The development of a new toxic 1080 bait for fox control: 'Pro-bait'.

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Abstract

1) An initial trial revealed foxes were 11% less likely to ingest Probaits than dried meat baits. Possible reasons for this difference were i) foxes found Probaits less palatable than dried meat baits or ii) Probaits were less available to foxes due to weathering or greater invertebrate damage
2) Enhancements to Probaits were undertaken and these included assessing the use of flavour enhancers to increase Probait palatability to foxes and assessing the field longevity of baits which incorporated an invertebrate repellent (Coopex) and/ or a different binder which made the Probaits harder
3) Seven flavour enhancers were field tested to determine the product most palatable to foxes. The flavour enhancers tested were hexylamine, caproic acid, a commercial 'beef' flavour, a commercial 'chicken' flavour, monosodium glutamate, honey and a combined beef and honey mixture.
4) Field trials were undertaken to compare the longevity of dried meat baits and several variations in Probaits ingredients. Six bait types were investigated: 1) a standard Probait, 2) a Probait incorporating the invertebrate deterrent 'Coopex' 3) a 'hard' Probait including a different binder, 4) a hard Probait plus 'Coopex', 5) a FoxOff bait, and 6) a dried meat bait. Samples of each bait type were trielled in four locations: Collie, Manjimup, Kalgoorlie and Narrogin. Bait samples were collected after 1 day, 4 days, 7 days, 14 days, 21 days, 28 days, 35 days, 42 days, 49 days, 56 days, 63 days and 70 days. The change in the weight of baits and the proportion of the bait that had been removed by invertebrates were used to assess the differences in bait-longevity at each of the four sites.
5) When the most useful additional ingredients had been identified (i.e. chicken flavour enhancer and the invertebrate repellent 'Coopex") these were added to Probaits and the uptake of the 'improved' Probait by foxes was compared with that of dried meat baits in the field. Baits were labelled with biomarkers and delivered to Charles Darwin Reserve and Lochada station. Cyanide baiting was used to obtain fox carcases six weeks later.
6) Captive trials were undertaken to determine the risk of Probaits to non-target fauna (Martin *et al.* 2002). The two species identified to be potentially at risk from Probaits were *Phascogale tapoatafa* and *Dasyurus geoffroii*. Field trials were undertaken to examine the risk of using Probaits operationally on populations of these two species. The survival of radio-collared individuals of both species was monitored throughout toxic Probait baiting programs.

Introduction

In Western Australia foxes are controlled by repeatedly baiting prescribed areas with dried meat baits containing the poison 1080. Baiting programs are now undertaken in vast tracts of CALM estate and other landholdings. Large numbers of dried meat baits are used each year and these are currently obtained from the Department of Agriculture at a

cost of more than $1 each. In order to reduce the cost of its baiting programs CALM has decided to develop and manufacture an alternative, cheaper fox bait; 'Probait'. The new Probaits are anticipated to cost less than 50 cents each.

Before the new fox bait, 'Probait' could be used operationally it was necessary to ensure that it is as palatable to foxes as the currently used dried meat bait. A previous experiment indicated Probaits in their current form were 11% less likely to be ingested by foxes than the dried meat bait (Marlow 2000). The formulation of Probaits was therefore modified to try to increase the palatability of baits and to enhance the longevity of the baits in the field to increase their uptake.

Methods

Increasing the palatability of 'Probait' to foxes by including flavour enhancers

The original Probait sausage contained 70% minced kangaroo meat, 20% pork fat and 10% canine 'digest' (a commercial flavour enhancer for dog food). A salami-style binder is added to this mixture to promote the hardening of the final product. (This reduces the risk of non-target species being able to eat the bait). Each bait weighs approximately 80-85g at manufacture and is then heated to 60°C to promote the hardening process. The baits are then dried to approximately half their weight before delivery to the field.

A trial of seven flavour enhancers was undertaken to determine if any of these might be more palatable to foxes than others. The flavour enhancers trialled were honey, a commercial chicken flavour (Magnum Essence chicken flavour #110), ethyl caproate (i. e. caproic acid which smells similar to decomposing meat), hexylamine (also smells like decomposing meat), a commercial 'beef' flavour, a combination of the beef and honey flavours, and monosodium glutamate (a general flavour enhancer). These chemicals were compared for their appeal to foxes by placing in them in a simulated bait matrix lure on cyanide capsules. Two capsules were placed at each cyanide station, one with a 'test' lure and one with a standard lure. The lure on the capsule that killed each fox was assumed to be the more appealing. These trials were undertaken in a pastoral area on Karara and Thundalarra stations where no recent fox control had been undertaken. **S**tandard cyanide transects were used which included 100 bait stations per 20 km transect.

Determining if invertebrate repellents and binder induced hardness increase field longevity of Probaits.

A trial to determine the relative field longevity of six bait types was undertaken. Baits were monitored for 10 weeks at four sites: Collie, Manjimup, Kalgoorlie and Narrogin. The six bait types examined were 1) a standard Probait, 2) a Probait containing the invertebrate deterrent 'Coopex', 3) a Probait with a harder consistency, 4) a hard Probait containing 'Coopex', 5) a FoxOff bait, and 6) a dried meat bait. A sample of four baits of each of the six types was collected after 1 day, 4 days, 7 days, 14 days, 21 days, 28 days, 35 days, 42 days, 49 days, 56 days, 63 days and 70 days. The change in the weight of

baits and the proportion of the bait that had been removed by invertebrates were used to assess the differences in bait-longevity at each of the four sites.

Comparing the uptake of 'enhanced' Probaits by foxes

The composition of Probaits was modified when the results of the flavour enhancer, invertebrate repellent and bait binder trials revealed the optimum components for baits. The relative uptake of modified Probaits and dried meat baits by foxes was then tested at two sites in the semi-arid zone of WA: Charles Darwin Reserve (previously White Wells station) and Lochada station.

The two bait types were labelled with different biomarkers so that foxes that had ingested either of the baits could be identified. The biomarkers iophenoxic acid and tetracycline were used. Iophenoxic acid raises blood iodine levels and can be detected by analysing a blood sample. It was added to the baits as a solution. This solution was prepared by dissolving iophenoxic acid crystals in 100% alcohol. Each 0.6 ml of the solution contained 20mg of iophenoxic acid and this volume was added to each bait using a standard dosing gun. The baits were kept upright as they dried and the iophenoxic acid crystals remained in the bait as the alcohol evaporated.

The biomarker tetracycline produces a characteristic fluorescent ring in the canine teeth of any fox that ingests it. Tetracycline was added to baits as a powder using a modified syringe. This was used to insert 150mg of tetracycline into a hole that had been drilled into the side of the bait using an electric drill. The presence of this biomarker in foxes is observed by taking a very thin section of each of the fox's canine teeth using an Isomet saw. These sections are then examined under a microscope using ultraviolet light (450*nm*) that reveals the presence of fluorescent rings. A sample of seven captive foxes were fed tetracycline labelled baits and were killed with cyanide 16 days after they had ingested the bait. All of these foxes revealed tetracycline rings in their teeth.

Both bait types were delivered to both study sites at the standard baiting rate of 5 baits km^{-2} (i.e. 10 baits km^{-2} will be laid). On Charles Darwin Reserve dried meat baits were labelled with iophenoxic acid and Probaits were labelled with tetracycline. On Lochada station Probaits were labelled with iophenoxic acid and dried meat baits were labelled with tetracycline.

The baits were aerially delivered to both sites on 26/2/ 2004. An area of approximately 400 km^{-2} was baited at each site and the position of the plane when the baits were dropped was recorded using a GPS. Fox carcases were retrieved from both sites 6 weeks later using cyanide baiting (13-23rd March 2004). This interval was chosen because foxes are known to continue to take baits for up to six weeks after they are delivered (Thomson *et al.*, 2000). Cyanide baiting involved placing two cyanide capsules every 200 m along transects within the study site. The capsules were covered with a condensed milk/ icing sugar lure. Foxes died very close to where they bit the cyanide capsule and it was relatively easy to collect the carcases. Cyanide transects of 20 km in length were positioned each night and areas that revealed fresh fox tracks in the morning were

sampled repeatedly. Fox carcasses were retrieved as early as possible each morning. A blood sample was obtained from each fox and the canine teeth were collected.

The blood samples were frozen and sent to the Department of Natural Resources in Queensland to have their iodine concentration determined. The samples were not centrifuged because they had been obtained from foxes that had been dead for up to 12 hours. The iodine concentration of the whole blood samples was obtained by initially 'digesting' the sample with hot perchloric acid to release the protein-bound iodine and oxidising the iodine to iodide in the process. This iodide was then used to catalyse a caesium arsenic redox reaction which proceeded at a rate proportional to the iodide concentration. Over a set period of time the change in absorbance (measured at 420nm) is proportional to the iodide and consequently the total iodine concentration. Samples with concentrations higher than the standard ranges were re-run in dilution with deionised water.

A background sample of blood iodine levels was obtained from four foxes in Charles Darwin reserve on 26-27th February 2004 so that the level of IPA that indicated whether a fox had ingested a bait could be calculated. The concentrations of blood iodine observed in these foxes were added to those from samples obtained from another four foxes obtained during a trial undertaken in 1999.

Non-target species testing:

Probaits were also required to be no more of a threat to non-target fauna than dried meat baits. The native fauna in Western Australia has a natural tolerance to the poison used in the fox baits ('1080') but if small native animals eat sufficient material from a fox bait, they could still be at risk of being poisoned due to their smaller body size. The native species that are at potential risk from fox baiting campaigns in Western Australia were identified by considering the diet, weight and the tolerance to 1080 of all native species that are likely to be present in the baited areas (Martin *et al.* 2002). The species identified to be most at risk from toxic Probaits were *Phascogale tapoatafa (*brush-tailed phascogale*)* and *Dasyurus geoffroii* (chuditch*)*. The uptake of toxic Probaits by these two species was examined in field trials (see Marlow *et al.* 2005 and Morris *et al.* 2005 for reports on Probait uptake by brush tailed phascogales and chuditch respectively).

Results

The results of the field trial investigating the foxes' preferences for seven different flavour enhancers revealed the chicken flavour to be the only enhancer for which foxes had any preference (Table 1).

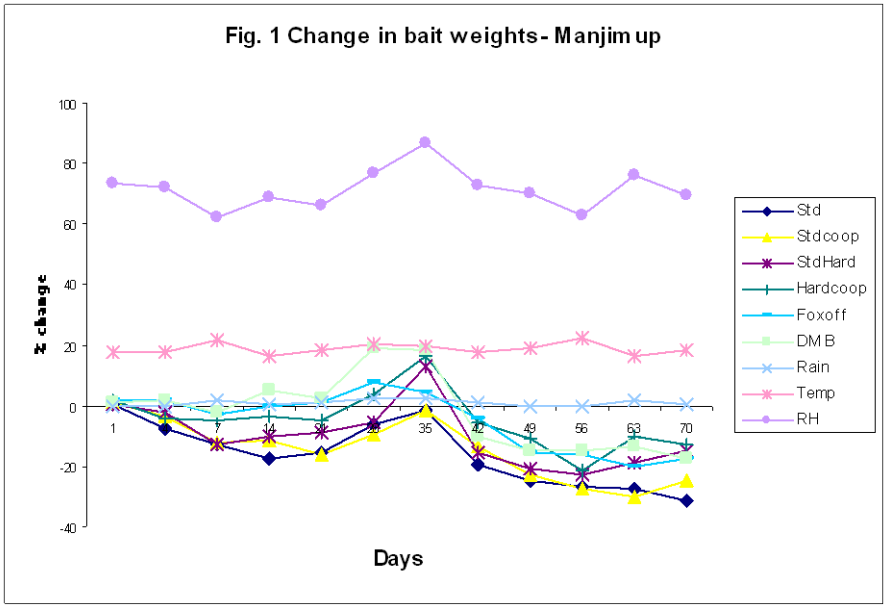
Table 1: Foxes' preferences for flavour enhancers

Flavour	Chi^2	Probability
Honey	0.2	n.s.
Chicken	8.88	$0.001<p<0.01$
Ethyl caproate	0.0	n.s.

Hexylamine	2.63	n.s.
Beef	0.7	n.s.
Beef and Honey	0.2	n.s.
Monosodium glutamate	0.2	n.s.

Bait longevity:

The assessment of each bait types' field longevity was confounded by the fact that any material remaining after invertebrate attack increased in weight when it rehydrated after rain. Thus a bait that had not been attacked by invertebrates and had not rehydrated after rain could weigh the same as a bait that had been extensively attacked by invertebrates and exposed to considerable rainfall. There was a trend for bait weights to be correlated with relative humidity (Figs. 1-4) but these relationships were not statistically significant ($p>0.05$).



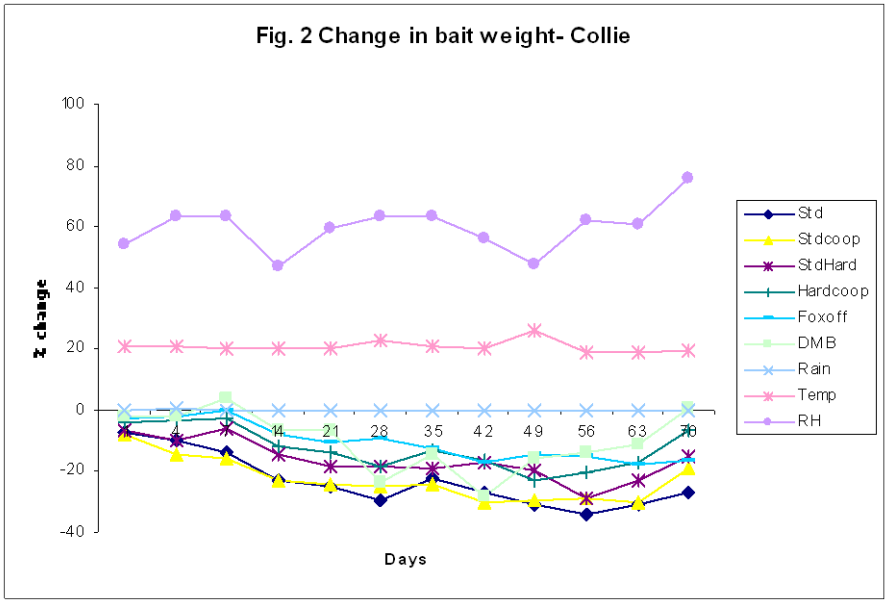


Fig. 2 Change in bait weight- Collie
% change
100
80
60
40
20
0
-20
-40
1
4
7
14
21
28
35
42
49
56
63
70
Days
Std
Stdcoop
StdHard
Hardcoop
Foxoff
DMB
Rain
Temp
RH

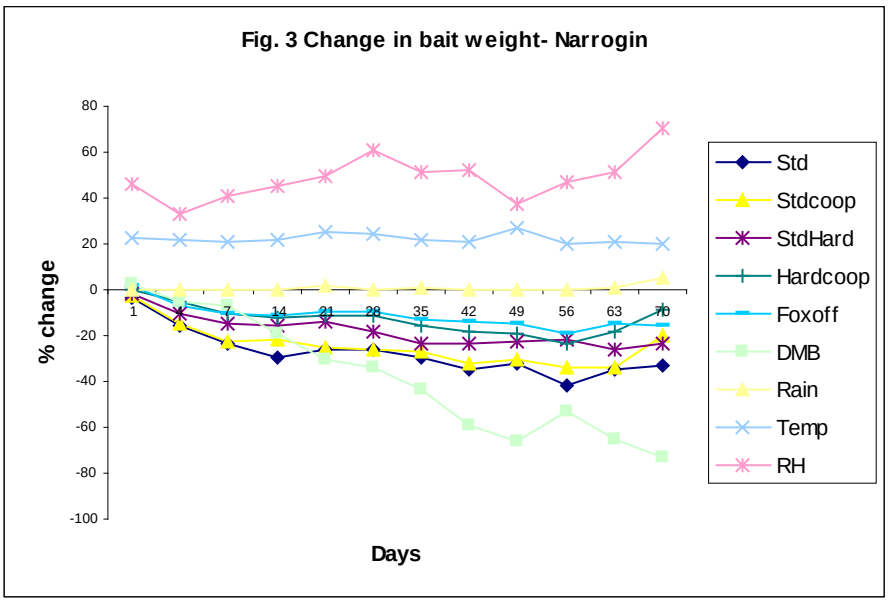
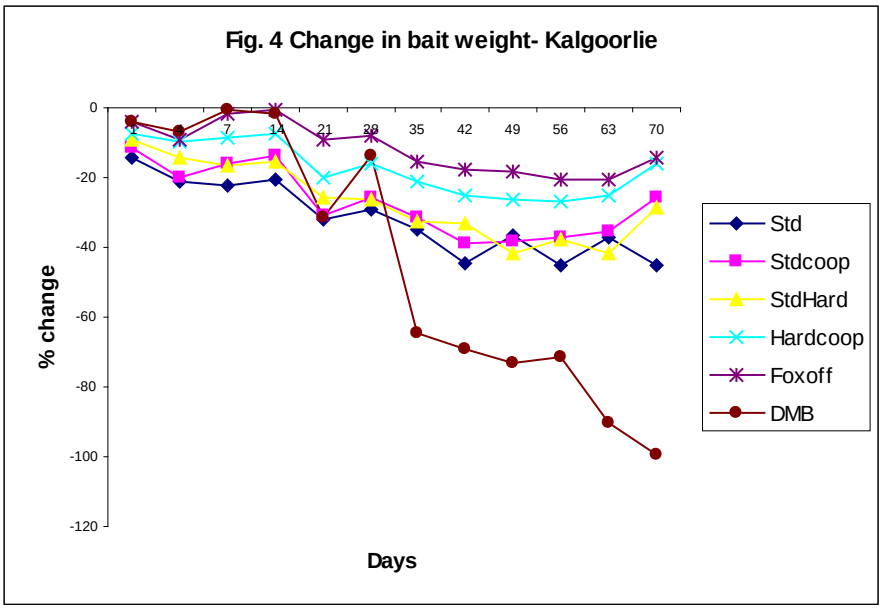


Fig. 3 Change in bait weight- Narrogin
% change
80
60
40
20
0
-20
-40
-60
-80
-100
1
7
14
21
28
35
42
49
56
63
70
Days
Std
Stdcoop
StdHard
Hardcoop
Foxoff
DMB
Rain
Temp
RH



Invertebrate attack:

The amount of invertebrate attack on each of the six bait types was recorded for all four sites (Figs 5-8). The overall impact of invertebrates was also investigated by combining the results from all four sites (Fig. 9). The standard Probait with Coopex withstood invertebrate attack significantly more than the standard Probait ($p=0.02$). Similarly the Hard Probait with Coopex withstood invertebrate attack significantly more than the standard Probait ($p=0.02$). However, increasing the hardness of the bait did not increase its longevity and there was no significant difference in the invertebrate attack between the standard Probait and the Hard Probait ($p=0.2$). The dried meat baits suffered most from invertebrate attack and were ingested significantly more by invertebrates than the standard Probait ($p= 0.001$). The invertebrate species responsible for the invertebrate attack varied between the four sites. At Manjimup and Collie the damage was done by *Demestes* beetle larvae and there were also dipteran eggs within the baits. At Narrogin and Kalgoorlie the main removal of bait material was by meat ants.

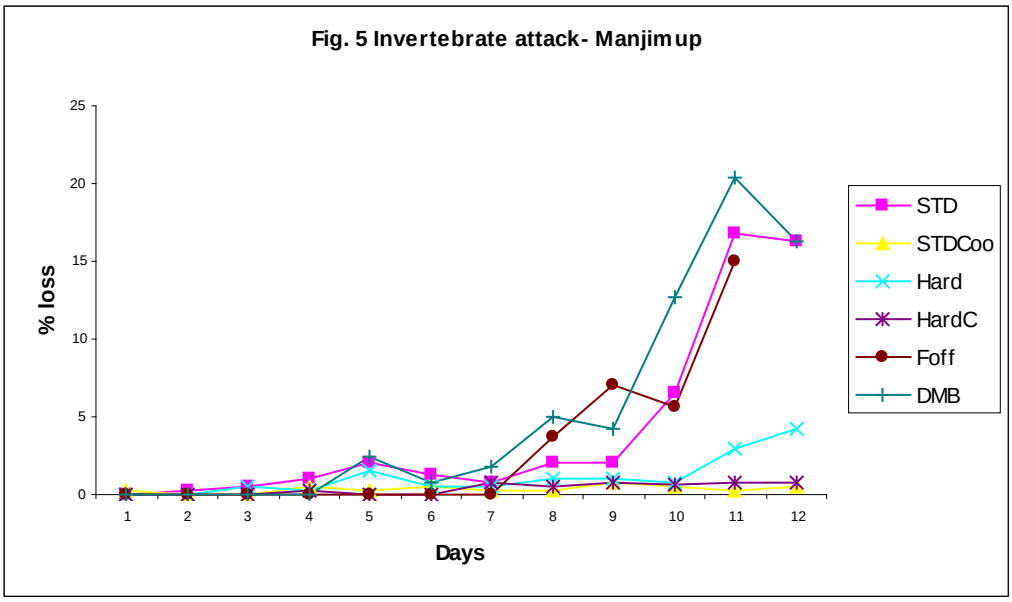


Fig. 5 Invertebrate attack- Manjimup
% loss
25
20
15
10
5
0
1
2
3
4
5
6
7
8
9
10
11
12
Days
STD
STDCoo
Hard
HardC
Foff
DMB

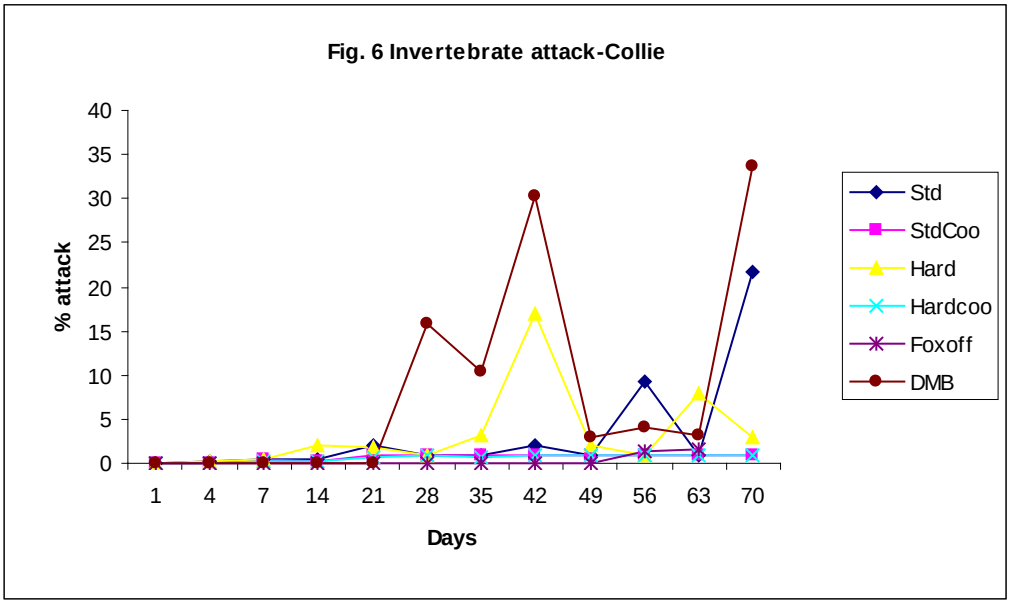


Fig. 6 Invertebrate attack-Collie
% attack
40
35
30
25
20
15
10
5
0
1
4
7
14
21
28
35
42
49
56
63
70
Days
Std
StdCoo
Hard
Hardcoo
Foxoff
DMB

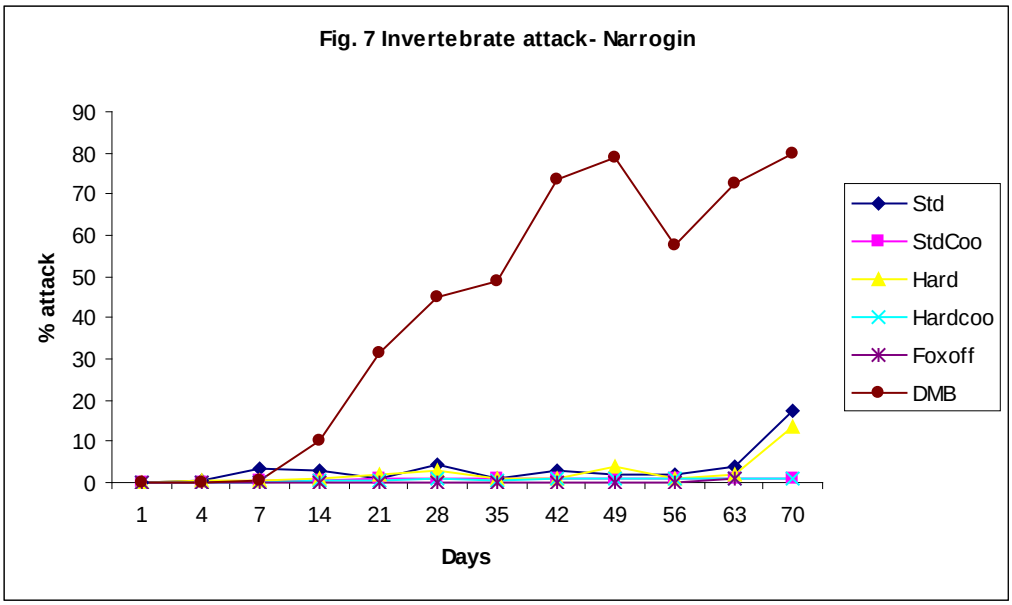


Fig. 7 Invertebrate attack- Narrogin
% attack
90
80
70
60
50
40
30
20
10
0
1
4
7
14
21
28
35
42
49
56
63
70
Days
Std
StdCoo
Hard
Hardcoo
Foxoff
DMB

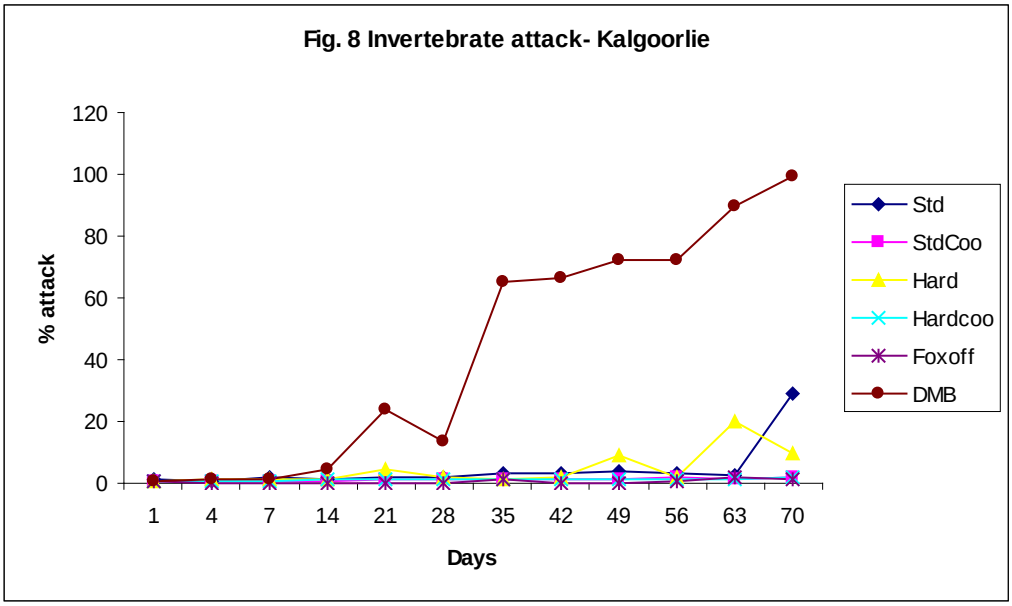
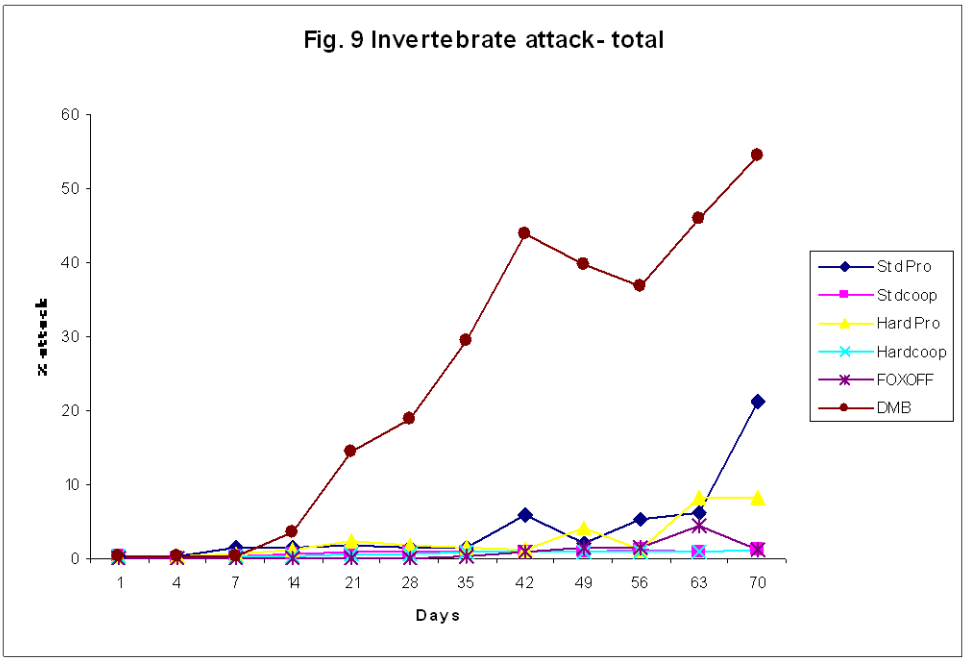


Fig. 8 Invertebrate attack- Kalgoorlie
% attack
120
100
80
60
40
20
0
1
4
7
14
21
28
35
42
49
56
63
70
Days
Std
StdCoo
Hard
Hardcoo
Foxoff
DMB



Comparing the uptake of enhanced Probaits and dried meat baits in the field

50 foxes were collected from Lochada station and 53 from White Wells station.

The rate of bait uptake for the two baits when results from the two study sites were combined was not significantly different (Table 2). The proportion of foxes that ingested each bait type was 87%.

Table 2: Uptake of biomarkers by foxes at White Wells and Charles Darwin reserve

	Dried meat bait	Probait	X^2	*p*
Lochada station	44/50	40/48	0.44	n.s.
Charles Darwin Reserve	46/53	48/53	0.38	n.s.
Total	90/103	88/101	0.00	n.s.

Non-target bait uptake trials

The studies of the uptake of toxic Probaits by brush tailed phascogales and chuditch revealed populations of both species were not at risk from operational Probait baiting programmes (Marlow *et al.* 2005 and Morris *et al*. 2005, respectively).

Discussion

In an attempt to increase the uptake of Probaits by foxes the palatability and field longevity of baits was 'improved'. Seven flavour enhancers were investigated to determine if foxes had any preferences and only the commercial chicken flavour revealed a positive response.

The field longevity of Probaits was investigated and bait longevity was shown to increase when invertebrate attack was reduced by adding an invertebrate repellent, 'Coopex'.

The formulation of Probaits was then modified to include the chicken flavour enhancer and the invertebrate repellent. The uptake of the modified Probaits was compared with that of dried meat baits. Baits were laid at the standard baiting rate of 5 baits km^{-2} (Thomson and Algar 2000) on both Lochada station and Charles Darwin Reserve. Baits were delivered by air and each bait contained a biomarker so that if it was ingested by a fox its presence could be detected. Cyanide baiting was then implemented and over fifty fox carcasses were recovered from both sites. There was no significant difference in the detection of biomarkers in foxes from either site and therefore no difference in the uptake of Probaits and dried meat baits had occurred.

Earlier captive trials had suggested the brush-tailed phascogale (*Phascogale tapoatafa)* and the chuditch (*Dasyurus geoffroii)* may be at risk from toxic Probait fox control programmes. Subsequent field trials revealed neither species to be at risk from the operational use of Probaits (Marlow *et al.* 2005 and Morris *et al.* 2005 respectively) .

Now that the uptake of Probaits has been shown to equal that of dried meat baits, and that the two non-target species considered to be most at risk from poisoning by Probaits have been shown to be survive toxic bait deliveries, CALM can switch to the exclusive use of Probaits in all its Western Shield sites. This has potential to ensure that supply of baits is uninterrupted and if savings are made the program may be expanded into new areas.

Summary

The uptake of modified Probaits by foxes is not significantly different to that of dried meat baits.

The uptake of modified Probaits by non-target species, the brush-tailed phascogale (*Phascogale tapoatafa)* and the chuditch (*Dasyurus geoffroii),* does not pose a threat to populations of these species in the field.

CALM can now switch to the exclusive use of modified Probaits in all its Western Shield sites. This will ensure an uninterrupted supply of baits and may lead to savings which will enable the Western Shield program to be expanded to new areas.

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