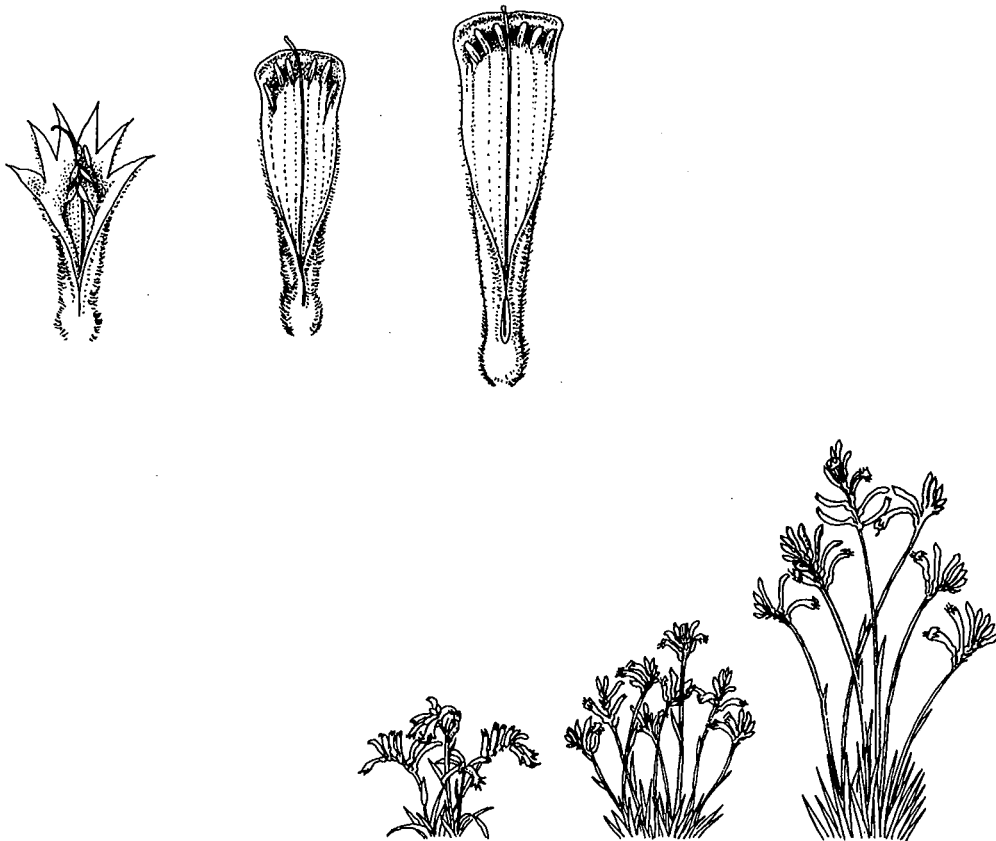


The Listing of Hybrids as Threatened Species

**PROCEEDINGS
and
RECOMMENDATIONS
of a
WORKSHOP**

HELD AT KINGS PARK AND BOTANIC GARDEN

30 JULY 1998



**DEPARTMENT OF CONSERVATION AND
LAND MANAGEMENT**

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Cover Page

A rare hybrid (centre) of *Anigozanthos humilis* (left) and *A. manglesii* (right) shows characteristics intermediate between those of its parents. From: Hopper, S.D. 1993. *Kangaroo Paws and Catpaws: A natural history and field guide*. Department of Conservation and Land Management. Perth.

BACKGROUND

One of the action items listed at the Threatened Species Scientific Committee (TSSC) meeting, held on 12 March 1998, required “.....*Dr Hopper to liaise with Dr Coates and arrange a workshop on the conservation significance of rare hybrids and natural hybridisation.....*”.

Dr Hopper agreed to host the workshop at Kings Park and Botanic Garden.

The meeting was chaired by the Department of Conservation and Land Management's Principal Research Scientist, Dr Tony Start, and organised by the TSSC.

The objectives of the workshop were to:

- discuss the implications of listing hybrids in different groups of flora and fauna;
- establish clear guidelines for listing hybrids; and
- ensure that the members of the TSSC were comfortable with the issue and interpretation of the criteria/guidelines.

Seven experts presented information on the occurrence and mechanisms of hybridisation in various flora and fauna groups before the participants addressed the objectives. This document contains their papers and the outcomes of the discussion.

Thanks to Dr A N Start for chairing the workshop, Dr S D Hopper for the use of the Kings Park and Botanic Garden's meeting facilities, and Dr K Atkins who provided valuable technical advice on this paper.

Finally, thanks to all those individuals who presented papers and participated in the discussions.

Mike O'Donoghue
Secretary (Flora)
Threatened Species Scientific Committee
4 December 1998

WORKSHOP PROGRAM

9.00 to 9.10	Convener's Welcome	Dr Tony Start
9.10 to 9.30	Introduction, overview of hybrid flora	Dr Steve Hopper
9.30 to 9.45	Hybrid flora in Western Australia	Mr Greg Keighery
9.45 to 10.00	Genetics aspects of hybrid flora	Dr David Coates
10.00 to 10.15	Hybrid invertebrates	Dr Mark Harvey
10.15 to 10.30	Hybrid amphibians	Dr Dale Roberts
10.45 to 11.00	Hybrid mammals	Dr Peter Spencer
11.00 to 11.15	Hybrid reptiles	Dr Ken Aplin
11.15 to 12.00	General comments and discussion	Dr Tony Start (facilitator)

CALM's CURRENT POLICY

Flora

The following extract is taken from the Department of Conservation and Land Management Policy Statement No. 9 "*Conservation of Threatened Flora in the Wild*" (December 1992).

"Plants which are protected flora declared under the Wildlife Conservation Act may be recommended for gazettal as declared rare (threatened) flora if they satisfy the following criteria:

- iv) In the case of hybrids, or suspected hybrids the following criteria must also be satisfied:*
 - (a) they must be a distinct entity, that is, the progeny are consistent within the agreed taxonomic limits for that taxon group;*
 - (b) they must be self perpetuating, that is, not reliant on the parent stock for replacement; and*
 - (c) they are the product of a natural event, that is, both parents are naturally occurring and cross fertilisation was by natural means"*

Note: The Director of Nature Conservation has agreed, before the Workshop, to amend (b) to include the words "*capable of being*" before "*self-perpetuating*". However, the CALM Policy Statement has not yet been formally amended.

Fauna

There is no reference to hybrids in CALM's Policy Statement No. 33 "*Conservation of Endangered and Specially Protected Fauna in the Wild*".

CONVENOR'S INTRODUCTION

A.N. Start

Welcome

Welcome. I am grateful to you for giving us your time today. I am also grateful to Steve Hopper for hosting this workshop at Kings Park and Botanic Garden and Mike O'Donoghue for arranging the program. In particular, I thank those of you who have also put time into preparing issue papers to help us set the scene. We look forward to hearing them.

Scope

There are many interesting facets to the issue of hybrids and conservation. However, today it is important that we maintain a focus on the workshop topic, **The Listing of Hybrids as Threatened Species**. Therefore, it would be appropriate if I start proceedings by outlining the scope of the topic we will be dealing with.

At their meeting on 12 March 1998, members of the Threatened Species Scientific Committee (TSSC) considered the issue of formally recognising the conservation value of rare or threatened hybrids. They concluded that it would be useful to obtain the views of a wider group of people who have expertise in that field. Thus, Action 6 from the meeting required ".....Dr. Hopper to liaise with Dr Coates and arrange a workshop on the conservation significance of rare hybrids and natural hybridisation.....". Charging two botanists with the task probably reflects the circumstances under which the Committee had usually encountered the issue. However, the scope of the workshop will not be limited to plants. Our agenda includes background papers from zoologists as well as botanists and the former will deal with invertebrates as well as vertebrates.

The Department of Conservation and Land Management has two Policy Statements which address the conservation of threatened fauna and flora in the wild. Policy Statement No. 33, *Conservation of Endangered and Specially Protected Fauna in the Wild*, is silent on the subject of hybrids. However, Policy Statement No. 9, *Conservation of Threatened Flora in the Wild*, defines the conditions under which hybrid or suspected hybrid plants may be recommended for gazettal as declared rare (threatened) flora. The policy requires that, in addition to the conditions which naturally-evolved taxa must meet:

- they must be a distinct entity, that is, the progeny are consistent within the agreed taxonomic limits for that taxon group;
- they must be self perpetuating, that is, not reliant on the parent stock for replacement; and
- they are the product of a natural event, that is, both parents are naturally occurring and cross fertilisation was by natural means.

This gives us a useful starting point for the discussion session.

Objectives

Having defined the scope, it is worthwhile reiterating the three objectives that were sent out with the agenda because we will have to keep them clearly in mind if the workshop is to produce outcomes that are useful to the TSSC. They are:

- Discuss the implications of listing hybrids in different groups of flora and fauna;
- Establish clear guidelines for listing hybrids; and
- Ensure that the members of the TSSC were comfortable with the issue and interpretation of the criteria/guidelines.

It will be easy to meet the first objective. The second one will be the key outcome provided that the members of the TSSC are comfortable with it. Many members of the TSSC are here so we should be able to accomplish the third objective. However, the TSSC will have to formally consider the outcomes of the workshop in session and accept them or not.

In either case, the TSSC will have to take the next step towards recommending modifying the policy (and, if necessary, legislation) unless it decides that the status quo is adequate. In any case, we should be able to give the Committee a good foundation from which to work.

GENERAL INTRODUCTION; FLORA

S.D. Hopper

All organisms are derived from evolutionary networks of mating relationships that stretch back through time (Hopper 1995). Natural hybridisation involves the mating of individuals from distinguishable populations, usually of distinct taxa, and the production of later-generation progeny from such mating. The key terms in the definition are: (1) "distinguishable populations", which would include populations differing in morphology, anatomy, biochemistry chromosomes, reproductive biology, ecology and/or behaviour; and (2) "later generation progeny", which include individuals produced through backcrossing of hybrids with parental individuals, or mating among hybrids themselves. The process has varied outcomes, from complete failure following mating, through successful fertilisation but subsequent abortion, to the production of hybrid offspring (sometimes sterile, sometimes not), introgression and the origin of new species. "Natural hybridisation" is, then, the above process happening in the wild, without direct human intervention while artificial or experimental hybridisation is the above process conducted through human manipulation. Natural hybridisation is relatively rare in nature, and especially so in the southwestern Australian flora, where hybrids number only 1,743 (0.6%) of some 300,300 specimens on the database in the Western Australian Herbarium.

Hybridisation may have beneficial or harmful impacts on the conservation of biological diversity. The process may lead to greater diversity (through hybrid speciation), enhanced population fitness, adaptation to new environments and improved gene dispersal, or it may result in outbreeding depression and genetic swamping, threatening small relict populations, rare species, or island endemics in particular. Ecologically, hybrid zones may support endemic insect fauna.

A sense of history and knowledge of ecological processes underpinning natural hybridisation is important to understanding present-day plants and their conservation. We need to appreciate evolutionary networks as much as we need the human networks required to manage for nature conservation.

The following abstract from Rossetto *et al.* (1997) is provided as a case study on hybrid conservation.

Eucalyptus graniticola is known from a single plant located on a granite outcrop southeast of Perth in Western Australia. Since its discovery in 1987, it has been uncertain whether this eucalypt is a relict species or a hybrid and, consequently, further study is required in order to devise appropriate conservation strategies. The similarity of features, such as leaf, bud and fruit morphology, to those of *E. rudis*, a common tree found in the vicinity, suggested that *E. graniticola* is a hybrid.

This study used random amplified polymorphic DNA (RAPD) analysis to demonstrate the addition inheritance of DNA markers from *E. rudis* and *E. drummondii*, the putative parent species, in *E. graniticola*. All the markers detected for *E. graniticola* using nine primers were shared with either *E. rudis* (40 per cent), *E. drummondii* (35 per cent) or both parent species (25 per cent).

The DNA fingerprinting results, combined with other factors, such as the segregation of cotyledon morphology, demonstrate the hybrid origin of *E. graniticola*. As a result, conservation of this rare eucalypt should rely more on *ex situ* propagation and storage than on active management.

The conservation of hybrids and hybrid zones is important and consistent with CALM's current policy in relation to the declaration of rare flora.

References

Hopper, S.D. 1995. Evolutionary networks: Natural hybridisation and its conservation significance. In: Saunders, D.A., Craig, J.L. and Mattiske, E.M. (Eds) *Nature Conservation 4. The role of networks*. Surry Beatty and Sons. Chipping Norton.

Rossetto, M., Lucarotti, F., Hopper S. D. and Dixon, K. 1997. *Heredity* 79.

WESTERN AUSTRALIA'S HYBRID FLORA

G. J. Keighery

The list of known hybrids of native vascular plants in Western Australia includes 2 Gymnosperms, 193 Monocotyledons (of which 159 are orchids), and 229 Dicotyledons (of which 105 are Eucalypts) i. e. there is a total of 424 taxa currently known.¹

There are, however, numerous examples of West Australian hybrids that have not yet been separately identified. Many of the hybrids may be under threat, predominantly from land clearing and some have almost certainly been destroyed.

¹ The full list of vascular plant hybrids recorded in Western Australia is not reproduced in this summary. It is available from Greg Keighery.

It is appropriate, therefore, to protect rare hybrids, as a means of conserving genetic diversity, and the processes that may ultimately lead to the genesis of new species. The protection of rare hybrids should also include associated vegetation and the areas in which they occur and should also include "rare varieties" as well as subspecies. Hybrids are often morphologically distinguishable and consistent, and able to replicate (even if) by asexual means.

Plant hybrids are but one form of significant genetic variation occurring below the formal taxonomic levels in vascular plant species. These include:

1) Breeding System Variants

An example of one such variant is the viviparous, pentaploid, seed sterile form of *Sporobolus virginicus*. Smith-White and Adam (1988) documented this form as only occurring as a large clone on the north side of the mouth of the Greenough River.

2) Polyploid Races, Semi-species and Biological Species

Polyploid forms are scattered through the flora of south western Western Australia. Examples include the tetraploid race of *Stylidium elongatum*, *Laxmannia omnifertilis* and the triploid/tetraploid races of *Stylidium utricularioides*. Many of these could be allopolyploids of hybrid origin which now form biological species that are reproductively isolated from their diploid ancestors. Literature searches demonstrate that approximately 10% of the flora of Western Australia contains polyploid races, but this is much higher in the arid zone, eg: *Senna* (numerous polyploid apomitic hybrid derived semi-species) and *Eremophila* (17% of the genus). Probably 30% of these races are of hybrid origin.

One example of a complex hybrid form at the diploid level has been documented between *Laxmannia sessiliflora* and *L. ramosa* at Collie by James et al (1999) where the resultant population has the morphology of one parent, the breeding system and habitat of the other and has undergone marked chromosomal repatterning. Studies by Coates reported in this volume suggests that this cryptic hybridization may be much more common than morphology suggests.

3) Morphological Forms

There are numerous distinct ecological races in the flora that show minor morphological separation, eg: Clay form of *Hakea trifurcata*, the Red Heartwood form of Tuart (*Eucalyptus gomphocephala*) and the Eagle Bay form of *Acacia pulchella*. Hybrids that occur between distinct populations within a single taxonomic may be as genetically and evolutionally important as those between formally named taxa. I have recorded a hybrid swarm between the normal *A. pulchella* and this form.

These hybrids between distinct unnamed forms may deserve recognition as much as the forms themselves.

4) Genetically distinct variants

There are numerous distinct ecological races that show minor morphological separation but major genetic distinction. Obviously these forms could hybridise and they and the parents should be recognised.

5) Hybrids

Currently there are 438² vascular plant hybrids (including 14 weeds) recorded from south western Western Australia, mostly Orchids and Eucalypts. Of particular note are six native-X-weed hybrids. These are a form of genetic pollution and, as plantings of congeneric material from Eastern Australia increases, could become a major threat to some rare taxa.

There is one obvious hybrid form worthy of listing: *Drosera nitidula* subsp. *omissa* x *D. pulchella*, which Lowrie (1989) has shown forms a pure population of many thousand plants on the northern shores of Lake Gnangara reproducing vegetatively by gemmae.

Even if only one percent of hybrids are worthy of listing there are 4-5 hybrids that currently fall into this category. The most likely genera to contain hybrids worthy of listing are: *Caladenia*, *Acacia*, *Drosera*, *Agonis*, *Eucalyptus*, *Kunzea* and some members of the family *Chenopodiaceae*. Outside the south west members of the genus *Senna* are likely candidates.

Conclusions

Hybrids, like other forms of intraspecific variations, are a natural part of the genetic variation of vascular plant species and hence contribute to the evolutionary potential of Western Australia's vascular plants. Legislation to preserve this variation should easily encompass hybrids as a necessary part of this continuum.

The listing of rare Western Australian hybrids as Declared Rare Flora, under the Wildlife Conservation Act, is consistent with existing CALM policy.

References

- Bates, R. 1985. *Checklist of Australian Terrestrial Orchid Hybrids*. Native Orchid Society of South Australia.
- Coates, D.J. and Hnatiuk, R.J. 1990. Systematic and evolutionary inferences from isozyme studies in the genus *Eremaea* (Myrtaceae). *Australian Systematic Botany* 3: 59-74.
- Craig, G.F. 1984. Reinstatement of *Spinifex sericeus* R.Br. and hybrid status of *S. alterniflorus* Nees (Poaceae). *Nuytsia* 5: 67-74.
- Hnatiuk, R.J. 1993. A revision of the genus *Eremaea* (Myrtaceae). *Nuytsia* 9: 137-222.
- Hopper, S.D. 1977. Variation and natural hybridisation in the *Conostylis aculeata* R.Br. species group near Dawesville, Western Australia. *Australian Journal of Botany* 25: 395-411.
- Hopper, S.D. 1978. Nomenclatural notes and new taxa in the *Conostylis aculeata* group (Haemodoraceae). *Nuytsia* 2: 254-264.
- Hopper, S.D., Purdie, R.W., George, A.S. and Patrick, S.J. 1987. *Conostylis*. Pp 57-110. In: A.S. George (Executive Ed.). *Flora of Australia. Volume 45*. Australian Government Publishing Service. Canberra.
- James, S.H., Keighery, G.J., Moorees, A. and Waycott. 1999. M. Genomic coalescence in a population of *Laxmannia sessiliflora* (Angiospermae, Anthericaceae): an association of lethal polymorphism, self pollination and chromosome number reduction. *Heredity* (in press).

² Based on literature records, Western Australian Herbarium records and Greg Keighery's personal data.

Lowrie, A. 1989. *Carnivorous Plants of Australia, Volume 2*. University of Western Australia Press, Nedlands.

Rechinger, K.H. 1984. *Rumex* (Polygonaceae) in Australia: a reconsideration. *Nuytsia* 5: 75-122.

Smith-White, A.R. and Adam, P. 1988. An unusual form of *Sporobolus virginicus* (L.) Kunth. *Western Australian Naturalist* 17: 118-120.

Toelken, H.R. 1996. A revision of the genus *Kunzea* (Myrtaceae): 1 The Western Australian Section Zeanuk. *Journal of the Adelaide Botanic Gardens* 17: 29-106.

Wilson, P.G. 1970. A taxonomic revision of the genera *Crowea*, *Eriostemon* and *Phebalium* (Rutaceae). *Nuytsia* 1: 3-155.

Wilson, P.G. 1980. A revision of the Australian species of Salicornieae (Chenopodiaceae). *Nuytsia* 2: 3-154.

Wilson, P.G. 1983. A Taxonomic Revision of the Tribe Chenopodieae (Chenopodiaceae) in Australia. *Nuytsia* 4: 135-262.

Wilson, P.G. 1984. Chenopodiaceae. Pp 81-317. In: A.S. George (Executive Ed.). *Flora of Australia Volume 4*. Australian Government Publishing Service. Canberra.

GENETIC ASPECTS (FLORA)

D. J. Coates

Hybridisation may result in a range of different genetic outcomes such as hybrid zones and movement of genes into one or both hybridising species (introgression), stabilised hybrid derivatives culminating in new species, occasional F1 hybrids and backcrosses. Studies undertaken in the *Stylidium caricifolium* species complex, where recent hybridisation had resulted in a hybrid zone and more ancient hybridisation, had resulted in a group of stabilised hybrid derived populations.

Research on the stabilised hybrid derived populations showed that they are fertile and produce comparable levels of seed to the parental species. Allozyme and chromosome data confirmed the hybrid origin of these populations. The allozyme studies also showed that these populations were characterised by unique novel alleles not found in either of the parental species that had presumably arisen during the hybridisation process or post hybridisation. This finding supported one view that these populations had arisen from a single hybridisation event and had subsequently expanded into the area they now occupy.

More recent preliminary studies, utilising chloroplast DNA markers, have not confirmed this single origin hypothesis. Rather they suggest the each population is the result of a different hybridisation event between the same two parent species.

These results suggest a far more complex hybrid origin of these populations than originally thought and indicate that a range of approaches and molecular markers may be required to correctly interpret hybridisation events.

The Western Australian flora, particularly in the southwest, is considered to be an ancient flora combining relictual species and suites of recently derived taxa. The complex evolutionary patterns evident in this flora are likely to be reflected in patterns of hybridisation.

To date hybridisation in this flora is considered to be less important than other mechanisms of evolutionary change and speciation. Hybridisation may, therefore, be more common and more important than previously thought but is difficult to detect and quantify in an extremely rich and diverse flora which contains a high proportion of closely related species.

Hybridisation has been shown to be a critical process in the evolution of plants and can lead to the evolution of new species through the process of "recombinational speciation" (See Rieseberg and Wendel, 1993). Therefore, the conservation of hybrids is an important part of conserving biodiversity and evolutionary processes.

References

- Coates, D. J. 1995 Chromosome re-patterning, genetic diversity and defining conservation units in Western Australian triggerplants (*Stylidium*). Pp. 9-20. In: Brandham, P. E. and Bennett, M. D. (Eds.) *Kew Chromosome Conference IV*. Royal Botanic Gardens. Kew.
- Coates, D. J. and James, S. H. 1996. Chromosome repatterning, population genetic structure and local speciation in southwest Australian triggerplants (*Stylidium*). In: Hopper, S.D., Chappill, J., Harvey, M. and George, A.S. (Eds.) *Gondwanan Heritage: Past, Present and Future of the Western Australian Biota*. Surrey Beatty and Sons. Chipping Norton.
- Reisberg, L. H and Wendel, J. F. 1993. Introgression and its consequences in plants. In: Harrison, R.G. *Hybrid Zones and Evolutionary Process*. Oxford University Press, New York.

INVERTEBRATE HYBRIDS

M. Harvey

Detecting invertebrate hybrids in the past has been fraught with difficulty, which has been mostly due to difficulties in determining the limits of species. Some older research which purported to find hybrids and hybrid zones amongst some Western Australian invertebrates, such as spiders, has been revised and updated, with evidence for interspecific hybrids now found to be lacking.

The most detailed work on Australian invertebrate hybrids was conducted during the 1970's and 1980's on various groups of stick insects and grasshoppers, with a combination of cytological and morphological work utilised to detect species, 'races' and intermediates. The basic conclusion of the work was that interspecific hybrids were unlikely to develop, and either failed to reach maturity or were totally sterile (usually as a result of incompatible chromosome arrangements). Intraspecific hybrids, usually between 'races', were detected, some of which had reduced fecundity.

Some of these hybrids possessed discrete morphological differences between the parent stock, for example, intermediate wing length between long-winged and short-winged parents in a Tasmanian hybrid zone.

Many examples of parapatry exist amongst the morabine grasshoppers mentioned above, both at the species and 'race' level, with no apparent areas of hybridisation.

There is no reason why hybridisation cannot contribute to the speciation process amongst invertebrates, but there is no confirmed evidence of this amongst the morabine grasshoppers, which have been the most extensively studied.

HYBRIDS, FROGS AND THEIR STATUS IN WESTERN AUSTRALIA

J. D. Roberts

Hybrids have been reported in four contexts in the West Australian frog fauna:

- (a) isolated instances of hybridisation between two species of apparently well-established status (eg. Main 1968; Aplin *et al.*, 1998);
- (b) back and inter-cross genotypes, possibly F1 hybrids in an area of broad overlap of two identifiably different taxa (*Pseudophryne* species: Main 1968; Roberts & Johnson unpublished data);
- (c) stable hybrid populations occurring in extended, narrow hybrid zones between readily differentiated taxa with essentially parapatric distributions (eg. *Litoria* species Cale 1991; *Crinia* species, Littlejohn 1959; Bull 1978, Bull and Blackwell 1978; possibly *Neobatrachus* species, Roberts 1997a); and

- (d) hybridisation may be involved in the origin of tetraploid species of *Neobatrachus* though the mechanisms of tetraploid origins are unresolved (Mable & Roberts 1997; Roberts 1997b).

These four situations represent radically different evolutionary problems with quite different implications for ultimate evolutionary outcomes and conservation status.

General problems relate to whether there are novel hybrid combinations that might represent either potential for new evolutionary units, or, effective avenues for gene flow between existing units leading to taxonomic sinking of one species name.

Rare hybrids represent an avenue for potential gene flow between species but this is only true if the hybrid phenotypes can mate successfully. Given the specificity of female response to male call in frogs this is unlikely unless the hybrid call contains elements essential to normal mate recognition in one or other of the hybridising taxa, or calls of the two taxa are very similar. In *Heleioporus* and *Crinia* hybrids were initially detected by their calls which were intermediate. Some shared features, eg. frequency could lead to further hybridisation but no data on fertility of F1 hybrids or viability of backcross or intercross hybrids is available. Conclusion: hybrids represent rare phenomena with some potential to affect long term evolution but of no major significance, particularly for conservation status. Moreover, hybridisation is so rare that no conservation planning is possible. If hybridisation is a result of habitat modification eliminating normal barriers to hybridisation then that is a conservation issue due to the potential impact on the parent species populations.

In this case of broad overlap with hybridisation, calls of males fall into one of two categories: characteristic of *P. guentheri* or *P. occidentalis*. There is some evidence of call structure displacement expected from reinforcement models (selection against hybrids, evolution of effective premating barriers – ie. better, own-species recognition).

Looks like a recovery from episodes of hybridisation leading to likely incorporation of alleles from alternate taxon without fusion or generation of new fixed hybrid forms.

Litoria is discussed as an example of stable, narrow hybrid zones, as many similar issues arise from *Crinia* hybrid zones.

Litoria moorei and *L. cyclorhynchus* are widespread in southwest WA and readily distinguished by differences in groin and back pattern. Groin pattern is particularly distinct: *L. cyclorhynchus* has bright yellow spots on a black background, *L. moorei* has a uniform light blue appearance.

The ranges of these two nominal taxa are parapatric but there is a range of intermediate phenotypes found in populations along the abutting range margins.

Analyses of adult male morphology confirm this pattern with some discontinuities demonstrated in relative size of the metatarsal tubercle at the same geographic location.

There are no call structure differences or differences observed are known to be related to body size or chorus density, and so could vary.

Allozyme studies of 30 loci (20 enzyme systems) showed only four loci with significant geographic variation in allele frequencies. There were no fixed differences so genotypes of hybrid pattern forms cannot be attributed to one or the other species.

From plots of genetic versus geographic distance, neighbourhood diameter was estimated at 43 km suggesting highly mobile frogs.

Immunological studies of albumin also show no differentiation (Maxson, Tyler & Maxson 1982) and unpublished analyses of RFLP patterns in mitochondrial DNA show no systematic differentiation but did show some local, single frog, variants (Wallis, personal communication). In summary, there is little or no genetic differentiation between these two taxa but a clear phenotypic difference.

How should we deal with status of a) hybrid populations b) the two nominal taxa? My conclusion is that there are no barriers to gene flow, the phenotype differences are primitive (they occur in related species from eastern Australia) and that the two taxa should be treated logically as one evolutionary unit and one conservation unit. That means no separate status for the hybrid populations.

Crinia hybrid zones are very similar, but the hybrid zone is narrower, with little or no genetic differentiation, but in this case there are no obvious phenotype differences except in male call.

Currently, both species pairs, *Litoria* and *Crinia* are named separately but there is no separate status given to hybrid populations. In both cases, "hybrid" populations contain intermediate or intergrade as well as apparently clean, parental phenotypes - they do not have any fixed genetic status.

Hybridisation in Western Australian frogs

Rare Hybrids

- Call differences: hybrid calls, genetic differences confirm F1 status (*Crinia*, *Heleioporus*)
- Chromosomally identified hybrids. (*Neobatrachus*)

Overlap with hybridisation. *Pseudophryne*

- Genetic and colour differences, call differences.
- Calls suggest no current hybridisation.
- Genetic data consistent with past hybridisation but probably not current.

Parapatric distributions, narrow hybrid zones

- Call differences (*Crinia* only)
- Little genetic differentiation (*Crinia*, *Litoria*, *Neobatrachus*)
- Chromosome differences (*Neobatrachus* only)
- Gene flow through hybrid zone (uncertain, yes for *Neobatrachus*)

Allopolyploid species

- Possibly *Neobatrachus*.

References

Aplin, K. P., Cowan M. A., Donnellan, S. C. & Roberts, J. D. 1998. Natural hybrids between the frogs *Heleioporus eyrei* and *H. albopunctatus* (Myobatrachidae, Anura) from southwestern Australia. Under revision for *Journal of Herpetology*.

- Blackwell, J. M. and Bull, C. M. 1978. A narrow hybrid zone between two Western Australian frog species *Ranidella insignifera* and *R. pseudinsignifera*: the extent of introgression. *Heredity* **40**: 13-25.
- Bull, C. M. 1978. The position and stability of a hybrid zone between the Western Australian frogs *Ranidella insignifera* and *R. pseudinsignifera*. *Australian Journal of Zoology* **26**: 305-322.
- Cale, D. 1991. *The interactions between Litoria moorei (Copland) and Litoria cyclorhynchus (Parker) at their common distributional boundary*. Unpublished MSc thesis, Department of Zoology, University of Western Australia.
- Littlejohn, M. J. 1959. Call differentiation in a complex of seven species of *Crinia* (Anura, Leptodactylidae). *Evolution* **13**: 452-468.
- Mable, B. K. & Roberts, J. D. 1997. Mitochondrial DNA evolution of tetraploids in the genus *Neobatrachus* (Anura: Myobatrachidae). *Copeia* **1997**: 680-689.
- Main, A. R. 1968. Ecology, systematics and evolution of Australian frogs. *Advances in Ecological Research* **5**: 37-86.
- Maxson, L. R., Tyler, M. J. & Maxson, R. 1982. Phylogenetic relationships of *Cyclorana* and the *Litoria aurea* species-group (Anura: Hylidae): a molecular perspective. *Australian Journal of Zoology* **30**: 643-651.
- Roberts, J. D. 1997a. Geographic variation in calls of males and determination of species boundaries in tetraploid frogs of the Australian genus *Neobatrachus* (Myobatrachidae). *Australian Journal of Zoology* **45**: 95-12.
- Roberts, J. D. 1997b. Call evolution in *Neobatrachus* (Anura: Myobatrachidae): speculations on tetraploid origins. *Copeia* **1997**: 791-801.

HYBRID MAMMALS

P. Spencer

There are no known rare mammal hybrid species occurring in the wild in Western Australia. However, there has been little research work done in the field in this State to date.

Some research has been carried out on Rock Wallabies at Macquarie University (see further notes below). However, no real systematic approach has been undertaken.

Some hybrids of *Petrogale* and *Thylogale* had been artificially produced in the laboratory.

A small number of viable *Thylogale* had apparently been produced in the Eastern States. Therefore, there is the potential for hybridisation to occur (in the Eastern States) where there are contact zones. However, there are no contact zones in this State for these species.

In mammalian systems in Australia, the best-documented studies are between species of *Petrogale*.

The following species are viable hybrids of Australian mammals:

Petrogale

Almost all species that have contact zones have been able to be hybridised in experimental conditions. Some of these are viable, others are not. Much of the work to date has been undertaken by Rob Close at Macquarie University. In WA the Warburton range rock-wallabies have been shown to contain hybrids.

Thylogale

Shown that there are viable F1 hybrids between the two species of pademelons (*T. stigmatica* and *T. thetis*). These two species have range overlaps.

Macropus

Macropus agilis x *M. rufogriseus*: Agile and Redneck wallabies produce viable F1 female crosses.

M. dorsalis x *M. parma*: Black-striped and Parma wallabies produce viable F1 female crosses.

Rodents

In respect to Australian native rodents Baverstock has compiled some data on endemic rodents, a large amount of which remains unpublished. However, hybridisation between species of native rats, particularly *Rattus* spp. has been noted.

Possums

T. caninus x *T. arnhemensis*: Mountain (Bobuck) and Northern Brush-tail Possum data (A. Kerle)

Ancient hybrid zones occur where, presumably, speciation occurs more frequently than in other areas of both species' distribution. Consequently, these zones should be given high conservation priority. Some natural hybridisation has, and will continue to, occur on various island/mainland species.

Issues

- What impact does fragmentation and environmental disturbance cause?
- What will occur in species regeneration (reintroduction) programs where geographical barriers may be overcome?
- Should hybrids be used to maximise or increase the genetic base of existing species?

HYBRIDS AND TAXA OF HYBRID ORIGIN IN THE AUSTRALIAN HERPETOFAUNA.

K. Aplin

Introduction

The following categories need to be clearly distinguished in any discussion of hybrids and taxa of hybrid origin within the WA reptile fauna.

1. Artificial interspecific crosses (product of interspecific matings in captivity; often involving pairs of taxa which do not occur sympatrically in nature).
2. Natural interspecific hybrids. These can vary in frequency of occurrence from rare to common, and the outcome can vary from infertile F1 hybrids through to full genomic introgression. Probably the most significant context is a 'hybrid zone' where two closely related taxa interbreed at medium to high intensity in a localised area of sympatry or parapatry. Hybrid zones are of special interest to evolutionary biologists and may also represent a significant factor in the origin of genetic diversity through reticulate evolution.

The International Code of Zoological Nomenclature (1985: Article 23h) specifically excludes individuals of hybrid origin from being the name bearing type of either of the specific parental taxa.

3. Discrete populations or taxa (ie., historical "individuals") which have arisen as a consequence of interspecific hybridisation. Such taxa may exhibit diploid sexual reproduction but can also be polyploid and may also exhibit various modes of clonal reproduction (most commonly, parthenogenesis). Considerable debate has taken place over the nomenclatural status of "taxa of hybrid origin".

The modern consensus is that formal recognition may be warranted in certain cases, but that caution needs to be employed in application of binomial nomenclature in situations where repeated, independent hybridisation events give rise to multiple clonal lineages (Frost & Wright 1988, Echelle 1990).

Survey of Western Australian Reptiles

Documented instances of each category involving at least one taxon native to WA(*) are listed below. Additional notes are provided in several cases.

1. Artificial interspecific crosses

Boidae

*Morelia spilota** x *M. amethistina*

*Morelia spilota** x *Liasis fuscus**

Elapidae

*Acanthophis praelongus** x *A. hawkei*

Scincidae

*Egernia stokesii** x *E. hosmeri*

*Tiliqua rugosa** x *T. scincoides**

*Tiliqua rugosa** x *T. nigrolutea*

*Tiliqua scincoides** x *T. nigrolutea*

2. Possible natural interspecific crosses

Elapidae

*Acanthophis pyrrhus** x *A. wellsei**.

*Pseudonaja affinis** x *P. nuchalis**.

Scincidae

?*Lerista maculosa** (possibly *L. lineopunctulata* x *L. uniduo*).

3. Populations or taxa of known/likely/possible hybrid origin

Typhlopidae

"*Ramphotyphlops*" *braminus** - triploid, parthenogenetic?

Gekkonidae

*Heteronotia binoei** (A6 x SM6 'cytotypes') - triploid, parthenogenetic populations.

Scincidae

?*Egernia stokesii aethiops** (possibly *E. stokesii stokesii* x *E. stokesii badia*).

*Menetia greyii** ('northern' x 'southern' species) - triploid, parthenogenetic? populations.

Summary

There are very few well-documented natural interspecific hybrids within the WA (and Australian) reptile fauna. To some extent, this reflects the lack of detailed taxonomic research in many groups, and the near universal lack of genetic investigation. In two genera, interspecific hybridisation has resulted in triploid parthenogenetic lineages.

These are of considerable scientific interest. The *Heteronotia* parthenogens are geographically widespread and abundant. The *Menetia* parthenogens are currently known from a single area in WA and represent the first recorded instance of polyploidy/parthenogenesis within the cosmopolitan family Scincidae.

One currently recognised subspecies of skink (*Egernia stokesii aethiops*) is of possible hybrid origin (*stokesii* x *badia* ?). One other "poorly-known" taxon (*Lerista maculosa*) may comprise individuals of hybrid origin.

As in most other vertebrate groups, hybridisation in reptiles only rarely ever gives rise to new evolutionary lineages (ie. 'taxa'). However, hybridisation may be more commonplace within the group than is currently understood.

Additional notes for examples

1. Probable hybrid zone exists between Giralalia and Cane River (Aplin & Donnellan *in prep.*). Not yet confirmed genetically.
2. Possible hybrids identified from within zone of distributional overlap, including northern metropolitan region. Not yet examined in detail and no genetic data available.
3. Taxon described from two specimens from different localities in the Shark Bay region. Possibly represent individuals of hybrid origin (*L. uniduo* ? x *L. lineopunctulata* ? Aplin *in prep.*).

4. A pan-tropical species of uncertain affinities. At least some populations are triploid parthenogens. Recorded from a few localities in northern Australia. Possibly introduced.
5. Widespread triploid clonal populations have apparently arisen on multiple occasions through crossing of different 'cytotypes' of *H. binoei* (representing different subspecies or species; taxonomy not yet resolved).

The clonal lineages are evidently long-lasting and have succeeded in colonising habitats not currently occupied by the sexual parentals. The fact of multiple origins will impose difficulties in application of binomial nomenclature to the clonal lineages.

6. The subspecies '*aethiops*' combines morphological features of each of *stokesii* and *badia* and occupies a geographic position on the boundary between 'coastal' (ie. continental shelf) and 'inland' habitats. It is thus possible that *aethiops* may be of hybrid origin; genetic data are needed to confirm this.
7. Hybrid populations are known from the Cape Cuvier area in WA and from northern SA. Triploidy has been confirmed for the latter population; both populations appear to be all-female. The taxonomy of *Menetia greyii* is very confused and a major revision is required.

References

- Echelle, A.A. 1990. Nomenclature and non-Mendelian ("clonal") vertebrates. *Systematic Zoology*. **39**: 70-78.
- Frost, D.R. & Wright, J.W. 1988. The taxonomy of uniparental species, with special reference to parthenogenetic *Cnemidophorus* (Squamata: Teiidae). *Systematic Zoology*. **37**: 200-209.
- ICZN (1985). *International code of Zoological Nomenclature*, 3rd Edition. University of California Press, Berkeley.

DISCUSSION SUMMARY

Mike O'Donoghue and A.N. Start

Workshop participants noted the existing CALM policy relating to flora hybrids

"Plants which are protected flora declared under the Wildlife Conservation Act may be recommended for gazettal as declared rare (threatened) flora if they [also] satisfy the following criteria:

iv) In the case of hybrids, or suspected hybrids the following criteria must also be satisfied:

- (a) they must be a distinct entity, that is, the progeny are consistent within the agreed taxonomic limits for that taxon group;*
- (b) they must be [capable of being] self-perpetuating, that is, not reliant on the parent stock for replacement; and*
- (c) they are the product of a natural event, that is, both parents are naturally occurring and cross fertilisation was by natural means"*

and agreed that thrust is appropriate but suggested that "were/" should be added to Clause iv (c) before the second "are". The Clause would read as follows:

"(c) they are the product of a natural event, that is, both parents were/are naturally occurring and cross fertilisation was by natural means".

The amendment would ensure that the intent of the Clause would be retained where one or both the parental taxa of stable hybrids may be lost.

The workshop noted that the Director of Nature Conservation has already agreed to change (b) by the addition of the words "*capable of being*" before "*self-perpetuating*" and endorsed the decision.

In reviewing the information provided on hybrid fauna, the workshop participants noted that there has been little call for listing hybrids so far, but noted that knowledge of the topic is limited and concluded that it would be appropriate for CALM policy to cover hybrids so that:

- a) if zoologists identify hybrid populations that warrant protection as threatened and valuable genetic entities, they will be able to prepare nominations which address the agreed criteria for assessment, and
- b) if the TSSC receives nominations, it will have criteria by which to evaluate them.

The participants also discussed various issues associated with the proposed revision of the Wildlife Conservation Act that relate to conserving hybrids, particularly "specially protected areas".

They emphasised the need to protect the process as well as the results of natural hybridisation involving native taxa. Consequently, it was agreed to recommend to the Department of Conservation and Land Management that there is a need to retain "specially protected areas" in the draft bill to allow for the protection of hybrid zones as well as threatened ecological communities.

In some circumstances hybridisation can be regarded as genetic pollution. Examples include:

- introduced taxa that hybridise with indigenous taxa of fauna and flora. The results could be detrimental to the conservation of native species, particularly where hybridisation might cause any indigenous species to disappear as a discrete genetic entity. This is a particular concern where it affects rare species.
- the range of one indigenous taxon extending until it comes into contact with another, threatened taxon with which it can interbreed. Again, it is possible that the threatened taxon could disappear as a discrete genetic entity. Corellas in south western Australia was cited as one example.

In some cases the progeny of these 'polluting' processes could be rare and/or threatened genetic entities. Nevertheless they should not be protected. The current wording of the Wildlife Conservation Act requires a taxon to "be in need of special protection" to be listed as threatened flora or fauna. The Act thus precludes such 'genetic pollutants' being declared "threatened species". It was agreed that a similar provision should be included in any revision of the Wildlife Conservation Act.

RECOMMENDATIONS

Workshop Participants recommend that:

1. CALM Policy Statement No. 9, *Conservation of Threatened Flora in the Wild*, be amended as follows:

The criteria which hybrid, or suspected hybrid plant, taxa must meet for declaration as rare (threatened) flora should read as follows (ie. underlined words being added):

- (b) they must be capable of being self perpetuating, that is, not reliant on the parent stock for replacement; and
- (c) they are the product of a natural event, that is, both parents were/are naturally occurring and cross fertilisation was by natural means.

2. CALM Policy Statement No. 33, *Conservation of Endangered and Specially Protected Fauna in the Wild*, be amended by inserting the following:

In the case of hybrids, or suspected hybrids, the following criteria must also be satisfied:

- (a) *they must be a distinct entity, that is, the progeny are consistent within the agreed taxonomic limits for that taxon group;*
- (b) *they must be capable of being self perpetuating, that is, not reliant on the parent stock for replacement; and*
- (c) *they are the product of a natural event, that is, both parents were/are naturally occurring and cross fertilisation was by natural means.*

3. Provision for declaration of areas as "specially protected areas" should be retained in the Wildlife Conservation Bill to allow for the protection (*inter alia*) of hybrid zones.
4. Any revision or replacement of the Wildlife Conservation Act should retain the concept of rare or threatened flora or fauna needing special protection before being considered for declaration as threatened under the Act.

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