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Short Communication

ACARICIDE RESISTANCE STATUS OF *HYALOMMA ANATOLICUM* AND *RHIPICEPHALUS* (*BOOPHILUS*) *MICROPLUS* TICKS COLLECTED FROM VARIOUS LOCATIONS OF HARYANA

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SUMMARY

Indiscriminate use of acaricides such as synthetic pyrethroids and organophosphates for control of ticks has led to the emergence of resistance against these acaricides. The present study aimed to determine the status of acaricide resistance in *Hyalomma anatolicum* and *Rhipicephalus* (*Boophilus*) *microplus* ticks. Fully engorged female ticks were collected from various Gaushalas of Hisar, Bhiwani and Charkhi Dadri districts and subjected to adult immersion test with discriminating dose (AIT-DD) against technical grade deltamethrin and coumaphos. Prevalence of resistance in *H. anatolicum* and *R.* (*B.*) *microplus* against deltamethrin ranged from 10%-60%, and 6.67%-76.67%, respectively, whereas, against coumaphos, 46.67%-73.33% and 13.33%-63.33%, respectively, showing high resistance against both deltamethrin and coumaphos in field ticks. Data generated indicate an immediate need of switching to newer class or advanced generation of acaricides.

Keywords: Acaricide resistance, *Hyalomma anatolicum*, *Rhipicephalus* (*Boophilus*) *microplus*

Ticks and tick borne diseases are widely prevalent throughout the world. Synthetic pyrethroids (SP) and organophosphates (OP) are being most commonly used for tick control in India. The greatest hindrance in control of ticks is emergence of resistance against acaricides (Rosario-Cruz *et al.*, 2009). Amongst various bioassays proposed for determination of acaricide resistance in ticks, adult immersion test with a discriminating dose (AIT-DD) is most rapid and simpler method for large scale screening of field ticks (FAO, 2004). The estimated cost for controlling tick and tick borne disease has been estimated more than 2000 crores annually for India (Minjauw and McLeod, 2003). Several workers have also reported development of resistance against OP and SP acaricides (Gaur *et al.*, 2016; Kumari *et al.*, 2018). Thus, the present study aimed to assess the acaricide resistance status against OP (coumaphos) and SP (deltamethrin) in *Hyalomma anatolicum* and *Rhipicephalus* (*Boophilus*) *microplus* ticks.

To check the acaricide resistance status, several fully engorged freshly dropped adult female *H. anatolicum* and *R.* (B.) *microplus* ticks were collected from various Gaushalas of Hisar (Shahpur, Dhobi, Mangali, Kaimri, Mignikhera), Bhiwani (Tosham) and Charkhi Dadri districts. Ticks were identified using available standard literature (Walker *et al.*, 2014). Adult engorged female ticks collected from different sites were kept for adult immersion test with AIT-DD in desiccators containing 10% KOH in BOD incubator maintained at 25 to 30 °C till start of experiment.

Stock solutions of technical grade deltamethrin (5,000ppm) and coumaphos (50,000ppm) (Sigma-aldrich) were prepared in acetone and methanol, respectively. For the experimental bioassay, AIT-DD, different concentrations of the acaricides were prepared in distilled water from the stock solutions and tested against adult engorged female *H. anatolicum* and *R.* (B.) *microplus*.

The bioassay was conducted according to the methods provided by FAO (2004) with slight modifications. The discriminating dose taken for coumaphos was 262 ppm (Kumar *et al.*, 2015) and for deltamethrin was 59.2 ppm (Shyma *et al.*, 2013). Ten healthy, engorged female ticks were immersed in 20 ml of the freshly prepared acaricide dilution for 30 minutes at room temperature with gentle and intermittent shaking. After 30 minutes, the acaricide solution was poured off and the ticks were dried gently on filter paper. The test was performed in triplicate. After that the ticks were sticked with ventral side up, onto double-sided sticky tape in a Petri dish, placed in desiccators (85 to 95% RH, 28±2 °C) for 7 days. After 7 days the number of ticks that have laid eggs were counted. The percentage resistance was calculated as per the formula, R (%) = (Nt/Nw) × 100, where R (%) refers to percent resistance, Nt refers to number of treated ticks laying eggs and Nw refers to number of untreated ticks laying eggs.

Acaricide resistance determined using adult immersion test with discriminating dose (AIT-DD) for deltamethrin (59.2ppm) and coumaphos (262 ppm) against *H. anatolicum* and *R.* (B.) *microplus* ticks. The percent resistance against deltamethrin was highest i.e. 60% and 76.6% in *H. anatolicum* and *R.* (*B.*) *microplus* ticks collected from Kaimri whereas lowest in the isolates collected from Charkhi Dadri. On the other hand, percent resistance against coumaphos was highest (73.33% and 63.33%) in *H. anatolicum* and *R.* (*B.*) *microplus* ticks

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collected from Mingnikhera and Dobhi, respectively. *H. anatolicum* and *R.* (*B.*) *microplus* ticks collected from Charkhi Dadri were found to be susceptible to deltamethrin as well as to coumaphos. The data generated could be attributed to the excessive use of synthetic pyrethroids (SP) and organophosphates (OP) under field condition. Similarly, deltamethrin resistance was also reported in *R.* (*B.*) *microplus* collected from Hisar using AIT-DD (Kumari *et al.*, 2018). Synthetic pyrethroid resistance accessed using AIT-DD bioassay has also been reported in sheep ticks collected from different regions of Karnataka (Sudharani *et al.*, 2018). Acaricide resistance in *H. anatolicum* and *R.* (*B.*) *microplus* ticks collected from Hisar (Haryana) and Churu (Rajasthan) against deltamethrin and diazinon has also been reported using adult immersion test and larval packet test (Gaur *et al.*, 2016). Kumar *et al.* (2015) evaluated coumaphos against three field isolates and found all of them susceptible using AIT and one of them showed level I resistance when larval packet test was conducted. Four isolates collected and tested for development of resistance using AIT from Uttar Pradesh were found to be resistant to deltamethrin and susceptible to coumaphos (Chigure *et al.*, 2018). Similarly *R.* (*B.*) *microplus* ticks collected from 6 districts in Uttar Pradesh were found to be susceptible to coumaphos and resistant to deltamethrin using classical AIT (Nagar *et al.*, 2018). Resistance against deltamethrin in *R. microplus* have also been reported by Fular *et al.* (2018) from 12 states of India. However in the present study, *H. anatolicum* and *R.* (B.) *microplus* ticks showed resistance to deltamethrin and coumaphos using AIT-DD. This is the first report of resistance development in ticks against coumaphos in India.

This emergence of resistance against deltamethrin and coumaphos in *H. anatolicum* and *R.* (*B.*) *microplus* ticks can be either due to increased expression of the detoxifying enzymes such as esterases, monoxygenase and glutathione-S-transferase or due to target site mutation like acetylcholinesterase and sodium channel gene. So there is a need to study the exact mechanism for development of resistance in these ticks against deltamethrin and coumaphos which may help in devising other alternate strategies to control these commonly available ticks in cattle and buffaloes in Haryana. Further, it is essential that farmers and extension workers should be skilled in acaricide rotation for control of acaricide resistance in ticks under field conditions.

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