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Efficacy of different concentrations of insecticides on potential mealybug predator Australian Lady Bird Bettle *Cryptolaemus montrouzieri* Mulsant under laboratory conditions

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Abstract
The present study on "Efficacy of newer insecticides on potential predator Australian lady bird beetle, *Cryptolaemus montrouzieri* (Mulsant) under laboratory conditions was conducted under controlled at Agriculture Entomology Section, College of Agriculture, Pune (M.S.) during the year 2017-18. The data indicated that there was significant difference in mortality of adult of *C. montrouzieri* due to different insecticides at various concentrations. Among the insecticides tested, Cyantranilliprole recorded significantly lower mortality (19.16%) and was found less toxic insecticide over rest of the insecticides followed by Buprofezin (25.00%), Flonicamid (28.33%) and Acetamaprid (38.33%), respectively. However in the treatment clothinidin (41.66%) highest mortality of adult of *C. montrouzieri* was observed.

Keywords: Mealy bug, lady bird beetle, insecticide

Introduction
Australian lady bird beetle *Cryptolaemus montrouzieri* Mulsant is one of the important potential predator as long–term solution because of its self perpetuating ability, persist even when the mealy bug is at low population densities in biological control of mealy bugs on different fruit crops and keeping the populations mealybugs below economic injury level. Thus, the Australian lady bird beetle is accepted by most of the fruit growers as a potential bio-agent for the control of mealy bugs on different fruit crops. In India, *C. montrouzieri* @ 20 per plant found solely responsible for the decline of *Mechanolicoccus. hirsutus* on sapota within two months in Bangalore [8]. In order to achieve the best pest control at higher level as well as safety to the consumers, integration of chemical insecticides and bio-agents has been followed as IPM strategies. Keeping the view in mind, this study was conducted to know efficacy of insecticides on *C. montrouzieri.*

Materials and Methods
Mass rearing of host insect mealy bugs, *Maconellicoccus hirsutus* Green on red pumpkins and *Phenacoccus solenopsis* Tinsley on sproutsed potato
Mass culturing of host insect, *M. hirsutus* and *P. solenopsis* is one of the basic needs in the laboratory experimentation of *C. montrouzieri*. The culture of mealy bug, *M. hirsutus* was collected from local orchards and kept in cages containing red pumpkins (*Cucurbita maxima* Duch.) while the culture of *P. solenopsis* made available from Central Institute of Cotton Research, Nagpur and multiplied on sprouted potatoes.
Mealy bugs were reared on red pumpkins to obtain the pure laboratory culture throughout the period of research work as per the method used by Singh (1978). For rearing of *M. hirsutus* mealy bugs, ripened red pumpkins with prominent ridges and grooves and bearing small stalk for easy handling were selected and cleaned with water to get rid of any dust on them. To prevent rotting, the pumpkins surface were treated with 0.1 percent carbendazim solution (Bavistin 50% WP @ 1 g/ lit water). The wounds on the pumpkins were plugged with paraffin wax. The ovisacs of *M. hirsutus* obtained from the stock culture of mealy bug and transferred over the pumpkins. The pumpkins infested with mealy bugs were kept on iron stands in the specially designed wooden cages (45× 45× 45 cm) with glass door in front, glass on top and backside and lateral sides fitted with wire mesh.

These cages were arranged on slotted angle iron racks in the rearing room. In order to maintain darkness at the rearing site, individual racks were completely covered with black muslin cloth. The matured *M. hirsutus* females were developed within 30 to 35 days from the fully infested pumpkins. For mass rearing of mealy bugs, *P. solenopsis* mealybugs, the iron trays of size 30× 25× 10 cm^3 were used for developing sprouts on potato. Sandy silt soil was filled in the trays with 5 cm layer and 10-15 whole tubers were placed at about 2 cm apart on a layer of soil in the trays which were covered with slightly moist soil. These trays were kept in the racks in the rearing room and watered gently. Temperature in the rearing room was maintained between 21 and 25ºC. Ovisacs of the mealy bugs obtained from the stock culture were placed over the sprouts and culture of mealy bug was developed within 25 days.

Mass rearing of predator, Australian Lady Bird Bettle, *Cryptolaemus montrouzieri* Mulsant on mealybug colonies:
The predator *C. montrouzieri* was reared after development of sufficient culture of *M. hirsutus* on pumpkins and *P. solenopsis* sprouts on potato. The adults of *C. montrouzieri* beetles were released on these pumpkins and potatoes inside the separate cages. Burlaps strips were attached to the fronts of each rack to accommodate pupating to *C. montrouzieri* grubs and the racks were again darkened. The emerging adults were collected in plastic vials. The beetles were also fed with honey–agar diet which consisted agar–agar powder 1 g, sugar 20 g, honey 40 cc and distilled water 100 ml intermittently. The diet was prepared by boiling sugar in 50 ml water in a beaker, in which 1 g agar–agar powder was added and again boiled for 30 to 60 seconds. In another beaker, 40 cc honey was diluted with remaining 50 ml water. This honey solution was mixed with the content of first beaker and again boiled for 30 to 60 seconds. Aril sized drops of this mixture were laid on a plastic sheet and allowed to cool for 10 to 15 minutes. After becoming sufficiently hard, these arils were fed to *C. montrouzieri* beetles.

Efficacy of different concentrations of insecticides on Australian Lady Bird Bettle *Cryptolaemus montrouzieri* Mulsant under laboratory conditions.
This laboratory experiment on Efficacy of newer insecticides on potential predator Australian lady bird beetle, *C. montrouzieri* under laboratory conditions was carried out in the Bio-Control Laboratory, Division of Agril., Entomology to study the efficacy of various insecticides at below recommended, recommended and above recommended concentrations to adults of *C. montrouzieri.*
Five different insecticides were tested against adults of *C. montrouzieri* following film or residue deposit method. The desired concentrations of insecticides were prepared by taking the known quantity of insecticides on weight/volume basis and dissolved in 1000 ml distilled water. The concentrations of test insecticides were worked out by calculating the quantity of toxicant using following formula:

$$\text{Quantity of insecticidal formulation required} = \frac{\text{Desired strength}}{\text{Percentage of insecticide in formulation}} \times \text{Quantity of spray material required}$$

Each treatment was replicated for four times. In all the cases, commercially available formulations were used as insecticide solution and was prepared fresh whenever required.
In each treatment, 1 ml of fresh aliquot of desired concentration prepared from commercially available formulation dissolved in distilled water was pipetted out and poured 0.5 ml on upper lid and 0.5 ml on lower lid of the petriplate (9 cm dia).
The lids were rotated so as to have a thin film of toxicant coated all over the inner surface of the plates and allowed it for drying under electric fan with medium speed for 5 minutes to evaporate excessive water.
Then sufficient numbers of adults (24 hr old) were collected from the mass culturing unit.
Ten such grubs were released in each treated petriplate duly closed with lids. Each insecticidal concentration was replicated four times with ten 24 hr-old–adults as treatment. Just before release, mealy bugs (*M. hirsutus*) egg masses were placed as food in the petriplates. The observations on mortality were recorded after 24, 48 and 72 hrs exposure of the grubs to the treated surfaces. Fresh mealy bug egg masses were provided at 24 hrs intervals as food [1].
Average per cent mortality of grubs as well as adults were work out for each treatment at different intervals. The mortality data thus obtained were further corrected by using Abbott's formula (1925) and transformed into angular values. The data were then subjected to statistically analysis by using Factorial completely randomized design [9].

Table 1: Classification system for recognizing adverse effects of pesticides on beneficial and non-target arthropods as given by IOBC (Hassan, 1992) [7].

Class	Category	% Mortality (Laboratory studies)
Class 1	Harmless	< 30%
Class 2	Slightly Harmful	30-79%
Class 3	Moderately Harmful	80-98%
Class 4	Harmful	> 99%

Table 2: Different insecticide concentrations tested to study their influence on mortality on adult of *C. montrouzieri*

Treatments	Name of Insecticide	Recommended Concentration (%)		
		Below dose	At dose	Above dose
T1	Buprofezin	0.03	0.05	0.07
T2	Clothinidin	0.001	0.003	0.005
T3	Cyantranilliprole	0.009	0.018	0.03
T4	Flonicamid	0.007	0.015	0.03
T5	Acetamaprid	0.001	0.002	0.004

Result and Discussion
Efficacy of different concentrations of insecticides to adult of C*ryptoleumus montrouzieri* by dry film method
Among various concentrations, the variation of dose above recommended level proved significantly unsafe for mortality. The mortality was 27.00% under below recommended level while it was 34.50% when the dose was above recommended level (Table 3)

Table 3: Efficacy of different newer insecticides and their concentrations on mortality of adult C*ryptoleumus montrouzieri* under laboratory conditions.

Tt. No.	Name of insecticide	Concentration of insecticides as per recommended dose											
		After 24 hours of spray				After 48 hours of spray				After 72 hours of spray			
		Below	Recomd	Above	Mean	Below	Recomd	Above	Mean	Below	Recomd	Above	Mean
T1	Buprofezin	7.50 (13.82)	12.50 (20.45)	20.00 (26.55)	13.33 (20.27)	15.00 (22.49)	17.50 (24.52)	25.00 (29.87)	19.16 (25.63)	17.50 (24.52)	25.00 (29.87)	32.50 (34.70)	25.00 (29.70)
T2	Clothinidin	22.50 (28.21)	25.00 (29.87)	27.50 (31.53)	25.00 (29.87)	32.50 (34.54)	35.00 (36.20)	37.50 (37.71)	35.00 (36.15)	40.00 (39.15)	42.50 (40.65)	42.50 (40.65)	41.66 (40.15)
T3	Cyantranilliprole	5.00 (9.21)	5.00 (9.21)	7.50 (13.82)	5.83 (10.74)	10.00 (18.42)	15.00 (22.49)	20.00 (26.18)	15.00 (22.36)	17.50 (24.52)	17.50 (24.52)	22.50 (28.21)	19.16 (25.75)
T4	Flonicamid	10.00 (15.85)	12.50 (20.45)	15.00 (22.49)	12.50 (19.60)	15.00 (22.49)	22.50 (28.21)	30.00 (33.04)	22.50 (27.91)	22.50 (28.21)	27.50 (31.53)	35.00 (36.20)	28.33 (31.98)
T5	Acetamaprid	22.50 (28.21)	25.00 (29.87)	27.50 (31.53)	25.00 (29.87)	30.00 (33.04)	35.00 (36.20)	35.00 (36.20)	33.33 (35.15)	37.50 (37.71)	37.50 (37.71)	40.00 (39.21)	38.33 (38.21)
Mean Concentrations		13.50 (19.06)	16.00 (21.97)	19.50 (25.18)		20.50 (26.19)	25.00 (29.52)	29.50 (32.60)		27.00 (30.82)	30.00 (32.86)	34.50 (35.79)	
	Factors	C.D.(0.05)	SE(m)			C.D.(0.05)	SE(m)			C.D.(0.05)	SE(m)		
	Insecticide	5.45	1.90			3.33	1.16			2.77	0.96		
	Concentration	4.22	1.47			2.58	0.90			2.15	0.75		
	Interaction(I x C)	NS	3.30			NS	2.01			NS	1.67		

*(Figures in parentheses are arc - sin transformed values)

Interaction effect

The interaction effect at 24 hour, 48 hour and 72 hour interval showed same pattern. The interaction effect of different insecticides and different concentrations was non - significant. The data was indicated that cyantranilliprole showed 5.83, 15.00 and 19.16 percent mortality at insecticide concentrations 0.009, 0.018 and 0.03% respectively, which indicate that it was harmless to the predator *C. montrouzieri*. Effects were found as above by laboratory testing, using limited numbers of test insects but there were deficit of information on this line. But some studies on other species of non - target insect found that cyantranilliprole was safe. Brugger *et al.*, (2010) [3] reported diamide cyantranilliprole have a favorable ecotoxicological profile and are relatively safe to insect natural enemies. From the data it was also observed that buprofezin found 13.33, 19.16 and 25.00 percent mortality at 0.03, 0.05 and 0.07% concentrations respectively; it was at par with flonocamid which killed 12.50, 22.50 and 28.33 percent mortality at 0.007, 0.015 and 0.03% concentration of test solution. It indicated that it was harmless to the predator *C. montrouzieri*. The study was in close agreement with Cloyd and Dickinson (2006) and Hallikatti *et al.*, (2009) [4, 6] who reported 10 - 20% and 18.29% mortality respectively. Anjitha (2010) [2] reported buprofezin as least toxic to the *C. montrouzieri.* Halappa *et al.*, (2013) [5] found buprofezin as safest causing minimum per cent mortality of 20.00 percent.

The data also indicated that clothinidin and acetamaprid were at par with each other and both were showed 25.00% mortality at below recommended level (*i. e.* 0.001%). Acetamaprid killed 33.33, 38.33 percent mortality a0.002, 0.004% concenrations respectively, while clothinidin observed 35.00, 41.66% mortality respectively at 0.003 and 0.005% concentrations. Both the insecticides observed to have slight toxicity to *C. montrouzieri.* Tank *et al.*, (2007) [10] recorded acetamaprid least toxic and caused 19.83% mortality while Halappa *et al.*, (2013) [5] recorded higher adult mortality by acetamiprid (93.33%). This variation was due to use of different formulations and concentration of test solution.

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