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Evaluation of antifeedant potential of insect growth regulators against phytophagous pest, *Papilio demoleus* (Lepidoptera: Papilionidae) under laboratory conditions

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Abstract

The feeding deterrence of three commercial Insect Growth Regulators; Diflubenzuron, Precocene II and Rakshak were evaluated at 1.0, 0.5, 0.25, 0.125, 0.06, 0.03, 0.015 and 0.007 per cent concentrations for larvae of the lemon butterfly, *Papilio demoleus* in the laboratory. Results showed that the Diflubenzuron, Precocene II and Rakshak treated larvae showed maximum antifeedant activity of 84.92, 83.60 and 83.38 at 1% concentration respectively. The antifeedant activity were increased with increasing concentration. All three Insect Growth Regulators exhibited significant antifeedant effect, and *Papilio demoleus* larvae on treated leaves quickly stopped feeding and dropped off treated leaves, resulting in no or minimal damage on the treated lemon leaves.

Keywords: Antifeedant activity, *Papilio demoleus*, insect growth regulators

1. Introduction

Pests are the serious problems to many of the major food and industrial crops grown in India, causing annual yield losses estimated to 30 to 60 per cent. Consequently India depended heavily on the use of synthetic pesticides. Synthetic chemical pesticides can be very effects on health and the natural environment and the quality of agricultural products. The lemon butterfly, *Papilio demoleus* is an economically important pest whose larval forms cause serious damage to citrus family in the field by devouring large quantity of foliage during the later stages of their development Bhutani and Jotwani (1975) [3], Srivastava, (1993) [18]. The caterpillars feed voraciously and cause extensive damage to nurseries and young seedlings leaving behind midribs only. Severe infestation results in defoliation of the tree [3] and leads to retarded plant growth and decreases fruit yield [11]. Information on the morphometry and biology of citrus butterfly on Sweet orange will be useful to evolve effective management strategy, against citrus leaf miner [12]. Nowadays, the use of synthetic pesticides due to their high efficacy and reliability of pest control has become popular [2]. Besides, these pesticides have some negative effects also causing ecological damage and health hazards. Currently, synthetic pesticides are used in controlling crops insect pests and have usually provided strong defence against insect pests. Worldwide, it is estimated that approximately 1.8 billion people engage in agriculture and most of them use approximately 5.6 billion pounds of synthetic pesticides to protect the food and commercial products that they produce [6]. Synthetic pesticides and their metabolites have high persistence in soil, water and crops themselves and therefore affect environment and the health of human being during preparation, application and the consumption of crops. [16]. Hence, most of the advanced countries have banned the practical use of few insecticides. Therefore, to minimize the environmental pollution and upsurge of secondary pest problems, there is an urgent need to bring a change in the pest control strategy from reliance on use of pesticides to less toxic and safe methods of pest control. In this context, the modern approaches in pest management i.e. the use of Insect Growth Regulators (IGRs) has become the most promising way of insect management because of their specific and non-toxic effect to human beings, domestic animals, fishes and beneficial insects including parasites, predators and pollinators Sabtharishi and Shankarganesh (2016) [13]. Many workers have reported the antifeeding of different Insect Growth Regulators (IGRs) on various harmful insects but no systemic work has been done on IGRs against various pests [19].

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The present work have been planned to evaluate the antifeeding effects of different Insect Growth Regulators (IGRs), viz.; Diflubenzuron (moult inhibitor), Triterpenoid azadirachtin; Rakshak (neem based), and Precocene on larvae of lemon butterfly (*Papilio demoleus* Linn.). In the present study *P. demoleus* L. was selected as a test species to evaluate the antifeedant activity by using the insect growth regulator hormones.

2. Materials and methods

2.1 Test insect

The lemon butterfly, *P. demoleus* is a key pest of citrus in India. It feeds voraciously on vegetative growth of citrus plants throughout the year. It is most destructive to citrus seedlings as well as new flushes.

2.2 Mass Rearing of *Papilio demoleus* In the Laboratory:

Eggs and early larval instars were collected from the lemon nurseries and lemon plants and reared in the laboratory on fresh lemon leaves food was supplied daily in Environmental Chamber and maintained at 28±1°C temperature, 75-80% R.H. Third instar larvae of desired age groups were sorted out. The fully grown larvae were allowed to pupate on branch

of lemon leaves. Soon after emergence, adults were transferred on potted plants of lemon covered with a glass chimney for egg laying. The mouth of each chimney was covered with a muslin cloth secured with a rubber band. The cotton bolls soaked with 10% glucose solution were hanged with the help of a thread to provide food for adults. The eggs laid on leaves were removed from the slits of lemon leaf margins and were kept in petridishes for hatching. The newly hatched larvae were transferred on soft, newly grown up leaves of lemon in petridishes with the help of camel hairbrush. The completely grown fourth instar larvae were sorted out and placed in a separate glass dish at room temperature for the experiment.

2.3 Preparations of different concentrations of test compounds:

Different concentrations of IGRs (Diflubenzuron, Precocene II and Rakshak) were prepared by adding desired quantity of distilled water. These IGRs have been registered in our country and are commercially available. For this purpose, the 10% stock solution was prepared for each test compound by the formula given below:

$$\text{Amount of test compound} = \frac{\text{Quantity of solution required} \times \% \text{ of solution desired}}{\text{Strength of formulation available}}$$

The desired concentrations of Diflubenzuron, Precocene II and Rakshak were prepared as from the stock solution by diluting with desired amount of distilled water.

2.4 Antifeeding test against *Papilio demoleus* Linn.

Leaf pieces of 2.0 cm sq. were cut from the lemon leaf by means of rectangular metal designed for this purpose and shrinkage was directly calculated by putting it on graph paper. Measured leaf pieces were dipped in Insect Growth Regulator Hormone formulation solution for 2 second and solvent was evaporated under fan for ½ hour. On treated leaf piece was kept in each Petridish having blotting paper over moist cotton

wool in order to avoid desiccation of leaves. Each treatment was replicated three times. On 24 hour starved larvae was released on the treated leaf material in each Petridish. Observations were recorded after 48 hours and area of leaf piece left over was measured. Leaf treated with solvent only was taken as control. The percentage of antifeedant activity was calculated using the formula ^[6] Percentage feeding and leaf area protected over control were calculated by the following formula:-

$$\text{Percentage feeding} = \frac{\text{Leaf area given} - \text{Corrected leaf area}}{\text{Leaf area given}} \times 100$$

$$\text{Leaf area protected over control} = \frac{\text{Percentage protection in treated} - \text{Percentage protection in control}}{100 - \text{Percentage protection in control}}$$

3. Result and Discussion

Antifeeding effectiveness of different IGRs against the larvae of *Papilio demoleus* was evaluated on the basis of leaf area protection and leaf area consumed over control as depicted in Table 1-3.

3.1 Effect of Diflubenzuron

It is inferred from the table 1 that the leaf area protected by Diflubenzuron was 84.92 per cent at its 1.0 per cent concentration. The lowest per cent protection of leaves i.e.

70.34 due to treatment was observed at the lower concentration of 0.007 per cent. The rest of the treatments behaved intermediary and the leaf area protected ranged 74.62 to 85.01 per cent when the concentrations used were 0.25 to 1.75 per cent. The consumption of leaf area by the larvae of *Papilio demoleus* was lowest in 2.0 per cent concentration of Diflubenzuron which resulted only 13.70 consumption of leaves followed by 0.5, 0.25, 0.125, 0.06, 0.03 and 0.015 per cent concentration which gave 14.99, 16.56, 18.41, 20.11, 22.18 and 25.38 per consumption respectively.

Table 2: Showing the antifeeding effect of Insect growth regulator of Diflubenzuron on the *Papilio demoleus* Linn.

S. No.	Concentration in per cent	Mean feeding per cent	Mean per cent concentration	Corrected per cent protection due to treatment
1.	1.00	13.70	86.30	84.92
2.	0.50	14.99	85.01	83.50
3.	0.25	16.56	83.44	81.78
4.	0.125	18.41	81.59	79.24
5.	0.06	20.11	79.89	77.87
6.	0.30	22.18	77.82	75.59
7.	0.015	25.38	74.62	72.07
8.	0.007	29.66	70.34	67.37
9.	Control	90.90	9.10	---

3.2 Effect of Precocene (II)

The data on per cent feeding and protection due to treatment (table-2) revealed that the per cent feeding decreased with increased in the concentration of the IGRs Precocene II. Its concentration of 1.0 per cent resulted only 14.90 per cent feeding of leaves, whereas its lowest concentration 0.007 elicited 26.71 per cent consumption of larvae. The rest of the treatments behaved intermediary showing the range of 23.00 to 15.88 per cent consumption. It has also been observed that percentage protection of leaf area increased with increase in

concentration as they showed 73.29, 77.00, 78.94, 80.54, 81.02, 82.67 and 84.12 per cent leaf protection in 0.25, 0.03, 0.06, 0.125, and 0.5 per cent concentration, respectively. All the used concentration proved superior over control in protecting lemon leaves by the larvae of *P. demoleus*. These compounds either inhibited JH biosynthesis or were inhibitors of enzyme action. These were synthetic analogs of precocenes, stimulators or inhibitors of JH degradation or acted in an antagonistic manner at the target tissue level i.e. JH receptor levels.

Table 3: Showing the antifeeding effect of Insect growth regulator of Precocene on the *Papilio demoleus* Linn.

S. No.	Concentration in per cent	Mean feeding per cent	Mean per cent concentration	Corrected per cent protection due to treatment
1.	1.00	14.90	85.10	83.60
2.	0.50	15.88	84.12	82.53
3.	0.25	17.33	82.67	80.93
4.	0.125	18.98	81.02	79.11
5.	0.06	19.46	80.54	78.59
6.	0.30	21.06	78.94	76.83
7.	0.015	23.00	77.00	74.69
8.	0.007	26.71	73.29	70.61
9.	Control	90.90	9.10	--

3.3 Effect of Rakshak

It is evident from the (table 3) that the leaf area protected by Rakshak was 85.90 per cent in case of its 1.0 per cent concentration. The lowest percentage protection of leaves i.e. 66.29 due to treatment was observed at 0.007 per cent concentration of rakshak. The rest of the treatment behaved intermediary and leaf area protected ranged 71.03, 73.29, 78.01, 80.99, 82.00 and 83.89 per cent when the concentrations used were 0.25, 0.03, 0.06, 0.125, and 1.75 per cent respectively. The minimum consumption of lemon leaves (15.10%) was noted at 1.0 per cent concentration. It was closely followed by 0.5 per cent which resulted in 16.11 per cent consumption. However, they proved superior to rest of the treatments. All the used concentration was found superior to control against *P. demoleus* Linn. (Table-3). Antifeedant is a chemical that inhibits the feeding without killing the insect directly, while the insect remains near the treated foliage and

dies through starvation [4]. The plant products and active compounds may act as antifeedants, disturb insect growth, development and inhibit oviposition. Azadirachtin had higher activity of feeding deterrence at 200ppm and reduced the food consumption, at 150 and 100ppm were found to have a moderate activity of feeding deterrence. Similar findings were reported earlier by the main active compound in the *C. odorata* wood, gedunin, had previously shown moderate antifeedant activity against many insect species [7], so antifeedants have some physiological or toxic actions on insects, depending on the treatment concentrations. Similar results were observed in *Spodoptera mauritia*, *Ephesia kuehniella* Zell and *Manduca sexta* when subjected to azadirachtin [20]. Similar observations were also noticed in *P. demoleus* treated with plant products such as andrographolide and Costunolide [17, 18].

Table 3: Showing the antifeeding effect of Insect growth regulator of Rakshak on the *Papilio demoleus*

S. No.	Concentration in per cent	Mean feeding per cent	Mean per cent concentration	Corrected per cent protection due to treatment
1.	1.00	15.10	84.90	83.38
2.	0.50	16.11	83.89	82.27
3.	0.25	18.00	82.00	80.19
4.	0.125	19.01	80.99	79.08
5.	0.06	21.99	78.01	75.84
6.	0.30	26.71	73.29	72.61
7.	0.015	28.97	71.03	68.12
8.	0.007	33.71	66.29	62.91
9.	Control	90.90	9.10	--

4. Conclusion

It is concluded that, these concentrations should enhance the management of lepidopterous pests in the vegetable agro ecosystems because they do not persist in the environment, have unique modes of action, low mammalian toxicity, and may be potentially compatible with natural enemies. These various concentrations of Insect Growth Regulators were having a profound effect on larval reduction of *P. demoleus*. These research works can be of great importance for the farming community in many areas of the developing world. The major thrust of this work is its adaptability for use by

small scale farmers plagued by the challenge of not being able to afford conventional pesticides on the market.

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