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BEHAVIOURAL AND ELECTROPHYSIOLOGICAL RESPONSES OF THE COCONUT PEST, *RHYNCHOPHORUS FERRUGINEUS* (COLEOPTERA: CURCULIONIDAE) TO HOST, NONHOST AND ALTERNATE HOST PLANT VOLATILES

NEELAKANTHI E. GUNAWARDENA and M.N.A. SWARNAKANTHI
Department of Chemistry, University of Kelaniya, Kelaniya.

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Abstract: Steam bark distillates of the typica and aurancica varieties of *Cocos nucifera* (host plant) *Terminalia catappa* (Kottamba) and *Albizzia amara* Boiv. (Albizzia) (nonhost plants) and *Areca catechu* L. (Puwak) and *Areca concinna* Thw. Enum. (Lentheti) (alternate hosts) were subjected to electrophysiological and behavioural assays to compare the attractiveness to the coconut pest, *Rhynchophorus ferrugineus* F. (Coleoptera: Curculionidae).

In the electroantennogram (EAG) assay, the steam distillates of *C. nucifera* form aurancica elicited the highest response of 79.1% (relative to standard) and the form typica (young bark) 55.6% to the antenna of *R. ferrugineus* both values being significantly different ($p<0.05$, ANOVA, Scheffe's test) from those of the nonhosts *T. catappa* and *A. amara* (10.9% and 5.9% respectively). EAGs of old bark (51.4%) steam distillate of *C. nucifera* form typica did not differ significantly from that of the young bark distillates (55.6%). The two alternate hosts, *A. catechu* and *A. concinna* showed moderate EAG activities (29.4% and 25.9% respectively) between those of host varieties and nonhosts.

In a behavioural assay (choice test) using an olfactometer the steam distillates of host palms were more attractive ($p<0.05$, ANOVA, Scheffe's test) to the red weevil than those of the nonhosts. *C. nucifera* form aurancica was more attractive (79.9% attraction to the baited arm) than typica. Between young and old palms of *C. nucifera* form typica, no significant difference ($p>0.05$, ANOVA, Scheffe's test) in the behavioural activity was observed. The alternate hosts showed moderate activity.

Key words: Coconut pest, EAG and behavioural assay, host and nonhost volatiles, *Rhynchophorus ferrugineus*.

INTRODUCTION

Host plant recognition of insects is known to be primarily based on olfaction.[1] An array of relationships between host plant volatiles and Coleopteran behaviours have been recognized in the recent years.[2-4] There are ten described species in the genus *Rhynchophorus* and all are suspected to use palms as hosts. Seven species are considered major pests on *Cocos nucifera* Linneaus,[5] in different geographical locations throughout the world. *Rhynchophorus ferrugineus*, F. (Coleoptera: Curculionidae) commonly known as the red weevil, is a major pest on the coconut palm in Sri Lanka.[6] Nearly 10% of the damage on coconut in Sri Lanka is attributed to this pest.[7] It is known that this pest uses *Phoenix dactylifera* (date palm), *Borassus flabellifer* (palmyrah palm), *Oreodoxa regia* (sago palm), *Metrozylen sagu* (serdang palm), *Livistonia chinensis*, *Arenga saccharifera* (sugar palm), *Phoenix sylvestris* (toddy palm), *Corypha umbraculifera* (tailpot palm) etc. in addition to *Cocos nucifera* (coconut palm) as the host.[8] In Sri Lanka the red weevil uses all varieties of *C. nucifera* (family; Palmae) *viz* .form typica (Pol), aurancica (Thambili) and nana (Kundira) as the preferred host while it is known that other palms belonging to palmae such as *Caryota urens* (Kithul), *Areca catechu* L (Puwak) etc. are alternate hosts.

In a previous behavioural bioassay it was shown that the host finding process of the red weevil is chemically mediated. In a choice test this pest was strongly attracted to the coconut bark steam distillate.[9] On the basis of the above, a comparative study has been carried out presently to investigate the preference of the red weevil in the following steam distillates; 1) host, alternate hosts and nonhosts, 2) form aurancica and typica, 3) form typica old and young palms.

Chemical ecological studies such as the above, would provide a basis for understanding pest infestations. The knowledge will eventually be important in saving the vulnerable varieties of the coconut as well as other palms from red weevil attack. Currently, the red weevil infestations are not satisfactorily controlled.

METHODS AND MATERIALS

Chemical stimuli: Fresh bark (1kg) each of *C. nucifera* form typica young (<15 y) and old (>15 y), form aurancica, *A. catechu, A. concinna, T. catappa* and *A. amara* were cut into small pieces and steam distilled for 5 h. The aqueous layer was extracted with diethyl ether (30 ml x 3) and the combined ether layers was subjected to evaporation of the solvent under reduced pressure down to 1 ml.

EAG assay: Newly emerged adult *R. ferrugineus* aged between 1-5 d were maintained in a laboratory and fed with sugar cane and water. Using the isolated antenna EAG assay of the above steam distillates were carried out by the procedure described previously.[11,12] 4-Methyl-5-nonanol (ferrugineol), the active component of the aggregation pheromone[13] was used as the internal standard. Each compound was assayed on three male and three female antennae. Each test compound was presented to the antenna six times and so was the standard. The effects due to the time and the individual antenna was removed by the use of an appropriate equation. All statistical comparisons of mean EAG responses of *R. ferrugineus* were done by the use of ANOVA and Scheffe's test unless otherwise stated.

Laboratory bioassay: Choice test was performed on a Y-shaped olfactometer[10] as described previously[9] using steam distillates[9] (0.01 ml) and 5-6 unsexed weevils in a batch. Seven replicates were run on each steam distillate. Mean percentage of weevils attracted for a particular steam distillate was compared by χ^2 - test and the attractiveness of different steam distillates were compared by ANOVA, Scheffe's test.

RESULTS

The mean responses of both female and male antenna of *R. ferrugineus* to the standard stimulations (10 μg ferrugineol) were of high magnitude. No significant difference ($p<0.05$, Student's test) in EAG standard was found between males (X=0.84 mV, SD ± 0.18) and females (X=0.80 mV, SD ± 0.15) and therefore, the data were pooled. The EAG activities of the bark steam distillates of test substances are presented in Figure 1.

Results of the EAG assay showed that the percentage EAG responses (compared to standard) of the weevils to their host palms, *C. nucifera* (form typica, 55.58%, SD ± 11.56: form aurancica 79.1%, SD ± 6.94) were significantly higher ($p<0.05$) than those of nonhosts, *T. catappa* (EAG 5.95%, SD ± 4.39) and *A. amara* (EAG 10.96%, SD ± 1.02). The alternate hosts, *A. catechu* and *A. concinna* elicited EAGs (25.92%, SD ± 7.37 and 29.4%, SD ± 8.95 respectively) lower than those of the hosts and higher than those of the nonhosts but statistically not significantly different from either (Fig. 1). The attractiveness of the old and young bark distillate of *C. nucifera* (typica), did not show significant difference ($p>0.05$) (EAGs 51.4%, SD ± 10.39: 55.5%, SD ± 11.56 respectively).

In the behavioural assay of the host varieties, the highest percentage of weevils were attracted to the baited arm by the form aurancica (79.9%, SD ± 5.3) and form typica (young) (74.7%, SD ± 7.6) at statistically equivalent levels. The steam distillates of young and old palms of *C. nucifera* (typica) did not differ significantly in their attractiveness (74.7%, SD ± 7.6 and 66.5%, SD ± 5.4) respectively. Towards the alternate hosts, *A. catechu* and *A. concinna* the attraction of weevils were moderate (57.1% SD ± 7.6 and 54.2%, SD ± 9.4 respectively) and differed statistically ($p<0.05$) from those of *C. nucifera*. Nonhost plants, *T. catappa* and *A. amara* elicited lowest attraction of 24.2%, (SD ± 4.2) and 25.7%, (SD ± 4.7 respectively) significantly different ($p<0.05$) from those of the host and alternate host (Table 1).

DISCUSSION

From the large number of recent investigations focused on the insect semiochemicals it has become apparent that the host finding behaviour of many species is primarily mediated by broad overlapping blends of volatile compounds.[14,15] This is consistent with the broad host ranges exhibited by many insect species.[16-19] The electrophysiological and behavioural assay yielded consistent results in the present study. The antennal receptors of the red weevil seems specially selective to the volatiles from palms belonging to the family Palmae. There may be a specific blend of compounds that make Palmae attractive to the weevil.

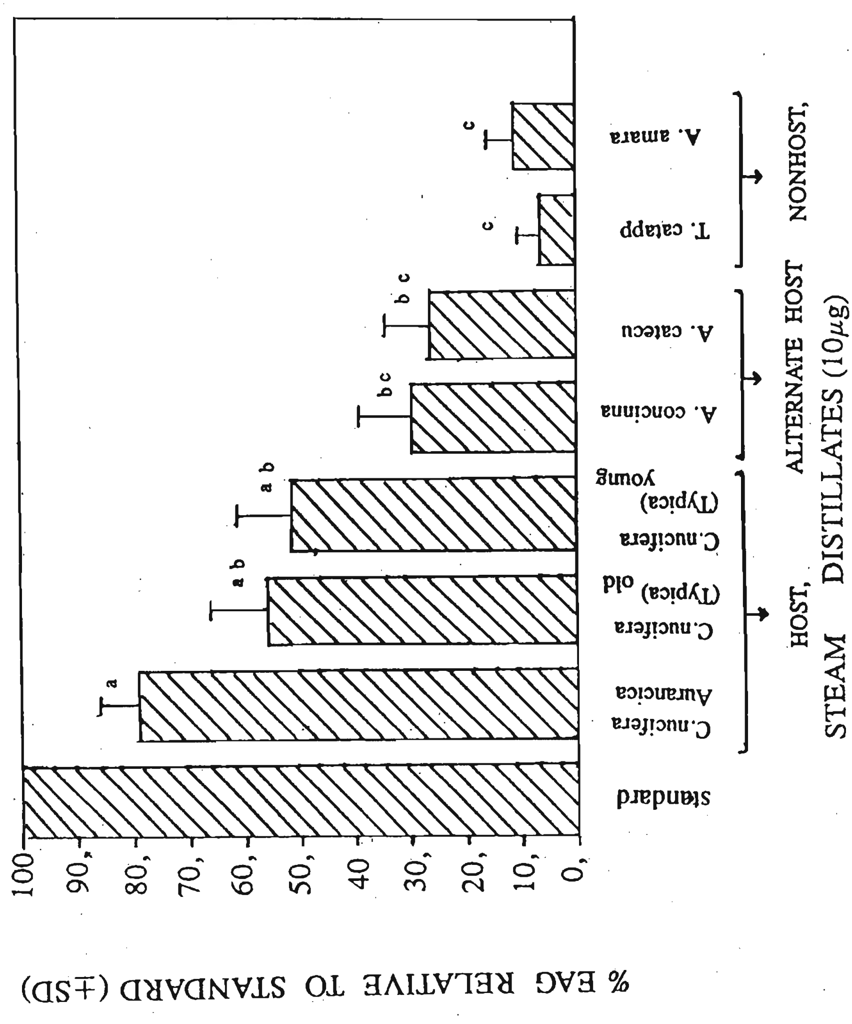

Figure 1: Mean EAG (Relative to standard) responses of *R. ferrugineus* to steam distillates of host, alternate hosts and nonhosts. Vertical bars indicate standard deviation. Means not followed by the same small letters are significantly different ($p<0.05$, ANOVA, Scheffe's test).

Table 1: Olfactometer bioassay (choice test) with adult *R. ferrugineus*.

		% mean of responding weevils**		% mean of non responding weevils
Stimulus#	Total No. of insects used*	Baited Arm A	Non-baited Arm B	Insect Tube + C
C. nucifera aurancica	39	79.9%[a,f] (5.3)	14.9% (4.3)	5.2% (2.4)
C. nucifera typica (young)	39	74.7%[a,b,f] (7.6)	17.6% (4.7)	7.7% (3.1)
C. nucifera typica (old)	38	66.5%[b,f] (5.4)	9.6% (4.5)	23.9% (5.8)
A. catechu	35	57.1%[c,f] (7.6)	10.1% (5.4)	32.8% (6.3)
A. concinna	35	54.2%[c,f] (9.4)	12.1% (6.7)	33.7% (5.5)
T. catappa	35	24.2%[d,f] (4.2)	20.0% (3.3)	55.8% (3.2)
A. amara	35	25.7[d,f] (4.7)	20.1% (4.3)	54.2% (5.9)
Distilled water	35	8.1%[e] (3.7)	7.9% (2.0)	84.0% (5.3)

\# 0.1 ml of the concentrated steam distillates were used as the bait.

* Each batch of insects consisted of 5-6 insects and 7 batches were used to assay one test substance.

** Values for standard deviation are given in parenthesis.

a-e Comparison between baited arms for different hosts. Means not followed by the same letter are significantly different, $p<0.05$ (ANOVA and Scheffe's test)

f Comparison between baited and non-baited arms for the same host are significantly different at $p< 0.05$ (χ^2-test).

Of the family Palmae, the preferred palm is *C. nucifera* as evident by the EAG and behavioural assay having greater attraction of the weevil compared to the other two palms belonging to palmae, Areca. Comparative studies on the the weevil incidences on *C. nucifera* and alternative hosts are not available in Sri Lanka; nevertheless it is known that coconut is highly preferred by the red weevil. The present study provides a verification on the above field observations.

C. nucifera form aurancica is believed to be the most vulnerable variety for red weevil attack and this field observation is well in agreement with the present results where the steam distillate of aurancica had the highest EAG (79.17%) and behavioural activity (79.9%) being significantly different ($p<0.05$) from those of the alternate hosts as well. In contrast, the activity of the form typica did not differ significantly from those of the alternate host.

An interesting observation is that the red weevil antenna did not show a significant difference ($p>0.05$) in the response to the young and old *C. nucifera* (typica) bark steam distillates. However, the red weevil incidents reported on young coconut palms are much higher than the older palms. One possible explanation is that the hardened barks of older palms have fewer injuries. Older palms become attractive to the weevils at the cut outer ends of the inflorescence when they are tapped for toddy and perhaps this indicates the high potency of the older palms as attraction sites once they are injured. In a separate study, the introduction of the red weevil larvae to the older coconut palms were shown to be unsuccessful mainly due to the hardness.[20] The difference in field activity could therefore be attributed to physical than chemical factors.

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