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Original Research Article

Laboratory Evaluation of Difethialone for Bait Shyness in Indian Desert Gerbil, *Meriones hurrianae* Jerdon

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A B S T R A C T

Due to the development of problem of bait shyness with acute rodenticides and resistance against first generation anticoagulants, it has become essential to develop rodenticides to overcome such shortcomings. A second generation anticoagulant rodenticide, Difethialone was evaluated in laboratory conditions for bait shyness in Indian Desert Gerbil, *Meriones hurrianae* Jerdon. Most palatable concentration of 0.00093% was used for preparing poison baits. Results of this laboratory evaluation showed a highly significant difference ($P<0.001$) in the total daily consumption of most preferred poisoned bait and less preferred plain bait. This is indicative of the absence of bait shyness and poison aversion in gerbils towards Difethialone.

Introduction

Rat control is of prime importance in pursuit of minimizing the losses caused by them. Chemical control has been found to be most effective against rodent depredations as it ensures greater control in least possible time. The chemicals used in rodenticide formulations are of varied nature and origin having different modes of action. Chronic rodenticides are the multiple dose poisons and include the anticoagulant compounds, which are slow acting and have cumulative effects. They are much selective in their activity and are safer to the non target species and the environmental hazard is also minimal (Sharma, 1989). Development of bait shyness and resistance to the first generation anticoagulant rodenticides led to the development of a new series of hydroxycoumarin derivatives. These are the single shot second generation anticoagulants effective against resistant rats and mice and can be fatal after a single feeding. The active ingredient of the second generation anticoagulants is used at very low levels and the initiation of symptoms is delayed so the rodent is unable to associate the onset of illness due to poisoning with particular bait, and thus it prevents bait shyness. Difethialone is considered as a second generation anticoagulant rodenticide (Saravanan *et*

al., 2003). Its anticoagulant activities on several rodent species in field and laboratory conditions have been described by Lechevin, (1987), Lechevin and Poche, (1988). Difethialone is thus tested in the laboratory in the present study for bait shyness in the field rodents, *Meriones hurrianae*.

Materials and Methods

Test Animal

The Indian desert gerbil, *Meriones hurrianae* was used as the test species in the study. It is the major pest in the arid and semi-arid regions of Northern India and poses a threat to Kharif crop fields and grasslands. Gerbils captured from the local surroundings were acclimatized to the laboratory conditions, maintained on a palatable diet (Rat feed: Hindustan Lever Ltd., New Delhi, India) for 7 days and fresh water was made available ad libitum.
Test Rodenticide - Difethialone, a second generation anticoagulant rodenticide is a product patented by LIPHA OF France and is marketed under the trade name LM - 2219. Its empirical formula is - $C_{31}H_{23}BrO_2S$ and the chemical formula is 3-[3-(4'- bromo [1, 1'-biphenyl]-4yl)-1, 2, 3, 4-tetrahydro-1-1naphthlenyl]-4-hydroxy. The technical compound is a liquid concentrate containing LM-2219 (1.25 gm/litre).

Bait Formulation

Calculated quantity of the technical concentrate containing 1.25 gm/litre Difethialone was mixed with bajra flour + 2% sugar + 2% refined groundnut oil to obtain the desired concentration of the poison bait. Plain bait was prepared without the rodenticide and contained all the ingredients.

Mode of administration of rodenticide - Cage baiting was done for the conduction of bait shyness tests.

Experimental Design

The experiment was conducted to evaluate bait shyness in adult gerbils of both the sexes. The test was completed in two phases.

Phase A

Healthy adult gerbils were given two types of flour baits, the most preferred and the second preferred flour bait for three days. Water was made available ad libitum.

Phase B

The most preferred flour was mixed with 2% groundnut oil and 2% sugar in calculated amount of Difethialone concentrate (1.25 gm/litre) to prepare poison bait of 0.00093% concentration (most palatable concentration). This poison bait was given to the gerbils of Phase B for four days, starting from the fourth day and given simultaneously with plain bait (second preferred). Water was given ad libitum. Difference in consumption of the two baits was analysed statistically.

Results and Discussion

The experiment was carried out in two phases as shown in the table.1.
In the Phase A of the experiment, the consumption of bajra, i.e. the most preferred food was more than the less preferred food i.e. wheat. A highly significant ($P< 0.001$) difference was noticed in the mean daily consumption of bajra and wheat.

Table.1 Bait Shyness with Difethialone (0.00093% concentration) in *Meriones hurrianae* Jerdon

Phase A

Carrier food	Mean daily intake of food (gm/kg. body wt.)			Mean total daily intake (gm/kg. body wt.)	Rank of preference
	1st day	2nd day	3rd day		
Bajra	136.20 ± 0.88	143.61 ± 0.85	138.72 ± 1.10	139.51 ± 1.88	1
Wheat	93.20 ± 0.52	109.15 ± 1.10	101.30 ± 1.00	101.21 ± 3.97[c]	2

Phase B

Carrier food	Mean daily intake of food (gm/kg. body wt.)				Mean total daily intake (gm/kg. body wt.)	Rank of preference
	4th day	5th day	6th day	7th day		
Bajra 0.00093 %Difet-hialone	138.94 ± 0.79	142.36 ± 0.80	139.91± 0.71	127.84 ± 1.2	136.51 ± 3.13	1
Wheat	112.30 ± 0.60[c]	113.64 ± 0.39[c]	117.65 ± 0.60[c]	116.19 ± 0.66[c]	114.94 ± 1.25[c]	2

Values are expressed as mean ± S.E. Data significantly different from Bajra is indicated as c: P<0.001

In Phase B of the experiment, the test rodenticide, Difethialone was added to the most preferred food i.e. bajra flour. On the 4th day, the consumption of bajra (along with the toxicant) decreased non significantly (P< 0.05). The intake of poisoned bait bajra was still higher than that of plain bait, wheat. On the 5th day, (2nd day of poison baiting), the consumption of bajra increased again as compared to the plain bait and the difference in the intake of the two was highly significant (P< 0.001). The consumption of poisoned bait bajra decreased on the 6th day (3rd day of poison baiting) but it was significantly high (P<0.001) than the intake of less preferred plain bait. With the onset of sickness due to poisoning, the total intake of poison bait reduced. The mean daily intake of poisoned bait bajra decreased on the 7th day also. The difference between the intake of bajra (poisoned bait) and wheat (poison free bait) was noted to be highly significant (P<0.001). Highly significant difference in the total daily intake of most preferred but poisoned food and less preferred but plain food indicates the absence of bait shyness and poison aversion for Difethialone. Also, the poison bait shows good palatability. It has been found palatable and efficacious in both laboratory (Nahas *et al*., 1989) and field tests (Marshall, 1992, Saxena *et al*., 1992) against Norway rats and mice.

The phenomenon of bait shyness is of much practical importance from the point of view of optimizing the control success in several rodent species. First generation

anticoagulant rodenticides generally have shorter elimination half-lives (Vandenbroucke, 2008) and require higher concentrations to accumulate the lethal dose, and are less toxic than second generation agents. Cholecalciferol products for rodent control require relatively high concentrations of active ingredient, which may lead to bait aversion (Arora *et al*., 1992).The usually slow onset of toxic effects of second generation anticoagulants allows the baits to be prepared with very low concentrations of active ingredient. It prevents the rodents from associating illness with the particular rodenticide. The rodents feed repeatedly on such rodenticide baits without showing any bait shyness symptoms. Single day exposure of the rodenticide resulted in effective kill with three different concentrations viz. 0.0025%, 0.00125% and 0.00375% in no-choice and choice feeding tests in house rat, *Rattus rattus* (Kaukeinen, 2000). The second generation anticoagulants are also effective against rodent strains that are resistant to first generation anticoagulants and are sometimes referred to as "superwarfarins" (Kotsaftis, 2007).

Eradication of rodents surviving acute and first generation anticoagulant bait poisoning need to be prebaited for at least a week to overcome bait shyness. The poisoned bait is applied subsequently. During this period, the gerbils may cause immense damage. Bait shyness studies have not been reported with Difethialone in any of the earlier works. In the present study, Difethialone is found to have an edge over the acute and the first generation anticoagulant rodenticides due to the absence of bait shyness and good palatability in gerbils as studied under laboratory conditions and can prove to be very effective in controlling gerbils in the field conditions too.

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