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Biological efficacy of amitraz against the three host tick *Rhipicephalus sanguineus*

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Resistance of ticks to acaricides has spread dramatically in recent years, so changing of acaricides periodically is needed to avoid resistant strains (Khalaf-Allah 1996). Amitraz has been proved to be effective against *Amblyomma variegatum* and *Boophilus decoloratus* ticks of cattle (Karaguki 1996, George *et al.* 1998) and *Hyalomma dromedarii* in the camel (Jacquiet *et al.* 1994). The present study was undertaken to assess its efficacy as an acaricide against the 3-host tick *Rhipicephalus sanguineus*.

Amitraz was obtained as an emulsifiable concentrate containing 12.5% w/v active ingredient. The concentrations used in the studies were made using triple distilled water as to give 150 ppm, 200 ppm, 250 ppm, 300 ppm and 350 ppm.

Engorged adult *R. sanguineus* ticks were carefully detached from tick infested dogs attending the outpatient departments of the Veterinary College, UAS, Bangalore, and from the SPCA shelter, Bangalore; after confirming that the dogs ware not sprayed with any insecticide during the preceding 6 months. Male and female ticks were collected in separate specimen tubes and immediately transferred to the laboratory for the *in-vitro* studies.

Acaricidal efficacy against adult male ticks

The filter paper method described by Sabnis *et al.* (1984) was followed with some modifications. Five Whatman No.1 filter papers (11.0 cm) were impregnated with 1 ml of various concentrations of the insecticide solution. The filter papers were dried under the ceiling fan and kept in petri-dishes. Ten male ticks were released on the filter paper and covered with petri-covers. Observations on tick mortality were recorded after 24 hr, after exposure to insecticide and percentage mortality was calculated for each group.

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Inhibitory effect on oviposition in engorged female ticks

The tea-bag method of Gladney *et al.* (1972) was followed for all concentration's of the insecticide and the sixth group served as control. The ticks were dipped in the insecticide solution for exactly 40 sec and then solution was drained through filter paper and thereafter ticks were dried by placing them in-between layers of filter paper. The same procedure was followed for controls using distilled water for dipping. The ticks were then placed individually in labeled glass vials. The mouth of vials was closed with a piece of muslin cloth and fastened with rubber band. The vials were placed in a desiccator containing saturated solution of calcium chloride so as to maintain a relative humidity of 80-85 %. The oviposition, number of eggs laid and hatching period of the eggs were recorded/day.

Larvicidal efficacy

The larvicidal efficacy of amitraz was evaluated by a modification of the method described by Stendel (1969). Six strips of Whatman No.1 filter paper measuring about 15cm long and 2cm wide were taken, of which 5 were impregnated with various concentrations of the insecticide in the same manner as for the adult male ticks and the sixth was dipped in distilled water. They were dried under a fan and were used to line the inner surface of the specimen tube of the size 7cm long and 2cm wide. One-week-old larvae, which hatched from the eggs that were laid by the control ticks in the previous experiments, were used for the study. A batch of larvae was released (25) into each of the specimen tubes. The mouth of tube was covered with a double layer of muslin cloth and fastened with rubber band. The mortality was recorded at 3, 6, 9, and 12 hr and percentage mortality for each group was calculated.

Ovicidal efficacy

The efficacy of the acaricide on eggs was determined by the tea-bag method (Gladney *et al*, 1972). The tea bags were prepared from heat sealable rice paper. The bags were prepared by cutting into (10cm × 4cm) strips, folding length-wise and sealing them on both sides with a soldering iron to

make a flat envelope (4cm × 5cm).

Dilutions of the different concentration of the insecticides were taken in a small beaker and a control group was dipped in distilled water. A known number of eggs were taken in each of 4 bags before test and sealed and dipped in known concentration of acaricide for 10 sec and 50 sec. The eggs from each tea bag were transferred to separate test-tubes after 24 hr. They were kept under observation for hatching and the percentage hatchability was calculated.

Acaricidal efficacy against adult male ticks

After 24 hr of exposure to the insecticide, there was 100% mortality in 250 ppm, 300 ppm and 350 ppm concentration group and there was no mortality in the control group.

The therapeutic efficacy of amitraz on *Boophilus microplus* (Canestrini) on cattle was studied. The efficacy in each of the 4 observed concentrations (0.0088, 0.0131, 0.0174, and 0.0231% AI) of amitraz wa excellent (>99% control) (George *et al.* 1998).

Inhibitory effect on engorged female ticks

The groups of ticks dipped in various concentrations viz. 250 ppm, 300 ppm and 350 ppm did not lay eggs. At 200 ppm concentration, eggs laid did not hatch and eggs laid by ticks dipped in 150 ppm hatched after 15 days; whereas the controls laid eggs hatched in 5-8 days. Thus the acaricide inhibited oviposition in female ticks even at lowest concentration of 200ppm.

The efficacy of amitraz on cattle ticks was assessed by susceptibility tests, spraying and dipping trials (Karaguki 1996). Tests on the susceptibility of 3 tick species, *Amblyomma variegatum* and *Boophilus decoloratus* to amitraz (technical grade-purity 98.6% w/w). More than 50% of the detached engorged females failed to lay eggs. The remainder laid few eggs, and these had a low hatching rate of 0-2%, compared with 90-98% in the controls.

Larvicidal efficacy

At 12 hr after exposure to the insecticide there was 100% mortality in all the groups exposed to various dilutions of insecticide and no mortality in the control group. Therefore, amitraz is a potent larvicidal agent.Sabnis *et al.* (1984) using the filter paper impregnation method studied the efficacy of propetamphos against the larvae of *Rhipicephalus sanguineus* and found 100% mortality in all the groups exposed to the various dilutions of the insecticide and 10% mortality in the control group. The efficacy of propetamphos against the larvae of *Boophilus* spp and *Rhipicephalus* spp was studied by using the dipping technique (Rajeshwari 1984) and found 100% mortality in all the groups exposed to the insecticide.

Ovicidal efficacy

The eggs dipped at the concentration of 150 ppm had 80% and 90% hatchability at dipping of 50 sec and 10 sec respectively; the eggs hatched after 22-24 days. At 200 ppm concentration the eggs hatched in those that were dipped for 10 sec with 80% hatchability and the eggs hatched after 25 days, whereas the eggs dipped at concentrations of 250 ppm, 300 ppm and 350 ppm did not hatch at dipping for 10 and 50 sec.

SUMMARY

The acaricidal efficacy of amitraz was studied against the three-host tick *Rhipicephalus sanguineus* at the concentrations of 150, 200, 250, 300 and 350 ppm. It was found to be an effective insecticide against adult male ticks, engorged female ticks, ova and larvae at the concentrations used.

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