Development inhibitory effect of *Acacia auriculiformis* extracts (Coquillett) (Diptera: Tephritidae) on *Bactrocera cucurbitae* (Coquillett) (Diptera: Tephritidae)

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**ABSTRACT**

The effect of acetone and water extracts of *Acacia auriculiformis* A. Cunn. bark was investigated on biology of *Bactrocera cucurbitae* (Coquillett). The eggs, larvae and adults were treated with different concentrations (1, 5, 25, 125 and 625 ppm) of the bark extracts. Both the extracts significantly prolonged the larval period and total developmental period. Percentage pupation, percentage emergence, oviposition and egg hatching decreased in a dose dependent manner. The LC₅₀ values were lower for the acetone extract as compared to water extract indicating that acetone extract was more toxic than water extract. The present findings demonstrated the potential of the plant extracts in controlling melon fruit fly.

**Keywords:** *Acacia auriculiformis*, plant extracts, *Bactrocera cucurbitae*, development.

**INTRODUCTION**

Plants contain thousands of compounds which are virtually an untapped reservoir of pesticides that can be used directly or as templates for synthetic pesticides. India being a tropical country has a vast resource of plants, a large number of which have not been screened for their pesticidal properties. *Acacia auriculiformis* A. Cunn (Fabaceae) is an important medicinal plant. It has a rich source of tannins and terpenoids along with polyphenols (Singh and Sehgal, 2001). It’s antimutagenic, cytotoxic and antioxidant activities have been reported (Singh et al., 2007a, 2007b, 2007c). However, it has not been explored for its pesticidal potential.

*Bactrocera cucurbitae* is distributed widely in temperate, tropical and sub-tropical regions of the world but India is considered as its native home (Dhillon et al., 2005). It damages over 81 plant species and plants belonging to the family Cucurbitaceae are preferred most which include fruits of bitter gourd (*Momordica charantia*) and muskmelon (*Cucumis melo*), snap melon (*Cucumis melo* var. *mondatica*) and snake gourd (*Trichosanthes anguina* and *T. cucumeria*) (Sapkota et al., 2010). Plant products (Shivayya and Kumar, 2008), *Arisema* sp. (Kaur 2009) Imapct of methoprene (Ihsan ul Hag et al. 2010) on this pest was reported. Therefore, in the present study it was envisaged to investgate the effect of the extracts of the plant on the development of the melon fruit fly, *Bactrocera cucurbitae* (Coquillett).

**MATERIALS AND METHODS**

**Plant material and extract preparation**

*A. auriculiformis* procured in the month of September was identified by comparing it with the specimen available in the herbarium (Voucher No. 6422) of the Guru Nanak Dev University. The fresh bark of *A. auriculiformis* was selected, washed and dried in oven at 30°C overnight and ground to fine powder with grinder. The extracts of the bark powder were prepared by dissolving the bark powder in acetone and water and these were concentrated in rotary vacuum evaporator. The dried extracts were weighed and preserved in sealed bottles in a refrigerator after making stock solutions of the required concentrations. The desired amount of stock solution required for making various test concentrations for the experiment was calculated as given below:

\[
\text{Amount of stock solution required (ml)} = \frac{\text{Concentration required}}{\text{Concentration of the stock solution}} \times \text{Amount of diet (ml)}.
\]

**Insect**

The infested bitter gourd, *Momordica charantia* (L.) and guava, *Psidium guajava* L. were collected from the vegetable market and scanned for melon fruit fly, *B. cucurbitae*. The flies were identified on the basis of criteria given in the taxonomic keys (Kapoor, 1993; White and Elson-Harris, 1992). The cultures of *B. cucurbitae* were maintained in insect culture room under a constant photoperiod (10L: 14D), temperature (28±2°C) and relative humidity (70-80%). They were fed on a diet of sugar solution and protein incorporated with vitamin E.

**Experiments with eggs**

About 100 gravid females were released for 6-8 h in a wire mesh cage, in which 3-4 pumpkin pieces were placed for oviposition. The pumpkin pieces were removed and the top layer of the pumpkin flesh was teased with the help of
scalpel and a needle in the saline water. The eggs were collected from water with the help of camel hair brush, washed in distilled water and placed on the sterilized filter paper to soak the extra water. These eggs were dipped in 5ml solution of various concentrations of bark extracts for one minute. The eggs were dried on filter paper and transferred to petri dishes with moist filter paper. These Petri dishes were kept in B.O.D. incubator and observed at a dial periodicity for egg hatching. All the five concentrations of bark extracts had six replications with 20 eggs in each replication.

Experiments with larvae
The first instar, second instar and third instar larvae were collected after 48-56 hours, 64-72 hours and 88-96 hours respectively and used for the experiments. The larvae of same age were procured by releasing around 100 gravid females in a cage for oviposition on pumpkin pieces for an interval of 6 to 8 hours. The larvae were transferred to artificial diet containing different concentrations of the plant extracts along with control. There were six replications for each concentration of all the extracts, each having 15 larvae for each experiment. Observations were made for larval period, pupal period, total developmental period, percentage pupation and percentage of adult emergence.

Experiments with adults
Pumpkin pieces (2.5×1×1.5) cleaned and incubated for 48h at 27±1°C and 65±5% R. H. were dipped in different concentrations of extracts for 30 seconds and allowed to dry at room temperature. Five pairs of 20 days old flies were transferred to small wire cages (15×15×16 cm). These flies were provided with protinex and sugar solution as their food and plant extract impregnated pumpkin pieces for oviposition. Water treated fruit served as control. Three replications were provided for each treatment including control. Pumpkin pieces were removed after 6 hours and dissected under a binocular microscope to count the number of eggs laid by the female flies.

Statistical analysis
The data was subjected to one way analysis of variance (ANOVA), using SPSS (10.0 for window) computer software programme to assess the significance of observations on the development period, pupation and emergence of B. cucurbitae. The LC₅₀ value was calculated for adult emergence through probit analysis (Finney, 1971) and the means were separated using Tukey HSD test (Assistat v7.5beta).

RESULTS AND DISCUSSION
In tropical countries having a rich biodiversity of plants, botanical pesticides are fast emerging as a viable component of integrated pest management. The toxic effect of plant extracts is due to the synergistic effect of secondary compounds which are postulated to have evolved for the plant’s defense. These compounds confer protection to crops through reduction of fitness to insect herbivores. Although the plant extracts may be less toxic than the conventional pesticides (Koul et al., 2008) but they are relatively safe and ecofriendly.

Eggs
From the present study it was evident that extracts of A. auriculiformis had an adverse effect on the development of B. cucurbitae. A concentration dependent decreased egg hatching was observed when melon fly eggs were treated with different concentrations of A. auriculiformis acetone and water extracts. Between the two extracts, acetone highly reduced the egg hatching (45.75%) than water extract (49.15%) at 625ppm (Figure 1). Reduced egg hatching in B. cucurbitae has also observed by Nair and Thomas (2001) after treatment of eggs with different concentrations of Acorus calamus L. (Acoraceae) extracts. Similar impact was reported in peach fruit fly, Bactrocera zonata (Saunders) (Mahmood and Shoeib, 2008) and western cherry fruit fly (Renden et al., 1998). The egg shell in Diptera is comprised of vitelline membrane, waxy layer and chorion (Margaritis, 1985a). The chorion consists of several proteins which are crosslinked via di- and tri-tyrosine bonds and provide the egg shell with the required hardness and elasticity. The enzyme, Peroxidase which is a functional and structural component of the chorion involved in the hardening process is inhibited by Phloroglucinol, a phenolic compound produced during the breakdown of plant polyphenols (Mindrinos et al., 1980; Margaritis, 1985a, 1985b, 1985c; Keramaris et al., 1991). Decreased egg hatching observed in the present study could be attributed to the secondary compounds particularly polyphenols present in the bark extracts of A. auriculiformis which might have interfered in the protein synthesis or cross linking of proteins.

Figure 1. Effect of extracts of A. auriculiformis on egg hatching of B. cucurbitae

Larvae
The larval period and total developmental period of the insect prolonged in the first instar and second instar
larvae, however it was reduced in the third instar larvae (Tables 2 and 3). This could be a stage specific response of the insect to the plant extracts. All first instar larvae were dead at 625 ppm concentration of both the extracts. As observed in eggs, acetone extract has more impact than water extract on larval too. In a study conducted by Mustafa and Al-Khazraji (2008), delayed larval development of the mosquito, *Culex pipiens molestus* Forskal was observed with the extracts of *Azadirachta indica* Jack. Previously, 12 different New Zealand *Acacia excelsa* gymnosperm species extracts were tested against larvae of the mosquito, *Culex quinquefasciatus* Say (Tauro et al., 2005), *C. quinquefasciatus*, *A. stephensi* and *A. aegypti* (Sakthivadivel and Daniel, 2008). Larvicidal effects of other species of *Acacia* has been reported against *Aedes aegypti* (L.) and *Culex quinquefasciatus* (Say) mosquito larvae (Tauro et al., 2004; Chaubal et al., 2005), *C. quinquefasciatus*, *A. stephensi* and *A. aegypti* (Sakthivadivel and Daniel, 2008). Larvicidal effects of *Pavonia zeylanica* and *Acacia ferruginea* have been reported in the late third instar larvae of *C. quinquefasciatus* (Vahitha et al., 2002). Other plant extracts such as *Parthenium hysterophorus*, *Peganum harmala* L., *Sawsswera lappa* C. B. Clarke and *Valariiana*...
Table 4. Percentage emergence of *B. cucurbitae* when first instar, second instar and third instar larvae were treated with different extracts of *A. auriculiformis*.

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<td>83.33±5.58°</td>
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**Significant at 1% level ; Means within a column followed by the same letter are not significantly different according to the Tukey test (P=0.05).**
Influence of Acacia auriculaformis extracts on Bactrocera cucurbitae.

Figure 2. Effect of extracts of A. auriculaformis on the oviposition of B. cucurbitae.

The present study clearly revealed the susceptibility of B. cucurbitae to both the extracts of A. auriculaformis as indicated by their deleterious effects on growth and development of melon fruit fly. Further studies are being undertaken to isolate and identify the bioactive compounds from the most potent extract and evaluate its effect on the melon fruit fly.

REFERENCES


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