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RESEARCH ARTICLE

INFLUECNE OF SOLASODINE A PURE COMPOUND OF *SOLANUM AVICULARE* ON FEEDING ACTIVITY AGAINST *PAPILIO DEMOLEUS* L (LEPIDOPTERA: PAPILIONIDAE) LARVAE

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ABSTRACT

The use of plant products as pesticides against crop pests is gaining importance in recent years due to environmental and health hazards posed by synthetic organic pesticides. Plant materials comprises rich source of phytochemicals which are highly toxic to many insect species without phytotoxic properties. The lemon butterfly *Papilio demoleus* is an important pest whose larval forms causes serious damage to citrus family, in the field the caterpillars feed on leaves and prefer blossoms and young ones. In the present study to assess the influence Solasodine, which was isolated from the *Solanum aviculare* was used the feeding intensity of *Papilio demoleus* larvae to evaluate its antifeeding activity. A large leaf disc 36.5 sq.cm of lemon was used. Solasodine was sprayed with different concentrations such as 50, 100, 150, 200ppm uniformly on either side by using an atomizer and placed in a Petri dish. Single *Papilio demoleus* fourth instar larva was introduced into each Petri dish for observation at 24hrs, 48hrs and mean average of the ten sets were taken for the activity. The consumed area was measured graphically and also with Planimeter and the percentage leaf area protection was also calculated. Parallel control Petri dishes were maintained for each experiment. Solasodine showed a potent antifeedant activity against fourth instar larva of *Papilio demoleus.*

INTRODUCTION

The world population is expected to reach 9 billion by 2050. This global population growth of 2 to 3 billion people over the next 40 years, so increase in food demand of 70% by 2050 (Bruinsma, 2009; UNDESA, 2009). To feed the burgeoning population, we need to produce more food. Providing ample food for the ever-growing global population is only the first part of the challenge the second and more important part is to produce this in a safe and sustainable manner (Kumar, 2012; Suresh Kumar, 2013). According to the FAO report, US $120 billion losses worldwide were caused by 20–40% decrease in crop yield due to the attack from pathogenic organisms and insect pests (Zhou, 2001). Controlling this polyphagous pest has become the challenging work in agriculture field. To ensure the stable and high output of crops, huge amount of pesticides were applied to control the pests and this not only caused serious environmental pollution but also induced in a wide range of pesticide resistance (Armes *et al.*, 1997). Meanwhile by applying these chemical pesticides different varieties of pest predators were killed and the ecological balance was destroyed, thereby causing pest resurgence and a greater outbreak of secondary pests (Jiang *et al.*, 2000). Due to this reason many researchers have involved on alternative control methods. Botanical and microbial pesticides are having advantage over chemical pesticides by its highly effective, safe and ecologically acceptable nature. In recent years biopesticides have been gaining increased attention and interest among those concerned with developing environment friendly, safe and integrated crop management for pest management (Leonard *et al*., 2000). Natural products derived from plants and microorganisms have been used for insect control (Tang *et al.,* 2009). Azadirachtin, Forskoin, Sesamin and some other natural products, bacterial and viral based insecticides were used to control different pests. Most of the pesticides from microorganisms have been isolated from entomopathogens and the terrestrial environment (Zhang, 1996).

Papilio demoleus L. (Lepidoptera: Papilionidae) is one of the most serious pests of cultivated citrus fruits that are of high economic value in local and international markets (UNCTAD, 2006; FAOSTAT, 2010). *Papilio demoleus* was also known as citrus swallowtail and lime butterfly the widely distributed species occur in both the tropical and sub tropical regions of the world including India and the Indo-Pacific region (Vane-Wright and DeJong, 2003). *Papilio demoleus* attacks all species and varieties of citrus including lime, orange and lemon (Lewis, 2009). Its larvae are voracious feeders with older instars being the most damaging. The larvae prefer young foliage, and heavy infestation can lead to complete defoliation of orchards and nurseries (Lewis, 2009; Satya Singh *et al.*, 2011). In the present investigation an attempt was made to

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evaluate the antifeedant activity of solasodine, a plant extract against *Papilio demoleus* larvae.

MATERIALS AND METHODS

Insect Maintenance

Eggs and young larvae of *Papilio demoleus* were collected from citrus fields with no history of pesticide use, and reared in the laboratory at 27° ± 2°C and 75 ± 5 % R.H. and 12 hrs light –12 hrs dark photoperiod. Larvae were kept in glass troughs and provided with fresh citrus leaves daily. Adults were maintained in ventilated cages and provided with a 10% honey solution for feeding and fresh citrus leaves twigs for oviposition.

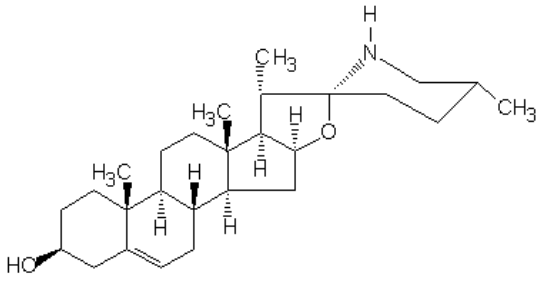
Test Compound

For the present investigation Solasodine, a natural product which was isolated from *Solanum aviculare* was provided by natural products division, Department of Chemistry, Osmania University, and Hyderabad. The family Solanaceae comprises a number of plants widely known for medicinal value. The natural products of medicinal significance such as steroidal lactones, glycosides, alkaloids and flavanoids were isolated and used for various studies from this family (Klocke *et al.,* 1989).

SOLASODINE ($C_{27}H_{43}NO_2$)

Solasodine is a steroidal alkaloid commonly present in Solanum species such as *Solanum aviculare*. Solasodine is a natural precursor for the preparation of steroidal hormones and drugs which are used in contraceptives, arthritis and behavioral drugs. It is also used as a starting material to prepare steroid compounds with glucocorticoid-like properties, which have selective cytotoxicity against certain cancer cells including squamous cell carcinoma. Solasodine is also reported to have anti-inflammatory activity and antiproliferative activity against human colon and liver cancers.

Chemical Structure of Solasodine



Preparation of test solution

Acetone was used as the solvent in preparing the test solutions since the solubility of the test compound was very high in acetone. 1% stock solution was prepared using acetone and 200, 150, 100, 50ppm concentrations were prepared by dilution method from the stock.

Antifeedant activity evaluation method

A large leaf disc 36.5 sq.cm of lemon was sprayed with Solasodine uniformly on either side by using an atomizer and placed on moist filter paper in a Petri dish. Single *Papilio demoleus* fourth instar larva was introduced into the Petri dish. Controls of *Papilio demoleus* were maintained with similar leaf discs simultaneously. 10 such Petri dishes were taken for each experiment. The setup was kept for observation at 24hrs, 48hrs and mean average of the 10 sets were taken for the activity. The consumed area was measured graphically and also with Planimeter. The percentage leaf area protection was calculated as suggested by earlier authors (Ben Jannet *et al.,* 2000; Duraipandiyan *et al.,* 2011).

$$\text{Antifeedant activity} = \frac{\text{Leaf area consumed in control – treated leaf}}{\text{Leaf area consumed in control + treated leaf}} \times 100$$

Statistical analysis

The results are expressed as Mean ± SD and data was statistically analyzed by one-way ANOVA, with the level of significance set at $p<0.05$ using SPSS software.

RESULTS AND DISCUSSION

Table 1. Mean and SD of undamaged leaf area (sq.cm) and antifeedant activity with different concentration treatments of Solasodine

Conc in ppm	No of Insects	Mean ± SD After 24 hrs	Mean ± SD After 48 hrs	Antifeedatnt activity after 24hrs	Antifeedatnt activity after 48hrs
200	10	30.57 ± 0.71*	25.38 ± 0.81*	78.65	73.25
150	10	27.28 ± 0.75*	22.57 ± 0.96*	68.66	67.44
100	10	24.63 ± 0.84*	19.06 ± 1.31*	61.39	61.07
50	10	21.58 ± 0.85*	15.71 ± 1.02*	55.77	53.27
Control	10	18.69 ± 0.61*	10.92 ± 0.67*	------	-----

Mean and SD values are significant at $P < 0.05$.

The botanical extracts from the plant leaves, roots, seeds, flowers and bark in their crude form has been used as conventional insecticides from centuries. One plant species may possess activities with a wide range, for example extracts from the neem tree are antifeedant, oviposition deterrent, repellent and growth-regulating (Schmutterer, 1995; Raguraman, 1997). Effect of seed extract on the fecundity and fertility of *T. castaneum* and *T. confusum* of *J. gossypifolia* were reported by Khanam *et al.,* (2008). The MeOH extracts of *J. gossypifolia* were assayed for their toxicity against the early fourth instar larvae of *C. quinquefasciatus* (Rahuman *et al.,* 2008). The antifeedant activity of Solasodine in the present investigation showed highest activity at 200ppm concentration that was 78.65% and 73.25% respectively at 24hrs and 48hrs exposure. Undamaged leaf area was observed at 24hrs and

48hrs of Solasodine treatment with various concentrations. At 50ppm treatment the undamaged leaf area after 24hrs and 48hrs were 21.58 ± 0.85sq.cm and 15.71 ± 1.02 and undamaged leaf area at 200ppm treatment that was observed at 24hrs and 48hrs treatment 18.69 ± 0.61 and 10.92 ± 0.67 sq.cm respectively.

In the present study, the plant product Solasodine significantly reduced the feeding activity but did not prevent it entirely. The antifeedant effect of Solasodine is the key antifeedant ingredient in the *Solanum aviculare* result from stimulation of specific deterrent chemoreceptor on the mouthparts, together with interference of the perception of phagostimulants by another chemoreceptor (Mordue Luntz, 2000). Although the larvae could discriminate between treated and untreated food, solasodine leaves did not remain unharmed to *Papilio demoleus* in test which may be related to the nature of the preparation, and the insect must taste it in order to respond to it. According to Danielson (1996) and (Hiiesaar *et al.*, 2009) a substance that inhibits insect feeding only for a defined time or rate is referred to as a relative antifeedant; in contrast to absolute antifeedant that is described as substance which the insects refuse to eat in any case. In our test the feeding activity of was reduced by 3 - 5 times as in comparison with the control. The test compound revealed a moderate antifeedant activity even at a low concentration that is 50ppm, where it showed 55.77% and 53.27% antifeedant activity against *Papilio demoleus* for 24hrs and 48hrs respectively. All the values were statistically significant over control ($P \leq 0.05$) (Table 1).

Efficiency of conversion of ingested food (ECI), Relative consumption rate (RCR), and efficiency of conversion of digested food (ECD) also decrease as extract concentration increase. ECI is an overall measure of an insect's ability to utilize the food that it ingests for growth. A drop in ECI indicates more food is being metabolized for energy and less is being converted to body substance. ECD also decreases as proportion of digested food metabolized for energy. When IV instar larvae of *Papilio demoleus* were fed with Solasodine in their diet, growth rates decrease as extract dosage increased. This corresponded to a decrease in the consumption rate. It is likely that this decrease in consumption rate is due to the antifeedant nature of the extract and this account for the majority of the decrease in growth rate of *Papilio demoleus.* It indicates that ingested Solasodine may be exhibited some chronic toxicity. This effect can produce a reduction in the rate of embryonic development, alteration of metamorphic, occurrence of super numary larval stages, inability to molt and even death of the insect. Similar results were also seen with hirtin and *T. connaroides* extract when tested against *Peridroma saucia* and *S. litura* (Xie *et al*., 1994) and *Trichilia americana* extract against the larvae of *Spodoptera litura* (Deborah *et al*., 2001). However, hirtin also showed similar effects on nutritional indices of *P. saucia* larvae. It would appear that the extract acts primarily as an antifeedant and has to be ingested for any toxic effects to be seen. This result coincided with earlier results of Kannan who had isolated violacein from *Chromobacterium violaceum* claimed more than 80% antifeedancy at 1000ppm against H.armigera (Kannan, 2008). Xiang *et al.* (2011) isolated novel macrocyclic lactone from *Streptomyces microflavu*s, showed high acaricidal activity against adult mites and nematocidal activity against *Caenorhabditis elegans*. Pavunraj *et al*. (2011) demonstrated that *Pergularia daemia* leaf extract showed antifeedant activity of 70.3% and 71.82% against H. armigera and S. litura respectively. The present results also agree with the findings of Muthu *et al*. (2010), who reported that the ethyl acetate extract of *Atalantia monophylla* exhibited 78.67% antifeedant activity against *Earias vittella*. Jeyasankar *et al.* (2010) performed the antifeedant activity of ethyl acetate crude extracts of *Syzygium lineare* leaves and obtained significant antifeedant activity of 91.58% against the fourth instar larvae of S. litura. Many researchers have confirmed that ethyl acetate extracts of different plants showed antifeedant activity against *Anisomeles malabarica*, S. litura, L. orbonalis, and E.vittella etc. (Jeyasankar *et al.*, 2010; Elango *et al*., 2011; Pavunraj *et al*., 2012).

Conclusion

In the present studies Solasodine was used to evaluate the antifeedant activity against *Papilio demoleus* fourth instar larva and it is proved to be suitable to use at field level without any harm to natural enemies with worker safety. This study clearly allowed a fast screening for antifeedant activity and toxicity. These findings would be useful in promoting research aiming at the development of new agent for pest control from indigenous plant resources. Further it is concluded that the made of action of Solasodine to be a combination of antifeedant action and pest digestive toxicity.

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