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Laboratory evaluation of pyriminyl used as a rodenticide against the lesser bandicoot rat, *Bandicota bengalensis**

BY JOE E. BROOKS AND PE THAN HTUN
Rodent Control Demonstration Unit, World Health Organization, Rangoon, Burma

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SUMMARY

The properties of pyriminyl (*N*-3-pyridylmethyl-*N'*-*p*-nitrophenyl urea) as a rodenticide against the lesser bandicoot rat (*Bandicota bengalensis*) in Rangoon, Burma, were investigated in the laboratory. The acute LD 50 and LD 95 dose of orally administered pyriminyl for *B. bengalensis* were found to be 6·7 mg/kg and 23·0 mg/kg of body weight respectively. When caged bandicoots were given a choice between plain and poisoned baits, the optimum rodenticidal concentration in the bait was found to be 0·25–0·5 %. Symptoms of pyriminyl poisoning appear from 1 to 4 h after feeding starts, giving individual animals time to consume from 2 to over 30 LD 50 doses of 0·5 % pyriminyl before feeding stops. Deaths occurred from 4 to 96 h after either oral dosing or free-choice feeding. There appeared to be no significant aversion to the poison at 0·25 % or 0·5 % concentration in foods. The potential hazards and use of pyriminyl as a field bait against populations of *B. bengalensis* are discussed.

INTRODUCTION

In parts of India and Burma the lesser bandicoot rat, *Bandicota bengalensis*, has within recent times become an urban pest rodent of significant importance. In rapidly expanding cities such as Bombay, Calcutta, Madras and Delhi the relative prevalence of this species as a member of the urban small mammal fauna has increased at the expense of *Rattus rattus* and *R. norvegicus* (Deoras, 1969; Rao, 1947; Seal & Banerji, 1969; Spillett, 1968). In our own observations in Rangoon, we have noted that during the past 45 years *B. bengalensis* has displaced *R. norvegicus* from outdoor habitats and the latter species now lives almost exclusively indoors (Walton *et al.* 1977).

Control of the lesser bandicoot rat is necessary both for economic and public health reasons. In grain storage facilities and food shops, bandicoots consume and contaminate large quantities of human food (Parrack, 1969). The lesser bandicoot causes economic damage to structural foundations, streets and sidewalks. The species is also of considerable importance because of its association with plague. *B. bengalensis* has been found susceptible to *Yersinia pestis* infection and isolations of the organism have been made from the species in several areas of India (Bhat-

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nagar, 1966). Evidence of natural plague-infected lesser bandicoots was detected recently by serological methods in extensive areas within Rangoon (Brooks *et al.* 1977).

Almost nothing is known about the toxicity of rodenticides to *B. bengalensis*. Harrison & Woodville (1948, 1950) reported some investigations on the use of barium carbonate against this species in Rangoon and found the optimum concentration to lie between 10% and 20%. Barium carbonate, however, is no longer generally used in rodent control.

Zinc phosphide in concentrations of 1–5% has been widely used in the control of lesser bandicoots (Barnett & Prakash, 1975; Deoras, 1968). Srivastava, Tripathi, Pandya & Awasthi (1968) reported an LD 50 for zinc phosphide against *B. bengalensis* of 2·51 mg/kg body weight. This degree of toxicity seems extremely doubtful in light of the LD 50 of zinc phosphide for various species of *Rattus* of between 23 and 41 mg/kg (Barnett, Cowan, Radford & Prakash, 1975; Hood, 1972; Schoof, 1970). In our own investigations we have observed the LD 50 of zinc phosphide against female lesser bandicoots to be 26·5 mg/kg (unpublished observations). Much work yet remains to be done on the characterization and efficacy of zinc phosphide against this species.

Muktabai, Krishnakumari & Mujumder (1968) reported an LD 50 of 90 mg/kg with magnesium ammonium arsenate but found this material was generally unpalatable at concentrations required in field baits. Norbormide was found by several investigators (Deoras, 1965; Kapoor, Ramisivan & Krishnamurthy, 1965) to create no mortality in *B. bengalensis* at dosages of 0·05% or more. Because of this paucity of information on acute toxic rodenticides used against *B. bengalensis*, we undertook a study of the response of this species to several acute toxicants. One of the materials studied was a relatively new rodenticide, pyriminyl (*N*-3-pyridylmethyl-*N'*-*p*-nitrophenyl urea), also known as RH-787 and Vacor. Pyriminyl is reported to act as a metabolic inhibitor of nicotinamide in the rat and several other animals (*Chem. Eng. News*, 1975) causing death from paralysis and pulmonary arrest. This paper reports the results of our laboratory evaluation of this material against *B. bengalensis* from Rangoon, Burma.

METHODS AND MATERIALS

The animals

Bandicoots were captured in wooden live-traps made locally, usually baited with dried fish. Captured animals were returned alive to the laboratory while still in the traps. Animals used in cage tests were run out of the traps into dark bags, weighed, sexed and dusted with insecticide to kill ectoparasites. The insecticide used, pirimiphos methyl, has a very low toxicity for rats (oral LD 50 about 2000 mg/kg) and there was no apparent effect on the animals. Rats were then caged individually and given laboratory meal (a locally-milled diet consisting of 8 parts whole dried fish meal, 8 parts pressed peanut meal, 25 parts crushed rice and 59 parts whole wheat meal and wheat flour) with free access to water and allowed to acclimate to cage conditions for a period of several days to a week.

Animals used for intubation studies were simply run out of the traps into a dark cloth bag, sexed, weighed and, without anaesthesia, given a dose of pyriminyl (using the method described by Redfern, 1971). They were then caged, given food and water and set aside for up to 6 days to observe mortality.

LD 50 determinations

Pyriminyl was administered orally by syringe and a ball-tipped 18-gauge needle. Technical crystalline pyriminyl was dissolved in propylene glycol in concentrations ranging from 0·025% to 0·5%. This allowed a range of dosages from 2·5 mg/kg to 50 mg/kg. Controls received propylene glycol only, in amounts approximating what the poison-dosed animals received. The results of the acute toxicity assay were analysed by the method of Litchfield & Wilcoxon (1949).

Efficacy determinations

Caged bandicoots were given a free choice between two food cups containing plain laboratory meal only for several nights. If food consumption was normal, then pyriminyl was substituted in one of the food cups but offered in the same dietary base for two additional nights. Food cups were alternated in placement each night and were staggered alternately from cage to cage when first placed in order to avoid the bias of position preference. Pyriminyl was offered in baits in concentrations ranging from 0·05% to 1·0% by weight. All amounts eaten each night from each cup were measured to the nearest 0·1 g. Poisoning symptoms and mortality effects were observed for up to 8 days after the poison offering. A longer observation period of mortality was used for bait-fed rats because death was delayed longer than in stomach-tubed animals, especially at the lower concentrations.

Latency

Observations on latency (the period between the beginning of feeding and its termination due to the onset of poisoning symptoms) were carried out by offering rats caged individually a no-choice feeding upon normal laboratory diet containing 0·5% pyriminyl. Five animals, two males and three females, were observed quietly from nearby in the evening and the times and duration of feeding were noted. The animals had been previously preconditioned to feeding in the evening under a lamp.

RESULTS AND DISCUSSION

Oral intubation tests

The results of oral intubation tests with pyriminyl against lesser bandicoot rats are given in Table 1. While the acute oral LD 50 value is given for each sex, there was no significant difference between the sexes in their response. In fact, a highly significant correlation exists for the dose–response curves between sexes, $r = 0{\cdot}996$ ($P < 0{\cdot}001$, Simpson, Roe & Lewontin, 1960). The combined-sex LD 50 figure of 6·7 mg/kg compares closely to the figure of 4·75 mg/kg given for male Norway

Table 1. *Toxicity of Pyriminyl to* B. bengalensis

(Means with standard errors.)

	Males	Females	Total
No. rats tested	58	76	134
Body weight (g)			
Mean	250·7 ± 15·9	272·5 ± 10·8	263·1 ± 9·2
Range	50–585	71–449	50–585
LD 50, mg/kg	6·2	7·2	6·7
(95 % fiducial limits)	4·0–9·5	5·0–10·3	5·0–9·0
LD 95, mg/kg	—	—	23·0
(95 % fiducial limits)	—	—	14·6–36·1

rats, the most sensitive known species (Peardon, 1974). The acute oral LD 95 was estimated to be 23·0 mg/kg.

The first visible signs of illness in rats given a 50 mg/kg dose appeared between 1 and 2 h after intubation, with deaths occurring between 3½ and 5½ h. At doses of 25 mg/kg symptoms first appeared between 3 and 4 h and death occurred from 5 to 18 h after dosing. Some animals given lower doses died as long as 96 h later. The mean time to death for 89 rats at all doses was 1·73 days. Thus, the period elapsing between dosing and both the onset of poisoning symptoms and time to death are related to the dosage.

Poisoning symptoms and pathology

Pyriminyl is thought to act through metabolic inhibition of nicotinamide in the rodent body. We found the primary symptoms of pyriminyl poisoning in the lesser bandicoot rat are (1) a sluggishness that sets in about 1–3 h after intubation, (2) the animal then drops, with its back hunched and the fur erected, (3) shortly thereafter it falls onto its side or lies flat with the hind limbs extended behind, (4) flaccid paralysis of the hindlimbs becomes noticeable about 2–3 h after intubation, (5) breathing becomes laboured and (6) death occurs from 4 to 96 h later. When animals receive the material in food, we usually found them dead 18 h later or lying prostrate with hind-limb paralysis; infrequently the fore limbs were paralysed. Usually, even though paralysed, the animal was able to lift its head and act defensively by attempting to bite. Occasionally we noted a thick lachrymal secretion from the eyes both in dead rats and those still alive but paralysed.

At autopsy the prominent finding was moderate to heavy haemorrhage of the lungs (although we rarely noticed evidence of bleeding from the nostrils). Haematuria was seen in about 5 % of the rats. Gastrointesinal irritation and haemorrhage were seen less frequently.

Several rats received sub-lethal doses of pyriminyl, became paralysed in the hind limbs but did not die. If food and water were offered where the rat could pull itself with its fore limbs (if these were not affected), the rat would survive for a prolonged period (5–10 days) and several recovered completely from the paralysis within 7–10 days. We observed no residual damage to the hind limbs and full

Table 2. *Observations on duration of feeding by* B. bengalensis *on bait containing* 0·5 % *priminyl*

Sex	Weight (g)	Time spent in feeding (min)	Duration of feeding period (min)	Amount of food consumed during feeding period (g)	Result (18 h)
♀	371	61	200	17·4	Dead
♀	289	24	225	5·9	Dead
♀	319	30	222	8·0	Dead
♂	331	44	52	10·4	Dead
♂	455	17	230	6·9	Dead

Table 3. *Results of giving lesser bandicoot rats a choice for* 2 *days between plain bait and bait containing pyriminyl*

Conc. (%)	Sex	Mean body weight (g)	Mortality	Mean bait intake (g)		Mean and range of doses (mg/kg) of active ingredient taken by rats that	
				Poison	Plain	Died	Survived
1·0	♂	305	4/4	1·6	11·6	40 (0–88)	—
1·0	♀	315	9/9	7·4	8·7	252 (17–1271)	—
0·5	♂	371	5/5	2·1	7·6	29 (18–36)	—
0·5	♀	314	7/7	3·7	9·1	60 (26–115)	—
0·25	♂	412	4/5	5·9	2·1	39 (28–55)	17
0·25	♀	317	11/12	5·7	9·3	46 (17–58)	?
0·1	♂	332	5/5	6·8	1·1	21 (16–25)	—
0·1	♀	335	4/5	10·9	7·8	28 (25–36)	52
0·05	♂	304	3/5	11·1	10·6	15 (11–24)	40 (24–55)
0·05	♀	345	1/5	15·2	14·5	22	22 (9–29)

motor control was regained. In the wild, however, a rat with full paralysis of its hind limbs would probably perish or be easily killed when found above ground.

Onset of poisoning symptoms

The time between the start of feeding and the onset of warning symptoms that stop feeding are summarized in Table 2. In direct observations of lesser bandicoots at nightly feedings we noted the minimum time for duration of feeding to be 52 min and the maximum to be 230 min. The maximum time might be even longer since our observations were discontinued at 250 min. One female fed for 48 out of the first 60 min after food cups were placed. During that period she consumed 13·7 g of pyriminyl bait. Calculating from her body weight, she had consumed 184 mg/kg of the active ingredient (about 27 LD 50 doses) yet she was still feeding on the same bait up to 200 min from the start of feeding. From these observations we conclude that the period between the start of feeding and the termination due to onset of warning symptoms varies from about 60 to greater than 240 min. This

Table 4. *Chi square analysis of mortality in lesser bandicoot rats (data from Table 3)*

Source of variation	χ^2	D.F.	*P*
Concentration	12·60	4	0·01–0·02
Sex	0·159	1	NS
Sex × concentration	4·04	4	NS
Total	16·80	9	0·05–0·06

Table 5. *Results of giving lesser bandicoot rats a choice for 2 days between plain banana and boiled rice bait and bait containing pyriminyl*

Conc. of pyriminyl (%)	No. of rats ♂	No. of rats ♀	Mean body wt (g)	Mortality (%)	Mean bait consumed* (g) Poison	Mean bait consumed* (g) Plain	Mean and range of doses (mg/kg) of active ingredient taken
0·5	2	8	331	100	8·0	6·7	102·7 (6·6–273·0)
0·25	–	9	287	100	12·1	12·8	66·3 (36·0–241·1)
0·25	6	2	266	100	6·2	10·5	41·2 (8·2–109·9)

* Not corrected for evaporative water loss.

means that lesser bandicoot rats feeding on 0·5 % pyriminyl baits should have ample time to consume multiple LD 50 doses.

Free-choice feeding tests

We used titrated quantities of pyriminyl in laboratory meal to establish the optimum concentration under laboratory conditions. Pyriminyl was offered in baits in concentrations ranging from 0·05 % to 1 % by weight.

The results are summarized in Table 3. At 1 % a complete kill was obtained in two different trials. At 0·5 %, again 100 % mortality resulted in several trials and animals consumed between 2 and 15 LD 50 doses. At 0·25 % and lower concentrations we failed to kill more than 90 % of the animals on test. Pyriminyl appears to be somewhat unpalatable to lesser bandicoot rats at concentrations of 0·5 % and above but not of such a degree as to prevent consumption of lethal amounts.

A chi square analysis of the mortality data is given in Table 4. Mortality was not significantly influenced by sex but was by concentration of rodenticide. Combining results for both sexes, the mortalities obtained were 100 %, 100 %, 88·2 %, 90 % and 40 % at concentrations of 1 %, 0·5 %, 0·25 % 0·1 % and 0·05 % respectively. From these results we concluded that the optimum concentration of pyriminyl in bait materials was around 0·5 %.

Additional bait trials were carried out using a potential field bait, a mixture of boiled rice and ripe bananas. The results (Table 5) indicated that a 0·25 % concentration of pyriminyl could be expected to give as good a kill as would 0·5 % when using a moist bait. Similarly, Cowan, Srihari & Sridhara (1977) obtained

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100 % mortality using pyriminyl at 0·25 % in a rice flour and groundnut oil bait against *B. bengalensis* in India.

Pyriminyl offers an alternative to zinc phosphide as an acute rodenticide against *B. bengalensis*. Before general use in the field, however, pilot field trials using both the 0·25 % and 0·5 % concentrations will be necessary to determine the optimum concentration required in field baits. Information is also needed on the toxicity of pyriminyl to other pest rodent species that occur along with *B. bengalensis*.

Pyriminyl is known to be toxic to humans and to certain domestic animals, especially cats. The evidence presented of the antidotal properties of nicotinamide (Whitmoyer Laboratories, 1975*a*, *b*) offer promise that pyriminyl may be a relatively safe material to use. Despite the potential of an antidote, the material should always be regarded as a hazardous compound and every effort made when using it to avoid situations where contamination of food-stuffs or accidental poisoning of humans and domestic animals could occur.

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