old ice cream containers, buckets and the like. This species also breeds in large numbers in swimming pools which have "gone off" due to lack of maintenance, especially at the end of winter and spring after periods of not being used. *Culex quinquefasciatus* readily bites humans at night and will enter buildings in search of bloodmeals.

Aedes notoscriptus does not breed in heavily polluted water, but does breed in all of the clean water situations which are also colonised by *Culex quinquefasciatus*, especially where the water contains dead leaves. This species is a vicious biter both at night and during early morning and late afternoon, especially in shaded situations. It appears to be particularly prevalent in lush, shaded ferneries and other lush cool areas provide excellent harbourage and the opportunity to attack unwary gardeners.

MOSQUITO BREEDING IN NATURAL SITUATIONS IN THE DIFFERENT REGIONS OF WA

A. SOUTH WEST LAND DIVISION

For the purpose of discussing mosquitoes the south-west land division is the area which receives predominantly winter rainfall and lies south-west of a line drawn between Kalbarri and Esperance.

In more arid areas of this region the mosquito fauna is dominated by temporary ground pool breeding *Aedes* species, however in the wetter areas mainly located southwest of a line between about Bunbury and Albany, more permanent water breeders of the genera *Culex*, *Anopheles* and *Coquillettidia* can also be very abundant and cause serious nuisance problems.

Aedes camptorhynchus This is the most widespread and abundant mosquito throughout the south-west land division. It breeds in a wide variety of temporary and seasonal water bodies following winter/spring rain, as well as brackish wetlands and tidal saltmarshes fringing estuaries. It is also the dominant mosquito in many areas of the wheatbelt, where overclearing has led to soil salinity in many low-lying areas and the colonisation of salt-tolerant samphire (*Sarcocornia* sp.) vegetation.

There appears to be a very strong association between high density breeding of *Aedes* and *Sarcocornia* vegetation. This probably reflects the inclination of egg-laying female *Aedes camptorhynchus* to choose the base of *Sarcocornia* plants as favoured oviposition sites.

This species readily disperses 3-5km in search of bloodmeals, especially in heavily wooded areas, and bites humans viciously at night and during the day in mild or cloudy and humid weather.

Aedes vigilax As described in the Kimberley section, however in the south-west this species only occurs between December and March/April following high tides in estuarine areas. It only appears to occur in high numbers from Bunbury northwards.

Aedes alboannulatus This species has a widespread distribution throughout the south-west land division, including the Perth metropolitan area. It breeds in a wide variety of rockpools and temporary ground pools, both sunlit and shaded, and will also breed in containers under similar conditions as *Aedes notoscriptus* (i.e. not polluted water, but fresh water with dead leaves etc.). It is generally only present in moderate numbers, however it is a significant nuisance because it bites humans viciously both at night and during the day in the shade or in mild weather during winter and spring when it is most prevalent.

Aedes clelandi This species occurs throughout the wetter areas of the southwest land division west of a line between Albany and Lancelin. It is usually univoltine (one generation per year), with adults biting only during spring (August-October). It breeds in a variety of temporary freshwater ground pools, including roadside ditches, pools in paddocks and seasonal swamps, especially tea-tree swamps. It usually prefers sunlit and clear water (i.e. not turbid).

The adults of this species bite viciously, both at night and during the day, and it appears able to disperse at least 1-3 km away from breeding sites.

Aedes hesperonotius This species is similar in habits to *Aedes clelandi*, however its breeding sites appear to be largely confined to tea-tree swamps, so it is less widespread and more localised in distribution than *Aedes clelandi*.

Aedes ratcliffei This species occurs throughout the wetter areas of southwest land division west of a line from Perth to Albany. It is usually univoltine (one generation per year), with adults active between September and November each year. This species appears to breed mainly in paperbark swamps where the water is clear and shaded, but contains a high tannin content.

The adults of this species bite humans viciously both by day and night, but do not appear to disperse more than 1km away from their swampland breeding sites. Whilst it can be a severe nuisance close to breeding sites it appears to be fairly localised in distribution.

Aedes sagax This species was covered in the section on arid zone mosquitoes. Within the south-west land division it occurs in the wheatbelt areas as a significant nuisance in spring, especially when seasonal winter and early spring rainfall is above average.

Culex annulirostris This species is covered in the section on Kimberley mosquitoes. In the south-west land division it occurs mainly in heavily vegetated permanent or semi-permanent lakes and swamps and only reaches significant nuisance levels during the summer months (November to March) when water temperatures are consistently above about 18°C.

Coquillettidia sp. nr. linealis This species occurs on the Swan Coastal plain and breeds in permanent heavily vegetated swamps and wetlands. It is most prevalent in wet years when high water levels maximise the area of *Typha, Baumea* and other thick emergent vegetation in contact with water; the larvae of this species depend upon the interface area between water and thick emergent vegetation for their development. This species is active from October onwards until water levels recede away from emergent vegetation within its breeding sites due to seasonal drought (between December and March according to seasonal conditions).

This species bites humans viciously at night and appears to travel up to 3km in search of bloodmeals.

B. THE KIMBERLEY REGION

The spectacular and varied natural environment in this region provides a wide range of temporary, seasonal and permanent mosquito breeding sites. These breeding sites are colonised by no less than 55 species of mosquito, several of which occur in numbers sufficient to constitute a significant pest problem, and which readily bite humans, as follows:

Aedes vigilax This species breeds in huge numbers in coastal areas throughout the region when heavy rain or high tides cause temporary pooling of water on its saltmarsh breeding sites. It is the most significant pest species in coastal areas throughout the region because, as well as occurring in huge numbers it also bites viciously at any time of day or night and

disperses up to 100km inland, and in significant numbers up to 20km. It is most prevalent in October - December and March - May in the Kimberley.

Aedes normanensis This species breeds in temporary freshwater pools on river floodplains and along creek lines following flood rains. Whilst it does not appear to breed at such high densities as *Aedes vigilax*, the sheer size of its breeding sites often mean that it occurs in plague numbers following heavy rain. It occurs in greatest numbers after the first heavy seasonal rains in November - January, during the wet season if there is a dry spell, and immediately following the last heavy rains of the season, usually in March or April. It is a vicious biter of humans, both at night and during early morning and late afternoon, and appears to readily disperse up to 2-3 km. It will also bite at any other time of the day when conditions are fairly still, humid and overcast.

Aedes lineatopennis This species only occurs in significant numbers in the lower Ord River region around Kununurra. There its occurrence and habits are similar to those of *Aedes normanensis*.

Aedes tremulus This species breeds in treeholes as well as some artificial containers such as rainwater tanks. It appears to breed most readily in rot-holes in old rivergums (*Eucalyptus camaldulensis*), Coolibah Trees (*E. microtheca*) and ghost gums. it occurs at low to moderate density throughout most of the Kimberley region and at higher densities along Sturt Creek and at Derby. It does not appear to disperse more than a few hundred metres at best, but is a vicious biter, especially early in the morning for the first hour or two after dawn in still conditions. It is most prevalent at the end of the wet season (March - April).

Other floodwater *Aedes* **species** Several species of *Aedes* which can breed in plague numbers following heavy rain occur in the more arid areas of the Kimberley south of Halls creek and Fitzroy Crossing. These include *Aedes bancroftianus, Aedes pseudonormanensis, Aedes eidsvoldensis* and *Aedes* sp. #85. These will be covered in more detail in the next section covering the Pilbara Region, where they are more widely distributed and occur in more heavily populated centres.

Culex annulirostris This is the most common and widespread species in the Kimberley Region, and breeds in a wide variety of temporary, seasonal and permanent freshwater sites where some emergent and/or marginal vegetation occurs. It also breeds at very high densities in sewage oxidation ponds where marginal vegetation (especially couch grass) is allowed to colonise their margins, and in unmanaged overflows from these lagoons. It bites mainly during the first 2-3 hours after sunset and again around dawn, although it also bites during the night and sometimes during the day if humid and overcast weather prevails. It occurs in greatest numbers during the second half of the wet season (February - April) and the first half of the dry season, when semi-permanent billabongs and large river floodplain pools persist to provide breeding sites. This species appears to readily disperse 2-5km in search of bloodmeals and occasionally up to 10km.

In the Ord River region this species breeds year round in both the heavily vegetated swampy margins of Lake Kununurra and in poorly maintained irrigation ditches and drains (i.e. those with heavily vegetated margins).

Culex sitiens This species is common in coastal areas of the Kimberley, where it breeds in the larger and more persistent brackish and saline pools remaining on saltmarsh areas following heavy rainfall or high tides. It appears to breed most abundantly in saltmarsh areas where human activity inadvertently results in the impoundment of water, e.g. roads with inappropriately sited culverts. It bites viciously and mainly at night and can disperse up to 30km in search of bloodmeals. It occurs in greatest numbers during the wet season (December - April), when higher water tables allow greater persistence of large brackish groundpools.

Mansonia uniformis This species breeds in heavily vegetated permanent freshwater, especially in association with *Cumbungi (Typha)* and various species of water lilies. It bites viciously at night, but in WA only occurs in large numbers in the swamps adjacent to Lake Kununurra during the latter half of the dry season (July-November).

Anopheles annulipes s.l. This species breeds in a wide variety of permanent, semipermanent and temporary freshwater sites, and occasionally in moderately polluted or brackish water sites. It bites viciously at night and occasionally during the day, and is most prevalent during the mid-dry season, especially around Lake Kununurra.

Anopheles bancroftii This species breeds in shaded, vegetated, permanent and semipermanent freshwater sites throughout the Kimberley, but in WA it only appears to breed in large numbers in the Ord River region, especially around the swampy margins of Lake Kununurra. It bites viciously at night and during the day in shade. It is most abundant during the early dry season (April-June) each year.

C. THE PILBARA REGION

Despite the arid and rugged nature of the Pilbara region, several species of mosquito occur in plague numbers following infrequent heavy rainfall, and two further species are significant pests as a result of tidal flooding of saltmarshes. In the Pilbara mosquito plagues following heavy rainfall occur mainly in flatter areas where runoff is less than in more rocky terrain, and also in coastal areas. Because of the need to survive long and irregular drought periods, the Pilbara mosquito fauna is usually dominated by temporary ground pool breeding *Aedes* species.

Aedes vigilax As for the Kimberley Region, but most prevalent following heavy summer rainfall when saltmarsh flooding occurs over wider areas and rates of water infiltration into the soil are reduced by higher water tables.

Aedes normanensis As for the Kimberley Region, however this species only appears to occur in significant numbers within the De Grey River catchment when the river floods after cyclones. Even then it does not appear to occur in plague numbers as it can in the Kimberley Region.

Aedes tremulus As for the Kimberley Region, but it only appears to occur at low to moderate population levels.

Aedes bancroftianus

See section on arid zones

Aedes pseudonormanensis This species is usually the most widespread and abundant mosquito throughout the inland Pilbara following heavy rainfall. It is also abundant following heavy rainfall in the SE Kimberley from Halls Creek south, and has been recorded in lower numbers from parts of the inland arid zones following summer rain.

It breeds in a variety of freshwater temporary ground pools especially in sunlit, turbid water. Larvae and pupae of this species have been observed at very high densities in residual pools along creeklines and on river floodplains when floodwaters have receded (often very rapidly), e.g. Strelley River, creeks east of Pardoo, Devils Creek (south of Karratha) and the creeks in the "Pound" at Balgo Mission. It bites humans viciously during both day and night, especially around dusk and appears to disperse several kilometres when the weather is not too hot, dry and windy.

Aedes sp#85

See section on arid zones.

Aedes alternans

See section on arid zones.

Culex annulirostris As for the Kimberley region, however the restricted nature and (usually) shorter duration of its seasonal wetland habitats in the Pilbara usually mean it is less prevalent than in the Kimberley. It is also prevalent in sewage oxidation ponds that are inadequately maintained, as for the Kimberley, however in the Pilbara it appears to be replaced by *Culex quinquefasciatus* during winter in these situations, probably due to cooler water temperatures.

Culex sitiens As for the Kimberley region; it has been particularly prevalent on the Wickham saltmarshes near the sewerage overflow.

Anopheles amictus As for the Kimberley Region, but generally less abundant and persistent as is the rainfall in the Pilbara (usually!).

Anopheles annulipes As for the Kimberley Region, but generally less abundant and persistent, as are the semi-permanent freshwater habitats in which it breeds.

D. ARID ZONES (GASCOYNE, MURCHISON, INTERIOR, GOLDFIELDS)

These regions are discussed together here because of the general similarities in mosquito faunas which have been recorded from the intermittent mosquito/virus collecting trips to these regions. Patterns of mosquito activity are similar to those in the Pilbara, and are usually characterised by sudden and relatively short-lived plagues of mosquitoes following soon after heavy rainfall. Again, and for the same reasons, the mosquito fauna in these regions is usually dominated by temporary ground pool breeding *Aedes* species.

Aedes vigilax As for the Kimberley Region, however, this species is dominant during the summer months in coastal areas of the Gascoyne region around Carnarvon and Shark Bay.

Aedes sagax/sp.#85 The taxonomy of these two closely-related species is unclear. Typically *Aedes sagax* occurs in the more temperate latitudes of these arid zones whereas *Aedes* sp.#85 occurs in the more tropical latitudes. However individuals which appear on the basis of morphology to represent both species (as well as some which look like intermediates) have been collected at both Onslow and the Kalgoorlie/Menzies/Leonora area. These two poorly defined species breed in plague numbers in a wide variety of freshwater temporary ground pools throughout the inland arid zones following occasional heavy rain (e.g. rainfall associated with cyclones). The adults bite humans viciously during both day and night and appear to be able to disperse at least 1-3km away from breeding areas.

Aedes bancroftianus This species occurs following occasional heavy rainfall and has been recorded throughout the tropical and temperate arid zones as well as the Pilbara and the southern Kimberley as far north as Louisa Downs Station, and as far south as parts of the wheatbelt. It was the dominant species collected at the end of the wet season at Balgo Mission in 1978. It bites humans viciously both at night and during the day and appears to disperse at least 1-3 km in search of bloodmeals. Larvae of this species occur in a variety of temporary ground pools, especially residual pools along creeklines after the creeks have flooded and the water levels subsided.

Aedes eidsvoldensis This species has been recorded throughout the tropical and temperate and arid zones as well as the Pilbara and the southern Kimberley as far north as Cherrabun Station and the Billiluna Community on Sturt Creek. It has been recorded in moderate to large

numbers at Billiluna, Leonora, Menzies, Kalgoorlie and Carnarvon and in plague numbers at Minilya north of Carnarvon. Its preferred breeding sites appear to be temporary ground pools along larger creeklines and low-lying claypan areas following occasional heavy rainfall. There appears to be an association between abundant breeding of this species and the heavier, reddish soils in these arid zones rather than other sandier soil types. This species bites humans viciously at night and during daylight in humid, still or cloudy conditions, and appears able to disperse at least 1-3kms in search of bloodmeals.

Aedes sapiens This large mosquito has been recorded in moderate to large numbers at Carnarvon and in the Kalgoorlie/Menzies/Leonora area following occasional heavy rainfall. Little is known of its larval habitats, but this species bites readily during the day and early evening.

Aedes alternans This very large mosquito has been recorded throughout the arid zones as far south as Coolgardie, and also throughout the Pilbara and most of the Kimberley as far north as Wyndham. Its larvae are predatory upon the larvae of other mosquito species, and it breeds in both temporary ground pools following heavy rainfall and brackish/saline pools left after high tides. It usually only occurs in low to moderate numbers (like most predators), but when it occurs it is noticeable because of its large size and vicious biting habits, and because it bites mainly during the day.

Aedes sp #71 This species occurs throughout the temperate and tropical arid zones, but has only been recorded in moderate to large numbers in the Kalgoorlie/Menzies/Leonora and Mount Magnet areas. Its breeding and biting habits appear to be very similar to *Aedes pseudonormanensis*, to which it appears closely related.
5. WHY MOSQUITOES CANNOT TRANSMIT AIDS

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(reprinted from: Rutgers Entomology New Jersey Mosquito Control Association Website: http://wwwrci.rutgers.edu/insects~insect/aids.htm, accessed 15 July 2004)

Media releases concerning the possibility of mosquitoes transmitting AIDS (Acquired Immune Deficiency Syndrome) were common when the disease was first recognized, and the subject is still addressed by tabloids that seek captivating headlines to increase their circulation. The topic was initiated by reports from a small community in southern Florida where preliminary evidence suggested that mosquitoes may have been responsible for the higher on average incidence of AIDS in the local population. The media was quick to publicize claims that mosquitoes were involved in AIDS transmission despite findings of scientific surveys of the National Centers for Disease Control (CDC) that clearly demonstrated that mosquito transmission of AIDS in that community appeared highly unlikely. Nevertheless, media releases perpetuated the concept that mosquitoes transmitted AIDS, and many people still feel that mosquitoes may be responsible for transmission of this infection from one individual to another.

There are three theoretical mechanisms which would allow blood-sucking insects such as mosquitoes to transmit HIV (the virus that causes AIDS).

1. In the first mechanism, a mosquito would initiate the cycle by feeding on an HIV positive carrier and ingest virus particles with the blood meal. For the virus to be passed on, it would have to survive inside the mosquito, preferably increase in numbers, and then migrate to the mosquito's salivary glands. The infected mosquito would then seek its second blood meal from an uninfected host and transfer the HIV from its salivary glands during the course of the bite. This is the mechanism used by most mosquito-borne parasites, including malaria, yellow fever, dengue, and the encephalitis viruses.

2. In the second mechanism, a mosquito would initiate the cycle by beginning to feed on an HIV carrier and be interrupted after it had successfully drawn blood. Instead of resuming the partial blood meal on its original host, the mosquito would select an AIDS-free person to complete the meal. As it penetrated the skin of the new host, the mosquito would transfer virus particles that were adhering to the mouthparts from the previous meal. This mechanism is not common in mosquito-borne infections, but equine infectious anemia is transmitted to horses by biting flies in this manner.

3. The third theoretical mechanism also involves a mosquito that is interrupted while feeding on an HIV carrier and resumes the partial blood meal on a different individual. In this scenario, however, the AIDS-free host squashes the mosquito as it attempts to feed and smears HIV contaminated blood into the wound. In theory, any of the mosquito-borne viruses could be transmitted in this manner providing the host circulated sufficient virus particles to initiate reinfection by contamination.

Each of these mechanisms has been investigated with a variety of blood sucking insects and the results clearly show that mosquitoes cannot transmit AIDS. News reports on the findings, however, have been confusing, and media interpretation of the results has not been clear. The average person is still not convinced that mosquitoes are not involved in the transmission of a disease that appears in the blood, is passed from person to person and can be contracted by persons that share hypodermic needles. Here are just some of the reasons why the studies showed that mosquitoes cannot transmit AIDS:

Mosquitoes Digest the Virus that Causes AIDS

When a mosquito transmits a disease agent from one person to another, the infectious agent must remain alive inside the mosquito until transfer is completed. If the mosquito digests the parasite, the transmission cycle is terminated and the parasite cannot be passed on to the

next host. Successful mosquito-borne parasites have a number of interesting ways to avoid being treated as food. Some are refractory to the digestive enzymes inside the mosquito's stomach; most bore their way out of the stomach as quickly as possible to avoid the powerful digestive enzymes that would quickly eliminate their existence. Malaria parasites survive inside mosquitoes for 9-12 days and actually go through a series of necessary life stages during that period. Encephalitis virus particles survive for 10-25 days inside a mosquito and replicate enormously during the incubation period. Studies with HIV clearly show that the virus responsible for the AIDS infection is regarded as food to the mosquito and is digested along with the blood meal. As a result, mosquitoes that ingest HIV-infected blood digest that blood within 1-2 days and completely destroy any virus particles that could potentially produce a new infection. Since the virus does not survive to reproduce and invade the salivary glands, the mechanism that most mosquito-borne parasites use to get from one host to the next is not possible with HIV.

Mosquitoes Do Not Ingest Enough HIV Particles to Transmit AIDS by Contamination

Insect-borne disease agents that have the ability to be transferred from one individual to the next via contaminated mouthparts must circulate at very high levels in the bloodstream of their host. Transfer by mouthpart contamination requires sufficient infectious particles to initiate a new infection. The exact number of infectious particles varies from one disease to the next. HIV circulates at very low levels in the blood--well below the levels of any of the known mosquito-borne diseases. Infected individuals rarely circulate more that 10 units of HIV, and 70 to 80% of HIV-infected persons have undetectable levels of virus particles in their blood. Calculations with mosquitoes and HIV show that a mosquito that is interrupted while feeding on an HIV carrier circulating 1000 units of HIV has a 1:10 million probability of injecting a single unit of HIV to an AIDS-free recipient. In laymen's terms, an AIDS-free individual would have to be bitten by 10 million mosquitoes that had begun feeding on an AIDS carrier to receive a single unit of HIV from contaminated mosquito mouthparts. Using the same calculations, crushing a fully engorged mosquito containing AIDS positive blood would still not begin to approach the levels needed to initiate infection. In short, mechanical transmission of AIDS by HIV-contaminated mosquitoes appears to be well beyond the limits of probability. Therefore, none of the theoretical mechanisms cited earlier appear to be possible for mosquito transmission of HIV.

Mosquitoes Are Not Flying Hypodermic Needles

Many people think of mosquitoes as tiny, flying hypodermic syringes, and if hypodermic needles can successfully transmit HIV from one individual to another then mosquitoes ought to be able to do the same. We have already seen that HIV-infected individuals do not circulate enough virus particles to result in infection by contamination. However, even if HIV-positive individuals did circulate high levels of virus, mosquitoes could not transmit the virus by the methods that are employed in used syringes. Most people have heard that mosquitoes regurgitate saliva before they feed, but are unaware that the food canal and salivary canal are separate passageways in the mosquito. The mosquito's feeding apparatus is an extremely complicated structure that is totally unlike the crude single-bore syringe. Unlike a syringe, the mosquito delivers salivary fluid through one passage and draws blood up another. As a result, the food canal is not flushed out like a used needle, and blood flow is always unidirectional. The mechanics involved in mosquito feeding are totally unlike the mechanisms employed by the drug user's needles. In short, mosquitoes are not flying hypodermic needles and a mosquito that disgorges saliva into your body is not flushing out the remnants of its last blood meal.

For more in depth information on this topic see Staff Paper #I, Do Insects Transmit AIDS?, OTA series on AIDS-Related Issues, Health Program, Office of Technology Assessment, United States Congress, Washington, D.C. 20510-8025.

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6. MOSQUITO BIOLOGY AND BEHAVIOUR: CASE EXAMPLES AND IMPLICATIONS FOR CONTROL

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INTRODUCTION

Mosquito control is not a matter of applying pesticide to the nearest body of water or bushland and hoping for the best. This approach is unlikely to achieve the desired result and may impact adversely on other, non-target animals. Instead, a range of carefully planned measures that takes into account important differences in the biology and behaviour of different target mosquito species must be considered.

Different mosquitoes breed in very different habitats, have diverse requirements for breeding and survival and behave differently. These differences impact substantially on how, when, where and whether we control particular mosquito species.

This chapter reviews aspects of the biology and behaviour of mosquitoes in relation to control strategies and requirements. Three common Australian mosquitoes are used to illustrate different strategies and requirements for different target species. The examples used are by no means all-inclusive, and should not be regarded as applicable to all pest or vector mosquito species. However, the concepts presented provide a guide to aspects of mosquito biology and behaviour that should be considered when developing control programs or policies to effectively minimise contact between humans and particular mosquito species.

BIOLOGY AND BEHAVIOUR

The biology and behaviour of mosquitoes is discussed in detail elsewhere in the 'Mosquito Biology and Ecology' chapter by Professor Richard Russell on page 2. This section is intended as an overview only, referring to those aspects that impact upon the requirements and strategies for control. Table 1 summarises some aspects of the biology and behaviour of mosquitoes that influence mosquito control.

LARVAL HABITAT

Worldwide, there are about 3500 different mosquito species of which more than 200 occur in Australia. Over 90 species have been recorded in Western Australia (WA) alone. As a group, mosquitoes have colonised a vast array of aquatic environments for breeding, including such diverse habitats as tree holes, floodplains, sewage ponds, saltmarshes, water-filled debris/rubbish, leaf axils, coastal rock pools, gutters, irrigation ditches, billabongs, wheel ruts, septic tanks, storm water drains and many more. However, individual mosquito species may be restricted to particular geographic regions or even specific habitats within a region, and most have preferred larval habitats, selected by the adult female when she lays her eggs. Within a particular habitat other factors such as sunlight or shade, the presence or absence of emergent vegetation and prevailing winds, which can drive larvae towards one end of a breeding site, may also be important. It is essential to locate the specific site of breeding of a problem species in order to attempt control of the larval (aquatic) stages of that species.

LOCATION OF HABITAT

Location of breeding habitat in relation to human habitation and activities is important. Some species breed specifically in man-associated habitats and are therefore only likely to be abundant in urban or agricultural settings. Many species breed exclusively in natural environments and are only regarded as pests if humans visit or live near such habitats. Alternatively, some species associated with natural habitats can be a problem if they are capable of dispersing large distances into populated areas in search of a bloodmeal. These are important considerations for determining the need for, means and extent of control. Dispersal ability of relevant species must also be taken into account when considering the location of new residential developments and projects involving agricultural practices or manipulation of water that may promote breeding of mosquitoes.

SIZE AND DISTRIBUTION OF HABITAT

Size (extent) and distribution of larval habitat varies enormously. Some species breed in multiple, small breeding sites (e.g. leaf axils, tree holes, domestic containers). It may not be possible to treat these with chemicals on a regular basis, so other strategies such as source reduction are required to achieve a reduction in breeding. Controlling mosquitoes breeding in larger, more contiguous habitats (e.g. saltmarshes, temporary freshwater ground pools, etc) may be possible if resources to develop large-scale chemical application or source reduction programs are available. In reality, many rural communities in Australia do not have the resources to implement programs of sufficient magnitude to effectively control breeding of the principal rural pest species associated with widespread flooding or a series of exceptionally high tides. Under these circumstances, timely and well-targeted public warnings and education about how to minimise exposure to mosquitoes become extremely important.

OTHER CHARACTERISTICS OF HABITAT

Consideration should also be given to other characteristics of breeding sites of some species before attempting chemical control. Delivering the correct dose of chemical to larvae in deep water bodies or sites that are frequently inundated (e.g. by tides or man-made activities) may be difficult or impossible. Products that require several days of contact with target species to ensure effectiveness (e.g. some s-methoprene products) can be diluted to sublethal concentrations in environments where ongoing flushing occurs. Dense emergent, fringing or floating vegetation in some habitats can reduce the amount of applied larvicide that actually gets into the water. This too may result in only sub-lethal concentrations of product coming into contact with the target.

ENVIRONMENTAL CONDITIONS

TEMPERATURE

Temperature plays a major role in the development and survival of mosquitoes. Some species are temperature-limited. This means they only breed when environmental temperatures are within certain ranges. Examples of these are discussed later in this chapter. Familiarity with the temperature ranges of target species is essential. For example, there is no value in monitoring or attempting to control the summer saltmarsh mosquito in mid winter in the southwest of WA.

Temperature also plays an important role in the rate of development of larval mosquitoes. Some species may take weeks to develop, or even enter 'larval hibernation' under winter conditions. The same species may develop from egg to adult in as little as 4-5 days in summer. This has fundamental implications for the 'window of opportunity' for controlling mosquitoes as larvae. An entire generation of mosquitoes may pass through the stages at which some larvicides are effective in as little as 2-3 days in summer. This presents a major problem if there are large areas of larval habitat that require treatment.

Temperature also has a profound effect on adult mosquitoes. Extremes of temperature may be detrimental to adult populations, reducing numbers to below nuisance levels relatively quickly. In contrast, milder temperatures may assist their survival and dispersal, leading to persistence of severe nuisance populations and a potential disease risk for a considerable period.

RAINFALL AND TIDES

Rainfall, tides, humidity, wind, predators, pathogens and many other environmental factors also play important roles in determining when and where mosquitoes breed. It is not possible to review all of these in detail here. However, monitoring of rainfall and tides can act as a useful predictor of mosquito breeding in some settings.

In particular, a rapid rise in the numbers of *Aedes* species that use temporary ground pools can be expected soon after rains, floods or high tides that top up ground pools around which their desiccation resistant eggs have been deposited. Similarly, container and tree-hole breeding *Aedes* species also require a rise in the water level to flood their eggs. This can result from rains, human activities such as the use of sprinklers for irrigation, or other sources. Further water level increases (e.g. more rains, tides, irrigation, etc) are required before additional generations of these ground pool or container *Aedes* will hatch. Monitoring and control measures for these species may therefore be necessary almost immediately after a significant rainfall, flooding or tidal event.

In contrast, the eggs of *Culex*, *Anopheles*, *Mansonia* and *Coquillettidia* mosquitoes are not desiccation-resistant and are deposited directly on or in existing water bodies. Unlike *Aedes* mosquitoes, several generations of these species may occur after heavy rains, floods or tides before nuisance or plague populations are reached. However, these species can also persist in large numbers long after *Aedes* populations have dwindled. The need to monitor and control *Culex*, *Anopheles*, *Mansonia* or *Coquillettidia* species may be greatest after rains, flooding or high tides that create abundant breeding habitat. However, some may also breed more or less continuously in favourable habitats, with or without a rise in water level.

BEHAVIOUR/ECOLOGY

EGGS

Many *Aedes* species undergo installment hatching, where only a proportion of viable eggs hatches in response to a given environmental trigger. This is a survival mechanism that ensures that some eggs remain to recolonise a breeding site in the event that individuals from earlier hatchings do not survive. It has important implications where eradication is being attempted (e.g. following incursion of an exotic mosquito species such as *Aedes aegypti*). In the case of *Ae. aegypti*, which breeds in domestic containers, several cohorts of larvae are possible from one batch of eggs if the container is flooded repeatedly. To overcome this, it may be necessary to use separate approaches to killing eggs and larvae. Alternatively, such sites must be monitored and treated for several rounds of flooding to ensure no additional breeding will take place.

LARVAE

The larvae of some species can be extremely cryptic or evasive within their breeding site and great care needs to be taken to ensure that they are not missed during monitoring. The larvae of *Mansonia* and *Coquillettidia* species do not surface, but remain attached to the submerged stems of aquatic vegetation. These are virtually impossible to monitor directly and therefore indirect methods of monitoring are usually required.

Larvae of different species feed at different water levels within a breeding site. *Aedes* species are typically bottom feeders whereas *Anopheles* species usually feed at the surface. *Culex* species generally feed in the middle range below the surface and, as mentioned above, *Coquillettidia* and *Mansonia* remain attached to submerged vegetation. Thus, it is important to ensure that the formulation and method of application of chemical used to control a larval population, particularly for those chemicals that must be ingested, will convey the active ingredient to the appropriate level within the breeding site.

ADULTS

Some mosquito species are autogenous. This simply means the female can lay an initial batch of eggs without needing to obtain additional protein via a bloodmeal. From a control perspective, this has two important implications. Firstly, subsequent generations of mosquitoes can occur at a breeding site despite a successful control program that targets adult females leaving that breeding site in search of a bloodmeal. Secondly, autogenous species may remain near their breeding site for longer than non-autogenous species, while they mature their first batch of eggs. Consequently, the timing of movement away from a breeding site in search of a bloodmeal will differ between populations of autogenous and nonautogenous species. Different timing of application of adulticides may therefore be required to control such populations moving into residential areas.

The biting behaviour of different mosquito species varies substantially. Some bite mainly at dusk and/or dawn, some mainly during the day and others only at night. Some mosquitoes only bite outdoors, while others will enter dwellings in search of a bloodmeal. Some will only bite when environmental conditions are extremely favourable (e.g. mild temperature, high humidity and little or no wind). Others are hardier and will bite under most conditions. Strategies to control or avoid adult mosquitoes must be tailor-made to suit the biting behaviour of the target species. For example, adulticide application should be timed to coincide with the period of peak activity of the adult population. Repellents and protective clothing should be recommended where humans may come into contact with day-biting species, whereas flyscreens or bednets may be more appropriate in areas where indoor, night-biting species are abundant.

Biological/behavioural/ecological considerations		Example
Breeding habitat	Type of breeding habitat	Tidal saltmarsh - runnelling is only suitable for some saltmarsh situations
	Physical characteristics of breeding site	Regular flushing, high organic content, deep water, etc may pose problems for control
	Location of habitat (in relation to human habitation)	Determines severity, and timing of problem and need for control
	Natural, man-made or domestic breeding	Source reduction may be attempted for many man-made breeding situations. Public should be encouraged to reduce domestic breeding
	Extent/number/abundance of breeding sites	Very different control strategies are required for different types, sizes of breeding sites.
	Presence/absence of vegetation	Thick emergent vegetation may prevent application or limit effectiveness of larvicides.
	Shady, sunlit or windy	Larvae may be concentrated in shady, sunlit or downwind areas only.
	Regular flushing of breeding site	Frequent tides or other sources of flushing may dilute slow-acting larvicides.

Table 1: Examples of the biology and behaviour of mosquitoes that may determine the specifics of mosquito control programs for different species.

Table continued on next page

 Table 1 (cont): Examples of the biology and behaviour of mosquitoes that may determine the specifics of mosquito control programs for different species.

Biological/behavioural/ecological considerations		Example
Timing/seasonality	Temperature/season	Many species are temperature restricted, e.g. <i>Cx. annulirostris, Ae. vigilax, Ae.</i> <i>camptorhynchus</i>
	Water temperature	Determines rate of development of larvae, dictates timing of monitoring and larval control
	Rainfall, tides, temperature	Useful predictors of timing of breeding of some important species.
Eggs	Instalment hatching of eggs	Several cohorts of larvae may result from one batch of eggs. Ongoing control/monitoring may be necessary (eradication of exotic species)
Larval behaviour	Region of active feeding within breeding habitat (e.g. surface, substrate)	Ensure larvicides requiring ingestion are delivered to appropriate site
	Preference for open water or water with dense fringing or emergent vegetation	May determine ability to deliver larvicide to target species
	Evasive or cryptic nature of larvae	May make direct monitoring difficult or impossible – seek alternative methods
Adult biology	Autogeny (egg laying before a blood meal is taken)	Affects timing of movement of adults away from breeding site
	Vector competence (ability to transmit a particular disease/pathogen)	Determines need to control a species (for health purposes)
Adult behaviour	Dispersal capabilities (distance/'hardiness')	Important for determining how far from human habitation that control may be necessary
	Timing of dispersal/bloodseeking	Determines when adults are active and therefore when methods for adult control are likely to be effective. Also important for warnings to public
	Host preference – man-biting or not?	It is not necessary (or productive) to attempt control of species that do not regularly feed on humans
	Day-biting	Repellents and protective clothing recommended
	Preference for entering dwellings	Fly screens or bed nets may be required to prevent exposure to these species.
	Cues for bloodseeking	Some species may not enter CO ₂ traps. Other methods (e.g. ovitraps, emergence traps, animal traps may be required)
Other issues	Environmental significance of breeding habitat	Chemical control or source reduction may not be permitted in conservation areas. Residential areas should not be located close to conservation areas that also breed vector mosquito species.

This table is intended as a summary only. Additional information is available in other chapters of this manual; Liehne, 1990; Lee *et al.*; 1988, MCAA, 2008; or Russell, 1993.

Cues for blood-seeking and host preference of adult mosquitoes are also important considerations. Some species will not readily enter carbon dioxide-baited traps, so other methods such as ovitraps or animal-baited traps are used for surveillance. There is little benefit in controlling species that do not bite humans (unless they are a known maintenance vector of a human pathogen). Some species are vicious biters, whereas others may be

extremely timid. A small or moderate size population of vicious biting mosquitoes may be perceived as a much greater problem by the public than a large population of a timid or non-man-biting species. This should be taken into account when interpreting adult monitoring results.

Finally, and very importantly, only some mosquito species are vectors of pathogens that cause human disease. Potential or known disease-carrying species should be targeted for control in preference to nuisance-only species, particularly if a mosquito control program is operating with limited resources.

CASE EXAMPLES

AEDES VIGILAX: THE SUMMER SALTMARSH MOSQUITO

This vicious-biting mosquito is a major pest species in many coastal regions of Australia. Ae. vigilax is also an important vector of Ross River (RRV) and Barmah Forest (BFV) viruses.

In WA, *Ae. vigilax* has been implicated as a vector of RRV and BFV in coastal areas from the Kimberley to the Swan Coastal Plain in the south-west.

OCCURRENCE

Ae. vigilax breeds in coastal saltmarshes and



brackish swamps around Australia except along the south coast. It is normally associated with saline and brackish ground pools that remain after spring tides or heavy rains on saltmarsh, mangroves, brackish reed swamps, tidal creek extremities and other similar habitats.

Ae. vigilax will readily disperse substantial distances (between 5 and 50 kilometres) from its breeding sites in search of a blood meal. It is one of the most important pest and vector species for coastal communities within 5-20 km of its coastal breeding sites. However, it will also affect communities further inland if environmental conditions assist dispersal.

Breeding of *Ae. vigilax* is temperature limited. In northern areas of its range it can breed yearround if tides or rains permit. However, in southern areas of its distribution (e.g. south-west of WA and south coast of NSW) it only breeds during the warmest (summer) months. It is not found at all along the south coast of Australia.

BIOLOGY/BEHAVIOUR AND IMPLICATIONS FOR CONTROL

Breeding habitat

Proximity to the coast (and tidal saltmarshes) should be considered when assessing the likelihood of problems attributable to this species. Inland communities are unlikely to experience a major problem with *Ae. vigilax*. In southern areas of its range, the time of year (daily temperatures) should also be taken into account. For example, there would be no requirement for control of *Ae. vigilax* in the middle of winter on the south coasts of WA or NSW.

Coastal communities surrounded by *Ae. vigilax* breeding habitat may face a severe problem that can be difficult or virtually impossible to control. The capacity for dispersal of this species means that breeding sites that are a considerable distance from human habitation may still

pose a problem, irrespective of whether effective control has been achieved at nearby sites. Dense vegetation may assist dispersal by providing harbourage (resting sites) for the adults. Buffer zones in which vegetation has been cleared or reduced may help to reduce the potential for adults to disperse into residential areas.

Larviciding should be regarded as the principle method of control for *Ae. vigilax* where possible. Regrettably, in many cases the scale of the problem and the inaccessibility of *Ae. vigilax* breeding sites after spring tides or heavy rains make it almost impossible to effectively control this species in some regions.

The vast areas of *Ae. vigilax* larval habitat that may require treatment are minor when compared to controlling a population of adults that has dispersed over tens or hundreds of square kilometres. Limited access to areas of harbourage, unfavourable conditions for correct dispersal of the chemical and the sheer size of the region that may require treatment mean that adulticiding is often ineffective.

Even effective adulticiding may only provide short-term control, because more adults will invade residential areas once the chemical has dissipated. Despite this, adulticiding strategies such as barrier fogging or residual surface sprays can play an important role in situations where larval control is simply not possible. Furthermore, control of adult mosquitoes is a key strategy during an outbreak of mosquito-borne disease. Under these circumstances it is important to reduce exposure of communities to existing populations of infected adult mosquitoes, which cannot be accomplished by controlling larval populations.

Adult female *Ae. vigilax* deposit their eggs on the substrate (soil/mud) or vegetation of receding temporary pools on the upper salt marsh or salt-affected wetlands. The eggs hatch and larvae develop more or less synchronously when high tides or heavy rains subsequently flood these pools. Breeding areas can be very extensive (i.e. up to many hundreds of hectares) and difficult or impossible to access and treat with larvicides using ground-based equipment. In these situations aerial application of larvicides is the only option available. Unfortunately, this may not be practical (for financial and logistical reasons) unless there are sufficient funds to enable an aerial application program. Aerial application is expensive and requires specialised equipment and well-trained staff.

Breeding of *Ae. vigilax* can occasionally occur in response to inundation of saltmarsh breeding habitat from man-made sources, including storm water run-off, sewage overflows and various forms of impoundment. This should be avoided, with careful planning or modification and maintenance of such facilities. It is not acceptable to resort to chemical control measures for mosquito breeding problems that can be prevented or resolved by proper design and maintenance.

Many breeding sites of *Ae. vigilax* are environmentally significant wetlands. It may not be appropriate to undertake chemical or physical control of mosquitoes in such habitats. This can pose a dilemma for public health authorities faced with a need to control a known vector species breeding in a habitat that environmental authorities would like to conserve.

Source reduction may be a realistic control method of *Ae. vigilax* in situations where physical alteration of the environment is acceptable. This can include selectively filling, draining or flushing of low-lying or disturbed areas of breeding sites.

The obvious limitations for control of *Ae. vigilax* described above mean that new residential areas should be located outside the dispersal zone of *Ae. vigilax* when planning future developments. This should be made clear to agencies and individuals responsible for decisions about planning in mosquito-affected regions.

Egg/larval biology and behaviour

The eggs of *Ae. vigilax* are desiccation-resistant. They survive for months and possibly years on dry breeding sites, waiting for inundation of the site. Widespread, high-density breeding can commence soon after inundation. Control programs for this species must be in place *before* inundation occurs. There will not be time to prepare a control program once breeding has been initiated.

The rate of development of *Ae. vigilax* larvae increases as water temperature increases. In hot (summer) conditions this species can develop from egg to pupa in as little as 4-5 days. Field monitoring to determine the timing of breeding prior to control efforts for *Ae. vigilax* is essential.

The rapid life-cycle means that there may only be a narrow window of opportunity to control a generation of larvae of this species after a tidal or rainfall event. Consider having to control hundreds of hectares of inaccessible saltmarsh breeding within 2-3 days in mid-summer. The only way this can be carried out effectively in the short time available is to apply larvicide by helicopter. This is an expensive and recurrent exercise and must always be accompanied by pre- and post-treatment monitoring to ensure effectiveness.

Hatching of a new cohort of *Ae. vigilax* eggs requires a rise in the water level. Thus, breeding usually occurs in response to high tides, heavy rain or occasionally inundation of breeding sites by human activities. There is little point searching for larvae in the absence of such environmental triggers. Instead, it is more appropriate to wait a few days (depending on temperature) after the next high tides or rain to commence surveys. Experience and local knowledge (field monitoring) will be required in order to become familiar with environmental triggers for breeding at different locations.

Ae. vigilax larvae browse on the substrate (bottom) of their breeding sites. Larvicides that must be ingested in order to be effective must disperse through the water column, rather than staying on top of the water.

Adult biology and behaviour

Ae. vigilax is autogenous. Females deposit small batches of eggs even before they find their first bloodmeal. This ensures survival of another generation. Monitoring is required to determine when a cohort will start to move away from the breeding site into residential areas. Under some circumstances it may be necessary to attempt control (adulticiding) of *Ae. vigilax* adults while they are still concentrated at the breeding site. However, this may have serious adverse impacts on other saltmarsh fauna if the wrong chemical is used and should be carefully planned.

Female *Ae. vigilax* will bite readily during the day and particularly at dusk. *Ae. vigilax* is a hardy species and may bite in hot, dry, windy conditions that are generally unfavourable for other species. It is a particularly vicious biter and catches of only 20 adults in a CO₂-baited trap usually represent a significant pest problem. It will also enter unscreened dwellings in search of a bloodmeal.

The vicious, day-biting characteristics of this species will mean that public demand for control in affected areas is likely to be high, despite the difficulties involved. Providing information about the problem to the public is an important adjunct under these circumstances. In particular, warnings about the need for repellents and protective attire throughout the day should be made available.

CO₂-baited traps are suitable for monitoring of *Ae. vigilax* adult populations and will provide a good indication of the severity of a current problem. Unfortunately, complaints from the public are also another common means by which the severity of an *Ae. vigilax* problem is assessed.

AEDES NOTOSCRIPTUS: THE CONTAINER MOSQUITO

Ae. notoscriptus is one of the major domestic pest species in Australia. Field investigations and recent laboratory studies suggest that Ae. notoscriptus may be an important carrier of RRV in urban areas.

RRV has been isolated from *Ae. notoscriptus* in residential areas in the south-west of WA.

OCCURRENCE

Ae. notoscriptus occurs throughout Australia,



including most areas of WA. It is found in natural habitats associated with tree and rock holes. *Ae. notoscriptus* has also successfully colonised a wide range of peri-domestic habitats, which is the main reason for its status as a major domestic pest species in Australia. Such habitats include artificial containers, tyres, drums, pot plant drip trays, roof gutters, rainwater tanks, bird baths, abandoned fish ponds, leaf axils (e.g. bromeliads), abandoned swimming pools and stormwater sumps/drains.

Ae. notoscriptus does not disperse far and is most commonly noticed as a pest species within 400m of breeding sites. In southern areas *Ae. notoscriptus* is found throughout the year, but only in low numbers in the coldest months. In northern areas it can breed throughout the year but is most abundant during the wettest months. However, the close association of this species with artificial container sites in domestic settings can result in breeding throughout the year.

BIOLOGY/BEHAVIOUR AND IMPLICATIONS FOR CONTROL

Breeding habitat

Breeding sites of *Ae. notoscriptus* are often small, but diverse and abundant, particularly after widespread rains. Control of *Ae. notoscriptus* in natural forest environments can be very difficult. It is not realistic to attempt to gain access to and treat so many individual and often cryptic sites after heavy rains. Adulticiding may be the only viable control option in circumstances where a community is at risk of exposure to RRV from this species in densely vegetated or forest regions.

Control of *Ae. notoscriptus* in urban/domestic environments is also a challenge due to the diverse nature and abundance of breeding sites across a suburb. In many instances breeding of *Ae. notoscriptus* results from poor management of water in ponds, bird baths, pet water bowls, gutters, pot plant drip trays, leaf axils and many other types of backyard container. This is preventable, and it is important to educate the public and enlist their help in minimising the potential for breeding of this species in their own backyards.

The preventable nature of urban breeding of *Ae. notoscriptus* means that residential zoning and planning are not as critical for this species, in comparison to *Ae. vigilax* or *Cx. annulirostris.* Exceptions to this are subdivisions located immediately adjacent to or in forested areas with tree-holes and/or rockpools that may breed *Ae. notoscriptus* (and may also harbour vertebrate hosts of mosquito-borne viruses). Buffer zones of up to 400m between such environments and residential areas will reduce interaction between *Ae. notoscriptus* and residents.

Egg/larval biology and behaviour

The eggs of *Ae. notoscriptus* are laid singly at the waterline on the inside of containers. Rainfall or artificial water sources (reticulation, fountains, etc) that raise the water level will trigger hatching of eggs.

Preventing domestic breeding of *Ae. notoscriptus* can be as simple as emptying water from artificial containers a few days after they have been flooded. As with *Ae. vigilax*, the development rate of *Ae. notoscriptus* is temperature dependent so containers should be checked and emptied more frequently in warmer weather.

Where removal of water from a container is not possible, biological forms of control such as introduction of fish (only in man-made situations) or application of a bio-larvicide may be appropriate. Restricting access of adults to potential breeding sites such as rainwater tanks will prevent breeding. This is achieved by ensuring the tank and its inspection port are completely sealed and that the inlet and overflow are fitted with robust mosquito-proof screening.

Application of slow-release insecticides may be necessary to control breeding of *Ae. notoscriptus* (and other species) in roadside stormwater sumps/gullies that do not completely drain. Some local governments have ongoing larviciding programs for this purpose. However, a longer-term solution for these sites is to modify and maintain them so that they do not retain water for more than a day or two.

Adult biology and behaviour

Adult *Ae. notoscriptus* do not disperse far from breeding sites. For this reason, breeding sites in urban areas should be possible to locate if adult monitoring indicates a significant pest problem. Adults will bite in the shade during the day with a peak in biting around dusk. Harbourage (dense vegetation) is important for survival and for day biting of this species. Removal of dense vegetation may reduce the potential for adults of this species to persist in suburban gardens.

Ae. notoscriptus is quite a vicious biter of humans in cool, humid or shady conditions. The species will readily enter CO_2 -baited traps and counts of > 30 *Ae. notoscriptus* per trap indicate that there is a pest problem. Avoiding shady, densely vegetated areas during the day, using repellents and avoiding being outdoors around dusk will reduce exposure to this species in the event that control of larval populations has not been possible.

Ae. notoscriptus is a competent vector of RRV and BFV in the laboratory and RRV has been isolated from this species in urban outbreaks of RRV disease in several states, including WA. Consequently, it should be regarded as a mosquito of public health importance, particularly in urban areas into which RRV or BFV may be introduced from rural areas.

CULEX ANNULIROSTRIS: THE COMMON BANDED MOSQUITO

Cx. annulirostris is the most common and widely distributed pest species in Australia.

It is a major vector of arboviral disease in Australia. It is the principal vector of Murray Valley encephalitis and Kunjin viruses and an important vector of RRV and BFV in many regions.



OCCURRENCE

In northern Australia, *Cx. annulirostris* can breed year-round, but is most abundant in the midlate wet season and first months of the dry season. Breeding of *Cx. annulirostris* generally occurs when the mean temperature is above 17.5° C. Therefore, it is most abundant in summer in southern areas of Australia. It may 'over-winter' in southern areas as adults, resting in various harbourage sites.

Cx. annulirostris is mainly associated with natural freshwater habitats but also breeds in agricultural, container and polluted water (effluent) freshwater environments, and occasionally in mildly brackish water. Breeding is typically associated with shallow, open sunlit sites with emergent or fringing vegetation.

It is found in reed swamps, temporary flooded grasslands with organic matter in sub-coastal and inland areas and sewage effluent and organic wastewater. It will colonise some domestic containers and breed readily in semi-polluted water in storm drains. It will also colonise poorly maintained irrigation channels and ditches and other habitats associated with irrigation for agriculture.

Cx. annulirostris will breed in abundance in artificial wetlands such as those constructed as compensation basins or for stripping nutrients from residential stormwater runoff.

BIOLOGY/BEHAVIOUR AND IMPLICATIONS FOR CONTROL

Breeding habitat

Proximity to freshwater breeding sites should be considered when assessing the likelihood of problems caused by this species. Communities situated more than 10-15 km from such sites are unlikely to experience a major problem with *Cx. annulirostris.*

Breeding in natural habitats is often widespread and difficult to control due to scale of the problem and inaccessibility of sites. As with *Ae. vigilax*, small communities may have no alternative but to resort to the use of adulticiding (barrier fogging, residual surface sprays) in the event of an outbreak of disease. It may be possible and is desirable to treat natural habitats that are in very close proximity to residential areas with an appropriate larvicide.

The capacity for dispersal of *Cx. annulirostris* is less than for *Ae. vigilax*, but it may still travel several kilometres from breeding areas. Dense vegetation can assist dispersal by providing harbourage for the adults. Buffer zones in which vegetation has been cleared or reduced will help to reduce the potential for adults to disperse into residential areas.

Some natural breeding sites of *Cx. annulirostris* are environmentally significant wetlands. It may not be appropriate to undertake chemical or physical control of mosquitoes in such habitats (refer to the chapter on environmental considerations in mosquito management by Ms Susan Harrington on page 100 for more details).

Appropriate planning is required to ensure that new residential developments are not located within the dispersal zone of *Cx. annulirostris*. As with *Ae. vigilax*, the potential public health and mosquito control problems that can be experienced with *Cx. annulirostris* should be brought to the attention of urban planners.

Furthermore, engineers, planners, landscape architects, environmental consultants and environmental agencies should be informed about requirements for design and maintenance of water treatment and management facilities. If possible, sewage lagoons and constructed wetlands should be located well away from residential areas. Agricultural and irrigation projects or wastewater and stormwater management infrastructure should be designed and maintained in a manner that will not exacerbate breeding of *Cx. annulirostris*.

Source reduction is the method of choice for control of *Cx. annulirostris* in man-made situations such as stormwater sumps, drains, gullies, vegetated sewage ponds/outfall areas and irrigation ditches and channels. Most breeding in these habitats can be prevented with appropriate design, good water management practices and removal of emergent or fringing vegetation. This type of control is discussed in detail in the chapters on physical (page 72) and cultural (page 93) control of mosquitoes by Mr Peter Whelan. Repeated use of chemicals should not be regarded as an acceptable alternative to source minimisation or reduction for control of *Cx. annulirostris* breeding in man-made situations. Indeed, insecticide use should be minimised where possible to reduce environmental impacts and potential for development of resistance. Ultimately, source reduction will also be far less expensive than recurrent application of chemicals.

Cx. annulirostris can breed throughout the year in poorly maintained sewage lagoons, irrigation channels, etc in northern Australia. This creates a public health risk and enhances colonisation of nearby seasonal natural habitat at the start of the next wet season. Consequently, the potential for breeding of *Cx. annulirostris* in such sites should be eliminated.

There is an increasing trend towards the use of artificial (constructed/engineered) wetlands for removing nutrients from stormwater or as compensation basins for stormwater management. These must be carefully designed and maintained to prevent high levels of breeding of Cx. *annulirostris* (and other potential vector species). Professor Richard Russell deals this with in detail in the chapter on constructed wetlands in Australia on page 198.

Egg/larval biology and behaviour

The eggs of *Cx. annulirostris* are not desiccation-resistant. Instead, they are laid in rafts directly on the water surface and the eggs hatch within a few days. Breeding occurs in more or less continuous cycles, generally in sites that are permanent waterbodies. However, *Cx. annulirostris* can also colonise newly formed freshwater sites quite rapidly. Breeding is not synchronous like *Aedes* species and may not commence as reliably after a specific environmental event. Therefore, regular monitoring is required to determine whether a breeding site has been recently colonised by this species.

Larvae of *Cx. annulirostris* are frequently associated with dense fringing or emergent vegetation. Thus, care is required when monitoring to ensure that they are not overlooked, particularly as early instar larvae. Effective application of larvicides in densely vegetated sites can also be difficult. This is another incentive to keep potential man-made breeding sites free of vegetation.

Breeding of *Cx. annulirostris* is temperature restricted. The time of year should therefore be taken into account in southern areas of its range. For example, there would be no requirement for control of *Cx. annulirostris* before the mean daily temperature has risen above 17.5° C in southern areas of Australia.

Adult biology and behaviour

Female *Cx. annulirostris* feed on a range of mammals including humans. They feed mainly at dusk or after dark. Therefore, adulticiding should be carried out at or after dusk to ensure maximum contact between the insecticide and blood-seeking adults.

Cx. annulirostris is a 'less vicious' biter than *Ae. vigilax* and CO_2 -baited trap counts of around 100 indicate a current pest problem. People with protected, screened dwellings will not experience a problem with *Cx. annulirostris* unless they venture outside after sunset. Personal repellents, protective clothing and well maintained flyscreens on tents and caravans should be recommended for people camping in areas where this species is abundant.

Sentinel chickens are an effective method of monitoring flaviviruses (such as MVE virus) vectored by *Cx. annulirostris* as this species feeds on birds as well as mammals. *Cx. annulirostris* is a very competent vector of several important mosquito-borne viral diseases in Australia, including the potentially fatal Murray Valley encephalitis virus. Mosquito control and public warning and education programs are necessary for communities affected by this species.

SUMMARY

Differences in the biology and behaviour of different mosquito species will determine the approach to controlling them or minimising their contact with people. Available information about problem species in a region should be used to assist with defining breeding sites and to develop the most effective control program possible with available resources. The control program will need to integrate source reduction and public education and warnings with strategic use of larvicides and/or adulticides.

Ongoing monitoring of breeding and adult activity is essential. Such monitoring determines the method(s) and timing for control measures. Over time, it will produce an understanding of environmental conditions or human activities that can lead to breeding. The evaluation of control efforts is also critical to improving long-term mosquito control strategies.

SUGGESTED READING

- 1. Lee, D.J. *et al.*, (1980-1989) The Culicidae of the Australasian region. Volumes 1-12. Australian Government Publishing Service, Canberra, Australia.
- 2. Liehne, P.F.S. (1991) An atlas of the mosquitoes of Western Australia. ISBN 0 7309 4635 5, published by the Health Department of Western Australia.
- **3.** Russell, R.C. (1993) Mosquitoes and mosquito-borne disease in Southeastern Australia. Department of Medical Entomology, Westmead Hospital, Westmead, NSW and Department of Medicine, University of Sydney, NSW. ISBN 1 875398 38 4.
- 4. Mosquito Control Association of Australia Inc. (2008) Australian Mosquito Control Manual. ISBN 0-646 35310-1.

ACKNOWLEDGEMENTS

The photographs of *Ae. vigilax* and *Cx. annulirostris* were obtained from the website of the Department of Medical Entomology, University of Sydney and Westmead Hospital, Australia: <u>http://medent.usyd.edu.au/</u>

Information on flight range and CO2 trap pest thresholds for some species was obtained from Whelan, P.I. (1997) "Problem mosquito species in the Top End of the NT, pest and vector status, habitats and breeding sites" Medical Entomology Branch, Northern Territory Department of Health and Community Services.

7. THEORY OF MOSQUITO CONTROL

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Before initiating or considering the initiation of a mosquito control programme there are a number of questions that should be addressed in order to ensure an effectual, economical and environmentally acceptable programme.

WHY is a mosquito control programme being contemplated? Is it for the prevention of disease amongst the human or animal populations, or is it because the 'quality of life' of the human community is affected by the biting of pest mosquitoes as either a seasonal occurrence or as a year-round problem?

Either of the above reasons may justify the implementation of a control programme, but each will involve considerations and decisions on issues related to social, economic, environmental and political implications before, during and after the course of the programme.

WHEN will the control programme begin and when will it be terminated? With an area troubled with pest mosquitoes the answer simply may be that control should be undertaken and continued for as long as the pest poses a problem.

However, if the control of a mosquito-borne disease is the primary concern, the signals indicating the requirement for initiating or terminating control procedures may not be so obvious. The appearance of human clinical disease will almost certainly signal the need to start control operations, but an 'early warning system' that reduced the risk of human disease would be a great advantage. This is not an easy problem to overcome, but surveillance of pathogen activity in sentinel animals and/or vector mosquitoes, and the monitoring of vector population abundance may provide such a pre-epidemic signal.

With respect to an anticipated outbreak of disease, there are options for either 'pre-emptive' or 'reactive' control action. There may be strong arguments for control to begin early in the 'season of activity' of either the target species or the pathogen, or it may be more appropriate to wait until there are definite indications that transmission of a disease pathogen is imminent or has already begun; the nature and likely severity of the disease in question will also influence decisions.

The effects of the control operations on non-target species and on the environment in general, and with regard to particular factors such as the development of insecticide resistance in the target species, will need to be considered. Such implications are particularly relevant to both long term programmes and intensive short term programmes.

WHERE will the control operations be directed? Should there be an attempt at widespread eradication of the target species from the locality (area, region, state, country) under threat, or could the protection of the human population at risk be more effective by concentrating on a more pragmatic and economical restricted control approach?

There is little to be said in defence of the 'eradication' approach and much to be said for a 'restricted control' proposal. Except in a few special circumstances, the eradication of a nondomestic mosquito species should be considered untenable, and it is generally preferable to consider a local programme of mosquito population management directed at those species that are most likely to contact the human population at risk and that is requiring protection. A concept of '**barrier control**', the creation of a '**buffer zone**' around the community at risk, is often advocated as this approach can provide economies in deployment of funds for manpower, equipment and supplies, and yet can be quite effective in protecting the human population. The effectiveness of this concept depends on intensive and efficient mosquito control within an area surrounding the human community with a view to separating the community from the mosquitoes emanating from outside the buffer zone. The success of the technique depends very much on adequate knowledge of the habits and habitats of the target mosquito species and particularly the range of dispersal of that species from its larval habitats outside the 'buffer zone'.

In any situation where disease is being transmitted, the prime objective is the interruption of transmission as soon as possible, and this 'barrier' method can result in a break in contact between humans and vectors, and the transmission of the pathogen can be interrupted effectively and economically.

HOW will the mosquito programme be devised, organized and supervised? The methodologies and technologies available for incorporation in modern mosquito control programmes are discussed elsewhere in this manual, but the important considerations relate to the procedures of planning, implementation and evaluation. The choice of control technique/s will involve considerations of availability, economy and efficacy, with particular tailoring of the recommendations to the specific circumstances of the particular target species and the local area.

In an attempt to pre-empt disease transmission, a control programme may be mounted early in the vector season and directed primarily against the vector mosquito's larval populations to retard the build-up in adult populations for the time that the pathogen is expected to become active. Control programmes directed against a vector species during an outbreak of disease, when the pathogen is already being transmitted, may be principally directed against the adult mosquito population, in an attempt to reduce the numbers of those mosquitoes already infected with the pathogen and likely to be involved in transmission within the immediate future.

It should be remembered that the larval control programmes will not have an immediate effect on adult population numbers, and are generally not appropriate for interruption of disease transmission in epidemic circumstances, however larval control should be undertaken in conjunction with adult control during an epidemic (if at all possible) to inhibit replenishment of adults if the pathogen is still active in vertebrate reservoirs or hosts.

Although adult control programmes reduce the overall age (which is most important in the dynamics of transmission) of the vector population, larval control programmes can actually increase the age of the adult population (by reducing the input of younger individuals) and thus create an 'older' and, from a disease perspective, a more dangerous vector population.

THEREFORE, before beginning a control programme we must be able to define the problem as accurately as possible, formulate objectives that are practicable, select the appropriate control measures carefully, know how we can monitor the effectiveness of the operations during the programme, and establish procedures for an evaluation of the control operations at the completion of the programme. The information required for these components of the overall task is obtained with **surveys**.

Surveys can be used to:

- (i) determine the need for a control programme
- (ii) guide the planning of the programme
- (iii) monitor the operation of the programme
- (iv) evaluate the programme

and can be categorized as follows:

1. Initial Survey

The object is to define the nature and extent of the problem; the survey should indicate whether a control programme is necessary, and if so, its extent and points of concentration. This survey can be used to determine:

- the species of mosquito present
- source/s (breeding sites)
- relative densities
- dispersal or flight range

With continuing surveillance it is possible to gather data on:

- life cycle of the various species
- host feeding preferences, times and places
- resting sites
- seasonal fluctuations
- susceptibility to candidate insecticides

Such investigations provide important baseline data for later comparisons, particularly the density indices (both adult and larval) and the susceptibility to insecticides in case of suspected resistance. Unless baseline data is collected prior to the commencement of a control programme, proper evaluation at a later stage is hindered or even precluded.

2. Operations Survey

The object is to monitor the effectiveness of the control programme; the survey should

- provide information on the current status of the population of the target species, and can be used to monitor pathogen infection rates in vectors
- be used to guide operations and modify procedures if control is found to be unsatisfactory.

3. Evaluation Survey

The object is to assess the effectiveness of the programme; the survey should

- evaluate the costs (and consider the benefits) of the programme
- estimate the effective reduction in mosquito populations
- identify any residual problems
- provide information useful in maintaining or modifying the routine operations for future programmes.

Methods and techniques for conducting these surveys will be presented and discussed elsewhere.

8. MOSQUITO SURVEILLANCE AND MONITORING TECHNIQUES

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1.0 INTRODUCTION

Mosquito surveillance is the process of providing information on aspects of mosquito populations by carrying out surveys. It is a vital part of any mosquito control program and should be started before any direct control begins. The underlying aim of carrying out any surveillance program is to determine the what, where, when and why of any possible mosquito problem. When it has been established that a control program is necessary, the ongoing surveillance program will assess whether the mosquito populations are being reduced and, more importantly, if the control program is achieving reductions in pest problems or mosquito borne disease. The results of the mosquito surveys are used to;

- Determine the need for a mosquito control program.
- Plan the program by providing adequate information to allow decisions on the type and extent of control.
- Guide the day to day activities of the program.
- Permit evaluation of the effectiveness of the control program.

Mosquito surveillance can be separated into four phases corresponding to the four periods of a control program. These stages are the preliminary phase, the base line data phase, the operational phase and the evaluation phase. The object and the details of mosquito surveys are different for the different stages of a control program.

2.0 THE PRELIMINARY PHASE

The object of the preliminary phase is to define the nature and extent of the mosquito problem. This will indicate whether a control program is necessary and if so, the extent of the area to be controlled and particular areas of priority. The preliminary phase should be completed before the base line data phase is put into operation, although sampling during the preliminary phase can provide some of the base line information. The preliminary phase should incorporate three elements that include an information search, drawing vector control maps and initial sampling of adult and larval mosquitoes.

2.1 INFORMATION SEARCH

The first step in the initial survey is to gather together all the relevant files, literature and references that will be relevant to a mosquito control program in a particular area. All the information should be organised and accessible for ready reference.

Contacts should be established with Local, State and Federal Authorities for information and help. The information needed should include:

- a. What species of mosquitoes are potential problems in your particular area? Is it a pest problem or a potential disease problem?
- b. When do the mosquito problems occur? Is it a seasonal problem and what is a likely reason for seasonality? What data is available from medical records and reports on previous mosquito borne disease outbreaks, including seasonal distribution and the spatial distribution of cases?
- c. What information and equipment is needed to carry out mosquito surveys and control operations in your area?
- d. How can the mosquitoes be identified?
- e. What is the relevant biological information on the problem species, including habitat preferences and seasonal abundance?
- f. What are the local climatic/environmental variables that may affect local mosquito populations?
- g. What is the distribution of the major mosquito habitats?

h. Is there any mosquito collection data from past records?

2.2 VECTOR CONTROL MAPS

The next step is to draw up a preliminary vector control map. This map should be updated as the preliminary surveys progress. The vector control map should show all the relevant details of the nature and extent of the mosquito problem and enable the planning of mosquito surveys and future control operations.

Vector control maps usually relate to a town and should extend at least 2 km and ideally up to 10 km from the urban boundary. All maps of the local area for at least 10 kilometres around the town should be assembled, including vegetation maps, topographical maps and maps of built-up areas. These should be incorporated onto one map. The map should show the location and nature of actual and potential mosquito breeding sites, such as rivers, creeks, lakes, dams, marshes, storm water drains, borrow pits, depressions, sewage ponds, mangroves and dense forest. Maps can be hard copies or electronic interactive maps. Examples of a stylised vector control map is shown in Figure 1.

Incorporated on the map should be the residential areas, night recreation areas, roads, railways, access routes, industrial plants, concentration of animals, areas of pollution, and tidal influenced areas. Google earth images and aerial photographs are of particular value in an initial survey, so that vegetation zones can be used as a guide to mosquito habitats and access to particular habitats can be planned. This map can be updated by additional information as it becomes available and by a reconnaissance survey, to verify the accuracy of the details.

Marked on the vector control map should be a buffer zone around urban areas based on the flight range of the most important species of mosquitoes in that area.

Generally the buffer zone will be in the order of 1.6 to 4 kilometres from the perimeter of the urban residential development. The buffer zone should expand at breeding sites or points of dense vegetation which are continuous with areas inside the buffer zone. Most of the mosquito monitoring and control activities should be carried out within the buffer zone unless the initial monitoring indicates a need to expand the area of operation.

2.3 PRELIMINARY SAMPLING

For vector control operations, the initial larval and adult sampling sites are determined from the vector control map. The various methods and detailed procedures for larval and adult sampling are dealt with in Section 6.0.

When a complete picture of the mosquito fauna of an area is required, every available type of breeding sites including crab holes, plant axils, receptacles, creeks, swamps, water filled depressions and any other water accumulation should be considered and examined for mosquito breeding. However for well established pest and disease problems, it is only necessary to examine and sample the potential breeding sites of the principal problem species.

With preliminary surveys, there is no substitute for a lot of legwork, and a determined effort to penetrate into all vegetation zones or areas of likely mosquito breeding. The golden rule of larval surveys is to sample around the entire boundary of any potential mosquito breeding area. This is very important, at least initially, to determine if a particular habitat or part of a habitat is more prolific than others, and to find the source or sink of the water in the breeding site. The initial larval sampling should be carried out in the likely mosquito problem period, and be completed in a few weeks, with initial adult sampling being conducted at the same time to detect possible undetected sources.

During preliminary sampling, consideration should be given to the selection of suitable permanent sampling stations for larvae and adult mosquitoes. This choice will be based on a quantitative assessment of the breeding sites and on the relative numbers of adults or larvae present.

The preliminary adult sample sites should be many as possible near all the probable breeding sites located during the initial larval survey and at relevant points in and around the buffer area as determined from the information on the vector control map. The number of trap sites can then be reduced down to a manageable number for routine sampling. The most productive and the most informative trap sites are chosen for routine sample sites. Once the routine adult sample sites are chosen, usually within the first two months, these should not be changed, so that base line information can be gathered to allow accurate assessment of changes over the season and years, and to asses control measures.

The initial survey must note the particular habitats where mosquitoes are found, so that knowledge of the preferred habitat for each species can be compiled. Note that mosquito populations can change dramatically in a few weeks, both in size and species composition. These changes can occur with variations in tides, rainfall and vegetation, or due to other factors that are less obvious.

3.0 BASE LINE DATA PHASE

During the base line data phase the permanent larval and adult sampling situations selected during the preliminary phase are regularly sampled. All climatic data is collected and organised, including information on rainfall, tides, temperature and any other climatic variable that is likely to affect mosquito populations. During this phase there will be dynamic changes occurring in the mosquito habitats and all of these changes should be noted and correlated with the larval and adult sampling. Base line data on mosquito complaints and details of mosquito borne disease should also be compiled for later comparison.

This phase should last at least 12 months to cover the major habitat changes and seasonal variations. It is during this phase that plans for a control strategy should be formulated. Strategies for disease control or vector control should be examined so that an integrated control program can be drawn up.

4.0 **OPERATIONS PHASE**

When a mosquito control program is implemented, it needs to be guided by regular larval and adult sampling of the selected sample points of the area being controlled. Sampling should be from the same points and with the same regularity as the base line data phase. These regular and ongoing surveys will indicate the current status of adult and larval populations in the control area. Changes in habitats can occur seasonally or with artificial influences and give rise to mosquito population fluctuations. Usually the adult sampling program or the larval sampling will show the response to the changes.

Sometimes there will be public complaints that may not tie in with the sampling data and require additional or supplementary larval or adult sampling. At other times, the adult sampling data will show increases that are not obvious in the larval sampling data. Additional larval surveys are then required to locate any additional mosquito breeding or to determine the reasons for the increased adult levels.

The regular operations survey should allow areas of mosquito breeding to be defined and quantitatively assessed so that ongoing priorities for control can be decided.

5.0 EVALUATION PHASE

After control measures have been carried out, it is most important to assess their effectiveness and to identify any remaining problems.

5.1 LARVAL EVALUATION

Evaluation of larval insecticide control operations should be done on the day or the day after control, with the results compared with a pre-control survey. Areas that have been missed can be re-treated and any operational and technical difficulties should be reviewed and rectified.

5.2 ADULT EVALUATION

Evaluation of larval control includes the comparison of adult population indexes before and after larval control. Evaluation of larval control using adult mosquito information is generally imprecise because the adult population will take some time to decrease, and there may be dispersal into control areas from other areas. In assessing the efficiency of adult control programs, it may be necessary to carry out age determination assessments on the sampled females to determine whether emergence or re-invasion has occurred.

5.3 ENGINEERING EVALUATION

The assessment of engineering measures such as draining and filling should include a comparison of twelve months sampling data before and after the completion of the engineering measures. The evaluation of engineering measures will usually require a short and longer term assessment to cope with possible gradual habitat changes in the years after engineering measures.

5.4 DISEASE EVALUATION

Disease evaluation will reflect the real benefits of the mosquito control program. Different parameters of disease can be compared with the base line data after control measures have been completed. If disease parameters are not decreasing, the original hypothesis and vector control strategies need to be critically evaluated.

6.0 MOSQUITO SAMPLING TECHNIQUES

6.1 LARVAL SURVEYS

6.1.1 General

The purpose of larval surveys is to find out where and when the mosquitoes are breeding and what type of habitat they are breeding in. Generally we can divide the sampling procedures into the sampling of ground water habitats and artificial receptacle sampling. Most mosquito control operations are concerned with ground water habitats, while receptacle sampling is a more specialised survey to gain information on receptacle breeding *Aedes* species.

6.1.2 Ground Water Habitats

For field sampling of ground water habitats, it is important to traverse the entire margins of the breeding site to determine the entry or exit points and possible source of the water. Permanent indicator sample points that represent the habitat should be chosen after extensive initial sampling. Sampling should then be quantitative so that the relative importance of all the breeding sites or habitats can be assessed.

In an extensive breeding site, this may include a point in each vegetation or water type known to be a breeding site. These permanent points should be at sites where there is year round

access. These permanent larval sampling sites should be sampled at least once per month over the twelve month period, using the same sampling techniques, and recording all the important variables as shown on the larval collection form. For tidally affected areas, these sampling frequencies will have to be coordinated with the tides. Increased frequency of sampling is necessary for transition periods between times of little mosquito activity and times of increased activity, such as the start of summer or the start of the wet season.

All sample sites should be marked on the vector control map. Additional larval samples should be taken at different points throughout the year to make sure the permanent points are efficient indicators of larval breeding sites.

The sampler must be careful not to change the nature of the sample site itself by repeated sampling. These permanent larval sampling sites will enable an assessment of how the breeding habitat and the species and numbers of mosquito larvae changes over a 12 month period. It will pin-point those important factors in the habitat that lead to fluctuations in mosquito numbers.

6.1.3 Artificial Receptacle Sampling

Artificial receptacle sampling is primarily to detect the presence of *Aedes aegypti* or other related *Aedes* species in an area, or to determine the receptivity of an area for *Aedes* species introduction. The presence or receptivity is assessed by recording all the available information on the number, type, and characteristics of various receptacles in each particular area. The detection of these *Aedes* species can be assisted by adult sampling techniques, but usually the specialised techniques such as ovitraps or receptacle surveys are required.

Ovitrap sampling is a specialised sampling technique using special egg laying substrates dipping into water in a dark coloured receptacle. The ovitraps are usually left out for a week and then the paddles or ovistrip are inspected for eggs. The eggs can be hatched, and either the larvae or the emerged adults are examined for species determination.

An artificial receptacle breeding survey is carried out to determine all the relevant particulars of receptacle breeding mosquitoes in a certain area. The primary aim of a receptacle survey is to examine artificial receptacles, but natural receptacles are also examined.

The priorities for receptacle sampling can be guided by the initial ovitrap data. Detailed receptacle surveys can then be carried out in certain suburbs or limited areas.

Aedes receptacle surveys record all the relevant receptacle information for each premise. If the number of premises is very large, as in a large town, a sample of premises can be made by selecting a number of streets at random and then sampling each property in that street. If the town is small, as many premises as possible should be sampled for receptacles.

For exotic *Aedes* species, particular attention should be given around airports, seaports, boat berthing facilities, dry dumps and industrial areas.

6.1.4 Equipment for Larval Sampling

- a. An enamel dipper painted white on the inside with a relatively long handle. A deep soup ladle is ideal. A long piece of wood can be attached to the dipper for difficult to reach situations (see Figure 9).
- b. White enamel pans, trays or pale coloured buckets. These have the advantage that more water can be sampled at one time when larval populations are not particularly concentrated. They are not convenient in shallow water or where there is a lot of vegetation obstruction.

- c. A pipette or dropper. Ensure that the tip of the pipette is wide enough to allow large larvae and pupae to be sucked up.
- d. Small stoppered or topped vials. These can be glass or plastic. Mosquito larvae can be collected live into these small vials or into vials of 70% alcohol (ethanol) or 70% methylated spirits.
- e. Note book, labels and pens. Labels are best written in pencil and placed inside the collection receptacle with larvae.
- f. A bulb pipette. This is a large bulb with flexible tubing that can be utilised for sucking water plus larvae out of crab or tree holes or plant axils.
- g. A pale bucket, a long piece of rope and a powerful torch for examining wells and rainwater tanks.
- h. If you have an inaccessible area, a 4 wheel drive vehicle is an advantage. An all terrain vehicle such as the Argo is extremely useful in large swamp situations.
- i. Suitable clothing, such as hat, overalls, rubber boots and carry bags.

6.1.5 Procedures for Ground Water Habitat Sampling

Mosquito larvae are usually found where surface vegetation or debris are present. In larger bodies of water, larvae are generally confined to marginal areas or floating surface materials.

- a. Before looking for larvae, examine the data of the adult catches to see what species you are looking for. This will give you an idea of the preferred habitats of your species, with its water type and vegetation requirements.
- b. Examine the vector control map and aerial photographs for vegetation patterns and likely areas of mosquito breeding. Plan the access route and plan your specific search sites. If a large area is of uniform vegetation then examination at selected points only can reduce the amount of work.
- c. When searching for mosquito larvae, the searcher must be prepared to walk and to penetrate through thick vegetation into the selected points chosen on the aerial photographs. There is no substitute for legwork and perseverance.
- d. When approaching a margin of a water body, it is important to note the vegetation patterns. The different types and habit of grass, reeds, or other vegetation may be clues to deciding exactly where the mosquitoes are likely to be and which habitats must be sampled.
- e. When you have selected particular habitats look at the water first before disturbing it with the feet, the ladle or by shadows. Note the presence of fish and other predators and look for larval activity.
- f. Use the dipper at likely places. If looking for *Anopheles* larvae let the top layer of water run into the dipper or skim the top layer of water. Very shallow water at the extreme edges or the water on the top of floating algae is also a source of *Anopheles* larvae. With *Culex* larvae the dipper sample will need to be deeper and next to clumps of vegetation or grass. When sampling for larvae, proceed carefully, as disturbance and shadows cause larvae to go to the bottom. Let water run into the dipper from vegetation clumps and scoop the dipper up just before it fills up with water. With *Aedes* larvae you need a quicker motion as they will dive rapidly to the bottom, (see O'Malley 1995).
- g. Record the number of dips made. Usually these are in multiples of ten, with dips being made only in likely places (after you have established the types of habitat where the larvae are).

- h. Transfer the larvae to the vial with the aid of the pipette. Take a water sample in another vial for salinity and pH examination.
- i. Record all habitat information on a notebook or form with a code label inside the vial. The degree of pollution, vegetation, degree of shade, water colour, possible salinity and predators present should be noted.
- j. Note the larval instars present and relative proportion of each instar. *Culex* species eggs should also be searched for, especially at the side of pools or where scum has been blown by the wind. Note all larval information on collection forms. Set forms for direct larval recording are more convenient for information gathering, identification records and later compilation. An example of a set larval form is shown in Figure 3.
- k. Surveys of larvae should be made at least once per month, with increased frequency during the breeding season or major habitat changes, to establish the time required between surveys.
- I. For mosquito species such as *Mansonia and Coquillettidia*, larvae may be found by pulling up aquatic and semi aquatic plants and washing them in a pale coloured bucket to dislodge them from the vegetation, or pumping out water from a bottomless isolation drum placed over emergent vegetation and pouring all the sampled water though a fine sieve.
- m. Sometimes there may be a need to muddy up pools and sit and wait for larvae to rise to the surface.
- n. Some species, such as tree hole breeders or crab hole breeders require sampling with a piece of flexible tubing or more specialised equipment. Trees such as figs and *Poinciana trees* that have areas between the main branches capable of holding water are productive sites. Other trees such as boabs, mangroves and eucalypts can have hollow broken branches with water inside the hollow. These sites need to be looked at a few days after rain.
- o. With salt marsh mosquitoes, it is important to time the search for larvae two to three days after the highest tides of the month or rain.
- p. For fresh flood water *Aedes*, inspection of rain filled depressions is needed two to three days after rain.

6.1.6 Ovitraps and Procedures for Aedes Ovitrap Sampling

Ovitraps or egg traps are special traps used for detecting the presence of *Aedes* receptacle breeding mosquitoes. There are a number of types of ovitraps, using various materials as the receptacle and various substrates used for egg laying.

The types range from sticky ovitraps for adult sampling as the mosquitoes lay eggs, lethal ovitraps which aim to impart insecticide to adults as they lay eggs, and general ovitraps which sample eggs.

General ovitraps use glass jars or plastic buckets as receptacles. Generally glass jars have the advantage of being clear to allow ease of inspection of the sides and bottom for eggs or larvae. The glass is unsuited to egg laying, so very few eggs are attached to the sides, reducing the loss of eggs for analysis.

The disadvantage of glass is the need for painting or encasing black protectors for a dark background to prevent breakages. Other disadvantages are the fragility, weight and additional space required in transportation.

Plastic buckets are generally light, stackable, cheap, and do not need painting or enclosure in a black cover. Plastic buckets can be black but red is seen by mosquitoes as dark and allows

ease of inspection of scum and eggs on the sides. The disadvantage of plastic is that *Aedes* tend to lay eggs on the sides, as well as the paddle or ovistrip.

The types of paddles or ovi-strips include 'Masonite' paddles, red velour strips, or red painted tongue depressors. Generally 'Masonite' paddles have the advantage of roughness, grooves and a wide wet wick zone favouring egg laying. Red velour has the advantage of a wide coverage of the receptacle, good wick area and ease of observation for counting eggs. It can also be impregnated with an insecticide to act as a lethal ovitrap to kill adults as they lay. Tongue depressors need to be roughened and painted red to improve suitability for egg laying.

Standard ovitrap procedure

- a. A standard ovitrap consists of a black receptacle with a 'Masonite' paddle attached to the inside of the receptacle in a vertical position, with the rough side facing inwards. The paddle has a white painted line half way up the paddle as a water level indicator.
- b. Ovitrap sites should be secluded, shaded, low to the ground, near vegetation and protected from rain and animal disturbance. If possible the ovitraps can be placed between two bricks or stones, or behind or under a suitable object such as a wash trough.
- c. The ovitrap should be placed near or within fifty (50) metres of human habitation.
- d. Water is added to the ovitrap to the white line on the paddle or receptacle. The water should be fresh and can be either tap water left to stand for a few hours or rainwater. Ovitraps should also have a food source for the larvae, with aged grass infusion water or fish flakes ground fine to provide suitable food for both young and older larvae. Ovitraps for Quarantine purposes should have a methoprene pellet added each month to prevent any possible adult escapees.
- e. The ovitrap should be numbered, and the number, town, location, the date placed, and the date retrieved should all be recorded in a record book or an information system.
- f. The ovitrap should be left for a week and then reinspected.
- g. On inspection, it should be noted in the record whether the trap is tipped over, dry or otherwise disturbed by ants, frogs or polluted.
- h. If there is any remaining water in the trap, pour it into a clear glass jar and inspect if for mosquito larvae. Larvae are collected alive into a small receptacle for rearing or identification.
- i. If there are only fourth instar larvae present, then all the larvae can be put into a vial of 70% alcohol and labelled with an identification number.
- j. If there are any younger larvae they should be reared to fourth instar, together with larvae reared from the eggs on the paddles. The identification number should correlate with information in the record, including the ovitrap number and the location. The labelled vials should be filled to the top with alcohol and stored adequately to prevent breakage until they are submitted to specialists for identification, along with the records or form.
- k. The paddles with attached eggs are left in a plastic bag for one day and allowed to dry out gradually. They are then placed in fresh water in trays, together with any young larvae from the ovitrap collection in step h. The eggs can then hatch and the larvae are reared to fourth instar for identification. Hatched paddles should be inspected for unhatched eggs under the microscope and then dried and re-flooded if necessary. The paddles are then sterilised and scrubbed to remove any old hatched eggs.
- I. The replacement ovitrap is refilled, with a new paddle, fresh water, and the ovitrap replaced for later inspection. If the ovitrap cannot be inspected within one week, the

ovitrap should be collected and replaced with a fresh ovitrap or left in position until dry and then collected for inspection. The ovitrap and paddle should both be identically labelled and attached together.

m. For regular ovitrap monitoring purposes, all ovitraps should be collected every six months, and taken to the laboratory where they should be filled completely with fresh water to hatch all eggs and examined after one week. Then the ovitraps should be thoroughly cleaned with boiling water and scrubbed to remove any old eggs, and then repositioned.

6.1.7 Procedures for Receptacle Sampling

- a. If possible, contact the person responsible for the property and inform them of your intentions and reason.
- b. Examine the entire premises, both indoors and outdoors for any receptacles, and note the type of receptacle and the presence of water or larvae in each receptacle.
- c. Each receptacle should be sampled for larvae.
- d. The first step in sampling larvae is to look carefully at the surface of the water for larvae or pupae.
- e. With a ladle, gently lower the ladle deep into the receptacle so that larvae can be seen against the white background of the ladle. The ladle can then be slowly extracted with the larvae and water.
- f. The receptacle can then be emptied carefully into a white bucket or tray for further examination.
- g. Tyres can be prised open and the inside water can be scooped with a ladle, ensuring that the ladle makes contact with the bottom of the tyre.
- h. In rainwater tanks, the bucket and a length of rope can be used, with the bucket lowered down into the water to provide a pale background for detecting larvae. The torch should be used to examine for the presence of larvae. Water should also be extracted from the tap into a bucket and examined.
- i. Blocked roof gutters can be checked for breeding by using a ladder or a long stick with a mirror attached to one end. A sure sign of a blocked gutter is a dripping down pipe a few days after the last rain.
- j. The axils of trees and other plants with leaf axils such as bromeliads and lillies can be sampled with a bulb pipette.
- k. All the larvae found should be collected and put in the vials with an identification number. The identification number should correspond to notebook information on the town, street, premises number, date, type and water characteristics of the receptacle. The vials should be filled with 70% alcohol. If there are many larvae, all stages of larvae should be collected, with different looking larvae included.
- I. If there are only first and second instar larvae, they should be collected into a clean receptacle with original water, labelled, and reared to a later instar.
- m. The larvae can be reared by taking the sample in a sealed receptacle back to a base, loosening the top of the receptacle, and leaving the receptacle in a cool shaded place until the larvae develop.
- n. For overseas arrival boats, and in other areas where exotic *Aedes are* suspected, receptacles can be sampled by looking for the presence of eggs. Scrapings can be made from just above where the water line would have been in the receptacle. The scrapings can be made by a paint scraper or a chisel and collected into a receptacle.

The scrapings are put into a labelled sterile receptacle for later microscope examination or rearing of the eggs.

- o. If a return trip to an area is anticipated in three to four days, fresh water can be added to any dry receptacles or tree hole axils where *Aedes* eggs are suspected, and the water can be collected after a few days for examination.
- p. The labelled larval samples, together with records should be submitted to recognised experts for species identification or confirmation.

6.2 ADULT SURVEYS

6.2.1 General

The purpose of adult sampling is to get an indication of a mosquito species presence or population fluctuations in time or space. For regular adult sampling during control programs, the method chosen may not necessarily be the method that catches the most number of mosquitoes, as long as it accurately reflects population changes.

Once-off adult sampling rather than regular sampling from the same trap sites is of limited value in a mosquito control program, but is useful to determine the presence of various species in initial surveys or to locate particular areas of high mosquito activity. Once-off surveys need to be timed during potentially productive periods and include as many methods as possible to increase the accuracy of species records.

There are a number of ways to collect adult mosquitoes, and the method used will depend upon the aim in making the collection. All of the adult collection methods have a bias that will be more or less applicable for certain species. Direct quantitative comparisons can not be made between species from different trap type collections unless the bias of each trap is known. If there is little information on the mosquito fauna in an area, it is best to use as many different methods as possible to ensure that you collect as many species as possible.

For routine monitoring purposes, a single method is usually most convenient. Once a method is chosen, it is important that the method and equipment is standardised, so that comparisons actually reflect the variations in the mosquito population and are not the result of a change of trap type or location. The types of adult trapping methods are outlined below. Examples of adult collection record forms are shown in Table 3 and Table 4.

6.2.2 Human Biting Collection

These collections are usually the simplest and most direct way to sample a mosquito problem. It involves collecting the mosquitoes that are about to bite a person. If you wish to find out which mosquitoes are causing a problem for people, the obvious and most accurate method is to carry out a human biting collection. The collection site must be selected out of the wind, near vegetation, in the area to be assessed. If this is in a town or near a residence it should not be in a lit area or under street lights.

Most biting collections are carried out at night, just after sundown for a defined period, but there are variations for different species. The collector sits quietly with one or both legs exposed and collects mosquitoes that are about to bite. The mosquitoes are usually collected by the use of a mouth aspirator (sucking tube) (see Figure 9) or a specialised vacuum mechanical aspirator. A dull or red light torch is necessary for night collections. The mosquitoes are transferred from the sucking tube to a mesh covered paper cup or collected in the detachable receptacle on the mechanical aspirator. They can be killed prior to being identified by placing the receptacle in the freezer for 10 minutes.

The freshly killed specimens can be pinned or placed between layers of loose tissue paper in pill boxes or small tins. Pinned specimens should be correctly labelled and forwarded for identification in special pinning boxes (see Figure 10 and Figure 11).

The choice of the time to conduct biting catches in any particular area should be made on the basis of catches of 10-20 minutes duration every hour over a 24 hour period. This will establish peak biting times for each species so that standardised time catches can be made from week to week or season to season. Generally the first hour after sunset is used as a standard period for man biting collections and collections are usually made for 20 to 30 minutes. If the adult mosquito population is very high, the collection time can be shortened. Collection should be recorded on standardised collection forms (see Table 3) for later comparison and analysis.

A variation of this technique is the human attraction collection, where the collector uses an aspirator or wide net to collect mosquitoes attracted to the vicinity of collector before they attempt to bite. This is useful for rapid surveys for day biting species, where the mosquitoes can be relatively easily seen. Human attraction collections are usually made in sheltered vicinities where adult mosquitoes tend to harbour during the day. For dengue vector *Aedes*, these collections can be made using a sweep net around the legs over a 5 minute period.

6.2.3 Animal Collections

Mosquitoes can be collected from tethered animals. This method involves collecting mosquitoes that are attracted to a particular animal and does not necessarily include those mosquitoes that are attracted to humans. This method, using a mechanical aspirator and protective clothing for the collector, can be used safely and painlessly to catch large numbers of mosquitoes.

6.2.4 Animal Bait Traps

For animal bait trap collections, the animal can be enclosed in a trap set out for a night time collection. The trap is designed so that mosquitoes can enter easily but can't find their way out. Mosquitoes can be collected from the trap the following morning by the use of a mouth or mechanical aspirator such as a small car vacuum with a catching attachment. Magoon traps are large specially built animal bait traps housing an animal inside a net protected cage and surrounded by a trap body that allows mosquitoes easy access but limited egress.

Animal bait collections do not give the same results as human biting collections. There are variations in the attractiveness of various mosquito species to different animals, and animal bait collections may be less suitable to assess the relative numbers of various species of mosquitoes that prefer to bite people in particular. They are however safer than human biting collections when there is a risk of mosquito borne disease, and are usually more convenient than direct animal collection.

6.2.5 Window Traps

These traps are mounted on windows in houses or special experimental huts, and rely on trapping mosquitoes as they enter or leave via a limited route. They are mainly used in programs such as malaria programs, where an assessment of the numbers and species that enter and leave houses is used to determine likely vectors.

6.2.6 Net Traps

Net traps are used around a person or an animal and rely on the habit of mosquitoes approaching low to the ground and then trying to leave by flying up. Mosquitoes enter under the net to feed on the bait animal and are then restricted in their escape by the insect net. A collector catches the mosquitoes from the inside of the net. This method can be used to collect large numbers of mosquitoes, especially when a large bait animal is utilised. It can

provide good specimens and can be used with little risk of the collector getting bitten. A person can be substituted for a bait animal and if further protected by an inner net, this can be a safe and convenient way to assess species specifically attracted to people.

6.2.7 Carbon Dioxide Traps

Dry ice will attract some species of mosquitoes. Cylinder traps with entry funnels utilising carbon dioxide are constructed so as to allow entry to insects attracted to the carbon dioxide but restricting their exit. This method is used when relatively clean catches of mosquitoes are required and but will only catch certain species of mosquitoes that are both attracted to CO2 and will enter the funnel.

A variation of this method is the passive trap developed by Scott Ritchie and colleagues in Queensland. The passive trap uses no mechanical suction, with mosquitoes attracted to below the trap by cylinder delivered CO2. The mosquitoes tend to fly up into the open bottom of the trap and are killed and retained on the inside of the trap with insecticide impregnated honey baits on the walls as they feed. The baits can be used as a virus surveillance tool by analysis for arboviral dna expectorated by the feeding mosquitoes. The advantages of this trap is that it requires no battery power, and can be regulated by timers to the cylinder regulators to catch large number of mosquitoes over certain periods of the day for up to a week between services.

6.2.8 Light Traps

Light traps are the most commonly used adult surveillance technique. There are many different types of light traps that can be used to attract and trap mosquitoes. Light can be used by itself or in combination with carbon dioxide. There is a variation in species and numbers collected using light alone or light plus carbon dioxides as attractants. The advantages of these traps are that they can be operated all night and can sometimes collect large numbers of insects. They can be permanent or simple and portable, and there is a variety of power sources available.

Light traps can be used to obtain the relative numbers of some species of mosquitoes that are active at night. The CDC light trap is usually made up of a small incandescent bulb, a fan and a catching receptacle (Figure 12). It is suspended about 2 m above the ground in a sheltered position but with a wide range of view. It should be placed in a position where it does not compete with other light sources and care may have to be taken to prevent ants from devouring the catch. Traps are set before sunset and collected after sunrise. To increase trap catches, a supply of "dry ice" or gas from a bottled source can be discharged near the trap entrance. Other modifications can by made to increase the trapping efficiency, such as the CFG trap that incorporates counter flow geometry where modified air flow increases the catch.

The Mozzie Magnet trap utilises counter flow geometry and a propane gas bottle to both power the trap and supply the CO2, and catches mosquitoes continuously into alcohol or dry over a period of days to weeks. The advantage of traps like the Mozzie Magnet trap is longer term and larger catch trapping without an operator, but has disadvantages in sensitivity to movement, fungus in the catches, and technical difficulties in operating continuously.

Special carbon dioxide baited light traps are commonly used as a standard adult mosquito monitoring technique in Australia. The EVS (Encephalitis Virus Surveillance) trap consists of an insulated can that contains the dry ice, a small battery driven fan incorporating a "grain of wheat" light suspended below the can, and a collecting receptacle attached to the trap body. Various wavelengths of light can be used to attract different species of mosquitoes.

If light alone is utilised, trapping should be done on similar moon phases, as attractiveness of the light will vary considerably. When a permanent trap site is chosen, it should not be

changed during a program. Trap results can vary markedly from one site to another due to the proximity of the vegetation, exposure to wind, the effect of lights and other less obvious factors. Trap results are recorded on the standardised collection forms (see Table 4).

6.2.9 Truck Trapping

A large rectangular mouthed funnel shaped vehicle mounted insect trap can be used to collect only flying insects. This is a direct sample of what is flying and has no bait bias that may be present in the trap methods. This method can be used to indicate the times of flight activity of different species of mosquitoes. Collections are usually made over a fixed route at regular intervals. It can also be useful in assessing the efficiency of adult vector control treatments by comparison of pre and post control sampling. One disadvantage is the height of the mouth of the trap on the vehicle, which gives a bias to a certain height of collection.

6.2.10 Spray Catches

Aerosol spray catches inside houses can be carried out to determine what species and numbers are inside houses. Mosquitoes are collected from sheets laid on the floor.

6.2.11 Resting Station Collections

Most adult mosquitoes rest in the day in cool, dark, humid places. Careful searching may locate particularly productive resting places, from which regular collections can be made. These collections can then give an idea of the relative mosquito population. These particular sites are usually productive for only one and two separate species. Some typical sites include wells, under bridges, in storm water drains, verandas, hollow logs, dense fern patches, overhangs on creek banks, and caves. The mosquitoes are collected with an aspirator, small hand held adapted vacuum collector, or a large powered suction sampler.

6.2.12 Fay Traps

The Fay trap is a day-time trap which is quite specific for *Aedes aegypti* adults of both sexes. It is based on the attraction of contrasting gloss black and white panels and involves a wind orientated cover and a cylinder, housing a battery operated suction motor and a suspended collection bag. The trap is placed near a suspected *Aedes aegypti* breeding location to establish the presence of *Aedes aegypti*.

6.2.13 BG Traps

This trap was originally designed to attract the dengue mosquito *Aedes aegypti*. However, this trap also attracts other *Aedes, Culex* and *Anopheles* mosquitoes. The trap consists of a collapsible white cylinder with white mesh covering the top. In the middle of the mesh cover is a black tube through which a down flow is created by a 12V DC fan that causes mosquitoes in the vicinity of the opening to be sucked into a catch bag. The catch bag is located above the suction fan. The air then exits the trap through the mesh top. This design generates ascending current and it is claimed to be similar to that produced by a human host, both in its direction, geometrical structure, and chemical composition of the attractants. Attractants, a combination of lactic acid, ammonia, and fatty acids are given off by the BG-Lure®. The lure releases the long-lasting attractant for up to five months. There have been reports that the lure is not very effective for some species. The trap can be made much more effective for Aedes species by the addition of solid dry ice or CO2 gas from a cylinder and regulator. Models can vary in size and there are 240v power models for continuous operation. Other lures such as octanol could be used.

6.2.14 Gravid Traps

This is a trap designed to collect gravid *Culex* mosquitoes and is most effective for the collection of mosquitoes of the *Culex pipiens* complex. The trap consists of a pan over which is suspended a suction trap similar to a standard CDC miniature light trap. An attractant such

as hay infusion is added to the water in the pan and the attracted gravid mosquitoes are sucked into the collecting receptacle by the battery operated fan. The trap can be set over night or all day. Collecting gravid mosquitoes increases the chance of collecting mosquitoes infected with pathogens.

6.1.15 Sticky ovitraps

Sticky ovitraps are composed of acetate sheets with an adhesive facing inwards to the inside of the ovitrap and placed inside an ovitrap suitably baited with water and feed or organic rich water. Female receptacle breeding mosquitoes are attracted to the ovitrap and are caught on the adhesive. This method is useful in exotic *Aedes* sampling to determine the presence of adults that come to lay eggs, or to remove them from the population.

7.0 RECORDING DATA

The results of all collections should be entered on standardised collection forms. Examples of collection forms are shown in Table 2, Table 3 and Table 4. Essential items to be recorded are locality, date, collector's name, sampling station, type of collection, number of mosquitoes, sex of mosquitoes, species of mosquitoes, population index (larvae per dip, number of adults biting per hour), and meteorological and habitat data.

All collection forms should be collated and kept in date order as permanent records of the program. In addition, ongoing tabulation and graphing of all results should be maintained on a weekly basis so that a quick visual inspection will show the current status of mosquito populations. Other variables such as tide data, river height, temperature, control operations, and any other locally important characteristic, should be incorporated into the visual presentation or analysed with the monitoring data to gain an insight into the reasons for population fluctuations. The monitoring data should also be analysed with information on pest thresholds or cases of mosquito borne disease to determine whether the control program is achieving the ultimate aims.

The surveillance results should be critically examined at least once per month to determine underlying causes for variations and whether the variations can be attributed to any of the measured variables. Indications of variation that can be attributed to one particular variable should be critically examined and tested wherever possible, either in the field or the laboratory.

Each year the whole program should be examined to determine annual patterns of abundance and any reasons for annual variation. Annual Reports should be compiled outlining all aspects of the program and include up to date vector control maps, changes in procedures and equipment, and details of all control operations. Assessments and conclusions about the surveillance program and the overall control program should be made so that annual progress can be assessed.

If the surveillance programs and an integrated control program have been properly planned and carried out, you should see reductions in mosquito populations and corresponding reductions in potential or actual mosquito borne disease.

8.0 REFERENCES

Dale P and Morris C (Editors), Panel of authors (2002). 'Mosquitoes, surveillance and disease'. In 'Australian Mosquito Control Manual', *Mosquito Control Association of Australia,* revised edition 2002.

De Little SC, Bowman DMJS, Whelan PI, Brook BW, Bradshaw CJA (2009). 'Quantifying the drivers of larval density patterns in two tropical species to maximise control efficiency'. *Environmental Entomology 38 (4): 1013-1021*

Bisevac L, Franklin DC, Williamson GJ, Whelan PI. 'A Comparison of Two Generic Trap Types for Monitoring Mosquitoes Through and Annual Cycle in Tropical Australia' (2009). Journal of the American Mosquito Control Association 25(1):58-65

Hagstrum DW. (1971). Evaluation of the standard pint dipper as a quantitative sampling device for mosquito larvae. *Annals of the Entomological Society of America*. 64: 537-540.

Kay BH and Jorgensen WK (1986). Eggs of *Aedes vigilax* and their distribution on plants and soil in south- east Queensland saltmarsh. *Journal Australian Entomological Society* 25:267-272

Kurucz N, Whelan PI, Carter JM and Jacups S (2009) 'Vegetation parameters as indicators for salt marsh mosquito larval control in coastal swamps in north Australia' *Arbovirus Research in Australia* 10:84-90

Kurucz N, Whelan PI, Carter JM and Jacups S (2009). 'A geospatial evaluation of *Aedes vigilax* larval control efforts across a coastal swampland, Northern Territory, Australia' *J. Vector Ecology* 34 (2):317-323

O'Malley C (1995). 'Seven ways to a successful dipping career'. Wing Beats, reprinted in Bulletin of Mosquito Control Association of Australia, 8(2):19-20

Service MW (1993). Mosquito Ecology: Field sampling methods. London: Elsevier Science Publishers Ltd.

Yang G, Brook BW, Whelan PI, Cleland S and Bradshaw CJA (2008). 'Endogenous and exogenous factors controlling temporal abundance patterns of tropical mosquitoes' Ecological Applications 18(8), pp. 2028-2040.



Figure 8: Vector control map

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Figure 9: Equipment for mosquito surveys
Table 2: Mosquito larvi	al colle	ection	form																			
SHIRE OF:					PRE	-TREA	TMEN		RVAL	SURVE	Y RE	PORT						Date:				
	5	ATER I	рертн	_	WATEF	SUALI	Ł	LAR	VAL DE	NSITY (/m²)			JISTRIB	UTION				INSTAI	K) (%)		
Site name or number	01-0	J0-25	52-50	>20	(Ͻ [°]) qm∋T	PH Conductivity	(mɔ/ɛm)	01>	001-01	000,1-001	10,000 <	estimated total area	of site (m [*]) Edge (use '1' to	indicate) Nidespread (use '1'	estimated % of site	area requiring	-	=	Ξ	≥	əsquq	Total
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Rainfall (during 3 weeks	s prior t	O SULVE	ey)					+	2 1	construction	se veu	sist wit	h or h	e relev	ant to	determ	ining ne	ed and	hest ti	ming fo	L L	<u>, 101</u>
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High tides (during 3 weel	ks prior	to sur	vey)																			
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 Table 3: Adult survey form (man biting collection)

ADULT SURVEY

MAN BITING COLLECTION : INDOORS/OUTDOORS

AREA No:	COLLECTION S	ITE:	
STATION No:			
SEASON:	TEMPERATURE:	START	END
WET DRY	HUMIDITY :	START	END
CLOUDY FINE RAIN	WIND SPEED:	START	END
_	DIRECTION:	START	END
No. Of COLLECTIONS:	HOME:	LOW LEVEL	PIERS
COLLECTION METHOD:	SCREENED	UNSCREENED	SCREENING
PAROUS RATE:	WALLS: BRICI	K 🗌 TIMBER 🗌	FAULTY DLACK BLACK VENEER

TIME	TOTAL N	ο.			SPECI	ES	
11ME	An	С	CUP NO.	NO.FEED	ANOPHELINE	CULICINE	
6-7							
7-8							
8-9							
9-10							
10-11							
11-12							
12-1 AM							
1-2							
2-3							
3-4							
5-6							

TOTAL No. per M/H COLLECTION DATE SIGNATURE OF COLLECTOR Copyright Department of Health Western Australia 2013

 Table 4: Adult mosquito trapping record

Trap site:	
Trap location details:	

Time set:	
Time collected:	

Overnight weather conditions:

Collection number:

Details of identified mosquitoes						
Species	Females	Males	Total			
Total						





Figure 6. Touch polish to pleural aspect.

Figure 7. Adjust height of point with setter.





Figure 1. Pinned specimens in box.



Figure 2. Box ready for packing.



Figure 3. Adding cardboard protector.

PREPARATION OF INSECTS FOR SHIPPING



Figure 4. Adding cotton cushion.



Figure 5. Exterior packing.

Figure 12: Light traps



EVS- CO2/LIGHT TRAP

9. ADULT MOSQUITO SAMPLE HANDLING PROCEDURES FOR TRANSPORTATION

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For relatively small samples up to 200 mosquitoes

Outlined here is the preferred method for the preparation of adult mosquito samples containing below 200 individuals for transportation. This method may be used for larger sample sizes <u>if</u> multiple sample vials are used and labelled appropriately. For example: trap site name plus vial 1 of 3, 2 of 3, and 3 of 3 etc. <u>All</u> vials should be labelled with the trap location and the date collected.



A sample vial with a sachet of salt. This is to absorb moisture from the sample to prevent mould growth.



A piece of tissue is firmly pushed into the vial to create a partition and cushion layer above the salt.



A piece of tissue with one layer removed is placed over the vial and a well space created as shown.



Mosquito sample is carefully transferred into the well space.



The tissue edges are then folded over <u>gently</u> to enclose the mosquito sample and allow the lid to be screwed on.

*NOTE: DO NOT FORCE SAMPLE OR COMPRESS AT ANY STAGE.

Figure 13: Preparation for transportation of adult mosquito samples below 200 individuals

For large sample sizes above 200 mosquitoes

Below is the preferred method for the preparation of adult mosquito samples that contain above 200 individuals for transportation. While providing cushioning and preventing sample damages, the alternate layering of mosquitoes and tissue paper limits moisture pockets within the mosquito sample therefore, reduces the incidence of mould growth. Specimens will then be presented in good condition for identification.



Sample vial with salt added and single layered tissue pieces for packaging.



A piece of tissue is firmly pushed into the vial to create a partition and cushion layer above the salt.



A portion (*Less than* 200 individuals) of the mosquito sample is placed on top of the tissue cushion.



Another layer of tissue is gently placed over the 1st portion of mosquitoes.



The remainder of the sample (*Less than 200 individuals*) is placed on the previously laid tissue.



Finally, the last tissue piece is placed loosely on top to prevent mosquitoes from bouncing around in transit.

***NOTE:** DO NOT FORCE SAMPLE OR COMPRESS AT ANY STAGE.

Figure 14: Preparation for transportation of adult mosquito samples above 200 individuals

10. THE PHYSICAL CONTROL OF MOSQUITOES

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BACKGROUND

The physical control of mosquitoes is a method of source reduction. It is any physical method used to reduce or eliminate mosquito breeding sites. It is a planned modification of the environment that physically removes water or makes that water unsuitable for mosquito breeding.

Any mosquito control program must be planned after taking all available relevant information into account. It must be based on a definition of the problem, and all aspects of the problem must be considered including:-

- a) the geographical scope of the problem;
- b) the nature of the problem;
- c) the species concerned;
- d) the flight range of the species;
- e) the location of breeding sites in relation to people;
- f) when the problem occurs;
- g) habitat preferences of the mosquito to be controlled;
- h) local control possibilities;
- i) the environmental impact of the control methods;
- j) the cost of the various control methods.

These preliminary considerations should allow a decision to be made on the type of control that is most appropriate for any particular problem.

Physical control has many advantages over other forms of control and is especially advantageous in offering a long-term solution. However, physical control can be relatively expensive and require specialized resources. A cost benefit analysis is often helpful in deciding the most suitable method, particularly where regular maintenance is required.

The most common physical control methods are dealt with in detail in the following sections:

- Filling (page 72)
- Draining (page 73)
- Water management (page 79)
- Management of sewage treatment facilities (page 82).

FILLING

The physical filling of mosquito breeding areas with sand or earth is the ultimate mosquito control method, as it removes sources of mosquitoes permanently, with no recurrent maintenance costs of periodic inspection and insecticide control. This method has the advantage that it can be done by relatively simple measures and can add to property values.

SMALL FILLS

Small amounts of earth or other material can be used to fill in any small depressions that hold water. Areas treated could include sand or gravel borrow pits, uneven land clearing, uneven settling of sanitary landfills, water scoured depressions, abandoned ditches, tree stump holes, abandoned wells or septic tanks, and small mining scrapes or ditches (costeans). Attention to the final slope is most important. This method is cost effective for small volumes or when fill material is locally available, but can become uneconomical with increasing volumes and large cartage distances.

NATURAL FILLS

Natural method of filling can be used to achieve a supply of fill when artificial fill is unavailable or uneconomical. Natural fills are entirely dependent on the nature of the site to be filled but can often be achieved with relatively little capital outlay. Types of situations where this method could be used include:

- a) Silt laden streams being redirected to fill depressions;
- b) retention dams to trap silt;
- c) wind-fills with the use of sand-trapping vegetation.

HYDRAULIC FILLS

Hydraulic filling is the process where silt-laden water is pumped by pipe into swampy or lowlying areas that are to be reclaimed. This is a capital-intensive process and is usually only viable if the land reclaimed can be used.

Particular consideration is needed to ensure that the water used in the process does not become a breeding site, both in the fill area or where it is discharged. Other considerations include attention to contouring and final slope for drainage, the levelling to avoid depressions, the provision of a vegetation cover to stop erosion, and internal drainage and erosion prevention measures.

Hydraulic fills are often used in:

- a) dredging harbors and rivers;
- b) the disposal of mining waste.

SANITARY LAND FILLS

The disposal of urban garbage and rubbish can be used to eliminate mosquito breeding sites and improve land values. Dumping into flooded areas should be avoided because this can actually lead to an increase in mosquito problems. In flood prone areas drainage provisions for both preventing off site inflow and providing on site drainage need to be incorporated into the land fill operation.

There must also be consideration of the final soil cover, the final slope, and the final vegetation cover.

DRAINING

Drains are generally constructed structures to convey stormwater, waste-water, or other sources of water by a flow path or formal channel from a catchment or source to a discharge point. Drains can be part of an organized drainage system or a specific measure to drain depressions or temporary flooded areas. Generally drains in small catchments in undeveloped areas only convey water during or for a few hours after rain, and if they have adequate slope, dry up soon after the rain ceases. However drains with larger catchments or from perennial or artificial sources of water, contain water for relatively long periods and can be significant sources of mosquitoes.

Stormwater drains, particularly those near urban areas or industrial developments that receive organic or nutrient rich wastewater, can be prolific sources of mosquitoes. The mosquito species commonly found in freshwater drains include *Culex quinquefasciatus*, the brown house mosquito, *Culex annulirostris*, the common banded mosquito, and *Anopheles annulipes*, the common Australian *Anopheles*. Drains near tidal areas, particularly in the tropics, can be substantial sources of *Aedes vigilax*, *Verrallina funerea*, *Cx. Sitiens*, *An. farauti* and *An. hilli*. As these species include those that are serious pests of people and potential vectors of disease, the presence of large numbers close to people can pose serious public health problems.

Mosquito breeding generally occurs in drains when the flow is intermittent, the water has high nutrient levels, and the channel is overgrown with semi aquatic or aquatic vegetation. Breeding also occurs in residual pooling as the drain dries out after ceasing to flow or after tidal inundation. Those drains that are capable of sustaining fish or aquatic bugs and beetles are normally not significant sources of mosquitoes. Drains can be constructed with provisions to reduce or avoid mosquito breeding. Generally well-constructed impervious drains are cost effective in the long term, as they are not subject to the need for regular maintenance or expensive rehabilitation, and have little requirement for ongoing insecticide control operations.

Drainage can be a very efficient method of preventing mosquito breeding, but drain construction without due consideration of good design features can result in new and prolific mosquito sources. Adequate considerations of these features in the planning of drainage systems can reduce expensive and ongoing maintenance requirements, avoid additional mosquito populations, and reduce potential public health problems.

GENERAL CONSIDERATIONS

The amounts of water to be removed when considering drainage for mosquito control may often be small. These may not be significant to the civil engineer or the farmer but may be of great significance as a mosquito breeding site. Drain features for mosquito control often need to be less sophisticated than civil engineering considerations for storm water control. The water does not always need to be contained, and only needs to drain away over a four to five day period before mosquito larvae can mature, rather than a matter of hours required for civil engineering drains.

The choice of the drainage method for mosquito control (draining, pumping or tide control) and type of drain depends on the local capabilities, the size and nature of the mosquito problem, the relative cost, the terrain, the soil type, and ongoing maintenance considerations.

The needs for drains can sometimes be reduced. Any land clearance or development operation should include the rectification of small depressions by grading and selective filling, and the inclusion of infiltration technology, so that the finished landscape is free draining without the water being concentrated in defined flow-paths.

Sometimes land development does not fill a number of depressions, or stormwater is concentrated to low lying areas by sealed or impervious areas (buildings, roads). Often it is cheaper to drain small to moderate depressions rather than fill them. In these cases, drainage can often be achieved by simple open drains, such as narrow grassed swales or shallow gently sloped ditches to convey water to a stream, another drain, or to spread it over sloping ground for infiltration. For larger depressions or greater volumes, drainage may require a broad floodway. The equipment requirements for drainage are generally simple, ranging from hand tools, bobcats or graders. Usually the input of labor and money for these methods are relatively small, and the works can be carried out by general contractors without formal measurement.

Where larger water bodies need to be drained or if there is seasonal to continuous flow, a more formal drainage system may be required. The importance of good engineering design for larger scale drainage increases with the size and scope of the problem. Two points to remember are:

- a) the minimum structure should be used to achieve the desired result;
- b) drains should be rounded or 'U' shaped in cross section to ensure that flow can continue for relatively small volumes, and the drains are self cleansing.

The drainage of large areas or extensive water bodies needs a more thorough engineering appraisal. Drainage of large swamps and large catchment areas that have perennial flow may have permanent effects on the local or down stream environment. These larger engineering projects generally need hydrological, engineering and ecological expertise to ensure that the drainage achieves the desired results without causing deleterious effects. These large-scale engineering undertakings are not dealt with in detail in this paper.

There are a number of specific questions that should be considered before constructing drains. Adequate consideration of these questions can have a large outcome on whether the drains become sources of mosquitoes. The questions include:

a) What type of drain is required (open unlined, open lines, subsoil)?

The type of drain required is dictated by a host of physical and resource considerations, but the possibility of creating mosquito breeding should be a major consideration.

b) How much water needs to be drained?

The volume of water to be conveyed will determine the size of the drain required. If it is a small volume it may be drained easily in a matter of hours by a simple ditch. If it is larger, it may be drained by a drain of sufficient size over a period of up to 5 days without causing mosquito breeding problems.

c) When is the drainage necessary?

If the drainage is required only during rain episodes, there may be no need for any permanent concrete features. There may be little opportunity for mosquito breeding in the drain due to the regular flushing and the rapid draining or drying of the area when the rain ceases.

d) What is the ecological effect on the drained area?

If a deep-water area is drained, the drainage may reduce wave action or disrupt fish ecology. It may become a shallow area that permits emergent vegetation growth that could be conducive to mosquito breeding. If the drained area is only partially drained, or takes a relatively long time to drain, periodic refilling in a shallow vegetated wetland could cause a new mosquito problems.

e) Where is the outlet and what are the down stream or upstream effects?

If the outlet is to the open sea or a large river, it will generally be adequate to prevent mosquito breeding. If the outlet is to a sheltered area near the upper tide limit or to a small creek or a lake, the deposition of silt and subsequent cut off pools or vegetation growth could cause new mosquito breeding sites. If the drain includes drop structures, they may prevent the seasonal redistribution of fish to upstream areas.

f) What is to be done with earth spoil?

If the spoil is placed on the uphill side of a drain it will restrict flow into the drain. Earth spoil should be placed on the downhill side of a drain and have numerous breaks to allow drainage into the drain.

g) What maintenance considerations are necessary?

Open earth drains require regular maintenance, while subsoil drains require only occasional maintenance at a limited number of points. Maintenance procedures will require resources for the longer term, and capability of maintaining these resources should be considered before the drain is constructed. Maintenance also requires adequate access for inspection and rectification equipment, which may alienate larger areas of land.

TYPES OF DRAINS AND DESIGN CONSIDERATIONS

OPEN DITCHING (SURFACE DRAINS)

Open drains (OD) can be either simple unlined structures or well-constructed and shaped drains with full concrete lining. Open unlined drains (OUD) are often the cheapest and most simple form of drainage, but many aspects should be considered before construction. After due consideration, it may be clear that partial concrete lining is required, or that it is more practical to construct a subsoil drain.

Open unlined drains are usually subject to regular maintenance for weed control and silt removal. If the drains are in urban or industrial settings, they often contain organic pollution and vegetation, and usually become prolific sources of mosquitoes. Upgrading unlined drains to permanent concrete drains is often justified in urban and industrial areas where the flow needs to be contained totally within the drain and where high flows or velocities are experienced. Upgrading can be justified if regular mosquito control or excessive maintenance is required. Often mosquito breeding in larger drains is seasonal and occurs with dry season low flows. In these cases the upgrading may only require a central dry season flow concrete insert to facilitate flow and silt movement. These low flow inserts can drastically reduce maintenance requirements by removing the long-term water and nutrient sources that encourage vegetation growth and subsequent silt accumulation.

Some of the major aspects that should be considered before the construction of open drains have been outlined below. They are discussed more fully in the listed reference, (Pratt et al., 1972).

a) Dimension of drain required

Drains for mosquito control only need to be of dimensions that will drain an actual or potential flooded mosquito breeding site over a period of 3-5 days, ie the drain should be as small as possible to achieve the desired aim. For relatively flat topography in undeveloped areas, gently shaped grassed floodways with shallow central inverts can adequately cater for periodic storm flows.

Often the time required for drainage to occur before mosquito breeding can be completed will dictate the dimensions of the required drain. Flooded areas that come under tidal influence at any time of the year should drain within three (3) days because of the shorter larval time for saline water species, while all freshwater areas should drain within five (5) days. These time frames incorporate a safety margin when extended rain occurs. If a drain overflows its banks during periods of high tides or heavy rainfall, the overflow water should drain back in to the drain within the above times. Storm floodways or drains may receive continuous seepage or wastewater during dry periods. The seasonal volume of these low flows will determine the dimension of a low flow concrete invert.

b) Route and layout of the drain

Generally drains should follow the shortest route from catchment to discharge point. If there are small drainage problems adjacent to main drainage routes, smaller lateral drains can be constructed to those areas. For extensive, flat, poorly draining areas, a fishbone lateral feeder system may be required. Drains should be as straight as possible, to shorten the length for construction and maintenance, and enable more rapid flow. There should be no sharp bends that result in bank erosion or require rectification measures. Any bends, drain junctions or discharge points may need erosion prevention structures, and may require access for silt removal.

c) Grade of area to be drained

The grade of the area to be drained will determine the need for erosion control structures and the dimensions of the drain.

The finished grade should be adequate to drain the area over a number of days, but not so steep that erosion occurs. If steep areas are to be drained, adequate erosion control drop structures will need to be installed. A grade of at least 0.01% to 0.05% is required for concrete drains or drains with concrete inverts. Often grades for mosquito control drains can be considerably lower than for civil engineering purposes. Greater slopes between 0.1 and 1% are required for grassed or earth drains where minor silt and vegetation accumulation can cause pooling in the drain.

d) Side slope of drain

The side slope of the drain will determine the stability of the drain, and is largely determined by soil type. Drains in rock and clay can approach the vertical, while in most compact soils a 45° slope is required. In sandy soils or sand, slopes less than 45° are required. The sides of drains with appreciable flow and erosion prone soils may require erosion prevention measures such as rock baskets or vegetation mats.

e) The location of berm spoil

When drains are constructed in low lying or level areas, any berm or spoil should have regular breaks to allow lateral drainage into the drain such that no pooling occurs outside the drain. In sloping situations, the berm should be placed on the downhill side of the drain to prevent ponding uphill of the berm. The spoil should always be placed at a sufficient distance from the drain so that is cannot erode or slump into the drain. Earth spoil on the berm can cause week growth or ecological change and may require removal or spreading. Soil spreading in very flat areas such as salt marshes may still cause shallow pooling and may require lateral drains through the disturbed areas to the central drain.

f) The final depth of the drain

Drains should only be as deep as required to gain sufficient grade to enable timely flow to a suitable discharge point. Drains for mosquito control generally should be as shallow as possible. They can be smaller if occasional overflow does not cause problems. The invert of any drain should not be below the seasonal water table. Deep drains promote continued seepage into the drain. This may result in perennial water within the drain, encourage continuous vegetation growth, and result in long term mosquito breeding.

g) The end point

Drains with dry season low flows, or considerable and prolonged flows, should discharge directly into daily flushed tidal areas, large channels or creeks, or large bodies of deep water. Drains with low flows should never discharge to flat, ill defined, low lying, poorly drained areas.

The end point for 100 year flood drains without dry season low flows should be just below the maximum high tide level in tidal areas, or just below the 100 year flood level for larger receiving rivers or water bodies.

The invert of the end point of drains with dry season low flows near tidal areas should be below the average high tide level or to a natural well-defined tidal creek that drains freely at low tide. A channel could be dug back from a tidal creek to satisfy this requirement.

h) Soil type and erosion potential

Open unlined drains of moderate grade through unconsolidated or erosion prone soils will require the installation of erosion prevention structures (such as stone and iron mesh gabions) wherever there is a likelihood of erosion within the drain.

If there is a potential for soil erosion or appreciable silt movement in development areas, drains through the area should include silt traps. Silt traps should be constructed upstream of the discharge point to freshwater or tidal creeks. Drains, erosion prevention structures, and silt traps should be installed as a first stage of development, particularly if the drainage discharges into relatively flat areas or to creeks or another water body. Any silt trap should have access for regular maintenance and silt removal.

i) Potential for dry season low flows

Urban or industrial areas can often produce dry season low flows in drains. The ultimate standards for urban drains with low flows are impervious underground pipes or open lined channels with central low flow inserts. Central impervious low flow capabilities are essential where there is a likelihood of dry season low flows. Low flows generally occur from institutions with large lawn areas or garden beds that are watered by automatic sprinkler systems (eg hospitals, schools, parks and shopping-centres). Low flows are also highly likely in areas that generate waste-water from watering, processing or washing operations (eg service stations, industrial areas, plant nurseries and shopping centres). In general the larger the catchment, and the more institutions and non-residential developments within the catchment, the greater the probability of dry season low flows in drains servicing the catchment.

j) Drain maintenance

There should be no vegetation, cut off pools or silt deposits in drains. Drain maintenance such as silt removal, weediciding or vegetation and debris removal for earth lined drains should be programmed on an annual basis. Maintenance easements should be included alongside all open earth lined drains. Drains that discharge into dams or lakes will require periodic silt removal at the discharge point into the water body to prevent the establishment of aquatic and semi aquatic vegetation.

SUBSOIL DRAINAGE (UNDERGROUND DRAINS)

The types and size of subsoil drains will need to suit local conditions. Subsoil drains can either be impervious or pervious, depending on the need to either carry or dissipate the water. Subsoil drainage may be expensive initially but can reduce maintenance and access problems. The decision to construct subsoil drains as opposed to open drains will generally be made on cost and access considerations.

Pervious drains include underground terracotta, perforated pipe, rubble, sand or gravel drains. Sometimes pervious drains can be constructed unintentionally by the breakdown of jointing or leaks in pervious pipe systems.

Pervious drains are used to remove surface water and allow infiltration of wastewater over a larger area. Pervious drains generally can handle only limited amounts of water before saturating the surroundings and causing surface pooling. They are also not adequate for long term drainage, as the pore spaces become compacted or filled with earth and blocked by root growth.

Impervious drains are the most common drainage systems in urban and industrial developments. Generally they are constructed of concrete or PVC pipe sections and convey water to a suitable end point. Impervious drains generally have adequate slope and if they are round in cross section, convey even small amounts of water and are self cleansing. Their main advantage is the reduced access requirements for inspection and machinery, and the reduced maintenance costs. Their main disadvantage is their high initial capital cost.

PUMPING

Pumping of excess water can be a useful method to drain areas by collecting the water from one point and lifting it so that it can flow to another water body or to a suitable disposal site. Pumping can be used when there are depressions below tide or river levels, or the gradient is too small for simple draining or ditching. Pumping has an energy requirement, but with innovation this can be supplied by solar or wind power, as well as electrical pumps. It can be considered for small-scale drainage in some situations, as well as for large projects.

TIDE CONTROL

Draining large low-lying areas or drains inundated by tides can be facilitated by tide exclusion. Tide gates or rock or earth barriers can be installed in drains and across tidal flats to restrict or prevent the inundation of areas below maximum tide level, and then allow the water to drain through a gravity operated gate or porous rock sections during low tide levels.

Tide gates can vary from large sophisticated flap valves at the end point of large underground pipes or open channels, to simple hinged barriers in an open earth drain. There can be varying combinations of ditching, dyking, pumping and tide gates in large scale projects. One of the biggest considerations for tide control is the ecological effect on upstream habitats. Tide exclusion in drains can cause a proliferation of freshwater semi aquatic and aquatic reeds upstream of the tide barrier. Sometimes freshwater vegetation in drains or marsh areas can be controlled by allowing occasional tides to inundate the vegetation at critical times in the growth stages.

WATER MANAGEMENT

Water management is an important method for controlling mosquito production in constructed water impoundments, farm ponds, sewage ponds, salt marshes and irrigated land. Water management usually involves a good knowledge of the biology and ecology of the mosquito species to be controlled. For small and less sophisticated water storage areas, this can require regular assessment and regular decision making. For large water storage structures, good engineering design can avoid maintenance and regular management.

LARGE AREA WATER STORAGE

Proper reservoir preparation is most important. Aspects to consider are:-

- a) design they must have steep banks which are non-eroding and have short margins;
- b) vegetation there needs to be vegetation clearing between high and low water levels;
- c) silt traps these must be used to prevent silting by inflow streams in the upper reaches;
- d) deepening, levelling or bottom sloping this is to remove residual pools when areas dry out;
- e) the prevention of seepage through dam banks or walls;
- f) levy bank containment for flood periods;
- g) vehicle access around margins.

For large water storage, water management techniques such as alternating the water level can be used to strand flotsam and marginal vegetation. The greatest potential for mosquito

breeding in large water storage occurs in the shallow upper reaches where silt input and shallow water can give rise to dense aquatic and semi aquatic vegetation.

FARM PONDS & DAMS

- a) Need to have a steep clean shore line with little or no vegetation.
- b) Access for animals must be restricted to a narrow area of the margin to prevent pugging.
- c) There should be a minimization of organic pollution to discourage thick vegetation growth.

BORROW PITS

These are holes caused by the excavation for sand or gravel during road construction or mining activities.

Borrow pits should be:-

- a) self draining; or
- b) alternatively deep enough and large enough to have steep clean sides and capable of maintaining a permanent or seasonal population of fish or aquatic beetles or bugs.

SALT MARSHES

Salt marshes are important sources of mosquitoes but these habitats are also very important ecologically. Control methods in these areas must be closely examined against necessity for control, the economics of control, and the possible deleterious ecological effects.

Many aspects of research are needed before salt marsh manipulations are carried out. The biology of the problem mosquito species must be well known, including its habitat preferences, its effective predators, when is it a problem, and which available engineering method is best suited to the local conditions and likely to achieve long term control.

OPEN MARSH DITCHING

Open tidal marsh ditching in areas of little water level fluctuation can be used to promote natural tide circulation. It relies on predaceous fish controlling mosquito larvae. For areas of large tidal fluctuations, open marsh ditching is used to drain the marsh area at low tides. The ditching can be:-

- a) Simple. Connection of low depressions by runnelling or simple ditches.
- b) Complex. An engineered approach with an organized drainage system of main drains and lateral feeder drains. Seasonal freshwater inputs into marshes can require larger drains to prevent flooding and the drains usually require silt prevention and removal considerations. Generally lateral drains should be as shallow as possible so that they drain at low tides.

SHALLOW FLOODING

The flooding of areas using a dyke or levied marshes. In this method the higher tides overtop a low artificial bund to create a more permanent water area. Considerations include:-

- a) the prevention of the flooded area from drying out;
- b) the stocking of the flooded area with fish;
- c) the provision for excess water to escape;
- d) likely vegetation or salinity changes and the effect of these on mosquito production.

DEEP FLOODING

This is usually a more expensive method than shallow flooding. It involves the flooding of an area by using levy banks or dykes that are not topped by storm water or high tides, or by

excavation of shallow swamps to provide artificial lakes. Deep flooding requires at least a large wet season water input or a periodic tidal input.

Considerations include:

- a) The necessity of a spill way or outlet for excess water;
- b) the depth of water to prevent emergent vegetation;
- c) the necessity for top up water in addition to creek or river inlets;
- d) the presence and effectiveness of fish and other mosquito predators;
- e) the ecological effect of increased water retention.
- f) the need for silt removal at inflow sites

RECLAMATION

Reclaiming swamps for agriculture or housing use can be carried out by constructing levies against tides or river water. Considerations are:

- a) the need for very secure levies;
- b) the need for tide gates or pumps;
- c) the need of an internal drainage system;
- d) the consideration of possible marsh subsidence.

IRRIGATED AREAS

Irrigated areas can be very large sources of mosquitoes. Mosquito production can be possible in the four components or irrigation systems.

IMPOUNDMENTS

Considerations are:-

- a) any impoundment must be cleared of vegetation before filling;
- b) depressions and low lying areas should drain freely into the main impoundment;
- c) a margin must be maintained in a vegetation free state;
- d) flotsam should be annually removed by stranding;
- e) inlets points should be maintained silt free;
- f) water-level fluctuations should be practiced when necessary.

Irrigation Conveyance

Considerations are:-

- a) the need for correct layout of water input and drainage discharge;
- b) irrigated areas to be properly graded;
- c) the correct amounts of water only must be applied;
- d) the drainage system must remove excess water without pooling.

DRAINAGE SYSTEMS

Consideration are:-

- a) excess irrigated water must be adequately disposed of;
- b) drainage ditches must be maintained to prevent weed growth, seepage and siltation;
- c) the disposal point must be either into the sea or a large river or lake, or a vegetation free impoundment.

ENVIRONMENTAL MODIFICATION OF BREEDING SITE

Environmental modification includes any change that may be induced into the ecology of the mosquito, such that it will reduce or eliminate that species in that particular area. Modifications are usually only justified when a major vector species needs to be controlled, or when there is little possibility of creating an area that is suitable for another potentially troublesome species. Various environmental modifications include:-

VARIATION OF THE SALINITY OF THE WATER

- a) changing the water to fresh water;
- b) changing the water to salt water.

THE FLUCTUATION OF WATER LEVELS BY ARTIFICIAL MEANS

This can remove vegetated edges by drowning or drying the vegetation.

WATER SURFACE AGITATION

Water falling into ponds by planned waterfalls can create surface agitation. Removing trees or other screening materials around ponds can make wind agitation more effective.

FLUSHING

Small dams can be constructed so that there is a periodic release of water by an automatic flush system to flush out pools or creeks.

ADDITION OF POLLUTED WATER

The suitability of a breeding place can be changed by redirecting water of a different pH or organic content.

REMOVAL OF SHADE

Elimination of trees or shrubs can eliminate some species that require shaded breeding grounds.

THE PLANTING OF TREES

This can create shade or can increase water removal by evapo-transpiration. Shading can dramatically reduce both micro-organisms or aquatic and semi aquatic vegetation, and thus create an un-favourable environment for species requiring sunlit or vegetated breeding sites.

INCREASE OF THE RETENTION TIME OF WATER

If water is retained in an area for longer periods, changes in aquatic vegetation or predator populations may make the breeding site unsuitable or unproductive.

MANAGEMENT OF SEWAGE TREATMENT FACILITIES

Sewage treatment facilities can be major sources of pest and vector mosquitoes (Whelan, 1981, 1984, 1988). Nutrient rich sewage and sewage effluent has the capacity to produce enormous numbers of mosquitoes. As treatment facilities are usually relatively close to communities, these mosquitoes can cause large and continuous pest and potential public health problems.

The mosquito breeding associated with sewage treatment facilities is usually associated with their inadequate design, operation and maintenance, or faulty methods of effluent disposal or dispersal Some of these problems can be rectified in the planning stages, while others need consideration in the operational phase. There is a need for increased awareness of the nature of the potential problems among designers, operators and regulators of sewage treatment facilities. This section outlines the problems, and suggests design and operational practices that can reduce mosquito breeding.

MOSQUITO SPECIES ASSOCIATED WITH SEWAGE

CULEX QUINQUEFASCIATUS: 'THE BROWN HOUSE MOSQUITO'

This species usually breeds in organically polluted water near human communities. It is frequently found breeding in high numbers in unsealed septic tanks and primary sewage ponds, although it is sometimes found in organically overloaded secondary sewage ponds. This is a very appreciable pest species wherever favourable breeding sites exist. The females rarely travel more than 2 km from their breeding sites.

CULEX ANNULIROSTRIS: 'THE COMMON BANDED MOSQUITO'

Culex annulirostris is one of the most common mosquitoes in Australia. The most prolific artificial breeding places are in secondary sewage treatment and evaporation ponds, and sewage pond effluent (Whelan 1984). The larvae are most frequently found in calm, sheltered areas where vegetation offers protection from disruptive wave action and aquatic predators. The females of this species can disperse up to 10 kilometres from the breeding site (Russell 1986), although the highest concentrations are usually found within 3 - 4 km of appreciable breeding sites.

CULEX GELIDUS: 'THE FROSTY MOSQUITO'.

This species is a relatively recent introduction to Australia and is found in northern Australia from S-East Qld, across the NT to North West WA. The larvae are usually found in high organic sites such as abattoir or other animal waste-water storage or contaminated areas, as well as sewage ponds and effluent disposal sites. Females are not found in high numbers unless traps are set in close proximity to breeding sites.

ANOPHELES ANNULIPES S.L.: 'THE COMMON AUSTRALIAN ANOPHELES'

This species usually breeds in open, sunlit, temporary and permanent freshwater ground pools, streams or swamps. It is not found in septic tanks and is rarely found in sewage treatment ponds, but it can frequently be found in sites of disposal of sewage effluent, particularly where the effluent flows into shallow, grassed areas. The females can disperse up to 2 km from their breeding places.

MOSQUITO BREEDING AND SEPTIC TANKS

Mosquito breeding in septic tanks is entirely dependent on mosquito access into the tank, and is usually due to damaged or missing tank tops and inspection manholes or unscreened vents. *Culex quinquefasciatus* is the principal mosquito species found breeding in septic tanks. Septic tanks that overflow, or absorption trenches that are faulty or in water logged soil, can result in surface pooling of untreated or partially treated sewage that can become prolific sources of *Cx. quinquefasciatus* or *Cx. annulirostris*.

In general, the close fitting inspection covers of fibre-glass septic tanks are less likely to provide mosquito access than concrete prefabricated tanks. Concrete tanks with a flat concrete top slab invariably have a small space between the top slab and the tank walls that can be sufficient to allow mosquito access. Inspection manholes or access points that are of faulty design or damaged can also allow mosquito entry.

Concrete tank tops and inspection manholes can be simply mosquito proofed by applying a sand and cement mixture or a silicone sealant to all joints. All vents to septic tanks including gully traps and breather vents should also be screened to prevent mosquito entry.

Mosquito breeding can be detected relatively easily by opening the inspection manhole and disturbing the interior of the tank with a stick. Any observed adult mosquito activity indicates that sealing of the tank is required. If septic tanks are correctly sealed there is no need for insecticide treatment.

MOSQUITO BREEDING AND PACKAGE SEWAGE SYSTEMS

Generally package sewage systems are relatively small facilities where the sewage treatment is carried out in tanks or chambers under an active process, rather than a large passive pond system. One of the finished products can be a variable volume of treated effluent that is usually disposed of by infiltration, sprinkler dispersal, evaporation or discharge to natural water bodies. Mosquito breeding does not usually occur within the treatment facilities, but inappropriate disposal of the effluent produced can cause pooling or ecological changes to receiving water, which results in breeding sites for *Cx. annulirostris* or *An. annulipes*. The precautions and remedies outlined below for sewage pond effluent disposal apply equally for package sewage systems.

MOSQUITO BREEDING AND SEWAGE TREATMENT PONDS

Sewage treatment in pond systems is one of the most common methods of sewage treatment in Australia. Usually the ponds are sited in a relatively low-lying area where a gravity assisted sewerage system delivers untreated sewage. The sewage is treated in a series of ponds involving a passive or active primary pond, secondary aerobic ponds, and final evaporation or holding ponds for dispersal or disposal.

DESIGN CONSIDERATIONS

DISPOSAL OF EFFLUENT

Appropriate planning for the final disposal is an important part of site selection. Disposal near the coast is relatively easy, but can lead to major problems in inland areas if appropriate techniques are not employed.

WIND

Ideally ponds should be located in open windy areas away from steep hills or fringing tall or dense vegetation. Wind and the associated wave action play an important part in preventing mosquito breeding by disrupting the larvae and pupae at the water surface. Wind action can also prevent the spread and hinder the growth of algae, floating aquatic ferns (*Azolla sp.*) and duck weed (*Lemna sp.*), or concentrate flotsam into discrete areas for ease of removal. These floating plants and flotsam should generally be regularly removed as they can shelter the larvae from both wave action and aquatic predators. There are some situations where an even and complete cover of floating duck weed (*Lemna sp*) has shown very good mosquito larval suppression, and these situations generally need to be in wind protected areas or have emergent barriers to prevent wind disruption.

DRAINAGE

The choice of a site should consider the necessity to drain the ponds for maintenance without thereby creating swamps or pools of stagnant water. Effluent release from the final pond is usually suitably arranged, but provision for emptying the intermediate ponds into suitable areas for maintenance is often overlooked.

Site design should ensure that there is no disruption to storm water drainage pathways caused by any of the works. Diversionary drains around facilities that have seepage or dry season low flows should be constructed with concrete inverts and should discharge to suitable endpoints. The permeability of pond walls and groundwater seepage must also be considered and catered for. Evaporation areas or effluent dispersal areas should be suitably graded and well drained, and have surface bunding and diversionary drains to prevent any overland storm flow from entering the disposal or dispersal areas.

Ponds or embankments constructed near tidal areas need to ensure existing tidal drainage patterns are maintained. If these are blocked, problems with salt-water species of mosquitoes breeding in impounded water can be expected.

ACCESS

Site design should allow for all weather access around the entire installation. Weed and tree growth maintenance, fire prevention, and erosion and siltation rectification in diversionary drains may require regular access. Seepage from ponds or diversion of site surface water can cause swampy conditions that require vehicle access for inspection and periodic mosquito control.

POND DIMENSIONS

POND SIZE

Sewage pond size is primarily determined by engineering parameters related to design flow rates, pollution loading, and the required effluent quality. Frequently there is little consideration given to the effect of pond size on mosquito breeding. Adoption of oversized ponds, either from inaccurate predictions of sewage volume or a desire to provide for future capacity, can lead to the ponds becoming shallow, thickly vegetated swamps, capable of breeding large numbers of mosquitoes.

Consideration should be given to staging of pond construction, or the use of multiple ponds, although the use of smaller multiple ponds may inhibit wind and wave action. In most cases, it is the margins of the ponds which provide the mosquito breeding sites. Multiple small ponds can result in increased total margin length. Installing peninsular barriers to reduce short-circuiting sewage flow through a large pond can also markedly increase the margin length. Increased margins require more capital expense with edge treatments, and increased maintenance costs.

POND DEPTH

Selection of pond depth is usually dictated by the function of the pond, namely primary, secondary, or evaporation. Adequate allowance must be made for solids deposition, particularly in primary ponds, otherwise excessive deposits will lead to emergent vegetation and mosquito breeding.

Profiling the pond base, with the deepest side at the pond entry, can help, particularly if there is a seasonal variation in sewage input. For evaporation ponds, particularly those with earth sides or those that may be continually full, require a depth of 1-2 m or more to prevent the intrusive growth of emergent semi aquatic reeds such as *Eleocharis sp.* and *Typha* sp.

CONSTRUCTION DETAILS

VERTICAL CONCRETE MARGINS

Vertical concrete margins have proved to be the most satisfactory means of controlling mosquito breeding by promoting wave action, and maintaining margins free of vegetation and debris.

Concrete walls can be pre-cast for remote locations or constructed in situ, and are cost effective in the long term. The walls should be deep enough to allow for a wide variation in water level and should have a horizontal or slightly sloping bench at the inside base of the walls above the bottom level of the pond, to discourage establishment of vegetation and reduce silt accumulation immediately adjacent to the pond wall. Sealed verges around the top of the banks are desirable to facilitate maintenance and to prevent the erosion of soil into the pond. Walled ponds may still have problems, particularly in primary ponds, with flotsam and

wind blown debris in the corners, and silt accumulation near sewage entry points. Truncated or rounded comers and multiple or variable entry points can help to reduce these problems.

UNLINED EARTH BANKS

Sewage ponds with unlined earth banks have the greatest capacity for mosquito breeding, particularly those with gentle slopes where marginal vegetation can establish. These are accordingly not recommended, except as a temporary or emergency measures. The banks should be constructed using impervious materials such as compacted clay. If neglected, unlined earth banks can become either eroded, or overgrown with grass, shrubs and trees. Corrective measures can then be a major undertaking.

OTHER LININGS

Various systems have been used to line earth banks as a temporary measure to reduce the growth of vegetation, but they have not been entirely satisfactory. Stone pitching of the margins is not satisfactory as it does not offer sufficient deterrent to vegetation growth, and mechanical maintenance becomes difficult. Overlapping cement or iron sheets have been used, but have problems with damage and stability, resulting in subsequent weed growth. Various types of bituminous or plastic sheeting have also been tried, and have shown promise as short to medium term solutions. Problems encountered include inadequate anchoring, weed growth, ultraviolet deterioration, and human interference. The more modem ultraviolet resistant heavy-duty plastics, anchored with earth mounds back from the rim of the ponds, have been more successful.

Sloping concrete margins have been tried in a number of locations. While better than unlined ponds, they have the drawback that wave action is damped by the slope. Dust and organic matter can also build up and enable vegetation to establish. It is important that the margins have a slight rim and sealed verges to prevent side wall erosion and subsequent accumulation of soil and vegetation at the water margin.

MAINTENANCE

Before commissioning sewage pond systems, a general survey of the whole site should be conducted to ensure that mosquito breeding sites have not been created inadvertently by borrow pits formed during pond construction, pools of water resulting from site drainage works, or pooling caused by road access. Any problems should be rectified before the ponds are commissioned, so that they do not become a routine maintenance problem

Pond maintenance is a vital part of pond management. The highest levels of maintenance will be required for earth lined primary ponds and final or evaporation ponds with low and seasonally variable effluent flow rates. Some form of maintenance will be required even for ponds with vertical concrete margins and sealed verges. Even those ponds of good design in favourable locations, with ideal effluent characteristics, must have adequate provision for people and resources to carry out a regular and defined maintenance program

Aspects of maintenance frequently overlooked include; the regular control and removal of vegetation on the margins or the pond verges, the regular removal of flotsam, alga mats or aquatic plants from the pond margins, and the repair of cracks and other failures that can allow increased soil moisture levels on or near the banks and subsequent vegetation growth.

For some ponds, a program of water level management may be adopted which alternately floods and strands marginal vegetation or flotsam. The form of possible maintenance will depend heavily on the pond design, effluent parameters, and staff experience.

Regular and adequate maintenance to prevent mosquito breeding is not common in many sewage treatment facilities. If there is any anticipation that proper maintenance will not be carried out regularly, a maintenance-free design should be chosen.

EFFLUENT DISPOSAL OR DISPERSAL

PROBLEMS

Many sewage treatment facilities give insufficient consideration to the disposal of treated effluent. It has been assumed that effluent after 'adequate treatment' is no longer a problem, and can therefore be left to run down the nearest flow line. In fact, this effluent often forms flooded, overgrown, stagnant pools that create very productive mosquito breeding grounds.

In some situations effluent has been directed into sand dunes or sandy situations in the belief that infiltration would provide a satisfactory disposal method. This is totally inadequate because the high organic loads of the effluent and resultant algae invariably seal against infiltration and result in extensive pooling of effluent.

Even after extended treatment in secondary and evaporation ponds, the resulting 'treated' effluent still retains a great capacity to breed mosquitoes. It still has relatively high nutrient levels that lead to high alga and vegetative growth, and can disrupt freshwater ecology including that of the fish predators of mosquito larvae. Even high quality tertiary treated effluent with low nutrient levels may be sufficient to cause pooling, ecological change, and mosquito breeding, if not disposed of adequately.

LARGE EVAPORATION PONDS

Evaporation ponds either of intentional or ad hoc' design have commonly been used as an effluent disposal method. Large evaporation ponds are rarely filled to capacity for the entire year, and in many instances are just bunded areas that store effluent against escape to other areas. Because of their large area, the variable inflow, and seasonal variations, large evaporation ponds can become shallow, flooded, swamps with dense weed and reed vegetation. Evaporation ponds that dry up and are then seasonally inundated by rain or effluent release can become breeding grounds for floodwater mosquitoes.

The aspects to be considered in designing large ponds to reduce mosquito breeding, include:

- initial and regular removal of all emergent vegetation within the evaporation area,
- levelling of the floor of the evaporation area,
- division of the evaporation area into a number of smaller areas,
- constructing a sloping floor to concentrate the water in a 'sink' area at the effluent entry point;
- concrete lining of the 'sink' area on the floor of the evaporation area and concrete lining of embankments.

However, incorporating some of these aspects into the design can be prohibitively expensive. The alternatives are to have a regular maintenance program, which could be more expensive in the longer term, or to choose a more suitable method of effluent disposal.

SMALL EVAPORATION PONDS

The use of a series of relatively small concrete lined evaporation ponds can be a very effective method of effluent disposal. The best designs incorporate a series of ponds that can progressively fill by gravity overflow. Such a system may be expensive to construct, particularly if the evaporation area required is relatively large. However, the method has the advantage of being relatively maintenance free and can cope with variations in effluent volume.

DISPOSAL TO THE SEA

Disposal direct to the sea or to a daily flushed tidal area is one of the most suitable methods for treated effluent disposal. It is important that the disposal outlet is to the open sea, or a large creek or river with considerable tidal movement, and that there are no deleterious environmental or public health concerns with the disposal site. Disposal at the lower end of a relatively long, narrow or tortuous tidal creek can result in effluent build up in the creek, which can be pushed by incoming tides higher up the creek line and overflow or pool in areas where mosquito breeding sites can develop.

Disposal onto large flat, inadequately flushed, tidal areas can create breeding sites not only for freshwater species of mosquitoes, but also for salt and brackish water species.

DISPOSAL TO RIVERS

The suitability of discharge to rivers depends upon the volume of flow in the river, the seasonal variability of flow, and the downstream effects of the disposal. When the flow in the rivers or creeks is small or subject to wide seasonal variation, this method can result in eutrophication or ecological and vegetation changes, which invariably lead to mosquito breeding.

DISPOSAL TO LAND

SPRINKLER DISPERSAL

This method has been relatively successful in areas where there have been particular problems with other disposal methods. It is most successful onto areas with well-developed stands of trees that are on soils of good permeability. In these situations, the final effluent can be automatically and periodically dispersed via a system of overhead sprinkler heads. Fire damage in natural vegetation can be rectified by the construction of an underground pipe system with steel uprights and metal sprinklers. Sprinkler irrigation can also be undertaken using small-volume under-tree micro sprinklers in plantation settings. However weed maintenance can be a problem with smaller systems, and is generally only suited to plantations or organized irrigation areas where regular maintenance is practical or cost effective.

Ideally, sites should be relatively flat but have adequate drainage to cope with rainy periods. Feeder lines to spray heads should be laid out along contours, rather than at right angles to contours, so that effluent pressures are equal and effluent will be retained in the lines after spraying rather than permitting continued flow to the lowest spray head. This will avoid creating semi permanent pools of effluent at the base of one sprinkler head.

The area required will depend upon the volume of effluent to be disposed, and the long-term absorption capacity of the soil and the vegetation. Precautions are required to ensure that effluent contaminated run off after rain episodes cannot pool in nearby depressions, flow lines or creeks.

Sprinkler dispersal can be used for tree and pasture growing or landscape watering, but the National Health and Medical Research Council Guidelines for the Reuse of Waste-water must be adhered to (NH&MRC, 1979). This can include fencing, adequate signs and chlorination. Tertiary chlorination can provide a high quality effluent for drip irrigation and recreational area watering but may need filtration.

If tertiary treated chlorinated effluent is held in open temporary storage ponds, the ponds should be constructed as for secondary sewage treatment or evaporation ponds. Alga and other microscopic growth will still occur and lead to marginal vegetation growth and ideal conditions for mosquito breeding. Reuse of tertiary treated effluent for landscape watering may require freshwater flushing of the distribution pipes immediately after effluent dispersal to prevent odour problems resulting from anaerobic action on the retained effluent in the pipes. Sprinkler disposal using high volume spray units has been successfully used in some areas, but potential problems include regularly moving the spray units, and over-watering, leading to pooling and rising water tables.

DRIP IRRIGATION DISPERSAL

Disposal by irrigation drip systems requires a high standard of effluent, usually with a tertiary chlorine treatment, to prevent drip blockage by algae. Drip systems can be used for both small or large-scale disposal, but is usually only suitable for plantation situations where the vegetation growth at each drip site can be practically and economically maintained. Drips held off the ground can reduce root blockages of the drip. Generally drip systems are only suitable for the dispersal of small volumes of effluent per unit area or periodic release, and are relatively expensive because of their high maintenance requirement.

SMALL FURROW IRRIGATION

This method is useful for relatively small volumes of effluent on sandy soil in low rainfall areas. A feeder channel is used to deliver effluent to a ploughed area of small furrows that slope gently away from the feeder channel. Disposal is by infiltration into the sandy soil. When infiltration becomes less efficient, the flow is directed to an adjacent ploughed area, and the original area is allowed to dry out and is re-ploughed.

This system requires a considerable amount of attention and maintenance, but has a low capital cost.

CHANNEL INFILTRATION

In this system, permanent infiltration channels are constructed and effluent flow is directed down a number of groups of channels, which are alternatively spelled and maintained. The method can be used on less porous soils than is possible for furrow irrigation. If this method is used for the irrigation of tree or bush crops, intensive monitoring of water tables and salinity levels is required to ensure viability of the crop in the long term. Problems with larger scale use have included high capital cost of infrastructure, high labour input, regular weed and erosion control in the channels, rising soil salinity, and elevated water tables.

FLOOD BAY IRRIGATION

The degree of land preparation for flood bay irrigation is usually considerable, as a system of correctly graded flood bays is necessary to allow for efficient flooding and to prevent pooling in or at the end of the flood bays. The bays are periodically or alternatively flooded by a distribution feeder channel, and the effluent is allowed to evaporate or infiltrate in the bays over a period of 3 - 4 days. This method has been used successfully to grow irrigated pasture and tree crops.

Problems with flood bay irrigation arise during rainy periods, when extended flooding of the bays with nutrient enriched water can result in mosquito breeding. Small flood bay systems are suited for relatively small effluent volumes, arid areas where surface evaporation is high, or in situations with good soil infiltration. Some of the problems with flood irrigation systems can be reduced by using an automatic siphon and a multi discharge distribution channel to release effluent periodically and evenly over the flood bay. Generally flood systems require at least two separate bays so maintenance and spelling can be carried out.

BIOLOGICAL CONTROL

Biological control, though not generally efficient or applicable to primary ponds, can be a very efficient means of controlling mosquito larvae in secondary and evaporation ponds.

The major biological control agents are fish, aquatic beetles and aquatic bugs. Fish can control mosquito larval numbers directly by eating the larvae, or indirectly by reducing algae which provide protection from other predators or wave action. Fish are usually only suitable for the higher oxygenated waters. Several native species have shown promise including the rainbow fish (*Melanotaenia sp*), Pacific blue eyes, and gudgeons. Wildlife regulations must be observed when considering using fish as biological control agents.

Marginal vegetation such as couch grass and reeds should be eliminated or kept to a minimum, so that fish can have physical access to the mosquito larvae. Actively growing reeds with upright stems may not restrict fish access. However, when these reed species die or lodge over, they prevent physical access for the fish and enable mosquito breeding. Vegetation can be removed by slashing, weedicide or burning (see Whittle 1993), and can have a major reduction on larval numbers by improving biological control and removing physical shelter for larvae.

Aquatic beetle larvae (family Carabidae) and aquatic bugs (family Belostomatidae) can be very efficient mosquito larvae predators in secondary and evaporation ponds. The aquatic bugs are able to live in higher organic water than the aquatic beetle larvae, and can be present in enormous numbers. Again, physical impedance by thick vegetation at the margins will reduce the effectiveness of these predators. Vegetation problem areas should be eliminated by physical removal or weedicide application. Insect predators can achieve almost total control of mosquito larvae in sewage ponds of suitable water quality, and steep or vegetation free margins.

CHEMICAL CONTROL

The aim of chemical control of mosquito larvae should be to apply the minimum amount of insecticide to prevent the production of adult mosquitoes. Chemical control should not be used as a long-term strategy in sewage treatment areas, in order to avoid insecticide resistance and unwanted effects on non-target organisms. Weedicide application to mosquito breeding sites often provides more efficient short to medium term control and can greatly reduce insecticide requirements. However, it may be necessary to apply insecticides during the initial operational period or when proper maintenance has not been carried out. The insecticides of choice to control mosquito larvae in sewage ponds and effluents are temephos, methoprene or *Bacillus thuringiensis var. israelensis (Bti*). Correct rates for temephos must be strictly adhered to, as overdosing can kill fish and other aquatic insects.

MOSQUITO SAMPLING

Regular inspections should be carried out in sewage ponds and their effluents to determine whether mosquito larvae are present and to determine the necessity for weed or chemical control. Chemical control with temephos, methoprene or *Bti* may be necessary at weekly or longer intervals. The presence of pupae indicates that control should have been conducted at shorter intervals. If only first and second instar larvae are present, then either biological control is quite efficient, or the mosquitoes have just started to breed in that area, and continued monitoring is necessary.

Mosquito larval or pupa samples can be collected by dipping into sheltered vegetated zones with a soup ladle. Any larvae collected should be stored in small vials with 70 % alcohol or methylated spirits, together with information on collection locality, site, date and collector.

Adult specimens that have been collected as they bite or harbor can be killed by freezing and packed loosely in tissue paper in a small box, together with all the details of collection. Larval, pupa and adult specimens should be sent to an entomologist for identification or verification.

Chironomid midge pupae or adults are often in very high numbers near sewage treatment facilities and are frequently mistaken for mosquitoes. Their presence has often resulted in control programs being instituted where none was necessary.

SUMMARY

In the past, the design of sewage treatment and effluent disposal facilities was usually dictated solely by engineering and microbiological principles. Little attention was paid to the possibility of breeding mosquito populations close to habitations, and the resulting potential pest and public health problems. Appreciation of this potential risk, followed by the application of simple design principles and adequate maintenance, can reduce these problems. Biological control can often be used to supplement good design features. Chemical control can be used under certain situations but should be reserved as a short-term remedy until permanent solutions or maintenance measures can be implemented.

REFERENCES

Brisbane City Council Water Sensitive Urban Design Practice Note Series. Practice Note 6 Constructed Wetlands.

Brogan B, Whelan PI, Carter J and Lamche G. (2002). 'Rectification and control practices in major salt marsh mosquito breeding site, Darwin, NT'. The Northern Territory Disease Control Bulletin 9:4:16-21.

Chironomid midge and mosquito risk assessment guide for constructed water bodies, Midge Research Group of WA August 2007.

Dale P and Morris C (Editors), Panel of authors including Whelan P.I. (2002). 'Drainage considerations for mosquito control'. In 'Australian Mosquito Control Manual', Mosquito Control Association of Australia, revised edition 2002.

Dale PER, Chapman H, Brown MD, Ritchie SA, Knight J, Kay BH,(2002). Does habitat modification affect oviposition by the salt marsh mosquito, Ochlerotatus vigilax (Skuse) (Diptera: Culicidae)? Australian Journal of Entomology 41: 49-54.

Department of Medical Entomology, University of Sydney. Freshwater Wetlands (natural and constructed). Mosquito production and management.

Jacups S, Warchot A and Whelan PI (submitted) 'Anthropogenic ecological change and impacts on mosquito breeding and control strategies in salt-marshes Northern Territory Australia.

'Guidelines for preventing biting insect problems for urban residential developments or subdivisions in the Northern Territory', Medical Entomology, NT Department of Health and Community Services 1997.

Kurucz N, Whelan PI and Porigneaux P. (2002). 'Mosquito control in Ilparpa Swamp – A Big Step Forward'. The Northern Territory Disease Control Bulletin. 9:1:22-23.

Kurucz N, Whelan PI, and Daly C.(2003). 'Mosquito control at Hickey's Lake, Katherine, NT'. Bulletin of the Mosquito Control Association of Australia. 15:1:22-25.

NH&MRC (1979), 'Guidelines for Re-use of Wastewater, Australian Water Resources Council, National Health and Medical Research Council, Department of National Development, Canberra.

Policy for the design of off site sewage ponds and the disposal or reuse of sewage pond effluent. Department of Health and Community Services, June, 1997.

Pratt, H D., Littig, K S., & Barnes, R C. (1972), 'Mosquitoes of public health importance and their control', United States Department of Health, Education & Welfare, Centre for Disease Control, No. 72 - 8140, Atlanta, Georgia, USA

Russell, R C. (1986), 'Dispersal of the arbovirus vector Culex annulirostris Skuse (Diptera: Culicidae) in the Murray Valley of Victoria Australia', Gen App Ent, vol. 18, pp. 5-9.

Russell, R. C. (1999), 'Constructed wetlands and mosquitoes: Health hazards and management options – An Australian perspective', Ecological Engineering 12 (1999) 107 – 124.

Turner PA, Streever WJ, (1999). Changes in productivity of the saltmarsh mosquito, Aedes vigilax (Diptera: Culicidae), and vegetation cover following culvert removal. Australian Journal of Ecology 24: 240-248

Turner PA, Streever WJ, (1997). The relationship between the density of Aedes vigilax (Diptera : Culicidae) eggshells and environmental factors on Kooragang Island, New South Wales, Australia. Journal of the American Mosquito Control Association 13: 361-367.

Whelan, P I. (1981), 'The vulnerability and receptivity of the Northern Territory to mosquito borne disease', Transactions of the Menzies Foundation Vol 2, Living in the North, pp. 165-171.

Whelan, P I. (1984), 'Mosquitoes of public health importance in the Northern Territory and their control', Northern Territory Department of Health, Darwin.

Whelan PI. (2007). 'Mosquito Control in Leanyer Swamp'. Northern Territory Disease Control Bulletin. 14:2:19-20.

Whelan PI. (2007). 'Mosquito Vector Control in the Northern Territory'. Northern Territory Disease Control Bulletin. 14:2:12-18.

Whelan, P. 1, (1988), 'Mosquito breeding and sewage treatment in the Northern Territory', 'Water', J Aust Water & Wastewater Assoc, vol 15, no. 5, pp. 34-37.

Whelan, P. I. (1994), 'Construction practice near tidal areas in the NT', Bulletin of the Mosquito Control Association of Australia, Vol. 6, No. 1, pp. 18-39.

Whelan PI. (1990). Urban stormwater drainage and its effects on mosquito breeding habitats in an intertidal creek at Casuarina, Darwin NT', Bull. Mos. Control Assoc. Aust. 2:1:12-28.

Whittle RK, Linthicum KJ, Thande PC, Wagati JN, Kamau CM, Roberts CR, 1993. Effect of controlled burning on survival of floodwater Aedes eggs in Kenya. Journal of the American Mosquito Control Association 9: 72-77.

11. THE CULTURAL CONTROL OF MOSQUITOES IN AUSTRALIA

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INTRODUCTION

There are certain aspects of the way we live that enables person/mosquito contact to occur. Much of the person/mosquito contact can be reduced by changing the patterns of doing things or by taking certain precautions. Many of these cultural aspects of mosquito control can be undertaken by the householder. Cultural control must rely on public education. The public must be informed of where mosquitoes breed, how to control them around their premises, and how to practice mosquito avoidance and personal protection.

SOURCE REDUCTION AROUND PREMISES

- Eliminate water holding containers such as tins, tyres, pot plant drip trays, plastic sheeting, plastic containers, palm fronds, disused machinery and jars. These can be punctured, burned, buried, filled, stored under cover or broken.
- Inspect and clean all roof gutters at the start of and in the middle of the rainy season.
- Checks all animal water containers weekly for larvae. They should be emptied, scrubbed out, and refilled once a week.
- Prevent leaking taps or irrigation systems which can maintain semi-permanent ground pools.
- Replace water in plant propagation containers every week, or use wet sand to prevent a free water surface.
- Maintain household swimming pools or empty them when not in use for considerable periods.
- Screen all water tanks and septic tanks inlets, outlets and inspection points to prevent the entry of mosquitoes.
- Flush disused toilets once a week.
- Avoid over-watering lawns. This can lead to high water tables and ground pools, or cause run off to storm water drains and hence create permanent pools at drain end points.
- Treat unsealed water storages such as water tanks, drums, water features, or other containers with methoprene pellets, mosquito larvicide films, or a small volume of kerosene plus oil of cloves in a 10:1 ratio, on a regular basis until sealed or otherwise rectified.
- Remove weeds and grass from drains to enable them to drain freely.
- Keep fish ponds and tanks stocked with fish.
- Empty water filled flower vases, ornamental urns, pots and bird baths once per week.
- Put a teaspoon of salt in any ant traps in the house.

SELF PROTECTION FROM MOSQUITOES

Mosquitoes can reach sufficient numbers in various localities to be considered serious pests. The bites themselves can be painful and extremely annoying, and people suffer varying degrees of reaction to bites (Lee 1975). However the possibility of the spread of various diseases by their blood sucking habits to either humans or animals is a more serious outcome.

Mosquitoes can carry viruses such as Murray Valley encephalitis, Kunjin, Ross River, and Barmah Forest viruses which cause human disease (Russell 1995).

Female mosquitoes bite to take blood from their host, which is necessary for the development of eggs.

Mosquitoes show considerable variation in their preference for hosts. Some species feed selectively on cattle, horses, marsupials, amphibians, birds or humans, while other species are relatively indiscriminate feeders.

The time of feeding varies for different species. Many mosquitoes feed just after sunset while others are more active at other times including late in the night, in the late afternoon, or in the early morning.

The place of feeding by mosquitoes is varied. Some species, such as the brown house mosquito, readily entering houses to feed on people, while others will only bite people outdoors.

When a mosquito bites, fine stylets sheathed in the proboscis are inserted into the skin. Blood is sucked up through one of the channels in the stylets, while saliva is injected down an adjacent smaller channel. This saliva contains substances that the human body recognizes as foreign and this often stimulates a histamine like bite reaction. Sometimes the saliva can contain viruses or other pathogens that can cause disease.

Some people can become very sensitive after being bitten and suffer various lumps, blistering, or a general reaction from further bites. The bites may itch for days, producing restlessness, loss of sleep and nervous irritation. Scratched bites can lead to secondary infections and result in ugly scars. However some people become tolerant to particular species and suffer little after-effects from repeated bites.

Mosquitoes create problems in the enjoyment of outdoor activities, causing a reluctance to enter certain areas after sundown or forcing people to be confined to mosquito-proof areas at certain times of the year. Personal protection and avoidance measures can offer considerable protection from bites, as well as offering protection against mosquito-borne disease.

PLAN WHERE YOU LIVE

Residential areas should be planned with mosquito breeding sites in mind. For example in Darwin, new urban residential areas must be 1.6 km away from extensive and uncontrolled or uncontrollable mosquito breeding sites. These buffer zones aim for a physical separation from significant mosquito breeding sites. Continuous tree belts or smaller breeding sites between residential areas and large breeding areas should be avoided or rectified, as these provide shelter and access routes into residential areas.

AVOIDANCE

A sensible precaution to prevent mosquito attack is to avoid areas that are known to be near breeding areas or have high mosquito activity.

The upper high tide areas near tidally influenced creeks or low lying areas, particularly in salt marsh habitats, can be significant sources of salt marsh mosquitoes, particularly *Aedes vigilax*, *Aedes camptorhynchus* and various other pest mosquitoes (Russell 1995). The period of high salt marsh mosquito activity is usually during the late spring or summer in temperate latitudes and in the late dry season and early wet season in tropical latitudes. Generally they are prevalent for one to two weeks after the highest tides of the month or appreciable early summer or early wet season rain. Dense tree or shrub vegetation near the breeding sites should be avoided during the day over this period. Pest and disease problems during the

evening and night often occur within 1 and up to 3 km of productive breeding sites (Whelan, Merianos et al. 1997).

Areas of high mosquito activity include the large seasonally flooded areas associated with rivers or drainage lines, flooded coastal swamps, extensive reed swamps and lagoons, ill defined or poorly draining creeks, extensive irrigation areas, and wastewater disposal facilities. Densely shaded areas near these habitats should be avoided during the day, and accommodation areas should ideally be at least 2-3 km from extensive areas of these habitats.

If camping or selecting house sites near creeks, rivers, lagoons, or other vegetated water bodies, choose localities of the water body which have steep margins or little marginal emergent vegetation, have swiftly running water with little marginal pooling or vegetation, or do not arise from or empty into a nearby swamp area. Exposed beaches or cliffs away from mangrove or estuary areas are preferred sites to avoid mosquitoes. In more inland areas, locations on hills or rises at least 3 km from ill defined drainage lines, poorly flowing creeks and seasonally flooded areas should avoid the worst mosquito problems.

In residential areas, a local source of mosquitoes may be the cause of the problem. Check nearby potential artificial sources of mosquitoes such as disused swimming pools, receptacles such as tyres and drums, blocked roof gutters, old fishponds, or localized ponding of drains. Sites with mosquitoes breeding can be rectified by physically removing the source, by draining or filling, or through the use of insecticides.

SCREENING

The best method of avoiding attack at night is to stay inside insect-screened houses. Screens can be made of galvanized iron, copper, bronze, aluminium or plastic. Near the coast, iron or copper screens are not recommended because of the corrosive action of salt sprays.

Screens should be of the correct mesh, fit tightly and be in good repair. Mosquitoes frequently follow people into buildings and for this reason, screen doors should open outward and have automatic closing devices. Insecticides such as alpha-cypermethrin, lambda-cyhalothrin, deltamethrin, or bifenthrin sprayed on or around doors and screens can give added protection against mosquitoes, but care is needed as some insecticides affect screens.

It is advisable to use a tent that is mosquito proof when camping near potential mosquito breeding areas.

MOSQUITO NETS

Mosquito nets are useful when camping or in unscreened houses near mosquito breeding areas. White netting is best as mosquitoes accidentally admitted into the net are easily seen and killed. The net should usually be suspended over the bed and tucked under the mattress. An aerosol knockdown (not residual) pyrethroid spray can be used to kill mosquitoes that enter the net. Care is needed not to leave exposed parts of the body in contact with the net, as mosquitoes will bite through the net. Nets can be made more effective by impregnation with permethrin (Lines et al. 1985).

MOSQUITO PROOF CLOTHING

Head nets, gloves and boots can protect parts of the body that are not covered by other clothing. Head nets with 1-1.5 meshes to the centimetre are recommended for good visibility and comfort, and additional treatment of the net with a repellent will discourage mosquito attack. Thick clothing or tightly woven material generally offers protection against bites. Light coloured, long sleeved shirts and full-length trousers are recommended. For particular risk areas or occupations, protective clothing can be impregnated with permethrin to give added

protection (Burgess et al. 1988). Sleeves and collars should be kept buttoned and trousers tucked in socks during biting insect risk periods. Protection is very necessary near areas of salt marsh, mangroves, or large fresh water swamps where the various species of mosquitoes may be very abundant during the day in shaded situations, as well as at night.

REPELLENTS

Relief from biting insect attack may be obtained by applying repellents to the skin and clothing (Schreck et al. 1984). Many repellents affect plastics and care is needed when applying them near mucous membranes such as the eyes and lips.

Repellents with the chemical diethyl toluamide (DEET) at between 5 and 20 percent (50-200g/litre) or picaridin give the best protection. Brands of repellents, such as Aerogard, which are formulated to repel flies are not efficient against mosquitoes. Brands such as Rid, Tropical Strength Aerogard, Bushman, Muskol, or Repel are more effective. Brands and formulations such as botanicals consisting of eucalyptus oil, tea tree oil, sandalwood or other plan oils offer only partial protection.

Application of repellent over large areas of the body or on extensive areas of children is not recommended, particularly those repellents with high concentrations of DEET. Protection from mosquito penetration through open weave clothes can be obtained by applying a light application of aerosol repellent to the exterior of clothing. Repellents should be supplementary to protective clothing and should not be regarded as substitutes.

Personal repellents are available as sprays, creams or gels. The creams or gels usually last longer than the aerosol formulations. Repellents can prevent bites from 2 to 4 hours, depending on the repellents, the species of biting insect, or the physical activity of the wearer.

There are some new metofluthrin and allethrin vapour active pyrethroid spatial repellents on the market where there is passive or active evaporation from impregnated strips or pads. These have been shown to be very effective in preventing landing or biting of many species of mosquitoes, even in outdoor situations within a close (up to 3-5m) surround of the devices, or within rooms in more enclosed areas. Devices on the market at present include gas powered devices and mosquito lanterns using a candle and allethrin pads.

Electronic insect repellers based on claims of ultrasonic or audible sounds acting as a repellent do not offer any protection against mosquitoes. They are based on a false premise and have been found to have no repellent effect under scientific testing (Curtis 1986). Electronic ultrasonic repellers do not repel mosquitoes and should not be relied upon for personal protection (Mitchell 1992).

Plants with reported insecticidal properties such as neem trees and the 'citrosa' plant have not been shown to act as mosquito repellents just by growing in the vicinity of people (Mitchell 1992, Matsuda et al. 1996). Growing or positioning these plants near evening activity areas will not prevent mosquito attack.

ANIMAL DIVERSION

Camping upwind near congregations of stock or domestic animals will serve to divert mosquitoes or biting midges to alternative hosts. Similar considerations can be made when planning residential sites and animal holding areas in a rural situation. Dogs or other mammals of darker colour tend to attract some species of mosquitoes more than lighter colours and can divert some pest problems from people in close vicinity in outdoor situations in the evening.

LIGHTING DIVERSION

Many mosquito species are attracted to light. This can cause pest problems in unscreened houses or when camping. The use of yellow incandescent bulbs or yellow fluorescent tubes rather than white light will reduce the attractiveness of lights to insects, with red lights offering even more protection. An incandescent or ultra violet light placed at a distance from a house or camp can serve to attract insects to an alternative area. This is more effective if the light is closer to the breeding site, between the breeding site and the accommodation area. The attractive lights should not be close to accommodation or directly down wind of accommodation areas. Light proof curtains or lights screening can be very effective in reducing the attraction of biting insects to areas that are illuminated at night.

ADULT INSECT CONTROL

If mosquitoes have entered a screened area, they can be knocked down with pyrethroid aerosols. Care should be taken by reading the label to ensure only knockdown aerosols suitable for spraying in the air are used in proximity to people or food.

Other devices that can be effective at killing and/or repelling biting insects include mosquito coils containing various pyerthroids (Charlwood & Jolley 1984), electric insecticide pads containing allethrin or bioallethrin, or passive vapour emanators containing metofluthrin. These devices are generally more effective in relatively enclosed areas such as patios or inside open unscreened buildings or where there are only slight breezes. They should generally be backed up with other measures such as suitable protective clothing or personal repellents.

Large scale adult biting insect control can be achieved for short terms (hours) by using portable or industrial fog generators, backpack misters, or heavy duty ultra-low-volume aerosol generators to knock down active adult insects. The insecticides of choice in these machines are maldison or bioresmethrin. Control relies on good access, open vegetation, small droplet size and light breezes in the direction of the breeding or harbouring sites. Application should only be during the peak biting insect activity period of those insects actually causing the problem. which is usually the late evening and early night.

Application of short term residual insecticides such as maldison, permethrin or other pyrethroids sprayed on surfaces or nearby vegetation can sometimes give short term (1 day) relief when large numbers of mosquitoes or biting midges are present near accommodation or outdoor use areas (Helson & Surgeoner 1985). These can be applied according to label recommendations with the aid of a garden sprayer. There are some aerosol products available as outdoor yard or patio repellents. Control will only be temporary and re-invasion will usually occur within hours or from one to a few days, depending on the species, nearby vegetation, proximity to breeding sites, environmental conditions and times of activity of the pest species.

Longer term residual insecticides such as bifenthrin, alpha-cypermethrin and lambdacyhalothrin can give effective barrier control up to 4 to 6 weeks. Bifenthrin sprayed on low vegetation, lawns, fences, and walls where mosquitoes harbor has shown promising reductions of between 94 to 99% of mosquitoes for periods of 4 weeks under tropical conditions (Standfast et al 2003) and similar protection has been shown for mosquitoes for up to 6 weeks for both bifenthrin and lambda-cyhalothrin (Trout et al 2007).

INSECTOCUTORS AND INSECT TRAPS

Electric insect insectocutors and other trap or killing devices that use an attracting light or carbon dioxide have been claimed to clear areas of biting insects and thus protect people. These claims have not been substantiated in outdoor situations with people nearby. While trap devices can attract biting insects, as well as a range of other insects, these devices can not be

relied on for protection from biting insect attack (Mitchell 1992). When used in outdoor situations it is possible that they can increase local problems by attracting insects to the vicinity of people. Attractive odours and carbon dioxide emitted by humans then divert the insects from the trap device to the people.

TREATMENT OF BITES

Relief from bites and prevention of secondary infection can be obtained by the application of various products, either to the skin or internally. The effectiveness of various products is variable, depending on individual reaction. Skin application products include proprietary products such as Eurax, Stingose, Medicreme, Katers lotion, Dermocaine and Paraderm creme, and non-proprietary products such as tea tree oil, papaya or paw paw ointment, eucalyptus oil, aloe vera gel, ice, or methylated spirits.

Ice packs applied to the general bite site will usually give immediate relief for painful and itchy bites, and reduce the swelling or blisters resultingfrom mosquito bites. The sooner the ice pack is applied after bites or reactions, the better the relief, and can often avoid more intense reactions. Paw paw ointment has been claimed to offer similar relief in some people.

Other products for internal application for more general symptoms include antihistamine products such as Phenergan, Telfast and Vallergan. Check with your doctor or pharmacist for products and the latest product and safety information.

EMERGENCY BITING INSECT PROTECTION

There are a number of emergency measures that can be taken when exposed to biting insects with no available protection. Sheltering downwind next to smoky fires can offer considerable protection. Burning dung or aromatic and oil producing foliage from plants such as *Hyptis* (horehound), *Calytrix* (Turkey bush), *Melaleuca* species (Paper bark) and *Eucalyptus* species (gum trees) can make the smoke more effective. A small native plant *Pterocaulon serrulatum* (warnulpu) which has sticky strongly aromatic leaves, are burnt or the moist leaves rubbed on the skin by Aborigines in the Katherine district to repel mosquitoes (Aborigines of the NT 1988). Climbing relatively high trees or choosing locations exposed to the wind can offer protection from some species.

Other emergency protection measures include coating the skin with mud, rubbing exposed areas with the leaves of certain plants such as eucalypts or tea-trees that contain volatile oils, or burying yourself in shallow sand with some form of head protection. If all else fails, keep running. The best form of protection, and the most comfortable, require an awareness of the potential problems and adequate preparation.

REFERENCES

- Burgess N., Carter S., Dodd G., & Shirley C. (1988), 'Permethrin fabric treatment for the protection of personnel from biting insect and other arthropod attack', International Pest Control, vol. 30, no. 6.
- Charlwood J. D., & Jolley D., (1984), 'The coil works (against mosquitoes in Papua New Guinea), Trans Roy Soc Trop Med Hyg, vol. 78.
- Curtis C. F. (1986), 'Fact and fiction in mosquito attraction and repulsion', Parasitology Today, vol. 2, no. 11.
- Helson B. & Surgeoner G. (1983), 'Permethrin as a residual lawn spray for adult mosquito control', Mosquito News, vol. 43, no. 2.
- Lee D. J. (1975), 'Arthropod bites and stings and other injurious effects', School of Public Health & Tropical Medicine, University of Sydney.
- Lines J. D., Curtis C. F., Myamba J., Njau R. (1985), 'Tests of repellent or insecticide impregnated curtains, bednets and anklets against malaria vectors in Tanzania', WHO VBC/85.920.
- Matsuda B. M., Surgeoner G. A., Heal J. D., Tucker A. O., Maciarello M. J. (1996), 'Essential oil analysis and field evaluation of the citrosa plant Pelargonium citrosum as a repellent against populations of Aedes mosquitoes', Journal of the American Mosquito Control Association, vol. 12, no. 1, pp. 69-74.
- Mitchell L. (1992), 'Mythical mosquito control', Wing Beats, vol. 3, no. 2, Florida Mosquito Control Association.
- Russell R. C. (1995), 'Arboviruses and their vectors in Australia: an update on the ecology and epidemiology of mosquito borne viruses', Review of Medical Veterinary Entomology, vol. 83, no. 4.
- Schreck C. E., Haile D. G., Kline D. L. (1984), 'The effectiveness of permethrin and deet alone or in combination for protection against Aedes taeniorhynchus ', Am J Trop Med Hyg, vol. 33, no. 4.
- Standfast H, Fanning I, Maloney L, Purdie D and Brown M.(2003), 'Field evaluation of Bistar 80SC as an effective insecticide harbourage treatment for biting midges(Culicoides) and mosquitoes infesting peridomestic situations in an urban environment. Bull.Mosq.Control Assoc Aust. Vol. 15 (2).
- Trout RT, Brown GC, Potter MF and Hubbard JL. 'Efficacy of two pyrethroid insecticides applied as barrier treatments for managing mosquito populations in suburban residential properties. J Med Entomol. 44(3): 470-477.
- Whelan P. I. (1990), 'Biting midge investigations near Darwin and their implications for urban planning'. Proceedings of the National Conference on Biting Midge, Surfers Paradise, February 1990.
- Whelan P. I., Hayes G., Montgomery B. L. (1997), 'Biting midge surveillance in Darwin harbour, Culicoides ornatus (Diptera: Ceratopogonidae) abundance and dispersal', Proceedings of the Seventh Symposium "Arbovirus Research in Australia", Second Conference Mosquito Control Association of Australia, Surfers Paradise.
- Whelan P. I., Merianos A., Hayes G., & Krause V.(1997), 'Ross River virus transmission in Darwin, Northern Territory, Australia', Proceedings of the Seventh Symposium "Arbovirus Research in Australia", Second Conference Mosquito Control Association of Australia, Surfers Paradise.

Whelan P 2004. 'Personal protection from mosquitoes and biting midges in the NT'. Northern Territory Disease Control Bulletin. 11:2 p. 18
12. ENVIRONMENTAL CONSIDERATIONS IN MOSQUITO MANAGEMENT

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Mosquitoes breed in association with wetlands (estuaries, lakes, streams), in man-made situations and in disturbed natural environments. As larvae they have an important role in breaking down detrital material and in some cases, they are predators of larvae of mosquitoes and other insects. In turn, mosquito larvae are eaten by other invertebrates, fish and birds. As adults, mosquitoes provide food for other animals (e.g. birds and frogs) and they are pollinators of flowering plants.

As a consequence of their importance in food chains, the removal of larval or adult mosquitoes will have impacts on other species in the local environment. These other animals may be permanent inhabitants or occasional visitors such as migratory waterbirds that visit the area because of the abundance of food (perhaps including mosquito larvae). The choice of management techniques will determine the extent of the impact on the environment and may also influence whether there are indirect economic consequences (e.g. by damaging nursery areas for fish and crustaceans, thereby affecting the sustainability of the local fisheries industry).

Mosquito managers must be aware that official approval may be required to undertake mosquito management action in many wetland environments. It is advisable for managers to investigate the requirements of the relevant environmental agencies as a first step in any mosquito management program to be undertaken in the natural environment.

AVOIDING THE NEED FOR MOSQUITO MANAGEMENT

Mosquito management is only necessary if people and mosquitoes come into contact, either in residential or recreational areas within dispersal range of mosquito breeding habitats. While recreational activities can be abandoned if mosquito numbers are unacceptably high, the considerations relating to residential location are more complex.

Good town planning is the key to minimising this problem and therefore it is important that proximity to mosquito breeding sites is taken into consideration when approving planning applications. If inappropriately-sited development is approved, residents find that they are exposed to mosquito nuisance and disease, and make demands of local and State governments to fund expensive mosquito management programs. Sadly, the wetland environment, which the residents were attracted to in the first place, can then be compromised by mosquito management having to be undertaken. There is also the very real possibility that mosquito management programs will be ineffective in some years due to repeated tides and other circumstances, resulting in residents being exposed to intolerable levels of mosquito nuisance. Also, currently-effective mosquito control agents may become ineffective over time due to the development of chemical resistance and therefore, the long-term effective control of mosquito populations may become increasingly challenging.

Mosquito avoidance by individuals should also be encouraged. Leisure activities should be planned for times when mosquitoes are least active. Personal preventive measures (i.e. wearing protective clothing, using personal repellents) may remove the need to carry out environmentally and economically unacceptable mosquito management activities in some situations. Appropriate signage that warns of seasonal mosquito and mosquito-borne disease activity may be appropriate in high-risk locations and/or in locations where mosquito control is not undertaken.

DETERMINING THE NEED FOR MOSQUITO MANAGEMENT

A good understanding of local conditions is crucial to making timely, environmentally and economically responsible decisions about management activities. Regular monitoring of larval and adult mosquitoes is essential and will allow the determination of whether a response is really necessary. Environmental and public health considerations will help to determine the most appropriate management options. Working with environmental managers will minimise unacceptable impacts to non-target species and to irreplaceable wetland environments.

ENVIRONMENTAL POLICIES AND LEGISLATION

Many wetland habitats have an official conservation classification. It is important that mosquito managers ascertain the international, national and regional level of significance of wetlands in their areas so that any necessary approvals for mosquito management activities can be sought from the appropriate environmental authorities. To fail to do so could have legal implications.

INTERNATIONAL OBLIGATIONS TO WETLANDS

The Ramsar Convention (Convention on Wetlands of International Importance) is an international treaty for the conservation and sustainable utilization of wetlands which was signed in 1971. The treaty uses a set of criteria for identifying Wetlands of International Importance (<u>http://www.ramsar.org/cda/en/ramsar-about-sites-criteria-for/main/ramsar/1-36-55%5E20740_4000_0</u>) and now includes 2,131 sites. A list of current Ramsar protected wetlands can be found at http://www.environment.gov.au/cgi-bin/wetlands/alphablist.pl.

A number of wetland sites are also protected under bilateral agreements between the Australian Government and countries within East Asia. These agreements have been established to conserve migratory birds and currently include three agreements:

- JAMBA Japan-Australia Migratory Bird Agreement (<u>http://www.austlii.edu.au/au/other/dfat/treaties/1981/6.html</u>)
- CAMBA China-Australia Migratory Bird Agreement (<u>http://www.austlii.edu.au/au/other/dfat/treaties/1988/22.html</u>); and
- ROKAMBA Republic of Korea-Australia Migratory Bird Agreement (http://www.austlii.edu.au/au/other/dfat/treaties/2007/24.html).

NATIONAL OBLIGATIONS TO WETLANDS

From a National perspective, there is the Directory of Important Wetlands in Australia that currently lists 851 wetlands that have qualified as being nationally important. The Directory lists the currently recognised wetlands of significance and builds knowledge of the variety of wetlands and associated flora and fauna (<u>http://www.environment.gov.au/water/publications/environmental/wetlands/directory.html</u>).

The National Heritage List has been established to list locations of "outstanding heritage significance to Australia" and includes natural, historic and indigenous places of heritage value. A number of wetland sites have been established under the National Heritage List (<u>http://www.environment.gov.au/heritage/about/national/index.html</u>). These sites are required to have management plans and heritage strategies in place and mosquito management needs to meet these goals.

However, the key piece of legislation to protect the environmental at a National Level is the Environmental Protection and Biodiversity Conservation Act (EPBC Act) 1999

(<u>http://www.environment.gov.au/epbc/</u>). This Act allows the Australian Government to join forces with the States and Territories in a national scheme for the protection of matters of national environmental significance. The objectives of the EPBC Act are to:

- Protect the environment, particularly areas of national significance;
- Conserve biodiversity;
- Provide a streamlined assessment and approval process;
- Enhance protection of natural and cultural significance;
- Control the international movement of wildlife; and
- Promote ecologically sustainable development through conservation.

The EPBC Act will be investigated in more detail below.

WETLAND PROTECTION IN WESTERN AUSTRALIA

Within each State and Territory, local legislation may also inhibit mosquito management practices. In Western Australia (WA), the Department of Parks and Wildlife (DPaW) should be contacted for advice on the conservation status of wetlands where mosquito management activities are proposed.

Regionally significant wetlands may be covered by a range of legislation, policies or reports. Examples of this for Western Australia include the:

- Bush Forever Policy which builds a framework to ensure the protection and management of bushland in the Perth Metropolitan Region and to provide policy for landuse planning and decision-making (<u>http://www.planning.wa.gov.au/publications/</u>5911.asp);
- Environmental Protection Agency (EPA) "System" Conservation Reserves The EPA divided WA into 12 "Systems" and has assessed each for its unique values, setting aside areas according to their floral, faunal, geological and recreational values.
- Environmental Protection Swan Coastal Plain Lakes Policy (1992) was developed to
 protect the environmental values of lakes across the Swan Coastal Plain. The policy
 states that filling, draining, excavating, polluting and clearing of these lakes an offence
 unless authorised by the EPA. The Policy can be found at: <u>http://www.epa.wa.gov.au/
 epadoclib/1090 EPP_SCPL92.pdf</u>.

ENVIRONMENTAL PROTECTION AND BIODIVERSITY CONSERVATION ACT 1999

The Commonwealth *Environmental Protection and Biodiversity Conservation Act (EPBC Act)* came into force on 16 July 2000. It has implications for mosquito management programs, whether they involve physical site modifications or the application of chemical mosquito control agents. The website of the Commonwealth Department of Environment and Heritage should be consulted for information about the *EPBC Act*: www.environment.gov.au/epbc

The EPBC Act establishes an environmental assessment and approval system that is distinct from the State system. It provides that a person must not impact on a matter of national environmental significance, except where certain processes have been followed and/or certain approvals obtained. Proponents must seek a determination from the Commonwealth Minister for the Environment and Heritage on the status of their proposal. State governments are not able to provide a determination, but the WA Environmental Protection Authority should be approached in the first instance for advice on how to proceed.

Wetlands of international importance (Ramsar wetlands) are one of the matters of national environmental significance under the EPBC Act. Actions taken outside the boundaries of a Ramsar wetland may still require approval if there could be an impact on a Ramsar-listed wetland.

Existing projects

Any project that commenced and was authorised under Commonwealth or State law prior to 16 July 2000 does not require approval under the EPBC Act. However, an enlargement, expansion or intensification of an existing project will be treated as a new project and will require approval.

Application process

Applications for approval must be referred to the Commonwealth Minister for the Environment and Heritage who will decide on the appropriate assessment approach to be undertaken and determine whether the proposed project can be approved on the basis of the assessment. Local and State Government agencies are under no obligation to ensure that any approvals they may grant have also received the necessary EPBC Act approval. **The responsibility lies with the proponent.** It is suggested that initial inquiries are made to the WA Environmental Protection Authority.

POTENTIAL IMPACTS OF BIOLOGICAL CONTROL

A number of organisms have been investigated for the purpose of managing mosquito larvae including bacteria, protozoa, fungi, nematodes, predatory invertebrates and fish. However, the only agents that show real prospects for biological control are bacteria and fish.

The bacterial "bio-larvicides" Bacillus thuringiensis var. israelensis (*B.t.i.*) and Bacillus sphaericus (*B.s.*), are highly target specific and where larviciding is to be used, should be considered as the first choices whenever possible.

Fish that eat mosquito larvae are an effective long-term control in man-made situations such as domestic water containers and artificial ponds. Native fish that can be considered for domestic situations include pygmy perch, spangled perch, western rainbow fish and guppies. However, if introduced fish are to be used, take care to avoid accidental escape of the fish into natural environments.

In lakes, streams and estuaries there will often be native fish which provide some control. The addition of introduced fish (mosquito fish or Gambusia, carp etc) into the natural environment can lead to the destruction of a previously balanced ecosystem. These fish may compete with the native fish for food, eat their eggs and reduce water quality. Predatory invertebrates which are visual feeders (such as dragonfly larvae and water beetle larvae) may not be able to survive in the turbid conditions created by some introduced fish. Seek the advice of the Department of Fisheries or the Department of Environment and Conservation when considering introducing fish to a wetland.

There is some evidence that the presence of fish and some invertebrates, such as backswimmers, discourages female mosquitoes to avoid laying their eggs in water bodies. Female mosquitoes may be responding to visual, olfactory or contact cues.

POTENTIAL IMPACTS OF PHYSICAL CONTROL

The physical modification of breeding sites may involve the filling or draining of areas that hold water, or the removal of vegetation with the aim of making the site unsuitable for mosquito larvae. In some cases these changes may reverse the damage created by previous human impacts (e.g. filling wheel ruts or re-opening natural channels that have been closed by vehicle movements). Provided that human impact does not reoccur, the physical modification will be a more-or-less permanent solution and will require only occasional maintenance, such as clearing of vegetation from drainage channels.

Such changes may eliminate mosquito breeding from the area concerned, but they may also inadvertently make the habitat unsuitable for other aquatic species. Reversing man-made damage would appear to be advantageous. However, in some situations when widespread loss of natural wetlands has occurred, species such as waterbirds may have come to rely on mosquitoes breeding in the man-made site as a food resource. Therefore, when proposing physical modifications of any site, take note of non-target aquatic life and contact environmental authorities if concerned.

Larvae of Coquillettidea and Mansonia mosquitoes have adaptations to allow them to live attached to submerged stems of floating or emergent wetland vegetation. Consequently, the management of these mosquito species may require the reduction of wetland vegetation. Inevitably there will be an impact on other animals which make use of the vegetated area for feeding and protection, including other invertebrates and birds. Similarly, the destruction of forest vegetation to limit the harbourage of adult mosquitoes may have undesirable effects on other animals. Manipulation of the environment should not be undertaken before environmental managers are consulted.

Runnelling

"Runnelling" is a low-impact method of physical modification for mosquito control that has been used with success in a few saltmarsh areas in Australia, including a few small sites in WA. Runnels are small-scale channels that allow tidal flushing of pools within saltmarsh habitats, but do not drain the marsh. The channels are shallow (10 - 20 cm deep and 30 - 60 cm wide), with a spoon-shaped profile. They are designed to follow natural drainage lines and have a very low gradient to avoid erosion. They work by allowing mosquito larvae to be flushed from pools into the main estuary where they are likely to die or be eaten, and they alsoallow fish to move into the pools to feed on larvae.

Runnelling has the potential to be effective in reducing mosquito numbers, however larviciding may still be necessary in some circumstances. Consequently, even when runnels have been installed, larval monitoring of the saltmarsh will be required as an ongoing activity.

The installation of runnels involves making a physical change to a marsh and therefore appropriate environmental approvals must be gained before proceeding. The Department of Environment and Conservation should be approached initially for advice on the requirements. It is likely that an Environmental Assessment and Management Plan (EAMP) will be required, as well as surveys of the site topography and soil profiles (including acid sulphate soil profiles) and for Aboriginal heritage and artifacts.

POTENTIAL IMPACTS OF CHEMICAL CONTROL

Chemical control of adult and larval mosquitoes involves the application of substances that are toxic, physically damaging or hormonally disruptive to mosquitoes in order to kill them or disrupt/slow their development. The degree to which these applications may impact on the broader wetland ecosystem will depend on the agent chosen and the scale of the application. Other animals in the environment (non-target species) may be at risk of exposure, and therefore only the registered rates and application methods must be used in order to prevent or minimise unintended impacts. Over-application is surprisingly easy, for example when applying larvicides by hand or when equipment has not been properly calibrated. Therefore, preparation and careful, accurate application is essential to avoid killing or impacting non-target animals.

There may be situations when conservation considerations demand that no management measures be undertaken due to the risks to non-target species or because the mosquitoes themselves are important as food for other animals in the habitat.

A varied or integrated approach to larval mosquito management, where larvicides with different 'modes of action' are alternated or rotated, will maximise the effectiveness of the larvicides and may also minimise the non-target impacts over time. Limiting the application of larvicide to the area where larvae occur (often the edge of water bodies) will save money, but will also ensure that accidental over-application will not have impacted on non-target animals in the untreated areas, allowing them to recolonise the water body.

Routine applications of insecticide can produce unexpected and disastrous results in particular situations. For example, an application of granular temephos (Abate 50SG[®]) for control of larval chironomid (non-biting) midges at Forrestdale Lake (Perth, WA) in 1984 was linked to the deaths of 240 waterbirds. The cause of the deaths is not entirely clear, because they may have been stressed by other factors. However, the aerial application of Abate 50SG[®] at 1.0kg/hectare to an almost dry lake meant that the waterbirds were exposed to high levels of temephos either by ingesting granules of temephos or by feeding on temephos-affected midge larvae in very shallow water. As a consequence of this incident, the Department of Environment and Conservation limits the use of temephos in lakes under its control to water depths of greater than 30cm. As a general rule, the Department of Health encourages mosquito managers to use the larvicides *B.t.i., B.s.* and s-methoprene in preference to temephos. However if temephos must be used, the application rate should be reduced when average water depth is less than 30cm.

The application of adulticides (fogging) will kill non-target flying insects contacted by the chemical. Great care must be taken to avoid the drift of synthetic pyrethroid adulticides across wetlands because of the high toxicity of these chemicals to fish. Residual surface spray adulticides will kill non-target insects and spiders landing or crawling on treated surfaces. Non-target insects that may be affected include those which are natural predators of mosquitoes as well as honey bees and biological control insects. No mosquito manager wants to be responsible for a fish-kill or the loss of an expensive agricultural biocontrol insect and the political fall-out which would follow!

When determining the most appropriate larvicide or adulticide to be used for mosquito control, attempt to use those that are the most specific in their action (i.e. kill the target insect, but not other insects or other animals). A regular monitoring program will allow environmentally preferable larvicides to be used, but the choice of larvicide will depend on the predominant larval stages present in the wetland. For example, *B.t.i.* and *B.s.* are appropriate when larvae are at instar stages I to early IV, but will not be effective against late IV instar larvae, as they have ceased feeding and will not ingest the toxin. S-methoprene is best applied to late stage larvae, because the concentration of the 'active ingredient' in the water body must be maintained to prevent the larvae from pupating and emerging as adult mosquitoes. If the larvicide is applied early there is more chance that tides or rainfall could cause dilution of the chemical and allow the larvae to complete their development.

Post-treatment larval monitoring is an essential and environmentally responsible part of a mosquito management program. In addition to assessing the effectiveness of the application against the target mosquito larvae, any adverse impacts on non-target insects or other animals (e.g. dead fish, birds, frogs, mayflies etc) should be recorded and reported to the Department of Environment and Conservation.

13. PERSPECTIVES ON PESTICIDE SAFETY

PESTICIDE SAFETY SECTION

WA Department of Health (revised by Geoff Harcombe)

INTRODUCTION

This paper is an overview of pesticide theory and safety principles. It provides information on the main statutes and Standards relevant to the safe use, handling, storage and disposal of pesticides.

Other aspects referred to include; the use of pesticides for mosquito control, health effects and personal protection.

Pesticides can play a significant and important role in integrated pest management (IPM) and are of prime importance when severe pest outbreaks occur.

The use of pesticides is not likely to diminish in the foreseeable future. The careful selection and application of these chemicals is essential to achieve an effective outcome, with minimal risk to public health.

Note: The pesticide groups mentioned below is not exhaustive, however, make up the bulk of those available in the control of common pest species.

WHAT IS A PESTICIDE?

Pesticides are chemical substances used to control or kill pests.

The definition provided in the Health (Pesticides) Regulations 2011 and the Agricultural and Veterinary Chemicals Code Act 1994 is as follows:

'Pesticide' means an agricultural chemical product as defined in the Agvet Code of Western Australia section 4. The definition of an "agricultural chemical product" is defined under the Agricultural and Veterinary Chemicals Code Act 1994 and states:

- 4 (2) subject to subsections (3) and (4), an agricultural chemical product is a substance or a mixture of substances that is represented, imported, manufactured, supplied or used as a means of directly or indirectly:
 - (a) destroying, stupefying, repelling, inhibiting the feeding of, or preventing infestation by or attack of, any pest in relation to a plant, or place or a thing; or
 - (b) destroying a plant; or
 - (c) modifying the physiology of a plant or pest so as to alter its natural development, productivity, quality or reproductive capacity; or
 - (d) modifying an effect of another agricultural chemical product; or
 - (e) attracting a pest for the purpose of destroying it.
 - (3) An agricultural chemical product includes a substance or mixture of substances declared by the regulations to be an agricultural chemical product.
 - (4) An agricultural chemical product does not include:
 - (a) a veterinary chemical product; or
 - (b) a substance or mixture of substances declared by the regulations not to be an agricultural chemical product."

PESTICIDE TYPES

(i) Organic Compounds

SYNTHETIC							
Pyrethrins	Pyrethroids	Pyrroles	Neonicotinoids	Insect Growth Regulators	Organo- phosphates	Carbamates	
Pyrethrin	Permethrin	Chlorfenapyr	Imidacloprid	S-methoprene	Maldison	Propoxur	
	Bioresmethrin				Dichlorvos	Aldicarb	
	Bifenthrin				Temephos	Carbaryl	
	Cypermethrin				Chlorpyrifos		
	Deltamethrin						



(ii) Inorganic Compounds

Examples include Copper compounds - copper sulphate, Arsenicals- arsenic trioxide

(iii) Other Pesticides

FUMIGANTS	RODENTICIDES	HERBICIDES	FUNGICIDES
Methyl bromide	Brodifacoum	Glyphosate	Mancozeb
Ethylene dibromide	Bromadiolone	Simazine	Benomyl
Chloropicrin	Coumatetralyl	MCPA	Iprodiene

(iv) Description of Organic Compounds

- (a) Organophosphates
 - Chemically unstable and generally non-persistant
 - Synaptic Poisons (see below)
 - Cholinesterase Inhibitors or Nervous System Poisons. That is, they exert their toxic action by inhibiting the cholinesterase enzymes in the Central Nervous System. This results in the accumulation of acetylcholine, causing interference with the neuromuscular junction. This results in rapid twitching of voluntary muscles, blurred vision, dizziness and ultimately, paralysis.
 - Atropine is the antidote (no longer available at pharmacies).
 - Highly toxic to birds

(b) Carbamates

- Similar to organophosphates, in their action
- Enzymes return to normal more quickly after poisoning with carbamates
- Atropine is the antidote (no longer available at Pharmacies).

- (c) Synthetic pyrethroids and Pyrethrins
 - Low toxicity to mammals
 - Axonic poisons (attacks the nerve fibre)
 - Quick acting, broad spectrum of activity
 - Breaks down quickly in the body
 - Highly toxic to bees and fish
 - Breaks down quickly in full sunlight
 - Adheres to synthetics, difficult to remove from carpets and furniture

(d) Neonicotinoids

- Act as a neurotoxin, interferes with the transmission of nerve impulses
- Translocated through plant roots when applied to soil
- Highly toxic to bees
- Moderately toxic to some birds
- Binds well to soil with organic content
- Breaks down rapidly when exposed to sunlight or high Ph water
- Very efficacious at low levels

(e) Pyrroles

- Highly toxic to birds
- Kills a wide range of insects (including termites)
- It is undetectable to insects, hence non-repellent
- Has much potential for the treatment of mosquito nets
- Low toxicity to mammals
- (f) Insect Growth Regulators
 - Low toxicity
 - Breaks down quickly in the body
- (g) Bacteria
 - Non toxic

TOXICITY AND HAZARDS

The toxicity of a pesticide is the ability of the substance to cause harm or injury once it reaches a target organ in the body.

Hazard is the risk of contamination or poisoning when a chemical is used or applied.

A hazard evaluation therefore differs considerably from a toxicology review.

For example, a pesticide may be found during toxicological assessment to be extremely toxic. However, in the hazard evaluation process, it may be determined that the hazard is low, as small quantities are used and in a manner which prevents over-exposure from occurring, except for say, in the case of equipment failure or product misuse. Conversely, a pesticide could be of low toxicity but pose a high hazard if the common use of the product involves long periods of exposure without protective equipment, due to a lack of specific information on the label.

TOXICITY MEASUREMENT

Toxicity is measured in terms of LD_{50} (Lethal Dose 50%). This is the amount of an undiluted substance, which when applied to test animals in a single dose, kills 50% of them.

This applies to both oral and dermal routes of entry into the body.

The LD_{50} is used to determine the acute toxicity of a substance and expressed in mg/kg of body weight. It does not indicate the likelihood of chronic disease occurring as a result of long periods of exposure to low concentrations of a chemical.

The dose, length of exposure, and route of absorption are the other important variables beside toxicity.

NO OBSERVABLE EFFECTS LEVEL (NOEL)

NOEL is defined as the maximum dose of a poison that can be given over a stated period without producing detectable ill effects.

PESTICIDE SCHEDULING

Pesticides are placed into Schedules indicating the level of hazard associated with using the product. The level of hazard is determined by reviewing the toxicology of the active constituent, its concentration, product formulation and any other ingredients, including solvents. These schedules are set by the Commonwealth Government and adopted under State Poisons legislation.

Pesticides are classified as either exempt from Scheduling (unscheduled) or included in Schedule 5, 6 or 7 of the classification.

SCHEDULE	TOXICITY	LABEL INFORMATION
Unscheduled	VERY LOW	Read safety direction before opening or using
Schedule 5	LOW	CAUTION Keep out of reach of children Read safety directions before opening or using
Schedule 6	MODERATE	POISON Keep out of reach of children Read safety directions before opening or using
Schedule 7	HIGH	DANGEROUS POISON Keep out of reach of children Read safety directions before opening or using

 Table 5: Pesticide classification schedule

MODES OF ENTRY

Pesticides can enter the body in any of the following ways:

- dermal
- inhalation
- oral

Dermal absorption is a very likely means of poisoning by pesticides due to occupational exposure. Over exposure can result from:

- splash or spillage of pesticide concentrate when mixing:
- prolonged exposure to spray drift
- increased absorption of pesticide due to cuts or abrasions or from chronic skin disease
- wearing clothes which have been contaminated due to overspray or drift

The rate of absorption through the skin varies with the type of chemical formulation and the area of the skin contaminated. This is an important factor often overlooked. Different areas of the skin can absorb chemical at varying rates.

Forearm	1.0
Palm	1.3
Ball of Foot	1.6
Abdomen	2.1
Scalp	3.7
Forehead	4.2
Ear canal	5.4
Scrotal area	11.8

Table 6: Relative skin absorption rates by body location

EFFECTS OF PESTICIDES ON HUMANS

Some early signs of poisoning or over exposure to pesticides are a change in behaviour, involving irritability, excitement or depression. Dizziness, headache or nausea may be present. Higher exposure may result in vomiting and uncontrolled twitching of the muscles. Severe cases of poisoning may result in fits and convulsions. Many of the health effects of pesticide exposure are not specific to pesticide poisoning, and may be due to other causes. This makes diagnosis of pesticide poisoning, very difficult.

Other health considerations when selecting a pesticide include whether it is:

- carcinogenic
- mutagenic
- teratogenic

Information on the acute and chronic health effects of exposure to a particular pesticide is available on a Material Safety Data Sheet (MSDS). At the time a pesticide is purchased, an MSDS may be obtained from your local pesticide distributor. It is important that any person using pesticides has access to an MSDS and has read the additional information regarding the pesticide prior to use.

PESTICIDES USED TO CONTROL MOSQUITOES

The following provides a guide to the toxicity and degree of hazard associated with some of the commonly used pesticides for mosquito control. **Note:** Prior to deciding on the use of a particular pesticide, please check with your chemical supplier to determine its availability and quantity in stock.

Trade name	Active Ingredient	Group	LD50	(mg/kg)	Schedule
			Oral	Dermai	
A sug 1/ Othering	Aduiticides (togging)	004	5000	<u> </u>
Aqua-K Olnrine	20g/L deltamethrin	SP SP	304	>5000	5
Dipthor LILV Insecticide	25g/L Cypermethin		2194	>0040	5
Twilight LILV Mosquito	89g/L Opernetnin 89g/L Phenothrin 89g/L	JF	5104	>2000	0
Adulticide Concentrate	Piperonyl butoxide	SP	>5000	>2000	5
Pyrethrin	4g/L Pyrethrins 24g/L Piperonyl Butoxide 719g/L Liquid Hydrocarbon	SP	NOEL: 10mg/kg	Not available	5
Pyrocide Mosquito Adulticiding	118g/L Pyrethrins 592g/L Piperenyl butevide	SP	>3000	>2000	5
	1169g/L Maldison		> 1000	> 1000	6
		UF	>1000	> 4000	0
Hy-Mal Insecticide	1150g/L Maldison	OP	5500	>2000	6
Maldison ULV insecticide	1180g/L Maldison	OP	1500	>4000	6
	Adulticides (barrier/ re	sidual tre	atments)		
Bifenthrin Aqua Termiticide and Insecticide	100g/L Bifenthrin	SP	505	>2000	6
Biflex Aqua Max	100g/L Bifenthrin	SP	505	>2000	6
Brigade T&O Insecticide/Miticide	80g/L Bifenthrin	SP	632	>2000	6
Isopthor Aqua	80g/L Bifenthrin	SP	632	>2000	6
	Larvici	des			
Abate 10 SG Mosquito Larvicide Granules	10g/kg Temephos	OP	>5000	>5000	6
Graybate 10 SG Mosquito Larvicide Granules	10g/kg Temephos	OP	>5000	>5000	6
Graybate 50 SG Mosquito Larvicide Granules	50g/kg Temephos	OP	>5000	>2000	6
Biopren 4GR Mosquito Larvicide	4g/kg (S) - methoprene	IGR	>5000	>2000	Unscheduled
Biopren 50 Liquid Mosquito Larvicide	50g/L (S) - methoprene	IGR	>5000	>2000	Unscheduled
Nomoz Mosquito Larvicide Pellets with ProLink	40g/kg (S) -methoprene	IGR	>5000	>2000	Unscheduled
ProLink – ProSand Mosquito Growth Regulator	4g/kg (S) - methoprene	IGR	>5000	>2000	Unscheduled
ProLink Liquid Larvicide Mosquito Growth Regulator	50g/L (S) - methoprene	IGR	>5000	>2000	Unscheduled
ProLink Pellets Mosquito Growth Regulator	40g/kg (S) - methoprene	IGR	>5000	>5000	Unscheduled
ProLink XR Briquets	18g/kg (S) - methoprene	IGR	>34,000	>2000	Unscheduled
Teknar 1200 SC Biological Larvicide	Bacillus thuringiensis Var.israelensis	Bacteria	>5000	>2000	Unscheduled
Vectobac AS Biological Larvicide	Bacillus thuringiensis subsp. israelensis	Bacteria	>5000	>5000	Unscheduled
Vectobac G Biological Larvicide	Bacillus thuringiensis subsp. israelensis	Bacteria	>5000	>5000	Unscheduled
Vectobac WG Biological	Bacillus thuringiensis	D	5000	5000	
Larvicide	subsp. israelensis	Bacteria	>5000	>5000	Unscheduled
Aquabac 200 GR	Bacilius inuringiensis subsp. israelensis	Bacteria	>5000	>2000	Unscheduled
Larvicide	2362	Bacteria	>5050	>5050	Unscheduled

Table 7: Pesticides commonly used in mosquito control

SP = Synthetic pyrethroid, OP = Organophosphate, IGR = Insect Growth Regulator **Table courtesy Geoff Harcombe 2006 - 2011 (updated 2013)**

LEGISLATIVE REQUIREMENTS

Legislation and Australian Standards

To control the use of pesticides and fumigants in Western Australia, legislation has been developed and enacted. There are two guidelines which will provide assistance to those involved in pest management, one a national document which is risk based, the other, WA specific for local government. Some of these guidelines can be found at the following web addresses:

Guide to the management of pesticides in local government pest control programs can be found at:

http://www.public.health.wa.gov.au/cproot/2663/2/A%20guide%20to%20the%20management %20of%20pesticides%20in%20local%20government%20pest%20control%20programs%20in %20Western%20Australia.pdf

Guidelines to safe use of pesticides in non agricultural workplaces: <u>http://www.public.health.wa.gov.au/cproot/2158/2/Guidelines%20for%20the%20safe%20use</u> <u>%20of%20pesticides%20in%20non-agricultural%20workplaces.pdf</u>

Quick contacts for the use of pesticides in Western Australia can be viewed at: <u>http://www.public.health.wa.gov.au/cproot/3775/2/Quick%20Contacts%20for%20the%20use%</u> 20of%20Pesticides%20in%20WA%20Feb%202011.pdf

However, there are three pieces of legislation relating to pesticide use that are mandatory, they include:

(i) Legislative Control

The current legislation in Western Australia to control the use of pesticides and fumigants is the Health (Pesticides) Regulations 2011. The 1956 Regulations were repealed on the 1st February 2011 along with the gazettal of the new Regulations.

The Regulations remain the principle instrument for the control of pesticide and fumigant use in Western Australia, where it affects public health. Some of the relevant provisions of the Regulations are cited in the paper.

(ii) Australian Standard 2507 - 1998

This Standard provides guidance for the safe storage and handling of pesticides, and is now a legal requirement for those who hold a Pest Management license and store pesticides on their business premises.

(iii) <u>Code of Practice:</u> Disposal of Pesticide Residues from Pesticide Spray Applications. This Code provides guidelines for the safe disposal and reuse of pesticide residues.

(I) HEALTH (PESTICIDES) REGULATIONS 2011

Table 8: Main aspects covered by the Health (Pesticides) Regulations 2011.

Regulation			
5	Definition of a Pest management technician		
6	Defines a Pest management treatment		
7	Registration of a business		
48	Information on a license card		
52	Review of licensing decisions		
62	Storage and handling of registered pesticides		
63	Fumigation		
77	Record of pest management treatments		
84	Pesticides to be kept and used safely		
86	Transportation of pesticides		
88	Signage and warning light when spraying from vehicles		
89	Application of Pesticides in Public Places - warning signs		
94	Disposal of registered pesticides		
95	Disposal of used pesticide containers		
100	EDPH permits		
104	Powers of a public health official		
109	Liability of employers for acts of employees		

The full Health (Pesticides) Regulations 2011 can be viewed at: <u>http://www.slp.wa.gov.au/pco/prod/FileStore.nsf/Documents/MRDocument:21385P/\$FILE/Hea</u> <u>lthPesticidesRegs2011-00-b0-01.pdf?OpenElement</u>

(II) AUSTRALIAN STANDARDS 2507-1998 THE STORAGE AND HANDLING OF PESTICIDES

The above Standard can be referred to in discussing storage installations and personnel safety. This Standard is called up under current Health (Pesticides) Regulations 2011 in Western Australia for those persons who are licensed and store pesticides at their place of business. It is recommended that the storage of pesticides should be in accordance with the Standard and also apply to those not licensed.

STORAGE INSTALLATIONS

The discussion on these installations covers siting, construction and ancilliaries required for pesticide stores.

PERSONAL SAFETY

This section discusses emergency communication arrangements, basic personal hygiene precautions and protective equipment requirements.

(III) CODE OF PRACTICE: DISPOSAL OF PESTICIDE RESIDUES FROM PESTICIDE SPRAY APPLICATIONS

This Code of Practice applies to both licensed pesticide technicians and State/Local Government, and is an additional requirement for the disposal of pesticide residues. The decontamination of spraying equipment at the site of application and the re-use of pesticide residues are the main features of this Code.

Where Local Governments are unable to dispose of pesticides, they may need to contact the following companies:

Environmental Recovery Service Ph: 1800 118800

OR

Tox Free Solutions Ph: 1300 869373

OR

The Dept of Conservation and Environment Ph: 08 6467 5000

SAFETY GUIDELINES FOR USE OF PESTICIDES

Before using any pesticides for the control of mosquitoes or other uses, you should familiarize yourself on how the pesticide works on its target and its potential human health and environmental impacts. Before mixing formulations for use, consult the Material Safety Data Sheets (MSDS)

Material Safety Data sheets (MSDS) can be found at the following web sites:

- The Australian Medical and Veterinary Medicines Authority: Check the PUBCRIS registration information at: <u>http://services.apvma.gov.au/PubcrisWebClient/welcome.do</u>
- Chemical Suppliers websites: For example, Garrards Pty Ltd displays all MSDS for the chemicals they sell: <u>http://www.garrards.com.au/</u>

MSDS's provide useful information on the product being used. Make sure you understand the chemical before using it. The following information is important to check before mixing and applying pesticides:

- The product name (make sure it is the correct MSDS for the pesticide you plan to use);
- The chemical Group that the pesticide is classified as;
- The formulation (what the pesticide is to be diluted with;
- Emergency phone number in case of accidents;
- The risks of using the pesticide (Section 2: Hazard Identification);
- First aid requirements if exposed to the pesticide (Section 4: First Aid Measures);
- Understand the implications in case of fire (ensure pesticides are stored correctly and al appropriate signage is displayed for firefighters (Section 5: Fire Fighting Measures);
- Accidental release measures provides information on accidental spillage and clean-up procedures (Section 6: Accidental Release Measures);

- How to handle and store pesticides (Section 7: Handling and Storage);
- Check the personal protective guidelines prior to using a pesticide (Section 8: Exposure controls/Personal protection);
- The physical and chemical properties of the pesticide (Section 9);
- The stability and reactivity of the pesticide (Section 10);
- The toxicology information (Section 11);
- The ecological/environmental information (section 12) and the potential impacts on non-target organisms; and
- Disposal information (Section 13).

A copy of all MSDS's for each chemical in your arsenal should be stored in a folder on site for easy reference and to be provided to fire fighters and emergency personal in case of fire or accident. The MSDS's should be easy to locate and maintained in an up-to-date fashion at all times.

PESTICIDE LABELS

The Pesticide label also provides valuable information and should be referred to before use. The label provides the following information:

- Product name and Pesticide group;
- Schedule heading (warning sign);
- Storage and disposal information;
- Safety and first-aid directions;
- Directions for use;
- Rates of application;
- Application methods and timing;
- Pesticide resistance; and
- Precautions for use.

Pesticide labels can be found at the following websites:

- The Australian Medical and Veterinary Medicines Authority: Check the PUBCRIS registration information at: <u>http://services.apvma.gov.au/PubcrisWebClient/welcome.do</u>
- Chemical Suppliers websites: For example, Garrards Pty Ltd displays all MSDS for the chemicals they sell: <u>http://www.garrards.com.au/index.php?option=com_msds&view=all&letter=a<emid=11</u>
- Understanding Pesticide Chemical Labels can be found at the APVMA website: <u>http://www.apvma.gov.au/use_safely/docs/understanding_labels_booklet.pdf</u>

A copy of all labels for each pesticide stored at your premise should be maintained and kept up to date in a folder for easy use and reference. This information is useful to staff applying pesticides, but also provides important information to staff and emergency personnel.

PRACTICAL ASPECTS OF PESTICIDE SAFETY

SELECTION AND CARE OF PERSONAL PROTECTIVE EQUIPMENT (PPE)

Exposure to pesticides can occur during:

- transportation
- mixing
- storing
- loading
- application
- working in treated crops
- decontamination of equipment
- disposal
- decontaminating spillage
- handling of contaminated clothing

When involved in any of these activities, personal protective equipment should be worn in accordance with instructions provided on the pesticide label. *Remember:* the label on pesticide containers is a legal document, which must be complied with.

OVERALLS

Overalls should be long sleeved to afford you the best protection. They are available in a variety of materials, depending on the type of work to be undertaken. Cotton/polyester overalls are suitable for hot weather; however need to be washed at the end of each day as they will absorb pesticide. Chemical resistant overalls are more suitable to heavy duty or continued handling of pesticide as they are less likely to absorb chemical and prevent dermal absorption of pesticide.

WATERPROOF CLOTHING

Waterproof clothing should be worn when particularly toxic chemicals are being used or if there is an increased risk of contamination due to spray drift, or handling concentrate. This too should be washed after use.

GLOVES

- Unlined PVC or Nitrile rubber.
- Elbow length.
- Lining should be avoided as this can absorb pesticide and become contaminated. Unlined gloves are far easier to clean.
- Don't use leather or cotton gloves as these absorb pesticides and hold it close to the skin.
- If there is a chemical odour inside the gloves, discard immediately.
- Check regularly for pinpoint holes, by filling with water and squeezing. Discard if holed.
- Gloves should be worn over protective clothing and turned up at the ends, this assists in reducing pesticide from reaching unprotected parts of the body.

BOOTS

- PVC or rubber with steel caps.
- Wear overalls outside boots.
- Should be unlined, as the lining will absorb pesticide
- Wash regularly both inside and out to remove contamination.

PVC APRON

- Wear whenever mixing or handling concentrate, to deflect spillage away from the groin region. (Remember the high absorption rate of this area!)
- Wash after use.

HAT

- Waterproof hat if possible, with a broad brim
- Should be washed at the end of the day

RESPIRATOR

- Wear whenever handling concentrates and applying pesticides.
- Cartridge for agricultural chemical must be used. (Check to ensure organic vapor type).
- Simple dust filters are of no use on their own, although they are useful when used in combination with an organic vapor cartridge.
- Adjust fitting of headband to give a good tight seal against the face.
- If you are clean shaven, your respirator will fit better than with facial hair.
- Change filters regularly one filter should last up to 8 hours. Change filters if pesticide can be detected regardless of the length of time they have been used.
- Do not expose the inside of the respirator to spray drift if contaminated, remove and wash immediately
- To wash, remove cartridges and wash face piece in warm soapy water. Rinse thoroughly and dry.
- Store the cartridge in a well sealed container to maximise the life span of the cartridge. Remember, the cartridge will keep on working, even when not being worn.
- Regularly check that the O-rings are flexible, are not damaged and that the rubber of the respirator has not perished in any way.
- If you can smell chemical while using the mask,
 - a) The fit is poor, OR
 - b) The charcoal is exhausted, OR
 - c) The cartridge is unsuitable for the job at hand.

FIRST AID

- Carry a basic first aid kit with you when preparing or applying pesticides. First aid kits should be maintained, item replaced when used and checked annually.
- Carry eye wash equipment and sufficient potable water to enable the eye and other parts of the body to be irrigated or decontaminated; up to 20L is appropriate and can be used to wash eyes, hands and to decontaminate affected areas.
- A compression bandage is also important in case of snake bite.

14. THE CHEMICAL CONTROL OF MOSQUITOES

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Mosquito-Borne Disease Control Branch Department of Health

CHEMICALS FOR CONTROLLING MOSQUITOES

There are four main types of chemicals that can be used for mosquito control:

INSECTICIDES

Insecticides are used to kill mosquitoes at various stages of their life cycle, reducing the problem by reducing the population. Insecticides used to kill mosquito larvae are called larvicides, and insecticides used to kill adult mosquitoes are called adulticides.

SURFACE FILMS

Most species of mosquito larvae rely on the surface tension of water to enable them to attach to the surface and breathe. A thin film of oil, kerosene or other substance breaks this surface tension, so the larvae can no longer attach to the surface, and subsequently drown.

REPELLENTS

Personal repellents are applied to the body to ward off direct mosquito attack (repellent formulations are available for animals as well). Spatial repellents, such as mosquito coils and citronella candles, can be strategically located to deter mosquitoes from entering an area.

HERBICIDES

Herbicides can be used to control vegetation that may provide mosquito egg-laying sites or harbourage for larval or adult mosquitoes.

These notes focus mainly on the use of insecticides for mosquito control; however there is also information at the end of the notes on the use of surface films and repellents. A detailed table has been included at the end of the chapter which lists most of the currently registered products for mosquito control in Western Australia.

MOSQUITO CONTROL – WHERE DO INSECTICIDES FIT IN?

The use of insecticides is undoubtedly the most common form of mosquito control at a global level. However, it is wrong to assume that this popularity suggests that insecticide control is the most effective, easiest or cheapest control method available.

An important issue facing any mosquito control program is that of sustainability. To be sustainable, a program must be able to achieve the long term goals for which it was put in place. To do this, the program must:

- be effective in controlling mosquitoes and mosquito-borne disease;
- be cost justifiable and cost effective; and
- have a minimal or acceptable level of environmental impact.

Only by employing a combination of methods with careful consideration of the problem at hand, can these criteria be met. This control approach is known as Integrated Pest Management (IPM). In adopting an IPM approach to mosquito control, the use of insecticides should be seen as a complimentary strategy to other methods.

Where possible (in terms of cost, engineering and environmental considerations), physical control or source reduction methods should be used in preference to insecticide control.

PHYSICAL CONTROL

- Offers efficient, low maintenance, long term control that can at the very least reduce the recurrent labour and product costs of using insecticides.
- When well-planned can have minimal impact on the environment.
- When incorporated in the design stage of artificial or man-made developments, such as constructed wetlands, can provide highly effective mosquito control from the outset.

Insecticides have a place in mosquito control, in particular when controlling vectors during disease outbreaks. However, insecticides should be used sensibly to maximise their benefits while minimising any disadvantages.

MOSQUITO INSECTICIDES - GENERAL CONSIDERATIONS

A number of aspects need to be considered when selecting insecticides for mosquito control. These include:

COST

Insecticides can be very costly. The reason lies in the fact that the main costs of using insecticides (labour and product) are recurrent. Over large areas or where mosquito breeding is frequent, these costs quickly add up. Cost may also be a factor in the choice of the insecticide used. Some insecticides cost considerably more than others, but offer other benefits that may make them more economical in the longer term.

SPECIES

Previous chapters focused on the biology and behaviour of mosquitoes, how this can differ amongst species, and how this information is a very important consideration in planning effective control. This is a foundation principle of this course and a theme that will reoccur frequently. It is also a major consideration in insecticide control. Thus we need to identify the mosquito species we are trying to control, understanding their biology and behaviour and choose an appropriate insecticide for maximum control.

RESISTANCE

History has shown many examples of where inappropriate use of insecticides leading to the development of resistance in the target insect. Resistance can develop in an insect population when individual insects with a genetic predisposition to resisting the effects of the insecticide survive the treatment and breed. Over time, this genetic trait can start to build up in the population. This usually starts off very slowly, but once resistance occurs it can quickly spread throughout the population. Often, when the first signs of resistance are noticed, it is too late to do anything about it.

There are a number of ways to defer the development of resistance in mosquitoes by:

- developing control programs that do not rely on insecticides; using other control methods where possible and appropriate.
- rotating insecticides with different modes of action.
- leaving wild populations of mosquitoes that do not need to be controlled alone. This enables recolonisation of the treated area by the non-resistant genotype.

Most experts believe, however, that the high reproductive rates and short generation times of mosquitoes mean that the development of resistance to a particular insecticide, over time, is inevitable.

MONITORING REQUIREMENTS

The effective use of insecticides for mosquito control requires monitoring. This applies when controlling both larval and adult mosquitoes. No program can continue to be efficient and therefore cost-effective without appropriate monitoring. A pre- and post-treatment monitoring program offers the following benefits:

- it identifies the target species, providing valuable information for the planning of management measures;
- it gives warning of the impending need for management;
- it enables management measures to be timed for maximum effect;
- it allows assessment of the effectiveness of management measures; and
- ultimately allows refinement of the program.

THE ENVIRONMENT

The use of insecticides always involves some level of environmental impact. This problem can be magnified when insecticides are used inappropriately. Severe impact on an ecosystem is undesirable and can be counterproductive to mosquito control efforts in the long term. The issue of environmental impacts is covered in Chapter 12., "Environmental Considerations in Mosquito Control" on page 100.

PUBLIC AND OPERATOR SAFETY / LABEL REQUIREMENTS

Insecticides are often used because of their inherent toxicity and some are not only toxic to mosquitoes but also humans. Public and operator safety must be considered when using insecticides. By law and without exception, an insecticide can not be used in any manner contrary to that described on its label without the permission of the Australian Pesticides and Veterinary Medicines Authority. Label instructions are also designed to help the user maximise the effectiveness of the product.

For more information see Chapter 13., "Perspectives on Pesticide Safety" on page 106.

PREPARATION

An ounce of preparation is worth a tonne of perspiration when it comes to using insecticides for mosquito control. Apart from monitoring, preparation includes knowing what resources are available for mosquito control (particularly personnel), what equipment is available and the logistics of obtaining the required insecticide.

Mosquitoes don't wait for anybody, and when ideal mosquito breeding conditions arrive, there will generally only be a short opportunity (as little as 2-3 days) to do anything meaningful about the impending problem. A program must be well planned and prepared to swing into action quickly to cover any contingency.

YOU CANNOT ERADICATE EVERY MOSQUITO

It is simply not possible or environmentally desirable to eradicate every last mosquito from an area. This is something that the person responsible for mosquito control and the general public both need to understand.

INSECTICIDES FOR MOSQUITO CONTROL

Insecticides used for mosquito control can be divided into two main groups. Products that are used to kill larvae are called larvicides, and products used to kill adults are called adulticides. Both have advantages and disadvantages that need to be evaluated and applied to the mosquito problem requiring control. Some of these are considered here.

In general, where insecticide control of mosquitoes is required, larviciding should be used in preference to adulticiding where possible.

LARVICIDES - ADVANTAGES:

- The targets of a larvicide (mosquito larvae) are confined to their breeding site. The area that requires treatment is clearly defined and is generally much smaller than the area that would need treatment once the larvae have emerged as adults and have dispersed.
- Because the larvae are confined to clearly defined areas, effective treatment using a larvicide, which should aim for >95% kill rate, can greatly reduce the subsequent adult population.
- New generations of larvicides available tend to be very target specific, thus their impact on the environment can be controlled through appropriate use.
- Larvicides kill mosquitoes before they pose a health or nuisance risk.
- By killing the mosquitoes as larvae, they don't get the opportunity to breed. Theoretically, if you are able to significantly reduce the population at this stage of their life cycle, and at an early stage of their breeding season, it should make ongoing control easier.
- Some larvicides are available in controlled release formulations, giving longer residual control.
- For small breeding sites, the application of a larvicide does not require any specific or expensive equipment.

LARVICIDES - DISADVANTAGES:

- Mapping breeding sites is time consuming, can require a lot of man-power, and generally needs to be done when mosquito control is also required.
- Mosquito breeding sites can often be difficult to define in areas where breeding is infrequent. Sites can also be difficult to access and thus difficult to treat.
- Larviciding is an inflexible control method. Treatment must be carried out within a short time period (when the opportunity presents itself), particularly with new generation larvicides.
- In situations where breeding occurs over large areas, especially if the target species can travel considerable distances, effective larviciding can quickly become uneconomical.
- There are no larvicides registered to control mosquito pupae.

ADULTICIDES - ADVANTAGES:

- Some forms of adulticides offer longer lasting residual effects.
- Once mosquitoes have emerged, adulticiding is the only control method available. This is particularly important for vector control during disease outbreaks.
- Adulticiding is a flexible control method that can be utilised (weather conditions permitting for fogging) as required.
- Adulticiding offers a level of mosquito control that can be effective at a much lower cost than larviciding, particularly in situations where communities are surrounded by vast areas of land with high mosquito breeding potential.

ADULTICIDES - DISADVANTAGES:

- Some forms of adulticiding (fogging) offer only very temporary control, as adult mosquitoes will migrate into the control area from adjacent uncontrolled areas.
- Adulticides are not as target specific as larvicides, and can affect many other insects. This environmental impact can be very significant and is undesirable.
- Adulticiding by way of fogging requires dedicated equipment that can be expensive if the capacity to treat a large area is required (as is often the case).
- The effectiveness of some forms of adulticiding is largely determined by the weather.
- The timing of adulticiding (i.e. fogging) is critical. If the target mosquitoes are not active at the time when the fogging is carried out, the effectiveness of the treatment is greatly reduced.
- Due to the recurrent cost of labour and insecticide, long term adulticiding programs for large areas can be very expensive.
- Access to areas where adulticiding (fogging) is required is not always available. Larger fogging machines are usually used from vehicles.
- Adulticides must be used with care around waterways. Synthetic pyrethroid products are lethal to fish and care must be taken to avoid adverse impacts.

CHOOSING AND USING A LARVICIDE

Table 1 provides a list of larvicides registered for use in Western Australia as of July 2004. The list provides a range of products, although depending on circumstances, the most commonly used larvicides are those containing s-methoprene, *Bacillus thuringiensis* var. *israelensis* (Bti), *Bacillus sphaericus* or temephos. These four actives will be discussed in more detail. Some of the other actives from the table can be very useful however, particularly in man-made environments (eg. septic tanks, sewerage ponds etc.).

Chemical Type	Active Ingredient	Environmental Hazard			
Chemical Type		Bees	Fish	Aquatic	Birds
Bio-larvicide	Bti	No	No	No	No
	Bs	No	No	No	No
Organophosphate	Temephos	High	High	High	High
Insect Growth Regulator	s-methoprene	Low	Medium	Medium	No

Table 9: Currently registered active ingredients for control of mosquito larvae in WA

[Adapted from Australian Mosquito Control Manual, Mosquito Control Association of Australia, 1998 (revised 2002)]

Be warned that the registration status of any product may change. If you are unsure of the registration status of any product, confirm it by contacting the Australian Pesticides and Veterinary Medicines Authority (APVMA) or using the PUBCRIS database over the Internet (<u>http://www.apvma.gov.au</u>). The Mosquito-Borne Disease Control (MBDC) section of the Department of Health (DOH) and chemical suppliers can also advise you of the registration status of any product.

S-METHOPRENE

S-methoprene is an insect growth regulator (IGR). Products containing this active work by mimicking a juvenile hormone (JH) that is present in the larval stage of the mosquito's life cycle.

Levels of JH are highest during the early instar stages of a mosquito larva's development. As the larva progresses to III and IV instar, the level of JH drops considerably. When s-methoprene is added to the breeding environment it is readily absorbed by the larva through contact. Because the s-methoprene mimics the JH, the larva's hormone levels become unbalanced. The desired effect is to prevent adult mosquitoes emerging from the pupal stage.

FORMULATIONS

A number of different s-methoprene formulations are currently registered for mosquito larval control in Western Australia. These are summarised in the following table:

 Table 10: S-Methoprene formulations are currently registered for mosquito larval control in Western

 Australia

Trade Name	Formulation	Application Rate	Control Period
Prolink [®]	Liquid	220-360ml/ha	One larval generation
Biopren [®] 50	Liquid	220-360ml/ha	One larval generation
Prosand®	Granule	3-5kg/ha	One larval generation
Biopren [®] 4GR	Granule	3-5kg/ha	One larval generation
Prolink Pellets [®]	Pellet	3-4kg/ha	30 days min
NOMOZ®	Pellet	3-4kg/ha	30 days min
Prolink XR Briquets [®]	Briquette	1 per 10-20 m ²	150 days max.

(NOMOZ[®] is the equivalent to the Prolink Pellets[®], however it is packaged in quantities to meet the domestic market as opposed to the commercial market.)

CONSIDERATIONS

S-methoprene products have a number of advantages when used for larviciding:

- They are very target specific, meaning they have very little impact on other aquatic species and hence the environment.
- They are not considered toxic to humans.
- The pellet and briquette formulations offer extended periods of control. This is especially useful for difficult to treat breeding sites, or for the control of species such as *Coquillettidia* and *Mansonia*, which are very difficult to monitor.
- They have a distinctly different mode of action to most products available a positive for resistance management.
- The briquette formulation is the only larvicide registered for use in drinking rainwater tanks.

There are also a number of important factors that need to be considered when using smethoprene products for larval control:

• The timing of any treatment is vitally important when using the granule and liquid formulations. Ideally, the treatment needs to occur when most larvae are late II to early IV instar. The treatment is useless once larvae have pupated. Hence, intensive pre-treatment monitoring is necessary for maximum efficiency.

- A disadvantage of using s-methoprene is that the effectiveness of the treatment can only be accurately determined by the number of adults that emerge. If the treatment fails, there are no second chances to larvicide again.
- S-methoprene does not provide a quick kill as some other more inherently toxic insecticides do.
- The briquettes can be difficult to use effectively in areas where flushing of the water-body occurs. Soft mud and loose sediment can cover the briquettes, inhibiting the release of the active ingredient. Tethering of the briquette in a mesh bag tied to a stake may be required. This will also prevent the briquette being washed away.
- The longer lasting formulations (pellets and briquettes) are expensive, though their residual activity tends to make them cost effective in the longer term.
- S-methoprene binds readily with organic material, meaning that the larvicide has reduced effectiveness in heavily organically polluted water (e.g. sewage ponds).

BACILLUS THURINGIENSIS VAR. ISRAELENSIS (BTI)

The larvicide Bti is a product of the activity of the bacteria *Bacillus thuringiensis* var. *israelensis*. Toward the end of its life cycle, the Bti bacteria sporulate, producing a crystalline endotoxin that is particularly lethal to mosquito larvae. In the manufacture of Bti, this endotoxin is collected and sold in a variety of formulations.

Bti goes to work after being ingested by the mosquito larva. Once the larva ingests the crystal, the action of certain gut enzymes converts the crystal into a number of other products. These by-products destroy the epithelial lining of the larva's gut, leading to death.

Bti is not a biological control agent. While being naturally produced by Bti bacteria, the crystals work as a chemical larvicide. They are incapable of reproduction and do not persist like a true biological control agent.

FORMULATIONS

A number of Bti formulations are currently registered for use in Western Australia. These are summarised in the table below:

Trade Name	Formulation	Applpication Rate
Vectobac [®] 12 AS	Liquid	0.3 – 1.2 L /Ha
Teknar [®] 1200SC	Liquid	0.3 – 1.2 L /Ha
Vectobac [®] G	Granule	7 kg/Ha
Vectobac [®] WG	Wettable Granule	125 – 800 g/Ha

 Table 11: Bti formulations currently registered for mosquito larval control in Western Australia

CONSIDERATIONS

Bti products have a number of advantages when used for larviciding:

- Of all the larvicide products available, Bti (along with *Bacillus sphaericus* detailed below) has the least impact on other life in the aquatic environment. It is the most environmentally friendly larvicide available in WA.
- Bti kills mosquito larvae within 24 hours, so the effects are readily noticed. This gives an opportunity to assess the success of the treatment while the mosquitoes are still confined to their aquatic breeding environment.
- Bti has a shelf life of up to 2 years if stored correctly.

There are also a number of important factors that need to be considered when using Bti products for larvae control:

- The activity of the product when applied is limited to approximately 24 hours.
- Bti must be applied when larvae are at stages of development where they are still feeding. Pupae and late IV instar larvae are not affected by the treatment. Therefore, comprehensive pre-treatment larval monitoring is required to maximise the efficiency of the product.

BACILLUS SPHAERICUS (BS)

This formulation is a biological control agent or a bio-larvicide. The formulation (known as Vectolex[®] WG) is produced from the bacterium *Bacillus sphaericus*.

Its mode of action is similar to Bti in that it goes to work after being ingested by the mosquito larva, destroying the epithelial lining of the larva's gut, leading to death. The one difference however, is its ability to replicate, producing more toxic spores resulting in a residual treatment that can last up to three weeks.

Like Bti, Bs has minimal effects on non-target species. This product is ideal for using in polluted waters such as sewage treatment lagoons, as it does not readily bind to organic matter.

FORMULATIONS

There is currently only one Bs formulation currently registered for use in Western Australia (shown in the table below):

Table 12: Bs formulations currently registered for use in Western Australia

Trade Name	Formulation	Application Rate
Vectolex [®] WG	Wettable Granule	0.5-1.5 kg/ha

CONSIDERATIONS

This Bs product has a number of advantages when used for larviciding:

- Along with Bti, it is the most environmentally friendly larvicide available in WA it has the least impact on other life in the aquatic environment.
- Bs kills mosquito larvae within 48 hours, so the effects can be easily monitored.
- Bs has a residual effect of up to 21 days.
- Bs has a shelf life of up to 2 years if stored correctly.

There are also a number of important factors that need to be considered when using Bs products for larvae control:

• Bs must be applied when larvae are at stages of development where they are still feeding (late I to early IV instar). Late IV instar larvae and Pupae are not affected by the treatment. Therefore, comprehensive pre-treatment larval monitoring is required to maximise the efficiency of the product.

TEMEPHOS

Temephos is an organophosphate compound. As is typical of this family of chemicals, temephos is an anti-cholinesterase agent. Such products disrupt the functioning of enzymes that are critically important to the nervous system. This leads to paralysis affecting vital bodily

functions, including the respiratory system, and a quick death. Temephos works by being absorbed through the skin of the larva, so it is able to be used from I to IV instars. It is **not** registered to be used for pupae.

Temephos has been used widely in Western Australia in the past; however it is no longer used as commonly, with a wider choice of more "environmentally friendly" products on offer.

FORMULATIONS

There are two temephos formulations registered for use in Western Australia. These are summarised in the following table.

Table 13: Temephos formulations currently registered for use in Western Australia

Trade Name	Formulation	Application Rate
Abate [®] 10SG	Granule (for Aerial use)	5-10kg/ha
Graybate [®] 10SG	Granule (for Aerial use)	5-10kg/ha
Graybate [®] 50SG	Granule (for Aerial use)	1-2kg/ha

CONSIDERATIONS

Temephos products have a number of advantages when used for larviciding:

- Temephos kills larvae quickly, so the results of control efforts can be assessed soon after treatment.
- It can be used on I IV instar larvae
- If a contact poison (quick kill) must be used for larviciding in a natural environment, temephos used at label rates is likely to have a relatively low environmental impact compared to other contact poisons.

However, there are a number of important factors that need to be considered when using temephos products for larvae control:

- Temephos binds readily with organic material, meaning that the larvicide is not as effective in heavily organically polluted water.
- Temephos is not a selective insecticide, and can have significant impacts on aquatic invertebrate fauna.
- Temephos is toxic to humans, and care must be taken in its use.

CHOOSING AND USING AN ADULTICIDE

The term adulticide is very broad. When applied to mosquitoes, it can cover any insecticide used to control adult mosquitoes. Technically, this includes many common household fly-sprays, a range of surface sprays to control adults on surfaces they come in contact with, through to the formulations used in Ultra Low Volume (ULV) techniques.

Commonly throughout Australia, when discussing adulticiding or fogging, people are referring to the use of products in ULV or thermal aerosol generators. Until recently people relied heavily on ULV and thermal fogging for adulticiding, however there is also now a residual surface treatment that has been specifically designed for control of mosquitoes, biting midges and flies.

While these notes focus on only a few adulticides, there are also other products available that should not be dismissed when implementing mosquito control measures.

BIFENTHRIN RESIDUAL ADULTICIDE

There are now residual surface treatments designed for the control of mosquitoes. These products are made from the synthetic pyrethroid, bifenthrin, and are marketed under several names. Some of these include – Bifenthrin Aqua Termiticide and Insecticide; Bistar[®]; Brigade T&O; Biflex AquaMax; and, Isopthor* Aqua. They work by knocking down adults which land on surfaces where the products have been applied. Most surface sprays have repellent properties, so while the main idea is to get the adult to land on the surface to get a knock down, they are actually deterred by the repellent. These newer products have reduced repellency properties and so mosquitoes will readily land on surfaces where it has been applied.

These bifenthrin adulticides can be used in two ways, as external barrier treatments and as indoor residual surface treatments. For outdoor barrier treatments the manufacturers recommend treating any surface area that a mosquito would rest on, including shrubs and hedges, tall grasses, surfaces surrounding entertainment areas, basically anywhere mosquitoes will harbour. **Bifenthrin products should never be used near wetlands or water bodies as they can be lethal to fish and aquatic organisms.** They can also be applied to interior surfaces and harbourage sites.

Bifenthrin residual adulticides can be applied using a range of applicators such as power sprayers, hand-held pump sprayers and backpack sprayers which are operated to produce large droplets to wet the target surface and to minimise spray drift. The product **must not** be applied using ULV or fogging equipment because these produce small droplets that may result in drift across sensitive areas.

Bifenthrin offers a new approach to mosquito control. In the past it has been recommended to remove harbourage areas in order to deter mosquitoes. By incorporating this product into a control program it would be more effective to provide good harbourage areas (treated with bifenthrin) that will attract the adults, resulting in a greater knock down rate.

Some of the advantages of using these products include:

- They can be applied at any time of the day (compared to traditional adulticide application in the early morning or evening when mosquitoes are active);
- The equipment used for applying bifenthrin is much cheaper than fogging equipment;
- It is residual can last up to two months; and
- No odour or staining.

Like any insecticide there are also important factors to consider when using these products:

- They are not target specific; and
- They can be lethal to fish and aquatic organisms and must be applied as large droplets to avoid spray drift.

REPELLENT TREATMENT OF CLOTHING AND BEDNETS

The treatment of clothing and bednets with mosquito repelling chemicals may be necessary in situations where there is no formal mosquito control, in particular, by those who sleep or camp outdoors; e.g. tourists, mine exploration workers and some community members. Their use may also be appropriate when mosquito numbers are extreme or disease risk is high.

Perigen[®] 500 (active - permethrin) and K-O Tab[®] (active - deltamethrin) are two products that can be used for mosquito net dipping. Perigen[®] 500 can also be used for dipping clothes. Both of these products have repellent properties, and so they deter the adults from landing on

mosquito bed nets and clothes. In the event that an adult lands on the treated material, the result will be fatal for the mosquito.

Some of the advantages of using these products include:

- residual action in the right conditions, Perigen[®] 500 may only need to be applied every 6 months, and K-O Tab[®] every 12 months;
- easy to apply; and
- relatively cheap.

There are also some important factors to consider when using these products:

- not target specific; and
- lethal to fish and aquatic organisms if the product finds its way into waterways or other aquatic environments.

FOGGING

There are numerous active ingredients and associated tradenames registered for use for fogging in Western Australia. Some of these are detailed in the next paragraph, however for the purpose of simplicity; this section will focus on two active ingredients found in formulations applied using thermal and ULV aerosol generators.

The synthetic pyrethroids, cypermethrin (tradenames: Cynoff[®]25 ULV and Dipthor ULV); deltamethrin (tradenames: Cislin[®], Aqua-K Othrine[®]), phenothrin (tradename: Twilight ULV) and pyrethrins (tradename: Pyrocide) are all registered for use for fogging in Western Australia. Reslin[®] (active: Bioresmethrin) was previously widely used Australia-wide, but it is no longer commercially available, although some local governments may still be using up old stock. Other similar products containing Bioresmethrin are still available for use for fogging.

Another product which has been used in this state for many years (though less commonly used in recent years) is the organophosphate, maldison (tradenames Malathion[®], Fyfanon ULV and Hy-mal[®]). Like all insecticides, these actives have advantages and disadvantages. These have been outlined below for two of the different actives.

Synthetic Pyrethroid Products (e.g. Dipthor ULV, Pyrocide and Aqua-K Othrine[®]):

Advantages:

- they are odourless;
- they knock down mosquitoes quickly; and
- they have very low toxicity to mammals (humans)

Disadvantages:

- they are more expensive than other adulticides;
- they are lethal to aquatic life, particularly fish and must not be allowed to drift across waterways;
- they are not target specific;
- known respiratory irritant to humans; and
- can be toxic to birds.

Maldison Products (eg Malathion[®], Fyfanon ULV and Hy-mal[®]):

Advantages:

- cost, they tend to be cheap compared to other adulticides;
- they break down quickly in the environment;
- low toxicity to mammals (humans); and

• their impact on the aquatic environment is lower than other adulticides.

Disadvantages:

- They are malodorous (they stink!);
- don't kill mosquitoes quickly (not a quick knockdown);
- are not target specific; and
- are a known respiratory irritant in humans.

A BRIEF GUIDE TO A SUCCESSFUL FOGGING CAREER

A myth exists that fogging does not require any monitoring of the target population, or at the very least, only requires monitoring of adult numbers to see if the chemical treatments have been successful. To maximise the effectiveness of adulticiding treatments, both larval and adult monitoring of breeding sites is helpful to allow you to know whether larviciding has been effective and to anticipate the timing of the emergence of adult mosquitoes and establish the optimal time for adulticiding, if needed.

As with all mosquito insecticides, fogging chemicals must be applied in accordance with the product label. Most adulticides stipulate: 'DO NOT use over or immediately adjacent to any water body or water course. DO NOT contaminate streams, rivers or waterways with the chemical or used container.' This is because of the risk of toxicity to non-target aquatic organisms. For example, bioresmethrin should never be drifted across water bodies due to its toxicity to fish.

Mosquitoes will tend to congregate around their breeding site post-emergence to mate and obtain an initial sucrose meal. A window of opportunity exists then, to undertake an adulticiding treatment before the mosquitoes disperse.

Also remember that some aerosol generators can be used to apply larvicides. If you intend to purchase a machine for a mosquito control program, keep this in mind.

The time of the day that the fogging is undertaken is also very important. Fogging is most effective when the target species is active. This ensures mosquitoes have the greatest chance of coming into contact with the aerosol, as they are not harbouring in vegetation. Studies have shown that aerosols disperse more effectively in open to moderately open terrain.

Meteorological conditions have a major impact on the success of fogging. The parameters that need to be considered include:

- *Wind speed/ direction* Some horizontal wind movement is required to drift the aerosol across the target area. However, too much wind disperses the aerosol too quickly, reducing the time the aerosol is available for controlling the mosquitoes.
- *Humidity* Low humidity can greatly affect any aerosol that uses water as a carrier, potentially causing the droplets to evaporate before they reach the target mosquitoes.
- **Atmospheric stability** Turbulence can affect aerosol movement across the target area. It is best to undertake adulticiding when conditions will be most stable, this tending to be in the early morning or late afternoon/early evening.

Finally, make sure all fogging equipment is well maintained. Thermal, and in particular ULV aerosol generators, are finely engineered to deliver optimal performance. However, this performance can only be guaranteed by keeping equipment in a well-maintained state of readiness.

A lot of the maintenance these machines require is simple and can easily be done by the operator. Follow the manufacturer instructions and ensure that equipment is serviced as required. In remote locations, it might be advisable to keep a limited supply of spare parts handy, particularly parts with a limited working life that cannot be easily replaced. The equipment supplier will be able to give you advice with regard to this.

It is important to let others know about your intended fogging schedule. Members of the public with respiratory sensitivities may need to be warned and may request that you turn off the fogging equipment as you pass their residence. Householders with caged birds or fish ponds may wish to cover their cages and ponds to prevent exposure of their animals. It may also be advisable to warn the local fire brigade about your fogging plans to prevent unnecessary callouts in response to clouds of white 'smoke' generated by thermal fogging machines.

MOSQUITO INSECTICIDE APPLICATION - EQUIPMENT

There is a wide variety of equipment available for applying mosquito insecticides. This is reflected in both the cost and operational parameters of the equipment.

It is important to obtain full manufacturers and manual specifications in order to assess the suitability of the equipment that might be used in the control program. The following information is provided as a guide only, and is intended to provide a basic idea of the types of application equipment available.

HAND APPLICATION (WITH GLOVES!)

This technique is only applicable to the use of granular larvicides and only suitable for very small breeding sites e.g. neglected backyard swimming pools, small isolated pools in roadside ditches, septic tanks, stock troughs etc.

HAND OPERATED COMPRESSION SPRAYERS

These are suitable for small areas requiring treatment with liquid larvicides (wettable powders and emulsions). Some surface adulticides can also be applied using this equipment.

MOTORISED SPRAYERS

These can be either backpack or vehicle mounted and can be used for application of either granular or liquid formulations of larvicides and for small scale adulticiding operations.

THERMAL FOGGERS

These can be vehicle mounted or hand-held portable machines. They produce dense white clouds of fog which consist of insecticide and a distillate carrier for control of adult mosquitoes and disperse downwind for up to 100 metres. Take care when using thermal foggers near vegetation because of the risk of starting a fire.

COLD FOGGERS

These can be vehicle mounted, portable (backpack) or aerial attachments for helicopters or fixed wing aircraft. The latter are rarely used in Australia. These foggers produce an almost invisible aerosol spray of concentrated insecticide which disperses downwind for up to 100 metres. Some of these types of foggers also have the ability to be used in larvicide applications.

AERIAL APPLICATION (LARVICIDES)

Various outboard and inboard hoppers are used in both helicopters and fixed wing aircraft for granule application. Spray booms may also be available for application of liquid larvicides.

REPELLENTS

Regional or even localised mosquito control is often not possible, especially during disease epidemics that tend to occur following extreme conditions such as floods, when mosquito breeding is widespread. This is typically the case in remote areas where effective mosquito control is often cost-prohibitive, and WA has plenty of remote areas! In these situations the only means of preventing mosquito bites is using personal preventative measures which afford protection at an individual level.

Wearing long loose clothing when outside, and sleeping in a mosquito proof tent or under a mosquito net are all effective in reducing or preventing mosquito bites. However, when you simply cannot avoid exposure of skin to biting mosquitoes, repellents can be very effective in preventing mosquito bites.

Repellents are chemicals which are either extremely distasteful to mosquitoes, leading to avoidance by the mosquitoes, or which mask the olfactory (smell) attraction of a person to mosquitoes. Diethyl toluamide (DEET) has long been considered the most effective mosquito repellent, and it has a remarkable safety record. A range of DEET based repellents are available in Western Australia, containing various concentrations of DEET, some up to 80%.

The DOH recommends the use of DEET based repellents that contain up to 20% DEET and not more. Repellents with this concentration of DEET have been shown to be effective. As with all chemicals, the instructions on the use of the product should be followed. DEET based repellents do have warnings about the use of the product on young children and prolonged and excessive use of the product.

The DOH also recommends the use of repellents containing the active ingredient Picaridin, which is very similar in most regards when compared to DEET. Other active ingredients that have some mosquito repellency action include citronella oil, dimethyl phthalate, indalone and ethyl hexanediol. However, studies have shown DEET and Picaridin products to be more consistently effective. As a general rule with regard to the use of repellents on infants, children and pregnant women, it is recommended that you read the product label and use accordingly.

For further information on repellents:

http://medent.usyd.edu.au/arbovirus/mosquit/repellent_guidelines_2011.pdf

SURFACE OILS & FILMS

Surface oils are the oldest form of chemical mosquito control, having been used for many years before the introduction of pesticides and modern repellents. Like some insecticides, surface oils are not specific in their control of mosquito larvae, and are likely to have major impacts on other aquatic invertebrates that utilise the air-water interface. For this reason, they should never be used in natural environments. In the right situation however, they can prove to be an extremely effective larvicide.

Surface oils rely on the fact that mosquito larvae need to get to the surface of water to breathe. Oil and similar products (e.g. kerosene, paraffin oil and Aquatain AMF) reduce the surface tension of the water. This prevents the larvae from being able to attach to the surface to breathe, and so they drown.

Surface oils are not effective against all types of mosquitoes and so it is important to monitor the situation to determine if this would be a suitable control method. For example, Coquillettidia and Mansonia mosquitoes do not breathe at the surface, and therefore they are not necessarily controlled in this manner. These types also tend to breed in environments where there is a lot of natural vegetation present in the water, and the use of surface oils in these environments is not recommended. Mosquitoes can breed rapidly and in large numbers in rainwater tanks, causing huge nuisance problems, even in the metropolitan area. Surface oils are an effective method to control mosquito breeding in rainwater tanks. However, the DOH recommends that tanks should be maintained so they prevent mosquitoes gaining access to the water, by being completely sealed and screened at the inlet. Even the smallest of holes can be enough to allow an adult to enter and lay her eggs.

In situations where completely sealing tanks is not possible, kerosene mixed with castor oil (1% castor oil to 99% kerosene mix) can also be used, but should not be applied if the water is used for drinking.

Aquatain AMF® is a recently registered product containing polydimethylsiloxane that works in the same way as oils and other substances described above, in order to prevent mosquito larvae from breathing at the water surface. It is for use in standing water in domestic/suburban areas such as gutters, ponds, blocked drains, water tanks, septic tanks and old tyres. This product is not appropriate for use in natural environments or in treatment areas exceeding 100m2. Aquatain AMF® is not yet registered for use in drinking water.

USEFUL	INTERNET	SITES
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Organisation	Internet Address	What you'll find
Australian Pesticides and Veterinary Medicines Authority	http://www.apvma.gov.au	Lots of useful information, including the PUBCRIS database (details the registration status of insecticides)
MSDS online	http://www.msdsonline.com/	Over 300,000 MSDS's
Mosquito Control Association of Australia	http://www.mcaa.org.au/	Lots of very useful mosquito control information. Also has some great links to other sites.
Dept of Medical Entomology. Uni of Sydney & Westmead Hospital	http://medent.usyd.edu.au/	Lots of very useful mosquito and mosquito-borne disease information. The only Australian site of its kind.
Bureau of Meteorology	http://www.bom.gov.au/	Weather and tidal forecasts

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15. (S)-METHOPRENE; HISTORY, MODE OF ACTION, AND MONITORING

DAVID WALKER

Pacific BioLogics Pty Ltd

HISTORY

The existence of Insect Juvenile Hormone (JH) was discovered in 1965 and its molecular structure was solved in 1967. It was subsequently discovered that excess JH was detrimental to insect development and that JH levels decline during normal development of insects through the juvenile stages. JH levels are high during immature development and fall prior to pupation. JH is critical during larval growth, but it must be at low levels for the insect to undergo metamorphosis into adults.

Carl Djerassi worked for Syntex in the 1950's and 60's, he worked with hormones and was involved in the development of the first human birth control pill. Ironically he is sometimes known as the "Father" of human birth control! The discovery of insect JH got him thinking that insect control might be possible via hormones. He formed Zoecon Corporation in Palo Alto in the late 1960's (later to become Wellmark) and started work on insect JH analogues.

Initially 21 analogue compounds were synthesized, isolated, and screened. Of these; Methoprene, Hydroprene, and Kinoprene showed promise and later became commercially available. Initially these compounds were termed "biorational" by the USEPA and later "biochemical" compounds. They are now classified as Insect Growth Regulators (IGR) for resistance management purposes.

Isolation of methoprene was relatively easy, next came the hurdle of basic synthesis and isomeric selection. Basic screening showed activity on a variety of pests including mosquito larvae. Niche market use was most appealing as crop uses were perceived as impractical due to a number of reasons but most notably the slow action of methoprene. Today methoprene continues to be used for a range of pest control activities; in particular mosquitoes, cockroaches, fleas, and stored grain pests.

Developing formulations for mosquito control proved difficult. Methoprene is photo-liable and subject to rapid UV light degradation in outdoor environments. Microbial action is also another pathway for degradation. Formulating end use products took many years and it wasn't until the late 1970's that the first products were registered in the US. Activated charcoal, micro-encapsulation, and slow release substrates were used in the developed formulations.

Methoprene is a mix of two enantiomers, or configuration isomers. The lefthanded (S-sinister, ~90%), and the righthanded (R-rectus ~10%) isomers. It was found that the (R) isomer was far less active on mosquito larvae and had more non-target effects than the (S) isomer and so through the 1980's manufacturing processes aimed to remove the (R) isomer. This made manufacturing more expensive but gave a more active and environmentally safe molecule.

Rachel Carson's book "Silent Spring" published in the early 1960's caused pesticide use to come under increasing media, government, and public scrutiny. The US Food Quality Protection Act (FQPA) of 1996 significantly increased regulatory pressure on many classes of pest control products. This brought methoprene into the spotlight due to its unusual mode of action. Methoprene has been registered by USEPA for more than 30 years and has been very extensively studied. There have been 100's of non-target, acute and chronic effect, trials done with methoprene, and in spite of this, the updated USEPA Re-registration Eligibility Document (RED) Fact Sheet indicates minimal or low toxicity to non-target organisms. Typical margins of

safety are greater than 100 times application rates. High margins of safety combined with extremely low use rates and high efficacy, results in very useful products for mosquito control.

Today, all methoprene products used for mosquito control in Australia are formulated with (S)methoprene and application rates are extremely low. In many instances, efficacious applications of (S)-methopene are below detectable levels even with the most advanced laboratory equipment and the only way to test for (S)-methoprene presence post application, is with mosquito larvae bioassays.

MODE OF ACTION

(S)-methoprene is a true analogue of the mosquito's own juvenile hormone. During the fourth larval instar, juvenile hormone (JH) levels naturally drop to very low levels. If the insect is exposed to (S)-methoprene at this stage, JH levels are artificially raised and maintained at higher than normal levels. The higher than normal level of JH during the latter instar stages prevents the insect from developing into normal pupa. Depending on dose, mortality may occur as larvae or pupae; at low doses, emergence of sub-optimal adults may even occur. These adults may be deformed and/or show behavioural changes that prevent them from mating, flying, and biting.

(S)-methoprene is absorbed through the exoskeleton of the insect as well as being ingested during normal feeding. High doses can often cause larval mortality but at normal label rates mosquitoes die at the pupal stage as they do not get the hormonal message to emerge and eventually use up their energy reserves during this non-feeding stage. It is important to have (S)-methoprene present at pupation for the best efficacy, hence treating later instars gives better results. Once pupation occurs (S)-methoprene does not effect the mosquitoes if there has been no previous exposure. If the larvae are prematurely removed from the (S)-methoprene source it is possible that they can recover and continue to develop normally, however, adults that emerge in this instance are often compromised.

When mosquitoes are exposed to (S)-methoprene during any larval stage, their development time slows down considerably. It can be observed that development times stretch out to weeks instead of days. In some cases mortality occurs when larval habitat dries up and larvae simply run out of water.

MONITORING (S)-METHOPRENE EFFICACY

(S)-methoprene acts by inducing morphological changes which interfere with normal development. These effects are not immediately apparent. Because the effect seen is neither total larval mortality nor widespread mortality immediately following pupation, the number of emerged viable adults is the only criterion for accurately assessing control. Checks by dip counts during larval and pupal stages are valid only for subjective evaluation by trained inspectors familiar with the subtle observable effects of labelled rates of (S)-methoprene on larvae and pupae.

After application of (S)-methoprene to 2nd, 3rd, or 4th instar larvae, direct toxic effects on larvae are generally not likely to be observed. They will continue to develop and will pupate. Pupae will also live for a short time but will eventually die. Adults will not emerge. Infrequently, a few adults may be seen at the water surface, but will have abnormalities preventing flight etc and will not survive.

Subjective post application evaluation of (S)-methoprene treated areas is routinely practiced by many professional mosquito control experts. Inspectors who have familiarised themselves with subtle changes in appearance and behaviour of treated larvae and pupae advise that a

general determination can be made on whether or not an effective level of (S)-methoprene has been delivered to the breeding site. Some of the subtle changes to look for are:

- 1. Abnormally light coloured larvae and /or pupae.
- 2. Less than normal larval vigour or pupal mobility.
- 3. Malformation of treated larvae or pupae.
- 4. Pupae may tend to be straighter than their normal "comma" shape.
- 5. Failure of treated larvae to advance to the next instar stage of their development or to pupate within a normal time frame.

Any one or all of these abnormalities may be found in a treated area. Close observation during the first few uses of (S)-methoprene may allow the field operator to forecast results of future treatments.

APPLICATION CHECKLIST

Things to do and look for:

- 1. Treat 2nd, 3rd, and/or 4th instar larvae, not pupae or adults (1st instar larvae are so small they are not readily detectable).
- 2. Wait until treated larvae have pupated. Then collect pupae and transfer to a laboratory to observe for emergence of adults. This does not take into account if there has been any larval mortality and hence should always be considered an under-estimation of control.
- 3. Observe pupae for several days, since mortality of (S)-methoprene treated larvae generally occurs after pupation (careful observation is necessary since dead pupae decompose rapidly and thus are not easy to see).
- 4. You could monitor emerging adults at the treatment site. This requires that emergence traps be placed in treatment areas to capture adult mosquitoes as they emerge.
- 5. To accurately measure the control percentage a known number of larvae must be collected and placed in floating bioassay rings prior to treating the area. These can be monitored post treatment and pupae collected for adult emergence monitoring. Total control will be the number of caged larvae minus the emerged adults (if any).

Things that you should avoid doing:

- 1. Do not take dip counts of larvae after treatment for the purpose of performance evaluation. Normal looking larvae may be present.
- 2. Do not take dip counts of pupae after treatment for the purpose of performance evaluation. Normal looking pupae will be seen but may not develop into normal adults.
- 3. Do not think the treatment has failed if some adult mosquitoes are flying in the treated areas; they may have flown in from nearby untreated areas.
- 4. Do not treat again because larvae and pupae are present after application. This is normal. The effectiveness of (S)-methoprene treatments can only be measured by lack of adult emergence.

As with most mosquito larvicide applications, monitoring the results is usually observed and anecdotal rather than rigorously scientific. (S)-methoprene is no different in this regard; however, it is possible to monitor and predict efficacy of applications through careful observation and judgments based on field experience.
16. PRACTICAL CONSIDERATIONS FOR CHEMICAL MOSQUITO CONTROL

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INTRODUCTION

Operational aspects of mosquito control programs and equipment are discussed in detail in the chapter by Scott Severn in this manual. This chapter provides some additional practical information that mosquito control officers should use to ensure safe and effective chemical control of larval or adult mosquito populations. However, it is not intended as a 'stand alone' resource and should be referred to in conjunction with Scott Severn's chapter. Furthermore, the information will only be helpful if it is accompanied by a sound knowledge of the biology of the target species and the mode of action of the chemical being used - aspects that are covered in detail in other chapters of this manual.

Reference should also be made to detailed information in the manual of the Mosquito Control Association of Australia, as well as specific texts relating to specialised protocols or programs. Anyone routinely applying chemicals should undertake training in their safe use, handling and storage. Operators must also refer to information and instructions supplied with specialised equipment and chemicals to ensure a best-practice approach.

In particular, this chapter covers some of the basic equipment used in the application of larvicides and adulticides, personal protective equipment and clothing, safety considerations, maintaining a chemical inventory and calibration and maintenance of equipment.

EQUIPMENT

There are two main categories of equipment for mosquito control: operational equipment and equipment for personal protection and safety.

Equipment for the application of larvicides and adulticides for mosquito control varies from simple hand held spreaders, or even just gloves, to sophisticated ultra low volume foggers and aerial application equipment. The equipment required for your particular circumstance will be determined by the size, accessibility and number of sites to be treated, the life cycle stage(s) that are being targeted, the preferred chemical for the job, and ultimately your budget.

There are many brands and models of equipment marketed for mosquito control – far too many to mention here. However, some of the basic categories of equipment include:

- Measuring containers (for accurate measurement of chemicals)
- Mechanical or motorised knapsack (liquid/granule application for larval mosquito control)
- Pressure knapsack (liquid application for larval mosquito control)
- Thermal fogger (adult mosquito control)
- ULV cold fogger (adult mosquito control)
- Mister/mistblower (for application of liquid residual surface adulticides)
- Vehicles [car/light truck, boat (or other aquatic craft), quad bike, etc to access mosquito breeding sites and to transport equipment and chemicals]
- Secure, insulated shed with concrete floor (chemical storage)
- Helicopter (liquid/granule application for 'large-scale' larval mosquito control)

Personal protective equipment and clothing (PPE) is essential for any form of insecticide use and includes:

- Gloves PVC or similar
- Overalls
- Boots not leather
- Goggles and safety glasses
- Respiratory equipment with appropriate cartridge type
- Hat
- Face shield or safety glasses
- Ear muffs or plugs

Additional protective equipment may be required for specialised operational equipment or for applying particular chemicals or formulations.

SAFETY

Personal and public safety is critically important when applying chemicals, and must be given high priority. For these reasons it is essential to use and maintain PPE. Operators should undertake an accredited pesticide handling course, and managers should ensure that any contractors or operators they employ have received appropriate training in the safe handling and use of pesticides and hold a current pesticides licence with the Department of Health's Pesticide Safety Branch. Please also refer to the chapter by Geoff Harcombe entitled 'Perspectives on Pesticide Safety' on page 106 of this manual for more details.

It is a legal requirement that any container or packaging for mosquito control chemicals is supplied with a label. The label contains important information about where and when the chemical can and cannot be used, as well as information about correct application rates, appropriate diluents or carriers, required PPE, storage, shelf life/expiry date and more. Therefore, always read the label before using a chemical and use according to label rates and directions. Never apply a chemical in a manner that does not comply with label instructions or advice.

Any consignment of chemical of more than 30 litres or 30kg must also be supplied with a Material Safety and Data Sheet (MSDS). The MSDS and label contain important information on the product name and active ingredient, suppliers name and address, health hazards, precautions for use, safe handling and storage. Never use batches of chemicals with altered, damaged or missing labels.

Personal and public safety can be further enhanced by adhering to some additional simple principles. Avoid unnecessary handling of chemicals. Instead, calculate the amount of chemical and diluent/carrier required for a particular job and use it all. Ensure equipment is properly calibrated to avoid under or overdosing. Avoid spray drift, a problem that is most common with liquid applications where droplets of less than 150µm are required (e.g. ULV and thermal fogging). Make sure you advise the public about the nature, timing and location of chemical control. Under no circumstances over-apply chemicals: this is potentially dangerous to human health, is a waste (of chemical, and therefore your budget) and will have undesirable environmental impacts. Similarly, under-application should also be avoided as this will be ineffective (also a waste) and may promote development of resistance to the chemical in use (as well as other chemicals in the same group) in the target mosquito species.

MANAGING CHEMICAL STOCKS

Chemicals that have passed their expiry date may no longer contain the required amount of active ingredient. Therefore these should be disposed of appropriately and not used. Use of old chemicals may expose targets and non-targets to sub-lethal concentrations of the active ingredient, resulting in failed control and promoting development of resistance to the active ingredient.

Ordering and storage of chemicals is also an important consideration for mosquito control programs. Chemical requirements for the upcoming mosquito 'season' should be anticipated and stocks ordered accordingly. Do not wait until after a major rainfall or tidal event to do this as stocks are likely to be hard to obtain at short notice or if roads have closed due to the very event that has triggered your requirement to undertake control.

However, some chemicals have a very limited shelf life, so avoid stockpiling more than is likely to be used within the shelf life of a chemical formulation. Manage batches carefully, using oldest stocks first. This will be greatly facilitated by maintaining a detailed log book. Store all chemicals in an appropriate (secure), temperature controlled facility. Never store chemicals in the cabin of a vehicle. Note that some chemicals have strict requirements for their storage, such as a requirement for storage on a sealed floor or concrete slab. When purchasing new chemical stock, check to make sure there have been no changes to the label.

APPLYING CHEMICALS

A number of basic principles should be applied when considering applying an insecticide to control mosquitoes. These can be summarised into four simple points:

- Right insecticide (and formulation, with appropriate diluent and carrier)
- Right dose
- Right place
- Right time

You will be well on your way to ensuring effective and safe control of mosquitoes if you make sure that every step of your chemical control program fits these principles.

THE 'CHEMICAL MIXTURE'

Ensuring you are using the correct amount and dilution of a chemical is one of the fundamental, but often overlooked considerations for applying chemicals to control mosquitoes. The correct mixture will be a combination of the right amount of the chemical, a diluent (if required) and a carrier (if required). In the case of liquid larvicides and adulticides the mixture is often referred to as an emulsion. Calculating the correct mixture will be based on some or all of the following:

- Concentration of active ingredient in the supplied formulation (liquid, pellet, granule, etc);
- Recommended label rate for the situation;
- Size of area to be treated;
- Requirement for a carrier compound as well as diluent;
- Flow rate of application equipment;
- Viscosity of liquids;
- Desired droplet size (e.g. 10-20µm for ULV adulticides); and
- Rate of ground coverage (e.g. vehicle speed).

GRANULAR LARVICIDES

Granular larvicides can be applied by hand, granule applicator, mechanical (e.g. backpack) applicators and other means. Before applying, you will need to determine the flow rate of the

equipment. This can be done as a trial using old stock in a location that will not impact on the surrounding environment or members of the public. Alternatively, and depending on the equipment to be used, you may be able to identify the flow rate from the machine specifications.

Once the flow rate is determined, this can be adjusted to give appropriate application (e.g. 3kg/ha) for coverage speed (e.g. walking pace). Depending on the desired application rate, flow rate of your equipment and coverage speed, you may need to dilute granular chemical formulations further using a similar substrate (e.g. clean sand of similar diameter) to avoid over-applying. This is particularly pertinent when applying chemicals to small or patchy areas or when on foot rather than in a vehicle, which will cover more ground in the same time.

 Example: Backpack application of Prosand[®] [desired rate is 3kg/hectare (ha)]: Swath width of backpack is 10m At quick walking pace (6km/hr) this will allow coverage of 1 ha every 10 minutes Therefore backpack must be calibrated to 300g/minute to ensure correct application rate of Prosand[®].

LIQUID LARVICIDES (AND WETTABLE GRANULES)

These formulations can be applied by pressure knapsack, backpack liquid applicator, boom sprays and other equipment designed for liquid chemicals. Generally, these chemicals can be diluted in clean water. However, with some formulations [such as *Bacillus thuringiensis* var *israelensis* (Bti)] you will need to ensure regular agitation to avoid concentrates clogging supply lines and nozzles.

The flow rate of your equipment can be determined using water only, or refer to machine specifications. Once this is known you will need to vary the concentration of the emulsion/mixture or the flow rate to provide the correct application for your coverage speed (e.g. walking pace).

ADULTICIDES

Adulticides are generally used for emergency control of adult mosquitoes and other biting insects. They may also be the only alternative when larval breeding sites are too large or difficult to access, making larval mosquito control impossible. The main strategies used are thermal fogging, ULV (Ultra Low Volume) fogging and more recently, residual surface adulticides. The first two rely on the contact between tiny droplets of insecticide and flying adult mosquitoes, whereas residual surface adulticides are applied to surfaces on which adult mosquitoes are likely to land and be exposed to the chemical.

Fogging

ULV and thermal fogging are best applied in low wind conditions at and after dusk or around dawn, when the adults of most mosquito species are most active. Temperature inversions may assist droplet carriage and suspension. An average droplet size of $10-20\mu m$ (1 μm or micron = 1/1000 mm) is most effective against populations of flying adult mosquitoes. Droplets of under $10\mu m$ in size should be avoided as these will be subject to rapid evaporation and degradation of the active component. Large droplets (say >50 μm) should also be avoided as these will quickly fall to earth where they will have no effect on flying adult mosquitoes.

In dry conditions (with low humidity) it is important to use oil or other carriers (e.g. Carmel Carrier II) to coat droplets of some adulticides, thereby reducing evaporation and enhancing delivery of the chemical to the target mosquitoes. A number of different carriers are available for fogging and are listed below.

Carriers for thermal and ULV fogging				
Water (only for ULV fogging in humid environments – do not use water alone for thermal fogging) Oil-based carriers:				
Diesel (use neat)				
Kerosene (use neat)				
Carmel Carrier II (1:2 - 1:6 with water depending on humidity)				
Dynamate (1:2 - 1:6 with water depending on humidity)				
Ondina Oil – Shell				
Pharma 68 – Caltex				
Enerpar MOO2 - BP				

Some adulticides for thermal and ULV fogging already incorporate anti-evaporant oils in their formulation and therefore do not require additional carrier. Therefore it is important to read the label to determine the specific requirements for the product you are using.

Determining the correct amount and ratio of your chemical, carrier and diluent is critical in ensuring an effective (and legal) application. Following, are two examples of the considerations and calculations used for determining appropriate mixtures/ emulsions for a ULV fogger and a handheld thermal fogger.

Example 1: Formulation for ULV application of Aqua-K-Othrine[®] by large vehicle-mounted ULV fogging machine:

- Label rate = 50mL (of active)/hectare (ha*) (water-based application) *1ha = 10,000m²
- Dilution rate = 50mL per 450mL water (=1 in 10 dilution)
- Application rate of dilution (ie the formulation) = 500mL/ha
- Formulation tank of ULV fogger (e.g. MaxiPro4) = 58L
- \therefore 5.8L Aqua-K-Othrine[®] + 52.2L water (=1 in 10 dilution)
- Flow rate of machine is set at 250mL/min
- 30m swath will cover 0.5ha/min at 10km/hr (vehicle mounted equipment, travelling 167m/min)
- Sufficient formulation to treat ~116ha (should take ~3 hrs 52 mins to empty tank)
- [Alternatively, the flow rate of the machine could be increased to 500mL/min to allow the vehicle speed to be doubled to 20kph and cover 1ha/min (in 1hr 56 mins)]

Example 2: Formulation for application of Aqua-K-Othrine[®] by larger hand-held thermal fogging machine:

- Label rate = 50mL (of active)/hectare (ha) (water-based application)
- Dilution rate = 50mL per 4950mL water (=1 in 100 dilution)
- Application rate of dilution (ie the formulation) = 5L/ha
- Formulation tank of hand-held thermal fogger (e.g. lgeba TF 35) = 5.7L
- \therefore 57mL Aqua-K-Othrine[®] + 5.64L water (= 1 in 100 dilution)
- Flow rate of machine with Igeba dosage nozzle size 1.0 = 15L/hr
- Sufficient formulation to treat 1.14ha (should take ~22.8mins)
- ∴ need to cover a distance of 380m walking at 1km/hr, with a 30m swath fog, to accurately apply the 5.7L formulation tank to 1.14ha.

Calibration of new equipment and calculating the correct 'mixture or emulsion' for new chemicals may require some thought and time. However, once established, the procedure and mixture should

be written down so that it can be done routinely. For this reason it is essential that good procedural records are maintained and made available to all operators in your program.

Residual surface adulticides

This approach is being used increasingly in situations where residential, recreational or work areas are situated within mosquito flight range (dispersal range) of extensive breeding habitat that cannot be treated using larvicides for environmental, logistical or financial reasons. This includes employees or communities deployed in temporary housing (e.g. mining camps) or camping (e.g. military exercises) in mosquito-prone areas.

Several formulations of residual insecticides (e.g. bifenthrin) designed specifically for control of mosquitoes and biting midge are now available. These can be applied diluted appropriately in water using knapsack-style or pressure sprayers, motorised mist blowers or machine-mounted spray equipment. Residual surface adulticides must not be applied using ULV or thermal fogging equipment.

Residual surface sprays should not be applied to wet surfaces or immediately prior to or after rain, to avoid run-off of chemical. They should be applied as large droplets (a coarse spray) up to but not exceeding the point of run-off and in accordance with label rates per m² of area treated.

Residual surface sprays are designed to treat harbourage and resting sites of adult mosquitoes. Therefore, there may be some value in installing artificial surfaces such as shade cloth, or planting additional vegetation that can be treated in areas with sparse vegetation.

CALIBRATION AND MAINTENANCE

Calibration and maintenance of equipment is essential to ensure that chemical application is effective, not wasteful and doesn't adversely impact on the environment. There are a number of simple maintenance and servicing principles that will help to ensure that all equipment remains reliable and effective. These include:

- Replace all damaged or worn nozzles
- Check pumps and motors
- Test flow rates using old stock (granules) or diluent/carriers (liquids)
- Flush piping, hoses and tanks after every application
- Drain and flush all fuel tanks, formulation tanks pipes and lines immediately after application
- Keep log of hours of use, faults and malfunctions
- Stock spare parts: seals, nozzles and caps
- Service equipment as necessary and before upcoming season

Always remember to read and refer back to the two key sources of essential information related to chemical application:

Equipment: read manufacturer's instructions on use, safety and maintenance

Chemicals: read label and MSDS and use accordingly

If you are in any doubt about any aspect of your program, seek assistance from the equipment or chemical supplier, other operators, and the WA Department of Health (Mosquito-Borne Disease Control and Pesticide Safety).

17. CALIBRATION OF APPLICATION EQUIPMENT

DAVID WALKER

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KEY LEARNING OBJECTIVES FOR THIS CHAPTER

- 1. Understanding the importance of calibrating application equipment for mosquito control products
- 2. Learning the basics of calibration; enabling your to apply your knowledge to calibrating any type of application equipment and for any type of product.
- 3. Teach you the way to think like a "Bush Mechanic"
- 4. Ensure that you always read application equipment manuals and the labels of the products you apply
- 5. Enable you to confidently answer the questions "What rate did you use?" and "How did you work that out?"

THE IMPORTANCE OF CALIBRATION

All application equipment should be calibrated on a regular basis. The frequency will depend on the equipment being used and the operators involved. It's important to understand that waste and/or damage can be caused due to lack of accurate calibration of application equipment. Products should be accurately applied at the recommended label rates for the given situation.

- If too little product is applied the efficacy may be reduced
- If too much product is applied there is a greater potential for adverse environmental impacts
- Applying too much product can also be a waste of product that may have no discernable increase in efficacy
- Consistently using too little product will increase the probability of resistance developing to the active you are using
- Varying situations may require the use of different rates and will therefore require that you are able to adjust to those specific rates
- There may be compelling legal requirements to apply the label rates

CALIBRATION BASICS

The calibration of any application equipment can be performed with the understanding of three basic parameters; Speed, Swath, and, Output. Additionally, for liquid applications, you will also need to understand the role that Dilution and Droplet size plays to ensure calibration and applications are done accurately and with the best efficacy.

Speed – The speed refers to the speed that the delivery point moves. This could be the walking speed for ground based larviciding, the driving speed for ULV space applications, or the speed that the helicopter flies for aerial applications. This speed has an inverse relationship to the application rate, i.e. if other parameters stay the same, the faster the speed the lower the rate and vice versa. The speed determines one side of the application area per time. All equipment has a minimum and maximum speed that it will tolerate.

Swath – The swath refers to the spread of product from the delivery point. It may be in one direction or both sides of the delivery point. Usually the amount delivered will taper off at the extremes of the swath requiring some overlap. The swath determines the second side of the application area per time. Swaths may be determined by maximum spread or assigned within the equipments swath limits.

Output – The output is the amount of product delivered per time. This has a direct relationship to the application rate; the higher the output, the higher the rate. Adjusting output (including changing dilution) is the easiest way to fine turn the rate.

Dilution - For liquid applications, rate can be fine tuned by changing the dilution. It is an easy and accurate way to adjust rate when neat application of liquids are not required. Dilution can also effect penetration of product through canopy if liquid larviciding or ULV space spraying. It can also help give better coverage when liquid formulations are concentrated. If diluted too much, application equipment may not be able to deliver enough product and formulation tanks may not hold enough product to give efficient applications.

Droplet size – Droplet size for liquid applications will affect the efficacy of your application. This may be due to better drift, better deposition, better impingement on adults, better coverage, minimised evaporation, and maximised canopy or barrier penetration. Droplet size is a very complex subject that can't be covered in a few pages. It is best to understand that the droplet size is important and that advice should be sort if you are unsure of the requirements for your application.

COLLECT, MEASURE, AND CALCULATE

Once you understand the basics of the parameters above, all that is left to do is to measure them, then calculate your rate. It is then simply a matter of adjusting a parameter and then recalculating until you hit your target rate. There are many ways to do this. You can assign a couple of parameters then adjust the other to arrive at your target rate. The best way to understand this process is to get in the field and do it. You will quickly work out the best way to measure each parameter with the resources you have. Some equipment will come with manuals that have output values for various formulations so it is always a good idea to read the equipment manuals first as this can give you a great starting point for the basic parameters. Below are a few examples of how you might measure the basic three parameters. There are endless ways to do this.

Speed

- GPS
- Car speedometer
- Use time to cover a distance or distance covered in a time
- If measuring speed for ground application it is best to measure speed during an actual application

Swath

- For high volume liquid applications you can spray a cemented area to see the spread
- Use a line of cloth catch-bags for solid formulations
- Use a line of die-cards for aerial liquid applications
- It can be useful to use a swath width within the equipments capability i.e. if you can spray out to 6 meters at a stretch but can easily spray out to 4 meters then you may like to use 4 meters as your swath.

Output

- Spray liquid into a measuring cylinder for a given time
- For small hand-held equipment you could weigh the chemical tank after spraying for a set spray time
- Collect solid formulations over a set time into a catch-bag and weigh the collected product
- Fill a liquid or solid formulation tank with known amount of product and measure the time it takes to empty the tank

BRINGING IT ALL TOGETHER

Once you have a good understanding of all of the parameters and how to measure and adjust each one of them you are ready to calibrate your equipment. Calibration should be done for new equipment, when products are changed, and at regular intervals such as each season.

It is easiest to start by using 1 minute as your time unit and set or measure two parameters and lock them in. Then adjust the third to reach the target rate. For liquid applications you can use dilution to adjust the rate also. This is particularly true if you want a certain total volume for canopy penetration or for best droplet generation.

Below are three examples of calibrations with explanations of assumptions together with the mathematical workings. Work through these examples and make sure you understand how the end result was arrived at. Remember that understanding the general process is the key to being able to calibrate your particular application equipment.

EXAMPLE #1

High Volume Liquid Larvicide application using a truck mounted 500 Litre tank, a hose reel, and two operators on foot.

- We are going to use 1 minute as a time unit
- It is decided that the application will be done by two operators. One controlling the application wand and one helping with the hose. The vehicle will be parked in a suitable position and the application will be done along the entire reach of the fully extended hose. And then the vehicle will be moved so the next section can be treated.
- The product being used has an application rate of 1000 ml/Ha and is water dilutable
- Using water in the spray tank the operators "apply" the water to a cemented area at a speed and sweep of the application wand which they thought was sustainable in a normal larval habitat.
- In one minute the operators cover a distance of 33 meters of the cemented area.
 ⇒ 33 m per minute (SPEED)
- A comfortable sweep of the wand wetted a 3 meter wide area (SWATH)
- With the wand set on a course spray the operators sprayed into a large measuring jug and found that the sprayer had an output of 5000 ml/min (OUTPUT)

If swath is 3 m and 33 m is covered in 1 minute

- \Rightarrow in 1 minute 3m x 33 m or 99 m² is covered
- \Rightarrow or 99 m² / 10000 m² per Ha = 0.0099 Ha per minute
- ⇒ If the target output for the product is 1000 ml/Ha and we treat 0.0099 Ha/min
- \Rightarrow (1000 ml/Ha) x (0.0099 Ha/min) = ~ 10 ml/min target rate for product

Since we measured the output at 5000 ml/min and we need to put out 10 ml of product/min we need to dilute the product with water at a rate of 10 ml product + 4990 ml water

- ⇒ 1:499 dilution rate
- \Rightarrow Use 1 Litre of product and fill the 500 Litre tank with water.

EXAMPLE #2

We are going to do a ULV space spray application using a truck mounted ULV space sprayer with an operator driving the vehicle

- We are going to use 1 minute as a time unit
- The product being used has an application rate of 24 ml/Ha and is oil dilutable

- It is decided that the vehicle can be safely driven at 10 km/Hr while doing the application
 ⇒ 10000 m/Hr = 167 m/min (SPEED)
- Literature confirms that the ULV sprayer is capable of generating a lethal spray cloud out to 300 feet (91.4 m) but the roads being driven on for the treatment are 50 meters apart so the swath is set at 50 m (SWATH)
- Research also indicates that flow rates of ~ 200 ml/min with a pressure of 6 psi in the spray head will give droplet sizes ~ 20 microns and will give good penetration through the built-up area being treated. The sprayer is filled with oil and the spray line is disconnected and the flowrate is adjusted to 200 ml/min (OUTPUT)

If swath is 50 m and 167 m is covered in 1 minute

- \Rightarrow in 1 minute 50 m x 167 m or 8350 m² is covered
- \Rightarrow or 8350 m² / 10000 m² per Ha = 0.835 Ha per minute
- ⇒ If the target output for the product is 24 ml/Ha and we treat 0.835 Ha/min
- \Rightarrow (24 ml/Ha) x (0.835 Ha/min) = ~ 20 ml/min target rate for product

Since we set the output at 200 ml/min and we need to put out 20 ml of product/min we need to dilute the product with water at a rate of 20 ml product + 180 ml diluent oil

- ⇒ 1:9 dilution rate
- ⇒ 1 Litre of product to 9 Litres of diluent oil

EXAMPLE #3

We are going to do a granule larvicide application using a quad-bike fitted with a C-Dax rotary spreader.

- We are going to use 1 minute as a time unit
- The product being used has an application rate of 4 Kg/Ha
- Based on a GPS reading it is decided that the quad-bike can be safely driven at 10 km/Hr while doing the application
 - ⇒ 10000 m/Hr = 167 m/min (SPEED)
- Using a line of cloth collector bags set on the ground a number of passes confirms that the C-Dax spreader can throw product relatively evenly out 8 meters either side of the quadbike i.e. 16 meter spread (SWATH)
- With the slide open about half way the product flows steadily onto the spinning disc. Measuring the flow into a bucket shows about a 1.5 Kg per minute flow and doing a number of runs to empty at this setting with a known amount of product in the C-Dax hopper confirms that the flow-rate of the granule product at this setting is 1.5 Kg per minute. (OUTPUT)

If swath is 16 m and 167 m is covered in 1 minute

- \Rightarrow in 1 minute 16 m x 167 m or 2672 m² is covered
- \Rightarrow or 2672 m² / 10000 m² per Ha = 0.2672 Ha per minute
- ⇒ If the target output for the product is 4 Kg/Ha and we treat 0.2672 Ha/min
- \Rightarrow (4 Kg/Ha) x (0.2672 Ha/min) = ~ 1 Kg/min rate is needed

We could either increase the application speed or adjust the output down. Since it is decided that 10 Km/Hr is about the fastest safe speed we can do an application at, we'll adjust the slide in until our target rate of 1 Kg/min is reached. This may take a few goes but eventually you will reach your target rate and complete the calibration. Once the slide position for 1 Kg/min is found it should be marked and locked into position.

SUMMARY

- Always calibrate application equipment
- Think SPEED, SWATH, OUTPUT
- For Liquid applications also consider DILUTION and DROPLET SIZE
- Set or lock-in the first two parameters, and use the third to adjust the rate. You can also use dilution to adjust the rate if using liquid formulations.
- To help you calibrate equipment, cut some corners, and stay within regulatory requirements always read:
 - Equipment manuals and brochures
 - Equipment specifications
 - $\circ \quad \text{Product labels} \\$

Gaining an understanding of the basics set out in this chapter will enable you to apply the same logic to ALL application equipment so that you can calibrate any equipment for any formulation.

18. MOSQUITO CONTROL EQUIPMENT

SCOTT SEVERN

City of Mandurah

(Based on chapters from earlier course manuals by Nick Long, Mark Sheppard and Rob Hill.)

BACKGROUND

As discussed in the chapter 'The Chemical Control of Mosquitoes' on page 118, there are different approaches to mosquito control that can be used. Larviciding and adulticiding are two chemical control methods used within a successful mosquito program. As the control agents used are different, so too is the equipment required to apply the various forms of chemical. The different types of applicators, practical and maintenance considerations will be discussed in this chapter.

Some basic points to consider prior to treating are: -

- What lifecycle stage the mosquitoes are at (larvae/adult)
- How big the area to be treated is
- Environmental considerations eg. Access to site, weather conditions, non-target organisms
- Proximity to population
- Occupational Health and Safety & Environmental issues in respect to third parties, yourself and non target organisms.

BASIC EQUIPMENT REQUIRED FOR A MOSQUITO CONTROL PROGRAM

A. Larval monitoring

Dipper (soup ladle) with depth indicator on handle Pipette for larval/pupal samples Sample jars Note book or information sheet for data Pen (waterproof) or pencil Map of areas PVC boots or waders Thermometer for taking water temperature Petri dish Filter Paper Emergence Containers Larval traps

B. Adult monitoring

EVS/CO2 traps with batteries Collection jars Data sheet Dry ice Trap/Breeding site location map Esky

C. Mosquito identification

Adult and larval keys Dissecting microscope (6- 40x) Light source Jewellers forceps Pipette Petri dishes

D. Personal Protective Equipment (PPE)

Mosquito repellent Long sleeve shirt and trousers Boots - leather ankle height, steel capped, rubber soled, impervious or steel capped rubber Broad rimmed washable hat Mosquito net Sun screen Drinking water Communications equipment First Aid kit Gloves – PVC or Nitrile Reflective vest Respiratory equipment including suitable cartridge Face shield Goggles and safety glasses Ear muffs or plugs

E. Larviciding equipment

Granule applicator Liquid applicators Safety equipment (see above) Data sheet for recording chemical use Fuel containers for powered equipment Measuring jugs/cylinders. Scales Funnel Briquetting equipment. Dye cards for liquid application assessment

F. Adulticiding equipment

Thermal or ULV fogger Blower/ Mister Map of area to be fogged Data sheet for recording chemical use Safety equipment (see above) Flashing light

G. Other

Suitable vehicle for equipment and conditions Storage area for chemicals and equipment Wash down area to clean equipment Tools for general maintenance Spare parts for equipment. MSDS for chemicals/products being used Boat for access to isolated areas

NB: IT IS IMPORTANT THAT CHEMICAL HANDLERS HAVE A KNOWLEDGE OF PESTICIDE SAFETY. THIS SHOULD BE OBTAINED THROUGH COMPLETION OF AN APPROPRIATE TRAINING COURSE.

LARVICIDING

GRANULAR FORMULATIONS

There are a number of methods available for the application of granular larvicides, which are designed for coverage of small to extremely large areas. Originally most equipment was derived from the agricultural industry, however there now specific mosquito control equipment available today.

1. Bucket and glove

Operation: This technique is most appropriate for treatment of very small breeding areas and can only be used with granular formulations. For better coverage ensure any wind is blowing from behind you and this will also prevent any chemical being blown into your face.

Practical considerations: Obviously the area that can be covered is limited. Great care is needed to avoid over application. For example, an average handful (50-60gms) of a chemical that has a label application rate of 4kg/Ha (e.g. Prosand) will cover 125 to 150m² (an area of roughly 11.7m x 11.7m). Obviously, this will vary depending on the label rate for different chemicals or formulations. If a large area is to be treated, consider taking two containers half filled with granules. A 20kg bucket of granule larvicide can become very heavy in a short period of time especially in wet, saltmarsh areas.

Calibration: Get a rough idea of what 100 square metres looks like and practice applying the correct (label) rate using sand only. For large areas, other application methods are more appropriate because of the difficulty in ensuring an even and comprehensive coverage.

Maintenance: Self-explanatory. It is advisable to maintain a reasonable level of fitness.

2. Hand operated spreader

Operation: Basically a person operated spreader that hangs around the neck. The handle, when turned, spins a disc which the larvicide falls onto through a controlled gate. The gate can be adjusted to deliver various rates as required, with the direction in which it is spread also adjustable.



Figure 15: Hand operated granule spreader (Source: City of Mandurah)

Practical considerations: This type of spreader is usually hung around the neck. Whilst only weighing 8kg when full, this can still put strain on the neck and back muscles. Excellent for

areas of small to medium size. When using any chemical, always read and follow the label thoroughly prior to use, including use of appropriate PPE.(Note: be sure to close gate whilst moving around site as larvicide will continue to flow and loss of product is possible)

Calibration: Fill the spreader with the larvicide. Standing still, turn the handle and note the area covered by the chemical. Attach an empty bag to the outlet. With a constant rate, turn the handle for 1 minute. Empty the bag and weigh the contents. Do this a number of times to work out an average weight as this can vary. Using **Formula 1** (page 160) will give the amount being applied per hectare. Adjustments can be made by closing or opening the gate.

Adjusting your speed of travel during application will alter the amount of product being applied/ hectare. This may need to be taken into consideration during the calibration process.

Maintenance: Empty the spreader thoroughly after use. Clean any granules away from the opening and the adjustment handle. This area can become clogged with granules and set hard, causing the mechanism to seize. During the off season some parts of the machine can be removed and cleaned to ensure good operation when required.

3. Motorised spreader

Operation: This unit comprises a 2-stroke motor coupled to a blower and tank combination that is carried on the back. Granular larvicide is poured into the tank with air being forced in from the blower. The larvicide is blown out through the tank into the delivery tube and out onto the treatment area. The rate is regulated by twisting the delivery tube which opens an aperture allowing more chemical through. This is marked on the outside of the delivery tube for reference. The motor speed is controlled via a throttle handpiece located on top of the delivery tube that also varies the distance the chemical is dispersed.



Figure 16: Motorised spreader/mistblower (Source: Stihl website)

Practical considerations: The blower unit is quite heavy weighing approximately 25kg when full. With this in mind it is advisable to start the motor and put the unit on from the back of your vehicle. Trying to start the motor and put the unit on is near impossible in the saltmarsh areas. The unit can disperse granules over a distance of 15 metres in still air but this can be increased with the wind behind you. The motor is quite noisy so hearing protection is advisable.

Calibration: Fill the tank with granule larvicide. Attach a bag to the end of the granule delivery tube. With the motor running at full speed twist open the delivery tube to a pre-determined mark. Time this output for one minute and use **Formula 1** (page 160) to work out the amount being applied per hectare. Do this a number of times to work out an average weight as this can vary.

Maintenance: Ensure that the fuel tank has clean two-stroke fuel in it. Empty the chemical tank thoroughly after use. Become familiar with the mechanics of the machine and how it works. Referring to the operator's manual is a good start. Parts such as the spark plug, throttle cable and fuel hose are required to be regularly checked to ensure the equipment runs well. Do not leave chemical in the tank as it can cause corrosion.

4. Aerial Application

Operation: In Australia, helicopters are the most commonly used aircraft for mosquito control. Hoppers for spreading a granular larvicide are fitted either side of the helicopter or can be slung under the aircraft. These hoppers can be either mechanical or air operated. This refers to the method used to disperse the chemical from the hoppers which are controlled by the pilot. Mechanical hoppers utilise a spinning disc which has catch plates on it. The chemical flows through an adjustable aperture, onto the spinning disc and out through control vanes onto the treatment area. The rate applied per hectare is adjusted by opening or closing the aperture above the disc. Swath width covered by mechanical hoppers is around 16 metres (8 metres each side of the helicopter).



Figure 17: Mechanical hoppers being filled

(Source: Department of Health)

Air operated hoppers use a constant flow of air to distribute the granules. With less contact on the granule, the air operated hoppers have a number of advantages over the mechanical driven type. The air driven granules cover a greater distance out of the hoppers usually reaching a swath width of 20 metres (10 metres each side of the helicopter) and also give a more uniform spread pattern than the mechanically driven hoppers.



Figure 18: Air operated hoppers

(Source: City of Mandurah)

Practical considerations: Observing the flow of the larvicide can be difficult from the helicopter depending on the terrain below. Most hoppers will have a window or an electronic sensor that will let the pilot know when the hoppers are nearly empty. Wind is a factor that can influence a successful aerial treatment. High or gusty winds not only blow the granules off target but also make it dangerous to fly. With heights of 10 to 15 metres at speeds of around 40 knots, gusty or high winds are not a welcome addition to the aerial application. A copy of the Beaufort Wind Scale for estimating wind strength is provided at the end of this chapter.

During an aerial treatment, it is highly advisable for the observer to wear an approved helmet. These are designed to ensure safety and provide communication between pilot and observer and are essential for a successful aerial treatment.

Some other considerations to make prior to undertaking an aerial treatment may also include:

- 1. Pilots experience on type of helicopter being flown as well as total flying hours.
- 2. Civil Aviation Safety Authority (CASA) approval for low-level flying and agricultural rating where required.
- 3. The aircraft is fitted with CASA approved communication equipment and emergency related equipment eg. Emergency Locater Beacon.
- 4. Carrying capacity of the aircraft being used and adhered to.

The observer should have local knowledge, not only of the areas to be treated, but of other points of interest such as power lines and high tree lines. Enjoying flying or at least not succumbing to motion sickness is an advantage being the observer, as the flights may get a little bumpy at times. The area covered from flight to flight should not vary provided the helicopter is flown at a constant speed and the hoppers have been calibrated correctly. For example if Prosand[®] is used at the rate of 3.5 kg/Ha with hoppers that hold 160kg then approximately 45.71 hectares should be covered with each load.

Calibration: Correct calibration of the hoppers is imperative for a successful aerial treatment. Due to the large area usually being covered, an error in the application rate either way,can result in either over application or a sub lethal dose. There are two methods used to calibrate the hoppers for an aerial treatment. The first is by way of filling the hoppers with a small quantity of product, attaching a mesh catch bag over each hopper outlet and catching the amount of product distributed for 30 seconds. The quantity caught is then weighed and can be compared to a calibration table that has various rates pre-calculated. This can then be used to work out whether the hoppers need adjustment to achieve the desired rate of application. This should be done prior to any treatment and before hoppers are filled. This should also be carried out during and after the treatment to see if the hoppers have maintained the settings. Below is the formula used to work out these rates:

{Swath width (m) x ground speed (knots) x application rate (kg/ha)} / 324 = kg/min

This is then divided by 2 to give the kg/30sec and then divided by two again to give the kg/30sec per side/hopper.

The second method involves the use of catch bags which are set across the known swath width of the hoppers. These funnel shaped bags measuring 1m x 1m catch the chemical as the helicopter flies over the catch bags at a standard height and speed for application. The contents of the 8 catch bags are then weighed to determine the rate per hectare being applied. Not only does this give the rate being applied but can also indicate any problems with the hoppers. Catchbags can also be set up on the breeding sites to be treated to back up the previously mention calibration methods and may also to provide important information as to the way the product is being broadcast on a particular site or to a specific area within a treatment site.

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Figure 19: Catch bag setup



Figure 20: Helicopter flight over catch bags

(Source: City of Mandurah)

Maintenance: Often helicopters in Australia are used on a contract basis and as such the contractor usually performs maintenance. However some simple tips to keep the equipment in operational condition are: -

- 1. When filling hoppers for the catchbag calibration only fill with a small amount of product as equipment failure may result in product having to be removed from hoppers.
- 2. Check the condition of the product prior to introducing to hoppers damp product can clog up the delivery system.

- 3. Never assume that the hoppers have already been or remain calibrated always calibrate the hoppers as different products have different application rates and vibration can affect settings on adjustment controls
- 4. Inspect the hoppers prior to filling.
- 5. Never leave any granules in the hoppers as they may be used for a number of applications eg. Fertiliser spreading. Disused chemical can become affected by moisture and harden.
- 6. Do not stand or place heavy objects on the hoppers.
- 7. Ensure the lids are closed after filling the hoppers.
- 8. Keep the hopper outlets free from obstruction.

LIQUID FORMULATIONS

1. Compression sprayer

Operation: Ideal for pest control and industrial applications. Available in 5 litres or 12 litres.



Figure 21: Compression sprayers

(Source: Marolex Website)

Practical considerations: Used for small areas that need to be treated. The difficulty with these types of units is that you are required to interrupt your spraying, place the unit on the ground and use the pump to again pressurise the unit. This type of sprayer can be used to apply Bifenthrin barrier treatments - practically around households.

Calibration: Clean water is required for this operation and the size of the area to be treated will determine the amount of water to be carried to the treatment site(s). Brass nozzles are harder wearing and hence last longer than ceramic or plastic nozzles. Ensure correct PPE is worn during filling and use. Elbow length gloves, cartridge respirator and protective glasses may need to be worn. Always check and follow the label for the specific chemical and formulation you are using.

Maintenance: Never leave chemical in the tank. After use, flush the tank, hose and wand with clean water. Some chemical will cause corrosion and can erode seals, o-rings etc. To prevent this, Vitron o-rings and seals should be installed. Inspect and clean filters regularly so as to avoid blockages. Inspect the hose and wand sections on a regular basis for cracks or holes.

2. Knapsack sprayer

Operation: A back mounted unit with a 10-15 litre tank that is operated by a piston pump. The piston is actuated via the hand lever and forces compressed air into the backpack. The chemical is released through a hose to which a wand controls the amount and dispersal pattern of the liquid larvicide. Constant pressure is maintained by the continued movement of the hand lever. Battery operated knapsack sprayers are also available and these can make hand treatments less physically demanding. The unit consists of a 12 volt rechargeable battery pack located in the base of the unit that powers a pump with various speed settings. The battery can provide several hours of intermittent use and can be recharged over night ready for the next day's work.





Figure 22: Knapsack sprayer

(Source: City of Mandurah)

Practical considerations: The knapsack sprayer when full is quite heavy (15-20kg) therefore adequate padding on the straps is essential. Clean water is required for this operation and the size of the area to be treated will determine the amount of water to be carried to the treatment site(s). Brass nozzles are harder wearing and hence last longer than ceramic or plastic nozzles. Always check and follow the label for the specific chemical and formulation you are using.

Calibration: Fill the knapsack with clean water. Spray for 1 minute into a measuring jug and record the amount of liquid captured. Do this a number of times to establish an average as this can vary. Once an average has been calculated use **Formula 2A** (page 160) to determine the output of the sprayer in litres per hectare. From this, use **Formula 2B** (page 160) to determine the rate of larvicide to add to the tank.

Maintenance: Never leave chemical in the tank. After use, flush the tank, hose and wand with clean water and drain. Some chemical will cause corrosion and can erode seals, o-rings etc. To prevent this, Vitron o-rings and seals should be installed. Inspect and clean filters regularly so as to avoid blockages. Inspect the hose and wand sections on a regular basis for cracks or holes. Batteries should be disconnected.

3.All terrain vehicle sprayer

Operation: This comprises of a large spray tank; an electrical pump usually 12-volt and a hose with a handpiece attached. The 12-volt pump is connected to the battery supply of the vehicle carrying the spray tank. The pump delivers the chemical through the hose and out the adjustable handpiece or lance (Figure 9).



Figure 23: 12-volt sprayer mounted on ATV

(Source: City of Mandurah)

Practical considerations: The use of an All Terrain Vehicle (ATV) such as a quad wheel bike will greatly enhance the size of the area that can be covered by the operator. It is also highly advantageous to have two people working together; one operating the ATV and the other treating. This treatment method is best suited to easily accessible areas that can be easily navigated, without damaging the environment or tangling up the hose.

Calibration: Fill the All Terrain Vehicle sprayer with clean water. Spray for 1 minute into a measuring jug and record the amount of liquid captured. Do this a number of times to establish an average as this can vary. Once an average has been calculated use **Formula 2A** (page 160) to determine the output of the sprayer in litres per hectare. From this, use **Formula 2B** (page 160) to determine the rate of larvicide to add to the tank.

Maintenance: When finished spraying, flush the empty tank and pump with clean water and check the filter is free of foreign matter. Remove the spray unit from the ATV and store out of the weather. Regular maintenance of the pump is required such as checking the diaphragm, valves and electrical system.

4. Vehicle mounted sprayer



Figure 24: Ute mounted sprayer

(Source: Garrards Website)

Practical Considerations: When designing the layout of this type of spray unit, consideration must be given to chemical and equipment security and Occupational Safety and Health issues - reaching, lifting, climbing onto unit, etc.

As all the equipment is always exposed to the elements, quality equipment which is UV protected should be selected. A protective cover over the hoses and pump is a good idea. Tanks come in a variety of sizes as do the pumps and hoses. Smaller diameter hoses will be easier to handle in difficult terrain however the assistance of another operator can make life a lot easier. Automated retractable hose reels although expensive can also be worthwhile investment. Although tanks will come with filling levels already marked its worth while checking these levels to ensure they are correct and dilution rates are met. The tanks can also be utilised as a portable water source for knapsack and misting operations. A dump outlet fitted to the tank will also enable the tanks to be flushed easily.

Calibration: Fill the vehicle mounted sprayer with clean water. Spray for 1 minute into a measuring jug and record the amount of liquid captured. Do this a number of times to establish an average total. Once an average has been calculated use **Formula 2A** (page 160) to determine the output of the sprayer in litres per hectare. From this, use **Formula 2B** (page 160) to determine the rate of larvicide to add to the tank.

Maintenance: When finished spraying, flush the empty tank and pump with clean water and check the filter is free of foreign matter. Some chemical will cause corrosion and can erode seals, o-rings etc. To prevent this, Vitron o-rings and seals should be installed. Regular maintenance of the pump is required such as checking the diaphragm, valves and electrical system. During periods of inactivity or long extended storage the tank and pump should be flushed and drained.

It is important to remember the chemical stored in the hoses. As an example, 100 meters of 12mm hose can retain in the region of 15 to 20 litres of chemical.



Figure 25: Van mounted sprayer

(Source: Pest Education Services & Training)

Practical considerations: This unit has all the spray equipment built into the back of a van and has been installed with automatic, computerised, dosing equipment which provides accurate mixture of chemicals with water and provides you with a digital display for both chemical used and emulsion sprayed. The water reservoir in this unit has been tailor designed using an aluminium tank.

It is important that a perfect seal between the driving area and rear storage is achieved.

Calibration: Fill the vehicle mounted sprayer with clean water. Spray for 1 minute into a measuring jug. Do this a number of times to establish an average total. Once an average has been calculated use **Formula 2A** (page 160) to determine the output of the sprayer in litres per hectare. From this, use **Formula 2B** (page 160) to determine the rate of larvicide to add to the tank.

Maintenance: Like all machinery and equipment, the pump, motor and area within the van should be cleaned immediately after use and maintain in line with manufacturer's specifications. Some chemical will cause corrosion and can erode seals, o-rings etc. To prevent this, Vitron o-rings and seals should be installed.

It is important to remember the chemical stored on the hoses. As an example, 100 meter of 12mm hose can retain in the region of 15 to 20 litres of chemical.

5. Aerial application

Operation: The aerial application of liquid formulations is integrated within many mosquito management programs throughout Australia. Especially those where tidal conditions are more predictable, treatment is required to vast areas, and weather conditions are conducive to this type of treatment method. Many large programs are able to use this method at low dilution rates of between 3 and 5 lites per hectare. Liquid applications are delivered via Helicopters of various sizes and capabilities with delivery systems similar to that used in aerial agricultural spraying. A boom, supporting a number of nozzles at set intervals is attached to the underside of the helicopter. The liquid formulation having already been mixed is pumped into a storage tank attached to the underside of the helicopter. The liquid S-methoprene and *Bti* are applied via this method.

Many helicopters that are set up for liquid application have DGPS fitted. This is a guidance system that allows the pilot to position the helicopter so that the product is distributed evenly and precisely. This information can then be downloaded on to a mapping system and the exact details of how a particular site has been treated can be assessed. This is extremely useful data to both program managers and pilots that can assist in evaluating treatment processes and post treatment assessments.

Practical considerations: The dilution rate, droplet size, larval stage, site conditions and weather conditions are just some of the factors that need to be considered when in the planning stages of any aerial liquid treatment. The droplet size produced by different nozzles can range in size from anywhere between 20 and >300 microns. For most liquid larvicide applications a droplet size of between 200 – 300 microns is generally aimed for to assist in canopy and vegetation(e.g. *Juncus*) or samphire (*Sarcocornia*) penetration and also to minimise spray drift that is factor to be considered with the aerial application of liquids. Use of a liquid larvicide requires a large quantity of clean, fresh water that logistically may not be available. The entire spray system must be kept in good order. A small amount of dirt can block filters and nozzles when least expected such as halfway through an aerial treatment, requiring a landing to clean out the blockage. Agitation within formulation tanks may also be required for some larvicides such as Bti.

As with granular applications wind is a major factor when planning a liquid application and treatments are ideally undertaken in low wind conditions with temperature and relative humidity also a limiting factor. Still hot conditions are not ideal for aerial application of liquids.

Flight heights associated with liquid applications are usually lower than that of granular applications (4 - 5m) whilst application speeds are higher. This needs to be taken into consideration where terrain or vegetation may pose a danger to the pilot/helicopter or limit the ability to treat a site with dense tree canopy.

In addition to this breeding sites that encroach on residential areas may also need to be accessed prior to any aerial liquid application.



Figure 26: Aerial larvicide application

(Source: City of Mandurah)

Calibration: Use **Formulae 2A and 2B** to calibrate the booms and also for calculating the amount of chemical to add to the tank. The output of all the nozzles will need to be captured for a period of one minute. Some variation may occur between nozzles so an average needs to be taken. Dye cards can also be used to determine the number and size of droplets hitting the target area.

Maintenance: Never leave any unused larvicide in the tank. Always flush the system with clean water. As with the granular hoppers, liquid aerial applicators are usually owned by the contractor and as such maintenance is performed by the same.



Figure 27: Helicopter fitted with liquid application equipment

(Source: City of Mandurah)

CALIBRATION FORMULAE

Formula 1 : For use with granule applicators

To calculate the output of the spreader: -

1. Attach a bag to the outlet of the spreader

2. Collect the output for 1 minute.

3. Remove the bag and weigh the contents. (Do this a number of times to work out an average.)

4. Walking speed is taken at being 3km/h or .83mt/sec(this will vary depending on site conditions)

5. Determine swath of the spreader ie. 10mt for hand operated spreader and 15mt for mechanical.

<u>600[‡] x output of spreader in 1 minute</u> = output (kg/ha) swath width (metres) x speed (km/h)

^{\pm} where 600 is a constant to convert the use of mixed units (I/min, m and km/hr) to I/Ha.

Example: Hand operated spreader releasing 200gm in 1 minute.

$$= \frac{600 \text{ x} .200 \text{ kg}}{10 \text{ mt x 3 km/h}}$$

= 4 kg/ha

Formula 2 : For use with **liquid** applicators

A. To calculate sprayer output:-

Collect sprayer output for 1 minute by spraying into a measuring jug at a constant rate (Do this a number of times to work out an average.)

<u>600 x output 1 minute</u> = sprayer output (I/Ha) (A) swath width x speed

B. To calculate amount of chemical to add to tank

<u>Tank capacity x rate to be applied/Ha</u> = litres of chemical Sprayer output **(A)**

Example: 15l knapsack sprayer releasing 1000ml of liquid Bti (as wettable granule) in 1 minute

<u>600 x 1</u> = sprayer output (I/Ha) **(A)** 2 x 3km/h

 $\frac{600}{6}$ = 100 l /Ha

 $\frac{15 \times 300 \text{gm (from label)}}{100} = = \text{litres of chemical}$

= 45g of Bti wettable granule added to a 15l knapsack

ADULTICIDING

The use of insecticides to control adult mosquitoes is required when they are present in intolerable numbers and / or disease-carrying adults have been identified in close proximity of human populations. Adulticiding is a reactive approach to mosquito control and while it does have a place in an effective control program, should not be used as the primary means of control, unless in emergency situations after floods or when sites are inconspicuous. Some points to consider before undertaking adulticiding: -

- 1. Proximity to human population (some adulticides can have harmful side effects)
- 2. Non-target species present. Most adulticides will affect more than just mosquitoes.
- 3. Size of area to be treated and vegetation present.
- 4. Weather conditions at time of treatment. Light winds are ideal.
- 5. Ability to treat the area more than once so as to suppress adult numbers as a one off treatment may not be worthwhile.

Ground-based units are used for the majority of adulticiding operations in Australia. For that reason the information here will be for ground-based equipment. Most adulticiding has been done in the form of fogging however, with the introduction of Bifenthrin residual surface sprays in the last few years there is now an additional method to assist in control of adult mosquitoes. Residual sprays will be discussed further within this section.

FOGGING

Either a thermal fog or a cold fog (ULV) method is employed for distributing the insecticide. Insecticides will give better results if applied in small droplets of 10 - 100 micron. This smaller droplet size allows it to remain in the air for longer for a better chance of contact with the adult mosquito. With relation to target coverage, it is more economical to decrease droplet size than to increase the volume being applied. Anecdotal evidence suggests the effect of the adulticide will usually persist for only 2 - 3 days hence it is a short-term control agent.

Application of adulticides for mosquito control must be undertaken in strict accordance with label requirements and rates. Appropriate PPE and other safety measures are essential to ensure operator and public safety. These are discussed in the chapter 'Perspectives on Pesticide Safety' in this manual.

Basically, there are two types of foggers: thermal foggers and cold foggers.



Figure 28: Types of thermal foggers

1. Thermal fogging

Operation: Without going into great detail, a thermal fogger operates like a pulsejet or a resonant jet cycle. Once a fuel and air mixture has been ignited the cycle continues with a mixture of the insecticide diluted in water /carrier mixture being fed into the exhaust tube. Here the mixture is evaporated and becomes a fog. The carrier can be kerosene, diesel or more recently a specific carrier ie. Carmel Carrier[®] diluted with water. The density of the fog is controlled by the ratio of carrier to water mixture. The more carrier added the thicker the fog. Always refer to the chemical label and equipment manuals for correct ratios.



Figure 29: Vehicle mounted thermal fog unit in operation

(Source: Curtis Dyna-Fog website)

Practical considerations: Starting thermal foggers can be a bit tricky, so be patient. A slight breeze is helpful in moving the fog across or through the area to be treated. The Beaufort Wind Scale is a useful tool for gauging wind strength and is provided in Table 1. Check that the internal batteries have enough charge to start the fogger, otherwise start with the vehicle extension that plugs into the lighter housing. Make sure the exhaust is pointing away from people as it can backfire when first starting sometimes shooting a flame out the exhaust. A constant speed is required with either walking or vehicle usage to ensure application at the correct rate.

Calibration: To correctly calibrate a thermal fogger, a number of mixtures need to be established such as the insecticide and the carrier (if used). The density of the fog is regulated by the addition of more water to the carrier/anti evaporant. Always refer to the label for the correct mixing ratios. To collect the output of the fogger, disconnect the formulation hose and place this into a measuring jug. Measure the output for 1 minute. Using **Formulae 2A and 2B**, the correct fogger outputs and amount of chemical to add can be determined. Some of the common foggers have a swath width of 60 metres, but ask the manufacturer or supplier for specific details.

Maintenance: Ensure that clean fuel is used at all times, and that the fuel tank is emptied if the machine is to be stored for long periods. The exhaust tube requires a regular cleanout with a brush. Consult the manual for regular maintenance to keep the fogger in a good working condition. Correct gap setting on the spark plug, not leaving any chemical mixture in the tank and flushing the system are some important routine maintenance procedures.



Figure 30: Portable thermal fog unit

(Source: IGEBA website)

Operation: The Fog Generator operates on the principle of the one sided open jet-tube with a valve at the inlet side (carburetor) and an open outlet (fog outlet). This system - operating without any moving parts - with carburetor, mixer tube, combustion chamber and resonator constitutes an acoustical oscillation system at a certain frequency. This Fog Generator works at an operating frequency of about 100 cycles/second. Liquids (chemical formulations, oil, etc.) can be fed into the pulsating gas stream of the resonator at the outlet end. The high frequency of the gas allows the application and break up of solutions, otherwise susceptible to combustion or decomposition, due to the very short time they are exposed to the hot gas stream.

2. Ultra Low Volume (ULV or cold fog)

Operation: Sometimes referred as aerosol fogging, this form of fogging uses low amounts adulticide to cover large areas. Hence its name Ultra Low Volume. By using high volumes of air at low pressure, small droplets of a more uniform size are generated. If the droplet size is too small the wind can carry it away from the target area. Alternatively, if the droplet is too large it will fall to the ground before reaching the target. ULV sprayers can also dispense adulticide formulations in a more concentrated form as less diluent is used. The rate of the chemical applied is usually controlled by a remote control, which can be used from the safety of the vehicle cabin. The droplet size is varied by varying engine speed and therefore airflow.

Practical considerations: A light breeze of up to 10 km/hr when using the ULV equipment can be helpful in moving the chemical into a target area. The machine is quite noisy when operating so hearing protection is recommended. The motor will get hot so check that no hoses come in contact with the exhaust. When not in use for an extended period, either cover the unit or remove it from the vehicle to protect it from the weather. Large ULV units can be heavy so adjustments to your vehicle's suspension may be required. Always apply the chemical with the vehicles windows wound up and with the ventilation set to recycle to minimise intake and exposure to chemical drift.

Calibration: Formulae 2A and 2B can be used to calibrate ULV foggers, however it always advisable to contact the manufacturer or supplier. To capture the liquid, disconnect the hose that transfers the chemical from the main tank to the air blast head. With the motor running, capture the chemical in a container for 1 minute. The swath width on machines similar that shown in Figure 17are fixed and can be found by referring to the operators manual.



Figure 31: Vehicle mountable ULV fogger

(Source: Curtis Dyna-Fog website)

Maintenance: As with all motorised equipment, regular servicing of the motor is required. The air filter, fuel filter, changing of the engine oil are some items that will need to be checked periodically. Ensure that the filter/s that are incorporated in the chemical system are checked after each use. It only takes a couple of minutes to clean and will deter any buildup of chemical sediment.

Manufacturers' manuals should be read and servicing and maintenance procedures followed all the time.



Figure 32: Example of electric cold fogging equipment

(Source: Industrial Chemical Cleaner website)

The above equipment dispenses both oil and water-based products and can come with an optional float valve to give continuous flow capabilities without refilling the tank. It is used in areas where large space spraying is required. For example, several units can be hooked together with hoses with just one main water and/or chemical supply source.



Figure 33: Handheld fogging unit

This unit handles both water- and oil-based solutions - and has a one turn control knob to adjust particle size and degree of misting. The dispensing rate is easy to calibrate in the field to compensate for differences in liquid density or viscosity.

RESIDUAL SURFACE SPRAYS

As previously mentioned the introduction of water based Bifenthrin residual surface sprays has provided an additional option for adult mosquito and midge control. Bifenthrin has long been used for the treatment of termites and other pests, however in recent years the introduction of a number of new products has seen more interest for the use of these products for mosquitoes and midge. A number of these formulations have been trialled by various mosquito management programs throughout Australia for both mosquito and midge control. Results of these trials have shown that adult populations can be significantly reduced within areas that have been treated with these types of products. The product provides a residual layer that kills landing or resting mosquitoes and midge.

Most trials in Australia have been based around barrier applications to vegetation or external surfaces on houses, however in some countries these types of products are used internally on walls and furniture. The product can be applied with either a compression sprayer, knapsack sprayer or preferably by a powered blower/mister. The blower/mister application allows easier and faster application to the underside surfaces of leaves and branches where adults may harbour.

During application the liquid is applied to almost achieve a "run off" state. Once the product has dried it can provide several weeks of control depending on environmental conditions. Surfaces such as eaves, walls, window and door openings, outdoor entertaining areas and garden vegetation can be treated.

As with all adulticides strict accordance with label requirements and rates should be adhered to. Appropriate PPE and other safety measures are essential to ensure operator and public safety. It should be noted that these types of products pose environmental risks and are practically toxic to fish, aquatic invertebrates and bees with slight toxicity to birds.

1. Mist blower/motorised spreader (see Figure 16)

Operation: This unit comprises a 2-stroke motor coupled to a blower and tank combination that is carried on the back. Theses unit can be used for adulticide and liquid larvicide applications. The liquid product and appropriate diluent/carrier are added into the formulation tank and are blown out of the delivery tube as large (approx 200 μ m) droplets onto the target surface. The rate is regulated by the metering knob at the end of the delivery tube The motor speed is controlled via a throttle handpiece located on delivery tube that also varies the distance the chemical is dispersed.(up to 12m)

Practical Considerations: When applying products to vegetation treat those areas where mosquitoes or midge are likely to harbour in, ideally a band of vegetation extending away from the structure to be protected. If treatment to walls and eaves etc is required, it is advisable to test a small area for any discoloration prior to any large areas being treated. Wind conditions should be considered as spray drift can be an issue. When using with liquid larvicides the wind may be used to assist in the broadcast of product.

Maintenance Before use, ensure tank and lines are clean and free from contaiminants. Visually check the machine for any obvious signs of damage. Refer to the manufactures maintenance schedule for ongoing maintenance. If machine is to be stored for 3 months or more the fuel tank should be drained, carburettor run dry and stored in a dry location.

Table 14: Beaufort Wind Scale

(Source: Bureau of Meterology)

	Units in km/h	Units in knots	Description on Land	Description at Sea
CALM	0	0	Smoke rises vertically	Sea like a mirror.
LIGHT WINDS	19 km/h or less	10 knots or less	Wind felt on face; leaves rustle; ordinary vanes moved by wind.	Small wavelets, ripples formed but do not break: A glassy appearance maintained.
MODERATE WINDS	20 - 29 km/h	11-16 knots	Raises dust and loose paper; small branches are moved.	Small waves - becoming longer; fairly frequent white horses.
FRESH WINDS	30 - 39 km/h	17-21 knots	Small trees in leaf begin to sway; crested waveless form on inland water	Moderate waves, taking a more pronounced long form; many white horses are formed - a chance of some spray
STRONG WINDS	40 - 50 km/h	22-27 knots	Large branches in motion; whistling heard in telephone wires; umbrellas used with difficulty.	Large waves begin to form; the white foam crests are more extensive with probably some spray
	51 - 62 km/h	28-33 knots	Whole trees in motion; inconvenience felt when walking against wind.	Sea heaps up and white foam from breaking waves begins to be blown in streaks along direction of wind.
GALE	63 - 75 km/h	34-40 knots	Twigs break off trees; progress generally impeded.	Moderately high waves of greater length; edges of crests begin to break into spin drift; foam is blown in well marked streaks along the direction of the wind.
	76 - 87 km/h	41-47 knots	Slight structural damage occurs -roofing dislodged; larger branches break off.	High waves; dense streaks of foam; crests of waves begin to topple, tumble and roll over; spray may affect visibility.
STORM	88 - 102 km/h	48-55 knots	Seldom experienced inland; trees uprooted; considerable structural damage.	Very high waves with long overhanging crests; the resulting foam in great patches is blown in dense white streaks; the surface of the sea takes on a white appearance; the tumbling of the sea becomes heavy with visibility affected.
	103 km/h or more	56 knots plus	Very rarely experienced - widespread damage	Exceptionally high waves; small and medium sized ships occasionally lost from view behind waves; the sea is completely covered with long white patches of foam; the edges of wave crests are blown into froth.

STORAGE OF CHEMICALS

Safe storage of chemicals is required for a number of reasons.

- 1. To maintain chemicals in a safe, useable condition
- 2. To protect the surrounding environment and its inhabitants (including humans)
- 3. Duty of care and legal obligations

All chemicals should be stored according to Australian Standard (AS) 2507 - 1998, 'The storage and handling of agricultural and veterinary chemicals', and storage conditions should meet all State and Federal regulations. The following considerations may be relevant for chemicals for mosquito control, but are not exhaustive and should not be used as a definitive reference.

The storage area should be lockable, well lit and feature appropriate warning signs. The facility should be insulated to keep the temperature at a reasonable level, with a bunded concrete floor to contain and facilitate clean-up of any spills that may occur. Ventilation will allow fumes to escape but should be designed to ensure that the chemicals are not exposed to moisture. A washdown area is also required to keep the vehicle and equipment clean. Preferably a concrete area, this should also be bunded on 3 sides if possible with an approved drainage system. A personal washdown area that includes a shower and eyewash equipment should be readily available. Personal Protection Equipment is essential and should be maintained in top working condition.

Rotate the stock so as to use the older batch numbers first and do not over order your requirements for the season. Some buffer stock will be required to make allowances for delivery times, remote areas, etc. Always check the manufacture and expiry dates on all chemicals as they are delivered. Some chemicals have a short shelf life and must be used relatively quickly, for example Bti liquid.

If storing flammable chemicals, the correct fire fighting equipment must also be provided. With all dangerous chemicals it is advisable to keep a manifest of the quantities and names. Also the Material Safety Data Sheets must be up to date and copies are made available to members of the public if an inquiry is made.



Figure 34: Chemical storage with signage and ventilation

(Source: City of Manduarah)

19. MOSQUITO CONTROL - AN ENVIRONMENTAL HEALTH OFFICER'S PERSPECTIVE

SCOTT DANDRIDGE

Principal Environmental Health Officer Shire of Harvey

INTRODUCTION

The Shire of Harvey is approximately 110 kilometres south of Perth from its northern boundary and about 10 kilometres from Bunbury at its southern boundary. The Shire is approximately 1766 square kilometres in area with about 43 kilometres of coast line stretching to some 45 kilometres to the east of the Darling Scarp. At present, the population is about 22,000 of which just over half reside within 3 kilometres of the Leschenault Estuary and the environs of the Collie and Brunswick Rivers.

The Leschenault Estuary includes many tidally driven wetlands ranging from discreet areas of less than 10 m^2 to single sites of up to 20 hectares, with the total being approximately 50 hectares. These sites are extremely suitable for mosquito breeding, with brackish water rich in nutrients and providing protection from predators through an abundance of samphire vegetation.

Larval counts of up to 100,000 or greater per square metre are not unusual, resulting in tens of millions of larvae over a site.

Life style lots (zoned 'special residential' and 'special rural'), BBQ areas and a variety of recreational activities such as crabbing, prawning, fishing, camping and walking attract humans into major mosquito and RRv areas.

The picture is completed through large populations of Western Grey Kangaroos and other possible vertebrate host reservoirs being abundant in the region.

With mosquitoes, vertebrate hosts and humans, the Environmental Health Officer is expected to produce minor miracles to control potential disease outbreaks and placate the public's health risk concerns.

PROBLEMS OF A MEDIUM SIZED LOCAL AUTHORITY

RECOGNITION AND FUNDING

In dealing with seasonal mosquito control issues the Environmental Health Officer (EHO) may be confronted with a variety of obstacles when attempting to implement a successful program (Figure 35).

These can be internal or external to your local government's operations. Competing for funding and recognition for mosquito control within your local government can be difficult as these operations may not be recognised as high value or high profile. Unfortunately, sometimes it takes a "*mosquito plague of biblical proportions*" for increased funding and recognition.

Therefore it is essential for the EHO to ensure that your mosquito control program is recognised, especially during periods of low activity.



Figure 35: Issues to consider when managing a mosquito control program

OBSTACLES

- Planning Authorities, Local and State Government (continued residential development next to mosquito risk areas).
- Subdivisions. (*E.P.A No 40 Guidance Statement for Management of Mosquitoes by Land Developers* a guideline document only).
- Environmental Agencies (e.g. nutrient stripping basins vs. mosquito breeding).
- Public Health vs. Environmental concerns i.e. DEC (approval for residential development vs. control of mosquitoes, vs. environment).
- Engineers (e.g. compensating/drainage basins vs. opportunistic mosquito breeding).
 Constructed wetlands. (DOH publication "*Chironomid Midge & Mosquito Risk* Assessment Guide for Constructed Water Bodies").
- Ignorance of mosquito control methods.
- Receiving follow-up information on cases.

EXPECTATIONS

- Council's: good outcomes for residents, remembering the public generally believe good control is always possible, continued urban development, tourism, budget priorities.
- Department of Health: reduction in notifications, effective surveillance & control systems, expanded research, value for money, budget control, recognition by Planning Authorities.
- EHO's: good Public Health outcomes, effective surveillance & control, sufficient funding, access to expertise, increased professional knowledge, acknowledgement from Council & residents, recognition by Planning Authorities.
- Public: No mosquito nuisance or disease risk, response, knowledge.
- Media: information
- Politicians: kudos, budget control.
- Land Developers: no impedance to development, constructed wetlands, no section 70A notifications on titles, mosquito control handed to Council or DOH, knowledge.
OBTAINING FUNDING

The formation of a Contiguous Local Authorities Group (CLAG) allows access to funding assistance from the Department of Health:

- Criteria to form a CLAG are: actual or potential health risk in specific areas or season, definition of mosquito breeding sites and pre- and post-treatment monitoring of breeding sites.
- CLAG funding will currently allow 100% of aerial costs (helicopter hire) to be met by the State Government, and
- 50% State Government, 50% Local Authority aerial and ground larvicide cost
- Earthworks negotiated funding through MCAC (Mosquito Control Advisory Committee).

LOCAL GOVERNMENT BUDGET

It is extremely important for funding purposes to inform your Council on the success and or failure of the mosquito control program in an annual report. Details should include the following:

- A brief description of the season, including problems such as unpredictable tides and rainfall.
- Number of surveys undertaken, with brief details of the number of larvae generally observed (photographs are useful).
- Control measures undertaken, successes and failures. For example, the number of onground treatments, larviciding, adulticiding, aerial treatments, runnelling.
- Quantities of control chemicals used. Equate this to the number of hectares treated.
- Number of Ross River virus (RRV) and Barmah Forest virus (BFV) cases, consider geographical breakdowns to identify location of exposure, e.g. in a Local Government (LG) or out of LG.
- Future directions of Mosquito Control Program:
 - should include purchase of new equipment (budget);
 - education/public awareness campaigns (budget);
 - runnelling programs (budget);
 - short comings e.g. funding equipment, labour;
 - **BE AWARE of future residential** subdivisions & mosquito risk assessment.

PUT YOUR COUNCIL IN THE PICTURE!

LABOUR AND VEHICLES

Labour is always a problem in small to medium sized Local Government (LG) where one person undertakes multiple tasks, especially if outside staff and vehicles are required to assist in mosquito control. There is generally a conflict of priorities for staff and vehicles.

Consider employing a reliable part-time, seasonal, dedicated mosquito control person, who you can train. Make sure that you dedicate time to train & nurture a new officer. Talk to your neighbouring local government EHOs as there may be the opportunity to resource share. Resource sharing may include, labour, equipment, education and knowledge.

EQUIPMENT

The type of equipment required will vary depending upon your control program and site access. For example, a heavy reliance on adulticiding may be necessary where larval sites are inaccessible. It is important to have a range of equipment as this allows an appropriate control response. Whilst being a CLAG member allows the sharing of equipment it's important to maintain and increase your equipment inventory. The following describes the equipment and application methods available to the Shire of Harvey:

- Hand treatment.
- Mechanical knap sac for granular application.
- Mechanical knap sac for liquid application
- Hand pressure pump.
- Fogger ULV vehicle mounted.
- Knap sac ULV (cold)
- Thermal fogger hand held.
- Vehicle.
- Quad bike equipped with 2 liquid spray units
- Boat.
- Aerial helicopter provided by DOH.

- Insulated shed.
- Microscope
- Digital microscope camera
- Digital camera with **MACRO** ability
- Computer & screen
- 2 decimal point electronic scales (0-6000gm)
- 3 decimal point electronic scales for weighing mosquitoes (0-400gm)
- Mounting pins
- Fine point forceps
- 8 EVS CO₂ light traps spare parts

The Shire of Harvey control program primarily targets larvae by using the s-methoprene based products such as sand and briquettes, as well as Bti based products. There are some circumstances where adulticiding using ULV (cold) and thermal fogging can supplement larvae control where adult numbers pose a health risk.

Get to know your mosquito environment

Each larval breeding site may behave differently due to environmental factors such as:

- Water characteristics (tidal influences, flooding levels, salinity, depth, turbidity etc);
- Vegetation types (samphire, reed, grass etc);
- Exposure to sunlight;
- Temperature (ambient & water); and,
- State and local weather patterns.

Understanding your sites should allow you to predict when surveillance and possible control measures may be required. Subsequently, in the case of shared labour (outside staff) and vehicles this allows arrangements in advance of probable labour and vehicle requirements.

For example, from your local knowledge you may know that a particular site only floods after a 1.5m tide, while other sites may flood on a 1m tide. Consequently, this allows you to initially prioritise surveillance activities to particular sites. However, do not become complacent and ensure that all sites are checked! Understanding and identifying sites can allow you to document the risk posed and the environmental "triggers" for Council's future reference

To assist in understanding tidal and weather activity the following websites are very useful; <u>http://www.dpi.wa.gov.au/imarine/19263.asp</u> <u>http://www.bom.gov.au/oceanography/tides/MAPS/wa.shtml</u> The examination of tide charts prior to the forthcoming mosquito threat season, will allow a certain amount of pre-season prediction. Although time consuming, the "threat tides" can be highlighted on to a year planner calendar allowing a concise reference to the tidal activity in your area.

Tide graphs (Figure 36) indicate the following information which are useful tools.

- Predicted tide
- Actual tide
- Positive or negative residual: i.e. the level of water above or below the predicted level.



Figure 36: Port Authority tide chart for Bunbury (Source: Department of Planning and Infrastructure).

Further to the aforementioned web sites, there may be local websites such as your local Port Authority that may have "real time" data which is an extremely useful predictor and can be accessed from the office or from a mobile phone, whilst in the field.

Documentation

Good documentation is very important in understanding sites and will be of assistance in report writing and for historical referencing. Basic field notes should include (but not be restricted to) the following:

- Number of larvae (densities);
- Age of larvae (instars/pupae);
- Area of larval activity (hectares, m²);
- Water levels (tidal, permanent, semi permanent);
- Water level behaviour e.g. tidal activity, expected time for a site to dry out etc; and,
- Any other observations that you may think are relevant.

Remember post treatment observations and documentation is just as important as pre-treatment observations.

Digital photography is an excellent method of recording all aspects of field observation, especially with calculating larval densities and for showing the effect of s-methoprene on pupal development (Figure 37). Most photoshop type computer programs allow for the addition of text and the automatic recording of date and time.



Figure 37: S-methoprene affected pupae.

CALCULATING LARVAL DENSITIES

Larval densities can be fairly accurately determined by photographing a typical "larval dip" and counting the larvae on a hard copy photo. Approximately 100 standard dippers equates to 1square metre (m²).

For example: to calculate how many larvae there are per square metre and per hectare in a dipper containing 56 larvae (Figure 38).

Larvae per square metre (m^2) : 56 larvae per dipper x 100 = 5,600 larvae per m^2 .

Larvae per hectare (ha); 5,600 larvae per $m^2 x 10,000 =$ 56, 000, 000 larvae per ha. (10,000 $m^2 = 1$ hectare)



Figure 38: Dipper count of 56 larvae & pupae

PUBLIC EDUCATION

GET PEOPLE THINKING!

Public education is the final component of the Council's and the Leschenault CLAG's integrated mosquito control program. It was realised that the public had a role in a mosquito control program by:

- protecting themselves;
- preventing backyard breeding; and,
- being informed of the preventative roles undertaken by the Councils and the DOH.

The education component was developed after the CLAG was satisfied with the physical control of mosquitoes. There is however no reason why education cannot be developed in conjunction with the physical program but be aware that there may be some criticism that the money should be spent on "killing mosquitoes".

MEDIA

Initially the media such as local newspapers and community newsletters were utilised by writing a fortnightly article on control programs including the number of human cases etc. Where possible provide a photo.

Consider introducing a mosquito risk/awareness gauge into your Council Offices/Libraries and approach your local newspaper to publish the gauge weekly as a community service. Remember to change the gauge as required.

During seasons of high mosquito and disease activity the media and the public will demand answers as to why control is "not working". Talk back radio seems to be a particular conduit suited to criticize mosquito control programs.

The Leschenault CLAG during the 2010 -2011 season was contacted several times by the local ABC radio station to comment on local mosquito issues. It was decided to take this as a positive opportunity and a local station was requested and agreed to host a regular fortnightly live to air information segment. By undertaking this proactive approach, EHO's could control the information flow as the EHO's set the subject to be discussed. Furthermore, the public are informed and your Council will also consider this in positive manner.

Be honest in the information to be disseminated that is, don't be afraid to outlined the difficulties in achieving control or stating that there may be circumstances when control can not be achieved, e.g. extreme tidal conditions.

However, as an EHO I wanted to outline the public health role of EHOs and to explain the science in plain language so that the public grasp the complexities and the difficulties of mosquito control.

Further to the media, consider, using the Council's "messages on hold" facility, by placing a simple message about mosquito avoidance, prevention, backyard breeding etc during the risk season.

PUBLIC DISPLAYS & SIGNAGE

To further enhance education, an "in house" rudimentary photographic and text display was developed by the CLAG EHOs. The display was pinned to foam office partitions, which was time consuming, cumbersome and second rate. It was therefore decided to develop a professionally designed and produced publicity display which allowed uniform information to



be communicated in recreational centres, shopping centres, libraries and council offices, (Figure 6). The sharing of design costs made the high quality display affordable to all three CLAG LG's.

The display must be designed to be self explanatory, bold, portable and either static or staffed. If staffed, the addition of equipment (e.g. CO_2 light traps, specimens of live larvae etc) can supplement the display, however we have found that staffing the display is not very successful.

Figure 39: Mosquito warning sign

As a way of determining the effectiveness of the display, run a simple question & answer competition and of course offer a prize! This Shire offers approximately \$80 worth of personal repellents. Create a tear off form with 4-5 questions concerning the display or other mosquito related activities that your Council is undertaking along with an entry box.

This allows you to assess the approximate number of people seeing the display and the effectiveness of your messages.

The posting of awareness signs (Figure 39) in mosquito risk areas, inform the public of the risk. The text should be bold and simple but distinctive.



Figure 40: An interpretive shelter at a popular bird watching wetland

In conclusion, public education was self evolving from basic beginnings, culminating into varied and professionally produced:

- Signage (Figure 39);
- Interpretive shelters on walkways and wetlands (Figure 40);
- Pull up displays (Figure 41); and
- Television adverts, live radio interviews.

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Figure 41: The CLAG's four panelled pull-up display (2050mm H x 830mm W)

TELEVISION CAMPAIGN 2003 – 2004 (AND ONGOING)

BACKGROUND

In March 2003, the Leschenault CLAG anticipated a high activity season for 2003 – 2004. This was due to low activity for the past 3 years. It was therefore decided that the CLAG needed to not only to be vigilant with control, but to also promote the forthcoming season. A potential viewing audience of 231,300 people made television advertising an attractive proposition.

Based upon another local government TV advertising campaign promoting regional recycling, the CLAG approached the Bunbury based Golden West Network (GWN) to explore the idea of television advertising.

The advertising was planned to commence in October through to November 2003 which was considered to be the critical period. The adverts were to target personal protection and backyard/container breeding.

BUDGET

Based on the estimated costs, each CLAG members (3) Local Governments supported the initiative and provided $2,500 \times 3 = 7,500$ for the 2003 – 2004 financial year for a minimum period of 3 years.

The CLAG also made a formal application to Healthway and was successful in receiving a 50% contribution to the project cost, subject to a formal evaluation survey being undertaken. Part-way through the campaign, the Shire's of Busselton and Capel also contributed \$500 each to enable an extension to the advertising.

Total funding now available was \$13,000 which allowed the CLAG to dramatically increase the campaign from October 2003 to January 2004.

ACTUAL COSTS

Production (2 x 30 sec adverts)		\$ 1,065
Advertising (103 paid adverts* October 2003 – January 2004)		\$ 11,000
Evaluation report		\$ 884
Sundry		\$ 53
	Total Cost	<u>\$ 13,002</u>
	Budgeted Cost	<u>\$ 13,000</u>

*GWN also provided 119 free to air adverts in addition to the 103 paid adverts, at total of 222.

The CLAG worked closely with GWN and the EHO's scripted and directed the 2 x 30 sec adverts. The script needed to be in plain English and the visual message related to commonly seen land marks in the local area to complement the script. For example, when the verbal message referred to larvae and pot plants that is what was seen. Onsite filming took approximately 4 hours. Both visual and verbal messages needed to be kept simple. It is to be noted that the paid adverts were shown during prime time viewing.

The two adverts were developed with the following messages:

Message 1 - Personal Protection:

- Use repellents.
- Wear long loose clothing.
- Avoid the mosquito environment.

• Insect screening.

Message 2 - Backyard/container breeding:

- Empty bird baths etc every 5 days.
- Remove unused containers.
- Stock fish ponds.
- Mosquito proof rainwater tanks.

CAMPAIGN OUTCOMES

- The timing of the advertising coincided with the worst outbreak, with 1,570 cases of mosquito-borne disease recorded state wide and 804 cases in the South West alone. The Shire of Harvey recorded 56 cases from unadjusted data.
- The campaign was flexible, which allowed the CLAG to bring forward the commencement date, due to the DOH and the UWA mosquito surveillance system and to extend the advertising when further funding became available.
- The evaluation report was vital in assessing the value of the campaign.
- The advertising reached 74% of the population surveyed which equated to 171,000 people.
- Primary messages received were:

_	Wear long loose clothing	46%
_	Empty bird baths, pot plants etc.	36%
_	Avoid mosquito activity	35%
_	Remove or empty containers every 5 days	28%

LONG TERM OUTCOMES

Television advertising is now an integral part of the CLAG's annual health education program and included in Council's budgets. Furthermore, the Shires of Capel and Busselton both contribute financially, which allows prolonged advertising during the highest risk period and the production of further adverts.

EXAMPLES OF A SUCCESSFUL PHYSICAL MOSQUITO CONTROL PROGRAM

Keys to success:

- GET DIRTY!!
- Know your mosquito environment.
- Know your staff and equipment.
- SURVEILLANCE! SURVEILLANCE! SURVEILLANCE!
- Timing of treatment.
- SURVEILLANCE after treatment is just as important as pre-treatment.
- Initiative.
- Co-operation from neighbouring LG's.(speak to your EH colleagues)
- Modification of management plans.

CRIMP CRESCENT WETLAND – RUNNELLING PROGRAM

Crimp Crescent is a tidal wetland which can be inundated to about 6 - 8 hectares and can easily support larval numbers up to 100,000 per square metre. This site is basically flat with a grade of only about 400 mm across the site. Without further inundation this site will dry out

after about 2 weeks. Samphire is the dominant vegetation with peripheral paper bark, tea tree, reeds and salt tolerant grasses.

On-ground treatment with a mechanical knap sac could take up to 6 hours, or alternatively, about 10 minutes by helicopter. Under some tidal conditions this site can produce three cohorts in a single week. Unpredictable tidal conditions are a major problem. For example, an S-methoprene treatment based upon late instar larvae with predicted diminishing tide levels can be made useless with an unpredictable high tide which dilutes the s-methoprene levels. Post treatment surveillance is essential for early re-treatments. Due to these difficulties, other methods of control were investigated. It was found that this site was ideal for runnelling.

HOW TO ACHIEVE RUNNELLING

- Ascertain if runnelling is possible. Check with DOE if the location maybe subject to Acid Sulphate Soils.
- Undertake a 100 mm contour survey.
- Liaise and convince environmental authorities such as the DOE (Water and Rivers Commission) that runnelling is appropriate. Formal approval is essential.
- Seek approval from the Mosquito Control Advisory Committee (MCAC) (formal approval in writing is essential). Your application must include:
 - 100mm contour diagrams
 - proposed runnel locations
 - site description
 - proposed runnelling equipment
 - budget and program schedule

Purpose built equipment for the installation of runnels did not currently exist in Western Australia. Spinning equipment is ideal for cutting a 'U' shape runnel, however spinners are usually fitted to conventional tractors which have too much ground pressure and cause unacceptable damage. Tracked vehicles have ideal ground pressure characteristics, some less than the footprint of a man, but cut a square drain, not the ideal 'U' shaped drain.

INSTALLING RUNNELS

- Walk the site with the vehicle operator and strongly emphasise that minimal ground damage is necessary.
- Locate proposed runnels with survey pegs.
- SURVEILLANCE. Inspect the work several times a day (photos).

Continue surveillance after runnel completion and record appropriate observations such as:

- Larval mosquito numbers;
- Fish densities;
- Runnel performance (rectify any high points);
- Time for water to egress from the site;
- Revegetation;
- Erosion and silting; and
- Reduction in larvicide quantities.



Figure 42: Runnels at Crimp Crescent

BENEFITS FROM RUNNELLING

- Larval mosquito numbers were considerably reduced.
- Fish have increased in number and size.
- Considerable reduced use of larvicides.
- Permanency of control.
- Reduced adult numbers.
- Economic saving:
 - reduced surveillance time
 - reduced chemical cost
 - reduced helicopter hire
- Publicity it is something tangible that the public can grasp.

This project, from its infancy to completion, took about 3 years and a cost of \$5,500. The runnels were installed in March 2000 and continue to function extremely efficiently, requiring only minor maintenance with a shovel.

Runnelling systems (Figure 42) have been installed at major larval sties resulting in the dramatic reduction in the amount of larvicides applied.

BARR ISLAND: PROBLEM & SOLUTION

Barr Island is triangular in shape, approximately 500 by 300 metres, and is situated at the mouth of the Collie River and Leschenault Inlet about 1200 metres from the ocean (Figure 43).

In 1992 extremely high adult mosquito numbers were being trapped at a site at Point Duoro. Consecutive fortnightly trap counts were 1500, 1900 and 500 respectively. This was of grave concern as residences were only about 1000 metres to the east and it was mid September. Recent urban development within the City of Bunbury, now place some residences within a few hundred metres of Barr Island.

Point Duoro is controlled very successfully with about 4 - 5 kilometres of runnels. In October every metre of the runnels were walked to assess any possible problems (e.g. 4WD damage). None were found.

Question:

Where were these large numbers of mosquitoes coming from?

Rationale:

The traps are set late in the afternoon with the prevailing winds are from the west. Therefore, the breeding sites were likely to be to the west. A possible site was the Leschenault Peninsula. This was dismissed, as mosquitoes had to travel over 1.5 - 2.0 kilometres over the water. Current thought suggests that mosquitoes will not travel over open water for a reasonable distance.

The second possible site was Barr Island. No one had surveyed Barr Island, so three days after a high tide in early December 1992, I swam to Barr Island. It was revealed that a lagoon existed measuring approximately 200 metres x 50 metres, densely covered with samphire vegetation and was supporting about 20,000 larvae per square metre.

Solution:

The short term solution was to float sealed drums of Abate (Temephos) and treat the site by hand application. A long term solution was runnelling. Within seven days of being identified, a 100 mm contour survey was undertaken to assess the possible installation of runnels.

In late January 1993 an on-site meeting with officers from the Water and Rivers Commission (now the Department of Water) and DOH resulted in approval in principal to runnel Barr Island. Official approval was then granted and on the 13th October 1993 Barr Island was runnelled.

Points of interest:

- After the Barr Island site was identified and subsequently treated, trap counts at Point Duoro decreased from 100 in November to 6 in December.
- Average mosquito trap counts;
 - Sept to Nov 1992 **830** (pre runnelling)
 - Sept to Nov 1993 312 (post runnelling)
 - Sept to Nov 1994 **105**
 - Sept to Nov 2001 103
- The machine used to undertake the runnelling work, was a 1.5 tonne track machine that was barged to the island.
- The clay soil was ideal for stable runnels.
- Why was it only now that Barr Island caused a problem? The small lagoon had been formed over a particular stormy winter with sand being deposited at the northern end of the island.
- Since 1993 one more lagoon has formed and another partially formed. These are both now connected to the original runnel systems.
- Runnelling has provided a long term cost effective solution.
- Only very minor maintenance work (shovel) has been undertaken since 1993.
- The purchase of a 4m aluminium dinghy utilising CLAG funds with MCAC permission has allowed intermittent surveillance of Barr Island but also other islands in the Collie River.



Figure 43: Runnels on Barr Island

SUMMARY

In summary, for a successful program it is essential to have an "*Integrated Approach* "-with a combination of the following elements:

- Time & mosquito program recognition
- Knowledge and training
- Sourcing funds (State & Local Government)
- Surveillance, monitoring and field observations
- Larviciding (chemical control and the understanding thereof)
- Source reduction (runnelling)
- Residual surface treatments
- Adulticiding (last resort!)
- Public education
- Public warnings (DOH)

20. MARK RECAPTURE PROJECT TO DETERMINE DISPERSAL OF MOSQUITOES ORIGINATING FROM WETLANDS IN STRATHAM, SHIRE OF CAPEL, WESTERN AUSTRALIA

Colin Dent and Carla Webster

Shire of Capel

Many residential areas in south-western Western Australia (WA) are located in close proximity to major natural mosquito breeding habitat and many more are being developed due to aesthetic values of living near water. However, residents in such areas may be exposed to greater risk of contracting mosquito-borne diseases such as RRV and intense nuisance problems at certain times of year when disperse from breeding habitat into surrounding suburbs. Rapid population growth and subsequent pressure to develop new areas for residential housing is further exacerbating the problem. Census data show that the region of interest in this study (Gelorup – Dalyellup – Stratham Statistical Area 2) grew by 242% between 2001 and 2011 and the WA Department of Planning has forecast an annual average population growth rate of 4.5% in Shire of Capel until 2026, placing further pressure to develop land in what is one of the highest risk areas for Ross River virus (RRV) disease in WA.

The Shire of Capel developed a Mosquito Management Strategy in 2007 which outlines the following components involved in reducing mosquito borne disease risk; larval monitoring, adult trapping, hand and aerial larvicide treatments. Part of the Shire of Capel mosquito management program involved a mosquito mark recapture project based in Stratham. The mark recapture study aimed to develop a greater understanding of the quantity and distribution of mosquitoes within the Stratham area and the nuisance impact and potential disease risk posed to surrounding residential areas.

An overlay of 1, 3 and 6km was placed over the project site to identify where mosquitoes bred in the project site area and the area mosquitoes could potentially have an impact on. The overlay identified that breeding occurring in the project area could impact on the urban areas of Gelorup and Dalyellup. To confirm this hypothesis the mosquito mark recapture project was undertaken.

The objectives of the project were:

- To determine the dispersal distance of mosquitoes breeding in Stratham.
- To determine the concentration of mosquitoes within 1km, 3km and 6km of the wetland.
- To obtain a greater understanding of the quantity and distribution of mosquitoes within the Stratham area.

PROJECT SITE

The locality of Stratham is situated 186km from Perth on the south west coast of Western Australia between Bunbury and Busselton. There are 196ha of wetland running from Harewoods Road, Dalyellup to Rich Road, Stratham. Within these there is a section of wetland referred to as Muddy Lakes (Figure 2).

Stratham is primarily made up of rural land and 2000-5000m2 blocks. Neighbouring suburbs include Dalyellup, a high density urban development and Gelorup, similar in all respects to Stratham, special rural areas and heavily vegetated. All three areas experience high mosquito activity at varying times throughout the season. The only major recreational facility within the area is the Capel Golf Club on Bussell Hwy. There is also a small playground on Ramsay Rd which is located 150m behind the Capel Golf Club. Substantial areas of native bushland remain in both Stratham and Gelorup which support populations of Western Grey Kangaroo (*Macropus fuliginosus*), the primary natural host of RRV in south-western Australia.

Muddy Lakes is the remnants of what originally used to be Minninup Lakes until it was severed by a drainage system known as 5 Mile Brook. Now diverted, the area was drained to allow more access to land for farming purposes. Large drains were cut through the area that removed the water and diverted it out to sea. These drains are also used to remove highly acidic water created from acid sulphate soils present in the area. The drains are still present today although they are not maintained.

Acid sulphate soils occur naturally in the environment. Studies conducted by University of Western Australia identify the annual sulphuric acid export from Muddy Lakes to be 21.9 tonnes. Furthermore, studies conducted in 2009 identified that 462kg of aluminium and 17 tonnes of iron were discharged from Muddy Lakes during the wet season. Once disturbed, acid sulphate soils can cause numerous problems to aquatic life, vegetation, infrastructure and soil conditions. Very few aquatic organisms can exist in such conditions, however those rare species that can include mosquito larvae.

Mosquito activity in the area is relatively high, with large adult trapping numbers and high amounts of complaints coming from the area. The wetlands are regularly treated with chemicals to reduce mosquito numbers and although this has helped there are many factors affecting the success of these treatments. As previously mentioned, acid sulphate soils are an issue for mosquito management as they reduce the efficacy of the chemical treatments. Thick vegetation is another factor that can reduce the amount of chemical entering into the wetland and theoretically reduce the effectiveness of chemical treatments. A desktop survey was conducted to first identify the desired transect line for the initial trap and the transect lines intended to be used for the recapture. It was aimed to cover a wide area of trapping, but being mindful of limited human resources.

METHODOLOGY

Larvae were monitored at the study site so that an initial night of trapping could be conducted immediately after a new hatching to enable a large number of mosquitoes to be collected for marking and release. Additionally, recently hatched mosquitoes were more likely to survive the duration of the project and actively disperse in search of a blood meal.

The initial adult traps were placed on the western side of the wetlands along a 2600m transect line that ran north – south. The traps were not placed at any predetermined distance along the transect line. The trap sites were selected for their close proximity to the wetlands and previously noted high mosquito activity. The traps were set in the afternoon, left overnight and collected the next morning.

A total of 54,000 mosquitoes were captured for marking and release. The mosquitoes were anaesthetised by placing on dry ice for 30 seconds to 1 minute depending on the quantity of mosquitoes in the catchbag, weighed and placed in a large plastic bag where the fluorescent pink dust was lightly applied using a turkey baster (Figure 44). The mosquitoes were then

placed on a tarpaulin under a tree to recover and disperse. The mosquitoes that did not recover were weighed and removed from the estimated total of mosquitoes released.



Figure 44: Dusted mosquitoes after release

Five recapture transect lines were identified going in a north, north east, east, south east and south directions from the release site. Trapping sites were then selected along these transect lines at the closest interval to the 300, 600, 900, 1200, 1500, 3000, 4500 and 6000 metre points. Distances varied along each transect due to the limited availability of accessible locations to set the traps.

For the first night, recapture traps were placed at 300m and 600m along transect A, 1000m along transect B, 700m along transect C, 1200m along transect D and 500m along transect E. A further 17 traps were placed out along the five transects during the course of the study as shown in Figure 45. If a marked mosquito was found at the 3500m trapping point a further trap was placed at the 6000m mark. Each morning, collections were scanned using a UV light to detect any marked mosquitoes and weighed to estimate the total quantity collected. Marked mosquitoes were placed in separate specimen containers, dated, identified and recorded. Species identification was also carried out for all mosquitoes collected every second day.



Figure 45: Study location and recapture trap transects

RESULTS

An estimated total of 464,600 mosquitoes were caught during the mark recapture project. *Aedes camptorhynchus*, was the dominant species collected overall and accounted for 82% of the 83 marked mosquitoes recaptured in this study. Two thirds of the mosquitoes were recaptured within 1km of the release site and a further 20% within 3km and the majority (53%) were caught in a northerly or easterly direction from the release point (Figure 46). The furthest recapture was 9 days after release on the opposite side of the high density urban Dalyellup development from Muddy Lakes, 6470m from the release site, demonstrating that mosquito breeding in Muddy Lakes is impacting on surrounding residential areas.



Figure 46: Number of marked mosquitoes recaptured at each trap site

CONCLUSIONS

The mark recapture project determined the dispersal of mosquitoes originating from wetlands in Stratham, Shire of Capel, Western Australia. The project itself was a challenging project that stretched resources to the limit. The level of information and the greater understanding of the area surpass this, as we now know the extent of influence mosquitoes bred from this area could potentially have on residents.

A potential harbourage area has been identified that will need further investigation and if confirmed, community awareness to reduce mosquito borne disease risk should be targeted in the area, such as encouraging alfresco areas to be enclosed and personal protective measures to be undertaken. An increase mosquito trapping within Stratham would also help to monitor mosquito numbers in the area and identify if the large number of mosquitoes present was a seasonal influx or a regular occurrence.

The quantity of mosquitoes and recaptures that occurred within the 1km and 3km radius of the release and project site indicate careful consideration is required when new development proposals are within these limits of a wetland. In an ideal scenario any rezoning and subsequent subdivision of land close to major wetlands would not proceed because of the potential threat to human health. However, reality dictates that most subdivision proposals will receive approval, due to reduced land availability and the desire to live close to water bodies.

21. PLANNING, FUNDING, LEGISLATIVE AND PUBLICITY ASPECTS OF MANAGING MOSQUITOES

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This paper presents a selection of considerations relating to the management of nuisance and disease vector mosquitoes. It highlights the current policies and legislation that govern planning, health and Aboriginal heritage issues in Western Australia (WA). It discusses the funding of local government mosquito management programs, including the provision of State government funding for health-driven mosquito management. It covers the importance of public relations and information campaigns as aspects of integrated mosquito management programs.

PLANNING POLICY AND MOSQUITO MANAGEMENT

Officers and councillors of local governments with ongoing mosquito problems know the difficulty and cost of providing effective mosquito management programs for their residents. The ability to make new land available for development and the associated rates revenue must be balanced against the reality of having to provide effective and ongoing mosquito management for residents. People living adjacent to waterways know the frustration of living in a supposedly idyllic situation, but where mosquito nuisance and the threat of contracting mosquito-borne diseases such as Ross River virus are serious down-sides to their lifestyle.

Appropriate planning is essential to ensure that residents are not exposed to unacceptable levels of mosquito nuisance and to reduce the number of cases of mosquito-borne disease. Environmental officers and environmental health officers have an important role to proactively influence planning decision-makers to ensure the provision of adequate buffers between waterways and urban development in order to minimise the dispersal of mosquitoes into residential areas. The mapping of mosquito complaints over time allows a local government to determine the high risk areas within their area and to use this information to inform their recommendations on new development. For example, mapping complaints received over 10 years has allowed the City of Mandurah to show that approximately 80% of their public complaints are from people living within one kilometre of estuaries and tidal river systems.

There are several documents that define the principles of land use planning in Western Australia. The stated purpose of the Planning and Development Act 2005 is to provide for an efficient and effective land use planning system and to promote the sustainable use and development of land. The Western Australian Planning Commission (WAPC) Statement of Planning Policy No. 1: State Planning Framework provides a number of 'General Principles for Land Use Planning for mosquito management: 'The protection of environmental assets and the wise use and management of resources are essential to encourage more ecologically sustainable land use and development. Planning should contribute to a more sustainable future by adopting a risk-management approach which aims to avoid or minimise environmental degradation and hazards.'

The WAPC is able to impose a 'notification' on a land title when land being subdivided is affected by particular hazards. Since January 1999 it has been WAPC practice for notifications to be imposed on subdivision lots within 3km of wetlands in the Peel Region and the practice has been extended to other areas of WA. Some land developers are now volunteering to have

notifications on the titles of land within their subdivisions, perhaps recognising that warned residents may have less to complain about.

It is clear however, that there are a number of inconsistencies in the imposition of notifications. Until recently, localities in south-west WA were more likely to be assessed, while northern localities often were not. New lots are affected, but existing lots in older subdivisions are not. There may be implications of advising some and not all landowners of the mosquito risks and this inconsistency needs to be addressed in the future. It is not surprising that there are sometimes adverse responses from those impacted by the imposition of notifications.

Additionally, the WAPC can impose Conditions of approval on the subdivision proposal, such as the requirement for the proponent to prepare a mosquito management plan and to contribute funding towards the local government mosquito management program. Subdivision of particularly 'at-risk' locations, may be not supported, unless it can be shown that the mosquitoes can be managed e.g. by physical modification of environment. In cases where there is dispute over land use conditions and approvals, the State Administrative Tribunal is the arbiter.

Several years ago, a multi-agency committee chaired by the then Department for Planning and Infrastructure investigated planning approaches for dealing with the conflicts between development and exposure to mosquito nuisance and mosquito-borne disease. This lead to new approaches by the WAPC that require mosquito impacts to be assessed at several planning levels, thereby improving the level of consideration of the issue.

Clearly there are economically, socially and environmentally costly implications of approving urban development in areas affected by mosquito nuisance and mosquito-borne disease. These impacts include public health and mosquito management costs, as well as impacts on tourism and residential property values. There may be impacts on lifestyle due to the presence of mosquitoes and a reduced enjoyment of the outdoors. There may be adverse impacts on the wetland environment that are attributable to chemical applications and/or physical modification of mosquito breeding habitats, made necessary because of the proximity of nearby residential areas.

HEALTH LEGISLATION AND OTHER CONSIDERATIONS

In WA the *Health Act 1911* provides the power to make regulations and by-laws in relation to pest or vector management under Part IV (Sanitary Provisions), Part VII (Nuisances and Offensive Trades) and Part IX (Infectious Diseases).

Under the *Health Act 1911* and subsidiary legislation, local government has the following responsibilities in relation to mosquito management:

- Where required, preparing and implementing management programs for the control of nuisance and disease-carrying insects (e.g. mosquitoes, but also biting midge, March flies, stable flies, non-biting midges) or other arthropods;
- Ensuring assessments of private property are undertaken if required and where necessary requiring or carrying out control of insects and arthropods;
- Ensuring nuisance or disease-carrying insects breeding on 'non-private' land where such insects impact on residential areas are monitored and managed;
- Ensuring programs including community education, warnings, and avoidance strategies in regions affected by vector-borne diseases are developed and implemented;
- Ensuring the suitability and effective management of new subdivisions, proposed zoning/rezoning and other planning initiatives within a municipality, in relation to nuisance and disease-carrying arthropods;

 Where necessary, working collaboratively with the Department of Health and other State and Commonwealth agencies to manage outbreaks of indigenous vector-borne diseases and prevent incursion of exotic vectors and diseases.

A new *Public Health Bill* has been drafted by the WA Department of Health and is in the process of being considered by State Government. The intention is that it will replace the *Health Act 1911*. Rather than the reactive approach of the *Health Act 1911*, the *Public Health Bill* focuses on proactive public health prevention, making it potentially much more strategic with regard to mosquito management. It may also provide for local government to create Local Laws through the Local Government Act.

The Local Government Act 1995 provides another mechanism whereby local government can make and amend Local Laws for the 'good governance' of their district, without reference to the prescriptive *Health Act 1911*. Local Laws can cover issues such as owner-occupier responsibilities, powers of entry, and local government rights to undertake action on land that is not local government property. Most current Local Laws have used the *City of Perth Health Local Law 1993* as a model, either adopted with or without change.

In the event of a local government failing to or being unable to undertake management of nuisance or vector mosquitoes, the Executive Director, Public Health has the power to direct that the appropriate management or site clean-up is undertaken.

Local governments often experience problems relating to mosquito breeding occurring on private property. All landowners, except for the Crown (and this may change with the new *Public Health Bill*, see above), are subject to the same requirements for managing nuisance insects. In practice however, few landholders carry out mosquito management and few local governments enforce the requirement. The resulting uncontrolled breeding can have serious nuisance and public health implications for surrounding residents. Some local governments undertake the necessary mosquito management on the private land in question, but are seldom able to recoup the costs.

The WA Department of Health provides partial funding for some local government mosquito management programs through the Contiguous Local Authorities Group (CLAG) program (see section on funding below).

The new *Health (Pesticides) Regulations 2011* regulate the use of pesticides in WA. Compared to the previous Regulations, there is now a more risk-based approach, obsolete clauses have been deleted and the licensing provisions are better defined.

IMPACT ASSESSMENT OF DEVELOPMENT PROPOSALS

The *Environmental Protection Act 1986* gives the WA Environmental Protection Authority (EPA) the power to require Environmental Impact Assessments (EIA) to be undertaken for development proposals where there is either the potential to significantly impact the environment or there is public concern about the likely effects of the development. Currently, potential health impacts are assessed as part of EIA.

MOSQUITO MANAGEMENT AND ENVIRONMENTAL LEGISLATION

The Conservation Commission policy on mosquito control opposes in principle the control of mosquitoes on nature reserves, high conservation value wetlands and internationally recognised Ramsar wetlands. The preferred option is to use appropriate land use planning to reduce the risk of mosquito-borne disease or extreme mosquito nuisance. Juvenile hormone analogues/insect growth regulators are the preferred method of mosquito control, with minor earthworks being acceptable in some situations. Fogging is considered unacceptable.

The Commonwealth *Environment Protection and Biodiversity Conservation Act 1999* requires that any 'action' that is likely to have a significant impact on a 'matter of national environmental significance' (including Ramsar wetlands) must be referred to the Commonwealth Minister for the Environment for a determination. It applies to any mosquito management program commencing after 16 July 2000.

The *Conservation and Land Management Regulations 2002* require that a licence is obtained before mosquitoes are collected from conservation reserves.

ENVIRONMENT AND PLANNING

The WA EPA has two guidance statements of relevance to development of land that is impacted by mosquitoes. 'Environmental Guidance for Planning and Development' states that 'control of mosquitoes and midges in conservation category wetlands or waterways near residential areas may conflict with the management objectives for the conservation areas. Where protection of natural values is paramount, large buffers to residential areas may need to be considered.' Further, it states that 'the EPA encourages adequate setbacks to minimise the need for chemical controls and physical alterations of foreshore areas. If controls are needed, measures that ... are consistent with conservation significance ... should be adopted.'

The objective of 'Guidance Statement for Management of Mosquitoes by Land Developers' is to ensure that mosquitoes do not adversely affect the health, welfare or amenity of future residents and that the breeding of mosquitoes is controlled to the satisfaction of the Department of Health without adversely affecting other flora or fauna. It applies to development proposals that would be assessed by the EPA and includes consideration of the impacts of on-site and off-site mosquito breeding (within mosquito dispersal range; up to 5 kilometres) from the subject land. Proponents must include monitoring and management commitments for assessment by the EPA.

ABORIGINAL HERITAGE LEGISLATION

Proposals to undertake physical modification of mosquito breeding areas must include consideration of Aboriginal heritage issues. The Aboriginal Heritage Act 1972 (AHA) protects and preserves any site or object which is of significance to Aboriginal people. Under section 17 of the AHA it is an offence to disturb any Aboriginal site. The consent of the Minister for Aboriginal Affairs is required if a development is likely to impact a site.

For managers of mosquito management programs, Aboriginal heritage is an important factor for consideration when proposing any physical modification of a site. Aboriginal people extensively utilised wetlands, swamps, rivers and estuaries for a variety of purposes and as these sites are often relatively undisturbed, their cultural significance can be high. Therefore, early investigation and planning of the proposed project may save time in the long term. It is important to recognise that discovery of an Aboriginal heritage site does not necessarily preclude physical modification for mosquito management. The vast majority of 'developments' involving Aboriginal heritage issues are resolved in favour of the applicant. In the first instance it is recommended that proponents approach their Regional Office of the Department of Indigenous Affairs for advice. Assistance can be provided with organising a search of the Register of Aboriginal Sites and with contacting relevant Aboriginal individuals and communities. Further information is available at: www.daa.wa.gov.au

Native Title is a separate legislative issue and there are separate responsibilities for applicants proposing any development or site disturbance. It should be noted that public works may extinguish Native Title and therefore there is an onus on mosquito managers to make appropriate enquiries before deciding to implement any site modification.

FUNDING OF MOSQUITO MANAGEMENT PROGRAMS

The costs of mosquito management programs within a local government area are usually spread across all ratepayers in the locality whether they benefit or not. While the imposition of a 'Specified Area Rate' (SAR) is possible and has been applied to the management of impacts such as weed harvesting in canal estates, this mechanism has only recently been considered by WA local governments for financing mosquito control programs. Some of the considerations relate to the difficulty of determining the affected area, the expectation of affected residents that there will be no mosquitoes, and the time taken to administer the SAR.

Another approach is to require developers to contribute to mosquito management programs, however some local governments consider that the administration of these fees is onerous and raises expectations unrealistically. A better alternative could be to ask for developer contributions towards a specific aspect of the integrated program, such as funding an education campaign on mosquito avoidance measures.

Most commonly, local governments include an amount in the general Council rate to cover the cost of the mosquito management program. The total program cost can vary from small to enormous budgets, and there needs to be an acceptance that funding will need to be ongoing.

In WA, the Department of Health provides funding for health driven mosquito management to local governments under the terms of a Cabinet agreement made in 1990. Local governments with known mosquito breeding sites and locally acquired human cases of mosquito-borne disease are encouraged to link with neighbouring local governments to form Contiguous Local Authority Groups (CLAGs). Funding is available for 50 percent of the cost of larvicides and 100 percent of the cost of a helicopter (if large-area larvicide applications are needed), but not for adulticides or equipment. The multi-agency Mosquito Control Advisory Committee (chaired by the Department of Health) considers applications for CLAG formation and for annual funding. A review of the CLAG funding agreement is being undertaken and may lead to changes in the scope of the assistance offered to local government.

CONSULTATION AND COLLABORATION WITH OTHER STAKEHOLDERS

Mosquito managers should make it part of their jobs to consult with relevant groups in the community to keep them informed of the mosquito and mosquito-borne disease issues in the local area. Landcare, conservation and public interest groups are likely to be more supportive if they are aware of the need for appropriate planning around waterways and the various options for mosquito management. It is also important that mosquito managers consult with the Department of Agriculture and Food to ensure that mosquito management programs do not impact adversely on biological control programs for agricultural pests.

PUBLIC RELATIONS

Effective interaction with the public is essential to a good mosquito management program. The interaction involves receiving complaints, providing information on minimising backyard mosquito breeding and giving advice about mosquito avoidance measures. It is important to develop a mechanism for advising about of the timing of larviciding and fogging programs and to warn of outbreaks of mosquito-borne viruses. The development of a public profile is a key element to achieving public recognition of your mosquito management efforts and to avoiding political interference.

COMPLAINTS

Most complaints are received by telephone, although some may be made directly to field staff. It is important that all staff, including field staff, are regularly briefed on details of your organisation's management program so that they can provide up-to-date information. Complaints should be recorded and categorised. On-the-spot advice can be offered on ways of managing mosquitoes and avoiding bites. Information can be given about the details or timing of your organisation's mosquito management program. However, complaints about a neighbour's unmaintained swimming pool, for example, will require further investigation and action. Larval sampling or adult trapping may determine the location or extent of the problem and may require that chemical management measures are undertaken.

When undertaking adulticiding, particularly thermal fogging, it may be advisable to notify the Fire Services, in case the fog is reported as a fire and leads to an unnecessary call-out.

INFORMATION

Brochures

The preparation of an appropriate information brochure or series of brochures can save a lot of time in the long term. The Department of Health has a number of brochures on mosquitoborne diseases and on backyard mosquito breeding which can be used. Alternatively you may wish to seek the assistance from the Mosquito-Borne Disease Control staff when writing your own brochures.

Letter-box drops

Information about the timing of fogging programs can be provided to the affected areas via letterbox drops. This allows individuals with respiratory problems to request that the fogging machine be turned off as it comes past their residence. Letterbox drops can also be useful for delivering advice on mosquito avoidance in emergency situations (e.g. after a devastating cyclone), when normal communications lines are out of order.

Telephone 'on-hold' messages

Most State and local governments have telephone 'on-hold' messages and these can be used to remind the public about ways of avoiding exposure to mosquito bites and mosquito-borne disease.

Health warnings and newspaper columns

The Department of Health releases health warnings through the media when there are known health risks from mosquito-borne disease. These are always communicated to the local governments and Population Health Units in the affected area. Those receiving these warnings should forward them on to relevant community groups and make them available to the public.

The development of a good relationship with your local media will be time well spent and can save considerable funds when getting your organisation's message out to the public. For example, a regular column in a local newspaper can provide information on ways of avoiding

mosquito bites, methods of minimising household and backyard mosquito breeding, levels of mosquito nuisance (from adult trapping data), disease transmission risks (from numbers of disease notifications) or proposed dates and locations for larviciding or fogging activities.

Many local papers are happy to have a regular column and this method of disseminating information is free! Radio and television stations make free community service announcements and this can be a good avenue for publicising the need for avoiding mosquito bites at particular risk periods. Care should be taken to ensure that media coverage occurs at the relevant time of year only. If unfounded warnings are issued the public may become desensitised and ignore advice when disease risk is high.

Signage

Signs which warn of high risks of exposure to mosquito-borne disease can be placed strategically in camping and recreation areas. Advice on appropriate action to take to minimise exposure, as well as telephone contact numbers for further information should be included. Mosquito risk gauges, like bushfire risk gauges, can be placed on the roadside to warn the public of the current level of risk.



Figure 47: Mosquito risk gauge sign

EDUCATION

School programs

There are many examples of education programs for school children that cover facts about mosquitoes and their management. The 'Beat the Bite' campaign produced by the Mid-West Population Health Unit of the Department of Health is one example. School programs should be informative and fun in order to maximise learning effectiveness for the children involved as well as their parents. Interactive computer units may be appropriate for secondary school children.

<u>Displays</u>

Displays at shopping centres and community fairs are another way of getting information out to the public, particularly to adults. The display can be active (involving staff and equipment such as microscopes or mosquito traps) or passive (primarily with display boards). The use of the display should be timed to coincide with the main mosquito season. It should provide contact details for officers who can give further advice.

PUBLIC MEETINGS

An informed community is a much more supportive one, so don't shy away from attending or organising public meetings to provide information and to answer questions about management programs or disease risks. If you are doing your best to manage mosquitoes and there are still significant numbers of mosquitoes, then it may be helpful to explain the program to the affected residents and the reasons for any limitations in its effectiveness, rather than to inflame the community by avoiding hearing their grievances. In this way you and your program have a better chance of being understood and supported by the community, and you will be less likely to be frustrated by political intervention sought by frustrated members of the public.

22. CONSTRUCTED WETLANDS IN AUSTRALIA: CONCERNS, CONSTRAINTS, COMPROMISES & COMPLEMENTS FOR EFFECTIVE MOSQUITO MANAGEMENT

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CONSTRUCTED WETLANDS

Constructed wetlands provide a way of managing stormwater pollutants and improving quality of wastewaters. For urban and rural stormwaters, constructed wetlands provide for reductions in stormwater volume and pollutants, surface drainage and flood protection, erosion and sedimentation control, conservational habitat and visual amenities, and water reuse and recreational opportunities. For domestic, municipal, agricultural and industrial wastewaters, constructed wetlands provide removal mechanisms for suspended solids, heavy metals, organic material and contaminants (e.g. pesticides), nitrogen and phosphorous, and pathogens. These are accomplished because constructed wetlands comprise a variety of microenvironments with various physical and biological processes. Stormwater wetlands and wastewater wetlands can have different characteristics and objectives but for the purposes of this paper they will be considered together as constructed wetlands.

The impetus for installation of constructed wetlands for storm- and wastewater control and treatment (by local and state authorities and private industrial and residential developers), has brought a concern for consequent community health risks - particularly for the production of mosquitoes. However, engineering and water quality objectives often conflict with mosquito management principles, and compromises are required for mosquito population minimisation.

In many respects, the mosquito 'problems' and 'solutions' associated with constructed wetlands are site-specific. This is principally because the designs are usually objective driven and site specific, the mosquito problems are derived from regional and local characteristics of fauna and ecology, and situation, design and operational features limit the management options. However, despite the difficulties therefore inherent in generalising about constructed wetlands, there are some features in common that can be addressed, and there are some principles and practices that can be incorporated in wetland design, operation and maintenance that can minimise mosquito production.

This paper is a distillation of my experiences with constructed wetlands, and my interactions with water engineers, landscape architects and wetland botanists in the planning, construction and maintenance of wetlands for water management and treatment, in Australia over the past 10 years.

CONCERNS

There are two major types of constructed wetlands, Subsurface Flow and Surface Flow systems. Subsurface Flow systems typically do not provide a mosquito concern unless they are overextended with influent water (exceeding hydraulic capacity) or the subsurface media becomes blocked with solids, and surface water persists. Surface Flow systems are the more common type in Australia, and typically provide ideal mosquito habitat with their shallow vegetated surface waters. They may develop into complex aquatic ecosystems, with a variety of flora and fauna, but may take many years to mimic natural wetlands with their biodiversity; in the early years at least it is usually the primary colonising insects such as mosquitoes that

dominate, and mosquitoes can present pest and disease problems for nearby human communities. Throughout Australia, there are some species of mosquito that are of relatively greater concern than others, for reasons of greater pest or vector potential, although their local significance will vary with region, season and other circumstances:

- **Aedes** mosquitoes such as *Aedes alboannulatus*, *Ae. bancroftianus*, *Ae. flavifrons*, *Ae. procax*, *Ae. sagax*, *Ae. theobaldi* and *Ae. vittiger*, are typically found in temporary pools after seasonal rains but can exploit the margins of those wetlands where the water level fluctuates substantially. They can be significant pests (often as daytime biters) and some have been associated with early season arbovirus activity.
- **Anopheles** mosquitoes are potential vectors of malaria plasmodia. Anopheles annulipes s.l. is presumed to have been responsible for transmission in southern Australia in the past, *An. farauti* is an important vector for northern Australia, and *An. bancroftii* can be a substantial pest and is a potential vector in northern regions. Populations of these are enhanced by emergent vegetation and by surface algae that protects the larvae from predators and wind action.
- **Coquillettidia** and **Mansonia** species larvae have an obligate biological association with aquatic vegetation. *Coquillettidia linealis* larvae have an association with aquatic reeds, can be relatively abundant in southern Australia, and the species is known to carry viruses. In northern Australia, *Cq. xanthogaster* is a major pest and has a similar association with reeds. *Mansonia uniformis* breeds in wetlands throughout Australia, the larvae have an association with floating vegetation, and the species is an important pest and potential disease vector, particularly in northern Australia where it can be very abundant.
- **Culex** species can be important pests and arbovirus vectors, and typically they are the most important and often the dominant mosquitoes in Australian constructed wetlands. *Culex annulirostris* is a major pest and vector of viruses causing encephalitis and polyarthritis in many parts of Australia, and can breed in great abundance in natural or artificial wetlands. *Culex australicus* is closely associated with the above species in wetlands, and although it does not bite humans it has yielded viruses and may be involved in the maintenance cycles and sustain a reservoir of pathogens from which other mosquitoes may infect humans. *Culex quinquefasciatus* and *Cx. molestus* may be produced in significant numbers from treatment wetlands (and particularly the associated Gross Pollutant Traps and sedimentation ponds) when there is high organic content derived from introduced wastewater or from decaying vegetation.

The areas of Australia likely to be at greatest 'mosquito-risk' from constructed wetlands are those where natural wetlands are non-existent or few, or have been removed, and thus where generally there are insignificant populations of mosquitoes. In such areas, particularly in arid or semiarid regions, the establishment of wetlands can provide a high-risk situation with habitat for pest and vector mosquitoes, and perhaps for vertebrate reservoirs of mosquitoborne pathogens, that presents a major public health concern to local communities. Elsewhere, in regions with greater inherent mosquito habitat, constructed wetlands can still be a health concern, providing additional breeding and harbourage sites and bringing vector mosquitoes into closer association with pathogen reservoirs such as birds that are roosting or nesting, or mammals visiting for water or shelter.

CONSTRAINTS

Constructed wetlands come in a variety of forms, from relatively 'simple' on-line linear wetlands accepting residential stormwater to highly 'complex' multi-component types with online and off-line sections. Although constructed wetlands typically are constructed for the primary objectives of water containment (for various reasons), and for water quality improvement (for various parameters), there are often many secondary objectives to provide community values that impact on mosquito management; values such as:

- visual amenities
- habitats for local and migratory wildlife
- education and research
- active and passive recreation
- economic resources (crops, forests, etc.)
- energy sinks for sunlight and biomass
- cultural / heritage values

These wetlands often contain various component zones, and these can be variously classified, but are often described as follows:

- inlet zone with water control structures, and GPT and detention basin to trap larger pollutants and store surcharge stormwater, and perhaps an energy dissipater (e.g. riffle zone) to prevent erosion,
- deep-water zone and sedimentation pond to manage high stormwater flows and capture settleable solids, and with submerged plants for water quality,
- littoral zone with edge plants for bank protection water quality, habitat creation, recreation and aesthetics,
- macrophyte zone with reed beds for water quality,
- open water zones with islands for habitat and aesthetics,
- deep-water zone for water storage and further sedimentation, and with submerged plants for further quality improvement, habitat and aesthetics,
- outlet zone with water control structures to manage water levels, spillways and weirs to protect wetland during high flow and maintain ponded water during low flow.

Essentially, mosquito management in constructed wetlands must be considered to be almost site- and type-specific, and must take into account many issues related to the volume, rate and quality of the water entering the wetland, the engineering objectives which determine detention and treatment time, and the design characteristics where aesthetics and other objectives are important. Working with the engineers and the landscape architects at the planning stage is highly advisable; retrofitting the wetland for minimal mosquito production can be an almost impossible task. Notwithstanding the risks inherent in generalising about these habitats, there are a number of constraints to effective mosquito management that are associated with constructed wetlands:

The wetland must be close to or within the community - for local water detention, aquifer recharge or reuse purposes because of costs of pumping away, or to create an aesthetic amenity or wildlife habitat that will increase land and lifestyle values (although proximity increases 'mosquito risk').

The wetlands must receive relatively highly polluted waters – for their treatment and polishing before release into local river systems/estuaries (although polluted water increases production of some mosquitoes directly with increased nutrients and indirectly with increased vegetation growth and decreased predator populations).

The wetland must have a GPT – for retention of macropollutants, including floatables, and exclusion from wetland proper (although blockage and pollution buildup favours some mosquitoes).

The wetland must have large areas of dense vegetation – to provide for nutrient, nitrogen and phosphorous uptake (although dense vegetation promotes mosquito production and impedes mosquito and wetland management).

The wetland must have shallow water - to provide more favourable conditions for the emergent vegetation for the removal of nutrients from the water (although shallow water favours mosquito production).

The water must move very slowly - to allow for deposition of sediments and uptake of nutrients by biofilms on the vegetation (although slow-moving water favours mosquito production).

The wetland must have low slope batters that are grassed - to allow comfortable and safe public access to the edge of the wetland (although low-slope batters and consequent shallow margins favour mosquito production).

The wetland must not have any management plan or maintenance schedule – to reduce on-going costs and avoid public acknowledgment of potential mosquito problems (although water and vegetation must be managed to limit mosquito production as well as for the proper functioning of the wetland).

The wetlands will not produce significant numbers of mosquitoes - because currently there is no local mosquito problem (although local habitats may not be substantial and may be significantly increased by the wetland).

COMPROMISES

As already stated, assessments of risk associated with constructed wetlands and recommendations for mosquito management for wetlands are usually 'type' and 'site' specific. However, there are constructive approaches that can address the typical concerns listed above.

In general, constructed wetlands should be sited in open areas where wind action produces surface waves that disrupt larval respiration and inhibit the algae and floating plants that provide protection. Sedimentation areas intended to be 'deep', and vegetated zones intended to be 'shallow', provide different opportunities for mosquitoes. Gross pollutant traps at inflow points are often essential but potential for blockages and pollution build-up must be considered; rock flume riffle zones dissipate energy and are useful in maintaining edge integrity against erosion.

Ponds with simple shapes and a low edge to area ratio are less likely to be productive of mosquitoes. Heterogeneity of the basin and the shoreline promotes heterogeneity of the biota. Linear shorelines provide less area of refuge for larvae from predators than do convoluted shorelines. The presence of small coves and inlets provides for growth of dense vegetation, and accumulation of floating debris, that can protect mosquito larvae from wave action and predators. Channels running from deeper areas into marginal convolutions allow a connection of habitats and provide for access of predators into the refugia to reduce mosquito larval populations.

In general, surface flow wetlands use emergent plants for nutrient exchange, but other possible biofilm substrates include medium to large pebbles/stones. Shallow bays with pebble bottoms might be included in some systems to reduce the area of dense vegetation where that might be desirable. Such open water pebble-bottomed bays are less likely to support mosquitoes and they can be stocked with predatory fish to control the few mosquitoes that might colonise such a habitat.

Specifications for depths and margins of vegetated wetland bays and ponds, to prevent emergent vegetation contributing to mosquito problems, should approximate a steep (>30°)

edge and a deep (>1.3 m) bottom. Higher incidence of mosquitoes has been associated with smaller (<0.2 ha) rather than larger retention basins, but this can be site specific. With larger bays, profiling the bottom (to a slope of 0.01%) with the depth greatest at the inflow end can be advisable to prevent/concentrate pooling, particularly if there are periods of low flow, although if the flow is low enough to have the water disappear by infiltration, evaporation and/or transpiration within a week, then a lower forward slope may be appropriate. Poor drainage through inadequate grading, and poorly constructed outlet or recirculation structures, provide for mosquito problems; pools should be provided in deep sections for predators to survive periods of low flow, drawdown or drainage. Additionally, wetlands should not 'leak' water to pool nearby and create mosquito habitats separate from the wetland and inaccessible to predators.

To address the '**Constraints**' listed above:

If the wetland must be close to or within the community, then public education and mosquito management should be used to reduce threats.

- Ideally, constructed wetlands should be located away from the community, and beyond the flight range of the important local mosquito species. The flight range (dispersal) of mosquitoes varies with species, from a few hundred metres or less, to tens of kilometres or more, but there is relatively little data on the dispersal of many of the species associated with wetlands in Australia. Of the more important freshwater species, in NSW Cx. annulirostris is known to travel up to 12 km from breeding sites, Cq. linealis has been shown to travel at least 5 km, and An. annulipes has been reported to average little more than 1 km.
- Buffer zones related to the flight/dispersal range of the principal species of concern can be established, but there is no quantitative basis on which to determine the size of an effective buffer zone (such as that beyond the spread of 90% of the mosquitoes from the wetland) other than to use known dispersal data for local situations. However, these are influenced by a range of factors such as mosquito species and population size, topography and meteorological characteristics, availability of blood hosts and harbourage sites, and the tolerance level of the local human population.

If the water entering the wetland is polluted, then nutrient loadings should not exceed the carrying capacity of the wetland.

- Pretreatment of influent waters to secondary standards to reduce levels of dissolved organic matter can significantly reduce mosquito production through a simultaneous decrease of favourable conditions for mosquitoes and increase of favourable conditions for predators.
- Stormwater runoff from tarred roads or other surfaces with chemical deposits may be potentially lethal to mosquito larvae (and their predators), but the dilution involved often negates this effect. Other factors that may act to reduce larval populations (such as surface vegetation and predators) can be overridden by the attractiveness and growth support factors of polluted wastewater.
- Species inhabiting wetlands may change with increase in pollution. Culex quinquefasciatus and Cx. molestus can be expected to supplant other species in waters that become heavily polluted with organic matter, although Cx. annulirostris will continue in moderately polluted conditions. Additionally, influent water that is high in chemicals may kill vegetation and predators, and possibly the current mosquito population but the mosquitoes are likely to re-establish more quickly than the predators.

If the wetland must have a GPT, then it must be maintained and kept clean

• If it becomes blocked and retains stagnant polluted water it will be particularly attractive to mosquitoes, specifically Culex quinquefasciatus and Culex molestus.

If the wetland must have large areas of dense vegetation, then there should be a diversity of plant species and periodic maintenance to reduce density

- If subsurface zones are included in the wetland, particularly at the upstream end, this can reduce the amount of nutrient and the productivity of potential mosquito habitat water downstream, while maintaining opportunities for aesthetic features such as emergent marshland and open water wildlife habitat downstream within the overall development.
- A lower density of vegetation can be achieved by having a wetland system with a number of cells in series that provide for better mosquito management while achieving the same overall treatment effect (albeit usually through a greater length of wetland area).
- Emergent vegetation confined to deeper water areas (i.e. not at margins) is less favourable for mosquitoes, although short-circuiting of water flow has to be avoided to meet engineering objectives. There should be a maximisation of 'edge habitat' between emergent vegetation and open water. One-phase wetlands that have continuous vegetation throughout produce more mosquitoes than multi-phase wetlands that have vegetated regions separated by a region of comparatively deeper open water.
- The wetland should include areas of open (and deep) water of no less than 30% (ideally 50%) of the total area of the wetland. Emergent vegetation planted in a non-continuous pattern (e.g. islands surrounded by open water) mitigates mosquito populations; the open water allows wind and wave action to limit mosquitoes and encourages establishment of aquatic predators, and the combination of both deep and shallow areas allows a mixing of biota from both habitats, increases the diversity of invertebrates and vertebrates, including predators, and thus militates against mosquito production. Including areas of open water in the wetland typically will, however, mean a larger wetland is required to meet water treatment objectives.
- Some plants are preferable over others. Although this should be addressed locally for specific sites, a general rule is that emergent plants that form dense stands and floating plants that form tight mats should be avoided.
- The most common emergent macrophytes used in wetlands are species of the genera • *Phragmites, Schoenoplectus* and *Typha*, and these typically are associated with mosquito production - particularly when densely packed, and during periods of senescence or decomposition. Cumbungi (Typha spp.) is a native plant that is sometimes planted but otherwise often invades wetlands; it has an advantageous feature in that it very effectively slows the water flow and thus readily 'knocks-down' particulates. However, this 'blockage' feature is a disadvantage inasmuch as it promotes mosquito production within the plant community, and Typha has another disadvantage in that it raises the BOD because of all the plant material it drops into the water. As alternatives, the Bulrush Shoenoplectus validus has fewer disadvantages with BOD and blockages, and is preferred in Australia although in the USA it is reported to create blockage problems similar to Typha. The Common Reed Phragmites australis is also an invader like Typha, and can thus create problems, but is preferred over Typha because it creates fewer BOD concerns. Additionally, Typha species and Phragmites australis are also undesirable because they undergo senescence in winter in cooler regions, while other plants such as the rushes Baumea articulata, and Schoenoplectus validus and S. mucronatus, are green all year

round and thus are actively 'working', and do not create such problems by dying or dropping leaves.

- Floating vegetation that does not completely cover the water surface, may support a diverse mosquito fauna. The larval density of some species can be high in ponds with dense mats of floating vegetation (*Lemna*), but oviposition of other species can be inhibited by floating plants (*Azolla*) covering the surface. Small aquatic macrophytes such as Duckweeds (e.g. *Lemna* species) or *Azolla* have been associated with lower mosquito production than larger aquatic macrophytes such as Water Hyacinth (*Eichhornia crassipes*) and pennywort (*Hydrocotyle umbellata*). Large macrophytes such as *E. crassipes*, *Pistia stratiotes* (Water Lettuce), and *Salvinia molesta* (Salvinia) that can cover open water areas and provide habitat for *Mansonia* mosquitoes should be precluded from wetlands or strictly managed to limit their extent.
- Structural diversity of plants in the wetland is important; wetlands with a diverse array and patchy distribution of plant species will support greater animal diversity and produce fewer mosquitoes than those with continuous coverage of a single plant species. A mix of plants with different growth forms avoids a homogeneity that may favour mosquitoes, and also can avoid problems with disease and pests hitting all the component species. It is important to maintain vigour of the plant communities and thus exclude undesirable species such as those of *Typha* and *Phragmites*. 'Niches' occupied by different plants are important. *Bolboschoenus* is an early coloniser and is useful for open water invasion in new wetlands, but it is unreliable for the long term in many situations as it 'comes and goes'. The sedge *Cyperus exaltatus* is a 'clump' plant, and areas of open water persist within the veg community, which is good for mosquito management.
- When plants are intended for nutrient removal, they must be maintained in a healthy state and protected from pests and adverse environmental conditions, because dead plants will release nutrients back into the wetland and thereby increase the potential for mosquito production.

If the wetland must have shallow water, then depths of at least 30cms (and preferably 60cms) should be maintained where possible

- There should be some deeper pools (1-2 metres) and areas of open water (30% of total) to sustain populations of predators. Deeper pools or channels can be placed perpendicular to the direction of water flow, but deep water zones should not be adjacent to the margins or provide a direct connection from inlet to outlet if that allows short-circuiting of water flow and does not provide for the hydraulic or treatment objectives.
- Water levels should be kept constant (so as to deter Aedes species) but there should be a capacity for quick drawdown and draining if necessary see below).

If the water must move very slowly, then recirculating the water through the vegetation may allow an initial greater velocity

• However, while mosquitoes do not establish in fast moving water, the critical velocity probably varies with species and vegetation situation, is unknown for any species/situation, and almost certainly would be above that recommended for water treatment processes.

If the wetland must have low slope batters that are grassed, *then* a slope as close as possible to 1V:3H is advisable, and it is better not to use grasses such as *Paspalum distichum* (water couch) that 'trap' water and provide habitat for mosquitoes

- The vegetation used should allow the slope to drain completely to the wetland and the integrity of the 'edge' between the margin of the wetland and the terrestrial batter above the waterline should be maintained so as to preclude the formation of isolated pools.
- Vertical concrete steep edges for the margins are the ideal anti-mosquito measure, but often are unacceptable for aesthetic and/or public safety reasons. Lower slopes (e.g. 1V:3H) that allow for good drainage and limit shallow water habitat also can be a safety concern in many situations, and where still lower slopes (e.g. 1:8) are required then a short (200 mm) vertical hard edge lip can provide depth at the margins that will be unfavourable for mosquitoes.
- Terrestrial vegetation of a type (e.g. spiky or thorny, or otherwise impenetrable) that will prevent access to edges can be planted to protect a steeper slope/edge than might otherwise be allowed.

If the wetland does not have an adequately funded management plan, then the wetland will be colonised by mosquitoes

• This is almost inevitable, but the species and numbers of mosquitoes will vary with region, site and season, and type and condition of the wetland, and there may or may not be significant species/numbers produced.

If the wetlands produce significant numbers of mosquitoes, then investigations must be undertaken towards a risk assessment to indicate whether intervention is required

- A program of both larval and adult surveillance at/around the wetland can indicate the relative importance of the wetland compared with other local habitats. Surveillance of larvae will indicate the species and their relative abundance within the wetland but larvae of some mosquitoes, particularly *Coquillettidia* and *Mansonia* species, are not readily collected. Surveillance of adults can give an indication of mosquito production from the wetland.
- The 'risk' can be assessed with consideration of various ecological and epidemiological factors such as the various species and local habitats, their relative abundance, pest/vector status, feeding and flight behaviour, host and pathogen affinities, local pathogen endemicity, and other factors related to particular local circumstances.

COMPLEMENTS

Baseline data on mosquito populations should be acquired to guide control interventions.

 Baseline data on mosquito production in the area of the wetland should be obtained, preferably before construction of the wetland, so that the contribution of the wetland to the mosquito fauna/abundance of the area can be assessed. This is also important later if some aspect/feature of the wetland is to be significantly altered, particularly with respect to the nature of the vegetation and water quality, such as the introduction of wastewaters with high nutrient loads that may promote mosquito production and vegetation growth.

Physical interventions – water and vegetation management can help to minimise mosquito populations.

- Routine maintenance of the wetland to preserve the slope, line and depth of the margins by grading, eliminating shallows, and dredging siltation from deep pools and channels, can help mitigate mosquito populations.
- Managing water movement through the wetland, by pumping if necessary, can be helpful in reducing mosquito populations. Rock riffle zones within the system provide turbulence that is detrimental to larvae and raises oxygen levels to improve water quality. Other measures that reduce mosquito populations include mechanical aeration systems and fountains that reduce larval survival by disturbing the water surface, and sprinkler systems that reduce mosquito oviposition in the wetland.
- Lowering (drawdown) and raising the water level can be can be detrimental to *Anopheles*, *Coquillettidia* and *Culex* mosquitoes (variously stranding and drowning larvae) but may promote some *Aedes* species (conditioning and subsequently flooding eggs). Drawdown is most effective when mosquitoes are confined to the margins but can be counter productive in thickly vegetated habitats.
- Periodic draining can effectively interrupt mosquito production, and a wetland system with a number of bays in parallel allows for some cells to be drained for mosquito management while others continue to provide treatment for the influent waters. Prescribed times for drying of wetlands must be decided with a view to mosquito species, vegetation types, seasonal factors, and perhaps other considerations. Drainage regimes may need to prevent water remaining for more than five days, but such decisions are usually site specific.
- Harvesting, edging, combing, channeling or culling of plants will provide increased water movement and predator access, and reduce biomass production and consequent detritus deposition. However, regrowth can occur quickly and periodic (at least annual) plant management is usually required. Harvesting requires labour intensive activity or expensive equipment, and cut vegetation must be removed from the wetland as its decomposition will promote mosquito production; it also can be disruptive of sediments, resulting in mobilisation and export of pollutants from the wetland, and disposal of the biomass can be problematic. Burning of the vegetation can also mobilise the stored nutrients and create pollution problems. Vegetation control with herbicides on a large scale is usually contraindicated where water quality is a major objective and aquatic fauna is a concern. Notwithstanding the problems with vegetation control, the simple removal of marginal and floating vegetation, and associated debris, can have a significant impact on mosquito populations.

Biological agent interventions – predators of mosquito larvae can substantially reduce mosquito populations.

Although maintaining the wetland in a 'healthy' state will allow for the establishment of a diverse biota that will include invertebrate predators, these are generally unmanageable and the only practicable biological agents are larvivorous fish. These can be a valuable component of an integrated control program.

• Because of undesirable impacts on native fauna by introduced larvivorous fish such as the 'mosquitofish' *Gambusia holbrooki*, endemic native fish species should be given priority in mosquito control programs. However, *Gambusia* may be difficult to exclude if it is present

in local watersystems, and it can certainly contribute significantly to mosquito population reduction - providing water quality and vegetation density do not preclude its effectiveness.

Chemical agent interventions – pesticides can be used to reduce extraordinary levels of mosquito populations.

Pesticides can be quickly applied and provide rapid results at relatively low cost. However, chemicals should be used only against occasional episodes of heavy breeding and not as a long-term routine strategy, as prolonged use can lead to development of resistance in the populations and limit overall management options.

Relatively few chemicals can be recommended for use in wetlands, and their effectiveness will depend on formulation and local conditions - particularly vegetation density and water quality.

- Temephos is relatively target specific for mosquitoes and is generally suitable for use in environmentally sensitive freshwater wetlands. Weekly use may be required during summer months, and its persistence can be reduced to a few days in polluted or colloidal waters.
- Bacillus thuringiensis israelensis (B.t.i.) products are environmentally acceptable because
 of their relative specificity for mosquitoes amongst invertebrates and negligible toxicity for
 vertebrates, but they have little persistence and mosquito populations can rebound in 1-2
 weeks, even in non-polluted sites. Bacillus sphaericus has been recently developed to
 target Culex species in polluted waters where B.t.i. is ineffective; B.s. can persist and
 provide control for more than 3 weeks, but the product is not yet registered for use in
 Australia.
- Methoprene is environmentally benign because of its relative specificity for mosquitoes, and slow-release formulations can provide control over some months.

Flow-through rates, reuse and recycling of water in wetlands can decrease effectiveness of chemicals through dilution. Solid (e.g. pellets, granules and briquettes) rather than liquid formulations are usually required for thick vegetation which can be particularly difficult to penetrate (particularly when lodged), but uniform application of any chemical agent can be problematic in wetlands.

Most insecticides are ineffective in highly polluted waters such as may be found in treatment wetlands because of breakdown by bacteria and 'trapping' by colloidal matter. While less selective chemicals might be required for emergency control in such circumstances, where the situation is usually less environmentally sensitive, insecticide doses for waters with a high organic content might have to be at least double the recommended rates. However, it must be recognised that 'overuse' of larvicides can induce resistance in the target mosquitoes, and can be detrimental to the natural predators established in the systems and the functions of the wastewater treatment system, and thus work against the objective of mosquito management.

Integration of intervention options should be used to provide the optimal results.

- Complementary strategies and methodologies for interventions should be the general approach.
- Access roads for mosquito and vegetation surveillance and various interventions should be provided.
SUMMARY

Mosquito management should be an important objective in the planning, design, operation and maintenance of constructed wetlands. Pest and disease hazards caused by mosquitoes are important issues that must be addressed by those responsible for constructed wetlands. To create wetlands in areas endemic for mosquito-borne pathogens, and which might otherwise not have habitats producing large numbers of mosquitoes, could have severe public health consequences unless mosquito control strategies are incorporated in or anticipated for the wetlands.

Although mosquito management principles may appear to be incompatible with engineering and water quality objectives and operations of wetlands, practical compromises are feasible. More integrated research on the production of mosquitoes by constructed wetlands in various regions of Australia is needed, so that interaction between the various stakeholders can lead to informed and practical solutions, and appropriate mosquito management can become an integral part of constructed wetland technology.

SELECTED READING

Batzer DP and Resh VH (1992) Wetland management strategies that enhance waterfowl habitats can also control mosquitoes. J Am Mosq Control Assoc 8: 117-125.

Brown M, Beharrel M and Bowling L (1998) Chemical, Biological and Physical processes in constructed wetlands. *In*: The Constructed Wetlands Manual. NSW Department of Land and Water Conservation (Young R, White G, Brown M, Burton J, Atkins B, eds) Volume 1: 25-47.

Carlson DB and RL Knight (1987) Mosquito production and hydrological capacity of southeast Florida impoundments used for wastewater retention. J Am Mosq Control Assoc 3: 74-83.

Chanda DA and Shisler JK (1980) Mosquito control problems associated with stormwater control facilities. Proc New Jersey Mosq Control Assoc 67: 193-200.

Collins JN and Resh VH (1989) Guidelines for the Ecological Control of Mosquitoes in Non-Tidal Wetlands of the San Francisco Bay Area. California Mosquito and Vector Control Association and University of California Mosquito Research Program. Berkeley, California.

Land and Water Conservation New South Wales (1998) The Constructed Wetlands Manual. Volumes 1 & 2. Department of Land and Water Conservation New South Wales, Orbit Offset Pty Ltd, Sydney.

Dill CH (1989) Wastewater wetlands: user friendly mosquito habitats. *In*: Constructed Wetlands for Wastewater Treatment: Municipal, Industrial and Agricultural (Hammer DA ed.) Lewis Publishers, Michigan, U.S.A., pp. 664-667.

Dinges R (1978) Aquatic vegetation and water pollution control: public health implications. Am J Publ Hlth 68: 1202-1205.

Dunkerley G (1995) Using wetlands to treat wastewater: NSW EPA guidelines. *In*: Proceedings of National Conference for Water Quality Control. Townsville, September 1995: 235-244.

Ebipane R, Heidig E and Gibson DW (1993) Prevention of mosquito production at an aquaculture wastewater reclamation plant in San Diego using an innovative sprinkler system. Bull Soc Vector Ecol 18: 40-44.

Kadlec RH and Knight RL (1996) Treatment Wetlands. Lewis Publishers, CRC Press.

Kramer VL and Garcia R. (1989) An analysis of factors affecting mosquito abundance in California wild rice fields. Bull Soc Vector Ecol 14: 87-92.

Martin CV and Eldridge BF (1989) California's experience with mosquitoes in aquatic wastewater treatment systems. *In*: Constructed Wetlands for Wastewater Treatment: Municipal, Industrial and Agricultural. (Hammer DA, ed.) Lewis, Michigan, U.S.A., pp. 393-398.

Mian LS, Mulla MS and Wilson BA (1986) Studies on potential biological control agents of immature mosquitoes in sewage wastewater in southern California. J Am Mosq Control Assoc 2: 329-335.

Mitsch WJ and Gosselink JG (1993) Wetlands. Van Nostrand Reinhold, New York.

Mortenson EW (1983) Mosquito occurrence in wastewater marshes: a potential new community problem. Proc Calif Mosq Vector Control Assoc 50: 65-67.

Mulla MS, Zgomba M, Chaney JD and Darwazeh HA (1990) Spatial and temporal dynamics of mosquito larvae in a managed marsh. Mosq Contr Res, Univ Calif, Ann Rep, 1990, pp. 38-39.

O'Carroll G (1978) The mosquito abatement hazards of detention - retention facilities in New Jersey. Proc New Jersey Mosq Extermination Assoc 65: 158-165.

Orr BK and Resh VH (1990). Interactions among aquatic vegetation, predators and mosquitoes: implications for management of *Anopheles* mosquitoes in a freshwater marsh. Proc Calif Mosq Vector Control Assoc 58: 214-220.

Rajendran R and Reuben R (1991) Evaluation of the water fern *Azolla microphylla* for mosquito population management in the rice-land agro-ecosystem of south India. Med Vet Entomol 5: 299-310.

Reed SC, Cites RW and Middlebrooks EJ (1995) Natural systems for waste management and treatment. McGraw-Hill, New York.

Rupp HR (1996) Adverse assessments of *Gambusia affinis*: an alternate view for mosquito control practitioners. J Am Mosq Control Assoc 12: 155-166.

Russell RC (1994) Wetlands and mosquitoes - the pest and disease threats. *In*: Practical Aspects of Constructed Wetlands. Proceedings of a Workshop at Shortland Wetlands Centre, June 1994. NSW Department of Water Resources, Sydney, pp. 17-23.

Russell RC (1999). Artificial wetlands and mosquito control in Australia. *In*: Water Resources: Health, Environment and Development. (Kay BH ed.) Publ. E&FN Spon (Routledge), London: 141-159.

Russell RC (1999). Constructed wetlands and mosquitoes: Health hazards and management options – An Australian perspective. Ecol Eng 12: 107-124.

Russell RC, Hunter G and Sainty G (1999). Wetlands for stormwater management: water, vegetation & mosquitoes - a recipe for concern. *In*: Proceedings of the First South Pacific Conference on Comprehensive Stormwater & Aquatic Ecosystem Management. 22-26 February 1999. Auckland, New Zealand. Volume 2: 137-144.

Russell RC and Kuginis L (1995) Constructed wetlands and mosquitoes - some problems and some solutions. *In*: Proceedings of National Conference for Water Quality Control. Townsville, September 1995: 213-224.

Russell RC and Kuginis L. (1998). Mosquito risk assessment and management. *In*: The Constructed Wetlands Manual. NSW Department of Land and Water Conservation (Young R, White G, Brown M, Burton J, Atkins B, eds) Volume 1: 181-191.

Sainty G and Beharrell M (1998) Wetland plants. *In*: The Constructed Wetlands Manual. NSW Department of Land and Water Conservation (Young R, White G, Brown M, Burton J, Atkins B, eds) Volume 1: 122-138.

Schaefer CH, Miura T, Stewart RJ and Takahashi RM (1983) Studies on the relationship of mosquito breeding on rice fields and the use of sewage effluent for irrigation. Proc Calif Mosq Vector Control Assoc 50: 59-65.

Schimmenti F (1979) Mosquito control problems associated with and general guidelines for detention and retention basins. Proc New Jersey Mosq Extermination Assoc 66: 139-148.

Schmidt RF (1980) A two year study of multi-purpose water structures in Middlesex County, N.J. Proc New Jersey Mosq Control Assoc 67: 184-192.

Tennessen KJ (1993) Production and suppression of mosquitoes in constructed wetlands. *In:* Constructed Wetlands for Water Quality Improvement. (Moshiri GA ed.) Lewis Publishers (CRC Press), Michigan, U.S.A., pp. 591-601.

Tennessen KJ and Painter MK (1990) Mosquito Production in Constructed Wetlands for Treatment of Municipal Wastewater. Tennessee Valley Authority, Technical Report TVA/WR/AB-90/4.

Walton WE and Mulla MS (1989) The influence of vegetation and mosquitofish on *Culex tarsalis* abundance in duck club ponds in southern California. Proc Calif Mosq Vector Control Assoc 57: 114-121.

Walton WE and Mulla MS (1991). Integrated control of *Culex tarsalis* larvae using *Bacillus sphaericus* and *Gambusia affinis*: effects on mosquitoes and nontarget organisms in field mesocosms. Bull Soc Vector Ecol 16: 203-221.

Walton WE and Workman PD (1998) Effect of marsh design on the abundance of mosquitoes in experimental constructed wetlands in southern California. J Am Mosq Control Assoc 14: 95-107.

Walton WE, Mulla MS, Wargo MJ and Durso SL (1990). Efficacy of a microbial insecticide and larvivorous fish against *Culex tarsalis* in duck club ponds in southern California. Proc Calif Mosq Vector Control Assoc 58: 148-156.

Walton WE, Schrieber ET and Mulla MS (1990) Distribution of *Culex tarsalis* larvae in a freshwater marsh in Orange County, California. J Am Mosq Control Assoc 6: 539-543.

Walton WE, Workman PD, Randall LA, Jiannino JA and Offill YA (1998). Effectiveness of control measures against mosquitoes at a constructed wetland in southern California. J Vector Ecol 23: 149-160.

Whelan PI (1988) Mosquito breeding and sewage treatment in the Northern Territory. Water 15: 34-37.

White G (1995) The design of constructed wetlands in New South Wales. *In*: Proceedings of National Conference for Water Quality Control. Townsville, September 1995: 425-434.

White G (1998) A systems approach to constructed wetlands. *In*: The Constructed Wetlands Manual. NSW Department of Land and Water Conservation (Young R, White G, Brown M, Burton J, Atkins B, eds) Volume 1: 18-24.

White G (1998) Planning considerations. *In*: The Constructed Wetlands Manual. NSW Department of Land and Water Conservation (Young R, White G, Brown M, Burton J, Atkins B, eds) Volume 1: 51-61.

WHO (1982) Manual on Environmental Management for Mosquito Control. WHO Offset Publication No. 66. World Health Organization, Geneva.

WHO (1988) UNEP/WHO Interregional Travelling Seminar on Environmental Management Measures for Vector Control in Water Resource Development Projects. WHO/VBC/88.956, World Health Organisation, Geneva.

Wong THF, Breen PF and Somes NLG (1999) Ponds vs wetlands – performance considerations in stormwater quality management. *In*: Proceedings of the First South Pacific Conference on Comprehensive Stormwater & Aquatic Ecosystem Management. 22-26 February 1999. Auckland, New Zealand. Volume 2: 223-231.

23. ROSS RIVER AND BARMAH FOREST VIRUSES

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INTRODUCTION

Ross River virus (RRV) and Barmah Forest virus (BFV) are the two most common causes of mosquito-borne disease in Australia. The illnesses caused by infection with these viruses are not fatal, but can be very debilitating and may persist for weeks or months. Consequently, both viruses have a considerable impact on public health in the many regions of Australia where they can be active.

This chapter focuses on the ecology and epidemiology of these two important Australian mosquito-borne viruses, with an emphasis on Western Australia. In particular, the chapter includes information on:

- When and where people are likely to be infected with the viruses;
- Human disease caused by infection with the viruses;
- Mosquito vectors;
- Animal hosts;
- Environmental conditions that lead to outbreaks;
- Virus survival between outbreaks; and
- Surveillance (monitoring) and control programs for RRV and BFV.

BACKGROUND

RRV and BFV are mosquito-borne viruses, also known as *arbo*viruses (= *ar*thropod-*bo*rne viruses).

Both are classified into a 'group' (genus) of viruses known as *alphaviruses* but they are quite distinct from one another (i.e. BFV virus is not just a different 'strain' of RRV).

Arboviruses circulate (in the environment) between blood sucking insects (e.g. mosquitoes) and vertebrate animals. The insect is known as a **vector**, the animal as a **host**. All arboviruses replicate (multiply) in both the vector and the host.

Humans can only be infected from the bite of an infected mosquito (you cannot become infected directly from contact with another human or animal).

ROSS RIVER VIRUS

RRV disease was first described during outbreaks in NSW in 1928. The virus itself was first isolated from *Aedes vigilax* mosquitoes collected in 1959 at the Ross River, near Townsville, Queensland. The first human isolate was obtained in 1979-80 during a large outbreak on islands in the South-Pacific.

RRV is the most common cause of arboviral disease of humans in Australia: 1000s of cases are reported on average each year (Table 15).

Year	WA	ACT	NSW	NT	QLD	SA	TAS	VIC	Aust
1993	154	4	598	264	2251	773	10	2414	6468
1994	94	1	331	312	2998	28	24	115	3903
1995	298	2	236	369	1642	21	28	65	2661
1996	1437	1	1031	137	4880	56	76	286	7904
1997	712	9	1600	217	2361	635	12	1047	6593
1998	286	6	576	127	1946	67	9	135	3152
1999	621	8	952	157	2304	40	67	223	4372
2000	1068	16	747	145	1481	418	8	319	4202
2001	195	9	717	225	1567	142	13	352	3220
2002	125	0	182	63	885	42	117	37	1451
2003	660	1	492	120	2513	33	4	15	3838
2004	1093	6	698	233	2004	53	20	90	4197
2005	295	4	578	209	1180	155	5	98	2524
2006	842	10	1221	279	2610	317	14	211	5504
2007	546	13	842	299	2135	211	8	96	4150
2008	828	21	1154	263	2843	197	77	231	5614
2009	803	4	909	431	2145	328	29	92	4741
2010	390	22	1088	336	2379	451	39	421	5126
2011	850	8	579	184	1219	978	7	1312	5137
2012	1376	11	602	227	1947	219	19	287	4688
2013*	859	1	263	115	974	74	4	77	2367
Totals	13532	157	15396	4712	44264	5238	590	7923	91812
Annual average*	634	8	757	230	2165	258	29	392	4472

Table 15: Notifications of RRV infection to State and Territory health authorities, 1993 to June 2013

Annual average* 634 8 757 230 2165 258 29 392 4472 Data courtesy of the National Notifiable Diseases Surveillance System, Australian Government Department of Health and Ageing (these totals may differ from WA Dep't of Health figures due to differences in reporting methods and criteria).

* 2013 totals are to 18 June 2013 and 2013 figures are not included in calculation of annual average

GEOGRAPHICAL RANGE

RRV occurs in all states of Australia, including Tasmania. RRV has also been reported from PNG, Irian Jaya and islands in the Western Pacific.

In Australia:

Outbreaks of RRV disease may occur in the tropical north after heavy wet season rains or very high tides. Smaller numbers of cases can also occur in tropical regions at other times of the year.

Less frequent but acute outbreaks occur in arid and inland areas after heavy rains/flooding. Outbreaks occur in temperate (southern) regions when rains/tides enable mosquito vector populations to persist into the warmer months (late spring, summer or early autumn). Diverse climatic conditions, mosquito species and animal host species in different parts of Australia mean that RRV has several different 'ecologies'.

RRV IN WA

RRV disease was first described in 1956, in the south-west. The virus was first isolated from *Culex annulirostris* mosquitoes trapped at Derby in 1977. The first south-west isolate was from the southern saltmarsh mosquito, *Ae. camptorhynchus*, in 1987.

RRV was designated a notifiable disease (to be reported by doctors) in 1985. Over 15,000 laboratory-confirmed cases have been reported from WA from January 1984 to June 2013



(Figure 48). This is still an under-representation of true incidence of disease as many cases are not diagnosed or reported.

Figure 48: Monthly incidence of RRV disease in WA, 1984-June 2013. Data from 1 Jan 2000 onwards include laboratory reports received as well as doctor notifications. Numbers above graph bars are total cases in WA by financial year (July to June).

The direct cost of RRV disease to individuals in WA has been estimated at \$1,755.00 per case (D. Hendrie, J Ricciardone and M. Lindsay, unpublished results). Thus, the direct cost to the WA community since 1984 is likely to have been around \$27.6 million (AUD). Indirect economic impacts on tourism, real estate, vector control programs and quality of life have not been accurately determined but are likely to be many times this figure.

Statistics on the incidence of RRV disease from different sources may differ depending on whether lab reports, case follow-up questionnaires and other available records were used in the compilation of the data.

CLINICAL FEATURES OF RRV DISEASE (EPIDEMIC POLYARTHRITIS)

RRV causes a non-fatal but potentially debilitating polyarthritic disease in humans. The disease was originally referred to as epidemic polyarthritis but is now called RRV disease to avoid confusion with epidemic polyarthritis caused by infection with BFV.

Most patients are aged between 20 and 50 years, but infants, children and older people can also be affected. The ratio of males to females affected may vary slightly between outbreaks but on average is close to 1 : 1. The incubation period of RRV (time from infected mosquito bite to onset of symptoms) is 3-21 days (usually 7-11 days).

CLINICAL MANIFESTATIONS

1. Constitutional symptoms/effects

Fatigue Muscle pains (myalgia) Headache Flu-like illness Enlarged glands (lymphadenopathy) Sore throat Eye irritation Lethargy Fever Neck stiffness Depression Loss of libido Tingling or numbness of extremities

2. Rash

The rash can appear 11 days before to 15 days after onset of joint symptoms, mainly on limbs and trunk but also on hands, feet and face. It is normally not itchy.

3. Rheumatic (joint) manifestations

The onset of joint manifestations is often sudden and symmetrical. 'Peripheral' or small joints (fingers, wrists, toes, ankles, knees, elbows etc.) are frequently affected but other joints can also be affected. Joint symptoms range from pain only to acute swelling, redness and severe pain. Tenderness in tendons may also occur.

4. Course of the illness

There is considerable variation in the course of the illness between patients. Fever and rash usually subside within two weeks. Joint manifestations and 'emotional' effects may last much longer.

By 3 months, symptoms are no longer continuous: people have good days interspersed with bad days. Good days occur more frequently as time progresses.

Emotional stress, fatigue or alcohol may aggravate symptoms.

There is currently debate about the duration of symptoms of RRV disease. Retrospective analyses of patients infected during outbreaks in the late 1980s to mid 1990's suggest that many patients suffered symptoms for several months or even years. Recent smaller, prospective studies in eastern Australia suggest that symptoms of RRV may not persist for as long as previously thought, or that other, unrelated conditions may be responsible for some long-term symptoms. Variation in virulence between the causative strains of different outbreaks of RRV may also be responsible for differences in severity and duration of symptoms. However, carefully designed studies in different regions of Australia are required to confirm these theories.

5. Diagnosis

Some symptoms of RRV disease are similar to those caused by other (mosquito-borne and non-mosquito-borne) viral infections. Therefore, a blood test requested by a doctor is the only way of confirming infection with RRV.

Ideally, two tests, 10-14 days apart ('paired sera'), are needed to confirm recent infection. A single test for IgM antibody to RRV is indicative of recent infection, but IgM antibody can persist for several months, so recent clinical and travel histories and recall of exposure to mosquitoes are also important.

6. Treatment

There are no vaccines against or specific treatments (cures) for RRV.

Patients should be informed that symptoms may persist for weeks to months, but that most patients recover completely within a few months.

Rest is very important, especially in the early phase of the illness, and at the time of flare ups. Gentle physical therapies such as swimming and hydrotherapy can reduce symptoms in some patients.

Many patients will require simple analgesics (aspirin, paracetamol) to alleviate inflammation and pain during the course of their illness. Some may also benefit from non-steroidal antiinflammatory agents.

Depression is common in prolonged cases of RRV disease and may require active management. Emotional support and understanding are important if symptoms become chronic.

Information about some other potentially helpful treatments can be obtained from the Arthritis Foundation of Western Australia.

MOSQUITO VECTORS OF RRV

RRV has been isolated from over 30 mosquito species in Australia and several of these have been shown to be competent vectors of the virus in laboratory studies. Thus, RRV appears to have adapted well to markedly different environments around Australia, with different mosquito species acting as vectors in different regions.

The principal rural pest species are the major 'epidemic' vectors. These include temporary ground pool-breeding *Aedes* species such as *Ae. vigilax* and *Ae. camptorhynchus* in coastal regions and *Ae. normanensis*, *Ae. sagax* and others in inland regions. Plague numbers of these *Aedes* species may develop between one and four weeks after heavy rains or high tides. Soon after this is when the risk of transmission of RRV to humans is highest.

Some semi-permanent and permanent fresh water-breeding species are also important, especially in regions with more reliable rain such as the tropical north and temperate south of Australia. The most important of these is *Culex annulirostris*, but other species such as *Coquillettidia linealis* (eastern Australia) and *Cq.* species near *linealis* (western Australia) may play a role also.

There is now good evidence that *Ae. notoscriptus*, a peri-domestic container-breeding species, is an important vector of RRV, particularly for transmission of the virus in urban areas. *Ae. notoscriptus* has been shown to be a competent vector in the laboratory and RRV has been isolated from field caught specimens prior to and during RRV outbreaks.

The diverse range of vectors of RRV in different environments means that predicting vectors in one region based on results from another may be misleading. Instead, surveys that link local species with outbreaks of RRV disease, and interpreted with information on their vector competence, will define the likely vectors in a region more accurately.

VECTORS OF RRV IN WA

Over 90 species of mosquito occur in WA but only some are vectors of RRV. Different vectors are involved in different regions or at different times of the year in the same region, contributing to the different ecologies of the virus.

Table 16 lists the mosquito species from which RRV has been isolated in different regions of WA. These results, combined with studies of mosquito fauna during outbreaks of RRV and laboratory (vector competence) studies have implicated certain species as major vectors and others as likely vectors. Vector competence studies are required to confirm involvement of many of the other species listed.

Table 16: Mosquito species that have yielded isolates of RRV in WA (to 2013)

Region	Species*
Kimberley	Aedes (Mucidus) alternans (Westwood)
	Aedes (Neomelaniconion) lineatopennis (Ludlow)
	Aedes (Ochlerotatus) normanensis (Taylor)
	Aedes (Ochlerotatus) pseudonormanensis (Marks)
	Aedes (Ochlerotatus) vigilax (Skuse)
	Anopheles (Anopheles) bancroftii Giles
	Anopheles (Cellia) annulipes s.l. Walker
	Anopheles species (species undetermined)
	Culex (Culex) annulirostris Skuse
	<i>Culex (Culex) palpalis</i> (Taylor)
	Culex (Culex) quinquefasciatus Say
	Culex (Culex) sitiens Wiedemann
Pilbara	Aedes (Levua) daliensis (Taylor)
	Aedes (Macleaya) tremulus (Theobald) male (vertical transmission?)
	Aedes (Ochlerotatus) eidsvoldensis (Mackerras)
	Aedes (Pseudoskusea) bancroftianus (Edwards)
	Aedes alternans
	Aedes E.N. Marks' species No. 85
	Aedes normanensis
	Aedes pseudonormanensis
	Aedes vigilax
	Aedes vigilax male (vertical transmission?)
	Anopheles (Cellia) amictus Edwards
	Culex (Culex) australicus Dobrotworsky and Drummond
	Culex annulirostris
	Culex quinquefasciatus
	Culex sitiens
Gascoyne	Aedes bancroftianus
	Aedes normanensis
	Culex annulirostris
Murchison	Aedes (Ochlerotatus) sagax (Skuse)
Mid-west	Aedes bancroftianus
	Culex quinquefasciatus
South-west and	Aedes (Finlaya) alboannulatus (Macquart)
southcoastal	Aedes (Finlaya) notoscriptus (Skuse)
	Aedes (Ochlerotatus) camptorhynchus (Thomson)
	Aedes (Ochlerotatus) clelandi (Taylor)
	Aedes (Ochlerotatus) hesperonotius (Marks)
	Aedes (Ochlerotatus) ratcliffei (Marks)
	Aedes camptorhynchus male (vertical transmission?)
	Aedes vigilax

(table continued on next page)

Region	Species*
South-west and	Anopheles annulipes s.l.
Southcoastal (cont)	Coquillettidia (Coquillettidia) sp. near linealis Marks and Harris
	Culex (Culex) globocoxitus Dobrotworsky
	Culex annulirostris
	Culex australicus
	Culiseta (Culicella) atra Lee
	Tripteroides species (species undetermined)
Perth	Aedes notoscriptus ⁺
	Coquillettidia sp. near linealis

Table 3 (cont): Mosquito species that have	yielded isolates of RRV in WA (to 2008)
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*Known or highly suspected vector species on the basis of frequency and timing of RRV isolations are shown in bold.

[†]*Ae. notoscriptus* is shown for Perth metropolitan area on the basis of circumstantial evidence only. Refer to Table 4 for other species from which RRV has not yet been isolated in some regions but which are suspected vectors on the basis of vector competence studies, demonstrated vector status elsewhere and/or abundance during outbreaks.

VERTEBRATE HOSTS OF RRV

Vertebrate hosts are defined as animals that, when bitten by a mosquito that is infected with RRV, become infected themselves and develop enough virus in their blood (viraemia) to pass on the virus to other mosquitoes that feed on them. Most hosts are thought to remain infectious (for mosquitoes) for only a few days or weeks after becoming infected.

The main suspects for RRV are native marsupials, particularly macropods (kangaroos, euros, wallabies) and also brush-tailed possums in urban areas. There is evidence that some domestic mammals such as horses may act as hosts. Flying foxes have also been implicated in Queensland.

Birds are not thought to play a major role, unlike for the flaviviruses, Murray Valley encephalitis virus and Kunjin.

Humans may act as hosts during major outbreaks; e.g. South Pacific outbreaks in 1979-80; outbreaks in the south-west of WA in 1991/92, 1995/96, 2003/04 and 2011/12 (circumstantial evidence). Regular and rapid movement of infected (viraemic) people from one region to another may assist in seeding and spread of outbreaks. However, by the time a person develops symptoms of RRV disease they are almost certainly no longer infectious for mosquitoes. Consequently, there would be no benefit whatsoever in quarantining patients diagnosed with RRV disease.

The size of the animal host population and the proportion of that population with immunity to RRV probably play a key role in determining whether an outbreak can occur. High levels of immunity in a host population immediately following a major outbreak may reduce the potential for another outbreak in the short-term. As new non-immunes are 'recruited' to the virus transmission cycle, either through births (new generations) or migration, the potential for outbreaks gradually increases. This theory is supported by evidence that major outbreaks of RRV disease are generally not reported from the same regions of WA in consecutive years. Furthermore, the thee largest outbreaks (in 1995/96, 2003/04 and 2011/12, see Figure 48 on page 214) followed several years of well below average RRV activity.

ENVIRONMENTAL CONDITIONS

RRV activity is strongly influenced and limited by environmental conditions.

The environment affects mosquito vectors (breeding, longevity, feeding behaviour etc), vertebrate hosts (breeding, survival, migration etc), and rate of virus replication in the vector.

Environmental conditions also determine whether humans are likely to be exposed: e.g., outdoor activities, less-protective clothing in south-west summer.

Some important environmental conditions to consider in determining the risk of RRV activity in WA include *rainfall, temperature* and *tides* (in coastal regions).

Rainfall

A review of environmental precursors of outbreaks of RRV around Australia between 1896 and 1998 found that overall, rainfall seemed to be the single most important risk factor.

Studies in WA show that the biggest threat of RRV activity follows heavy rains that:

- create areas of temporary freshwater (river floodplains, creeks, floodways, seasonal wetlands, claypans, etc);
- create areas of temporary brackish water (if run-off flows onto brackish wetlands, salt marshes or low-lying salt affected areas);
- top up existing water bodies.

An example of the relationship between unusually heavy rains and high levels of RRV activity is shown in Figure 2 (Broome, western Kimberley).



Figure 49: RRV cases and monthly rainfall at Broome (western Kimberley), June 2010 – June 2013

Abundance of food and water created by heavy rains, possibly in the season prior to a major outbreak, may also enable opportunistic animal hosts of RRV (kangaroos and other marsupials) to breed more successfully. This results in larger numbers of non-immune recruits for the virus transmission cycle, thereby increasing the potential for high levels of virus transmission if mosquito vectors are also present.

Finally, and in contrast, evidence from WA's three largest RRV outbreaks (1995/96, 2003/04 and 2011/12) suggests that drought may have been an environmental precursor to these major epidemics. Breeding of mosquito vectors appears to have been suppressed during three consecutive years of very much below average rains prior to the outbreak. As a result, very little RRV activity occurred. This meant that the proportion of non-immune individuals in vertebrate host populations increased markedly. Once rainfall (and vector mosquito breeding) returned to normal, it appears the scene was set for a dramatic escalation of RRV activity.

Temperature

Low temperature limits virus replication in mosquito vectors, and may explain the low number of cases of RRV disease during most south-west winters (Figure 50, Figure 54).

High temperatures may evaporate water in breeding sites before aquatic stages of the mosquito life cycle are completed. High temperatures may also kill off adult mosquitoes before they have a chance to become infected, incubate the virus and pass it on in a subsequent bloodmeal.



Figure 50: Proportion of RRV cases by month of onset in the south-west of Western Australia, July 2002 to June 2013

The relationship between moderate daily temperatures and high levels of RRV activity in an arid region of WA is shown in Figure 51 (Exmouth, Fortescue region).



Figure 51: Cases of RRV disease and monthly average maximum and minimum temperatures, Exmouth (south-west Pilbara), 1989 – 1993.

Breeding of several vectors of RRV is temperature-limited. In south-west WA, *Ae. vigilax, Cx. annulirostris* and *Cq.* species near *linealis* only breed in the warmest months of the year (October to March). *Ae. camptorhynchus*, a major south-west vector, normally only breeds in cooler months of the year (March to November) but persists into summer in some epidemic years.

Temperature also affects people's behaviour, which may increase or decrease their likelihood of exposure to mosquitoes. For example, people are less likely to be outdoors in extremes of temperature. Previous outbreaks of RRV have coincided with peak holiday seasons in the south-west (summer) or Pilbara (autumn-winter). This clearly adds to the potential for exposure of humans to mosquitoes infected with the virus.

Tides

Two important vector species, *Ae. vigilax* and *Ae. camptorhynchus* breed in pools left on saltmarshes or brackish wetlands after high tides. Therefore, tide height and frequency is important in RRV ecology.

Epidemics in the south-west in 1988/1989 and 1999/2000 were largely attributable to a rise in the mean sea level off the south-west. The high water level resulted in very frequent inundation of estuarine salt marshes in the Peel and Leschenault regions during late spring and summer. Consequently, *Ae. camptorhynchus* and *Ae. vigilax* persisted in high numbers throughout summer. In 'normal' years the salt marshes are dry throughout most of the summer.

The effect of construction of the Dawesville Channel on breeding of salt marsh mosquitoes in the Peel region illustrates the importance of tide height and frequency. Winter populations of *Ae. camptorhynchus* in the region have actually decreased since the opening of the channel as constant submersion of the marshes in winter flushes out larvae and provides access for predators. Unfortunately, tidal inundation of the marshes in late spring and summer (RRV season) is now much more regular and has resulted in larger populations of *Ae. camptorhynchus* and *Ae. vigilax* at this 'high-risk' time of the year.

HUMAN INFLUENCES ON RRV ACTIVITY

Human activities that alter aspects of the environment that affect mosquito vectors, vertebrate hosts, or interaction of humans with mosquito vectors can (and do) lead to increases in the incidence of RRV disease. These may include irrigation, dams, other forms of water impoundment, location of residential areas in relation to known mosquito breeding sites, planting of vegetation that may act as harbourage for mosquitoes and availability of pasture/crops that enable artificially high levels of breeding of vertebrate hosts.

Some known or 'suspected' examples:

- 1. Construction of the Dawesville Channel has led to larger summer populations of mosquitoes (see above) and more frequent RRV activity in the Peel region.
- 2. Studies in the Peel region have found that the attack rate of RRV disease was significantly higher in suburbs within 2 km of saltmarshes and seasonal wetlands compared to the region as a whole (e.g. Figure 52). The risk of mosquito-borne disease must therefore be considered as part of urban planning for new residential developments. Planning clearly has the potential to influence how many people may be at excessive risk of exposure to RRV during major outbreaks.



Figure 52: Rate of Ross River virus cases per 1000 dwellings versus distance from mosquito breeding habitat in the Peel region, July 2002 to June 2012

- 3. Very high case attack rates are observed in some south-west residential subdivisions where animal hosts (such as western grey kangaroos) are abundant. Further studies are required to substantiate this apparent relationship.
- 4. Cases of RRV disease with onset dates well before the first wet season rains have been reported from Kununurra (north-east Kimberley). This activity was recently linked to dry-season breeding of the RRV vector *Cx. annulirostris* in the Ord Irrigation Area. As a result, methods for minimising this man-made impact on public health have been provided to the Shire and the agency responsible for management of the irrigation infrastructure.
- 5. Dryland salinity resulting from the broad scale clearing of native vegetation for agriculture in the Wheatbelt region has been strongly associated with increased breeding of the RRV vector *Ae. camptorhynchus* and potential for disease transmission.

SURVIVAL OF RRV BETWEEN OUTBREAKS

In arid areas, RRV may survive in desiccation-resistant eggs of some *Aedes* species. The eggs are thought to acquire the virus from their infected female parent. When the next rains (or tides) enable the eggs to hatch and develop, a small percentage of the next generation may *already* be infected. This is known as vertical transmission of the virus, and may account for the occurrence of cases soon after rain in arid regions. Vertical transmission probably explains the isolations of RRV from male mosquitoes in WA (Table 16), as male mosquitoes do not feed on blood.

RRV is probably maintained in low-level 'maintenance' cycles between mosquitoes and natural hosts in temperate and tropical areas where mosquito populations can survive throughout the year.

The virus may also be introduced or re-introduced from one region to another (e.g. some metropolitan suburbs during the 1988/89, 1991/92, 1995/96, 2003/04 and 2011/12 south-west outbreaks), presumably in infected humans or livestock.

TIMING AND LOCATION OF VIRUS ACTIVITY

Cases of RRV disease have been reported from most regions of Australia. However, the incidence varies dramatically from one year to the next (Table 1; Figures 1 and 6). The geographical range and seasonality of RRV at a national level was summarised on page 2 of this chapter. Following is a more detailed description of the timing, location and incidence of RRV disease in different regions of WA.

The prevalence of RRV disease depends on mosquito and virus activity and the number of people living in affected areas. 70% of cases occur in south-west (86.5% of population); 30% of cases in north and east WA (only 13.5% of population). This means that the case attack rate (number of cases per head of population) is often highest in northern and eastern WA (e.g. Figure 53).

Other factors may also contribute to the incidence of RRV disease. For example, awareness of the disease and reporting by medical practitioners, or the fact that only severe cases may visit the doctor could influence the perceived incidence. RRV activity coinciding with the summer holiday season almost certainly increase the actual incidence of RRV in the southwest of WA.

The cumulative number of cases reported in each month of the year between July 1998 and June 2013 for most major regions of WA are shown in Figure 54.

Kimberley

RRV is probably endemic (some cases occur every year). Most cases occur during or just after the wet season (Dec-May) (Figure 54A). Incidence is linked to the amount and pattern of wet season rain; east and north Kimberley cases are generally earlier than in the West Kimberley.

Some cases occur during the 'dry' possibly due to tidal mosquito breeding in coastal regions and in the irrigation area around Kununurra.



Figure 53: Rates of RRV disease per 100,000 residents in different geographical regions of WA during two major outbreaks in 2003/04 and 2011/12.

Pilbara

There is considerable variation from year to year: activity is much more 'epidemic' in nature. Incidence is related to the amount and pattern of rainfall. Mosquito breeding after high tides may also be responsible for some cases in coastal areas.

Most cases occur between January and May (Figure 54A). In the De Grey (north-east Pilbara) most cases are in January-May. In the Fortescue (south-west) most cases are in March-June. Cases are often reported with a date of onset between two and three weeks after first heavy rains (e.g. Figure 49).

Gascoyne, South-east (Goldfields), North east (Interior) RRV activity in these arid regions is irregular and highly opportunistic.

In most years little or no activity is reported, but acute outbreaks, with high case attack rates, may occur following heavy late summer or autumn rains (e.g. in 2000, Figure 6). Most cases occur between February and June (Figure 54B).

Midwest and Wheatbelt

RRV activity is sporadic in these regions, with cases generally occurring individually or in small clusters only. Overall, the risk of RRV is normally lower than most other regions (Figure 53).

Cases can occur following above average spring/summer rains associated with southern cold fronts, or after heavy rains linked to southern extension of monsoonal/cyclonic activity in autumn (Figure 54B).

Rural south-west (including Great Southern and south coast)

More cases of RRV disease are reported from the south-west than any other region in WA. To some extent, this reflects the relatively large population compared with northern and eastern WA. However, attack rates in some regions on the Swan coastal plain between Rockingham and Augusta have been as high as 1.7% of the population. This risk may also extend inland and further south in some 'epidemic' years.

RRV is thought to be endemic (always present) in the south-west, but most cases occur in the warmest months of year (November-March) (Figure 50, Figure 54C). The risk season in non-epidemic years is late spring and summer (September-January). In epidemic years, activity continues into early autumn (March-April).

Perth

Cases are reported from Perth every year, but case investigations show that many of these were exposed to infected mosquitoes elsewhere in WA.

Small clusters of locally acquired cases (no travel history out of metropolitan area) are reported in years with above average late spring/summer rains or following on from high levels of RRV activity in rural areas further south. Semi-rural and outer metropolitan suburbs generally experience the highest attack rates during metropolitan 'outbreaks' (Figure 55). This is probably linked to abundance of non-human animal hosts.

A high level of RRV activity on the Swan Coastal Plain south of Perth (e.g. Mandurah-Busselton) usually precedes an increase in RRV activity in Perth.

Even during outbreaks, the overall attack rate (cases per head of population) in Perth is less than 1/8th of that in rural areas further south (e.g. Figure 53).

Virus activity was widespread across Perth during a major outbreak in 2011/12, with 739 cases occurring across 211 suburbs. However, only eight suburbs recorded more than five cases in any one month. This suggests that virus activity in Perth occurs in relatively low-level, inefficient cycles.



Figure 54A-D: Seasonality of cases of RRV disease in different regions of WA. Graphs show cumulative (total) cases reported for each month of the year between July 2002 and June 2013 for A) Kimberley and Pilbara, B) Gascoyne, Midwest and Wheatbelt, C) Perth and Southwest D) Great Southern and Goldfields.

Table 17 provides details of the relative requirements for predicting and monitoring RRV activity in the different regions of WA discussed above. Details of the risk season, risk predictors, surveillance methods and known or suspected vector mosquito species in each region are also provided.

Table 17: Proposed need and methods for predicting and monitoring RRV activity in meteorological districts of WA, based on results obtained from studies at the Arbovirus Laboratory, Department of Microbiology, The University of Western Australia.

Meteorological district	Need for surveillance [†]	Risk season	Risk predictors	Surveillance methods	Vector mosquito species*		
North and East Kimberley	+++	Dec-Apr	Amount of wet season rain, Low daily max. temps after first wet season rains Outbreaks in Northern Territory?	Monitor risk predictors, including cases in Northern Territory, mosquito population size, species composition, and infection prior to and during risk season	Ae. vigilax (coastal), Ae. normanensis, Cx annulirostris, Ae. pseudonormanensis [‡] , Ae. eidsvoldensis [‡] (inland)		
West Kimberley	+++	Jan-May	Wet season rain followed by below average daily temperatures, time since previous outbreak, large populations of infected <i>Ae. vigilax</i> (coastal areas only)	Monitor risk predictors, size, species composition and infection of mosquito population prior to and during risk season	Ae.vigilax(coastal),Ae.normanensis,Ae. $pseudonormanensis^{\dagger}$,Cx.annulirostris^{\dagger}, (inland)		
De Grey	+++	Jan-Jul	Heavy rains followed by moderate temperatures, high humidity?, outbreaks in West Kimberley, south- west, large populations of infected <i>Ae.</i> <i>vigilax</i> (coastal regions), time since previous outbreak	Monitor risk predictors, size, species composition and infection of mosquito population prior to and during risk season	<i>Ae. vigilax</i> (coastal), <i>Ae.</i> E.N. Marks' sp. #85, <i>Ae.</i> <i>normanensis</i> [‡] , <i>Ae.</i> <i>eidsvoldensis</i> [‡] , <i>Ae. tremulus</i> , <i>Ae.</i> <i>pseudonormanensis</i> [‡] (freshwater coastal and inland)		
Fortescue	+++	May-Aug	Autumn/winter rains with moderate temperatures, outbreaks in south-west, large populations of infected <i>Ae.</i> <i>vigilax</i> , <i>Ae.</i> E.N. Marks' species #85	Monitor risk predictors, size, species composition and infection of mosquito population prior to and during risk season	Ae. vigilax, Cx. sitiens ^{\ddagger} (coastal), Ae. E.N. Marks' sp. #85 (coastal, inland), Ae. eidsvoldensis ^{\ddagger} , Ae. tremulus ^{\ddagger} , Ae. pseudonormanensis ^{\ddagger} (inland)		
Gascoyne	++	Feb-Jun	Autumn rains, possibly tides in coastal regions	Monitor risk predictors, size, species composition and infection of mosquito population prior to and during risk season	<i>Ae. vigilax, Ae. camptorhynchus</i> [‡] (coastal), <i>Ae.</i> E.N. Marks' species #85 [‡] , <i>Ae. eidsvoldensis</i> [‡] , <i>pseudonormanensis</i> [‡] (fresh water coastal and inland)		

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Table 17: (cont)Proposed need and methods for predicting and monitoring RRV activity in meteorological districts of WA, based on results
obtained from studies at the Arbovirus Laboratory, Department of Microbiology, The University of Western Australia.

Meteorological district	Need for surveillance [†]	Risk season	Risk predictors	Surveillance methods	Vector mosquito species*		
Murchison	++	Feb-Jun	Autumn rains, large populations of <i>Ae. sagax</i> , possibly <i>Ae. camptorhynchus</i> , cases in coastal, salt-affected areas.	Monitor risk predictors, size, species composition and infection of mosquito population prior to and during risk season	<i>Ae. sagax</i> (freshwater sites), <i>Ae. camptorhynchus</i> [‡] (salt-affected sites).		
North East	+	Apr-Jun	Probably heavy autumn rains leading to breeding of species in the genus <i>Aedes</i> and cases elsewhere in WA	Monitoring of risk predictors during risk season or after predisposing climatic conditions	<i>Ae. sagax</i> [‡] , <i>Ae.</i> E.N. Marks' species #85 [‡] , <i>Ae. pseudonormanensis</i> [‡]		
South East	++	Mar-Jun	Heavy autumn rains, time since previous outbreak, human cases in south-west, large populations of <i>Ae.</i> <i>sagax, Ae. camptorhynchus</i> and other temporary groundpool-breeding species	Monitoring of risk predictors, breeding and infection of <i>Aedes</i> during risk season only	Ae. sagax [‡] , Ae. camptorhynchus [‡] , Ae. E.N. Marks' species #85 [‡] , Ae. eidsvoldensis [‡]		
Eucla	-	none identified	-	not applicable unless human population increases considerably	-		
North Coastal	+	Jan-May	Heavy late spring/summer rains, human cases elsewhere in south- west	Monitoring of risk predictors, species composition and size of suspected vector mosquito population during risk season or after predisposing climatic conditions	<i>Ae. camptorhynchus</i> [‡] , possibly temporary freshwater ground poolbreeding species		
North Central	+	Mar-Jul	As for North Coastal	Monitoring of risk predictors, species composition and size of suspected vector mosquito population during risk season or after predisposing climatic conditions	<i>Ae. camptorhynchus</i> [‡] , possibly temporary freshwater ground poolbreeding species		

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 Table 17: (cont) Proposed need and methods for predicting and monitoring RRV activity in meteorological districts of WA, based on results obtained from studies at the Arbovirus Laboratory, Department of Microbiology, The University of Western Australia.

Meteorological district	Need for surveillanc e [†]	Risk season	Risk predictors	Risk predictors Surveillance methods				
South Central	+	Nov-Mar	Heavy summer rain, human cases elsewhere in south-west	Monitoring of risk predictors, species composition and size of suspected vector mosquito population during risk season only	<i>Ae. camptorhynchus</i> [‡] , possibly freshwater ground pool-breeding species			
Central Coastal	++	Oct-Mar	Late spring/summer rains, abnormally high tides?, high levels of activity in Peel, Leschenault localities, time since previous outbreak	Monitoring of risk predictors, species composition and size of suspected vector mosquito population during risk season only	Ae. camptorhynchus, Ae. vigilax, Cq. sp. near linealis, Cx. annulirostris [‡] , Ae. notoscriptus [‡]			
South Coastal	++++	Aug-Apr	Abnormally high spring and summer tides, above average late spring-summer rainfall, above average winter/spring temperatures, amount of time since last outbreak, large populations of <i>Ae. camptorhynchus</i> persisting into spring and summer, large summer populations of <i>Ae. Vigilax</i>	Monitoring of risk predictors (including Southern Oscillation Index), year-round monitoring of species composition and size of mosquito populations and rapid determination of infection levels in probable enzootic foci (Peel, Leschenault, Capel-Busselton localities)	Ae. camptorhynchus, Ae. vigilax, Ae. clelandi (coastal regions), other freshwater breeding species (Cq. sp. near linealis, Cx. annulirostris?) in inland areas			
Perth metropolitan area	+ - +++ (depending on location & incidence)	Nov-Apr	Above average late spring/summer rains, abnormally high tides?, high levels of activity in Peel, Leschenault localities, time since previous outbreak, larger than average populations of <i>Cq.</i> species near <i>linealis</i> and <i>Cx. annulirostris</i> mosquitoes, possibly <i>Ae. notoscriptus</i> and <i>Ae. camptorhynchus</i> also	Monitoring of risk predictors, species composition and size of suspected vector mosquito population during risk season only	Cq. sp. near linealis, Cx. annulirostris [‡] , Ae. camptorhynchus [‡] , Ae. vigilax, Ae. notoscriptus [‡]			

[†] ++++, essential, +++ highly desirable, ++ desirable, + limited surveillance when possible, based on number of cases and case attack rate, 1984-2011.

mosquitoes denoted by ‡ are included on the basis of abundance during outbreaks of human disease, unpublished studies (M.D.A. Lindsay, C.A. Johansen, A.K. Broom, N.M. Oliveira, A.E Wright, S.A. Harrington, J.S. Mackenzie, and others, unpublished results) or results from studies in other regions of Australia.



Figure 55: RRV cases per 100,000 residents by local government regions in the Perth metropolitan area during major south-west outbreaks in 2003/04 and 2011/12. In both outbreaks, rates were generally highest in outer metropolitan local government areas (denoted with an asterisk).

BARMAH FOREST VIRUS

BACKGROUND

Barmah Forest virus (BFV), like RRV, is a uniquely Australian mosquito-borne virus. It belongs to the same group of viruses (alphaviruses) as RRV. BFV was first isolated from *Culex annulirostris* mosquitoes collected at the Barmah Forest on the Murray River, northern Victoria, 1974.

Human infection with BFV was demonstrated in NSW in 1986. Human disease due to infection with the virus was first reported in 1988, also in NSW. Human cases and isolations from mosquitoes have now been reported from most Australian states (Table 5).

Similarities between symptoms caused by infection with BFV and RRV probably means that BFV disease has been under-diagnosed. Serological tests, now widely available, can differentiate between infection with the two viruses.

Nationally, the incidence of BFV disease prior to 2003 was about 18% of that of RRV disease. Between 2003 and 2012, the incidence has doubled to about 36% of that of RRV disease, although this ratio varies substantially from region to region and year to year. It is difficult to say whether this observed increase (e.g. Table 5, Figure 9) is due to increased virus activity, increased recognition and reporting of the disease, or both. Regardless, it is clear that BFV is a cause of mosquito-borne illness that requires serious consideration.

In New South Wales, 2% of the population has antibodies to BFV. In Queensland, 6.5% of people have been infected (compared to 31.6% for RRV). Approximately 0.23% of the

resident population in Queensland becomes infected with BFV each year. Limited surveys in the south-west of WA suggest that 1-2% of the population have antibodies to BFV.

Year	WA	ACT	NSW	NT	QLD	SA	TAS	VIC	Aust
1995	16	5	271	13	446	3	0	13	767
1996	52	2	172	30	573	3	0	85	917
1997	63	0	185	39	361	3	0	35	686
1998	18	1	134	24	330	1	0	17	525
1999	46	0	248	18	310	0	2	13	637
2000	58	0	197	11	345	12	1	19	643
2001	75	2	396	37	602	6	1	21	1140
2002	40	0	395	23	386	4	0	60	908
2003	21	1	451	14	869	2	0	8	1366
2004	70	2	401	22	581	7	0	16	1099
2005	79	0	449	51	680	41	1	14	1315
2006	179	8	642	130	950	187	0	31	2127
2007	129	6	574	91	822	60	0	25	1707
2008	156	7	530	75	1242	37	1	32	2080
2009	142	3	359	118	796	36	3	17	1474
2010	75	4	264	82	908	58	2	77	1470
2011	150	2	459	63	872	129	2	187	1864
2012	215	2	348	87	982	48	0	40	1722
Totals	13532	157	15396	4712	44264	5238	590	7923	91812
Annual average	634	8	757	230	2165	258	29	392	4472

 Table 18: Notifications of Barmah Forest virus infection received by State and Territory health authorities, 1995 to 2012.

Data courtesy of the National Notifiable Diseases Surveillance System, Australian Government Department of Health and Ageing (these totals may differ from WA Dep't of Health figures due to differences in reporting methods and criteria). 2013 data is currently not available.

BFV IN WA

BFV was first isolated from several mosquito species from the south-east Kimberley in 1989. The first reported cases in WA were from the Murchison, Gascoyne and Pilbara regions in 1992 and the Kimberley in 1993, during larger outbreaks of RRV disease. Several isolates of BFV were made from mosquitoes collected during this time.

There is no evidence of BFV in south-western Australia prior to 1993 (from mosquito/virus surveillance, human cases or serological testing of animals). The first true outbreak (~30 cases) occurred in the south-west of WA in the latter half of 1993 and early 1994, in the apparent absence of RRV activity.

BFV was made a notifiable disease (reportable by medical practitioners) in WA in June 1994. Since then, the virus has been annually active with moderate numbers of cases in different regions of WA in most years. The State's largest outbreak (199 cases) occurred in 2008/09 (Figure 9).The annual average number of cases of BFV disease in WA increased over 3 times from 42 cases per year from 1996/97 to 2004/05 to 145 cases per year from 2005/06 to 2011/12 (Figure 56).



Figure 56: Monthly incidence of BFV disease in WA, July 1996-June 2012. Data from 1 Jan 2000 onwards include laboratory reports received as well as doctor notifications. Numbers above graph bars are total cases in WA by financial year (July to June).

CLINICAL FEATURES OF BARMAH FOREST VIRUS DISEASE

1. Symptoms

Symptoms of BFV disease are generally similar to those of RRV disease: i.e. rash, joint manifestations and constitutional effects.

Experience from some outbreaks suggests that the rash may be more common (90% of cases) but rheumatic (50% of cases) and constitutional symptoms are milder than for RRV disease.

2. Course of the illness

Symptoms usually persist for several weeks, but may resolve quickly. Chronic joint symptoms and fatigue are less likely than with RRV disease.

3. Diagnosis

Diagnosis, as for RRV, is confirmed through blood test(s) ordered by a doctor, along with clinical presentation and travel history to affected areas.

4. Treatment

As with RRV, there is no vaccine or specific treatment for BFV disease. Methods used to manage symptoms of RRV disease (described above) are likely to be helpful.

MOSQUITO VECTORS OF BFV

As with RRV, BFV appears to have a broad range of mosquito vectors. The virus has been isolated from field caught specimens of over 20 species, and a number of these have been shown to be competent vectors in the laboratory.

Several major rural pest species including *Ae. vigilax*, *Ae. camptorhynchus* and *Cx, annulirostris* are regarded as important vectors. *Ae. notoscriptus* and *Cq. linealis* (*Cq.* sp. near *linealis* in WA) are probably involved in urban areas. As with RRV, other species may be important in specific regions or environments and further studies are required to define these more completely.

VECTORS OF BFV IN WA

Mosquito species from which BFV has been isolated in WA are listed in Table 19. In the north of WA, *Ae. vigilax* is commonly infected in coastal regions, whilst various fresh water-breeding *Aedes* and *Anopheles* mosquitoes and *Cx. annulirostris* have been shown to be infected with BFV in inland areas.

Ae. camptorhynchus appears to be an important vector in coastal regions of the south-west. *Cx. annulirostris* and *Cq.* species near *linealis* may be vectors in areas where fresh water with emergent or fringing vegetation is abundant. BFV has been isolated from *Cx. annulirostris* in the southern suburbs of Perth, indicating that transmission cycles can occur in the metropolitan area.

VERTEBRATE HOSTS OF BFV

Little is known about the animal hosts of BFV. Surveys in eastern and western Australia indicate that marsupials (kangaroos, wallabies, possums, koalas), cattle and horses are frequently infected. However, studies about the role of most species in amplifying the virus have not yet been undertaken. Possums cats and dogs have been shown not to develop viraemias that would infect *Ae. vigilax* feeding on them.

Isolates of BFV from around Australia are genetically very similar which suggests that the natural hosts of BFV may be more mobile than the macropods associated with RRV. The possibility that humans may be amplifying and moving the virus around Australia has not yet been ruled out.

ECOLOGY

The ecology of BFV was initially presumed to be fairly similar to that of RRV as both are alphaviruses, appear soon after heavy rains and may have similar mosquito species as vectors. However, data from recent outbreaks in WA and elsewhere in Australia suggest that the ecologies of the two viruses may differ considerably.

Outbreaks of BFV and RRV generally occur when vector mosquitoes are abundant. In some cases, outbreaks of both have occurred simultaneously, but in others, activity of one has been independent of the other. In WA, BFV and RRV have been isolated from different individuals of the same mosquito species collected in the same trap, providing clear evidence that the two viruses can co-circulate.

Case attack rates with BFV in WA appear to be much lower than for RRV during outbreaks, despite relatively high infection rates in mosquitoes.

It is not known whether BFV persists in the environment like RRV. It has been isolated in midwinter in the south-west of WA, suggesting it can survive in temperate regions in low-level maintenance cycles.

Region	Species*						
Kimberley	Aedes (Neomelaniconion) lineatopennis (Ludlow)						
	Anopheles (Cellia) amictus Edwards						
	Anopheles (Cellia) annulipes s.l. Walker						
	Culex (Culex) annulirostris Skuse						
	Aedes (Ochlerotatus) eidsvoldensis (Mackerras)						
	Aedes (Ochlerotatus) pseudonormanensis (Marks)						
	Aedes (Pseudoskusea) bancroftianus (Edwards)						
	Aedes (Ochlerotatus) normanensis (Taylor)						
	Aedes (Ochlerotatus) vigilax (Skuse)						
Pilbara	Anopheles (Cellia) amictus						
	Culex (Culex) annulirostris						
	Aedes (Ochlerotatus) normanensis						
	Aedes (Ochlerotatus) vigilax						
	Aedes E.N. Marks species #85						
Gascoyne	Culex (Culex) quinquefasciatus Say						
	Aedes (Ochlerotatus) eidsvoldensis						
	Aedes E.N. Marks species #85						
South-west	Coquillettidia (Coquillettidia) sp. near linealis Marks and Harris						
	Culex (Culex) annulirostris						
	Culex (Culex) globocoxitus Dobrotworski						
	Culex (Culex) quinquefasciatus						
	Aedes (Finlaya) alboannulatus (Macquart)						
	Aedes (Ochlerotatus) camptorhynchus (Thomson)						
	Aedes (Ochlerotatus) clelandi (Taylor)						
	Aedes (Ochlerotatus) ratcliffei (Marks)						
	Aedes (Ochlerotatus) vigilax						
Perth	Culex (Culex) annulirostris						

Table 19: Mosquito species from which Barmah Forest virus has been isolated in WA

*Known or highly suspected vector species are shown in bold. Vector competence studies are required to determine the importance of other listed species as vectors of BFV.

SURVEILLANCE OF RRV AND BFV IN WA

MONITORING OF WEATHER, MOSQUITOES AND VIRUS ACTIVITY

Early warning of impending outbreaks of RRV and/or BFV is essential to enable up-scaling of vector control programs and timely and targeted warnings via the media and through other networks to affected communities. Mosquito-Borne Disease Control (MBDC) in the Environmental Health Hazards Unit (EHH) of the WA Department of Health and Arbovirus Surveillance and Research Laboratory, at The University of Western Australia undertake a state-wide program of mosquito-borne disease surveillance for this purpose. The program also provides valuable research data to help focus mosquito management programs towards high risk areas and to target mosquito species of public health importance.

<u>Mosquito and RRV/BFV surveillance is carried out by:</u> the Arbovirus Surveillance Laboratory, UWA, in conjunction with The WA Department of Health and Local Governments.

<u>In the north and interior</u> the program consists of monitoring of rainfall, tides, temperature, human cases; opportunistic sampling of mosquito populations, viruses and immunity of vertebrate hosts.

<u>In the south-west</u> the program comprises ongoing surveillance of mosquito populations in high risk areas by monitoring selected trap sites located at and between vector breeding sites and residential areas. This is done using EVS/CO₂ traps at suitable intervals after high tides or rainfall. Trapping is carried out fortnightly during periods of likely virus activity (September-May) and monthly at other times of the year. Mosquitoes are also processed for virus isolation to determine infection rates.

Other elements of the program include: monitoring of tides, temperature, rainfall and human cases; analysis of virus strains; monitoring of immunity of vertebrate hosts; and development of new, more rapid, systems to enable testing of mosquitoes from throughout WA.

The overall aim of the program is to predict major outbreaks, enable implementation of effective mosquito control measures and media warnings to the public, leading to a reduction in the incidence of human disease.

ENHANCED SURVEILLANCE OF HUMAN CASES

Accurate information about when and where people are exposed to RRV, BFV and other arboviruses is important for improving the timing and effectiveness of risk management programs in WA. In particular, the information helps to ensure that limited resources for mosquito control programs are well directed, that public warnings and education are timely and that advice to planning authorities is accurate.

For this reason, EHH MBDC requests assistance from Local Government Environmental Health Officers with administering case follow-up questionnaires for all RRV and BFV cases. This involves a series of simple questions about each patient's date of onset of symptoms, travel history prior to becoming ill and recollection of biting mosquitoes.

The process by which the questionnaires are completed is as follows:

For all regions outside Perth (metropolitan area health services), Doctor's send notification of serologically (laboratory) confirmed cases of RRV or BFV disease to the relevant regional Population Health Unit. Staff from the Population Health Unit then fax a copy of the notification to the local government in which the patient resides. Sufficient contact details are generally available on the notification form to allow the Environmental Health Officer to telephone or visit the patient and administer the questionnaire. The completed questionnaire is then forwarded to EHH MBDC for analysis.

Within Perth, the process is as above, except that Doctor's notify the Communicable Disease Control Directorate of the Department of Health directly. An officer from that Directorate then forwards a copy of each notification to EHH MBDC, which in turn sends a copy through to the relevant metropolitan local government.

The RRV/BFV enhanced surveillance questionnaire was recently updated and simplified. The new version is also designed to be completed by the patient, if appropriate. A copy of the new questionnaire is appended to this chapter (see Appendix 1) for reference, but MBDC will provide electronic and hard copies to all local governments in WA, whenever the questionnaire is updated. Please contact EHH MBDC if you require copies of the questionnaire or have any questions about its use.

All completed questionnaires should be sent directly to EHH MBDC at:

Mosquito-Borne Disease Control, Environmental Health Hazards Unit Environmental Health Directorate Department of Health PO Box 8172 Perth Business Centre, WA, 6849 Phone: (08) 9285 5500 Fax: (08) 9383 1819 E-mail: mosquito@health.wa.gov.au

SUGGESTED READING:

- 1. Flexman, J.P., Smith, D.W., Mackenzie, J.S., Fraser, R.E., Bass, S., Hueston, L., Lindsay, M.D.A. and Cunningham, A.L. (1998) A comparison of the diseases caused by Ross River virus and Barmah Forest virus. *Medical Journal of Australia*, 169: 159-163.
- 2. Harley, D., Sleigh, A. and Ritchie, S. (2001) Ross River virus transmission, infection and disease: a cross-disciplinary review. *Clinical Microbiology Reviews*, 14: 909-932.
- 3. Susan P. Jacups, Peter I. Whelan, Bart J. Currie. (2008) Ross River Virus and Barmah Forest Virus Infections: A Review of History, Ecology, and Predictive Models, with Implications for Tropical Northern Australia. *Vector-Borne and Zoonotic Diseases*, 8(2): 283-298.
- 4. Johansen, C.A., Broom, A.K., Lindsay, M.D.A., Maley, F.M., Power, S.L., Gordon, C.J. and Smith, D.W. (2005) Surveillance of arboviruses in mosquitoes from the southwest of Western Australia between 2000 and 2004. *Arbovirus Research in Australia*, 9: 159-163.
- 5. Kelly-Hope, L.A., Purdie, D.M and Kay, B.H. (2004) Ross River virus disease in Australia, 1886-1998, with analysis of risk factors associated with outbreaks. *Journal of Medical Entomology*, 41: 133-150.
- Lindsay, M., Oliveira, N., Jasinska, E., Johansen, C., Harrington, S., Wright, A., Smith, D. and Shellam, G. (1997) Western Australia's largest recorded outbreak of Ross River virus disease. *Arbovirus Research in Australia*, 7: 147-152.
- Lindsay, M.D., Johansen, C.A., Smith, D.W., Wallace, M.J. and Mackenzie, J.S. (1995) An outbreak of Barmah Forest virus disease in the south-west of Western Australia. *Medical Journal of Australia* 162: 291-294.
- Lindsay, M.D.A., Breeze, A.L., Harrington, S.A., Johansen, C.A., Broom, A.K., Gordon, C., Maley, F., Power, S., Jardine, A. and Smith, D. (2005) Ross River and Barmah Forest viruses in Western Australia, 2001/01 – 2003/04: contrasting patterns of virus activity. *Arbovirus Research in Australia*, 9: 194-201.
- 9. Mackenzie, J.S., Lindsay, M.D. and Broom, A.K. (2000) Effect of climate and weather on the transmission of Ross River and Murray Valley encephalitis viruses. *Microbiology Australia*, 21(2): 20-24.
- 10. Mackenzie, J.S., Broom, A.K., Hall, R.A., Johansen, C.A., Lindsay, M.D., Phillips, D.A., Ritchie, S.A., Russell, R.C. and Smith, D.W. (1998) Arboviruses in the Australian region, 1990-1998. *Communicable Diseases Intelligence (Australia)* 22 (6) 93-100.
- 11. Russell, R.C. (2002) Ross River virus: Ecology and Distribution. *Annual Review of Entomology*, 47, 1-31.

- 12. Russell, R.C. and Kay, B.H. (2004) Medical entomology: changes in the spectrum of mosquito-borne disease in Australia and other vector threats and risks, 1972-2004. *Australian Journal of Entomology* 43, 271-282.
- 13. Williams, C.R., Fricker, S.R. and Kokkinn, M.J. (2009) Environmental and entomological factors determining Ross River virus activity in the River Murray Valley of South Australia. *Australian and New Zealand Journal of Public Health*, 33: 284-288.
- 14. Woodruff, R.E., Guest, C.S., Garner, M.G., Becker, N. and Lindsay, M.D.A. (2006) Early warning of Ross River virus epidemics: combining climate and mosquito surveillance data, *Epidemiology*, 17 (5): 1-7.
- Vally, H., Peel, M., Dowse, G.K., Cameron, S., Codde, J.P., Hanigan, I. and Lindsay, M.D. (2012) Geographic Information Systems used to describe the link between the risk of Ross River virus infection and proximity to the Leschenault estuary, WA. Australian and New Zealand Journal of Public Health. 36(3):229-35
- McIver, L., Xiao, J., Lindsay, M.D., Rowe, T., and Yun, G. (2010) A climate-based early warning system to predict outbreaks of Ross River virus disease in the Broome region of Western Australia. Australian and New Zealand Journal of Public Health. 34(1):89-90

Appendix 1: RRV/BFV enhanced surveillance questionnaire

Mosquito-borne DiseaseFollow-up Questionnaire

Background

All cases of mosquito-borne diseases that meet case definition criteria should be notified to the relevant Public Health Unit or Communicable Diseases Control Directorate of the WA Department of Health by the patient's GP.

Ross River virus (RRV) and **Barmah Forest virus (BFV)** diseases are the two most commonly spread by mosquitoes in Western Australia.

Murray Valley encephalitis (MVE) virus, although much rarer, can cause potentially fatal encephalitis in humans. It generally only occurs in the northern regions of Western Australia.

Infections with RRV or BFV can result in a range of symptoms that may last for weeks or even months. Infections with MVE can be severe and potentially fatal. Since there is no vaccine or cure for any of these diseases, the only way to prevent illness is to reduce the potential for interaction between mosquitoes and people.

This questionnaire is designed to assist in identifying the most likely time and place of exposure to mosquito-borne diseases. The Environmental Health Directorate of the WA Department of Health uses this information to define high risk regions and direct mosquito management priorities throughout WA.

Confidentiality

Information collected from this questionnaire will remain completely confidential. It will be used solely for the purpose of guiding the WA Department of Health to prevent the spread of mosquito-borne diseases. No information that identifies individuals will be made available outside the WA Department of Health.

Return completed forms

This questionnaire can be completed by the patient, medical personnel or local government Environmental Health staff. Please return the completed questionnaire by email, fax or post to:

Scan and Email: mosquito@health.wa.gov.au	Fax: (08) 9383 1819	Post: Mosquito-Borne Disease Control Environmental Health Hazards Environmental Health Directorate WA Department of Health PO Box 8172 Porth Business Contro WA 6849
		Perth Business Centre WA 6849

Further information

Please contact Mosquito-Borne Disease Control in the Environmental Health Hazards Unit on email <u>mosquito@health.wa.gov.au</u> or phone (08) 9285 5500 for further information or to request an electronic version of this questionnaire (that can be completed online and emailed back).

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Section 1 Patient Details				Today's Date	/	/				
Name:				Notification ID:		-				
Gender (tick box) \Box M \Box F				Date of Birth:	/	/				
Home street address (not PO Box):									
Town / Suburb: State: Postcode:										
Please indicate who completed this form (tick box and provide further details where required):										
Person with the illness										
EHO (Name & Local Govt) -										
Other (Name & position/relation to	oatient) -									
The following questions re	late to	the pe	rson v	with the illness						
1) Which disease/s did you have (ick more i	than one	box if yo	ou were diagnosed with n	nore than one	virus)				
□ Ross River virus (RRV)			ay Valle	y encephalitis (MVE)						
Barmah Forest virus (BFV)		□ Othe	r (e.g. K	unjin) <i>please specify</i>						
2) What is your occupation?										
3) Does your job (or usual daily ro	utine if yo	ou do no	t work)	require you to be mos	tly: 🗆 In	doors 🗆	Outdoo	rs		
4) Have you noticed mosquitoes a	t:	Home	e: 🗆 Ye	es □ No	Work:	res 🗆 No	C			
The following questions re	late to	the pa	tient's	s symptoms and p	ossible ex	posure	e			
5) What was the approximate date	you first	noticed	sympto	oms? (For common symp	otoms please s	see next q	uestion)			
/ / Day MonthY	ar	OR		early / mid / l	ate/	Year				
6) Listed below are common symp	toms. Ple	ease tick	the bo	exes if you experienced	l any of these	symptor	ms:			
Common RRV / BFV symptoms:	Com	mon MVE	sympton	ns:	Common Kunjin symptoms:					
Headaches Tiredness		lausea			Fever	□ Joint pai	in			
□ Sore muscles □ Nausea		leadache		Tiredness	□ Headache □ Nausea					
Skin rash		leck stiffne	ess	Fever	Neck stiffn	ess				
Painful / swollen joints		rowsiness	s, floppine	ess or irritability in children						
\Box Tingling in palms or soles of feet										
 7) Symptoms of RRV, BFV or MVI infected mosquito. Knowing whe where you were infected. Please indicate all suburbs or town 	E disease ere you h ns you vis	e first ap ave bee sited in t	pear be n during he 3 we	etween 3 days and 3 w g these 3 weeks , can l eeks before symptom	veeks after be help determin is began (e.g	eing bitte ne the mc g. Albany	n by an ost likely , Broome	place		
Joondalup) and tick the appropr	iate box.	Note: Mo	ore spec	ific details about these lo	cations are rec	quested of	n the next	page.		
Suburb / Town	Reside	Work	Visit	Suburb / Town		Reside	Work	Visit		
1)				4)						
2)				5)						
3)				6)				\Box		

The	following	section	relates	to	the	most	likely	place	the	patient	was	exposed	to
mos	mosquitoes in the 3 weeks before symptoms began												

- Please indicate the most likely place where you were bitten by mosquitoes in Section 2 (below);
- If there was more than one place, use **Section 3** (next page) to indicate another place of exposure to mosquitoes;
- Complete Part A if you know the street address of the location where you were most likely bitten by mosquitoes (*e.g. Lot 47 Thompson Road, Baldivis*); **OR**
- Complete Part B to describe the location if you do not know the street address (e.g. southern side of Thompson Lake in Thompson Park, near Johnson Street, Carnarvon WA);
- It is **important** that you provide **as much detail as possible.** We need to **identify the location** to a street or lot number or a particular part of a recreational area.

Section 2 Most likely place of exposure					
(Please answer Part A OR Part B and questions 8-12)					
Part A - Known street address					
House / Lot N°: Street:					
Suburb /Town:	State: Postcode:				
OR					
Part B - Geographical location					
Location description:					
Nearest Suburb/Town: State: Postcode:					
Nearest Landmark / Street intersection / Other detail etc to help us	is pin-point the exposure location:				
8) Please indicate approximate date/s you were at the above loc 1 st week of January; 6-10 th April):	cation in the 3 weeks before you became ill <i>(e.g.</i>				
9) Was the majority of your time spent at the above location:	□ Indoors □ Outdoors				
10) Did you notice mosquitoes at the above location?	□ Yes □ No				
11) Do you remember being bitten by mosquitoes at the above lo	ocation? Yes No				
12) Please provide further details if appropriate [e.g. Do yo same place who have become ill? What activities were you mosquitoes - indoors/outdoors, near waterbodies/in the bush, etc]	bu know of other persons who have been to the I doing (camping etc)? Where did you notice]:				

Section 3 Second most likely place of e	xposure							
(Please answer Part A OR Part B and questions 13-17)								
Part A - Known street address								
House / Lot N ^o : Street:								
Suburb /Town:	State:	Postcode:						
	OR							
Part B - Geographical location								
Location description:								
Nearest Suburb/Town:	Nearest Suburb/Town: State:							
Nearest Landmark / Street intersection / Other detail etc	c to help us pin-point the exp	oosure location:						
13) Please indicate approximate date /s you were at th	e above location in the 3 w	eeks before you became ill <i>(e.g. 1st</i>						
14) Was the majority of your time spent at the above lo	ocation:	□ Indoors □ Outdoors						
15) Did you notice mosquitoes at the above location?		□ Yes □ No						
16) Do you remember being bitten by mosquitoes at th	e above location?	□ Yes □ No						
17) Please provide further details if appropriate [e.g. Do you know of other persons who have been to the same place who have become ill? What activities were you doing (camping etc)? Where did you notice mosquitoes - indoors/outdoors, near waterbodies/in the bush, etc]:								
Other information								
Please Add any further details that may help us in definition lines for previous answers:	ing an location where you r	nay have been infected or as extra						

24. MURRAY VALLEY ENCEPHALITIS AND KUNJIN VIRUS ACTIVITY IN WESTERN AUSTRALIA

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Murray Valley encephalitis virus (MVEV) and Kunjin virus (KUNV) are two medically important flaviviruses that are enzootic (always present) in northern Australia, particularly the north of Western Australia (WA) and the Northern Territory (NT) (Figure 57). MVEV can cause potentially fatal encephalitis in humans. KUNV is usually associated with milder febrile illness and only occasionally causes encephalitis. The disease caused by these viruses was previously known as Australian encephalitis but is now generally referred to as Murray Valley encephalitis and Kunjin virus disease. MVEV has been responsible for epidemics or scattered cases of encephalitis in northern and eastern Australia and in Papua New Guinea (PNG). Approximately 5-10% of arboviral encephalitis cases in Australia are due to KUNV infection. MVEV and KUNV are members of the family *Flaviviridae* and are included in the Japanese encephalitis antigenic subgroup. KUNV is now considered a subtype of West Nile virus. Other flaviviruses found in Australia include Japanese encephalitis, Alfuy, Kokobera, Stratford, Edge Hill and the dengue viruses.

BACKGROUND

Epidemics of lethal encephalitis in the Murray Valley region of New South Wales (NSW), caused by an unknown agent, were reported as far back as 1916. The disease was known then as "Australian X disease". MVEV was first isolated from the brains of patients who died during a major epidemic of encephalitic disease in the Murray Valley region of southeastern Australia in 1951. Mosquitoes were recognised as the major vectors in the early 1950s but it wasn't until 1960 that the first field isolates of MVEV and KUNV were obtained from *Culex annulirostris* mosquitoes collected at the Mitchell River Mission in north Queensland. The next major epidemic was in 1974 when cases were reported from all mainland States.



Figure 57: MVEV activity in Australia (adapted from Williams et al. 2010)

MURRAY VALLEY ENCEPHALITIS AND KUNJIN VIRUS DISEASE

INCIDENCE OF DISEASE

Early serological studies have suggested that only 1 in about 1000 people who are bitten by an infected mosquito will develop symptoms of disease. However, studies at Billiluna (a small community in the southeast Kimberley region) and in the Midwest have shown that in some communities the clinical to sub-clinical ratio may be higher, with closer to 1:150 to 1:250 infections resulting in disease. Previous research by the Arbovirus Surveillance and Research Laboratory at the University of Western Australia (UWA) has shown that in most Kimberley Aboriginal communities 90% or more of children seroconvert to MVE by the age of ten. In Caucasians, higher seroconversion rates are observed in adults and these are usually correlated with the length of residence in the region. There is also a high seroprevalence of antibodies to KUNV in people living in northern Australia.

NUMBER AND LOCATION OF HUMAN CASES

The last major epidemic in Australia was in 1974 when human cases were reported from all mainland Australian states, and the fatality rate was 22% (13/58) (Table 20). Since 1974, cases have occurred in most years, mainly in northern Australia, with major outbreaks in WA in 1981, 1993, 2000 and 2011, and in central Australia in 2000, 2001 and 2011. One case of MVE occurred in NSW in 2008, coupled with unusual MVEV activity in sentinel chickens. This is the first time such widespread activity has occurred since 1974. Before 2000, the WA cases were principally from the Kimberley region (25/30) and 23% of cases were fatal. Most of the severe cases were Aboriginal infants or visitors to the Kimberley. These cases occurred during or shortly after the end of the wet season (February-June). Since 2000 most cases involved non-Aboriginal adults (residents or visitors), including one Canadian tourist who died of MVE in 2011. In 2000 extreme rainfall conditions occurred in northern and central Australia and cases of MVE were diagnosed in people as far south as Dongara in the Midwest region of WA. There were 16 clinical cases of MVE and two non-encephalitic cases reported during 2000, including two fatalities. In 2011 there were sixteen cases of MVE including one nonencephalitic case and three fatalities. This outbreak was also associated with extremely high rainfall.

Twenty eight cases of KUNV disease have been reported in Australia since 1974. Twelve were from WA, seven from the NT, five from Queensland, two from Victoria and two from NSW. Although the majority of KUNV disease cases were mild, a number of the NT patients from 2000 and 2001 developed encephalitis.

In 2011 there was also a large outbreak of arboviral disease (including disease caused by infection with KUNV and MVEV) in horses, preceded and concurrent with widespread seroconversions in sentinel chickens. The outbreak was characterised by neurological and muscular disease, and predominantly occurred on the east coast of Australia, and is thought to be largely due to heavy rainfall in the spring and summer of 2010/11. Surprisingly only one human case of MVE from NSW, two from SA and none from Victoria were confirmed, nor was there a confirmed case of KUNV disease in humans in eastern Australia during the same season.

SYMPTOMS OF MURRAY VALLEY ENCEPHALITIS AND KUNV DISEASE

Early symptoms of MVE include headache, fever, nausea, vomiting, neck stiffness, disorientation and dizziness. In severe cases, the infection progresses to obvious neurological disease, potentially leading to coma, paralysis and death. Mild cases usually make a complete recovery, but serious, non-fatal cases are often left with severe disabilities including paraplegia, quadriplegia and various mental deficiencies. Historically, young children and the elderly were most at risk and tended to develop more severe disease, however healthy adults were more commonly affected in 2000 and 2011. There is no specific MVE vaccine available at present.

Symptoms of KUNV disease can be similar, but generally less severe than MVE. KUNV can also manifest as symptoms similar to those caused by RRV infection, including polyarthralgia, without the neurological complications that can occur with MVE or severe KUNV infections.

Year	W	Ά	N	Т	QL	D##	NS	SW	V	IC	S	Α #	Total/
	MVE	KUN	year										
1974*	1		5		10		5		27		10		58
1978	4	1											5
1979	1												1
1981	7		1		2								10
1984	2									1			3
1986	1												1
1987			1										1
1988			3										3
1989	1												1
1990	1												1
1991	1	1	2		2			1		1			8
1992						1							1
1993	9		7										16
1994					1								1
1995		1											1
1997	2	2	1	3									8
1998	1	1											2
1999		1											1
2000	11	2	4	1							1		19
2001	_	1	3	2	1								7
2002**	2					-							2
2004			1			2							3
2005		•	1		1	1							3
2006	1	2											3
2008	1		0				1						2
2009	2		2			4							4
2010	0		1	4		I	~				_		2
2011***	9		4				2	1			2		19
E. TOTAL	57	12	36	7	17	5	8	2	27	2	13	0	186

Table 20: Number and location of cases of MVE and KUNV disease for years in which cases occured, 1974 – 2013.

[#]Case in 2000 was undifferentiated MVE/KUN infection; *Cases in 1974 were all reported as MVE; **One MVE case occurred in December 2001 but was associated with the 2001/2002 wet season; ***One MVE case and one KUNV disease case in NSW occurred in December 2011 but was associated with the 2011/12 wet season; ¹Preliminary data available to end of June 2013.

MOSQUITO VECTORS, VERTEBRATE HOSTS AND GEOGRAPHICAL DISTRIBUTION OF MVEV AND KUNV

MVEV and KUNV are both arboviruses (<u>ar</u>thropod-<u>bo</u>rne), and are transmitted by mosquitoes. *Cx. annulirostris* mosquitoes are the main vectors of both MVEV and KUNV in Australia and water birds, particularly herons, are considered to be the major vertebrate hosts. In nature, these viruses cycle between mosquitoes and birds and humans are only incidental hosts.

MVEV has been regularly isolated from mosquitoes collected in the north of WA. A number of virus isolates have also been obtained from the Northern Territory (NT), north Queensland, and southeastern Australia. MVEV has also been isolated from PNG but the virus does not appear to extend into SE Asia. KUNV has been isolated from similar areas within Australia, although it has been isolated more often from southeastern Australia. KUNV has also been isolated from pools of *Cx. pseudovishnui* mosquitoes in Sarawak, Borneo.

MVEV and KUNV are thought to be enzootic (always present) in the Kimberley region of WA, particularly in the Ord River area near Kununurra, and in the Top End of the NT (Figure 57). These viruses are, however, considered to be epizootic (occasionally present) in the Pilbara, Gascoyne, Murchison and Midwest regions of WA, central Australia and northern Queensland.
MVEV has been responsible for occasional epidemics of encephalitis in southeastern Australia, the last being in 1974. Since this time, MVEV has only rarely been detected in southeastern Australia, whereas KUNV has caused a small number of clinical infections in people since 1974. The greater occurrence of KUNV activity in southeastern Australia suggests that the ecology of KUNV may differ from that of MVEV and that KUNV may move more readily from one area to another. In addition, the often milder manifestations of KUNV disease may mean that the incidence of disease in humans may be under-diagnosed. The most recent detections on the eastern seaboard occurred in 2011, when cases of MVE were also diagnosed in people living in southeastern Australia and there were widespread reports of horses being affected after infection with KUNV, MVEV and RRV.

ENVIRONMENTAL CONDITIONS CONDUCIVE TO MVEV AND KUNV ACTIVITY

MVEV and KUNV activity increases during the wet season. In tropical Australia, the wet season usually extends from November/December to April/May and the amount of rain varies from year to year. In the Kimberley region, the mosquito numbers increase during the wet season, which can lead to an increase in virus activity. MVEV and KUNV activity is usually detected in the Kimberley and Pilbara regions each year, and activity can spread further south in years with unusually high rainfall in southerly areas.

In WA, cases of MVE and KUNV disease are rare but the majority of cases have occurred after heavy wet season rains and flooding in the Kimberley region and/or regions to the south or east. Although increased rainfall can lead to high levels of MVEV activity and human cases in the region (as in 1993), this does not always occur (Figure 58). Other factors, including the immunity and availability of vertebrate hosts and the time between outbreaks are also likely to be involved. Elsewhere in epizootic areas, there appears to be a strong association between amount of rainfall and flavivirus activity in sentinel chickens.



Figure 58: Rainfall (Kununurra) and human cases of Murray Valley encephalitis in the Kimberley region of Western Australia, 1989 – 2011.

It is generally believed that MVEV and KUNV moves from enzootic areas in the Kimberley to major epizootic areas further south following periods of heavy rainfall and flooding. It is proposed that virus movement could occur in several ways, including movement of infected

birds from tropical areas, or movement of infected mosquitoes in the wind from one region to another. Once the viruses have been "seeded" in a new area it is possible that they may survive in desiccation resistant eggs of some Aedes species. This mechanism of virus survival is known as vertical transmission. Some of the eggs can become infected via the infected parent. After the next heavy rains the eggs hatch and some of the next generation of mosquitoes may be already infected with the virus. This could be the situation in many areas of northern Australia where regular flavivirus activity occurs. It is also possible that MVEV and KUNV remain active in small enzootic foci that are difficult to detect, with activity increasing and spreading when environmental conditions become suitable.

MVEV AND KUNV SURVEILLANCE IN AUSTRALIA

Regular monitoring of MVEV and KUNV activity is conducted in WA, the NT, NSW, Victoria and occasionally in South Australia and Queensland (currently there are no sentinel chickens in Queensland). The individual program methodologies vary slightly from state to state. however the essence of each surveillance program is similar. Activity of MVEV and KUNV is detected by monitoring seroconversions in sentinel chickens at strategically relevant locations (Figure 59), sometimes supplemented with collections of adult mosquitoes for virus isolation in cell culture or detection using molecular methods. Serum samples are collected from sentinel chickens at weekly or fortnightly intervals and sent to the surveillance laboratories for serological testing. The aim of the surveillance programs is to provide early warning of increased levels of activity of MVEV and KUNV. Media warnings to increase public awareness (via newspapers, radio and television) are distributed by the relevant health departments. Regional Public Health Units may also send out modified warnings to Aboriginal communities in "at risk" areas. Media releases provide advice to residents and travelers of the increased risk of mosquito-borne disease and the need to take adequate precautions against mosquito bites. The information may also be used by relevant local governments and health departments to implement mosquito control measures. In order for the surveillance programs to be successful it is essential that sentinel chickens are bled on a regular basis to determine exactly when seroconversions to MVEV or KUNV occurred. The WA and NT surveillance programs continue throughout the year as MVEV and KUNV are enzootic in parts of these states. Other state programs (NSW, Victoria and South Australia) are conducted during the peak risk season (roughly October/November to April, inclusive). Sentinel chickens are replaced on an annual basis, and if large proportions of a given flock develop antibodies or die. Additional flocks have been added to the programs in response to increased activity of these viruses in epizootic areas or regions where they have recently emerged (eg WA in 2000).

MVEV AND KUNV SURVEILLANCE IN WESTERN AUSTRALIA

In WA, the Arbovirus Surveillance and Research Laboratory at UWA is funded by the WA Department of Health (DOH) to monitor arbovirus activity throughout the state. There are two major components to the flavivirus surveillance program: the sentinel chicken flavivirus surveillance program; and annual mosquito collections and virus isolations from northern WA.

SENTINEL CHICKEN *FLAVIVIRUS* SURVEILLANCE PROGRAM

During 2012/2013 there were 28 flocks located mostly in towns, two large dams and several Aboriginal communities in the Kimberley and Pilbara regions and a single flock in the Gascoyne region. Five additional flocks were located in the Midwest, Wheatbelt and Goldfields regions of WA. Seroconversions in sentinel chickens show that MVEV and/or KUNV were active in all of the last 14 years in WA (Figure 60 and Figure 61). MVEV activity has been demonstrated in most years in at least one centre in the Pilbara, every few years in the Gascoyne and less often in the Midwest/Wheatbelt and Goldfields. Overall KUNV is far less prevalent and has never been detected in the Gascoyne or Goldfields regions of WA.



Figure 59: Location of sentinel chicken flocks in Australia in 2010/11 (Knope et al. 2013).



Figure 60: Percentage of sentinel chicken serum samples testing positive for MVEV antibodies in each region in WA since 1999/2000.



Figure 61: Percentage of sentinel chicken serum samples testing positive for KUNV antibodies in each region in WA since 1999/2000.

MOSQUITO COLLECTIONS AND VIRUS ISOLATIONS

Flavivirus activity is also monitored by collecting mosquitoes from northern WA and identifying and processing them for virus isolation. Annual mosquito collecting trips are conducted towards the end of the wet season in the Kimberley region. The mosquitoes are collected in dry ice baited EVS (encephalitis vector surveillance) traps. The traps are set at or before sunset and retrieved close to sunrise the next morning. The mosquitoes are then frozen on dry ice and transported to Perth. The maintenance of an effective "cold chain" is important to keep any virus present in the mosquitoes viable until they are processed for virus isolation. In the laboratory the mosquitoes are sorted into pools of 25 mosquitoes or less and homogenized. The supernatants are then inoculated onto various cell lines and any viruses isolated are then identified by fixed cell ELISA using a panel of monoclonal antibodies.

MVEV and KUNV are regularly isolated from mosquitoes collected in the Kimberley region. Most of the isolates of MVEV are from pools of *Cx. annulirostris.* MVEV has been isolated from ten other mosquito species in WA, including male tree-hole breeding *Aedes (Macleaya)* species mosquitoes, providing circumstantial evidence for transmission of MVEV from adult female mosquitoes to progeny via the desiccation-resistant eggs, a potential mechanism for survival of viruses during drought. KUNV, Kokobera virus, Alfuy virus and Edge Hill virus have also been isolated from mosquitoes collected in northern WA. Increased MVE isolations are often obtained in years when heavy wet season rains and flooding occurred in the Kimberley region (eg., 1993, 1997, 1999, 2006 and 2009), however increased MVEV activity does not always lead to human cases of encephalitis (eg., 1999). Other factors such as vertebrate host immunity are also likely to be important. Efficient surveillance programs have an important role in reducing disease risk.

Opportunistic mosquito collecting trips are also carried out in the Pilbara and other areas of the northwest after heavy rainfall events likely to cause increases in MVEV activity. Although mosquitoes have not been collected regularly from the Pilbara region, a number of flavivirus isolates have been obtained from this region. MVEV has been isolated occasionally from *Cx. annulirostris* mosquitoes collected at Port Hedland, Newman, Karratha, Harding Dam and

Exmouth. In addition, isolates of KUNV, Alfuy, Kokobera and Edge Hill viruses have been obtained from mosquito collections in the Pilbara.

CONCLUDING REMARKS

The level and extent of MVEV and KUNV activity varies substantially from year to year. Environmental factors such as rainfall heavily influence mosquito vector breeding and survival, however rainfall alone does not predict increases in MVEV and KUNV activity. Other factors such as vertebrate host availability and immunity also have a major influence on the level of MVEV and KUNV activity in an area. The ongoing activity of MVEV and KUNV in WA and the NT and nationwide MVEV and KUNV activity in 2011 highlights the need for efficient surveillance programs across northern Australia. It is difficult to determine how many cases of MVE and KUNV disease are prevented by the early warning of MVEV activity given by these surveillance programs, however we feel that the results are essential to reduce the impact of this disease in "at risk" populations.

Suggested reading:

- Knope *et al.* (2013). Arboviral diseases and malaria in Australia, 2010-2011: annual report of the National Arbovirus and Malaria Advisory Committee 37:E1-E20.
- Marshall, I.D. (1988). Murray Valley and Kunjin encephalitis. In: The Arboviruses: Epidemiology and Ecology, Vol. III. T.P. Monath (ed). CRC Press Inc., Florida, pp. 151-189.
- Mackenzie *et al.*, (1998). Arboviruses in the Australian region, 1990-1998, *Communicable Diseases Intelligence* 22(6): 93-100.
- Broom *et al.*, (2001). An outbreak of Australian encephalitis in WA and central Australia (NT and SA) during the 2000 wet season. *Arbovirus Research in Australia* 8: 37-42.
- Broom *et al.*, (2001). Seroconversions in sentinel chickens provide an early warning of MVE virus activity in WA. *Arbovirus Research in Australia* 8: 43-47.
- Broom et al., (2005). An overview of the flavivirus surveillance program in Western Australia, 2001-2004. *Arbovirus Research in Australia* 9: 64-69.
- Broom et al., (2002) Epizootic activity of Murray Valley encephalitis virus in an Aboriginal community in the southeast Kimberley region of Western Australia: results of cross-sectional and longitudinal serological studies. *American Journal of Tropical Medicine and Hygiene* 67(3): 319-323.
- Broom *et al.* (2002) Investigation of the southern limits of MVE Activity in Western Australia during the 2000 wet season. *Vector Borne and Zoonotic Diseases* 2(2): 87-95.
- Johansen et al. (2010). Arbovirus and vector surveillance in Western Australia, 2004/05 to 2007/08. *Arbovirus Research in Australia* 10: 76-81.

25. DENGUE IN AUSTRALIA, ASIA AND THE PACIFIC

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The World Health Organization has nominated dengue as "the world's most important mosquito-borne virus disease". WHO currently estimates there may be 50 million cases of dengue infection worldwide each year, with some 2,500 million people at risk of the disease. Certainly, dengue is the most important arboviral disease afflicting humans in the Southeast Asian/Western Pacific Region, whether measured in terms of human mortality, or simply human infection and morbidity, and Australia (Queensland) is not excluded from its distribution.



Figure 62: Countries with dengue risk and potential distribution of Aedes aegypti (WHO).

Dengue disease is caused by four viruses (called Dengue 1, 2, 3, and 4), affects more than 100 countries in all continents except Europe (Figure 62 and Figure 63) and is spreading rapidly in many areas. It flourishes in conditions of poor housing, overcrowding, and inadequate sanitation, especially in cities of the tropics/subtropics of the Americas, Asia, Africa and the Western Pacific, in communities where there is a lack of piped water and an accumulation of rubbish, especially discarded containers and tyres where the vectors breed. In Southeast Asia, dengue ranks as one of the leading causes of hospitalisation and death among children.

In the past few decades there has been resurgence in dengue transmission over a wide area, with major outbreaks in Indochina, China, southeast Asia, and the Caribbean and northern

South America. In the Pacific region epidemics have occurred in many areas since 1971 (having been absent for some 25 years), and many travellers entering/returning to Australia from Southeast Asia and the Pacific have presented with the infection in recent years. Dengue Haemorrhagic Fever (DHF), the fatal form of the disease, in particular has increased (Figure 64).



Figure 63: Increase in distribution of dengue cases (WHO).

DENGUE - THE DISEASE

Dengue is usually regarded as a debilitating infection of comparatively short duration with a high attack rate but a low fatality rate. In fact, dengue is an acute febrile illness that may be seen in two clinical forms. The so-called 'classical' Dengue Fever (DF) form which usually affects older children and adults with fever, violent headache, severe pains in the muscles and joints, and often a rash, following an incubation period of 5-8 days, and lasts about 4-7 days; recovery is generally complete although convalescence may be long. The more severe form, Dengue Haemorrhagic Fever (DHF), involves bleeding internally and from the nose, mouth and gums, may be followed by severe shock (the Dengue Shock Syndrome (DSS)), and occurs most frequently in infants and young children; the initial fever phase is similar to that occurring in classical dengue but is followed by a sudden collapse which is frequently fatal. An estimated 500,000 cases of DHF (mostly children) require hospitalisation each year, and approximately 5% die although fatality rates can reach 20% without proper treatment. There is no cure and as yet no vaccine (although various types of vaccines are being developed).

The four dengue viruses are called serotypes, and are serologically related but immunologically distinct. This distinctness means there is no cross-immunity conferred between serotypes, and an 'enhancement' reaction following a second infection with a different serotype can lead to complications that can result in the haemorrhagic syndrome. Each serotype has been involved in uncomplicated dengue and in cases with haemorrhagic syndrome, but the actual cause of DHF is not completely understood.



Figure 64: Increase in numbers of dengue cases (WHO).

Local transmission of dengue in Australia results from the importation of the viruses (in humans) from regions with current activity, and we are often at great risk with the frequent activity that occurs in our neighbouring regions. In recent years, for instance, there has been extensive activity in Southeast Asia (with hundreds of thousands of cases) and the Pacific (with tens of thousands of cases), with major outbreaks in Vietnam, The Philippines, Indonesia and Singapore, and in Fiji, Cook Islands Tonga, New Caledonia and French Polynesia. The threat and actuality of dengue seemingly continues to increase each year.

DENGUE IN AUSTRALIA

Dengue was first recorded in Australia in the 1870s, in cases imported from Mauritius, and there were local outbreaks in Queensland in 1879, 1885, and in Qld and NSW in the 1890s; it was first reported from Western Australia in 1909, and was often active until the 1930s in the regions north of Carnarvon.

Numerous epidemics of dengue occurred in the late 19th and early 20th century. Past outbreaks were mostly confined to northern Australia, although outbreaks in Qld spread south into NSW in 1925 and 1942. Following 1955, when there was a large outbreak (15,000 cases) in Townsville, Qld, Australia was considered to be free of local dengue until 1981 when a major outbreak with an estimated 3,000 infections occurred in northern Queensland, presumably initiated by the introduction of virus with a traveller to a locality where the vector *Aedes aegypti* was abundant.

Cases of dengue infection are recorded in other parts of Australia, in returning travellers infected elsewhere. Dengue is, in fact, the most commonly diagnosed flavivirus infection in Australia with the great majority of cases being travellers returning from Southeast Asia and the southwest Pacific. However it is only in Queensland that the viruses can be transmitted by the mosquito *Aedes aegypti*, which no longer exists in other states (note, however, that it was

introduced twice in the past 5 years to the NT, to Tennant Creek and Groote Eylandt, but was subsequently eradicated from both locations).

When local dengue disease reappeared in Australia in north Qld with the outbreak of DEN virus type 1 in 1981-1982, it was after an absence of more than 25 years. The previous outbreak in 1955 in northern Queensland had been of DEN virus type 3. A decade later, during 1990-1991, DEN virus type 1 was again active in northern Queensland, and in 1992-93 a larger outbreak (> 1000 cases) of DEN type 2 occurred in Townsville and the surrounding areas of northern Queensland. Since these years there has been almost annual transmission of dengue in northern Queensland – always initiated from an infection acquired outside Australia.

This 'continuum' of outbreaks in due to an increase in dengue activity overseas, plus a high level of international travel into Cairns (approximately 600,000 arrivals/yr). For many years there had been only a few mild cases of dengue haemorrhagic fever in Australia, but in early 2004 there were two deaths from DHF and there has been one death in 2009. Further DHF cases could readily occur in Qld as in any dengue-endemic or dengue-receptive area of the world.

DENGUE AND ITS VECTORS

The normal cycle of dengue infection is considered to be human-mosquito-human. From feeding on an infected and viraemic human, the female mosquito is able to transmit the dengue virus after an incubation period of 8-10 days wherein virus infection, replication and dissemination result in infection of the salivary glands making the mosquito infective for life. In Australia, only *Ae. aegypti* is considered to be an effective vector of dengue. Dengue is normally associated with tropical or subtropical urban areas of the world where domestic or peri-domestic container breeding species of the *Aedes (Stegomyia)* subgenus, particularly *Ae. aegypti*, but also *Ae. albopictus* and other related species such as some of the *Ae. scutellaris* group of the Pacific region (e.g. *Ae. polynesiensis, Ae. scutellaris*), may act as vectors.

It is assumed that *Ae. aegypti* is the vector of greatest concern because of its close association with humans and their domestic habitat. *Ae. aegypti* is predominantly a day-biting mosquito whose larvae may be found almost exclusively in clean water in man-made containers such as water-barrels, rainwater tanks, wells, vases, tyres, bottles, tins, and most other water-holding containers found in the domestic environment, and also in some subterranean habitats (e.g. wells and telecommunication service pits) in urban areas. The adult prefers to rest and feed indoors, especially during the day. It also feeds almost exclusively on man, and its timid, 'flighty' feeding habits ensure that many feeds are needed to obtain a full bloodmeal. Thus one mosquito can infect many people/household and epidemics caused by *Ae. aegypti* are often 'explosive', with rapid increase in the number of cases.

There are past records of *Ae. aegypti* being found in WA, and extensively in the NT and Qld. The species was widely distributed in NSW, almost to the Victorian border. However, the distribution of *Ae. aegypti* in Australia has contracted remarkably since the 1940s; it has not been recorded from NSW since 1950 and disappeared from the NT sometime in late 1950s or early 1960s. The last collection in WA was 1970. Surveys undertaken during the 1980s in WA, NT and NSW failed to find the species, but it is known to be widely distributed in northern Qld, being particularly prevalent in northern coastal towns, and currently extends south as far as Gladstone on the coast and Chinchilla in the inland.

There is potential for the vector to become re-established in other states. The NT and northern WA are particularly vulnerable to introductions from southeast Asia, and the species has been imported (via ships and their cargo) on many occasions in recent years to the NT, but also to

Brisbane, Sydney, and to WA at Willie Creek near Broome. However, the dry and sparsely populated character of northern WA makes it very inhospitable for *Ae. aegypti* establishment, and it is very unlikely it would spread south to Perth.

There is a real risk that other exotic species known to be vectors of dengue, particularly *Aedes albopictus*, could be imported and become established in mainland Australia. *Ae. albopictus* is common in the Pacific and SE Asia, and numerous interceptions have been made at Australian ports, usually via ships carrying infested cargo, and it has recently become established in island of the Torres Strait. *Ae. albopictus*, being an outdoor mosquito that feeds on other animals as well as humans, is not as effective a vector of dengue as *Ae. aegypti* and is not associated with the explosive urban outbreaks such as those caused by *Ae. aegypti*. However, *Ae. albopictus* has the greater potential to become established in southern areas of Australia, because of its greater tolerance of colder climates and its ability to establish in peridomestic artificial and natural containers as well as domestic artificial containers. Thus, it poses a greater threat for more widespread transmission of dengue in this country, although the low human population and dry climate of WA would reduce the likelihood it would establish widely in WA.

DENGUE CONTROL IN N. QUEENSLAND

THE 1994 DENGUE FEVER MANAGEMENT PLAN FOR NORTH QUEENSLAND

Following the 1992-93 Townsville epidemic, Queensland Health developed a structured Dengue Fever Management Plan (DFMP) for North Queensland. The plan detailed integrated strategies, namely disease surveillance, vector control and health promotion, to prevent dengue outbreaks and ensure that the virus was eliminated, preferably before local transmission occurred. Underpinning the plan was the need for efficient laboratory surveillance for the early detection of cases, particularly imported ones, followed by rapid and thorough vector control. The ultimate success of the plan relied upon successful vector control through a collaborative effort between local government environmental health officers and Queensland Health personnel.

Ae. aegypti harbours in dark, sheltered areas such as under beds and tables, and inside closets. Selectively treating these sites with a residual pyrethroid insecticide, such as bifenthrin, deltamethrin or lambda-cyhalothrin, effectively reduced *Ae. aegypti* activity, and therefore dengue virus transmission, in foci of dengue activity during outbreaks.

Although the strategies in the 1994 DFMP had been satisfactory for nearly five years, a large and protracted epidemic of dengue 3 in Cairns, Port Douglas and Mossman in 1997-1999 proved very difficult to control. The epidemic demonstrated that dengue virus could spread very rapidly via movement of viraemic individuals to initiate multiple foci of disease that could not be adequately controlled using conventional methods. It also demonstrated the importance of 'ignition' premises, such as backpacker hostels catering to rapid turnover, high volume populations of travellers who could import the virus, and of 'dissemination' premises, e.g. schools, that could lead to rapid dispersal of virus through a community via students and staff. Cryptic breeding sites, including subterranean sites (e.g. sump pits) and elevated roof gutters, were an important source of *Ae. aegypti*. Not surprisingly, the virus evaded eradication, and eventually this led to 'burnout' and shortages of staff that severely hampered vector control operations.

In December 1998, Queensland Health established the Dengue Action Response Team (DART), a trained specialist team of three personnel. The sole responsibility of DART was to implement *Ae. aegypti* prevention and control strategies, including, where necessary, the interior spraying of premises. The DEN-3 epidemic ended within three months of the DART commencing activities.

ELEMENTS OF THE 2000-2005 DENGUE FEVER MANAGEMENT PLAN FOR NORTH QUEENSLAND

In 2000, a revised DFMP ("Dengue Fever Management Plan for North Queensland, 2000 – 2005") (DFMP 2000) was launched to incorporate new strategies, including those to be implemented by the DART. The objectives of the DFMP 2000 were (i) to recognise dengue cases as rapidly as possible through not only laboratory but also clinical surveillance, (ii) to respond to dengue cases, both imported and locally-acquired, with thorough and sustained vector control aimed at eliminating the dengue virus before it can spread to other urban foci, and (iii) to use a variety of education initiatives to maintain community awareness of dengue, its mode of transmission, and the need for individuals to take action to prevent *Ae. aegypti* from breeding in households and business premises.

Disease surveillance

Between December 1994 and May 2002, there were 69 notifications of imported dengue cases where the traveller was viraemic (and therefore infectious to *Ae. aegypti*) whilst in north Queensland. All four serotypes were imported during this time. The numerous outbreaks in the 1990s reflected the necessity to identify imported viraemic cases as quickly as possible. Dengue is a notifiable disease in Australia, and as such, the Tropical Population Health Unit (TPHU) must be notified of a case by laboratories or physicians. Because many imported viraemic cases are not promptly notified, local *Ae. aegypti* may become infected and on the wing before control measures can be initiated. Furthermore, several of the dengue outbreaks had no known origin, indicating that many imported dengue cases probably went unrecognised.

Although serological false positive results remain a problem, the timeliness of laboratory testing for dengue improved. Serological tests have been augmented by rapid immunochromatographic card or dipstick tests, while the polymerase chain reaction test has supplanted virus culture to detect dengue virus in serum, giving quicker results yet still providing valuable genetic sequencing information. Finally, careful patient interviews by public health nurses are needed to establish the patient's history, particularly whether a case is imported or locally acquired, and points of contact where the patient may have infected mosquitoes.

Vector control

Upon notification of either a dengue IgM +ve test result or a suspected imported case, the DART makes an immediate response. The points of contact (usually the case's residence and place of work) are mapped, and the intended vector control activities detailed. The DART conducts interior spraving and larval control/source reduction activities within 100 metres of a contact point, while local government personnel conduct larval control in a zone 100-200 m from these premises. Interior spraying involves application of a residual synthetic pyrethroid (e.g., bifenthrin, deltamethrin, lambda-cyhalothrin) to dark resting places inside a premise (under beds and tables; wardrobes, laundry areas) using a pump sprayer. Larval control includes removal of small containers while larger containers are treated with the growth regulator s-methoprene pellets or sprayed with aerosol surface sprays containing a residual pyrethroid insecticide (deltamethrin, permethrin, cypermethrin, etc.). Particular care is placed upon identification and treatment of cryptic subterranean and elevated breeding sites, such as sump pits and roof gutters, respectively. If multiple dengue cases are reported in a particular area, the response zone is expanded. Field data are recorded onto a palm-top computer, then imported onto a Geographical Information System (GIS) for mapping response activities, the latter particularly useful for follow-up treatments. Between outbreaks, the DART conducts preventive larval control activities at ignition and dissemination premises, such as backpacker accommodation and schools, respectively.

Health promotion

Health promotion played two critical roles in the DFMP 2000. During an outbreak, publicity in the form of media advisories, delivered by local media, was used to inform the public about areas with active transmission. Residents were advised to take precautions by clearing households and backyards of obvious breeding sites, using surface sprays to kill adult *Ae. aegypti* and taking personal protective measures to avoid bites. Secondly, a television-based mass media campaign was run on a paid schedule during the wet season to educate the public. The domestic habits of the vector and how to control it were discussed, along with symptoms and consequences of the disease. Programs to educate school children were also developed.

SUCCESS OF THE DFMP 2000

Since the implementation of the DFMP 2000, five outbreaks of dengue that occurred in north Queensland were relatively easily contained, and very little locally-acquired dengue was detected beyond the initial focus of activity (Tables 1-2). Furthermore, the number of cases acquired after initiation of control efforts was limited and the duration of outbreaks reduced. The latter was critical in helping to maintain a focussed effort by dengue control staff; burnout during the 70 week dengue 3 epidemic in 1997-99 contributed to the duration and spread of the outbreak. We considered that early case recognition, coupled with thorough interior spraying of premises and intense larval control efforts, were instrumental in the success of the plan to date. The utility of the current DFMP was also evident in early 2000 when approximately 2000 Australian Defence Force personnel and aid workers returned from duties in East Timor to north Queensland. Despite at least eight viraemic importations of dengue in these personnel, focussed and collaborative responses ensured that not one of the cases led to local transmission.

DENGUE: RE-EMERGENCE AS A SEVERE DISEASE IN 2003.

In March 2003, a large epidemic of Den-2 began in Parramatta Park, an older suburb in Cairns. This outbreak eventually spread throughout much of Cairns, and to Townsville, where 459 and 20 cases were recorded from Jan – July, 2003, respectively. The primary reason why the outbreak became so large was the late recognition/notification of the index case; 42 days elapsed from the time the viraemic patient arrived in Parramatta Park until the TPHU was notified of dengue activity in the area. While this outbreak was officially eradicated from Cairns in July 2003, it persisted in Townsville as a slow, often cryptic, outbreak totalling 58 cases up to July 2005. The outbreak was notable for a relatively mild disease, with many cryptic cases and few (ca. 5%) hospitalisations.

This was in stark contrast to the dengue outbreak that started in Yam Island, in the Torres Strait, in October 2003. The initial two cases notified to the TPHU had severe dengue, indeed categorically DHF, with liver failure. Both women were in intensive care for ca. 1 month, and both nearly died. Serology indicated both were secondary cases, with evidence of a previous dengue infection. Numerous other cases of 'fevers' were then reported from Yam Island Health staff. The DART responded, using interior spraying and larval control to eliminate dengue within 3 weeks. However, the outbreak spread to Thursday Island (TI), where a large amount of rubbish in yards contributed to high *Ae. aegypti* populations. While the epidemic on TI did not result in explosive transmission, a steady influx of ca. 10 cases/week kept DART and Torres health workers busy. The outbreak brought the first reported deaths due to locally acquired DHF/DSS for almost 100 years in Australia.

Control efforts were complicated by the emergence of dengue activity in both Cairns and Townsville. Indeed, during February – March, large outbreaks of dengue were simultaneously occurring in Thursday Island (TI), Cairns and Townsville. Thus, the 3-person DART team was severely under-resourced to deal with the outbreak. The escalation of dengue activity in TI, along with the DSS case, necessitated a large intervention to eradicate the virus on TI. Up to 20 workers, consisting mainly of Environmental Health Officers from throughout Queensland,

conducted emergency vector control on TI from 2-12 March. Despite vector control efforts since November 2003, large numbers of breeding sites were still found; indeed, 47 and 57 breeding sites were found in two different respective premises! Many of the containers were large, such as car bodies, disabled rainwater tanks and tyres, and thus will require major works to remove. For short-term immediate control, these were treated with methoprene pellets or Bistar (bifenthrin) (n = 544 premises). Dengue case residences were sprayed inside with lambda-cyhalothrin (n = 53). Finally, lethal ovitraps (n = 780) containing a red velour strip treated with bifenthrin were set in yards. Adult *Ae. aegypti* were monitored with sticky ovitraps (n = 60). Within three weeks, female *Ae. aegypti* numbers had fallen by 91%. After 289 dengue cases in the Torres Strait, the last dengue case occurred on TI on 1 April.

Lethal ovitraps (LO) were also successfully employed in Cairns and Townsville. While not all interventions were successful, in 2003 and 2004, respectively, 12/14 and 4/8 interventions involving LO had no further cases after control. It should be noted that all of these interventions involved larval control, while many had restricted interior spraying near case premises. Thus, the results, while encouraging, do not definitively validate the method.

RESPONSE TO DENGUE ESCALATION: AN UPDATED DFMP 2005

In response to the epidemics of dengue in 2003-04, Qld Health revised the DFMP in 2005. The new DFMP does not rely upon early case notification, as the late notifications are impossible to omit. Late notifications (> 30 days from arrival of index case onset until notification of TPHU) of dengue had led to both the Cairns epidemic in 2003 and the Torres outbreak in 2003-04. Thus the new DFMP emphasised sustained, preventative vector control to reduce *Ae. aegypti* populations in high-risk areas. Hopefully, the explosive epidemics such as the 2003 Parramatta Park outbreak can be prevented despite late notifications. Other areas emphasised in the new DFMP include: routine insecticide resistance testing, sticky ovitrap surveillance to target control actions, a regional Torres/Cape York dengue program with a DART position, organised rubbish clean-up in the Torres, trials to validate lethal ovitrapping, a new health education campaign in Northern Qld and the Torres Strait, and an updated palmtop computer - GIS database. There was also an addition of 3 new DART positions to bolster the capability of the new DFMP.

Since 2005, there have been relatively minor annual outbreaks that have had to be dealt with in northern Qld. However, at the end of 2008, a new outbreak that lasted until June 2009 and finally totalled more than 1000 cases (predominantly in/around Cairns), proved to be a major problem to manage with respect to vector control and required the recruitment of a substantial temporary workforce. Qld Health is currently reassessing their capability to respond to such outbreaks, with a view to upgrading aspects of the DFMP.

26. EXOTIC AND EMERGING MOSQUITO-BORNE DISEASES: AN AUSTRALIAN PERSPECTIVE

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INTRODUCTION

Emerging or re-emerging diseases are those that are either newly recognised diseases or are increasing in prevalence after a period of decline or previous elimination. Emerging and reemerging diseases are a world-wide phenomenon that may be exacerbated by factors such as environmental changes (eg., deforestation, irrigation) and increasing travel and/or transportation between countries. Examples of newly emerging or re-emerging diseases include severe acute respiratory syndrome (SARS), avian influenza, Ebola, HIV, Nipah virus and Hendra virus. Vector-borne diseases are no exception. In Australia alone, Japanese encephalitis virus (JEV) emerged in 1995, and Barmah Forest virus emerged as a disease entity in Western Australia in 1992. Furthermore, a number of exotic vector-borne diseases are of concern to Australia due to their potential for introduction and possible establishment in local transmission cycles. Examples include Rift Valley fever in Africa, chikungunya virus in southeast Asia, West Nile virus (WNV) in North America, Europe, Africa and southeast Asia, and malaria in tropical and subtropical areas. Movement of insect vectors is another concern, as the introduction and establishment of foreign mosquito species in Australia may have implications for transmission of exotic and indigenous mosquito-borne diseases. Examples of exotic mosquito species detected in Australia include the dengue, yellow fever and chikungunya virus mosquito vector Aedes albopictus, as well as Ae. vexans and Culex gelidus, which are likely to be involved in JEV transmission cycles. This chapter will focus on the risk of introduction and possible establishment of two medically important flaviviruses (JEV and WNV), one alphavirus (chikungunya virus) and the parasitic disease malaria.



Figure 65: Torres Strait islands and Cape York Peninsula.

JAPANESE ENCEPHALITIS VIRUS (JEV)

THE EMERGENCE OF JAPANESE ENCEPHALITIS VIRUS IN AUSTRALIA

JEV is closely related to the Australian flaviviruses Murray Valley encephalitis virus (MVEV) and Kunjin virus (KUNV). JEV first occurred in Australia in 1995, with cases of encephalitis in people living on Badu Island, in the Torres Strait (Figure 65). Three cases of Japanese encephalitis occurred in residents on Badu Is. in March and April 1995, two of which were fatal. Surveys of residents on islands in the Torres Strait and communities on the Cape York Peninsula revealed that people on Badu Island and outer islands had been infected with JEV, however people on inner islands and Cape York had not been infected. Isolates of JEV were obtained from people and from Cx. annulirostris mosquitoes collected on Badu Is. during the outbreak, providing circumstantial evidence that Cx. annulirostris was the likely vector. As a result of this outbreak, vaccination of people living in communities in the Torres Strait was carried out. In addition, a surveillance programme was established involving the regular bleeding of sentinel pigs located at strategic positions in the Torres Strait and Cape York Peninsula. Monitoring of sentinel pigs has shown that JEV was active in the Torres Strait in all years but one (1999) since 1995, and human cases were reported in the Torres Strait and mainland Australia (western Cape York Peninsula) in 1998. A wider investigation revealed that sentinel pigs on the northern peninsula and domestic pigs near the mouth of the Mitchell River had been infected with JEV. Sentinel pigs were removed from the Torres Strait at the end of 2005, and it is not known if JEV activity has occurred in the region since this time.

THE NATURAL HISTORY OF JAPANESE ENCEPHALITIS VIRUS

JEV has been implicated in outbreaks of encephalitis in many countries in southern and southeastern Asia, causing over 45,000 human cases each year, mostly in children. Consequently, JEV is the most important of the arboviruses causing encephalitis, in terms of clinical disease and fatality rate. JEV primarily exists in a transmission cycle between pigs, birds and *Culex* mosquitoes, particularly *Cx. tritaeniorhynchus*.

CLINICAL PICTURE

As with many arboviruses, most infections with JEV do not result in clinical disease. More than 90% are subclinical infections, with affected people not knowing that they have been infected. The ratio of clinical to subclinical infections ranges between 1:50 and 1:300. Clinical symptoms of infection with JE can range from a mild febrile illness, aseptic meningitis, through to acute encephalitis. Approximately 25% of cases are fatal, and about 50% of sufferers are left with permanent neurologic and/or psychiatric damage. Only about one quarter of cases fully recover. JE has an incubation period of between 6 and 16 days. The early stage (lasting 1 to 4 days) is characterised by an abrupt onset of high fever with headache, anorexia, nausea, vomiting, dizziness and drowsiness. Children may also suffer from abdominal pain and diarrhoea. The acute encephalitis stage (3 to 4 days duration) is characterised by an altered level of consciousness (ranging from confusion to delirium, disorientation, stupor, semi-coma or coma), sore eyes, convulsions, weakness of extremities, neck stiffness, tremors, incoordination, and paralysis. In uncomplicated cases, symptoms recede over 1 to 2 months, and neurological symptoms improve. However, severe cases may not regain consciousness, and death may occur within 5 to 9 days, or over an extended period associated with cardiopulmonary complications. Infection with JEV has also been associated with abortions in pregnant women in the first or second trimester of gestation.

DIAGNOSIS

Laboratory diagnosis of JE infections depends on detection of the virus or antibodies to the virus. Diagnosis usually relies on the detection of a four-fold or greater rise in antibody titre in acute phase (early) and convalescent (late) serum specimens. However, cross-reactions with closely related viruses may complicate diagnosis. New assays have been developed which increase the speed, specificity and sensitivity of diagnosis. As an 'arboviral encephalitis', JE is a notifiable disease in WA.

GEOGRAPHIC DISTRIBUTION

JEV is widely distributed in Asia and southeast Asia (Figure 66). Occasional outbreaks have also been recorded on Micronesian islands including Guam and Saipan. JEV is now known to be present in Papua New Guinea, the Torres Strait islands and rarely, on Cape York Peninsula.

VECTORS

Isolations of JEV have been acquired from more than 35 species of mosquitoes. Australian species found to transmit JEV in laboratory conditions include *Cx. annulirostris*, *Cx. sitiens*, *Cx. bitaeniorhynchus*, *Cx. quinquefasciatus*, *Cx. gelidus*, *Mansonia uniformis* and *Ae. vigilax*.

The most significant species in terms of outbreaks of disease in Australia are most likely to be Cx. annulirostris and Cx. sitiens. These species are very efficient transmitters of the virus in the laboratory, are widely distributed and are often abundant, particularly in northern Australia. However there is some evidence for variation in vector competence of different populations of Cx. annulirostris with genetically different strains of JEV.



Figure 66: Worldwide distribution of JEV [adapted from CDC website (www.cdc.gov/ncidod/ dvbid/jencephalitis/map.htm)

VERTEBRATE HOSTS

Natural infections with JEV have been documented in many domestic and wild animal species including birds (particularly ardeid water birds, including various herons and egrets), pigs, cattle, buffalo, dogs, horses, goats, sheep and insectivorous and fruit bats. The animals most important in terms of JEV transmission are probably pigs, some birds (particularly ardeid water birds) and bats. Animals that do not develop high levels of viraemia and therefore are unlikely to have a major role in JEV transmission include cattle and buffalo, dogs, horses and humans. The role of marsupials in JEV transmission is yet to be clarified. Studies at the Australian Animal Health Laboratory showed that Eastern grey kangaroos, Tammar wallabies and Agile wallabies failed to develop or developed low levels of viraemia during experimental infections with JEV. Possums developed higher levels of viraemia that may be sufficient to infect mosquitoes. However, the level of viraemia needed to infect a vector mosquito was not determined.

HOW DID JEV ENTER AUSTRALIA?

It is most likely that the recent incursions of JEV into Australia probably originated from PNG. Analysis of isolates of JEV from PNG and the Torres Strait showed that the isolates were genetically identical. Given that the southern coastline of PNG lies very close to the northern islands of the Torres Strait (less than five km), it is possible that viraemic vertebrate hosts such as birds, bats or humans, or windblown mosquitoes, may have introduced JEV into Australia.

COULD JE VIRUS BECOME ESTABLISHED IN AUSTRALIA?

Torres Strait Islands

Repeated JEV activity in islands of the Torres Strait, such as Badu Is., suggests that the virus is often re-introduced, or that the virus has become established. Given the small size of most islands in the Torres Strait, with limited susceptible vertebrate hosts, the establishment of JEV in permanent transmission cycles seems unlikely. However, the isolation of JEV from *Ae. vigilax*, which has desiccation-resistant eggs, is cause for concern. The potential exists for vertical transmission to occur via desiccation-resistant eggs of certain species of mosquitoes, in a similar way that MVEV is thought to be maintained during periods not conducive to vector mosquito breeding. Vertical transmission is a possible mechanism by which JEV may become established for long periods on a small island, particularly when environmental conditions are suitable for hatching of desiccation-resistant eggs annually.

Cape York Peninsula

The potential for establishment of JEV on mainland Australia remains uncertain. Whilst activity of JEV continues to be detected in the Torres Strait, potential remains for re-introductions of JEV into northern QLD, given favourable environmental conditions. Breeding colonies of potential vertebrate hosts (eq., egrets and herons) on western Cape York Peninsula, large populations of feral pigs (particularly along river systems and floodwaters) and often large populations of confirmed vector species of mosquitoes are cause for concern. To date, studies in northern QLD have failed to find any evidence of establishment of JEV, although field trips were limited to once or twice per year, during periods when vector mosquitoes were likely to be most abundant and flaviviruses most active. However, other flaviviruses (including MVEV and KUNV) were isolated from mosquitoes from Cape York. The effect of multiple flaviviruses circulating in these areas on the potential establishment of JEV is uncertain. Studies in PNG have shown that flaviviruses including JEV and MVEV can co-exist in a locality, suggesting that the presence of one flavivirus does not prohibit another. However, sequential infections in pigs with different flaviviruses showed that prior exposure to MVEV or KUNV prevented JEV from replicating in the animals after subsequent exposure to JEV. It is possible that high levels of other flavivirus activity in the area may not inhibit the introduction and low-level activity of JEV, but may inhibit large outbreaks of disease.

Northern Western Australia and the NT

If JEV does become established in northern QLD, the possibility exists for JEV to progressively spread from one area to another in transmission cycles between susceptible vertebrate hosts (eg., migratory water birds, feral pigs or possibly marsupials) and mosquito vectors. Indeed, movement of MVEV from endemic regions of the Kimberley and NT is thought to occur in such ways.

A number of potential avian hosts of JEV are common or moderately common in swamps, mangroves or rainforest and woodland areas across northern Australia, including numerous species of waterbirds, and breeding colonies of some of these species are found in the NT. Whilst feral pigs are not considered abundant in northern WA, they are an ongoing problem in the NT. It is also possible that some species of bats could potentially be involved in virus movement and maintenance in northern Australia. So too could certain species of marsupials, such as possums, wallabies and kangaroos. Further laboratory studies are required to confirm the role of these species in JEV transmission.

Some mosquito vectors are often abundant and widely distributed in northern Australia. Species occurring in Australia that have been confirmed or are likely vectors for JEV include *Cx. annulirostris, Cx. sitiens, Cx. quinquefasciatus* and *Ma. uniformis.* All of these species can reach plague proportions, particularly in northern Australia, during times when flaviviruses are most active. Furthermore, the apparent spread and increasing abundance of *Cx. gelidus* across northern Australia is cause for concern, given its confirmed status as a vector in other countries and the isolation of JEV from this species on Badu Is. in 2000. Many of these

species will readily feed on animals such as humans, pigs, cattle, horses, marsupials and birds, increasing the potential for involvement in these mosquito species and vertebrates in JEV transmission cycles. Other potential vectors include *Cx. bitaeniorhynchus, Cx. whitmorei* and *Ae. lineatopennis,* however these are not common species and are therefore less likely to have a major role in JEV transmission in Australia.

The close proximity between the coastline of the Kimberley region and NT to the Indonesian archipelago is also cause for concern, particularly given the regularity with which monsoonal low pressure systems and cyclones occur in the Timor Sea, potentially carrying infected mosquitoes into these northern regions of Australia. For these reasons, active surveillance of JEV and mosquito vectors in Australia, including northern WA, the NT and northern QLD, is essential. If JEV is introduced into the NT and northern WA, the presence of indigenous flaviviruses could potentially prevent establishment or large outbreaks of JE due to cross-protective immunity, as hypothesised in the preceding section.

PREVENTION AND CONTROL

Prevention of outbreaks of JE generally relies on surveillance of JEV activity in vector mosquitoes, vertebrate hosts or humans, vector control and implementation of vaccination programmes. Other non-specific methods to reduce the risk of humans being bitten by vector mosquitoes are discussed below.

Vector control

Following the outbreak of JE in the Torres Strait islands in 1995, larvicides and adulticides were used to eliminate vector mosquitoes, and residual surface sprays (such as deltamethrin, lambdacyhalothrin and permethrin) were applied to the inside surfaces of pig pens to kill mosquitoes that had taken bloodmeals from infected pigs. However, the effectiveness of spraying pig pens with residual surface sprays was not determined (Dr. Scott Ritchie, QLD Health, Cairns Tropical Public Health Unit, personal communication).

Vaccination

JEV vaccines are principally used throughout Asia to protect the at-risk populations, however vaccination is also recommended for travellers to high-risk regions. A number of risk factors for travellers have been identified. However, because of some adverse side effects, the cost of vaccination, and relatively low risk to travellers, vaccination is only recommended for visitors to epidemic or endemic areas during the transmission season, particularly if staying for more than 30 days, or if outdoor activities expose the traveller to vector mosquitoes. The only vaccine currently licensed for use in Australia is the formalin-inactivated mouse brain Biken® Nakayama or Beijing-1 (or both) vaccine. Following the emergence of JEV in northern Australia, this vaccine was incorporated into the childhood vaccination regime in the risk area. Some studies suggest prior exposure to closely related flaviviruses such as JEV, MVEV or KUNV viruses may enhance disease following subsequent exposure to a different flavivirus. This may have implications for vaccination of people in parts of Australia where other flaviviruses are known to occur.

Other methods for prevention

Alternative methods of prevention are primarily aimed at mosquito avoidance. The use of mosquito nets while sleeping at night (when vector mosquitoes are more likely to take blood meals) can be effective at reducing human cases of JEV. Bed nets treated with synthetic pyrethroids are much better at reducing human cases than are untreated bed nets. Other methods include screening houses against mosquitoes, wearing protective clothing, and the use of insect repellents containing N, N-diethyl-meta-toluamide (DEET) or picaridin. Changing animal husbandry practices, such as the movement of domestic pigs on Badu Is. to a communal piggery several kilometres from the community, can also reduce or eliminate JEV transmission.

Surveillance

After JEV emerged in the Torres Strait and Cape York Peninsula, sentinel pig herds were introduced pigs in the Torres Strait, Cape York Peninsula and NT (Jonathan Lee, AQIS, personal communication), and sentinel cattle in northern WA, for detection of seroconversions to JEV. However the cost associated with establishing and monitoring a herd of five pigs in remote areas was high. Furthermore, pigs are amplifying hosts of JEV, and contribute to JEV activity in an area, thereby posing a risk to nearby residents. In addition, multiple flaviviruses circulate in some parts of northern Australia, such that even young immunologically naïve pigs can become infected with more than one flavivirus in a season, complicating serological diagnosis of infection (Jonathan Lee, AQIS, personal communication). Thus, sentinel herds of pigs are no longer used for JEV detection in Australia. Trials are currently being conducted in the Torres Strait and Cape York Peninsula using alternative mosquito-based surveillance methods that rely on molecular tools for detection of JEV.

WEST NILE VIRUS (WNV)

WNV is another flavivirus that is related to MVEV and JEV. It is widely distributed throughout Africa, the Middle East, southern Europe, Russia, southern India and parts of southeast Asia. WNV has rapidly spread through the United States since its emergence there in 1999 (Figure 67). WNV activity has now been detected in almost every state in the United States as well as Canada, Mexico, Puerto Rico, and as far south as Argentina in South America. It is not known how WNV entered the United States, although genetic studies suggest the virus came from the Middle East, possibly via an infected mosquito on an aircraft or an infected traveller with sufficient levels of virus in the blood to infect a mosquito after arrival in the United States. The Australian virus KUNV is now recognised as a subtype of WNV.



Figure 67: Incidence of WNV neuroinvasive disease in the United States in 2010

THE NATURAL HISTORY OF WEST NILE VIRUS

Transmission cycle

WNV is primarily transmitted between mosquito vectors and birds. *Culex* mosquitoes, particularly those that feed on birds, have a major role in the transmission of WNV. WNV has also been isolated from *Aedes* and *Mansonia* species, suggesting possible involvement of these species in WNV transmission. In addition, isolations of WNV from ticks and the laboratory-confirmed vector competence of some species of ticks for WNV indicate ticks may have a role in transmission, and may be important in dispersal and over-wintering of WNV. Although transmission by mosquito bite is the typical mode by which people become infected with WNV, unusual modes of transmission including by blood transfusion, organ donation and breastfeeding have been reported. Numerous species of birds, particularly crows and pigeons, develop high levels of virus in the blood, providing an infectious bloodmeal for competent

mosquito vectors. Birds probably have an important role in movement of WNV from one area to another. In North America and Israel, outbreaks of WNV have been associated with high levels of disease and mortality in several species of birds. Humans and horses suffer from clinical disease, however they are usually considered 'dead-end' hosts of WNV.

Clinical features

In most instances, WNV causes an unapparent ('sub-clinical') infection, or an acute denguelike fever followed by recovery. However, some patients develop meningo-encephalitis. Encephalitis is most common in adults rather than children, and disease is most severe in the elderly. The mortality rate from encephalitis is 15-40%.

The incubation period is 1-6 days. Historically, the disease has been typically mild, with fever, headache, muscle aches, backache and weight loss. Sore lymph nodes, a rash and nausea are also common features. When progression to encephalitis occurs, the patient develops severe headache, confusion, altered consciousness, neck stiffness, cranial nerve palsies and general weakness. Most survivors of encephalitis have substantial residual neurological damage.

Diagnosis

Infection with WNV is similar to that described for JEV. Detection of rising antibody titres to WNV in sequential blood samples is diagnostic, although molecular methods are also used. Detection of IgM in cerebrospinal fluid is a good indicator of encephalitis. Serological tests for other flaviviruses are recommended to exclude the possibility of misdiagnosis due to serological cross-reactions. This is particularly important in countries like Australia, where a number of closely related flaviviruses exist.

RISK OF INTRODUCTION AND ESTABLISHMENT OF WEST NILE VIRUS IN AUSTRALIA

The potential for WNV to enter Australia is a cause for concern. Possible modes of entry include similar mechanisms hypothesised for the incursion of WNV into the United States, ie., via an infected mosquito on an aircraft. Disinsection procedures for aircraft arriving in Australia should minimise the chance that an infected mosquito will arrive in Australia (although ineffective aircraft disinsection has been suggested as a means by which malaria may have entered northern Queensland). Another potential mechanism of introduction is via viraemic vertebrate hosts. Indeed, a horse infected with WNV was imported into Australia from North America in 2002. Although the level of virus in the blood of horses and humans is usually considered insufficient to infect a bloodfeeding mosquito, further investigation into viraemia levels during infection is required to adequately assess the risk of transmission of virus to mosquitoes from these hosts. Another possible method of entry for WNV into Australia is related to the potential for the gradual spread of WNV from southeast Asia, where it has been isolated from mosquitoes (in Malaysia), possibly leading to the eventual introduction of WNV into Australia in similar ways that JEV is likely to have been introduced. The introduction of WNV through migratory birds is considered unlikely because of the route and length of migration.

Evidence in some countries (including Australia) has showed that closely related flaviviruses can co-exist, and that they do not interfere with their ability to spread, despite the fact that the antibodies produced to these viruses cross-react. Nevertheless, should WNV be introduced into Australia, the outcome is not certain. Given the widespread distribution of KUNV in Australia and the very close relationship between KUNV and WNV, it seems likely that Australian species of mosquito and birds would be capable of becoming infected with and amplifying WNV. Recent studies have shown that several Australian species of mosquito are able to become infected with and transmit WNV, including *Cx. annulirostris, Cx. quinquefasciatus* and *Cx. gelidus*. However, in Australia WNV will have to contend with KUNV for vertebrate hosts and vectors. If vertebrate hosts have previously been infected with KUNV,

it seems likely that KUNV-specific antibodies will protect against future infection with WNV. The level of pre-existing immunity in vertebrate hosts may limit the number of susceptible vertebrate hosts, thereby impeding the establishment and spread of WNV in Australia.

PREVENTION, DETECTION AND CONTROL

The methods for prevention and control of WNV in Australia are similar to those for JEV, with the exception that no human vaccine currently exists. Given that WNV does not currently exist in Australia, the primary aim is to prevent the introduction of WNV in mosquitoes or vertebrate hosts. Continuation of aircraft disinsection procedures is essential to prevent infected mosquitoes from arriving in Australia. Unfortunately, sentinel chickens cannot be used for surveillance of WNV in Australia as antibodies produced in hosts following infection with WNV cross-react with KUNV (ie., the infection would look like a KUNV infection). Until improved serological methods are available, surveillance currently relies on detection of virus in mosquitoes using a panel of virus-specific reagents or molecular methods.

CHIKUNGUNYA

Chikungunya (CHIKV) is an alphavirus related to Ross River and Barmah Forest viruses. It was first isolated from patients in Tanzania during an outbreak in 1952-1953. The name 'chikungunya' means 'that which contorts or bends up'. CHIKV has a widespread distribution in Africa, Saudi Arabia, India and southeast Asia. Large outbreaks have occurred in urban environments in the past. Between 2005 and 2007, CHIKV caused an explosive outbreak on islands of the southwest Indian Ocean, spreading to India and causing imported cases of disease in Europe, North and South America, the Caribbean and Australia. In 2007 there was a localised outbreak of chikungunya in north-eastern Italy.

THE NATURAL HISTORY OF CHIKUNGUNYA VIRUS

Transmission cycle

In urban environments CHIKV is primarily transmitted to humans by *Ae. aegypti* and *Ae. albopictus*. Recent laboratory studies in Australian mosquitoes showed that a number of species may be capable of transmitting CHIKV including *Ae. vigilax, Ae. procax* and *Coquillettidia linealis*. Humans are amplification hosts of CHIKV, and other vertebrates are not required for high levels of transmission to occur. In the recent outbreak in the Indian Ocean there was some evidence for non-vector early maternal-foetal transmission.

Clinical features

The incubation period for CHIKV ranges from 3-12 days. Illness typically begins with rapid onset of fever, arthralgia, headache, back pain, muscle pain, fever, sore lymph nodes and conjunctivitis. About 50% of patients develop a rash. People usually recover after a few days or weeks. Severe haemorrhagic symptoms can occur, sometimes leading to fatal illness. Severe disease is more common in children, the elderly and people with chronic conditions whose immune systems are compromised. A small percentage of sufferers can experience prolonged joint pain, stiffness and swelling. CHIKV infection has been associated with encephalitis on rare occasions.

Diagnosis

Diagnosis of CHIKV infection is similar to diagnosis of JEV and WNV. Virus can be detected in serum during the early days of illness. Rising antibody titres can be detected in paired sera. There may be some cross-reactions with other closely related alphaviruses. CHIKV is a notifiable disease in most parts of Australia.

THE RISK OF INTRODUCTION OF CHIKV INTO AUSTRALIA

The demonstration that a number of common Australian mosquitoes are vector competent for CHIKV has important implications for the receptivity of Australia to CHIKV should it be introduced. This is because the area of Australia that is receptive to CHIKV introduction becomes far greater than northern Queensland and the Torres Strait Islands, where *Ae. aegypti* and/or *Ae. albopictus* occur. In 2010/11 alone, 63 cases of CHIKV infection were notified across Australia, however all of the affected people acquired the infection overseas. Nevertheless, the possibility remains that local transmission of CHIKV could occur should infected travellers return to parts of Australia where local mosquito species that are capable of transmitting the virus are abundant.

MALARIA

Malaria is a parasitic disease transmitted by *Anopheles* species of mosquitoes. Five species of the parasite *Plasmodium* cause malaria: *P. ovale*, *P. malariae*, *P. vivax*, *P. falciparum* and *P. knowlesi*. *P. vivax* and *P. falciparum* are the most common. Infection with *P. falciparum* can be life threatening, causing multiple organ damage, coma and death.

TRANSMISSION CYCLE

Malaria is transmitted by female *Anopheles* mosquitoes. The parasite enters humans during blood-feeding of an infected mosquito. Initially the parasite infects and multiplies in the liver (

Figure 68). After several days, the parasites are released into the blood, infecting red blood cells, where they also multiply, eventually bursting the red blood cells and infecting more red blood cells. Severe disease or death may result if the parasite load becomes very high. Some of the parasites develop into sexual stages (gametocytes) in red blood cells. It is when the gametocytes are ingested by a mosquito during a bloodmeal that the infection may spread to the mosquito, thereby maintaining the transmission cycle.





(source: CDC website).

GEOGRAPHIC DISTRIBUTION

Malaria is found in tropical and subtropical countries of the world (Figure 69). High risk areas include rural areas in South America, southeast Asia (eg., Thailand, Indonesia, East Timor), the western Pacific (eg., Papua New Guinea, Solomon Islands, Vanuatu) and sub-Saharan Africa. These warmer countries are where the *Anopheles* mosquitoes breed in abundance and the parasite is able to replicate in the mosquito. It was estimated by the World Health Organization that 219 million clinical episodes of malaria occurred in 2010 and 660 000 deaths occurred in 2010. Most of those deaths (86%) were children.



Figure 69: Worldwide distribution of malaria

(source: CDC website).

SYMPTOMS

Symptoms appear after approximately 9-14 days following a bite from an infected mosquito, coinciding with the rupture of red blood cells. Typical symptoms include fever, shakes, sweating, headache, vomiting, muscle aches and other flu-like symptoms. Symptoms may recede for 2-3 days before relapses. If left untreated the infection can rapidly progress and become life-threatening, due to severe anaemia (ie., destruction of red blood cells) and/or damage to organs including the brain (ie., cerebral malaria), lungs or kidneys. Severe malaria most often occurs in people with no immunity or whose immunity is lowered, such as children and pregnant women.

P. vivax and *P. ovale* have dormant liver stages that can remain silent for years before causing disease. If untreated, the liver stages may reactivate and cause relapses after years without symptoms. Similarly, *P. malariae* has been known to persist for years in low levels in the blood if left untreated. *P. falciparum* can recur after apparent recovery due to either inadequate treatment of infection with a drug resistant strain.

DIAGNOSIS

Malaria is diagnosed through examination of a blood smear from a patient, using a microscope to detect parasites inside red blood cells. Alternative methods for detection include serological and molecular techniques. In *P. falciparum* malaria, additional laboratory findings may include anaemia, a decrease in the number of blood platelets, an elevation in the concentration of certain enzymes and the presence of abnormal bodies in the urine. Malaria is a notifiable disease in Australia. Travellers who have returned to Australia from an endemic region up to 2 years previously and has a history of fever should be tested, irrespective of whether antimalarial prophylaxis was taken. Furthermore, negative blood smears do not necessarily exclude malaria, particularly if antimalarials have been taken, and blood tests should be repeated, particularly if fever persists.

PREVENTION

Prevention of malaria in endemic areas is aimed at:

- 1. Avoiding mosquito bites by conducting vector control, using personal protective measures such as insecticide-treated bed nets and repellents (containing DEET or picaridin), avoid being outdoors between dusk and dawn when vector mosquitoes are more active, and wearing long, loose-fitting light coloured clothes; and
- 2. Preventing disease by using antimalarial drugs. The drugs do not prevent infection with the parasite, however they suppress the development of malaria parasites in the blood.

Travellers from non-endemic countries should take precautions against being infected with malaria when they visit malaria risk areas. Preventative medication is often needed that should be commenced 1 week prior to entering the affected country, so contact with a general practitioner or travel clinic is recommended in advance to organise antimalarial medication. Different antimalarial drugs are recommended for different countries depending on the strains of malaria present and the level of drug resistance.

MALARIA IN AUSTRALIA

Australia was declared free of malaria by the World Health Organisation in 1981, however the number of cases of malaria imported to Australia from endemic countries each year is high. On average 67 imported cases of malaria were reported each year between 2000 and 2012 in WA alone (Table 21). Most of these were P. falciparum and P. vivax.

Imported cases of malaria account for the majority of cases diagnosed in Australia. Few cases of locally acquired malaria are reported, and most of these occur in the Torres Strait, close to Papua New Guinea. However, local outbreaks of malaria have been reported at Cape Tribulation in northern Queensland in 1996 and 2002, where the source of infection was probably an infected person arriving from endemic areas. In addition, a case of 'airport malaria' was reported in a local resident in Cairns in 1996, when it is thought that an infected mosquito carried on an aircraft may have been responsible.

A number of Australian *Anopheles* species are probably capable of transmitting malaria. *An. farauti* s.l. is considered the most likely vector for transmission in northern Australia. It is also an important vector of malaria in coastal parts of Papua New Guinea and the Solomon Islands. The widely-distributed *An. annulipes* s.l. is considered a possible vector in southern Australia.

Other species such as *An. hilli* and *An. amictus* are also possible candidates. Environmental conditions conducive for maintaining endemic malaria transmission occur north of latitude 19°C (just south of Broome in WA; north of Tennant Creek in the NT and Townsville in Queensland).

ORGANISM	2000	2001	2002	2003	2004	2005	2006	2007	2008	2009	2010	2011	2012	Total
Plasmodium falciparum	28	15	14	38	21	66	70	51	60	43	34	35	19	494
Plasmodium malariae		1		2	1	2		3	3	1	3	2	1	19
Plasmodium ovale	1			3	5	2	2	6		3	1	3	1	27
Plasmodium vivax	25	22	13	8	2	5	17	14	19	24	16	6	5	176
Undetermined <i>Plasmodium</i> sp.	2	6	5		5	3	21	7	8	10	2	20	26	115
Mixed infection (unspecified)	0	2	0	8	1	6	6	2	3	5	2	0	1	36
Total	56	46	32	59	35	84	116	83	93	86	58	66	53	867

 Table 21: Notified cases of malaria in Western Australia with optimal dates of onset between 01/01/2000 to 31/12/2012

Notes:

1) Data current as at 19/06/2013 - table may vary from previous or future versions due to inclusion of additional enhanced surveillance data.

2) Source of data: Western Australian Notifiable Infectious Diseases Database (comprising Doctor's notifications to Public Health Units & Communicable Disease Control Directorate; Laboratory reports to Communicable Disease Control Directorate from participating pathology laboratories); Enhanced Surveillance Data (comprising case follow-ups from Environmental Health Officers; patient interviews; Doctor's comments on notification forms).

3) Data varies from official Western Australian Notifiable Infectious Diseases Database records due to inclusion of Enhanced Surveillance Data.

4) NB - All cases had a history of travel outside of Western Australia (i.e. none of the notified cases were acquired in WA).

5) This information is the intellectual property of the Mosquito-Borne Disease Control Branch of the WA Department of Health and may not be used for any purpose without prior permission.

SURVEILLANCE

Surveillance of malaria in Australia is aimed at rapid detection and confirmation of human cases, enabling a timely response to prevent local transmission. The travel histories of patients need to be investigated quickly and they must be advised to avoid being bitten by local mosquitoes (by remaining inside screened dwellings or using repellent and covering up with appropriate clothing). If local transmission of malaria is suspected (eg, a case of malaria in a person who has not travelled), or if there is any doubt that a patient with malaria has been bitten by local mosquitoes, an entomological survey of adult and larval mosquitoes should be conducted immediately by local health authorities to determine the likely species present and potential for malaria transmission. Rapid control measures may need to be implemented, including adult and larval mosquito control and public awareness campaigns to ensure that people take sufficient caution to avoid exposure to potentially infected mosquitoes. Good communication between relevant health authorities (eg. Public Health Units, Environmental Health Officers and the DOH Mosquito-Borne Disease Control Branch) is essential to facilitate a rapid response to assess and eliminate the risk of further transmission in the area.

MANAGEMENT OF MALARIA

The principles of treatment are to cure and prevent relapses in the patient, and to eliminate the possibility of transmission. Management aims to:

- 1. Eradicate malaria parasites using antimalarial medication;
- 2. Eradicate malaria gametocytes using a specific gametocidal drug to prevent transmission of the parasite to mosquitoes;
- 3. Eradicate dormant liver forms of P. vivax and P. ovale with appropriate medication; and
- 4. Prevent transmission to local mosquitoes by physically separating the patient from local mosquitoes until the patient is no longer capable of transmitting the parasite.

All cases of *P. falciparum* malaria (including co-infections), or cases where the species cannot be determined quickly, are admitted to hospital to prevent life-threatening complications. In addition, all co-travellers should be tested for malaria.

SUMMARY

A number of exotic and emerging diseases are of concern to Australia, several of which have been discussed here. Vigilant surveillance measures and good lines of communication between public health authorities must be maintained (and improved where necessary) in order detect incursions and/or monitor potential establishment of these diseases in local transmission cycles. Efforts must also be made to continue to monitor incursions and spread of exotic mosquito vectors that may be involved in the establishment of local transmission cycles of exotic and indigenous mosquito-borne diseases.

SUGGESTED READING:

- 1. Anonymous (2004). Malaria protocol. Guidelines for health professionals in the Northern Territory 4th ed June 2004. *The Northern Territory Disease Control Bulletin* 11: 6-19.
- 2. Jansen CC, Webb EC, Northill JA, Ritchie SA, Russell RC and van den Hurk AF (2008). Vector competence of Australian mosquito species for a north American strain of West Nile virus. *Vector-Borne and Zoonotic Diseases* 8: 805-811.
- 3. Knope K, Whelan P, Smith D, Johansen C, Moran R, Doggett S, Sly A, Hobby M, Kurucz N, Wright P, Nicholson J and the National Arbovirus and Malaria Advisory Committee (2013). Arboviral diseases and malaria in Australia, 2010-11: Annual report of the National Arbovirus and Malaria Advisory Committee. *Communicable Diseases Intelligence* 37: E1-E20.
- 4. Mackenzie JS, Johansen CA, Ritchie SA, van den Hurk AF and Hall RA (2002). Japanese encephalitis as an emerging virus: the emergence and spread of Japanese encephalitis virus in Australasia. *Current Topics in Microbiology and Immunology* 267: 49-73.
- 5. Mackenzie JS, Smith DW and Hall RA. (2003). West Nile virus: is there a message for Australia? *Medical Journal of Australia* 178: 5-6.
- 6. Murgue B, Zeller H and Duebel V. (2002). The Ecology and Epidemiology of West Nile Virus in Africa, Europe and Asia. *Current Topics in Microbiology and immunology* 267: 195-222.
- 7. Roehrig JT, Layton M, Smith P, Campbell GL, Nasci R and Lanciotti RS. (2002). The emergence of West Nile virus in North America: ecology, epidemiology, and surveillance. *Current Topics in Microbiology and Immunology* 267: 223-240.
- Russell RC. (1998). Vectors vs. humans in Australia who is on top down under? An update on vector-borne disease and research on vectors in Australia. *Journal of Vector Ecology* 23: 1-46.
- 9. Smith DW, Hall RA, Johansen CA, Broom AK and Mackenzie JS. (2009). Arbovirus Infections. *In* Manson's Tropical Diseases, 22nd Edition.
- 10. Vaughn DW and Hoke CH (1992). The epidemiology of Japanese encephalitis: prospects for prevention. *Epidemiological Reviews* 14: 197-221.

USEFUL WEBSITES:

- 1. World Health Organization International Travel and Health website: <u>www.who.int/ith/index.html</u>
- 2. Centers for Disease Control and Prevention www.cdc.gov/travel/diseases/htm#malaria

27. PLANNING A MOSQUITO MANAGEMENT PROGRAM

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INTRODUCTION

The management of mosquitoes is rarely as simple as taking one approach (e.g. the application of larvicide) to achieve an acceptable level of control. In general, an effective mosquito management program will be based on an integrated approach, that combines various methods to minimise interaction between mosquitoes and the public and to reduce the risk of mosquito-borne disease, and yet will be environmentally and economically sustainable over the long-term.

Mosquito populations will fluctuate from year to year (as well as seasonally) in response to changing environmental conditions. Therefore there will need to be corresponding flexibility in the resources available to the program.

Approaches to mosquito management can be direct or indirect. Direct interventions include the removal of breeding habitat by physical modification, the introduction of biological controls (e.g. predatory fish) or the application of pesticides. Indirect approaches reduce humanmosquito conflict, for example utilising planning mechanisms to create adequate buffers around wetlands and educating the public to avoid mosquitoes. Another important indirect approach is for mosquito managers to actively liaise and collaborate with other departments/authorities to ensure that storm water and wastewater management, the planting and harvesting of aquatic vegetation, the design of roads, prevention of animal and vehicle access, and the impact of land use (e.g. mining, irrigation, farming) are undertaken in such a way as to minimise the potential for mosquito breeding.

This chapter provides a generic framework and checklist to assist people developing a mosquito management plan. Related elements and considerations for mosquito management plans have been grouped together with a brief explanation about their significance to mosquito management.

This information is a brief coverage of the issues only, and it is intended that reference should be made to other chapters in this manual for more detailed treatment of the various aspects of mosquito management.

WHERE DO I START?

Knowing just where to start with a mosquito management plan may seem daunting, especially if there is little or no evidence of a previous program in your region. However, in most cases there is some information that will help you get underway. This can be used to decide when and where to start with surveys to define and manage the problem.

EXISTING INFORMATION

As a starting point, contact colleagues in your own and other organisations, including previous incumbents. The following will provide important clues about the sources and extent of mosquito impacts:

- Previous mosquito surveys or reports if they exist (within agency or contact the Environmental Health Hazards Unit (EHH) of DOH)
- Public complaints (most local governments keep a complaints register)

- Disease reports and case follow-up information [can be obtained from DOH (EHH Unit, Population Health Units)]
- Geographical survey:
 - location of man-made water infrastructure (belonging to council and other agencies) (e.g. sewage lagoons, constructed wetlands, rainwater and effluent re-use tanks, roadside drains and culverts)
 - maps, aerial photographs
 - o local knowledge
- Land ownership & responsibilities (council planners, Dep't of Land Administration)
- Applicable environmental legislation (council planners; environmental agencies)

BASELINE MOSQUITO SURVEYS

If there is no prior information about mosquito breeding sites, seasonal productivity and the most prevalent species, then the following baseline surveys will be essential.

- Larval surveys: survey all potential mosquito breeding habitats, natural and man-made
- Adult surveys: undertake adult mosquito trapping in a range of natural and domestic locations
- Timing of surveys: surveys should follow breeding triggers [e.g. rainfall, tides, human manipulation of water sources (irrigation, dam releases, backyard sprinklers, effluent re-use)] to maximise the effectiveness of the survey to locate breeding sites
- Prioritise surveys in areas closest to residential and recreational areas and work out from there

DETERMINING MOSQUITO MANAGEMENT NEEDS AND OPTIONS

The analysis of existing information and baseline mosquito surveys (above) will allow you to decide whether, when, where and how mosquito management should be undertaken.

Necessity (the need for control):

- Cases of mosquito-borne disease
- Severe nuisance (complaints, impact on quality of life)

Timing:

- Likely season(s) of nuisance and disease risk
- Triggering environmental conditions or human activities
- Timing of monitoring and treatments (larviciding/adulticiding) or other interventions in relation to season, breeding triggers, activity of life stages of the target species

Priority sites from nuisance and public health perspectives:

- Proximity of breeding sites to human habitation
- Productivity of sites (size of breeding area and density of larvae)
- Pest and disease vector status and biology of mosquito species emanating from site

Options for management within available resources:

- Cultural will the public respond to encouragement about personal preventive measures?
- Physical (source reduction) can the site be modified or removed to prevent breeding?

- Chemical larvicides (ground and aerial applications)
- Chemical adulticides (fogging and residual surface adulticides)
- Biological is it possible and appropriate to introduce mosquito predators (e.g. fish) to the site?

IN-PRINCIPLE SUPPORT

• Obtain initial in-principle support for a program based on the above (later, the organisation will need to accept the program as part of the core business plan to ensure ongoing commitment to funding and support)

OPERATIONAL ASPECTS

Once you've decided on the broad approach you will need to determine necessary resources, stakeholder support and involvement, and then implement the program.

DETERMINING BUDGET AND RESOURCES

- Manpower personnel required to undertake the management options identified above
- Equipment chemical application, earthworks, PPE, etc
- Chemicals, prices, number of treatments, area to be treated
- Advertising, educational and promotional material
- Vehicles

APPROVALS AND COLLABORATION

- Identify key partners/stakeholders
- Seek environmental approvals
- Seek aboriginal heritage and native title approvals
- Inform other departments/agencies about proposed program and liaise over potential conflicts (e.g. with agricultural biocontrol programs)
- Advise other departments/land-owners of management responsibilities and options

PUBLIC EDUCATION, ADVICE AND WARNINGS

The public have a key role and responsibility in any integrated program to manage mosquitoes. It is important that communities are kept informed and become stakeholders in achieving a successful program.

- Develop information displays and material for letter drops
- Undertake school and community education
- Promote the program and your key messages using local media
- Disseminate warnings when environmental and mosquito monitoring indicate a risk of mosquito-borne disease is likely
- Advise the public of planned chemical and physical mosquito control activities
- Inform and educate the public about their responsibilities for personal preventive measures and backyard mosquito control (e.g. septic tanks, rainwater tanks, fish ponds, roof gutters, pot plant saucers, tyres and other water-holding 'containers')

DETERMINING THE EFFECTIVENESS OF THE PROGRAM

- Post-treatment monitoring of larvae and adults
- Monitor public complaints
- Occasional inspection of physically modified sites
- Measure the coverage and impact (on behaviour) of your publicity and warnings
- Monitor human case notifications (although this may not necessarily indicate the effectiveness of the program because disease transmission depends on more than just adult mosquitoes)

SUPPORT AND RESOURCES TO MAKE A MOSQUITO CONTROL PROGRAM HAPPEN

Effective mosquito management is an ongoing commitment for the agency concerned. This means dedicating some time to ensuring the program is supported and adequately resourced over the long term.

ORGANISATIONAL COMMITMENT

- Secure long-term commitment to program from council/agency by adoption of strategic plan
- Achieve recognition of fluctuating nature of funding requirements
- Achieve recognition that program will evolve and grow over time
- Ensure local councillors, politicians and community are aware of and supportive of your program
- Promote the need for adequate buffers between residential areas and high risk areas for nuisance and disease vectors with planning staff in your agency

PROGRAM FUNDING

- Secure funding for current financial year
- Obtain commitment to long-term funding in line with the agency's adoption of your strategic plan
- Seek opportunities for collaborative funding (e.g. DOH CLAG funding program, mining companies, local industry)
- Investigate the possibility of developer contributions to funding control of mosquitoes affecting new residential subdivisions

DOCUMENT PROGRAM ACTIVITIES AND PROCEDURES

Deliberately develop an institutional memory of the program to prevent loss of knowledge and information when staff leave or are promoted through the organisation.

- Maintain thorough records/archive files on the mosquito management program
- Document activities and write an annual report/summary
- Archive copies of maps, aerial photos, equipment manuals, chemical labels and other operational resources
- Integrate your program onto your agency's Geographic Information System, if it has one

• Ensure that other staff receive training in running the program so that back-up is available when needed (e.g. during annual leave), and to avoid the loss of your knowledge and experience if you resign

ONGOING REFINEMENT OF THE PROGRAM

There will be an ongoing and indefinite need to review and refine the program. Additional breeding sites will be found and some others may be created by human activities. Alternative approaches to mosquito management may become available or desirable (e.g. due to the development of resistance to a particular chemical group).

Periodically, review achievements and results from several consecutive seasons to identify emerging trends or risks.

Join the Mosquito Control Association of Australia and attend their conferences to continue to develop your professional skills and knowledge in this field (see website below).

FURTHER READING:

• Mosquito Control Association of Australia Inc. (2008). Australian Mosquito Control Manual. For purchasing details, see http://www.mcaa.org.au

MOSQUITOES AS A QUARANTINE ISSUE

(3 papers)

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28. EXOTIC MOSQUITOES ARRIVING ON SEAGOING VESSELS: RECOMMENDED INSPECTION & ERADICATION PROCEDURES

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INTRODUCTION

Various States in Australia are concerned about the possible importation and subsequent introduction or establishment of *Aedes aegypti* (the dengue fever mosquito), *Aedes albopictus* (The Asian tiger mosquito) and other exotic *Aedes* mosquitoes (such as members of the *Ae. scutellaris* group which occur in South East Asia, New Guinea and the Pacific) from interstate or overseas. *Aedes aegypti* is only established in northern Queensland (Sinclair 1992) but importation of this species from overseas to areas where it is already established could introduce new strains of the species or exotic viruses. Any measures to prevent the importation of *Ae. albopictus* into mainland Australia should also intercept *Ae. aegypti*, as Australia (except for the recent establishment of this species on islands in the Torres Strait) is free of *Ae. albopictus* (Kay et al. 1990). One of the demonstrated methods of importation of these mosquitoes from overseas is via seagoing vessels and their cargo, including commercial vessels, refugee vessels, fishing vessels and seagoing yachts (Whelan 1981, Kay et al. 1990).

In northern Australia the greatest potential risk from overseas is by importing drought resistant eggs that have been deposited on the inside of a range of receptacles or containers. The receptacles likely to contain larvae or eggs are open or stoppered receptacles for transporting drinking water such as drums or jerry cans, vehicle tyres used for packaging, spare tyres that are exposed to the rain, and deck stored machinery and equipment. The most frequent mode of importation has been on illegal fishing vessels intercepted in Australian waters. Vessels with refugees or illegal migrants, smaller trading vessels and barges, and private yachts, have also been sources of exotic mosquitoes (Whelan 1991, Whelan and Tucker 1998).

With the establishment of *Ae. albopictus* in the Torres Strait, the possible importation of this species to mainland Qld coastal communities becomes a very appreciable risk. All islands in the Torres Strait should now be regarded as infested with this species, and vessels and cargo from the Torres Strait should be treated as if they are from overseas ports. This means not only traditional importation receptacles need to be considered, but domestic receptacles such as potplant drip trays, toys, shells and indeed any receptacle exposed to rain. As well, small vessels such as dingies and canoes now need to be regarded as potential modes of importation.

An active inspection program of overseas vessels is currently being performed by Quarantine authorities in at least major ports. The intensity of inspection or the emphasis on detecting mosquitoes is variable between States. These, and additional inspections of other risk areas, are vital in excluding these vectors from Australia. The procedures outlined below are a guide to carrying out the inspection and elimination measures to ensure that Australia remains free of additional vectors or disease problems.

EQUIPMENT

The collecting equipment required includes a torch, a bulb pipette attached to a flexible clear hosing, white painted ladle, bucket, sample bottles, tissues, labels, pens, pipette, forceps,

insect collection containers, aerosol knockdown insecticide spray, white sheets, insect catch net, and record forms. All this equipment should be readily available for each inspection. Onvessel larval or pupal control operations usually necessitate an interim treatment, and require cans of aerosol pyrethroid insecticide.

On-shore elimination procedures require require cans of aerosol insecticide, a 5 litre pressure sprayer with a residual pyrethroid insecticide (preferably alpha cypermethrin or lambda cyhalothrin, and a 5 litre pressure sprayer with liquid chlorine and detergent, marker paint, spray cans, recording equipment.

VESSEL INSPECTION PROCEDURES

- 1. All vessels should be inspected for adult or larval mosquitoes. If possible all vessels should be inspected for adult mosquitoes before the vessels enter a harbour.
- 2. All inspections should be carried out in a specific quarantine area that should be at least 1 km from shore. If the vessel is brought direct to a landing, the inspection and control of adult mosquitoes should be carried out en route before any landing is made.
- 3. Any adult mosquitoes observed flying in any areas, such as holds or cabins, should be knocked down with aircraft disinsection spray or collected in a net. Attempts should be made to collect the knocked down mosquitoes by laying out white sheets in closed areas before spraying. Any insects collected from the sheets should be packaged in a secure container with a light packaging of tissue paper. The container should be labelled and forwarded as soon as possible to a specialist for identification.
- 4. The vessels containing live mosquitoes should be completely sprayed with aircraft disinsection sprays. Any intercepting vessel capable of offering mosquito harbourage that has been alongside a foreign vessel with adult mosquitoes should also be sprayed after leaving the intercepted vessel. Holds and other dark humid areas, including under beds and in cupboards, should be sprayed with aircraft disinfection spray. Any vessels with adult mosquitoes should be thoroughly searched to establish the source of the mosquitoes. Any vessel on which 4th instar larvae and pupae, or pupal skins are detected, should be sprayed on the presumption that live adult mosquitoes are present.
- 5. Any receptacles capable of holding or collecting water should be inspected for water and mosquito larvae. In most instances this will require the use of a torch and a white ladle. Receptacles needing examination will include bottles, cans, tyres, machinery parts, drums, and drinking water storages such as jars, jerry cans, and large water tanks. A large bulb pipette or siphon should be used for difficult to access receptacles or water tanks. Any receptacle used to carry water, irrespective of whether it is sealed, should be inspected for larvae. Water from water holding tanks should be sampled for mosquito larvae by net, by filling a bucket of water via a drain tap, and by using a large bulb pipette, or manual suction pump. Hygiene precautions should be taken when sampling drinking water to ensure water is not contaminated by the ladle or other sampling techniques.
- 6. All mosquito larvae or larval and pupal skins detected in any receptacles should be collected into labelled vials of 70 % alcohol and forwarded to a specialist for identification on the day of collection, together with collection details and a report form.

Where there are secure facilities and prior approval has been given, larvae and pupae in receptacles arriving from South East Asia, Timor Leste, New Guinea, the Pacific and the Torres Strait should be collected live, placed in labeled secure containers with original water, and transported immediately to the State Health entomologist or entomology specialist, so that the larvae and pupae can be link reared to adults (live collection can be

dispensed with when routine DNA testing is available). This is to clarify the identity of possible unknown *Aedes* species entering Australia from the above areas. Secure facilities and ready access to a mosquito specialist is required to prevent the escape of live exotic mosquitoes.

7. For larger vessels, built-in freshwater tanks should be inspected to ensure that all openings are sealed or mosquito proof. Any filling points should be made mosquito proof (at least temporarily) with the aid of materials such as insect netting, until examination and treatment are complete, or the vessel leaves port.

ERADICATION PROCEDURES ON VESSELS

- 1. Once examined for mosquito larvae, all water in any receptacles (except that in built-in water storages) should be emptied into the sea or the receptacles sealed until the vessel leaves port.
- Any vessel on which adult mosquitoes, live larvae or pupae, or pupal skins are detected, should have all receptacles on board that are either holding water, or likely to have recently been mosquito breeding sites, treated with chlorine and detergent (Shortus and Whelan 2006) or sprayed with alpha-cypermethrin or lambda-cyhalothrin to the point of run off.
- 3. Receptacles which held water or dry containers likely to have been a mosquito breeding site, should be stored under cover if the vessel is due to remain in port for more than 5 days.
- 4. No receptacle which held water or dry receptacles likely to have been a mosquito breeding site should be allowed off a vessel until it has been fumigated, chlorinated or treated with a residual insecticide.
- 5. If the vessel is due to go to another Australian port and has receptacles which hold water, those receptacles should be chlorinated, treated with residual insecticide, sealed or stored under cover. Authorities in the next port of call should be advised of the potential risk.

ONSHORE ELIMINATION MEASURES

- 1. Onshore inspection and elimination measures should be carried out within one day of detection of live exotic *Aedes* adults or larvae in association with pupae in any receptacle on an overseas vessel that has docked at a port or landing facility. Onshore inspection and eradication should also be carried out immediately if any exotic mosquito larvae are detected in the onshore ovitraps.
- 2. All property owners and businesses in the immediate area of importation (generally 500 m is sufficient) should be briefed on the importation and the importance and details of the elimination measures. No potential receptacles, particularly tyres, should be allowed to be removed from the premises until treated.
- 3. Adult mosquito aerosol fogging operations should be carried out within 500 m of the importing vessel or ovitrap site, with particular attention to the interior of warehouses and sheltered areas, or areas that contain potential breeding and harbouring sites. The fogging should be done in the mid morning or late evening when mosquitoes are likely to be most active.
- 4. A receptacle search should be done within 500 m of the landing site or ovitrap site. All receptacles with water should be located, with each receptacle marked to indicate

orientation. The number and location of each receptacle, and the presence of water and larvae, should be documented.

- 5. All larvae in each receptacle should be collected and placed in individual labelled collection vials of 70 % alcohol. Pupae should be transferred live to secure vials with adequate breeding site water and hatched in a secure laboratory. All larvae and pupae should be identified on the day of collection to locate the highest risk receptacle.
- 6. All water holding receptacles should be treated with chlorine/detergent mixture or residual pyrethroid insecticide to the whole interior surface. Receptacles with exotic larvae should be highlighted to prevent movement and ensure additional scrutiny.
- 7. If time and resources allow, an evaluation of the presence of exotic eggs in any receptacle could be made before chlorination or insecticide treatment. For this procedure every receptacle with water should be completely emptied and dried. This is to ensure that no late instar larvae or pupae inadvertently missed can later develop to maturity and emerge. All previously wet receptacles or receptacles likely to have held water at the time of importation are then completely filled with tap water and left for 3 days before reinspecting for mosquito larvae, and sampling the larvae. Each water filled receptacle should then be treated with chlorine or residual insecticide.
- 8. When using chlorine, the chlorine and detergent application method (Shortus and Whelan 2006) should be repeated after 30 minutes to assist the kill of any unhatched *Aedes* eggs.
- 9. The remaining chlorine solution should be completely rinsed from all the water receptacles within one day of treatment.
- 10. Additional ovitraps should be placed in the detection area and inspected weekly for two months and again after the next rain episode.
- 11. The public and local businesses should be informed of the results of the inspection and eradication measures.
- 12. The procedures and the results of the inspections should be completely documented, and the report should be forwarded to the relevant authorities.

CONCLUDING REMARKS

These inspection and elimination procedures should be supplemented by an *Aedes* ovitrap and CO2 baited BG trap surveillance program at least around international port areas. The possibility of the introduction and establishment of *Ae. albopictus* and *Ae. aegypti* to various States in Australia is very real. There have been instances of the interception of these vectors in Darwin, Broome, Townsville, Cairns and Brisbane (Whelan 1991). If the vectors are introduced, particularly to the northern areas of Australia, there will be new pest mosquito problems and additional risks of dengue outbreaks and epidemics (Kay et al. 1984). Failure to detect an importation very early, and to react immediately and thoroughly, could result in the establishment of these vectors, ongoing and expensive control operations, and increased mosquito borne disease.
REFERENCES

- Nguyen H, Whelan PI, Finlay-Doney M, Ying Soong S (2010). 'Interceptions of *Aedes aegypti* and *Aedes albopictus* in the port of Darwin, NT, Australia, 25 January and 5 February 2010'. *Northern Territory Disease Control Bulletin* Vol 17(1): 29-35.
- Kay, B. H., Barker-Hudson, P., Stallman, N. D., Weimers, M. A., Marks, E. N. M., Holt, P. J., Muscio, M., Gorman, B. M., (1984), 'Dengue Fever - Reappearance in northern Queensland after 26 years', *Medical Journal of Australia*, vol. 140, pp. 264-268.
- Kay, B. H., Ives, W. A., Whelan, P. I., Barker-Hudson, P., Fanning, I. D. & Marks, E. N. M., (1990), 'Is *Aedes albopictus* in Australia?', *MJA*, vol.153, pp.31-34.
- Pettit W, Whelan PI, McDonnell J and Jacups S (2010). 'Efficacy of alpha-cypermethrin and lambda-cyhalothrin applications in tires to prevent *Aedes* mosquitoes breeding' *J Am Mos Control Assoc* Vol. 26, No. 4.
- Shortus M and Whelan PI (2006). 'Recommended Interim Water Receptacle Treatment for Exotic Mosquitoes on International Foreign Fishing Vessels Arriving in Australia'. *Northern Territory Disease Control Bulletin.* 13:2:32-34.
- Sinclair, D. P., (1992), 'The distribution of *Aedes aegypti* in Queensland, 1990-1992', *Bull of the Mos Cont Assoc of Aust*, vol. 4, no. 2, pp. 17-24.
- Whelan, P. I., (1981), 'The vulnerability and receptivity of the NT to mosquito borne disease', *Trans Menzies Found*, vol. 2, pp. 165-171.
- Whelan, P. I., (1991), 'Northern Territory remains free of dengue fever', *Bull Mos Control Assoc Aust*, vol. 3, no. 1, pp. 7-9.
- Whelan PI and Tucker G (1998). 'Exotic *Aedes* surveillance and exclusion from the Northern Territory of Australia'. Supplement to the *Bulletin of the Mosquito Control Association of Australia.* 10:3.
- Whelan PI, Kulbac M, Bowbridge D, and Krause V (2009). 'The eradication of *Aedes aegypti* from Groote Eylandt NT Australia 2006-2008' *Arbovirus Research in Australia* 10:. 188-199.

29. EXOTIC AEDES SURVEILLANCE PROGRAM OVITRAP SERVICING PROCEDURES

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INTRODUCTION

Ovitraps are special egg traps used to detect the presence or importation of dengue and yellow fever vector mosquitoes such as *Aedes aegypti* and *Aedes albopictus. Aedes aegypti* is absent from all States and Territories in Australia except Queensland (Sinclair 1992), while *Ae. albopictus* is not present in mainland Australia but poses a real threat (Kay et al. 1990, Whelan 1981, Whelan 1991).

Standard ovitraps consist of a water filled blackened jar with a porous paddle dipping into the water. These jars are particularly attractive egg laying locations for artificial receptacle breeding mosquitoes such as *Aedes* (Stegomyia) mosquitoes. They are usually situated at favourable sites around vulnerable points of entry such as international airports or seaports. However for jurisdictions such as the NT, WA and NSW, where importation of *Ae. aegypti* can occur from Queensland, the vulnerable entry points include domestic airports and shipping facilities, tyre businesses, caravan parks, plant nurseries and other businesses with Qld interstate connections, and Qld interstate transport companies and removalists. Those towns with direct road, sea or air connections to Queensland are vulnerable, with the highest potential occurring at sites closest to Queensland, and routes with the most traffic.

An ovitrap program can be a relatively cheap method to detect an exotic *Aedes* importation. However it is a detection method for an event that may happen with very low frequency. There may be many years of negative results except the recovery of domestic species already present in the jurisdiction. This can lead to complacency and a reduction in priority or allocation of resources. However it may only need one timely interception in 10 or 20 years of operation to achieve the objective of preventing the establishment of an exotic mosquito species.

An ovitrap program can only be useful if it is a thorough and continuous operation, with weekly or fortnightly servicing, and with people who can competently rear and identify mosquito larvae. There should also be a regular evaluation of the procedures and the results of the program. If ovitraps are operated effectively and efficiently, they can be early warning indicators for timely eradication procedures. Timely eradication can ensure the various States and Territories remain free of exotic species of *Aedes* mosquitoes and certain mosquito borne diseases.

EQUIPMENT REQUIRED

The equipment required includes:

Field equipment

- 1 litre black painted glass jars or clear glass jars inside black plastic casing containers (ovitrap jars)
- 3 sets of numbered 'masonite' paddles approx 16 cm long and 3 cm wide, with stainless steel wire hooks attached to one end

- A bottle of de-chlorinated water (tap water left to stand in containers). These are filled after each use and set aside for the following week.
- Results sheet or record book
- Observation form (to record any disturbance to ovitrap)
- Maps with exact locations of ovitraps
- Jar carry basket

Laboratory equipment

- Rearing trays (12 x 17 x 4 cm)
- Ground fish food.
- A supply of de-chlorinated tap water
- Labelled larval storage jars
- 70 % alcohol
- Microscope and associated equipment
- Larval identification keys
- Results and summary record sheets

THE STANDARD OVITRAP

The standard ovitrap consists of a 1 litre blackened screw top glass jar (with lid) and a masonite paddle $(15 \times 2 \text{ cm})$. The paddle is suspended vertically by a stainless steel hook onto the inside of the jar with the rough side facing outwards. Each jar, paddle, screw on lid and black plastic casing for the ovitrap jar is numbered with an identical number and each ovitrap has a different number. Three sets of numbered paddles are used on a rotational basis. One set is in the ovitrap, one set is being hatched and the other set is being sterilised and dried.

OVITRAP POSITION SELECTION

Ovitrap positions at vulnerable sites should be relatively secluded, shaded, low to the ground, near vegetation and protected from rain and animal disturbance. They should also be adjacent to areas where there is regular human activity. Female *Aedes* (Stegomyia) mosquitoes are generally attracted to humans for blood meals and require a nearby egg laying site after feeding. Ovitraps can be placed between two bricks or stones, or behind or under a suitable object such as a wash trough if they are prone to disturbance. If animals drink from the ovitraps, a wide mesh screen can be placed over the ovitrap, but the mesh should be no smaller than 2 cm to prevent hindrance to mosquito entry. The ovitrap should not be placed in a position with spider webs or inside very thick vegetation.

PREPARATION OF OVITRAPS FOR PLACEMENT

Ovitraps are prepared in the laboratory or special preparation room.

- 1. Fill the clean ovitrap jars with de-chlorinated water to depth of 10 cm (approx. 650 ml).
- 2 Add a few leaves and twigs of aged lucerne or some fine ground fish food.

- 3. Place the numbered clean masonite paddle into the corresponding ovitrap jar and screw on the lid.
- 4. Put the ovitrap jar into corresponding black plastic casing.
- 5. Load the ovitraps into the carry baskets ready for transportation to the sites.

OVITRAP PLACEMENT

The new ovitraps are transported to the selected ovitrap positions.

- 1. New ovitraps are positioned at the same time as exposed ovitraps are collected.
- 2. At the trap site unscrew the lid and hook the masonite paddle over the lip of ovitrap jar.
- 3. Ensure that the ovitrap jar, masonite paddle and black plastic casing have corresponding site numbers.
- 4. Ensure that the water in the jar is at the correct level and top up if required.
- 5. Place the ovitrap in the designated position.

OVITRAP COLLECTION

Exposed ovitraps are collected after the new ovitraps have been positioned.

- 1. Unhook the paddle and lower it into the jar.
- 2. Screw on the lid, (from the new ovitrap just positioned), to the exposed ovitrap to be collected. Make sure that the lid is screwed on tightly.
- 3. Recorded any disturbances to the ovitrap on the observation form (eg. invasion of ovitrap by ants or frogs, ovitrap stolen or vandalised, no water left in ovitrap jar, or jar tipped over).
- 4. Collect the exposed ovitrap and record the presence of larvae or pupae on the observation form.
- 5. Return collected ovitraps to the laboratory.
- 6. Any relocation of the trap position should be recorded on the results form and the field map.

The presence of any pupal skins indicates that inspections have been too far apart and the ovitrap servicing period should be shortened. This aspect should be brought to the attention of the supervisor immediately on return to the laboratory.

OVITRAP PROCESSING

All rearing should be done in a secure mosquito proof room or an insectary. The insectary should have warning signs on the entry and there should be restricted access. No insecticides should be used in the insectary and contact with insecticides should be avoided prior to entering the insectary.

Immediately on returning from the field:

- 1. Remove the paddle from the ovitrap and lay it out on clean paper towels (rough surface up) to dry for three days, in an ant free environment (use water baths or ant traps). Ensure that the paddles do not touch each other.
- 2. Tip water from ovitrap jar into a rearing tray (12 x 17 x 4 cm) that is marked with trap site number and the date of collection.
- 3. Remove any 4th instar larvae and place them into a collection vial that contains 70 % ethanol. Label the vial with the trap site number and the collection date.
- 4. If any live pupae are found, place them in a small container with water from the collected ovitrap (eg plastic urine sample container). Label them with the trap site number, date of collection and the number of pupae recovered. Place this container in a separate rearing container or cage. Check for adult emergence every day. Maintain the adults alive for approximately 24 hours after emergence, and then kill them by freezing for 10 minutes. Identify the adult mosquitoes and record the identifications on the record form.
- 5. Reared adults of common domestic species can be discarded. New or exotic species must be pinned and labelled by standard procedures and stored adequately in insect storage boxes or cabinets.
- 6. Collect any larval or pupal skins in the ovitrap and place in a vial with 70 % ethanol. Label the vial with the trap site number and the collection date. The larval skins can be used for identification.
- 7. Record on the results sheet if any larvae or pupae were found in the ovitrap jar.
- 8. Repeat for each ovitrap.
- 9. After three days the dry paddles are placed in numbered rearing trays and fully immersed in de-chlorinated water for 7 days with added fine ground fish food flakes in solution added under the water surface with a dripper.
- 10. Glass jars are examined by eye for mosquito eggs, dried for 3 days, flooded to the top and left for 1 week. This will allow hatching of any eggs on the side of the glass jar. If eggs were observed on the glass, sufficient food is placed in the jar when flooded, and observed daily for hatching. If no eggs are observed, a small amount of food is placed in each jar in case there are eggs present.
- 11. Any larvae hatched in the jars are placed in the corresponding rearing tray (see above).

If there is no hatching, the jars are sterilised by boiling water and washed in the dishwasher in preparation for reuse.

LARVAL REARING

1. Larval rearing should be carried out in an un-airconditioned mosquito proof room or insectary. The room will require adequate bench space for larval rearing trays. Each larval rearing tray may contain larvae over a 2 to 3 week period. If larval growth takes longer than 3 weeks, it may be necessary to raise the temperature of the room (oil heaters are useful).

- 2. Rearing trays are inspected every day after flooding of the paddles. Any dead larvae and 4th instar larvae are removed by pipette and transferred to a labelled vial of 70 % ethanol. Cross contamination between trays should be avoided by washing the pipette between each procedure.
- 3. Hatched larvae are reared in the rearing tray under a hygienic feeding program. The larvae are fed ground fish flakes (Excelpet Tropical Fish Food is satisfactory). The ground fish flakes are soaked in water for one hour and the suspension added with a pipette below the water surface to reduce bacterial problems on the water surface.
- 4. Food should be added at Day 1 to aid hatching and ensure food availability for newly hatched larvae. Additional food should be added as required during larval growth. Care must also be taken not to add too much food or to put fingers in the water as this can promote bacterial growth (evident by the presence of a white bacterial scum on the surface of the water). Any accumulation of old food should be removed with a pipette, avoiding cross contamination between rearing trays.
- 5. If any larvae appear different from *Ae. notoscriptus* or other common domestic species, a sample should be identified immediately.
- 6. The larvae in the trays are collected when they die or when they reach 4th instar stage. The larvae are identified with the aid of a microscope and larval identification keys, and the number of larvae of each species should be counted.
- 7. Common domestic larvae can be discarded after identification. Exotic species should be stored in 70 % alcohol in labelled vials and placed in a larval collection or reference facility. The identification of exotic larvae should be confirmed by a specialist taxonomist.

CLEANING OF OVITRAPS

- 1. After one week, paddles are removed from rearing trays, scrubbed clean to remove any old hatched eggs and scum, sterilised in boiling water and dried. When sterilising the paddles make sure all surfaces of the paddle are exposed to the boiling water.
- 2. Once all the larvae have been reared to 4th instar stage, or at the end of the 2 or 3 week rearing period, rearing trays are sterilised by boiling water and washed in a dishwasher.

RECORDING AND REPORTING OF RESULTS

- 1. Results of the identifications and numbers recovered from each ovitrap should be recorded on the results form at the end of each rearing period, and entered into a database or another recording method.
- 2. Any possible exotic mosquito species should be brought to the attention of the supervisor immediately.
- 3. Confirmed identifications of exotic species should be reported to the relevant authorities and they should be advised of the possible need to initiate immediate eradication procedures (see 'Exotic mosquitoes arriving on overseas vessels').
- 4. The yearly results should be tabulated in summary sheets. These should include the number of inspections, the number of times the ovitrap was positive, the number of larvae present, and the identity of all larvae. The results should be evaluated for under performing sites. Under performing sites should be considered for relocation unless they are close to high risk or demonstrated entry points.

SUMMARY

These procedures have been developed to enable the rapid detection of exotic *Aedes* larvae. They require a high degree of consistency and accuracy to be efficient and are, by necessity, long term programs requiring adequate allocation of resources. Ovitrap programs should be carried out with a corresponding source reduction program around the vulnerable sites to improve the likelihood of detection of importations. Once an exotic species has been detected and the identification confirmed, there should be a rapid and organised response, which will usually involve elimination procedures. If importations are detected rapidly, elimination measures have a high likelihood of success. An efficient *Aedes* ovitrap program can help to maintain Australia free of additional exotic mosquitoes and certain mosquito borne disease outbreaks.

REFERENCES

- Nguyen H, Whelan PI, Finlay-Doney M, Ying Soong S (2010). 'Interceptions of *Aedes aegypti* and *Aedes albopictus* in the port of Darwin, NT, Australia, 25 January and 5 February 2010'. *Northern Territory Disease Control Bulletin* Vol 17(1): 29-35.
- Kay, B. H., Ives, W. A., Whelan, P. I., Barker-Hudson, P., Fanning, I. D. & Marks, E. N. (1990), 'Is *Aedes albopictus* in Australia?', *Med J Aust*, vol. 153, pp. 31-34.
- Sinclair, D. P., (1992), 'The distribution of *Aedes aegypti* in Queensland, 1990-1992', *Bull Mos Cont Assoc of Aust*, vol. 4, no. 2, pp. 17-24.
- Whelan, P. I.(1981), 'The vulnerability and receptivity of the Northern Territory to mosquito borne disease', *Transactions of the Menzies Foundation*, vol. 2, Living in the North, pp. 165-171.
- Whelan, P. I. (1991), 'Northern Territory remains free of dengue fever', *Bull Mos Control Assoc Aust*, vol. 3, no. 1, pp. 7-9.

Whelan PI, Kulbac M, Bowbridge D, and Krause V (2009). 'The eradication of Aedes aegypti from Groote Eylandt NT Australia 2006-2008' Arbovirus Research in Australia 10: 188-199.

30. RECOMMENDED WATER RECEPTACLE TREATMENT FOR EXOTIC MOSQUITOES ON INTERNATIONAL FOREIGN FISHING VESSELS ARRIVING IN AUSTRALIA

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INTRODUCTION

Exotic *Aedes* mosquito larvae are commonly found in receptacles on equipment or in cargo that hold or have held water from overseas vessels arriving in the Northern Territory (NT) of Australia. This applies especially to international foreign fishing vessels (IFFV) from Indonesia, which are commonly intercepted fishing in Australian waters by the Royal Australian Navy (RAN) and Customs and detained in Darwin or Gove harbours. The drinking water storage receptacles aboard these vessels are often found to contain *Aedes aegypti* or *Aedes albopictus* larvae. Drinking water storage receptacles are the most commonly detected type of receptacle to transport exotic mosquito pupae, larvae or eggs into the NT. *Aedes* species eggs are desiccation resistant and can often be present in either water holding or dry receptacles. The eggs are laid just above the water level on the inner surfaces of receptacles. Approved procedures to treat drinking water receptacles only allow the use of chlorine, due to the residue concerns posed by the use of insecticides. These treatments are part of routine quarantine inspection and control procedures on vessels or aircraft in the 400 m quarantine zone around air and seaports.

As part of the previously recommended chlorination procedures, any water holding receptacles were emptied and treated with a chlorine spray to kill possible exotic Aedes eggs on inner surfaces^{1,2,3}. However, the previously recommended receptacle treatments that involved spraying the receptacle surface with a 1% active ingredient (AI) chlorine solution to the point of run-off did not adequately kill 100% of mosquito eggs⁴. This was due to the mosquito eggs not being exposed to the chlorine solution for a long enough period. The vertical position of the treated surface, the large clusters of eggs, the sometimes low relative humidity and the dilution of the chlorine solution are all factors that affected the efficacy of the previous treatment recommendations⁴.

Recent re-evaluations of the efficacy of chlorine against Aedes aegypti eggs, as well as the development of new egg treatment methods that use detergents, can be combined to provide an improved interim method of receptacle treatment. These recommendations are in response to requests by the Australian Quarantine and Inspection Service (AQIS) for suitable receptacle treatment protocols for drinking water receptacles aboard IFFV's. These recommendations will also hold for refugee vessels and general vessels carrying cargo that holds water, or other receptacles, from overseas. The recommendations are for procedures to treat drinking water receptacles containing, or likely to contain exotic mosquito pupae, larvae and/or eggs.

RECOMMENDATIONS

COLLECTION OF LARVAL AND ADULT SAMPLES

When a water storage receptacle breeding site is identified, it is important to collect larval, pupal and/or pupal skin samples before treating the receptacle. If the IFFV has landed in Australia before being intercepted, the collection of these samples allows an effective risk analysis of an exotic vector incursion to be undertaken. It will also allow a timely and adequate monitoring response to be set up in the event of a suspected or potential incursion, as well as

the rapid implementation of any control or eradication procedures that are required if there is a confirmed exotic vector incursion.

THE TREATMENT PROCEDURE

The recommended procedure for the treatment of drinking receptacles aboard IFFV's is to firstly pump, siphon, or pour out the stored water into the sea and to spray the inner receptacle surfaces with a chlorine/detergent solution to kill any eggs that may be present. All mosquito larvae and pupae would be killed by disposal into the sea, and any eggs remaining on the receptacle wall will be killed by the chemical treatment.

The chemical treatment comprises a mixture of 4.8 litres of a liquid sodium hypochlorite solution that has at least 10% AI, combined with approximately 200 ml of liquid dishwashing detergent. The 10% sodium hypochlorite solution can be purchased as 'liquid chlorine' (with at least a 10% AI) from most pool shops. The liquid concentrate detergent can be any major domestic or commercial brand name. The ingredients should be mixed thoroughly in a 5-litre pressure sprayer and applied liberally to the point of run off to all of the inner surfaces of the water storage receptacle. The receptacle should then be sealed with a lid or cover and left to stand. This process should be repeated after 30 minutes. Once the receptacle has been treated twice over a 60 minute period it should remain sealed and let stand for another 24 hours, after which it can be thoroughly cleaned and rinsed.

IMPROVEMENTS IN THE TREATMENT PROCEDURE

The two major problems with the previous chlorine surface treatment was that the chlorine solution did not remain in contact with the eggs for long enough to kill them, and that the 1% chlorine solution was too dilute to kill large numbers, and clusters of eggs. Recent studies have found that chlorine solutions with a 10% AI rate, when applied to *Aedes* eggs and left in contact with the eggs for at least an hour, and then left to incubate for at least 24 hours in a high humidity environment, can achieve a 100% mortality rate⁴. Other studies have found that by adding detergent to a chlorine solution, the mixture becomes a lot more viscous and can adhere to the walls of the receptacle for a longer period of time⁵.

Increasing the strength of the chlorine solution that is in direct contact with the eggs, increases the time that the treatment mixture is in contact with the eggs. Sealing the receptacle increases the humidity and hence reduces evaporation of the chlorine. These treatment procedures will provide a more effective control of Aedes eggs in IFFV's water receptacles than the previous recommendations.

The previous recommendations use only a 1% AI chlorine solution for surface treatment of eggs, which has been shown to be too dilute, as well as too thin, so that it runs off vertical surfaces too quickly 4.

REFERENCES

- 1. Ritchie S. (2001): Efficacy of Australian Quarantine procedures against the mosquito *Aedes aegypti*. Journal of the American Mosquito Control Association, 17(2): 114-117.
- 2. Whelan P. Lamche G. (2002): Recommended Chlorination Procedures for Receptacles Containing Mosquito Eggs for Quarantine Purposes, in: Bulletin of the Mosquito Control Association of Australia, Vol 14, No 3, p14-18.
- 3. Mosquito Control Association of Australia Inc. (2002): Control of Container Breeding and Exotic Mosquitoes, in: Australian Mosquito Control Manual, revised edition; p. G0-G12
- 4. Ritchie S. Montgomery B. (2005): Efficacy of chlorine against eggs of the mosquito *Aedes aegypti*. Unpublished.
- 5. Sherman C. Fernandez EA. Chan AS. Lozano RC. Leontsini E. Winch PJ. (1998): *La Untadita*: A procedure for maintaining washbasins and drums free of *Aedes aegypti* based on modifications of existing practices. American Journal of Tropical Medicine Hygiene, 58(2): 257-262.

Figure 70: Summary of procedures for the treatment of the inner surfaces of water receptacles found on overseas vessels

Step 1 Collect larvae, pupae and pupal skins from water storage receptacle.

Step 2 Empty, siphon or pump contents of receptacle into sea.

Step 3

Prepare chemical treatment mixture in a 5-litre pressure sprayer. The mixture includes 4.8 litres of liquid chlorine solution that has at least a 10% active ingredient, and 200ml of liquid concentrate detergent. Mix well by agitation of sprayer

Step 4

Spray all interior surfaces of receptacle liberally to run off with chlorine/detergent mixture using a 5litre pressure spray. Seal receptacle and let stand for 30 minutes.

Step 5

Open receptacle and re-spray all interior surfaces of receptacle liberally to run off with chlorine/detergent mixture. Seal receptacle and let stand for another 30 minutes.

Step 6

Leave receptacle sealed for another twenty-four (24) hours, allowing eggs to incubate. Rinse and wash receptacle.

Figure 71: Summary of onshore of receptacle handling and surveillance measures

1: Vessel or Cargo Receptacles

Remove receptacles from overseas vessels, tag with an information label, and store covered or under cover away from rainfall.

Tagged receptacles from vessels are treated by fumigation or chlorination methods.

Ensure the site is clear of other potential water-holding receptacles or if present, ensure they are regularly treated with a residual insecticide application.

2: <u>Receptacle survey</u>

Surveys in the 400m zone should be conducted on a routine basis and can be combined with treatment rounds.

3: Adult Surveillance

Adult mosquito trapping is set and collected weekly.

Ovitrap/s are set and collected weekly to fortnightly. If there are only early larval instars and no 4th instar larvae present, all larvae need to be reared further to the 4th instar.

Sentinel tyres, if used, are inspected and serviced on a weekly to fortnightly basis depending on temperature and the amount of shade present. Methoprene pellets are added to sentinel tyres on monthly servicing. Re-sampling for 4th instar larvae is required if only early instars are initially present.

Figure 72: Types of ovitraps



Table 22: Chlorination/Detergent solution ratio mix for the surface treatment of emptied or dryreceptacles (from Lamche and Whelan 2002, Shortus and Whelan 2005)

Chlorine Formulation	Total Volume	Water Volume to make Solution	Amount of Pool Chlorine/Bleach	Active Ingredient	Detergent Volume Required	Concentration of Chlorine in Spray	
Granular pool							
chlorine	1 L	960 ml	154 g	100 g	40 ml	10%	
(650g/Kg calcium	5 L	4.8 L	770 g	500 g	200 ml	10%	
hypochlorite)	10 L	9.6 L	1.54 Kg	1 Kg	400 ml	10%	
Granular pool							
chlorine	1 L	960 ml	143 g	100 g	40 ml	10%	
(700g/kg calcium	5 L	4.8 L	715 g	500 g	200 ml	10%	
hypochlorite)	10 L	9.6 L	1.43 Kg	1 Kg	400 ml	10%	
Liquid pool							
chlorine	1 L	293 ml	667 g ≈ 667 ml	100 g	40 ml	10%	
(150 g/L	5 L	1.46 L	3.35 Kg ≈ 3.35 L	500 g	200 ml	10%	
benzalkonium chloride)	10 L	2.9 L	6.67 Kg ≈ 6.67 L	1 kg	400 ml	10%	
Liquid bleach	11	Llao poot		105 a	40 ml	100/	
(125g/L sodium	1 L 5 I	Use neat		120 y	40 ml	1270 109/	
hypochlorite)	101	Use neal	4.0 L 0.6 l	020 y 1 25 Ka	200 ml	12% 10%	
		USE neal	3.0 L	r.zu ry	400 111	1 2 /0	

Table 23: Chlorination solution for treatment of receptacles holding water (\geq 30 minutes) (from Lamche and Whelan 2002, Shortus and Whelan 2005)

Chlorine Formulation	Water Volume in Receptacle	Amount of Chlorine/Bleach	Pool Active Ingredient	Concentration of Chlorine in Water Receptacle
Granular pool chlorine	20 L	100 g	65 g	0.33%
(650g/Kg calcium	10 L	50 g	32.5 g	0.33%
hypochlorite)	100 L	500 g	325 g	0.33%
Granular pool chlorine	20 L	100 g	70 g	0.35%
(700g/Kg calcium	10 L	50 g	35 g	0.35%
hypochlorite)	100 L	500 g	350 g	0.35%
Liquid pool chlorine	5 L	100 g ≈ 100 ml	15 g	0.30%
(150 g/Kg	10 L	200 g ≈ 200 ml	30 g	0.30%
benzalkonium chloride)	100 L	2 kg ≈ 2 L	300 g	0.30%
Liquid bleach	1.2 L	100 ml	4%	0.33%
(4 % sodium	10 L	833 ml	4%	0.33%
hypochlorite)	100 L	8.33 L	4%	0.33%

31. CLIMATE CHANGE AND MOSQUITOES / DISEASE IN AUSTRALIA: REAL CONCERN OR CONUNDRUM REVEALED?

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SUMMARY

- It has been widely stated that global climate is changing and these changes will bring increases in temperature, rainfall and sea-level.
- These environmental factors can increase the distribution and seasonality of some mosquitoes, and mosquito borne disease may be increased. However, these climate changes will not be uniform across Australia.
- The effects of warming and increased precipitation will have different effects on different mosquitoes, and the vertebrate hosts of the human pathogens, and under various local conditions virus transmission may be either enhanced or inhibited.
- Nationally, some regions can expect conditions that will bring a greater risk of mosquito transmission of disease-causing agents, while others will experience seasonal changes that are unfavourable for some mosquitoes and these pathogens.
- For WA, current CSIRO projections indicate increases in temperatures and tide levels in general, increased cyclones in the northwest and decreased winter rain in the southwest.
- Overall, geographical distribution of Murray Valley encephalitis virus activity in tropical regions may extend further southward, and seasonal distribution of Ross River virus activity in temperate regions may change.
- The natural cycles involving arboviruses are complex, the predictive value of the climate models is likely to change, and local regional scenarios in Australia will certainly vary.

INTRODUCTION

While global climate has fluctuated widely over the earth's history, from around the time of settled agriculture in the current inter-glacial period, humans have enjoyed a relatively stable climate. However, in recent decades significant changes have been observed and linked to global warming, consistent with changes in composition of the atmosphere induced by humans. Although aspects of these changes may be influenced by natural climatic long-term variability, it is now widely and reputably believed that the so-called 'greenhouse effect' is a significant influence in regional climate change. The most fundamental change has been an increase in temperature, affecting not just atmospheric conditions but also marine conditions that result in increased melting of the polar ice-caps and a consequent rise in sea levels.

Since the 1970s, the observed rate of global warming has been 0.15° C per decade, and current international projections are for a globally averaged warming of 1.4 to 5.8° C by 2100 relative to 1990, with a projected rate of warming of 0.1 to 0.5° C per decade. Associated with this warming, sea-level has risen over the 20th century by 1 to 2 cm per decade, and is projected to rise by 9 to 88 cm by 2100, or 0.8 to 8.0 cm per decade. All this is in addition to the 0.6° C of warming and 10-20cm of sea level rise that occurred over the 20th century prior to 1990; but the degree of change will not be uniform around the globe, and various countries

and regions must be assessed separately. This has been done in recent times with scenario simulations from climate models.

CLIMATE CHANGE FOR AUSTRALIA

Various models of climate change have been developed during the past two decades and there remains much uncertainty in this area of research. The concept of greenhouse gas emissions increasing temperatures, rainfall and ocean levels led to the development of relatively simple climate models during the 1990s, but in recent years more complicated versions have been adopted to take into account various other chemical and physical factors.

While all current global climate models show some degree of future warming in response to increases in concentration of greenhouse gases in the atmosphere, the sensitivity of the various models varies considerably. The general result from the newer model-generated scenarios is that previously predicted increases in temperatures and sea-level have been reduced considerably, and patterns of rainfall changes vary within and between models. This is not the place to provide an analysis of the various predictions from the different models, and so only the most recent scenarios provided by the Climate Impact Group of the CSIRO Division of Marine and Atmospheric Research, which has used the most scenarios of the Intergovernmental Panel on Climate Change (IPCC), are provided.

The average surface air temperature of Australia increased by 0.7°C over the past century (since 1910), with marked declines in rainfall (particularly along the east and west coasts) over the past 50 years, and CSIRO projections are such that if all Green House Gas (GHG) emissions ceased today, we would be committed to an additional warming of 0.2-1.0°C by the end of this century. However, GHG emissions are expected to increase (at least in the short term), since the momentum of the world's fossil fuel economy precludes the elimination of GHG emissions over the short-term. Over most of Australia, annual average temperatures are projected to increase; inland areas are likely to warm faster than the global average, while coastal areas and the tropics will warm at around the global average. Average precipitation in southwest and southeast Australia is projected to decline in future decades and there will be more droughts in areas where rainfall decreases. Some regions, such as central and northwestern Australia, may experience increases in rainfall and more extremely wet years. Coastlines will experience erosion and inundation from the increase in global sea level.

The overall spatial distribution of projected changes in annual temperature and precipitation for 2030 and 2070 are shown in Figure 73, while seasonal distributions are shown in Figure 74 and Figure 75.

TEMPERATURE

By 2030, annual average temperatures will be 0.4 to $2.0 \,^{\circ}$ C higher over most of Australia, with slightly less warming in some coastal areas and Tasmania, and potential for greater warming in the north-west. By 2070, annual average temperatures will have increased by 1.0 to $6.0 \,^{\circ}$ C over most of Australia, with spatial variation similar to 2030. The range of warming will be greatest in spring and least in winter. In the north-west, the greatest potential warming will occur in summer. Models indicate that future increases in daily maximum and minimum temperature will be similar to the changes in average temperature, contrasting with the greater increase in minima than maxima observed over Australia in the 20th century.

RAINFALL

Projected annual rainfall will tend towards a decrease in the southwest (-20% to +5% by 2030 and -60% to +10% by 2070), and in parts of the south-east and Queensland (-10% to +5% by

2030 and -35% to +10% by 2070). In some other areas, including much of eastern Australia, projected ranges are less certain (-10% to +10% by 2030 and -35% to +35% by 2070), while the ranges for the tropical north (-5% to +5% by 2030 and -10% to +10% by 2070) represent little change from current conditions.

In summer and autumn, projected rainfall ranges for most locations are relatively stable (-10% to +10% by 2030 and -35% to +35% by 2070), or will tend towards increase (-10% to +20% by 2030 and -35% to +60% by 2070) with the latter occurring mainly in parts of southern inland Australia in summer and inland areas in autumn. In some parts of northern and eastern Australia in summer and inland Australia in autumn, there is a projection for wetter conditions (-5% to +10% by 2030 and -10% to +35% by 2070). However, for the far southeast of the continent and Tasmania, projected rainfall will tend to decrease in both seasons (-10% to +5% by 2030 and -35% to +10% by 2070).

In winter and spring, most locations will tend towards decreased rainfall (-10% to +5% by 2030 and -35% to +10% by 2070). Projected decreases are stronger in the south-west (-20% to +5% by 2030 and -60% to +10% by 2070), while Tasmania will tend toward increases in winter (-5% to +20% by 2030 and -10% to +60% by 2070).

In areas where rainfall increases, there is likely to be more extremely wet years, and where rainfall decreases there is likely to be more dry spells. Most models simulate an increase in extreme daily rainfall leading to more frequent heavy rainfall events. This may occur even where rainfall decreases although reductions in extreme rainfall may occur where rainfall declines significantly. Increases in extreme daily rainfall are likely to be associated with increased flooding. Changes in average rainfall may affect the frequency of wet and dry seasons. To illustrate this effect, the CSIRO regional model for south-west New South Wales indicates that summers are likely to become about 15% wetter and springs about 10% drier by 2030, and the number of extremely dry springs and extremely wet summers may more than double after 2020.

EVAPORATION AND MOISTURE BALANCE

Higher temperatures are likely to increase evaporation. CSIRO projects increases in all seasons, ranging (annually averaged) from 0 to 8% per degree of global warming over most of Australia, and up to 12% over the eastern highlands and Tasmania. The increases tend to be larger where there is a corresponding decrease in rainfall. Average decreases in annual water balance (difference between evaporation and rainfall) range from about 40 to 120mm per degree of global warming. This represents decreases of 15 to 160mm by 2030 and 40 to 500mm by 2070. These decreases in moisture balance mean greater moisture stress for Australia. The simulated changes show the greatest consistency in spring, with decreases greatest over eastern Australia and generally ranging between 20 and 100mm per degree of global warming, while decreases in the western half of Australia range between 0 and 60mm.

FOR WESTERN AUSTRALIA

TEMPERATURES

Temperatures in WA have increased by about 0.8°C since 1910 but mostly since 1950, and daily minimums have increased more than the maximums. Warming has been greatest in the southwest and least in the northeast, and has been greatest in winter/spring and least in summer.

RAINFALL

Rainfall in the southwest has shifted to consistently drier conditions since the mid-1970s (with a decline of about 10%), associated with a shift in the global atmospheric circulation. The decline has been greatest in autumn and early winter (with a slight increase in summer), and simulations of future climate with enhanced greenhouse gases show a similar pattern of drier conditions for the southwest. This decrease has led to a stream flow reduction of about 50% and some ecosystems such as wetlands are under pressure. For the northwest, stronger tropical cyclones are projected, and surface and ground waters may be increased in some areas.

TIDE

Tide data from Fremantle have shown that sea level has increased almost 20 cm at a rate of 1.54mm p.a. since 1897, and has increased at almost 3 times the global rate since 1991. The rise is expected to continue through the 21st century (by up to 0.30m by 2040 and 0.88m by 2100), resulting in extensive changes to saline wetlands and erosion of beaches by up to 30m by 2040.

Overall, all the scenarios in current CSIRO projections indicate further increases in temperatures in WA in general, and further decreases in winter rain across the southwest region of WA.



Figure 73: Spatial distribution of projected annual changes in Australian precipitation (left) & temperature (right) in 2030 & 2070 (Figures from Kevin Hennessy, CSIRO)

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Figure 74: Average seasonal and annual warming ranges (°C) for around 2030 and 2070 relative to 1990. The coloured bars show ranges of change for areas with corresponding colours in the maps (Figures from Kevin Hennessy, CSIRO).



Figure 75: Ranges of average seasonal and annual rainfall change (%) for around 2030 and 2070 relative to 1990. The coloured bars show ranges of change for areas with corresponding colours in the maps. Ranges are not given for areas with seasonally low rainfall because percentage changes in rainfall cannot be as reliably calculated or applied in such regions (Figures from Kevin Hennessy, CSIRO).

IMPACT OF CLIMATE CHANGE ON AUSTRALIAN MOSQUITOES AND MOSQUITO-BORNE PATHOGENS

Mosquitoes require water for larval development and adult survival. Their progress through the immature stages is regulated by temperature, and the geographic distribution and seasonal activity of many species is determined by temperature. Additionally, the development (incubation period) of disease-causing pathogens in mosquitoes is influenced by temperature, and the capacity of individual mosquitoes to survive the incubation period and go on to transmit the pathogens is dependent on favourable temperature and humidity conditions.

The 'global-warming' scenarios of climate change have a capacity to produce more freshwater habitat through increased rainfall, and longer periods of seasonal activity for mosquitoes in both inland and coastal areas with increasing temperature, and sea-level rises are likely to produce more saline habitat through gradual inundation of coastal regions. Such scenarios have fuelled speculation that mosquito vectors and mosquito-borne pathogens will increase in distribution and activity, and give rise to increases in the prevalence and incidence of human disease as a result. In a general sense, the proposition that climate change will influence mosquito populations and pathogen activity should be seen as having some validity, and it is likely there will be some increases, but with which mosquitoes and which pathogens and where? Additionally, the influence of human lifestyle factors and public health services must also be considered when contemplating increased contact between humans and mosquitoes, and increased risk of infection with mosquito-borne pathogens.

There have been predictions that malaria will invade much of Australia with climate warming; however, when based simply on climatological data, and taking no account of parasitological, medical, epidemiological and social factors, such claims are absurd. Climate warming may bring an increased distribution of the most important malaria vector in northern Australia, *Anopheles farauti*, but it will be a relatively minor southerly extension of little significance and, anyway, there are alternative vectors already in southern Australia where malaria has been transmitted in the past (presumably by *Anopheles annulipes*) as far south as Perth in the west and Melbourne in the east. There may also be a concomitant slight extension of the favourable season for transmission, but there is little likelihood that malaria will become reestablished in Australia for reasons more associated with human demographics and public health responses than with climate and vector factors.

Additionally, propositions that the most southern Australian mainland cities such as Melbourne could, with a slight increase in temperature, be expected to begin experiencing epidemics of dengue, should not be entertained without considering that, even with some slight warming that will favour an increase in the distribution of *Aedes aegypti*, currently the only vector in mainland Australia, it is most unlikely there will be any substantial extension into southern Australia (where the species once existed but long ago disappeared). Of greater concern is the potential for reintroduction, establishment and dispersal of *Aedes aegypti* in currently climatically favourable regions of the Northern Territory and northern Western Australia where it previously was widely distributed, and also the introduction to mainland Australia of the secondary vector *Aedes albopictus* (recently established in the Torres Strait islands) that is more tolerant of cool climate conditions and could become established in the southern regions/capitals of Australia and provide a vector where none currently exists.

Although pronouncements on the extension of vector-borne diseases in Australia may be wellintentioned and aimed at raising awareness of these important issues, they are often promulgated by persons ignorant of the complexities of vector-borne diseases, and their simplistic nature can render them counterproductive to creating the required public health concern. Malaria in particular (but also dengue), is a relatively poor 'target' for concern with respect to Australia, and the 'warming-climate-bringing-more-mosquito-borne-disease' proposition can be more soundly addressed with respect to some of the local arboviruses, which are the most important of the mosquito-borne pathogens in this country although they are varied and active variously in different parts of Australia.

Of the Australian arboviruses most important in human infection, Murray Valley Encephalitis virus (MVEV) and Kunjin virus (KUNV) cause life-threatening encephalitis, and Ross River virus (RRV) and Barmah Forest virus (BFV) cause a debilitating polyarthritis. These viruses are zoonoses, diseases in humans caused by pathogens transmitted from other animals, and are associated primarily with rural areas - inland or coastal. With respect to vertebrate hosts of these viruses, wetland birds (e.g. waders such as herons) are the natural reservoir hosts of MVEV and KUNV, and RRV is thought to be associated primarily with native marsupials, but the reservoir hosts for BFV are not known and mammals and/or birds may be involved.

Outbreaks of arboviruses typically are driven by local increases in mosquito populations. Areas of increasing arbovirus activity have noticeably more wetland habitat and concomitantly larger mosquito populations than other areas; often there is an obvious association with local enhancements of wetlands of various types, and the movement of people into closer contact with wetlands and their mosquitoes. Notwithstanding the above-mentioned outline of the vector - host - virus associations, the field situations are rarely simple. The epidemiology of RRV, in particular, differs in different regions of Australia. Moreover, the epidemiology and clinical severity of different outbreaks may vary within a geographic region, and between seasons, because of the involvement of different species of mosquito and strain of virus in different times.

In coastal regions of Australia, from the south coast of NSW northwards, around the top of the continent and down to Perth in WA, the saltmarsh mosquito, *Aedes vigilax,* is the principal vector of RRV. In VIC and in WA south of Perth, *Aedes camptorhynchus* is the principal saltmarsh species and replaces *Ae. vigilax* as the major pest species and coastal vector. However, it is not simply a matter of one species replacing the other; the seasonal activity of the two species is quite different, with *Ae. camptorhynchus* more active in the cooler months leading up to summer and *Ae. vigilax* peaking in abundance in mid- to late-summer. South Australia has both saltmarsh species, with seasonal activity similar to that just mentioned. In general, seasonal activity of arboviruses typically is later further south when there is a common vector, because of temperature influences, but activity of RRV in southwest WA and eastern VIC actually precedes that in neighbouring northern regions because of the different seasonal patterns of the different vectors.

In inland areas, there is a general seasonal progression of RRV activity southwards as rising temperature in spring brings new season activity of various mosquitoes and summer progresses the major vector species *Culex annulirostris*. However, early season rains can mean that *Aedes* floodwater species may be responsible for springtime transmission following localised rainfall or regional flooding, prior to the expected *Culex* transmission season in summer and autumn. Summer activity typically is driven by increasing populations of *Cx. annulirostris*, but these may not eventuate if the spring surface water does not persist and there is no summer rainfall.

In simple terms, climate circumscribes the distribution of mosquito species (and thus mosquito-borne diseases where there are specific vector-virus associations), while weather influences the timing of vector populations surges and disease outbreaks. Rainfall and temperature have the potential to enhance vector abundance and extend distribution, increase vector development, reproductive and biting rates, shorten pathogen incubation period, and encourage adult longevity. All these factors are important in the dynamics of transmission and can promote infective potential and produce more effective and more frequent transmission. Wind is also important and changes in wind direction, velocity and frequency will also have an impact on adult mosquito populations, affecting the dispersal, survival, and other aspects of the general behaviour of many species.

RAINFALL CHANGES

In the most basic terms, production of mosquitoes is determined by the availability of suitable and sufficient larval habitat, and for freshwater species this is dependent on rainfall (either local or more remote in the drainage basin) for its creation and persistence. Saline habitats are likewise dependent on local tide movements, but habitats in saline environments can be renewed by rainfall as well as tide, particularly the depressions higher up on the marsh which are usually filled only by the highest tides.

The projected general decreases in winter rainfall and increases in summer rainfall will provide for a variety of potential effects dependent on season, region and mosquito species. A general decrease in winter rainfall, as predicted by models for most of Australia, will have a deleterious effect on winter and spring mosquitoes through reduction in habitat, and the reduced humidity may also impact negatively on overwintering adults of many *Culex* species. This might have a direct negative influence on arbovirus transmission in some areas, e.g. with Ae. camptorhynchus in the winter and spring in the southwest (WA), and in spring in the southeast (NSW/VIC). However, the projected increased rainfall in summer in those regions would enhance different mosquito species and could bring a seasonal shift in virus transmission, particularly if temperature and sea-level/tidal movements have a supportive influence. Although it is difficult to extrapolate from meagre field evidence, if vertical (transovarial) transmission by *Aedes* species in winter/spring is an important strategy for virus survival, then reduction of mosquito populations by decreasing winter rainfall may delay or perhaps preclude virus activity in some regions so affected. Thus, populations of floodwater Aedes species may be adversely affected in drier areas of western NSW and northwestern VIC, and in parts of WA, where such species are thought to be involved in maintenance cycles of virus.

Decrease in summer rainfall throughout Australia would reduce availability of habitat and mosquito populations, but current models indicate possible increases in summer rainfall in the southwest of WA and much of eastern Australia, particularly the southeast. This would increase availability of mosquito habitat in areas where RRV and BFV have been epidemic. Some models indicate a general increase in summer rainfall throughout Australia, and this will increase available habitat, particularly for freshwater marsh species such as *Cx. annulirostris* and *Coquillettidia linealis*, two important vectors in inland Australia.

Apart from increased rainfall, it is predicted there will be increases in heavier rainfall events and, in this context, it needs to be borne in mind that often there can be too much rain for some mosquito species. Larvae of *Culex* species and *Anopheles* species are often flushed away from protected areas in their habitats by flooding from heavy or continuous rain and, in tropical areas in particular, such extensive rainfall can delay the buildup of some species until late in the season and thus delay transmission. However, this varies with species and type of habitat, and some floodwater *Aedes* species in semi-arid tropical areas, e.g. *Ae. normanensis*, which is associated with a number of viruses, may have their populations enhanced by excessive rainfall resulting in a significant increase in risk of local transmission. As a consequence, populations of *Cx. annulirostris* that follow the floodwater *Aedes* species, utilising the resulting vegetated habitat, will be greater than otherwise expected and any virus activity initiated by the *Aedes* species.

For northern WA, stronger tropical cyclones are projected that could bring a southward extension of MVEV and KUNV, which are normally annually active only in the Kimberley region. Outbreaks of RRV are also associated with unusually heavy rainfall, implicating floodwater *Aedes* species such as *Ae. normanensis* as vectors, although *Cx. annulirostris* also becomes involved when the surface water persists. In southwestern WA, outbreaks of RRV have also been associated with extensive rain providing extraordinary opportunities for mosquito breeding in both saltmarsh (with no tidal involvement) and freshwater habitats, with

freshwater mosquitoes such as *Cx. annulirostris* and *Cq. linealis* supplementing the saltmarsh *Ae. camptorhynchus* as vectors.

Mosquitoes breeding in saltmarsh habitats in eastern Australia can also have their populations enhanced by summer rainfall inasmuch as depressions usually filled by high tides on a monthly basis may be renewed more often and produce more generations. This was seen to great effect in December 1994 through February 1995 at Batemans Bay on the south coast of NSW with the extraordinary *Ae. vigilax* populations resulting in a major outbreak of BFV. In eastern VIC, rainfall can produce large numbers of *Ae. camptorhynchus* from brackish habitats also affected by tidal influence, and a similar situation exists in eastern TAS with *Ae. camptorhynchus* and *Ae. flavifrons*.

Overall, increased rainfall will provide more larval habitat for many species and potentially provide for increased populations. However, stabilisation of some habitats will provide for increases in predator populations that will modulate mosquito populations to a greater or lesser extent. Increases in humidity and cloud cover associated with increased rainfall will enhance mosquito survival and bring warmer nights in southern regions, increasing the risk of virus transmission and extending the breeding season, respectively.

TEMPERATURE CHANGES

Distribution of mosquito species can be limited by temperatures, and thus tropical vectors could be expected to move further into currently cooler areas (provided appropriate habitat is present) when climate change brings a general increase in ambient temperature. Mosquitoes respond to local temperature increases in various ways; within limits, higher temperatures mean more rapid development for larval populations, and shorter times between bloodmeals, quicker incubation times for virus infections, and shorter life spans for adults, although the latter is modulated by higher humidity.

As an example of potential effects, the major inland vector of arboviruses in southeastern Australia, *Cx. annulirostris*, overwinters as inactive adults and becomes seasonally active when the average temperature exceeds 17.5°C, usually in late October in southern NSW and northern VIC, with peak populations occurring after average temperatures reach 25°C. With climate warming, the species might be expected to commence activity and reach high levels of abundance earlier, and maintain them longer; thus, the season of arbovirus activity could be initiated earlier and be prolonged, with potentially greater levels of transmission. However, it is likely that photoperiod may also influence the autumn retreat and spring emergence, and the details of this are unknown, but as photoperiod will not change with warming of the climate, the situation is more complicated than it might appear.

With respect to effects of increased temperature on adult mosquitoes, it should be remembered that a rise in overall ambient temperature dose not necessarily mean a rise in the temperature experienced by the individual mosquito. In general, adult mosquitoes select microenvironments for resting between feeding and oviposition, and these are usually cooler and more humid than the ambient. Such places may be more difficult to find with increasing temperatures, but if rainfall also increases and humidity stays high there is a compensation which protects against desiccation. Rate of extrinsic incubation of arboviruses will typically be expected to increase with temperature, although it is known to decrease at higher temperatures for some viruses. Feeding and oviposition sites may be created closer to resting sites, and reduce the risk of predation for adults - this would also be the case with increased rainfall. Changed conditions may increase or decrease predator populations; increased temperatures reducing larval habitats may promote pathogen infection in confined circumstances. Increased temperature will increase development rate, but produce smaller adults; these may be more prone to desiccation, but may feed more frequently to obtain sufficient blood for egg development and thus increase the incidence of pathogen, and they

may lay eggs more frequently but in smaller batches because of their reduced size. The relation between climate and mosquitoes and disease is thus complex and not one of physical climatology alone.

The increases in temperatures 'predicted' with climate change will, in principle, provide better conditions for some species in some circumstances. However, populations in many regions and localities will require concomitant increase in rainfall to ensure larval habitat and to maintain humidity for adult survival. Providing there is maintenance of suitable hosts nearby, the distribution and seasonal activity of some vectors may shift a little.

SEA-LEVEL CHANGES

The most important mosquito environment impacted by sea-level changes is the mangrove swamp, mudflat and saltmarsh system of the estuary. With sea-level rise, the extent of tidal flooding on high-tide flats will increase in the earliest stages and the networks of small tidal creeks will expand into estuarine floodplains. Later, tidal flow in the main channels will increase and the channels will probably widen. As creek networks become more extensive, freshwater plant communities will be killed by invading brackish waters and saline mudflats formed, flooded by highest spring tides. Further effects, such as erosion and sedimentation, will be influenced by the relative dominance of flood tides over ebb tides, reflecting tidal velocities and drainage basins. In both northern and southern WA, saltmarsh habitats may be increased significantly, expanding habitat for *Ae. camptorhynchus* and *Ae. vigilax*, as relatively large freshwater areas may be affected by saltwater intrusion, resulting in an extension of estuarine wetlands at the expense of the freshwater wetlands and floodplains. Concomitant increased tropical rainfall and wet season extension will enhance both the spread and productivity of mangrove and saltmarsh communities.

Given that mosquito habitat in such environments is usually comprised of depressions on mudflats or saltmarshes that are filled by the highest tides of each month, such habitat may be inundated with saline water as sea-level rises. Ultimately, the extent of resultant mosquito habitat will depend on topography and ability of vegetation to persist or establish under the new conditions. Depending on location and local circumstances, mosquito habitat may be destroyed in steeper topography, and habitat may be created in flatter topography.

The 1988/89 outbreak of RRV in southwestern WA was associated with higher (5.5cm) than average sea-levels; these gave higher daily tide levels which, accentuated by a series of southwesterly weather patterns, resulted in inundation of large areas of saltmarsh normally dry during summer months, and subsequent heavy breeding of *Ae. camptorhynchus*. The 1995 outbreak of BFV on the south coast of New South Wales was associated with very large populations of *Ae. vigilax*, generated from extensive flooded saltmarsh habitat resulting from a series of very high tides and subsequent rainfall that maintained the breeding environment.

Both these instances reflect the expansion of habitat, and the generation of extraordinary mosquito populations, which may occur in many areas with slight increases in sea-levels and be enhanced by increases in rainfall or major rain events. More constant inundation of brackish habitats would enhance the populations of *Culex sitiens* on the coasts of NSW, QLD, NT and WA, as the temporary pools persisted and favoured this species over *Ae. vigilax*. Under such conditions, *Cx. sitiens*, which currently is thought not to be a major vector of RRV, could become more important and displace *Ae. vigilax* as the principal concern in some coastal regions.

IMPACT ON VERTEBRATE HOSTS

The impact of climate changes on the birds and mammals that have been incriminated as hosts of the arboviruses, and on their role in the complex cycles of virus activity, must also be

recognised. Climate changes will invariably impact indirectly on virus activity through direct impact on the vertebrate hosts. Again, no simple relationship should be expected; however, while increased transmission of arboviruses in the various regions of Australia would lead to increased infection of native vertebrates and possibly a greater overflow to humans, there may be a longer-term influence of herd immunity in animal hosts that could be a modifying factor for ongoing seasonal activity, and the complex nature of the cycles will continue.

CONCLUSIONS

Although a vector presently confined to warmer regions may spread into currently cooler areas as these become warmer, it does not follow that pathogens they carry will necessarily cause disease in the newly invaded areas at the levels from whence they came - although epidemics may ensue if the pathogen can persist at low levels. Appropriate vectors and reservoir hosts are essential features in maintaining the cycles of mosquito-borne pathogens, under favourable (to <u>each</u> of the 'player's') environmental conditions.

Also, where there are a number of potential vectors currently extant in one region, and where various species are known to carry a virus from time to time, as occurs in both inland and coastal areas of Australia, the effects of warming and increased precipitation will have different effects on different species, and it must be considered that under various local conditions virus transmission may be either enhanced or inhibited.

The seasonal activity of the various species of mosquitoes in most areas of Australia is now relatively well known, particularly those species of importance as vectors of arboviruses. The potential for their seasons to be influenced by temperature can be predicted in the simplest sense. However, the effect of climate change, involving rainfall and sea-level as well as temperature changes, on the distribution and abundance of these species, and on their involvement in arbovirus cycles which involve different vertebrate hosts also likely to be affected by climatic factors, is definitely not simple to predict.

Additionally, the impact of societal issues such as the increased use of domestic airconditioning (promoting indoor activities) as the climate provides warmer weather, the establishment and enhancement of government mosquito management programs more widely in Australia, the development of more effective repellents and other personal protection measures, and greater education of the general public towards avoidance of risk can all contribute to countering increased mosquito activity and associated disease.

SUGGESTED READING

- Bryan JH, Foley DH, Sutherst RW. Malaria transmission and climate change in Australia. *Medical Journal of Australia* 1996; 164: 345-347.
- CSIRO. Climate Projections for Australia.

http://www.cmar.csiro.au/e-print/open/projections2001.pdf

CSIRO. Marine and Atmospheric Research.

http://www.cmar.csiro.au/ar/information/climatechange.html

- Githeko AK, Lindsay SW, Confalonieri UE, Patz JA. Climate change and vector-borne diseases: a regional analysis. *Bulletin of the World Health Organization* 2000; 78: 1136-1147.
- Hughes L. Climate change and Australia: Trends, projections and impacts. *Austral Ecology* 2003; 28: 423-443.
- IAWG (Impacts and Adaptation Working Group). *Climate Change Impacts for Australia*, CSIRO. 2001: 8pp.
- IOIC (Indian Ocean Climate Initiative). Publications on Western Australia climate projections. http://www.ioci.org.au/
- IPCC (Intergovernmental Panel on Climate Change) Data Distribution Centre: <u>http://ipcc-ddc.cru.uea.ac.uk/</u>
- IPCC (Intergovernmental Panel on Climate Change). Climate Change 2000: The Science of Climate Change. Summary for Policymakers and Technical Summary of Working Group. Cambridge University Press. 2001. 98pp.
- Jetten TH, Focks DA. Potential changes in the distribution of dengue transmission under climate warming. *American Journal of Tropical Medicine and Hygiene* 1997; 57: 285-297.
- Liehne PFS. Climatic influences on mosquito-borne diseases in Australia. In: Pearman GI, editor. *Greenhouse - Planning for Climate Change*. Leiden: CSIRO/EJ Brill, 1988; 624-637.
- Lindsay MD, Mackenzie JS. Vector-borne viral diseases and climate change in the Australasian region: major concerns and the public health response. In: Curson P, Guest C, Jackson E, editors. *Climate Change and Human Health in the Asia-Pacific.* Canberra: Australian Medical Association / Greenpeace International, 1997; 47-62.
- Mackenzie JS, Lindsay MD, Broom AK. Climate changes and vector-borne diseases: potential consequences for human health. In: Ewan CE, Calvert GD, Bryant EA, Garrick JA, editors. *Health in the Greenhouse. The Medical and Environmental Health Effects of Global Climate Change.* Canberra: Australian Government Publishing Service, 1993; 229-234.
- Preston BL, Jones RN. Climate change impacts on Australia and the benefits of early action to reduce Global Greenhouse Gas Emissions. CSIRO. 2006: 41pp.
- Reeves WC, Hardy JL, Reisen WK, Milby MM. Potential effects of global warming on mosquito-borne arboviruses. *Journal of Medical Entomology* 1994; 31: 323-332.
- Reiter P. Climate change and mosquito-borne disease. *Environmental Health Perspectives* 2001; 109: 141-161.
- Rogers DJ, Packer MJ. Vector-borne diseases, models, and global change. *Lancet* 1993; 342: 1282-1284.
- Russell, RC. Mosquito-borne arboviruses in Australia: the current scene and implications of climate change for human health. *International Journal for Parasitology* 1998; 28: 955-969.
- Sutherst RW. Global change and human vulnerability to vector-borne diseases. Clinical Microbiology Reviews 2004; 17: 136-173.
- Taubes G. Apocalypse Not. Science 1997; 278: 1004-1006.

32. BITING MIDGES OR 'SANDFLIES' IN THE NORTHERN TERRITORY

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Centre For Disease Control Northern Territory Department of Health Adapted from 'Biting Midges Or "Sand Flies" in the NT' 2003' PI Whelan. Northern Territory Disease Control Bulletin. 10:3:1-9.

INTRODUCTION

Biting midges are small blood sucking flies in the family *Ceratopogonidae*. They are commonly referred to as "sandflies" in northern Australia. The term "sand fly" is a misused term for a number of families of small biting flies. This includes the true sandflies (Family *Psychodidae*) which are not pests of humans in Australia, as well as black flies (Family *Simulidae*) which are serious pests in the inland areas of Qld and NSW following flooding, and the biting midges (Family *Ceratopogonidae*).¹





Figure 76: Simulium – a female "Black Fly"

Source: Insects of Medical Importance. British Museum, 1956

Biting midges are the major midge pest problem in Northern Australia.² A number of members of this family bite people in the Northern Territory. They include three species in the genus *Lasiohelea,* which are found biting in small numbers in shaded areas in or near dense forests during the day. A species of *Styloconops* is found in small numbers biting and swarming around the head on open sandy beaches during the day. The members of the *Culicoides* genus are more common, with many species and a wide range of breeding sites and biting habits.



Figure 78: Culicoides – a female "Biting Midge" Source: Entomology for Students of Medicine. Blackwell Scientific Ltd. 1962



Figure 79: Heads of *Ceratopogonidae* (Biting Midges) (a) *Lasiohelea* (b) *Culicoides* (c) *Styloconops* Source: Atlas of Common Queensland Mosquitoes. Queensland Institute of Medical Research, 1982

Thirty-three species of *Culicoides* have been recorded from the Darwin area.³ The *Culicoides* species include some species that don't bite vertebrates, some which preferentially bite cattle and other domestic animals, and the few species that are serious pests of people. The breeding sites include fresh water margins and cattle dung. Most of the serious human pest species breed in tidal and estuarine sites.

Culicoides midges are small, robust flies, approximately 1 mm in length with two wings usually showing a pattern of clear patches on a grey background. They have a short, forward directing proboscis or mouthparts for piercing skin and sucking blood.

Two species, *Culicoides flumineus* and *Culicoides* species near *subimmaculatus* can be severe human pests in mangrove areas across the Top End of the NT, with the latter also found in high pest numbers up to 1.5 km from certain mangrove areas.

One species, *Culicoides ornatus,* sometimes referred to as the "mangrove midge", is found in association with mangroves across northern Australia, and is usually responsible for severe biting midge pest problems near the coast. This species is a major pest because it occurs in very high numbers and has a habit of invading nearby residential or recreational areas.

Culicoides ornatus is becoming an increasing problem across northern Australia due to urban development encroaching nearer to their major breeding places.^{5,6,7} They can impose serious restrictions on outdoor activities within flight range of their mangrove breeding sites due to the extremely annoying and painful bites, and to the discomforting after effects of the bites.

BITES OF BITING MIDGES

It is only the female midges that bite. Biting midges do not transmit disease to humans in Australia. Their main human medical importance is as a biting pest.

Midges must take a blood meal for their eggs to mature. They do not, as is sometimes believed, urinate on people to cause discomfort. In the process of biting and sucking blood, they inject a salivary secretion that produces a skin reaction of varying intensity, depending on an individual's reaction. Bites usually produce a classic allergic response, with the first bite producing no noticeable effect, and the subsequent bites producing the reactions. If the exposure to midges is reasonably continuous, a process of desensitization may follow. People continuously exposed are usually tolerant to the bites, and generally have no reaction or show a mild reaction with a small red spot.⁸

The average reaction for newly exposed people is a red spot that develops a small dome shaped blister with a hole at the top. In people who are more sensitive to bites, the reaction may result in a red swelling over an area of a few centimetres. The bite area can be extremely itchy, and scratching is very difficult to avoid. Reactions may last 3 - 4 days with slowly decreasing irritation. Sometimes scratching breaks the skin and allows secondary bacterial infections that lead to unsightly sores and residual scarring.

TREATMENT OF BITES

Mild reactions from bites require little treatment other than the application of soothing lotions. Proprietary products such as Eurax, Stingose, Medicreme, Katers Lotion, Democaine and Paraderm crème can give relief from bites or prevent secondary infection. Useful nonproprietary products include tea tree oil, eucalyptus oil, aloe vera gel, paw paw ointment, or methylated spirits. Painful reactions to bites can be appreciably reduced by the intermittent application of ice packs to the bite site.

More severe reactions may need medical advice and systemic treatment using antihistamine products such as Phenergan, Telfast or Vallergan. Check with your doctor or pharmacist for available products and safety information.

BREEDING SITES OF CULICOIDES ORNATUS

Culicoides ornatus is by far the most common biting midge pest around the coast of the Northern Territory.³

This midge breeds in the highest numbers in the dry season in the mangrove mud in the creek banks of upper tidal tributaries around the mean high water neap tide mark. This corresponds to an area reached by tides from 4.8m to 6.0m ACD (Admiralty Chart Datum) in Darwin Harbor.^{4,9} The prime breeding sites are in a narrow zone in the upper section of the creek bank associated with the occurrence of pneumatophores of the mangrove species *Avicennia marina* on narrow creek banks. The prime dry season breeding site has an upper limit where the *Avicennia* reduces in height and predominance, and a lower limit where the creek opens out from the overhanging *Avicennia* canopy.⁴ Broad mangrove areas with many tidal tributaries will have a considerable area of breeding sites.

Other breeding sites of low to medium productivity occur at the front edge of the mangrove forest in the *Sonneratia* or woodland mangrove zone facing open water. These breeding sites are usually associated with mud substrates and not with sandy substrates. Narrow beach fringing mangrove areas are usually not appreciable sources of *Culicoides ornatus*, particularly in areas with sandy substrates.⁴

Another site exploited only in the wet season is in the *Ceriops* transition zone at the back of the creek bank forest. This is just below MHWS (Mean High Water Spring or average high tide mark) or 6.6m in Darwin harbor. This is where the mixed *Ceriops* starts in a transition from the taller creek bank mangroves to the smaller mangroves in drier, less frequently flooded areas only reached by tides from 6.5 to 6.8m.

The larvae are small active worm-like creatures that are confined to the surface mud. The larvae take in excess of 6 weeks to mature, when they change into a relatively inactive, air-breathing pupa. The pupa stage lasts only two to three days and the adults emerge around the time of neap tides.⁹

THE FLIGHT ACTIVITY OF CULICOIDES ORNATUS

The numbers of adults emerging from pupa cases is related to the lunar cycle, with sudden rises in numbers inside their mangrove breeding sites of the order of 16 times the number occurring on the previous day. The peak in emergence occurs in the two days around the neap tide, although emergence of adults can continue for up to 4 days after the neap tide.⁴

The adults mate soon after emergence. The males are short lived while the females stay in the mangroves to develop and lay their first batch of eggs. The females then start to disperse from the mangroves in an active flight inland in search of blood meals. The dispersal starts about 2 days before the spring tide, and reaches a peak around the day of the spring tide. They show a marked abundance around spring tides with full moons, but are also numerous around spring tides of the new moon.³

The adults seek shelter in winds above 8 km/hour, so that there is little tendency for them to be borne long distances by strong winds. Light breezes from their breeding areas will however aid their dispersal flight. They are active fliers and despite their small size, are relatively hardy insects.

Mass movements of adults can occur to a zone from 0.5 to 1.5 km from the mangrove margin of their major breeding sites, although they will move greater distances up creeks and rivers with dense tree cover that form avenues of humidity for dispersal. The dispersal is a purposeful one, with the midges actively flying away from the mangroves. Often higher numbers can be found up to 1.0 km from the mangroves compared to lower numbers in the mangroves or at the mangrove margin. Elevated hills or escarpments within 1.5 km of prolific biting midge breeding sites often exhibit higher biting midge numbers compared with lower adjacent areas. Minor pest numbers can be detected up to 3 km from the nearest mangrove margin that are prolific breeding sites .

Most *C. ornatus* bite in the morning and evening. There is a peak in biting activity in the one hour either side of sunset, with a smaller peak in the one-hour after sunrise of about half the sunset peak. However there is a low level of activity throughout the night.

SEASONAL ABUNDANCE OF CULICOIDES ORNATUS

The annual peak of *Culicoides ornatus* adults in the NT is in the August to October period in the late dry season, with lowest numbers in January and February during the wet season. Populations start to build up from the end of the wet season to the late dry season with a slight decrease in the coldest months of June and July. Populations start to decline rapidly after the first heavy rains occur. However pest numbers can still be present during the seasonal lows in the mid dry season and the mid wet season.

There are three different breeding sites in the mangroves, with varying seasonal productivity from the different breeding sites. Mangroves with small tidal tributaries that contain the prime creek bank breeding sites are dry season breeding sites. The greatest productivity from these creeks occurs in the August to October period. They are not significant sources of midges in the wet season.⁴ The back of small mangrove creeks in the *Ceriops* transition zones has moderate productivity in the wet season.^{4,9} Areas with extensive *Sonneratia* zones will have moderate productivity at least in the dry season⁴ and probably all year around.

Highest numbers of *Culicoides ornatus* occur for the four days around the full moon, with high numbers to a lesser extent, four days around the new moon.

PROTECTION FROM BITES OF CULICOIDES ORNATUS

AVOIDANCE

Culicoides ornatus bite primarily in the early morning or evening around sunrise and sunset. Attacks can occur in the daytime in shaded areas adjacent to the mangroves near major mangrove breeding areas or in dense creek vegetation that is continuous with the mangrove breeding places. They will continue to bite throughout a still, humid day and warm humid night, particularly in sheltered areas outside the mangroves but close to their breeding areas. Often there is only a little biting activity in the mangroves during the day during and just after the spring tide, as all midges have usually dispersed landward.

Landward areas that are close to and within one kilometre of broad areas of mangroves with many tidal creek tributaries, especially near densely vegetated creeks that run into the mangroves, should be avoided. This particularly applies to the two days either side of the spring tides in the August to November period. Spring tides on full moons have roughly twice as many biting midges as spring tides on new moons.³

Minimum pest problems occur in the June-July period during the mid dry season or in January and February in the middle of the wet season. During any month the least pest problems occur in the two to three days either side of the neap tide, particularly neap tides following a new moon. A calendar marked with the 4 days around full moons and new moons, with highlights of seasonal peaks of abundance, can serve as a good midge avoidance reminder.

Biting midges are active under calm conditions and are generally inhibited by wind. Wind protected areas adjacent to and within 1.5km of large expanses of mangroves should be avoided around the spring tide period. People in open areas exposed to winds will experience less pest problems compared to other areas.

Elevated houses and high rise buildings have less pest problems than ground dwellings. Although midges probably fly over dense tree canopies and can fly in appreciable numbers at least 3 metres above the landscape surface, they are generally more numerous lower to the ground surface.¹¹

The worst pest problems around Darwin include areas include landward areas adjacent to the mangroves and tidal areas of Sadgroves and Reichardt Creeks, Hudson Creek, Elizabeth River, and Buffalo Creek. The north shore of Frances Bay near Sadgroves Creek in the Charles Darwin National Park is a particularly troublesome area. This is due to the dendrite pattern of numerous narrow mangrove creeks and an extensive Sonneratia zone nearby. Urban areas of Stuart Park, the Narrows, and near Winnellie, which are closest to the Sadgroves creek mangroves, can experience seasonal moderate to minor pest problems. There are some minor pest problems near the lower reaches of Ludmilla creek and Alawa near Rapid Creek. Darwin city itself is relatively free from midges due to the relative lack of mangroves, the exposed cliffs, and the fact that the prevailing SE and NW winds do not blow from mangrove areas.

CLOTHING AND NETTING

Full-length trousers, socks and shoes, and long sleeved shirts will usually provide considerable protection from midge attack. Pale clothing is generally less attractive than dark clothing. Any exposed part of the body will still be subject to midge bites, with most bites occurring on the legs. Protective clothing should be supplemented with the application of repellants on exposed skin.

Clothing impregnated with permethrin or bifenthrin insecticide offers considerable protection for people continually exposed to biting midges. Impregnation involves soaking the clothing in a prescribed volume and concentration of certain formulations of the insecticide. Protective clothing such as overalls and mosquito nets impregnated with permethrin or bifenthrin will remain effective through one or two washes at the most, and will need reapplication. The insecticides in these treatments can kill the insects after they land on them, but they can also have the effect of interfering with the normal biting behaviour. Impregnated clothes with the additional use of insect repellents can provide extremely good protection.

Normal insect nets and screens are usually not adequate to restrict entry to midges unless the mesh is very fine. Tents screens in particular should have mesh diameter approximately half that of normal mosquito netting. Clothing, screens, netting or tents can be impregnated with permethrin or sprayed with permethrin, bifenthrin or repellents containing Deet to increase their efficiency.

Houses should have outward opening doors and insect screens to prevent entry when opening doors during midge activity.

REPELLENTS

Most repellents have limitations because of their short duration of effectiveness (about 2-4 hours) and their irritability to mucous membranes around the eyes and mouth. Care is needed with young children to avoid the spread of repellent to their eyes or mouth. Repellents are also removed by perspiration.

Repellents that contain Deet (diethyl toluamide) or Picaridin as the active constituent offer considerable protection. Mixtures of natural oils or oils with natural ingredients such as herbs or antiseptics are not as effective as repellents containing Deet or Picaridin. In general effective repellents require above 10% Deet and 9% Picaridin. Repellents in lotions are more effective than alcohol based spray-ons, while gels are the most effective formulations. Repellents can also be applied to mosquito netting or insect screens, although a sample application on a small piece of netting is wise as some repellents affect synthetics. Repellents containing relatively high amounts of Deet can melt some plastics, although those containing Picaridin don't have the same effect.

Other methods of repelling biting midges include the use of coils, repellent oil lamps, and electric, flame or gas powered vapor pads impregnated with insecticide. These work

satisfactorily in relatively closed situations such as rooms, or sheltered patio and veranda situations out of the wind, where a cloud of vapour or smoke can build up. The Mosquito Lanterns and Thermocell devices that heat an impregnated pad containing allethrin have been found to be very effective in a narrow zone around people outdoors in still to mild breeze situations. However they cannot provide satisfactory protection in windy and exposed situations.

Smoke from a fire with green leaves will give some protection in emergency situations. Burning aromatic and oil producing foliage of plants such as *Hyptis* (horehound), *Calytrix* (turkey bush), *Melaleuca* species (paperbark) and *Eucalyptus* species (gum trees) can give appreciable protection. Rubbing the skin with the leaves of some of these plants can also provide some protection, but this is not as good as recommended repellents.

The so-called "electronic mosquito repellers" that emits a frequency that is supposed to repel biting midges by imitating the noise of males do not work and offer no protection against biting insect attack.

There is an urban myth that taking Vitamin B1 or thiamin can act as a repellent. There is no scientific evidence that Vitamin B1 acts as a repellent, or helps to reduce the reaction to insect bites by developing some immunity to the bites.¹³ Other topical applications such as a DettolTM and baby oil mixture do offer some physical barrier to biting midges, but are not as effective as Deet or Picaridin based repellents. The best protection from biting insects remains the avoidance of the problem areas at times of abundance and the use of protective clothing in combination with efficient repellents.

USE OF LIGHTS

Biting midges can be attracted to lights. Houses in biting midge problem areas should have dull outside lighting, with little internal light visible from outside. Lightproof curtains that can be drawn at night offer a good alternative. Outside lights should be away from insect screens, as the midges attracted to the light can then penetrate the screens. Outside lights should be yellow (or red, which is even better) to reduce their attractiveness to biting insects. Attractive lights such as large incandescent bulbs or white or ultra violet fluorescent tubes positioned a distance away from a house or building can deflect biting midges to some extent. However rows of streetlights positioned between mangroves and residential areas are not effective barriers to midge dispersal inland.¹¹

THE REDUCTION OF VEGETATION

The reduction of vegetation around houses or recreation areas can reduce problems by removing shelter for the midges. A buffer of clear open space between the mangroves and residential areas can reduce biting midge numbers in a residential area, as long as the buffer is wide and subject to winds. However clear open buffers by themselves offer little protection unless they are at least 1 km wide. Mowing a wide margin around houses to eliminate dense grass can help reduce the available areas where midges can harbor.

THE USE OF ATTRACTANT TRAPS

There are a number of insect attracting traps on the market. They generally use light or carbon dioxide as an attractant and either trap the insect in a container, electrocute, or drown the insects. Some are more useful than others but can not be relied to give considerable protection from bites for unprotected people in close proximity to the traps. In most cases they attract biting insects to the general vicinity and these are then diverted to people, who are more attractive targets. Some traps can help to reduce the overall population, as long as there are enough traps, the biting insect population is relatively small, and the area is isolated from re-invasion from other areas. However most trapping techniques can not cope with the huge populations of midges at one time, and those not trapped still result in a pest problem.

EVALUATION OF BITING MIDGE PROBLEMS

The Medical Entomology Branch of the NT Department of Health has conducted numerous investigations into biting midge problems in the Top End of the NT.^{2,3,5} Potential problems have been investigated by trapping midges overnight using special carbon dioxide (CO²) baited traps. The number of midges collected can be counted or estimated by weight or volume and identified to species under a microscope in the laboratory.

The number of bites by biting midges that constitute a pest problem will largely depend on an individual. It has been suggested that over 60 bites per hour for most experienced biting midge workers are the thresholds of acceptability. For people unaccustomed to biting midge bites, even 1 to 5 bites per hour may be considered unbearable.

There is an approximate relationship between the number of midges collected in a CO_2 trap and the number of bites that can be expected at the peak biting period. For an unprotected person, the number of bites in an hour at the peak biting time is approximately one quarter of the number collected in a CO_2 trap over one night at the same position. Thus CO_2 collections of over 240 per carbon dioxide trap per night are likely to represent a pest problem (equal to over 60 bites per hour) to unprotected people with prior experience of biting midges. Collections of over 1000 per trap per night represent over 250 bites in an hour and would constitute a major pest problem. Trap collections of over 5,000 per trap would constitute a severe pest problem.³

The numbers of *C. ornatus* collected by CO_2 traps in different locations can indicate the magnitude of the human pest problem in each location. Trapping on a constant day in relation to the tide cycle over every month in a year can give an indication of the seasonal population fluctuations. Trapping at different distances from the mangroves and in different vegetation types can give an indication of the dispersal of midges into various areas.

CONTROL OF CULICOIDES ORNATUS

INSECTICIDE FOGGING FOR ADULT MIDGES

Insecticide fogging is the application of aerosol size particles directed against active flying insects. Insecticide fogging operations in residential areas by vehicle or hand held equipment are usually not very effective measures to eliminate pest problems, due to the rapid re-infestation of midges from nearby breeding and harborage areas. Sometimes re-infestation occurs very soon after the fog has cleared, although up to 12 hours protection can be achieved in some localized situations.

For effective midge control, the entire midge breeding and harboring area near residential development needs to be fogged each day over the 3-4 day period of peak emergence. This has to be timed to coincide with the time just after the midges have emerged and before they begin to disperse out of their breeding areas. This area would also have to be relatively isolated from other such areas to prevent re-invasion. Fogging also has to be carried out during the peak activity period in the evening and early morning.

For vehicle ground based operations, the fog has to be able to drift into the target area on favorable winds of the right velocity and in the right direction. This often reduces the opportunity for effective fogging. Fogs do not usually penetrate more than 50m into dense forested areas such as mangroves, monsoon forests and other thick vegetation.

One of the major problems is determining the level of control required. A reduction of *C. ornatus* numbers by 99% may be required to reduce a large pest problem to an appreciable level. This may be impossible to achieve for various operational purposes, and if there were still any remaining pest problem, the control would not be cost effective.

In the Darwin situation, the mangrove breeding and harboring areas are generally inaccessible, too wide, or too extensive for ground based application methods to effectively reduce midge numbers, although some temporary relief would be possible in some areas.

Aerial application of insecticides aimed at adult midges in breeding and harborage areas has given the best results in overseas investigations, but in some instances there has been immediate re-infestation. It is a difficult practice, as the breeding grounds have to be closely delineated and fogging must be based on an accurate forecast of adult emergence times. The fogging has to be with sufficient regularity to kill all the emerging dispersing females over the night and fog drift to nearby residential areas has to be avoided. Fogging is not carried out regularly for midge control in Australia and requires more local research. Fogging involves large continuing costs, which is often beyond the resources of many local authorities. Insecticide resistance and the killing of other insects pose additional potential problems.

BARRIER SPRAYING

The application of insecticides to create an artificial barrier or an insect killing zone around houses offers great promise as a new control method. The application of these residual insecticides to exterior walls, screens, patio plants, nearby hedging plants or lawns and other close vegetation may kill midges attracted to houses or people.¹⁴ Insecticides that can be used include permethrin, deltamethrin, alpha-cypermethrin, lambda-cyhalothrin and bifenthrin. Bifenthrin has the advantage over other similar insecticides, as it appears to have less of a repellent or agitation effect on insects, is less irritant to people, is ultra violet resistant, and binds very well to surfaces to give a good residual effect.¹⁴ As with all synthetic pyrethroids, it must only be applied as per the label and kept out of fish habitats.

INSECTICIDE CONTROL OF LARVAL HABITATS

Breeding site treatment by applying insecticides to kill larvae before emergence of adults is a possible control method but there have been very few examples of successful larval treatment in mangrove areas. Larval habitat treatment involves considerable costs and organization, which is impractical in extensive breeding areas such as those surrounding Darwin. Insecticides would need to have good residual qualities and be able to penetrate dense mangrove tree cover and mud in a tidal situation. Most insecticides with these qualities would generally kill non-target insects. The problem of accurately delineating all the significant breeding sites and the seasonal fluctuation of breeding sites pose additional problems.

ELIMINATION OF BREEDING HABITATS

Reclamation of mangroves has been successful in eliminating biting midge breeding sites in various localized situations. This usually requires large amounts of fill material which is neither cheap or readily available. For *Culicoides ornatus*, the reclamation needs to extend from near the average high tide level to below the outer mangrove forest. This may involve significant engineering considerations posed by deep mud and erosion of the filled area.

Reclamation would not be practicable in most of the Darwin area because of the extensive areas involved. The destruction of large areas of mangroves would be environmentally undesirable and unacceptable to public opinion. This potential solution would only be practicable in localized areas if the breeding site was small, in close proximity to residential development, was regarded as an area of reduced environmental importance, and the filling could create a stable shore environment.

There should be conclusive evidence that the site to be reclaimed is a significant source of biting midges and that the midges are significant pests to nearby residential development. Mangroves can be an indicator of biting midge breeding sites, but the presence of mangroves does not confirm any site as the breeding place. Other specific factors such as substrate types are involved in productive breeding sites.

BUFFER ZONES

There is some evidence that creating a buffer zone between urban residential development and mangrove areas can reduce the dispersal of biting midges into residential areas. Clearing of vegetation and mowing to allow wind disruption, or extensive streetlights or roads with active traffic in the buffer zone may enhance the buffer to some extent.³ However extensive testing of a modified buffer with lights and different vegetation types in Darwin have shown that unmodified buffers and lights by themselves are not effective barriers to *C. ornatus* dispersal from mangroves to urban areas. The effectiveness of buffers is generally related to the width of the buffer and the presence of blood sources or other attractions such as light in the buffer zone. However semi-urban residential or industrial development between mangroves and urban areas can reduce midge dispersal inland. In general, unmodified buffers need to be in the order of 1.5km, and modified buffers in the order of one kilometre to offer significant reduction in numbers.

PLANNING GUIDELINES TO PREVENT BITING INSECT PROBLEMS

The Medical Entomology Branch is involved in the planning process to reduce the effects of biting insects. Guidelines have been prepared for preventing biting insect problems in new urban and semi rural residential developments, industrial, and other developments.

In 1974 the planning for the new satellite town of Palmerston near Darwin included a buffer of at least 1-km from the mangrove boundary to urban residential development.¹² Palmerston is one of the few urban areas in Australia that has been specifically designed to minimize biting insect problems.

Good urban planning is required to;

- reduce the risk of biting insect pests
- recognize and avoid areas of biting insect breeding or harborage
- avoid costly and environmentally undesirable rectification methods
- avoid costly and ongoing biting insect control programs

The Medical Entomology Branch gives advice on what may constitute a potentially significant biting insect breeding site. In some instances detailed entomological investigations are necessary to gather sufficient information before the detailed planning stage. The avoidance of biting insect problems can be achieved in the initial planning process by consideration of development location, easements, buffer zones, and sub division design.

SELECTED REFERENCES

- **1**. Marks, E.M. (1982), "An atlas of common Queensland mosquitoes" by Elizabeth Marks with "A guide to common Queensland biting midges" by Eric Reye, ", Q.I.M.R. Revised edition.
- 2. Whelan, P.I. (1991a), "Biting midge investigations near Darwin and their implications for urban planning", In Proc. Nat. Conf. on Biting Midge, Surfers Paradise, Qld., 8-9 Feb, 1990.
- **3**. Whelan, P.I., Hayes, G., Montgomery, B.L. (1997), "Biting midge surveillance in Darwin Harbor, *Culicoides ornatus* (Diptera: Ceratopogonidae) abundance and dispersal", *Arbovirus Research in Australia,* vol. 7, pp.326-336.
- **4**. Shivas M. (2000) "The Larval Biology of *Culicoides ornatus* Taylor in mangroves near Darwin, NT. PHD. Faculty of Science NTU Darwin Feb 2000.
- **5**. Whelan, P.I., Booth, D. and Kelton, W. (1988), "Biting insect investigations and comment on the proposed Newtown (Weddell) Development, Darwin, N.T.", Department of Health and Community Services, Darwin NT.
- 6. Whelan, P.I. and Hayes, G.A. (1993), "Biting Insect Investigation Darwin South Stage I", Department of Health and Community Services, Darwin NT.
- **7.** Whelan, P.I. (1995), "Biting insect investigation Darwin South Stage II", Department of Health and Community Services, Darwin NT.
- 8. Lee D.J. (1975), "Arthropod bites and stings and other injurious effects". SPHTM, Inst. of Public Health, Uni. of Sydney.
- **9**. Shivas, M., Whelan, P.I., & Webb, C. (1997), "The characterization of emergence sites of the biting midge *Culicoides ornatus* (Diptera: Ceratopogonidae) in mangroves near Darwin, NT, Australia", in *Arbovirus Research in Australia*, vol. 7.
- **10**. Wightman, G.M. (1989), "Mangroves of the Northern Territory", Northern Territory Botanical Bulletin, No.7 Conservation Commission, Palmerston, N.T.
- **11**. Shivas M and Whelan P.I. (2001), "Biting midge research project March 1999-March 2001". Study for DLP&E, Darwin NT on dispersal biology and effect of lights and vegetation. Department of Health and Community Services, Darwin NT.
- **12.** Leihne, P.F. in association with Medical Entomology Branch DHCS (1985), "Mosquito and biting midge investigations, Palmerston 1982-85", Palmerston Development Authority.
- **13.** Khan A.A., Maibach H.I., Strauss W.G. et al. "Vitamin B1 is not a systemic mosquito repellent in man". Trans St John Hosp Derm Soc 1969;55:99-102
- 14. Standfast H., Fanning I. Maloney L., Purdie D. and Brown M. "Field evaluation of Bistar 80SC as an effective insecticide treatment for biting midges (Culicoides) and mosquitoes infesting peri-domestic situations in an urban environment" Bulletin Mos Cont Assoc Aust Vol 15 (2) July 2003.

33. MARCH FLIES -PUBLIC HEALTH IMPORTANCE AND CONTROL

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March flies (also sometimes known as horse flies or tabanids) are stout-bodied flies measuring 6mm to 25mm in length with large eyes. There are 3,000 species worldwide, with some 200 species in Australia, although there are likely to be many more undescribed species. March flies are known to be serious pests of humans, livestock, domestic animals and wildlife. They impact on the economy through affects on human and animal health, recreation, tourism and agricultural productivity.

The females of most species of March fly take a blood meal after inflicting a painful bite with their piercing mouthparts. March flies can be annoyingly persistent while attempting to bite. The blood is needed as a protein source for maturation of the eggs (100 - 1000 eggs) produced per blood meal), however some species are able to lay their first batch of eggs before taking the first blood meal (autogeny). Nutrients for normal activities are obtained from flowers and plant juices or from food reserves stored during the larval stage.

March flies can occur anywhere in Australia, and in a range of habitats. The adults are most active during daylight hours during the warmer months, particularly on calm, sunny days. In the tropics, they can continue to be active through the drier winter months.

Breeding occurs in damp soil, rotting vegetation, sand and rot holes in trees, with the larvae feeding on animal and plant material within the substrate. The life-cycle can take many months or even years, depending on the species and soil temperatures. After emerging, the adults mate and the females disperse from the breeding site in search of blood meals. The adults live for 3-4 weeks.

HEALTH IMPACTS

March flies are not known to transmit disease to humans or livestock in Australia. However, their bites can cause adverse allergic reactions in some people, causing skin redness and swelling, sometimes requiring hospitalisation. Livestock can suffer significant blood loss from repeated biting.

The allergic reaction occurs in response to the saliva injected by the fly to prevent the blood from clotting. The application of an ice pack or mild antihistamine may relieve painful bites. Secondary infection due to scratching the bites may require the application of antiseptic cream or systemic antibiotics.

A small, brown species, *Mesomyia tryphera*, which occurs in the Pilbara and Kimberley regions of Western Australia, appears to produce serious symptoms in some people, including hives, fever, wheezing and anaphylaxis. In these cases, if an epi-pen (an auto-injector of adrenaline) is available, it should be administered immediately and an ambulance should be called. People having such severe reactions may need to relocate to another region during the March fly 'season' to avoid further bites and life-threatening symptoms.

In recent years there have been more people reporting to hospitals in the Pilbara region with March fly bites. The peak period is from January to March, especially in the weeks following heavy rainfall. In one Pilbara location the local hospital reported over 25 presentations in one day. Another Pilbara hospital reported over 100 presentations in a month. Some patients

required antibiotics for secondary infections and a few presented with anaphylaxis. It is not known whether there is a real increase in March fly abundance or if the number of people affected reflects the increase in outdoor workers associated with the Pilbara mining 'boom'. Publicity about the March fly risk may also affect the number of people presenting at hospitals.



Figure 80: March flies. Left: common species in southwest WA (photo MD Lindsay); Right: similar to species in northwest of WA that is associated with severe allergic reactions (photo Frank E French, Georgia Southern University).

AVOIDANCE AND CONTROL MEASURES

March flies are known to be attracted to dark blue, so it is advisable to avoid wearing blue and other dark colours. Light-coloured loose-fitting clothing and insect repellents containing diethyl toluamide (DEET) will maximise protection when March flies are present.

Control of March flies using larvicides is generally not possible due to their extensive breeding areas and ability to fly long distances from breeding sites. The targeted use of adulticides may reduce populations around settlements. The biology of most species is not well known, complicating efforts at control.

Fly trapping may be useful for reducing populations in localised areas such as schools and workplaces. Blue- or dark-coloured surfaces, coated with a permanently sticky adhesive (such as Tanglefoot), can remove March flies from an area. Box traps utilise a tent painted black inside, positioned over a black target ball, to attract March flies.



Figure 81: Traps for March flies. Left: sticky trap; Right: box trap with black target ball.

34. ADULT MOSQUITO IDENTIFICATION AND GENERAL CONSIDERATIONS FOR MOSQUITO IDENTIFICATION

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It is not always only true mosquitoes that are noticed or collected during surveillance operations, or submitted by a member of the public concerned by swarms of 'mozzies' creating a visible nuisance. Many insects around a home may resemble mosquitoes to the lay observer and it is important to be able to tell which specimens in a collection of insects are actually mosquitoes, remembering that not all mosquitoes look alike and there is remarkable variation between species.

Adult mosquitoes (the Family Culicidae) are slender, fragile, long-legged insects, but there are similar-looking insects, particularly some of the non-biting midges (the Family Chironomidae) and the crane flies (the Family Tipulidae), which are quite often mistaken for mosquitoes by the inexperienced observer or collector, and these can create some confusion when identification is attempted.

Larval mosquitoes may be more readily recognised as the 'wrigglers' in a container, drain, or ground pool, but they are difficult to identify to species in most cases without some experience and the use of a microscope.

It is important for those interested in public health or pest control to not only be able to recognise mosquitoes from similar insects, but also be able to identify the particular mosquito species that may, or may not, be important as pests or vectors of disease.

Simply, an insect is an adult mosquito if it has <u>all</u> of the following three characteristics:

- i. If there is a **long proboscis** (biting organ) protruding from the head and this proboscis is several times longer than the head itself;
- ii. If there is only **one pair of wings**, and not two pairs or no wings;
- iii. If there are scales present on the veins of the wings and a fringe of scales on the hind edge (magnification will be required to see these veins and the scales).

Mosquito larvae are generally distinguished from other aquatic insect larvae of similar appearance by their swollen thoracic area and lack of leg-like appendages. Most common mosquitoes (e.g. *Aedes* and *Culex* species) also have a terminal breathing tube (siphon), but *Anopheles* species have no siphon, and *Mansonia* and *Coquillettidia* species have a modified siphon which attaches to subsurface aquatic plants.

It is true that adult mosquito species differ from each other in aspects of their biology, their habitats and behavior, and thus in their potential to be a pest or vector of disease:

- Some species rarely or never attack humans for blood and thus do not need to be included as targets in pest or vector control programmes.
- Some of those species that do attack humans are not capable of transmitting particular disease pathogens and thus do not need to be targeted in some disease control programmes.
- Some species will be found in very different larval habitats from others and control operations will need to be appropriately directed.

- Some species may be known to have developed resistance to one or more insecticides and control operations will need to be selected accordingly.
- Some species have very quick development cycles and control operation schedules will need to take this into account.

Thus it is **very important** to know how to accurately identify the range of species active in the locality, or at least to be able to correctly recognise the important pest and vector species which might be collected during surveillance operations, so as to be able to determine the requirements for control operations, properly plan and implement the operations, and evaluate their effectiveness.

Mechanisms have been developed to allow most mosquito species to be recognised on external characters of both the adult and larval stages. For the most part, mosquitoes have variations in structure of body parts, and colours and patterns of markings which are characteristic for a particular species; many of these are visible under relatively low power magnification and the recognition of these physical attributes can enable the correct identification of a biological species. Some of these characteristics, the structural details and ornamental appearance, can be recognised readily by even the inexperienced collector; however, others are more subtle and less noticeable, and the trained and practiced eye of an expert might be required to discriminate between similar species.

When a species is 'discovered' and first reported in a scientific journal, a detailed description with illustrations is usually published so that others who may encounter the species can recognise it and differentiate it from other species. This can be very important in a public health context, such as in the case where, of two very similar (perhaps superficially identical) species which occur together in the same area, only one is a disease vector (because of genetically based physiological or behavioural factors) and in need of control while the other is of no public health concern.

Although, in any particular area there may be only few species of importance as pets or vectors, once surveillance programmes are undertaken it is inevitable that a wider range of species will be encountered and it will be vital that the 'target' (i.e. the pest or vector mosquito of most concern) species can be differentiated from the others that will be found with it in the various collections. The relationships (spatial, temporal, and numerical) of the 'target' species within the overall mosquito fauna will be important factors which are crucial to the efficient planning, implementation and evaluation of control operations.

Although some characteristic markings may be large and/or obvious, and with practice some species may be tentatively identified with the naked eye, the majority of characters used in identification of mosquito adults require examination under at least a x10 hand-lens but usually a dissecting microscope (x5-x50). Some larvae may also be identified to genus, and with 'educated guessing' to species, upon viewing gross characters with the eye or a hand-lens, but for accurate identification of mosquito larvae it can be necessary to examine the specimen mounted under a coverslip on a glass slide under at least a dissecting microscope but often under a compound microscope with high power magnification (x100-x400).

Identification of adult mosquitoes at the simplest level is dependent primarily on observing and comparing colour patterns on the various parts of the body – the head and its proboscis, the thorax and its legs and wings, and the upper and lower surfaces of the abdomen. Coloured patches or discreet coloured bands or stripes are composed of scales of one colour contrasting with adjacent scales of another colour or shade.

Adult mosquitoes are usually identified using females only; dry ice-baited traps collect predominantly females, and because they are the ones that bite and transmit pathogens it is females that are of concern. However, males may be occasionally trapped and will also be amongst females that might be bred out from larvae and pupae. It is important to note that

identification aids are generally designed only for females, and males should not be used for identification unless no females are available; identification of males should be considered tentative, unless corroborated by identification of associated larvae or females.

To a great extent, identification is simply an acquired skill that improves with experience. However, it requires good aids, and is facilitated by having a good microscope and bright light source. Often, adults collected in traps are somewhat damaged by the buffeting involved and the coloured scales on the body parts, or even the body parts themselves, may be missing. Under such circumstances identification can be difficult, and may be thought impossible, but such damaged adults can be compared with undamaged individuals within the same (or similar) collection and often a correct identification ca be achieved.

Identification of larval mosquitoes is covered in a following section, and can be more or less difficult depending on the species concerned and the development stage of the larva collected. Most aids produced for identification of larval mosquitoes are designed for the mature (4th instar) larva and, because some characters change with development, may not necessarily be appropriate for younger larvae. Therefore, if larvae collected in the field are suspected to be younger than 4th instar they could be kept alive in water from the field site (or distilled water, or tap water <u>boiled</u> to eliminate the chlorine), and fed on <u>small</u> amounts of fish food, ground dog biscuit, or cereal for further development. Also, if larval identification aids are not available, or there is uncertainty about larval identification, then some of the larvae should be kept alive (and fed) to allow complete development through all stages, and the pupa, through to the adult stage (which can be kept alive by feeding on raisins or apple pieces) and the adult can then be examined to identify the species.

Male adult mosquitoes can usually be readily differentiated from females because the males have very hairy antennae (called plumose such as for a feather) and the palps on either side of the proboscis are long and generally of similar length to the proboscis (see diagrams); some female mosquitoes (e.g. *Anopheles* species) also have long palps but the antennae are not as heavily 'feathered'. Males have pincer-like claspers (sexual organs) at the end of the abdomen which can also be used to separate them from females.

Adult female mosquitoes can sometimes be separated usefully into genera before the species is determined, by using some characters that are reasonably obvious. Anopheles species females all have palps as long as the proboscis while females of Aedes, Culex, Coquillettidia and Mansonia all have palps which in most cases are much less than half the length of the proboscis. Aedes species tend to have the terminal segments of the abdomen tapering sharply or to specialised flattish plates; Culex species tend to have a blunt and rounded end to the abdomen; Mansonia species have the end of the abdomen truncated; Coquillettidia species have bluntish abdomens somewhat similar to Culex. These characters however, are not universal and not always readily discernible on all specimens; all identifications should be undertaken with use of an appropriate 'KEY' and with reference to illustration and descriptions when necessary.

Rather than working through a number of detailed descriptions to identify an individual specimen, a relatively simple device called a '**KEY**' (written and/or illustrated) is usually developed for a region or for a group of mosquitoes in order to provide a quick but accurate means of identification. This involves a stepwise comparison of particular characters, selecting between alternatives and eliminating species when they do not fit the description, until arriving at a conclusion which identifies the species at hand.

For rare species which may not often be encountered and thus not readily recognised even by the practiced expect, the recourse is usually to compare the mosquito at hand with original descriptions in scientific journals and with a collection of reference specimens.

An identification **'key'** is very much a tool, an artificial device constructed for the purpose of quickly identifying species, and it usually has a number of limitations. Generally, a **'key'** is designed for a particular region or group of species, and includes only that range of species know to occur in that region; thus if a species from another region, or one collected locally but previously not recorded from the region in question, is presented for identification, the **'key'** may provide an incorrect identification or possibly none at all.

There are other problems to be aware of when using a 'key'. Firstly, a damaged specimen may be difficult or impossible to 'key-out' because particular characters are not present (e.g. hind legs missing or scale patches rubbed). Secondly, some natural variation occurs between different (e.g. geographically separated) populations of the same species, and even within local populations, and some specimens may not fit the 'key' as well as others. Thirdly, with a few species, it is simply not possible to distinguish between some species (or one sex of some species) simply on external appearance, and biochemical or genetic techniques are required to discern between what are termed 'sibling' species; these are truly separate species and, although they appear to be identical when identified through a 'key' they can be expected to have different biological traits.

Therefore, it should be kept in mind that a '**key**' is just a tool; it is <u>not</u> infallible, and any doubts concerning identifications should be checked out by recourse to an expert or by consulting detailed descriptions, illustrations, or reference specimens.

Use of a '**key**' involves the reader in selecting, from two alternative descriptions (together called a couplet), that description of a stated part or parts of a mosquito that best matches the specimen in question being examined. That selected part of the couplet will then lead to another couplet where a further choice between two alternatives within a couplet is required, and eventually the selection process will come to an end with a species name for the specimen in question being examined.

To illustrate the process, a 'key' to identify animals follows:

1	-	The specimen has scales covering the body skin, and there are no visible limbs
	-	The specimen has hair or feathers on the body skin, and there are four limbs 2
2	-	The specimen has feathered wings as front limbs, and has feathers covering
		the body surface BIRD
	-	The specimen has hair on the body surface, and the front limbs are not wings 3
3	-	The specimen stands horizontal on 2 pairs of legs4
	-	The specimen stands vertical on 1 pair of legs6
4	-	The specimen has horns on its head 5
	-	The specimen has no horns on its head DOG
5	-	The specimen has a long tail and unbranched horns COW
	-	The specimen has a short tail and multi – branched horns DEER
6	-	The specimen has a tail
	-	The specimen has no tail

To correctly identify a human specimen the second alternative in each couplet should have been chosen to lead to the answer in couplet 6.

Some deficiencies of the above 'key' should be immediately obvious (e.g. a sheep or goat would not '**key-out**' because they would not satisfy either choice in couplet 5, and a cat or pig would '**key-out**' incorrectly as a dog), but it serves to illustrate the process whereby a human may be differentiated from a selective group of animals in a particular area.

Some '**keys**' use illustrations instead of descriptions and some combine both, but the principle is the same, a selection of the most appropriate alternative (from usually 2, sometimes 3 choices), and then following the sequence through to the identity answer.

The **keys** provided are aids to the identification of the species of Western Australia. They have been constructed using the most obvious and readily recognised characters so as to be more useful to the inexperienced collector. However, having regard to the foregoing discussion on the limitations of **keys**, they should be used in conjunction with detailed notes on characteristic features and illustrations of the species.

Apart from mosquito species known to be recorded from the region in question and which of those are of greatest importance as pests or vectors of disease pathogens, it is desirable to have an appreciation of a few very important species that are potential invaders of the local region, either from other Australian or international sources. The two most important species in this context are *Aedes aegypti* and *Aedes albopictus*.

Ae aegypti is a domestic container-breeding pest mosquito and the major international vector of the dengue viruses and yellow fever virus (it can also transmit Ross River and other viruses); the species is known within Australia currently only from Queensland although it has previously been present in Western Australia, the Northern Territory and New South Wales. The adult has characteristic features (a white lyre-shaped pattern on the top of the thorax and an unbanded proboscis) which distinguish it from other species in Western Australia. The domestic container-breeder *Ae notoscriptus* has a similar pattern on the thorax but has a prominent white ring in the middle of the proboscis.

Ae albopictus is currently not established within Australia but in recent years this Asian species has invaded and become established in many countries, principally via the used tyre trade, and (as with *Ae aegypti*) it has been detected in Darwin on fishing and trading vessels from Asia; it is of great concern because it is a domestic and peri-domestic container-breeding pest mosquito and an important vector of dengue viruses (it can also transmit Ross River virus). The adult has a distinctive single white stripe on the thorax and an unbanded proboscis; there are no similar species in southern Australia, but it might be confused in northern Australia with *Ae katherinensis* although the abdominal bands on this latter species are sub-basal while in *Ae albopictus* they are basal.

Detection of either of these species in Western Australia should be reported promptly to the Department of Health. All adult specimens suspected to be perhaps *Ae aegypt* or *Ae albopictus* should be referred to an expert for confirmation. Identification of larvae suspected to be *Ae aegypti* or *Ae albopictus* also requires expert assistance.

SPECIMEN STORAGE

For storage in reference collections, adult mosquitoes should be pinned and maintained in proper entomological boxes or cabinets where they will be safe from physical damage and biological destruction by mould and other insects and mites. If this is not possible, or for short-term storage or transport, the specimens can be stored in secure containers between sheets of tissue paper to prevent movement and consequent damage. Mailing of specimens to experts for identification requires safe packaging to avoid damage in transit and the inclusion of data on the date, locality and method of collection, and the collectors name.

Larval mosquitoes can be stored in alcohol solution (preferably 70% ethanol, although approx. 70% methylated spirits is acceptable for the short-term), but for reference purpose they are usually mounted in a preservative medium on a glass slide under a glass coverslip; in either case a data label detailing date and locality of collection, and name of the collector should be included (written in pencil on a slip of paper) in the bottle or attached by adhesive to the glass slide.

35. LARVAL MOSQUITO IDENTIFICATION

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LARVAL MOSQUITO RECOGNITION

Mosquito larvae or "wrigglers" are the characteristic aquatic stages of mosquitoes. Most people have seen these at some stage in a bucket of water or a disused fish pond, but may not be aware that there are many different species of mosquitoes, all with larvae that differ in habitats or appearance from each other. Mosquito larvae can inhabit almost any aquatic habitat, from salt water to the small amount of water that collects in the flower bracts of the native ginger plant. Mosquito larvae can be distinguished from other small aquatic animals and from each other by certain structural features, such as the presence or length of the siphon or air breathing apparatus, the shape of the spines at the base of the siphon , the number of spines or teeth on the sides of the posterior tip of the abdomen, or the number of hairs at certain locations on the head.

Mosquito wrigglers are all aquatic and are not found other than in aquatic situations. They are all air breathing and generally must return to the surface of the water to obtain air. They characteristically have no legs and are not red. They have a soft body comprised of three distinct segments, with some sections comprised of hardened plates. They have a number of hairs on the head and abdomen, have a pair of forward projecting antennae and possess an anal segment that has a saddle, a ventral brush and anal papillae.

Many mosquito larvae can sometimes be recognised from other insects and between genera by their characteristic motion in the water and their habit of returning to the surface of the water to obtain air. Most *Aedes* larvae have a characteristic sinuous motion through the water, *Culex* have an arching or slow flicking movement, while *Anopheles* have a quick flicking movement. *Culex* and *Aedes* larvae tend to hang down in the water from their siphon while *Anopheles* and *Uranotaenia* species tend to rest horizontal to the surface of the water.

The larvae all have a number of varied structures that are related to the habitat in which they live. It is useful to look at the appearance of various structures on the larvae and seek an understanding of how these structures relate to the particular habitat in which the larvae has been found. There are a number of structures that are useful for this purpose and this includes the length and shape of the anal papillae, the length of the siphon, the appearance and number of hooks and hairs on the siphon, the colour of the larvae, the particular structure of the hairs near the mouthparts and the shape and size of the lateral comb and pecten teeth. The taxonomy of mosquito larvae relies on many of these structures.

Most mosquito larvae can be recognised to genus level by the naked eye but for species identification it is often necessary to use at least a x10 hand lens or even greater magnification with a dissecting microscope.

It is vital to seek the true identity of any mosquito larvae found and to tie in this identity with the habitat in which it is found. Information on habitat can often help in the field identification of larvae. For example if the water is salty it is more likely that a particular *Culex* species is *Culex sitiens or Culex globocoxitus* rather than *Culex annulirostris*. Often the identity of the larvae is paramount in determining the control requirements of a mosquito problem. If larval identification is not correct, many dollars and time could be wasted in treating the wrong or an innocuous species.

When collecting larvae for taxonomic purposes, it is important to collect as many different looking larvae as possible and in the various stages, as there may be larvae of more than one species in the same habitat and at different growth stages. When collecting larvae, it is important to include a sample of sufficient water so that if some stages need to be grown to a later stage for identification there is sufficient field collected water for this purpose.

Mosquito larvae hatch from eggs. There are basically three types of eggs and these are characteristic for certain genera of mosquitoes and are related to the habitat in which they live. The three main types of eggs can be illustrated by *Anopheles* eggs which are laid singly and float on the surface of the water, *Culex* eggs that are laid in a raft and float on the surface of the water, and *Aedes* eggs that are laid singly on moist surfaces above the waterline. The details of the appearance of the eggs can be used to indicate the various genera present in a habitat or even the species, but generally the eggs are too difficult to find on a regular basis and are not discussed further here. However the awareness of eggs when sampling for larvae and their collection, wherever possible, will aid in the identification of the larvae and allow an understanding of the dynamics of the habitats.

Mosquitoes go through four growth stages. The pupae develop within the final growth stage. Pupae can be identified by various structures in much the same way as larvae but larval keys are generally easier to use and larval stages can be more readily obtained in any breeding site. However it is often useful to distinguish between Anopheline and Culicine pupae. *Anopheles* pupae have a broad trumpet shaped flare on the tip of the two respiratory "horns" on the dorsal surface of the thorax, while Culicine pupae do not generally have any such trumpet shaped enlargement.

Most of the taxonomic keys are based on the fourth larval stage characters. It is sometimes necessary to rear the earlier larval stages to the fourth stage for identification. This can be achieved by rearing the larvae in suitable quantity of field collected water. The water can be topped up with tap water boiled to remove chlorine. The later the stage of larvae collected, the sooner the adults can be reared and the less time and care is needed in rearing . The rearing of early instar larvae is difficult because they must be supplied with food and similar conditions to the natural habitat for a relatively long time. When collecting or rearing larvae it is important to keep a sample of the various growth stages because there may be more than one species in any larval sample and there may be differential deaths between the different species. All of the larvae need to be identified in any sample to determine the presence of different species.

When rearing larvae to adults it is important to keep a sample of the fourth instar larvae and the last larval skin after pupation occurs. Suitable labelling of the last larval skin is essential to enable a correlation between the larval stage and the reared adult.

LARVAL MORPHOLOGY

GENERAL

The fourth stage mosquito larvae is composed of soft tissue with a flexible outer skin to allow movements and growth, with some part of the body characteristically hardened with chitin. Usually the head and various structures on the posterior end of the larvae have hardened plates.

The body is divided into three recognisable sections, a head, a thorax and an abdomen. The head capsule usually has a lot of hairs on top of the head and near the mouth. The thorax is membranous and appears as a distinct short swollen region behind the head and contains quite a number of long protruding hairs. The abdomen is characteristically long and has structures at the posterior end that includes the air breathing apparatus, a lateral comb-like structure, a hardened saddle section associated with a fin-like ventral brush and a group of sausage-like protuberances called the anal papillae.

HEAD

The head capsule is comprised of hardened plates and bears two lateral eyes and a pair of forward projecting antennae. The shape of the head is generally oval to round, with the shape of the head (ratio of length to width), or the size in comparison to the thorax, used as an indicator of various genera or species. The length of the antennae in relation to the head is also used as a taxonomic character. The antennae usually have a tuft of hairs near the midpoint and the number of hairs and the positioning of this tuft is an important character. There are paired groups of hairs on the top of the head that have specific names or numbers that differ between the Culicine mosquitoes and the Anopheline mosquitoes. In Culicine mosquitoes the group of hairs labelled C5 and C6 are most often used in larval identification while in Anophelines it is the inner (C2) and the outer (C3) clypeal hairs that are most often used. Anyone interested in mosquito taxonomy should at least get to know the position and the names of these groups of hairs.

The head features a ventral mouth that has a group of brush-like hairs on each side. This group of hairs is usually very bushy, but it can be quite compact in those species that are cannibalistic or predacious, where the group of hairs has been modified into a seizing apparatus. The mouth brushes are used to fan a water current containing food into the mouth.

In *Culex* and *Aedes* species the head is characteristically face down and the larvae feed below the surface of the water or at the bottom of the habitat. In the *Anopheles* mosquito the head rotates 180 degrees and the mouth brushes draw in the very top surface layer of water which often contains micro organisms and debris such as pollen.

THORAX

The thorax is divided into three segments that has been fused together and the remnants of these sections can be seen by the three transverse lines of hairs on the top of the thorax. The thorax contains a lot of the muscle for larval movement and many of the longer hairs that aid in vertical support in the water. In many *Aedes* species which inhabit bottom situations and feed on fallen detritus, these hairs are much reduced, while in *Anopheles* species and *Culex* species that feed at the surface of the water, these hairs are well developed.

ABDOMEN

The abdomen is divided into ten segments with segment 8 bearing the siphon and segment 10 bearing the saddle and the anal papillae. Segment 8 contains the lateral plate or comb teeth. The abdomen usually has a lot of hairs laterally and dorsally. In various genera the shape and arrangement of these hairs can be characteristic. For example, star-like or stellate hairs are common in *Tripteroides*, while palmate hairs, much like a palm leaf, are very characteristic of *Anopheles* species. In *Anopheles* species these palmate hairs are dorsal in two rows and help to attach the abdomen of the larvae to the surface of the water as they feed.

In Culicine mosquitoes, the appearance of the lateral comb is one of the most important characteristics, while the shape of the siphon and the shape and size of the anal papillae is also quite important.

In *Anopheles* mosquitoes the details of the scoop of the spiracular plate and the arrangement of the head hairs are the most important taxonomic characters.

SIPHON

Anopheles larvae do not posses a lengthened siphon or breathing tube, but instead posses a spiracular plate which has two valves through which the larvae obtain air. This structure is suited to their feeding habit which is at the very top of the water surface. The scoop that is associated with this plate has a varied appearance and can be used to distinguish between some Anopheles species.

In Culicine mosquitoes the siphon is a very important taxonomic characteristic. The overall length and shape of a siphon is of particular use and is usually expressed as the siphon index. This is simply the ratio of the length divided by the width of the siphon at the base. This can be quite long in some *Culex* species that have a long siphon to enable them to inhabit waters where there is a very hot thin layer at the surface. It can be quite short in *Aedes* mosquitoes that feed on bottom debris, as a long siphon which would get tangled in the debris would be a disadvantage.

Some *Aedes* species have hooks at the tip of the siphon to enable them to anchor themselves to the bottom so that they can facilitate feeding in their particular habitats. The siphon also bears a row of spines or pecten teeth at the base of the siphon. The number and shape of these spines is a very good diagnostic tool. In *Aedes* species the presence or number of projections at the base of each of the pecten teeth is often a very important characteristic.

The siphon characteristically bears a variable number of paired groups of hairs on the ventral side. In *Aedes* species this is a single pair while in *Culex* it is in multiples. In some *Culex* species there maybe an additional lateral pair of hairs. These are not included when counting the number of groups of ventral hairs. Sometimes each pair may not be exactly opposite each other but usually they are the same number on one side as on the other side. If a group of ventral hairs is accidentally knocked off, the point of attachment can often be seen on the siphon as a darkened spot.

LATERAL COMB TEETH

The lateral comb teeth are very distinctive in number and shape and are very diagnostic. The lateral comb is used to clean the mouth brushes and the structure of the teeth on the comb relate to the food and the habitats of the larvae.

In some genera (*Uranotaenia*) they can be on a hardened plate while in other genera or species they can be stout spines or fringed teeth. In most *Culex* species there are a variable number of fringed teeth that make up the lateral comb. The shape and length of the various denticles or spines on these teeth is a very distinctive character. Whether the spines on the lateral comb have a distinct central denticle or there is a difference in the arrangement and shape of the denticles from the proximal to the distal groups of teeth can also be useful. It should be noted that the detailed structure of the lateral comb often requires the use of a compound microscope. Any identification of any larvae that requires the use of a compound microscope should always be checked by a person familiar with the taxonomy of mosquitoes.

ANAL SEGMENT

The main features of the anal segment are the saddle, the ventral brush and the anal papillae. The hardened saddle can be a complete cylinder or just present on the dorsal surface. In many *Culex* species the development of the saddle can be used to determine the larval stage, with the complete saddle not being present until the fourth instar stage. The saddle also contains various hairs that are important diagnostic tools. The very long projecting hairs on the posterior top of the saddle, sometimes with a fringe of teeth at the base of these hairs, is quite diagnostic in many *Culex* species. The ventral hair tuft or the ventral brush on the ventral surface of the anal segment is a single row of relatively flattened hairs that forms quite a thick brush with a fin like appearance that is used as an aid in locomotion. Some species

have a series of groups of hairs that are not attached to the main brush base and the presence and number of these pre-cratal tufts can be diagnostic.

The anal papillae are very useful tools and although their shape and length can change in relation to salinity they are very useful characters. The anal papillae are the salt regulating organs of the larvae. As a general guide, the shorter the anal papillae the greater the salinity of water in the habitat. The length of the anal papillae is usually expressed in relation to the length of the saddle. When there is a variable length of the anal papillae, the shape of the tip of the papillae, usually pointed or rounded, is an important character. There are four anal papillae and sometimes the dorsal pair can be a different length to the ventral pair. Care is needed in the preservation of mosquito larvae as these anal papillae can quite often be knocked off. Sometimes the anal papillae are relatively clear or white and cannot be seen against a white background when using a dissecting microscope. In these instances a black background will clearly show up the anal papillae.

LARVAL IDENTIFICATION

MORPHOLOGY KEYS

See diagrams of general Anopheline and Culicine mosquitoes and a key to the genera of West Australian mosquitoes. See also key and diagrams for *Aedes* species of quarantine importance in Northern Australia.

MOLECULAR TECHNIQUES

There are a number of genetic techniques available for the identification of morphological similar species in either larval or adult stages. Some of the introductory literature is listed in the references below.

REFERENCES

Beebe NW, Cooper RD, Corcoran S, Ritchie SA, van den Hurk A, Whelan PI. 2007. 'A Polymerase Chain Reaction-Based Diagnostic to Identify Larvae and Eggs of Container Mosquito Species from the Australian Region'. Journal of Medical Entomology. 44:2:376-380.

Beebe NW, Whelan PI, van den Hurk A, Ritchie SA, Cooper RD. 2005. 'Genetic diversity of the dengue vector Aedes aegypti in Australia and implications for future surveillance and mainland incursion monitoring'. Communicable Diseases Intelligence. 29:3:299-304.

Hill LA, Davis JB, Hapgood G, Whelan PI, Smith GA, Ritchie SA, Cooper RD and van den Hurk AF (2008). 'Rapid identification of Aedes albopictus, Aedes scutellaris and Aedes aegypti life stages using real-time polymerase chain reaction assays.'Am. J. Med Hyg 79:6, pp. 866-875.



Various types of Dipterous larvae

Stages in the life cycle of a chironomid



PI Whelan. Adapted from Williams (1980) Australian Freshwater Life

Figure 82: Dipterous larvae

PI Whelan. Adapted from Williams (1980) Australian Freshwater Life

Mosquito larvae

- All aquatic
- Air breathing
- No legs
- Not red
- Soft bodied with some plates
- Three distinct segments
- Hairs on head
- Antennae
- Anal segment with ventral brush and anal papillae

Larvae recognition

- Motion in water
- Return to surface
- Siphon
- Habitats
- Stages
 - Eggs, Four larval stages, Pupae
- Specimens for reference
- Keys

Figure 83: Mosquito larval features

Hardened plates Eyes Antennae Oval to round shape Hairs on head C5 and C6 Culex Inner and outer clypeal in Anopheles Mouth ventral with mouthbrush

Thorax

Three segmentsLong hairs



PI Whelan. Adapt from Dobrotworsky N V (1965) The mosquitoes of Victoria

Figure 84: Mosquito larvae - head and thorax





Figure 85: Mosquito larvae – abdomen segement VIII and IX





PI Whelan. Adapt from Lee D J (1944)

Figure 86: Mosquito larvae – siphon and anal segment

MOSQUITO SPECIES OF IMPORTANCE IN WESTERN AUSTRALIA & THE NORTHERN TERRITORY

Keys and Diagnostic Diagrams

ADULT MOSQUITOES

ILLUSTRATION OF MOSQUITO ADULT FEMALE



ILLUSTRATION OF MOSQUITO ADULT FEMALE

CHARACTERS USED TO IDENTIFY SPECIES

(Courtesy of R.C. Russell)



KEY TO THE COMMON ADULT MOSQUITOES IN THE SOUTHWEST REGION

(Includes several species commonly collected in traps which are not usually pest species*)

1. Palps as long as proboscis, wings with distinct patches of pale scales <i>Anopheles annulipes</i> Palps less than ¹ / ₄ length of proboscis, wing scales otherwise			
2. Hind tarsal segments with broad white bands			
3. Proboscis with narrow but distinct white band, scutum with distinct silvery lyre shaped pattern, femora and tibiae with longitudinal stripes			
4. Hind femur with pre-apical white band			
5. Tergal bands straight with distinctive curved lateral patches; summer species onlyAedes vigilax Tergal bands with median points and triangular lateral patchesAedes camptorhynchus			
6. Proboscis with distinct white band ~0.3 length of proboscis <i>Culex annulirostris</i> Proboscis unbanded			
7. Tergites devoid of bands or lateral patches, thorax with coarse bristles, drab species			
8. Tergites unbanded, but with lateral patches, scutumn with four (4) distinct longitudinal lines of golden or bronze scale			
9. Tip of abdomen rounded10 Tip of abdomen pointed12			
10. Tergal bands without lateral constrictions			
11. Sternites with large median patches of dark scales			
12. Scutum rich brown with broad creamy white bands laterally, tergal bands restricted to long median points extending the length of each segment			
13.Tergal bands with prominent median points, femora and tibiae mottled <i>Aedes sagax</i> Tergal bands otherwise, femora and tibiae unmottled14			
14. Scutum orange to reddish brown, dorsal abdomen with straight white bands, concave at edges			

MOSQUITO IDENTIFICATION CHART ADULT FEMALES

SPECIES FOUND IN THE MANDURAH AND PERTH METROPOLITAN AREAS Note: some other species may be found occasionally

- Aedes abdomen pointed (most), or square (Ae notoscriptus)
- Culex, Coquillettidia, Culiseta abdomen square or rounded

Anopheles annulipes

Fine bands on legs and palps. Wings with patches of pale scales. Long palps.

Aedes notoscriptus

Proboscis with narrow white band. Lyre-shaped pattern on thorax. Brilliant silver/white markings.



BANDED LEGS Aedes camptorhynchus

Dorsal abdomen has white bands with median points. Dense white scaling on lateral thorax.

<u>Aedes vigilax</u>

Grey proboscis with dark tip. Straight with bands on dorsal abdomen and offset lateral patches.





<u>Aedes alboannulatus</u>

White 'elbow' patch on hind legs. Scalloped white bands on dorsal abdomen. Mosquito often large.

Culex annulirostris

Proboscis with broad white band. Dorsal abdomen has white bands with median points. White leg bands narrow.





UNBANDED LEGS

Aedes ratcliffei

Longitudinal median pale stripe on dorsal abdomen. Thorax brown with creamy white lateral bands.



Coquillettidia sp. nr linealis

Pale stripes on thorax. Dorsal abdomen unbanded but with pale lateral patches. Large wing scales.





Aedes clelandi

Dorsal abdomen with straight white bands, concave at edges. Integument (skin) of thorax reddish brown.





Culistea atra

Slender, dark mosquito. No markings on legs or abdomen Golden scales on head and thorax.





Culex australicus

Scalloped white bands on dorsal abdomen. Ventral abdomen with line of distinct dark patches.



Culex globocoxitus

Straight pale bands on dorsal abdomen, bands wider at lateral edges. Pale scales tend to be golden.



Culex quinquefasciatus

Unusually pale and nondescript. Scalloped pale

bands on dorsal thorax (as for Cx. australicus).

Ventral abdomen without line of distinct dark patches





Aedes hesperonotius

Dorsal abdomen with straight white bands, convex at edges. Integument (skin) of thorax dark brown or black.

KEY TO THE COMMON ADULT FEMALE MOSQUITOES IN THE ARID ZONES (Gascoyne, Murchison, Goldfields and interior)

1. Palps as long as proboscis, wings with distinct patches of pale scales <i>Anopheles annulipes</i> Palps short, or no more than 2/3 length of proboscis2				
2. Palps 1/2 – 3/5 length of proboscis, large mottled species 10-12mm longAedes alternans Palps less than ¹ / ₄ length of proboscis				
3. Tip of abdomen blunt, proboscis with narrow but distinct central band, scutum with distinct silvery lyre-shaped pattern				
4. Tip of abdomen rounded				
5. Proboscis pale scaled (except tip), tergal bands with no median point, domestic species				
6. Hind tarsal segments with distinct wide white bands (≥1/6 segment)				
7. Scales on wing veins extensively mottled and triangular, tergal bands with median points				
8. Hind tarsal segments with narrow white bands				
9. Head scales (vertex) round and flat, hind femora and tibiae unmottled <i>Aedes bancroftianus</i> Head scales narrow and curved or upright and forked, hind femur and tibiae mottled				
10. Large species, integument black, median points and lateral patches on tergal bands extend length of segment				

usually less than length of segment......Aedes sagax and Aedes Marks sp. No.85

KEY TO THE COMMON ADULT MOSQUITOES IN THE PILBARA REGION (Includes several species commonly collected in traps which are not usually pest species*)					
1. Palps as long as proboscis, wings with noticeable patches of pale scalesAnopheles2 Palps short or no more than 2/3 length of proboscis					
2. Proboscis entirely dark scaled, dorsal abdomen with scales, pale wing scales greyish					
Proboscis apically pale scaled, dorsal abdomen with hairs, pale wing scales creamy					
3. Palps ¹ / ₂ - 3/5 length of proboscis, large mottled species 10-12mm long <i>Aedes alternans</i> Palps less than ¹ / ₄ length of proboscis					
4. Hind tarsal segments with distinct white bands (≥1/6 length of segment)5 Hind tarsal segments with white bands absent or narrow (≤1/8 length)8					
5. Small species, short proboscis, "hunched" appearance, tip of abdomen blunt, 4 wide tarsal bands					
6. Tip of abdomen blunt, proboscis with narrow but distinct central band, scutum with distinct silvery lyre-shaped pattern					
7. Scales on wing veins extensively mottled and triangular, tergal bands with median points					
coastal species					
8. Hind tarsal segments unbanded					

10.Head scales (vertex) round and flat, hind tibiae unmottled......Aedes bancroftianus Head scales (vertex) narrow and curved to forked and upright, hind femora and tibiae extensively mottled.....Aedes Marks sp. No.85 and Aedes sagax

12. White band on proboscis 0.3 length, tergal bands with median points....Culex annulirostris White band on proboscis ≤ 0.2 length, tergal bands straight, coastal species....Culex sitiens

13.Integument very dark grey, lateral tergal patches triangular......Aedes normanensis Integument reddish-brown, lateral tergal patches more rounded...Aedes pseudonormanensis

* Other rarer *Aedes* subgenus *Macleaya* could key out here (see Atlas of Mosquitoes of Western Australia; page 119.).

KEY TO THE COMMON ADULT MOSQUITOES IN THE KIMBERLEY REGION

1. Palps as long as proboscis Palps short or no more than 3/5 length of	proboscis
2. Palps and wings almost entirely black sc Palps and wings with noticeable patches	aledAnopheles bancroftii of pale scales
3.*Proboscis entirely dark scaled, pale wing Proboscis apically scaled, pale wing scal	g scales greyishAnopheles amictus les creamyAnopheles annulipes
4. Palps ¹ / ₂ - 3/5 length of proboscis, large m Palps less than ¹ / ₄ length of proboscis	nottled species 10-12mm longAedes alternans
5. Pale mottled species, wing scales broad Colouring and abdomen otherwise, wing	distinctive blunt curved abdomen, <i>Mansonia uniformis</i> scales narrow6
6. Hind tarsal segments 2 and 3 with white Hind tarsal segments 2 and 3 with distinct	bands absent, narrow or indistinct7 ct bands10
7. Distinct white band on proboscis Unbanded proboscis	
8.**White band on proboscis 0.3 length, ter White band on proboscis 0.2 length, ter	gal bands with median points <i>Culex annulirostris</i> gal bands straight <i>Culex sitiens</i>
9. Proboscis pale scaled ventr domestic species Proboscis dark scaled, abdomen po on dorsal thorax	rally (except tip), abdomen rounded, <i>Culex quinquefasciatus</i> inted, two distinctive lateral golden bands <i>Aedes lineatopennis</i>
10. Five (5) distinctive tarsal bands on hindFour (4) distinctive tarsal bands on hind	leg
 Tip of abdomen blunt, proboscis with distinct silvery lyre-shaped pattern Tip of abdomen pointed, proboscis grey pale, straight basal bands and pale, basa 	n narrow but distinct central band, scutum with
12. Tip of abdomen blunt, proboscis s bands on I-III, IV dark, V all white Tip of abdomen pointed, proboscis da bands on I-IV, V dark	hort, hunched appearance, broad, basal tarsal
* Other less common <i>Anopheles</i> species meraukensis (North and east Kimberley swar ** Other less common <i>Culex</i> species could be starckeae, <i>Cx</i> squamosus, <i>Cx</i> vicinus and <i>Cx</i>	es could key here; An. hilli (coastal) and An. mps). cey out here, Cx palpalis, Cx bitaeniorhynchus, Cx crinicauda.

***Old, damaged specimens of this species may appear to not have distinctive bands on hind tarsi.

Aedes alboannulatus



Aedes camptorhynchus Adult diagnostic features



Aedes clelandi Adult diagnostic feature





Aedes notoscriptus Adult diagnostic features





Aedes vigilax


Anopheles annulipes Adult diagnostic features



Anopheles atratipes Adult diagnostic features





Culex annulirostris Adult diagnostic features



Culex australicus Adult diagnostic features



Culex quinquefasciatus Adult diagnostic features



Culiseta atra Adult diagnostic features (from Liehne, PFS (1991) An atlas of the mosquitoes of Western Australia)



A SIMPLIFIED KEY TO THE FEMALES OF COMMON TOP END NT MOSQUITOES

Medical entomology branch. NT Department of Health and Community Services

This key is only a tool to help identify the most common species. Verification of any identification should be carried out from complete keys in "Culicidae of Australia" or submitted to an entomologist for confirmation.

General

Mosquitoes are two winged flies with a long proboscis and narrow wings with small scales. The legs are long and covered in scales. Only female mosquitoes bite and are distinguished from males by their much sparser antennae.

Key to Female Mosquitoes

Pictorial representation of the various steps corresponds to the same number in the key (Figure 1).

1. Palps as long as proboscis Palps not more than ¹ / ₂ length of proboscis	Anopheles2
 Palps all black and shaggy Palps all white and black bands 	Anopheles bancroftii
3. Hind legs with white bands at tip and base of each tarsi to make a broad band across the jointsHind legs not with broad bands across the joints of the tarsi	Anopheles hilli 4
4. Top and underside of abdomen almost entirely clothed with pale scales including down the middle of the underside of the abdomen Abdomen with few or no scales, or scales grouped into distinct pate	Anopheles amictus hes5
 Underneath of the abdomen with scales grouped into two distinct patches on each segment Abdomen with few or no scales 	.Anopheles meraukensis 6
6. Apical half of palps with three broad white bands and two narrow black bands. Apical ½ of proboscis usually white Apical half of palps with 3 narrow black bands, two black bands separated by a narrow white band	Anopheles annulipes Anopheles farauti s.l
7. Proboscis with a clearly defined white band not more than 1/3 length of proboscisProboscis without a complete white band, pale scaled on underside only, or mottled or all dark	
 8. Hind tarsal segment with large obvious basal white bands; hind tarsi 5 all white or almost all white Hind tarsi with very reduced white basal bands, hind tarsi 5 all dark 	
 Scutum with a lyre shaped pattern of narrow white lines, wings with dark scales Scutum mottled with pale scales. Wings with numerous pale scales 	Aedes notoscriptus Aedes kochi

10. Segments on top of abdomen with white bands produced into points in the middle. Band on proboscis about 1/3 length of proboscis. Dark bands on underside of abdomen interrupted with pale scales in the middle	Culex annulirostris
Basal bands on top of abdomen straight. Band on proboscis 1/5 or less of the proboscis. Bands on underneath of abdomen always complete	Culex sitiens
 Legs with tarsal segments banded or with many pale scales Legs with tarsi all black 	12 14
12. Proboscis black, tarsi segment 4 all black Proboscis mottled or extensively pale underneath	Aedes tremulus
 Proboscis mottled, wings mottled with broad scales. Tip of abdomen broad and deep Basal part of proboscis pale underneath, wings mostly dark scaled, tip of abdomen pointed 	Mansonia uniformis Aedes vigilax
14. Orange coloured mosquitoes, legs dark scaled with purplish reflection Brown to dark mosquitoes	Coquillettidia xanthogaster 15
15. Dark brown mosquito. Proboscis dark. Hind tibia and tarsi dark. Tip of abdomen pointed. Top of abdomen with white patches at edges of segments or with almost complete bands of pale scales Top of abdomen with basal white bands constricted at the edges. Hind tibia pale at apex. Proboscis with some pale scales undernear	16 th . <i>Culex quinquefasciatus</i>
16. Segments on top of abdomen with lateral white patches only Segments on top of abdomen with complete or almost complete sub-basal bands	Verrallina reesi Verrallina funerea



MOSQUITO SPECIES OF IMPORTANCE IN WESTERN AUSTRALIA & THE NORTHERN TERRITORY

Keys and Diagnostic Diagrams

LARVAL MOSQUITOES

ILLUSTRATION OF MOSQUITO LARVA AND

CHARACTERS USED IN LARVAL IDENTIFICATION

(Courtesy of R.C. Russell)





PI Whelan. Adapt from Darsie R F & Ward R A (1981)

KEYS TO MOSQUITO GENERA IN WESTERN AUSTRALIA 4TH INSTAR LARVAE

1.		Siphon absent; seta 1 palmate on most abdominal segments	ANOPHELES
		Siphon present; Abdominal seta 1 never palmate	2
2.		Ventral brush with a single pair of setae (sometimes with 1-2 supplementary	
		hairs in some individuals): Antenna short, without articulated apical	
		segment: sinhon with 2 or more subdorsal setae as well as subventral	
		sognon, siphon with 2 of more subdotsar source as were as subtorning	TRIPTEROIDES
		Without this combination of characters	3
2		Sinhan modified for minimum relations with a coloratized carry tooth	5
э.		Sipnon modified for piercing plant tissues, with a scierofized saw-tooth	4
		process at up	4
	-	Siphon not modified; or if so, without saw-toothed process	3
4.		Antenna with very long flexible flagellar segment	COQUILLETTIDIA
		Antenna with short rigid flagellar segment	MANSONIA
			Ma (Mnd) uniformis
5.		Antenna broad and flattened; some thoracic setae very long, others stellate;	
		tip of siphon with paired hooks and branched setae	AEDEOMYIA
		np or signoir mine pairos most of comments of the	Ad (Adv) catasticta
		Antenna, thoracic setae and sinbon otherwise	6
		Antenna, moracle source and signon other wise	Ū
		O' the still a located by the second se	
б.		Sipnon with subventral setae more numerous, or it with a single pair, then	7
		these arise at 0.2 or less of the distance from the base to apex	/
		Siphon with single pair of subventral setae arising at not less than 0.25 of the	
		distance from base to apex, usually more	8
7.		Siphon with subventral setae more numerous, occasionally forming	
		midventral row: never with separate pair arising at base	CULEX
	_	Siphon with single pair of subventral setae arising near base	CULISETA
		orphicht with onigre pair of successing court months from the	Cs (Cuc) atra
Q		Maxillary suture absent or incomplete: head setae 5-C or 6-C or both often	
0.	•	flattened herhod oning like comb often origing from lorge selerotized	
		hatened, barbed, spine-like, comb often ansing from large selerotized	LID A NOT A ENILA
		plate	UKANOTALNIA Ukano albasana
			Ur (Ura) aidescens
		Maxillary suture well developed; head hairs 5-C and 6-C sometimes single	
		and barbed, never spine-like; comb plate if present smaller	AEDES

See also the simplified pictorial adult key for south-west WA species by Peter Whelan and Gwenda Hayes on the following pages.

Simplified larval key to WA fourth instar mosquito genera

Peter Whelan and Gwenda Hayes

Centre for Disease Control Northern Territory Department of Health (Adapted from Liehne 1991 'An atlas of the mosquitoes of WA')

1a. Breathing siphon absent..... Anopheles



PI Whelan. Adapt from Russell RC (1993) Mosquitoes and mosquito borne disease in southeastern Australia

1b. Breathing siphon present..... 2



PI Whelan. Adapt from Marks EN (1982) An atlas of Common Queensland Mosquitoes

2a. Antennae short without articulated apical segment; ventral brush with a single pair of setae; siphon with 2 or more subdorsal

setae..... Tripteroides



PI Whelan. Adapt from Lee DJ (1944) An atlas of the mosquito larvae of the Australasian region



PI Whelan. Adapt from Russell RC (1993) Mosquitoes and mosquito borne disease in south-eastern Australia

3b. Breathing siphon not modified for piercing plant tissue...... 5



PI Whelan. Adapt from Marks EN (1982) An atlas of Common Queensland Mosquitoes, and Lee DJ (1944) An atlas of the mosquito larvae of the Australasian region



PI Whelan. Adapt from Russell RC (1993) Mosquitoes and mosquito borne disease in south-eastern Australia





PI Whelan. Adapt from Lee DJ (1944) An atlas of the mosquito larvae of the Australasian region



6a. Siphon with sub-ventral setae more numerous, or if with a single pair, then these arise at 0.2 or less of the distance from the base to

the apex......7



PI Whelan. Adapt from Marks EN (1982) An atlas of Common Queensland Mosquitoes



7a. Siphon with subventral setae more numerous, occasionally forming mid-ventral row; never with separate pair arising at

base..... Culex



PI Whelan. Adapt from Marks EN (1982) An atlas of Common Queensland Mosquitoes



PI Whelan. Adapt from Harbach RE & Knight K L (1980) Taxonomists' Glossary of Mosquito anatomy

8a. Head setae 5(C) or 6(B) or both often flattened, barbed, spinelike; comb often arising from large sclerotized plate...... *Uranotaenia*



PI Whelan. Adapt from Harbach RE & Knight K L (1980) Taxonomists' Glossary of Mosquito anatomy

8b. Head setae 5(C) or 6(B) sometimes single and barbed, never spine-like; comb plate if present smaller...... Aedes



PI Whelan. Adapt from Marks EN (1982) An atlas of Common Queensland Mosquitoes

Simplified larval key for south-west WA fourth instar mosquito species

1a.	Siphon absent	Anopheles
1b.	Siphon present	2
2a.	One tuft of ventral hairs on siphon	3 <i>(Aedes)</i>
2b.	More than one tuft of hairs on siphon	7 (Culex)
3a.	Head hairs 5 & 6 single	4
3b.	Head hairs 5 & 6 not single	5
4a.	Seta 1-S with three branches	Aedes notoscriptus
4b.	Seta 1-S with approximately 10 branches	Aedes vigilax
5a.	Head hairs 6 single and longer than 5	<i>Aedes clelandi</i>
5b.	Head hair 5 not single and equal to 6	6
6a. 6b.	Anal papillae pointed; lateral comb of 30-60 scales, fringed Anal papillae rounded; lateral comb of 16-28 scales denticulate	Aedes ratcliffei Aedes camptorhynchus
7a. 7b.	Antennae with milky white base; 6 groups of ventral hair tufts Antennae uniform brown/grey; 3-5 groups of ventral hair tufts with one group lateral	. <i>Culex annulirostris</i> 8
8a. 8b.	Antennae uniform grey; usually 3 groups of ventral hair tufts; siphon bottle shaped Antennae uniform brown; siphon not bottle shaped	<i>Culex quinquefasciatus</i> .9
9a.	Antennae uniform brown; 3-4 groups of ventral hair tufts; siphon long anal papillae short and rounded; anal segment setae 2, 3 branched	Culex globocoxitus
30.	of ventral hair tufts; siphon long; anal papillae usually about as long as saddle; anal segment setae 2, 2 branched	Culex australicus

See also the simplified pictorial larval key for south-west WA species by Peter Whelan and Gwenda Hayes on the following pages.

SIMPLIFIED LARVAL KEY FOR SELECTED SOUTH-WEST WA FOURTH INSTAR MOSQUITO SPECIES

Peter Whelan and Gwenda Hayes Centre for Disease Control Northern Territory Department of Health

1a. Breathing siphon absent..... Anopheles



PI Whelan. Adapt from Russell RC (1993) Mosquitoes and mosquito borne disease in southeastern Australia





PI Whelan. Adapt from Marks EN (1982) An atlas of Common Queensland Mosquitoes



PI Whelan. Adapt from Marks EN (1982) An atlas of Common Queensland Mosquitoes





PI Whelan. Adapt from Marks EN (1982) An atlas of Common Queensland Mosquitoes

4a. Setae 1-S with 3 branched tuft...... *Aedes notoscriptus* 4b. Setae 1-S with approximately 10 branched tuft......*Aedes vigilax*



5a. Head hairs 6 single and longer than 5 *Aedes clelandi* 5b. Head hairs 6 not single and equal in length to 5...... 6



PI Whelan. Adapt from Russell RC(1993) Mosquitoes and mosquito borne disease in southeastern Australia

6a. Anal papillae pointed; lateral comb of 40-60 scales, fringed...... Aedes ratcliffei



PI Whelan. Adapt from Liehne P S (1991) An atlas of the mosquitoes of Western Australia

6b. Lateral comb of 16-35 scales, denticulate; anal papillae rounded...... *Aedes camptorhynchus*



Aedes camptorhynchus

PI Whelan. Adapt from Liehne P S (1991) An atlas of the mosquitoes of Western Australia



PIWhelan. Adapt from Marks EN (1982) An atlas of Common Queensland Mosquitoes



PI Whelan. Adapt from Marks EN (1982) An atlas of Common Queensland Mosquitoes



PI Whelan. Adapt from Dobrotworsky NV (1965) The mosquitoes of Victoria



PI Whelan. Adapt from Dobrotworsky NV (1965) The mosquitoes of Victoria

Aedes alboannulatus

(from Russell, RC (1993) Mosquitoes and mosquito-borne disease in southeastern Australia)



Aedes camptorhynchus

(from Russell, RC (1993) Mosquitoes and mosquito-borne disease in southeastern Australia)





Aedes notoscriptus Larval diagnostic features

(from Russell, RC (1993) Mosquitoes and mosquito-borne disease in southeastern Australia)




Aedes ratcliffei Larval diagnostic features (from Liehne, PFS (1991) An atlas of the mosquitoes of Western Australia)



Aedes vigilax





Anopheles annulipes





Anopheles atratipes







Culex annulirostris



Culex australicus





Culex globocoxitus



Culex quinquefasciatus



Culiseta atra Larval diagnostic features (from Liehne, PFS (1991) An atlas of the mosquitoes of Western Australia)



A SIMPLIFIED KEY TO FOURTH INSTAR MOSQUITO LARVAE OF COMMON "TOP END" NT MOSQUITOES.

Centre for Disease Control Northern Territory Department of Health

This key is only a tool to help identify the most common species. Verification of any identification should be checked against a detailed description of that species or checked with complete keys or submitted to a medical entomologist for confirmation.

GENERAL

Mosquito larvae are aquatic larvae. They are found in many types of water in many different situations. They have a distinct head, thorax and abdomen, but no legs. The thorax is broader than the abdomen. Antennae are present. A compound microscope is necessary to key out many of these larvae.

Pictorial representations of the various steps correspond to the same number in the key (Figure 2)

1.	Siphon absent.	Anopheles2
	Siphon present	3
2.	Antennae with a large branched hair tuft at mid length. Inner clypeal hairs close together.	Anopheles bancroftii
	Antennae without mid length hair tuft. Inner clypeal hairs wide apart.	Other Anopheles species
3.	One pair of a group of hairs on the siphon near the midpoint of the siphon.	4
	Several pairs of groups of hairs on the siphon, sometimes reduced to small hairs.	8
4.	Lateral comb a row of pointed teeth arising from a lateral thickend plate and head hairs 5 & 6 single.	Uranotenia species
	Lateral comb not arising from a lateral thickened plate or if arising from a lateral plate then head hairs 5 & 6 are not single.	5
5.	Head about as broad as long	6
	Head distinctly broader than long.	7
6.	Lateral comb a patch of about 25 scales. Anal papillae on anal segment unequal.	Aedes notoscriptus
	Lateral comb with four long simple spines with base fused in a plate. Anal papillae are equal and long.	Aedes tremulus
7.	Siphon short about 1.6 index. Head hairs 5 & 6 single. Lateral comb with about 20 scales in a triangular patch	Aedes vigilax
	Siphon longer about 2.5 index. Head hairs 5 & 6 with 3 to 4 branches. Lateral comb 1-3 rows of scales.	Verrallina funerea

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8.	Mouth brushes modified for seizing prey. Saddle as long as siphon.	Lutzia halifaxii
	Mouth brushes not modified for seizing prey. Siphon longer than saddle.	9
9.	Head hairs 5 & 6 single or 2 branched. Lateral comb of fringed scales.	Culex (Lophoceraomyia) species
	Head hairs 5 & 6 more than two branched	10
10.	Head about same width as thorax. Antennae tuft arising about 2/3 from base of antennae.	11
	Head about 1/2 width of thorax. Antennae tuft arising at 1/2 or less from base.	14
11.	Antennae entirely grey. Siphon rather short and roughly bottle shaped. Three to four pairs of hair tufts on siphon.	Culex quinquefasciatus
	Antennae with basal 2/3 milky white, tips usually distinctly darker	12
12.	Lateral comb teeth with even fringe of fine denticles at apex.	13
	Lateral comb teeth, at least in distal row, with central denticles at apex produced into a spine	<i>Culex palpalis</i> or <i>Culex Vishnui</i> group
13.	Saddle cut away posteroventrally to half its length. Anal papillae short and rounded. Clypeal spines stout. Salt water species.	Culex sitiens
	Saddle not cut away. Anal papillae pointed and usually longer. Six pairs of hair tufts on siphon. Clypeal spines slender.	Culex annulirostris
14	Lateral comb teeth about 5-7 strong spines.	Culex bitaeniorhynchus
	Lateral comb teeth about 20 spines	Culex squamosus



MOSQUITO SPECIES OF QUARANTINE IMPORTANCE IN AUSTRALIA

Keys and Diagnostic Diagrams

ADULT & LARVAL MOSQUITOES

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Key for Aedes sp. larvae of Quarantine Importance in Northern Australia

Peter Whelan Centre for Disease Control Northern Territory Department of Health

This simplified key is for *Aedes* or species recovered from artificial containers or tree hole breeding sites. Any tentative identification of an *Aedes species* exotic to a State or Territory should be verified by a medical entomologist or specialist taxonomist.

1. Lateral comb a patch of about 25 spines, anal papillae distinctly unequal and pointed with ventral pair shorter; apex of saddle with spines; clypeal spines stout and strongly curved......Aedes notoscriptus Lateral comb a single row of spines, apex of saddle without spines; anal papillae relatively long, rounded at tips and equal or nearly so......2 2 Lateral comb a single row of 4-5 spines attached to a hardened plate .. Aedes tremulus 3 Lateral comb of about 10 (8-13) separate spines with conspicuous basal denticles 4. Lateral comb of 8-12 spines with apical portion longer than basal portion (3:2); hair 1, segment VII of the abdomen usually with 4 (3-4) branches, short, less than twice as long as hair 5, segment VII.....Aedes albopictus Lateral comb of 9-15 spines with apical portion shorter than basal portion (1:1 or less than 1:1); hair 1, segment VII of the abdomen, usually with 2 (2-3) branches, long, at least 2.5

* These species can not be readily separated as larvae. In some cases there is overlap with some of the larval characteristics between *Aedes albopictus* and *Aedes scutellaris* gp species which requires live larvae reared to adults for confirmation of identity.

Simplified larval key for *Aedes* species of Quarantine importance in Northern Australia

Peter Whelan and Gwenda Hayes Centre for Disease Control Northern Territory Department of Health (Figs, Adapted from Marks and Huang, Dobrotworsky)



2a. Lateral comb a single row of 4-5 spines attached to a hardened plate *Aedes tremulus*



3a. Lateral comb of about 10 (8-13) separate spines with conspicuous basal denticles *Aedes aegypti*



3b. Lateral comb teeth fringed basally...... 4







* These species cannot be separated as larvae

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pairs of ventral setae (VS)	Siphon index (SI) (length + width) & No. of		Anal gills (one pair each of upper & lower gills)		Lateral comb scale/ spine shape		Lateral comb	illustrations only]	Upper & lower head setae (5 & 6) [diagrammatic			ŝ	VEC
SI 2 & VS 1		Equal		Pointed with strong spines basally		A row of 9-11 spines	Contraction of the second	Long & single		Aedes aegypti	Target Contair	2477 Buckford	TOR VIGI
SI 2 & VS 1		Almost equal		Pointed & finely fringed basally	PAPP	A row of 8-12 spines	29724141419y	Branched along setae		Aedes albopictus	ner-Breeders	178 CN 167	LANCE -
SI 2 & VS 1		Distinctly unequal		Spatulate & fringed	24	Patch of 25-30 scales		Long & single		Ochlerotatus notoscriptus		NU 2002 UN	Identifyin
SI 2.3 & VS 1		Almost equal		Pointed & fused at base on plate	111	4-5 spines fused on plate		Branched at setal insertion		Ochlerotatus tremulus	Non-Ta	ANY CONTRACTOR	ng larval co
SI 2.0-2.5 & VS 1	THE REAL PROPERTY OF	Distinctly unequal		Pointed & finely fringed basally	XPV	Patch of 8-15 spines	8 P B A NY	Branched at setal insertion		Ochlerotatus palmarum	rget Container-	ALC: Y ALC: Y	ntainer-b
SI 3.5-4.5 & VS 3-4		Almost equal		Spatulate & fringed		Patch of 45-55 scales		Branched at setal insertion		Culex quinquefasciatus	Breeders	A DRUGGER OF A	reeding mo
SI 2.5 & VS 1 (no pecten)		Gills short & round		Absent (large plate bearing 2 setae)		(large plate bearing 2 setae)	And the second sec	Single		Toxorhynchites speciosus		(日)/	squitoes
If you find mosquito larvae or adults on international cargo, contact the AQIS Entomologist: 1800 020 504													

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 PATTERN ON
SCUTUM. ON SCUTUM







WHITE SCALE PATCHES ON ABDOMEN





SINGLE WHITE STRIPE ON SCUTUM



DISTINCTIVE <u>BOOMERANG</u> SHAPED SCALE PATCH









WHITE PATCHES SEPARATED FROM WHITE BANDS. ON ABDOMEN









CURVED WHITE BANDS ON ABDOMEN

, SCALES ARRANGED

VECTOR VIGILANCE Container-breeding mosquitoes



ght 2002.

Entomology, Brisbane

Aedes aegypti Adult diagnostic features



Aedes aegypti Larval diagnostic features (from Russell, RC (1993) Mosquitoes and mosquito-borne disease in southeastern Australia) 49,00044

Aedes albopictus Adult diagnostic features (from Huang 1972) ſo mid leg hind leg

Aedes albopictus Larval diagnostic features (from Huang, Y-M Contributions of the American Entomological Institute)



Aedes katherinensis Adult and larval diagnostic features (from Liehne, PFS (1991) An atlas of the mosquitoes of Western Australia)



Aedes scutellaris Larval diagnostic features (from Huang, Y-M Contributions of the American Entomological Institute)



GLOSSARY

(Courtesy of P.F.S Liehne "An Atlas of the Mosquitoes of Western Australia")

- ACUS: Sclerotised hook at the base of the siphon.
- ANAL VEIN: (A) Wing vein.
- ANAL PAPILLAE: Anal gills, organs which maintain correct water balance within the mosquito larvae. Attached to the end of the anal segment.
- ANAL SEGMENT: Terminal segment (X) of larva.
- ANTENNAL TUFT: A tuft of hairs inserted at about the midpoint of the larval antenna (seta 1-A).

ANTERIOR PRONOTUM: (APN) One of the lateral sclerites on the adult thorax.

ANTERIOR SPIRACLE: (AS) Mesothoracic spiracle on the adult thorax.

ANTHROPOPHILIC: Preferring a human host as a source of blood.

- APICAL: At the end furthest from the centre of the animal (i.e at the apex).
- AUTOGENY: Ability of the newly emerged female to produce on egg batch without first taking a blood meal.
- BASAL: At the end closest to the centre of the animal (i.e. at the base).
- CEPHALUS: Head (cephalic: of the head).
- CERCI: Pair female genital processes at the end of the adult abdomen extending beyond abdominal segment VIII in some species.
- CLYPEAL SPINE: The first of the cephalic setae (1-C) on the larval head.
- CLYPEUS: Larva: One of two chitinised plates forming the larval head capsule. Adult: A small plate directly below the torus (of little diagnostic significance).
- COSTA: (C) Wing vein at leading edge of the wing
- COXA: Articulated joint on the leg which joins leg to thorax
- CRATAL HAIRS: Modified setae on the larval anal segment where the bases of a number of paired hairs (4-X) have fused together to form a grid structure.
- CREPUSCULAR: Active at sunset and sunrise
- CUBITUS: (Cu) Wing vein
- DIMORPHIC: Occurring in two distinct morphological forms.
- DISTAL: See apical.
- DIURNAL: Activity pattern predominantly during daylight
- DORSAL: Upperside or back
- ENDEMIC: Surviving or occurring regularly or continuously in an area.
- ENDOPHILIC: Habit of feeding indoors.
- EPHEMERAL: Habitats which are marginal and suitable for mosquito breeding only on rare occasions (for example, arid zones with unpredictable and infrequent rainfall). Mosquito species which survive in such habitats.

EPICRANIAL PLATE: The lateral chitinised larval head plate holding the eyes and antenna.

- EPIDEMIC: Not occurring regularly or continuously in an area but reintroduced periodically from outside.
- EXOPHILIC: Habit of feeding outdoors.
- FEMUR: Segment of leg

- FLAGELLUM: Segment of the adult antenna. 15 flagella segments make up the adult antenna, each with a whorl of fine setae.
- FOSSA: (F) The area of the scutum just forward of the scutal angle.
- FRONS: The area on the adult head between the eyes.
- HALTERE: A vestigial wing in the adult which acts as a balance organ.
- HYPOSTIGIAL: Membranous area between the postspiracular area and the posterior pronotum.
- INSTAR: Larval development stage. It is generally the fourth instar larva that is of sufficient size to ensure identification to species level.
- LABIUM: See proboscis.
- LATERAL: At or towards the side.
- LATERAL COMB: A lateral patch of scales on the larval abdominal segment VIII.
- MEDIA: (M) Wing vein
- MENTUM: Chitinous toothed plate visible ventrally on the larval head. Used as a diagnostic feature. The overall shape and number of teeth on each side may be important for identification.
- MERON: Plate on the lateral adult thorax.
- MESEPIMERON: (MEP) A large rectangular sclerite on the adult lateral thorax.
- MESIAL/MEDIAN: At or towards the middle.
- MESOTHORAX: The middle thoracic segment. In the larva the three thoracic segments are fused but are defined by three distinct lines of setae. In the adult the mesothorax consists of the scutum and scutellum and most of the lateral sclerites and membranous areas.
- METAMERON: Plate on adult lateral thorax
- METATHORAX: The third thoracic segment. In the adult it consists of the postnotum and the metapleuron.
- MOUTHBRUSH: Dense group of hairs at the front of the larval head used to draw food particles into the mouth. In some species (*Ae alternans and Cx halifaxii*) these are modified for predation and appear as a few strongly thickened toothed bristles.
- NOCTURNAL: Biting activity at night.
- OCCIPUT: Area on the adult head above the vertex and next to the thorax.
- OCULAR SETAE: Setae on the eye border of the adult.
- ORNITHOPHILIC: Taking blood meals exclusively from birds.
- OVIPOSITION: Act of laying eggs by the female mosquito.
- PARATERGITE: A small area of the lateral adult thorax just below the scutum between the spiracle and the prealar area.
- PATHOGEN: An organism or agent of disease.
- PECTEN PLATE: The pectin are fused into a pectin plate on the lateral surface of segment VIII in larval anophelines.
- PECTEN TEETH: Row of small spines/teeth extending from base on ventral siphon.
- PLEURA: General term of thoracic segments and sclerites.
- POSTERIOR PRONOTUM: (PPN) One of the lateral sclerites on the adult thorax.
- POSTNOTUM: Part of the adult metathorax a broad plate extending from the scutellum ot the abdomen.

- POSTSPIRACULAR AREA: (PSP) A sclerite immediately below and behind the anterior spiracle on the adult thorax.
- PREALAR AREA: A raised bump on the dorsal tip of the sternopleuron.
- PRECRATAL TUFTS: Detached hairs on the larval anal segment inserted before the cratal hairs (4-X) on the grid.
- PRESCUTELLAR SPACE: (PSS) The area of the scutum medially before the scutellum.
- PROBOSCIS: The proboscis on the adult mosquito consists of an outer sheath enclosing elongate mouthparts.
- PROPLEURON: (PPL) A small raised bump above the coxa of the foreleg on the anterior edge of the adult thorax.
- PROTHORAX: The first of the three fused segments forming the thorax. In the adult it incorporates the anterior and posterior pronotum, the spiracular area and the propleuron.

PROXIMAL: See basal.

- PULVILLUS: Feathery structure in *Culex* species on the last tarsal segment (tarsi V)
- RADIUS: (R) Wing vein
- REMIGIAL BRISTLES: These bristles lie on the dorsal surface of the remigium on the wing.
- REMIGIUM: The common base of wing veins *R*, *M*, *Cu* and *A*.
- SADDLE: A chitinised plate which covers the dorsal surface of the anal segment. The saddle may completely ring the anal segment in some species.
- SCLERITES: Hardened (sclerotised) plates of cuticle, which are linked by membranous cutitcle to form the exoskeleton of the mosquito.
- SCUTAL ANGLE: (SA) A slight ridge on the scutum above the mesthoracic spiracle.
- SCUTELLUM: (Sc) A small plate joined posteriorly to the adult scutum. The scutellum has three lobes in culicines, but is a single lobe in anophelines.
- SCUTUM (S): The dorsal surface of the adult thorax.
- SETAE: Fine hairs or bristles on the adult and larval mosquitoes. A variety of larval setae are illustrated on page 366.
- SIPHON: The breathing organ attached to the larval abdominal segment VIII. Not present in anophelines.
- SIPHON INDEX: A measure of the ratio between the length of the siphon and its width at the base.
- SPIRACULAR AREA: (SP) A membranous area directly anterior to the anterior spiracle on the adult thorax.
- STERNITE: The ventral plate of each adult abdominal segment.
- STERNOPLEURON: A large sclerotised plate on the lateral thorax of the adult.
- SUBCOSTA: (Sc) Wing vein
- SUPSPIRACULAR AREA: (SSP) This is a continuation of the sclerite forming the pospiracular area and lies directly below the spiracle on the adult thorax.
- TARSI: Last five segments of the leg, the last segment usually have paired claws.
- TAXON: General term used to describe any unit in a classification hierarchy (species, genus, etc.)
- TERGITE: The dorsal plate of each adult abdominal segment.
- TIBIA: Segment of the leg.
- TORUS: The base of the antenna on the adult.

VECTOR: Species cable of or known to transmit a disease

VENTRAL: Underside or front.

VERTEX: Area above the eyes on the adult head.

VIRAEMIA: The period during which a virus is found in relatively high concentration in the blood of a vertebrate prior to the development of antibodies by the vertebrate. It is during viraemia that a vector can pick up the virus during a blood meal.

ZOONOSE: A human disease which has a basic survival cycle involving non-human vertebrates, may or may not be vector borne.

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