

# EFFICACY OF DIFENACOUM BAIT FORMULATIONS AGAINST SEMI-WILD AND WILD *RATTUS RATTUS* FROM THREE DIFFERENT GLOBAL LOCATIONS

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**Abstract** *Rattus rattus* is considered to have a less omnivorous diet than *R. norvegicus*, preferring fruit, seeds and grains. Therefore, the efficacy of existing commercial (Sorex) difenacoum rodenticide bait formulations against *R. rattus* was uncertain and studies were undertaken to determine their efficacy. The difenacoum bait formulations tested were one of the following a pellet, a paste, a gel, a wax block or an oat based bait. The rats used in the studies were either wild or semi-wild and in either a semi-natural (simulated use) or natural (field trial) environment at three different global locations. The results establish that the Sorex difenacoum-based formulations will provide good to excellent control of *R. rattus* in the field. The comparative performance of the baits can be attributed to their different composition and physical features and the general food preferences of the *R. rattus*.

**Key words** bait acceptance, bait palatability, rodent control

## INTRODUCTION

*Rattus norvegicus* is thought to have a less omnivorous diet, preferring fruit, seeds and grains. Therefore, the efficacy of five existing difenacoum bait formulations; a pellet, a paste, a gel, a wax block and an oat based bait, against these rats was uncertain. The aim of the studies was to determine the efficacy of the existing difenacoum bait formulations against *R. rattus* from three global populations.

## MATERIALS AND METHODS

### Rats

The *R. rattus* used were colonies of wild-derived rats bred and maintained in a self-contained pen. The Queensland *R. rattus* were collected by live trapping on Gatton Campus, University of Queensland, Australia. The *R. rattus* used in Tokyo were a natural wild population. The resistance status of all rats was unknown. The pre-test census/control diet was a standard rat and mouse expanded laboratory diet supplied by B & K Universal Limited, Hull, England. The Queensland pre-test census/control diet was a standard laboratory rodent diet used in Australia (Riverina Rat and Mouse Cubes produced by Riverina Stock Feeds). All difenacoum baits used contained a nominal 50 ppm difenacoum and were supplied by Sorex Limited.

### Test Pen

The test enclosure/pen, measured 2.5 x 1.1 x 2.1 m (l x w x h) with solid walls and floor and a wire mesh ceiling, containing a discrete colony of *R. rattus*. The Queensland pen measured 2.3m x 1.5m x 1.8m (l x w x h). All pens contained bedding, harbourages, toys for environmental enrichment with control diet and water being available throughout the study. The control diet was presented in a single container placed in the middle of the pen. For the UK studies, ambient conditions were  $21 \pm 2^\circ\text{C}$ , relative humidity  $55 \pm 10\%$ , under a 12 h light, 12 h dark light cycle. For the Queensland studies, the pens were housed indoors in a large steel roofed shed with concrete walls and floor, and the rats were under a natural day night cycle, daily maximum temperature  $18 \pm 2^\circ\text{C}$  and daily minimum  $14 \pm 2^\circ\text{C}$ .

### Feeding Studies

Choice feeding efficacy studies were undertaken using established guidelines accepted in the United Kingdom and EU under EU Directive 98/8/EEC, against populations/colonies of *R. rattus* from three

different global locations. The UK studies were undertaken on semi-wild rats in semi-natural conditions, which was a colony held permanently in a pen/enclosure. The Queensland studies were undertaken by the University of Queensland on wild rats caught locally, acclimatised and held in semi-natural conditions, *i.e.* a pen/enclosure. The Tokyo study was undertaken on wild rats in their natural environment, *i.e.* a field trial study.

### **Simulated Use Studies**

The pen studies consisted of an acclimatisation period of either 2 months (at UK) or 1 month (at Queensland) followed by a 3 day pre-test census period and a test period of up to 20 days. For each of the pen studies the pre-test census diet was always presented in the same container in the same position as for the acclimatisation period. The diet was weighed and then re-weighed after 24 h and the amount of diet taken was calculated by subtraction, this represents pre-test diet take. The amount of pre-test census diet taken was recorded for three consecutive 24 h periods.

The test period followed immediately after the pre-test census period. The control diet was as for the acclimatisation and pre-test census periods, *i.e.* the same diet in the same container in the same position. Four test bait points were placed in the pen. Each bait point consisted of an aluminium pot, containing 150 — 300 g of bait, placed on an aluminium tray. Two of the bait points were placed on the ground level and two bait points were placed on the shelf area. Bait and control diet were, if necessary, topped up to avoid complete takes. Daily takes of the bait at the four bait points and control diet at the central feeding point were measured every 24 hours by weighing.

### **Bait Palatability**

The palatability of the bait was determined by comparing bait take and control diet take. The two factors calculated for each study were palatability ratio and percentage acceptance of the bait. Palatability ratio (T/C) and percentage acceptance (% A) were calculated each day for the first 4 days. The Palatability ratio (T/C) = the total test bait eaten / total control diet eaten, the Percentage Acceptance (% A) = (Total test bait eaten / Total test bait + Total control diet eaten) x 100.

The mean of the first 4 days palatability ratios was calculated to give the overall palatability ratio for the bait. Beyond the first 4 days the T/C and % A were not calculated as symptoms of anticoagulant toxicity and mortality distorted results. During the test period, the bodyweight, sex and days to death of dead and moribund rats was recorded. At the end of the test period any survivors were weighed, sexed and culled. Control was assessed by recording the number of dead and surviving rats and calculating the % mortality. At both the UK and Queensland sites, where more than one study was undertaken on a bait formulation, the mean values from these studies was calculated to give the overall mean T/C and the overall mean % A. The test period was terminated after three consecutive days of no takes were recorded or total mortality was achieved or after 20 days, whichever was the sooner.

### **Natural Study**

The field trial was undertaken in a nine storey commercial building. The infestation was throughout the building with a restaurant at the centre of the infestation. The rats were reported to be extremely neophobic. Control diet (cut wheat) and tracking patches were used for the pre- and post-test census assessments. Control was assessed using pre- and post-test censusing and estimating the percentage mortality. The T/C was a qualitative calculation; T/C = initial bait take / pre-test census take.

### **Data Analysis**

Statistical analysis (using MINITAB) was carried out to establish if there was a significant difference between the palatability of the baits. This involved using analysis of covariance (ANCOVA) to compare the amounts of bait ingested with the total amount of food (bait plus control diet) ingested. The significance level used was the  $p = 0.05$  level.

## RESULTS AND DISCUSSION

The results of the Sorex and Queensland simulated use studies are summarised in Table 1 and Table 2, respectively. The results of the Tokyo field study are shown in Figure 1. The comparison of UK and Queensland results, using ANCOVA, is summarised in Table 3.

**Table 1.** Summary of results from pen studies (UK) on difenacoum bait against semi-wild *Rattus rattus*.

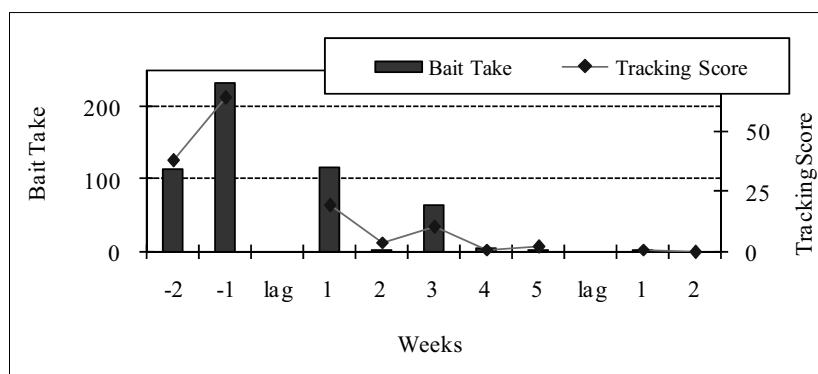
Difenacoum Bait	No. of Studies	Overall Mean % Control	Overall Mean T/C (Mean %A)
Oats	2	100	1.59 (59)
Paste	1	98	0.87 (46)
Pellet	4	97	0.81 (42)
Gel	1	100	0.54 (35)
Block	2	90	0.26 (21)

**Table 2.** Summary of results from pen studies (Queensland) on difenacoum bait against semi-wild *Rattus rattus*.

Difenacoum Bait	No. of Studies	Overall Mean % Control	Overall Mean T/C (Mean %A)
Pellet	3	97	0.55 (35)
Paste	2	96	0.45 (30)
Block	3	82	0.32 (24)
Gel	3	91	0.03 (3)

**Table 3.** Comparison of UK and Queensland results using ANCOVA.

Difenacoum Bait	P value	Interpretation
Pellet	0.39	No Sig Diff
Paste	0.000	Sig Diff.
Block	0.003	Sig Diff.
Gel	0.000	Sig Diff.



**Figure 1.** Summary of Tokyo field trial with difenacoum pellet bait; pre- and post census control diet and tracking score indicates activity.

### CONCLUSIONS

Generally, palatability/acceptance of the difenacoum baits was greater at the UK site than at Queensland. This could be attributed to the fact that at Sorex the rats were bred and maintained within the test pen while the Queensland rats were wild caught, albeit with an acclimatization period. It was expected the paste and gel baits would have greater palatability due to their moist texture and sweet / fruity flavour. However, for the gel bait, although it was still effective at controlling *R. rattus*, it was less palatable than expected. Although the wax block was not the most palatable bait formulation it still effected good to excellent control.

The Tokyo field study on the Sorex difenacoum pellet bait gave excellent control in a difficult environment. The results established that difenacoum based formulations will provide good to excellent control of *R. rattus* in the field. The comparative performance of the baits can be attributed to their different composition and physical features and the general food preferences of the *R. rattus*.

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