LABORATORY AND FIELD EVALUATION OF DIFETHIALONE, A NEW ANTICOAGULANT RODENTICIDE

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ABSTRACT: The efficacy of the newly developed anticoagulant rodenticide, difethialone was evaluated against various rodent species in laboratory and fields. Difethialone at 0.025% concentration in the form of loose bait (broken wheat and rice + veg. oil + garlic powder) gave absolute mortality in *Rattus rattus*, *Mus musculus*, *Funambulus pennanti*, and *Meriones hurrianae* during 'no-choice' tests in one day feeding. No significant difference was noted in poison bait intake and mean days to death between two and three days poison feeding. Mean days to death were ranged between 2.9 to 5.70 in all the species tested. In fields, baiting with the same loose bait (0.025%) was performed in the live burrows and 88.89% control recorded on the 4th day after treatment. In another trial, three large hay stacks situated near wheat, barley and mustard crops were also treated with difethialone (0.025%) and 90.47, 94.44 and 80.00% kills were recorded respectively. The results of the present investigation prove high potency and acceptability of difethialone against both domestic as well as field rodents.

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INTRODUCTION

Among mammals, order Rodentia is the largest group inhabiting the desert biome of India (Prakash et al. 1967). The world annual crop losses due to rodents was estimated to be approximately 30,000 million U.S. Dollar (Dubock 1982). In the view of increasing rodent problem many different methods have been devised to control rodent pests. However, in more recent times, chemical control using rodenticide has become the most widely used option. A range of products with considerable variation in their potency against the pest species have been evaluated. Zinc phosphide, an acute rodenticide, has been used predominantly over several years followed by chronic first generation anticoagulant rodenticide. Non-palatability and bait shyness towards acute rodenticide (Sood et al. 1978, Ojha 1978, and Rana 1980) and resistance to chronic first generation rodenticides (Boyle 1960, Deoras 1966, Mukthabai et al. 1981, and Taylor 1972) has limited their usefulness. Isolated cases of resistance to difenacoum and bromadiolone, the second generation chronic rodenticides have recently been reported. (Lund 1984). Search for improvised alternative rodenticide, vis-a-vis periodic screening of their bio-efficacy is considered to be of paramount importance. This has become more pertinent in the light of development of resistant population. Difethialone is a new anticoagulant rodenticide developed by Lipha of France which may fill up the lacunae between fast acting acute and slow acting chronic rodenticide.

An attempt has been made to evaluate the efficacy of this novel anticoagulant rodenticide against various rodent species in captivity and open fields.

MATERIAL AND METHOD

No-choice feeding test was conducted in laboratory on acclimatized, healthy adults which were fed on rat feed (Hindustan Lever Ltd., Bombay) and water *ad libitum*. Twelve individually caged animals were weighed, sexed and then exposed to difethialone (0.025% concentration) loose bait after 24 hr of starvation under 3 different experimental protocol.

1) 0.025% difethialone loose bait, 1 day feeding.
2) 0.025% difethialone loose bait, 2 day continuous feeding.
3) 0.025% difethialone loose bait, 3 day continuous feeding.

Poison bait consumption was measured, after which bait was replaced by normal laboratory diets. Deaths were scored for all the twelve days. Efficacy of difethialone loose bait was evaluated against *Rattus rattus*, *Mus musculus*, *Funambulus pennanti* and *Meriones hurrianae*.

The investigations to test the efficacy in field was carried out in a village Muhana, 22 km from Jaipur city.

The trials were conducted in a selected field of barley crop heavily infested with rodents. The study area measured 1500 sq. yards approximately. Pretreatment level of rodents infestation was estimated by adopting the live burrow count method (Barnett and Prakash 1975, Rao 1977). However, in order to ascertain the number of incumbents per burrow, twelve burrows were excavated and it was noted that only one individual inhabited each burrow. Therefore the number of live burrows were taken as the number of rats infesting the study area. The live burrows were plugged with wet soil + lime and marked with lime powder. The next day open burrows were taken as live burrows and treated with 20 gms of difethialone loose bait (0.025% conc.) contained in polythene bags, pushed inside each burrow. After treatment the burrows were again plugged and marked. The same procedure was repeated for all the 25 days of observation.

In the 2nd field trial, three large hay stacks situated near wheat, barley and mustard crop were selected. Population count was carried out by capture-recapture technique. Fifty packets each containing about 20 gms of difethialone loose bait (0.025% conc.) were placed at different points in the clay pots under the hay stack near wheat and barley crops respectively. While due to less population count in the hay stack near mustard crop, only 20 packets were placed under the hay stack. On the 7th day after treatment, carcasses and rats in moribund condition were collected.

RESULT AND DISCUSSION

Based on combined sex mortality data the results of no-choice feeding test are summarized in Table 1. The high efficacy of difethialone is confirmed by 100% mortality observed in all the three sets of experiments against *Rattus rattus*, *Mus musculus*, *Funambulus pennanti* and *Meriones hurrianae*.

Table 1. Efficacy of difethialone (0.025%) against different rodent species (no-choice feeding test). One hundred percent mortality rate was achieved in all tests (12 animals per group).

Rodent species	Bait exposure (days)	Mean poison bait intake (gm/kg b.wt.±SE)	Mean active ingradient intake (mg/kg b.wt.±SE)	Days to death	
				Mean	Range
Rattus rattus	1	148.00±1.41	37.00±0.31	4.50±0.36	3-7
	2	196.64±1.68*	49.16±0.42	3.00±0.52*	2-7
	3	202.25±4.20**	50.56±1.05	2.90±0.34***	2-5
Mus musculus	1	117.67±3.98	29.42±1.99	4.00±0.25	3-5
	2	145.71±6.24*	36.42±1.56	3.50±0.15**	3-4
	3	151.25±6.76**	37.81±1.69	3.00±0.25**	2-4
Funambulus pennanti	1	117.76±3.176	29.43±0.79	5.50±0.85	3-12
	2	161.04±1.00*	40.25±2.62	5.00±0.61*	2-9
	3	161.75±3.70**	40.43±0.92	5.20±0.35*	3-8
Meriones hurrianae	1	102.40±5.83	25.60±1.46	5.70±0.524	4-9
	2	160.50±2.75*	40.12±0.67	4.00±0.44**	2-7
	3	195.75±33.00**	48.93±8.25	4.10±0.24*	3-5

Student's 't' test, level of significance between:
(A) Mean poison bait intake on 1 and 2 days and 2 and 3 days.
(B) Mean days to death on 1 and 2 days and 2 and 3 days of bait exposure.
A. i) Significant difference between 1 day and 2 day mean poison bait intake values (*p 0.001).
ii) Non-significant difference between 2 day and 3 day mean poison bait intake values (**p 0.05).
B. i) Non-significantly different values (*p 0.05)
ii) Almost significantly different values (**p 0.05)
iii) Significantly different values (***p 0.01).

Table 2. Treatment of difethialone (0.025% conc.) loose bait under haystacks.

Location	Population count in hay stacks	Packets[a] placed (left)	Carcasses recovered (on 7th day)	Rats found in moribund condition	Percent kill
Near wheat crop	42	50(1)	29	9	90.47
Near barley crop	36	50(3)	32	2	94.44
Near mustard crop	15	25(5)	12	nil	80.00

[a]One packet contains 20 gm difethialone loose bait (0.025%).

The following Laboratory no-choice feeding study typifies difethialone's good acceptability. Statistically nonsignificant difference was observed in the mean poison bait intake values ($p > 0.05$) between two and three days of continuous feeding. However, significant difference in the values ($p < 0.001$) between one day and two days exposure period was observed. Mean days to death ranged from 2.9 to 5.7 in all the rodent species tested. No significant difference in the values for mean days to death was observed for all the three sets of experiment as the students 't' test level of significance between mean days to death on 1 day, 2 day and 3 days of bait exposure was incoherent (Table 1). Clinical signs consistent with anticoagulant toxicity (partial paralysis, perinasal and periorbital dark staining, lassitude) were seen in some animals after fourth day of exposure to the toxicant. The laboratory data's therefore suggests that difethialone (0.025% conc.) in single dose exposure will economise on the amount of rodenticide used.

The following two field trials typify high level of control achieved by difethialone (0.025% conc.) in different operational uses. In field studies, baiting with the same loose bait revealed 88.89% control success fourth day after treatment. Observations of 10th day after treatment indicated the same control success (88.89%). The results suggest that during the field trials maximum kill occurs between 4th to 10th day of exposure to the poison bait. Similar studies were carried out to test the efficacy of difethialone against two native field rodent species of Europe (*Arvicola terrestris* and *Pirymys*

duodecimostatus) with bait containing as low as 0.0025% of active ingredient (Lechevin 1987). The findings are further supported by Marsh (1988). He reported that difethialone has been found effective against warfarin resistant strains also.

Another field trial carried out under the hay stacks near wheat, barley and mustard crop, treated with difethialone (0.025% conc.) resulted in 90.47, 94.44 and 80.00% control success respectively (Table 2). Furthermore, no damage was noticed from these crop fields indicating that the control operation was successful. Difethialone shows some intrinsic beneficial selectivity towards non-target species. Even though a number of stray dog pets and other animals were around the treated area, there was not a single incidence of primary or secondary non-target toxicity. Major rodent species identified in field were *Meriones hurrianae*, *Tatera indica*, and *Rattus meltada*.

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