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Short Communication

Malathion resistance in red flour beetle (*Tribolium castaneum*) (Herbst) (Coleoptera: Tenebrionidae) from FCI godowns of Kerala, India

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Abstract

Laboratory studies were conducted during 2018-2019 in the Department of Agricultural Entomology, College of Horticulture, Vellanikkara, Kerala Agricultural University, Thrissur, to evaluate the susceptibility of different populations of *Tribolium castaneum* Malathion (Herbst) (Coleoptera: Tenebrionidae) collected from five FCI godowns of Kerala *viz*., Thikkodi, Olavakkode, Mulangunnathukavu, Angamaly and Valiyathura to malathion with respect to the laboratory maintained susceptible strain from IARI, New Delhi. Residual film bioassay was carried out using technical grade malathion. Angamaly population of *T. castaneum* showed 13.34 fold resistance with the highest LC_{50} value of 6949.80 ppm, while the IARI strain had the lowest LC_{50} of 520.76 ppm. Resistance ratios for Mulangunnathukavu, Valiyathura, Olavakkode and Thikkodi population were 11.82, 11.27, 10.99 and 10.95, respectively. Resistance to malathion in all field collected populations were uniform and homogenous.

Key words: Insecticide resistance, Malathion, *Tribolium castaneum*.

The world population is growing by approximately 83 million people every year. An increasing demand on the production of cereals and other food grains occurs due to this inevitable population growth. Since arable lands are diminishing, reducing yield loss is a way to increase the availability of food grains to the burgeoning humanity. Post harvest losses constitute a form of yield loss and accounts for one-third of the food produced every year (Gustavsson et al., 2011). Cereal crops suffer about 19 per cent loss by weight and 53 per cent loss on the basis of calorific content (Lipinski et al., 2013). Insect pests cause 30 – 40 per cent loss to the stored grains (Abass et al., 2014). The red flour beetle, *Tribolium castaneum* (Coleoptera: Tenebrionidae) is an important worldwide stored grain pest, causing postharvest losses of > 20 per cent in developing countries and up to 9 per cent in developed countries (Phillips and Throne, 2010). Larvae and adults of red flour beetle are external feeders which attack the damaged grains or farinaceous materials. They readily adapt to stored grain environment due to high fecundity and relative longevity (Boyer et al., 2012).

Even though several practices are available for stored grain pest management, use of insecticides is the quickest, effective and economic method to reduce the pest population to acceptable levels (Perez-Mendoza, 1999). However, selection pressure by insecticides had led to development of resistance to insecticides (Boyer et al*.,* 2012).

Tribolium castaneum ranks 19th among the 20 most insecticide resistant arthropods in the world (Whalon et al., 2008) and it has already been

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reported to have developed resistance against phosphine, methyl bromide, organophosphates, pyrethroids and insect growth regulators, which are commonly used for its management (Werner, 1997; Zettler and Arthur, 1997; Pimental et al., 2007).

Malathion, deltamethrin and dichlorovos are the insecticides recommended for the management of stored grain pests in FCI godowns. However, development of insecticide resistance has made the management of these pests a difficult task. In 1971 itself 37.76-fold resistance to malathion was reported in *T. castaneum* in India by Bhatia et al. (1971). Subsequently, resistance to malathion was reported from different parts of the country (Dhaliwal and Chawla, 1995; Srivastava et al., 2001). This study was carried out to determine the susceptibility of *T. castaneum* population collected from different FCI godowns of Kerala to malathion.

Different population of red flour beetle, *T. castaneum* were collected from five different godowns of Food Corporation of India (FCI) situated at different geographic locations of Kerala such as Thikkodi (Kozhikkode district; 11.494295, 75.625879), Olavakkode (Palakkad district; 10.807528, 76.628972), Mulangunnathukavu (Thrissur district; 10.596744, 76.206517), Angamaly (Ernakulam district; 10.191658, 76.374888) and Valiyathura (Thiruvananthapuram district; 8.470714, 76.924354). Susceptible strain of *T. castaneum* was procured from Division of Entomology, Indian Agricultural Research Institute (IARI), New Delhi, which was maintained without exposure to any insecticide for more than 35 years. The study was carried out at Pesticide Residue Laboratory, Department of Agricultural Entomology, College of Horticulture, Vellanikkara during 2018-19.

Populations of *T. castaneum* were reared in the laboratory at 30 ± 2°C and 80 ± 5 per cent relative humidity as per the method described by Bhatia and Pradhan (1968). About 15 – 20 adults obtained from different FCI godowns and IARI were introduced into separate plastic containers (17 cm x 11 cm) containing 250 g of sterilized wheat flour fortified with 5 per cent (wt./wt.) brewer's yeast. Ten such containers were initially prepared for each population and kept in a culture room protected with ant wells. After allowing five days for oviposition, the adults were sieved out and transferred to fresh rearing containers. This process was repeated in order to get a regular supply of adult insects of known age. Rearing containers were regularly examined and the wheat flour was sieved every day following the first sighting of adult beetle to obtain one day old adults. Such adults were maintained in plastic containers containing fresh wheat flour for 15 days. Adults (17 ± 2 day old) obtained in this manner was used for bioassay.

Susceptibility of six populations of *T. castaneum* was evaluated against FCI recommended insecticide, malathion, by residual film method of bioassay. Technical grade insecticide (Sigma Aldrich) dissolved in acetone was used for bioassay. Stock solution of 1000 ppm was prepared in volumetric flasks using acetone as solvent and was diluted serially to obtain required concentrations.

A preliminary bioassay was conducted by using wide range of concentrations to arrive at actual concentration required for bioassay for all the six populations, separately. Bioassay involved a minimum of six concentrations for each population, including control, which were replicated thrice. One millilitre of prepared concentration of insecticide was pipetted out into a 9 mm Petri plate. In the case of control, acetone alone was used. Petri plate was rotated thoroughly to obtain a thin and uniform film of insecticide/solvent over the surface of Petri plate. After air drying, ten adult insects were released into each Petri plate. Insects were pre-starved for a period of two hours before bioassay. Observation on mortality was taken after 48 h. Moribund insects were counted as dead. LC_{50} was calculated using probit analysis (Finney, 1971) for each population using Polo PC software. Mortality was corrected as per Abbott's formula (Abbott, 1925) whenever required. Resistance ratio for each insecticide was calculated using the formula given below.

Resistance ratio=

$$\frac{LC_{50} \text{ of Field collected population}}{LC_{50} \text{ of susceptible population}}$$

Angamaly population of *T. castaneum* was the least susceptible to malathion with LC_{50} value 6949.80 ppm. The LC_{50} of Mulangunnathukavu population was 6157.30 ppm, and that of Valiyathura population was 5873.02 ppm. Olavakkode and Thikkodi populations had LC 50 values of 5727.94 ppm and 5703.48 ppm, respectively. The susceptible IARI strain had the lowest LC_{50} of 520.76 ppm (Table 1).

Comparing the LC_{90} values, Angamaly population had the highest LC_{90} value (23042 ppm) and the susceptible strain from IARI had the lowest (1392.74 ppm). LC_{90} values of Mulangunnathukavu, Valiyathura, Thikkodi and Olavakkode population were 18540 ppm, 14030 ppm, 15525 ppm and 16121 ppm, respectively (Table 1).

Chi-square values of all the probit regression analysis were below the table values, which indicated that the populations were homogenous. On comparison of the fiducial limits of LC_{50}, it was observed that all the field collected populations had a significantly higher level of LC_{50} when compared with the susceptible strain. Among the various populations, Angamaly population showed 13.34 fold resistance to malathion (Table 2). Resistance ratio for Mulangunnathukavu population was 11.82, for Valiyathura 11.27, for Olavakkode 10.99, and for Thikkodi population it was 10.95.

Resistance ratio calculated based on LC_{90}, showed no change in the order of resistance among the field collected populations of *T. castaneum*. Resistance ratio was 10.07 for Valiyathura population, 11.14 for Thikkodi population, 11.57 for Olavakkode population, 13.31 for Mulangunnathukavu population and 16.54 for Angamaly population (Table 2).

Table 2. Resistance to malathion in different populations of *Tribolium castaneum*

Population	Resistance ratio	
	LC_{50}	LC_{90}
Mulangunnathukavu	11.82	13.31
Valiyathura	11.27	10.07
Thikkodi	10.95	11.14
Olavakkode	10.99	11.57
Angamaly	13.34	16.54

Comparison of Log dose – probit (ld-p) lines of different populations showed a significant rightward shift in the field collected population from that of laboratory maintained susceptible strain (Fig. 1).

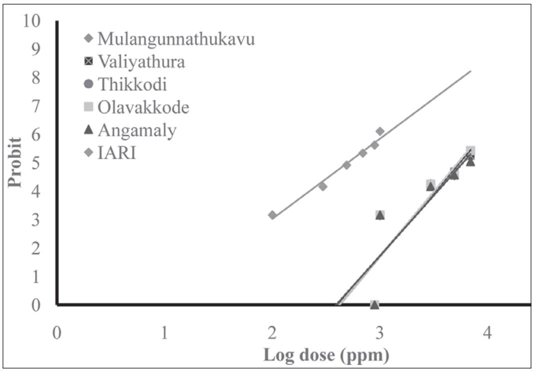

Figure 1. Log dose – Probit line of malathin

Table 1. Susceptibility of different population of *Tribolium castaneum* to malathion

Population	Heterogeneity		LC_{50} (ppm)(95 % fiducial limit)	LC_{90} (ppm)(95 % fiducial limit)	Slope
	d.f.	X^2			
Mulangunnath-ukavu	3	2.24	6157.30(4923.70- 8716.10)	18540(11897 – 46370)	2.68
Valiyathura	3	0.64	5873.02(4931.60–7583.60)	14030(9945 – 30177)	3.39
Thikkodi	3	2.58	5703.49(4679.50 -7574.90)	15525(10613 – 33139)	2.95
Olavakkode	3	2.64	5727.94(4648.70 -7769.90)	16121(10792 – 36288)	2.85
Angamaly	3	0.94	6949.80(5353.90-10987.00)	23042(13547 – 75302)	2.46
Susceptible	4	1.79	520.76(434.20 – 613.10)	1392.740(1085.29-2122.24)	3.00

LC_{50} = Concentration (ppm) calculated to give 50 per cent mortality; LC_{90} = Concentration (ppm) calculated to give 90 per cent mortality

The ld-p lines of all the field collected populations were of same slope and were cluttered together, indicating that all the populations tested in this study were homogenous in response to malathion.

Malathion resistance in *T. castaneum* has been a global phenomenon from 1962 onwards (Champ and Dyte, 1976). Resistance levels detected in the current study is in concurrence with the findings of studies Srivastava et al. (2001), wherein 0.725 to 24.53 fold resistance to malathion was detected in *T. castaneum* populations collected from 13 National Seed Programme (NSP) centres throughout the country. Similarly, 16-21 fold resistance to malathion in *T. castaneum* was detected by Rajak et al. (1973) and 17.25 fold resistance was detected by Dhaliwal and Chawla (1995).

This is the first report of malathion resistance in *T. castaneum* populations from FCI godowns of Kerala. The homogenous response observed in this study might be due to the same level and frequency of exposure to malathion, as FCI followed a common protocol for pesticide sprays across its godowns. Malathion has been used for insect pest management in FCI godowns from 1970s onwards (Champ and Dyte, 1976). Malathion sprays are being carried out in FCI godowns at an interval of 15 days as per the quality control specifications of FCI.

According to Arnaud and Haubruge (2002) malathion specific resistance in *T. castaneum* is wide spread and resistant phenotype has almost completely replaced the susceptible phenotype. The frequency of resistant gene is stable in the natural population even after the withdrawal of pesticide exposure. It was argued that there was little or no reproductive disadvantage between malathion resistant and susceptible strain (Haubruge and Arnaud, 2001; Arnaud and Haubruge, 2002). This could be the reason for the widespread and uniform detection of malathion resistance in *T. castaneum* in the present investigation. Hence efforts should be directed towards the management of malathion resistance in FCI godowns of Kerala.

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