Field evaluation of synthetic kairomonal attractants against major lepidopteran pests of castor

**Abstract**

Aim: The tobacco caterpillar (*Spodoptera litura* F.), semilooper (*Achaea janata* L.) and capsule borer (*Conogethes punctiferalis* Guenee) are the most destructive lepidopteran insect pests of castor. Developing new environmentally benign alternative monitoring and management system for these pests may reduce the frequency of application of insecticides and impact on the environment. Therefore, the objective of the present study was to evaluate the attractiveness of synthetic kairomones against these lepidopteran pests under laboratory and field conditions.

Methodology: Antennal responses of lepidopteran species to six synthetic kairomones (benzaldehyde, benzyl acetate, p-anisaldehyde, phenyl acetaldehyde, 2-phenyl ethanol and methyl salicylate) were studied by electroantennogram (EAG). Lures of three synthetic kairomones elicited strong EAG responses viz., phenyl acetaldehyde, benzaldehyde and 2-phenyl ethanol were prepared in single and combinations and field tested using three trap designs (funnel trap, sleeve trap and water trap) for trapping of lepidopteran pests in castor.

Results: Phenyl acetaldehyde elicited strongest EAG responses for female and male moths of *S. litura* (-1.518 to -2.221 mV and -1.756 to -2.214 mV, respectively). Female and male moths of *A. janata* showed significantly greater EAG responses to benzaldehyde (-2.573 to -3.336 mV and -2.377 to -3.396 mV, respectively). The highest EAG responses of female and male moths of *C. punctiferalis* were elicited by 2-phenylethanol (-2.563 to -3.152 mV and -2.475 to -2.903 mV, respectively). In field experiment, water trap baited with phenyl acetaldehyde + 2-phenylethanol recorded significantly higher moth catches of *S. litura* (6.8 moths/trap/wk) and *C. punctiferalis* (5.8 moths/trap/wk).

Interpretation: Combination of phenyl acetaldehyde and 2-phenyl ethanol in water trap can be used for monitoring of the lepidopteran pests in castor. The synthetic attractants has potential in developing management methods such as mass trapping or 'attract and kill system' for the lepidopteran pests of castor.
**Introduction**

Castor (Ricinus communis L.) is an industrially important oilseed crop. Its seed oil has multifarious applications in production of a wide range of industrial products viz., dyes, detergents, plastics, printing ink, linoleum, patent leather, ointment, polishes, surface paints, adhesives, lubricants and hydraulic fluids. India has 70 % of world area with 87 % of the total world production in castor (Sarada et al., 2015). The current castor production in the country is 17.33 lakh tonnes from 11.05 lakh ha with a productivity of 1568 kg ha⁻¹ (Anonymous, 2016). Though castor productivity in India is more than the world average, there are several production constraints in the traditional rainfed castor growing areas of South India, among which insect pests and diseases dominate the scenario. A large number of insect pests are recorded on castor at different phenological stages of the crop, but damage due to lepidopteran pests viz., tobacco caterpillar, Spodoptera litura F. (Noctuidae: Lepidoptera); semilooper, Achaea janata (L.) (Noctuidae: Lepidoptera) and capsule borer, Conogethes punctiferalis Guenee (Pyralidae: Lepidoptera) are of greater economic importance. The avoidable yield losses due to insect pests on castor cultivars ranged from 17.2 to 63.3 % during kharif season (Lakshminarayana and Duraimurugan, 2014).

So far, the use of synthetic insecticides has been a major approach for controlling these pests. However, the indiscriminate and injudicious use of wide spectrum insecticides induced insecticide resistance in insects, pest resurgence, toxic effect on non target organisms including wide natural enemies and environmental pollution (Lakshminarayana and Raoof, 2005, Singh et al., 2017). Developing a new environmentally benign alternative monitoring and management system for these pests may reduce the frequency of application of insecticides and impact on the environment. Sex pheromones of lepidopteran pests have been widely used for monitoring, however they are species specific and attract only males. Further improvement in monitoring and management of these pests might be achieved with traps that attract females or both sexes. Since lepidopteran moths are highly attracted to the volatile cues present in the floral parts of host plants for both ovipositing and nectar feeding, this could be used as the basis for monitoring or management strategy (Burguiere et al., 2001). In the recent years, kairomones, a type of semiocchemicals which include the chemical volatiles emitted by plants have been experimentally demonstrated as attractants for trapping of several lepidopterans (Toth et al., 2010; Landolt et al., 2011). Management by ‘lure and kill’ using floral attractants for trapping of several lepidopterans (Toth et al., 2010; Duraimurugan, 2014).

**Test insects** : Larvae of tobacco caterpillar, Spodoptera litura; semilooper, Achaea janata and capsule borer, Conogethes punctiferalis collected during rainy seasons (July to November, 2012 and 2013) from castor fields at Research Farm, ICAR-Indian Institute of Oilseeds Research, Rajendranagar, Hyderabad were used to initiate the mass culturing. The larvae of S. litura and A. janata were mass reared in laboratory on castor leaves (cv. VP1). The larvae of C. punctiferalis were reared on fresh castor capsules (cv. DCS-9) in Insect Breeding Dish (90 x 40 mm with air hole size of 40mm, HiMedia, Mumbai). The larvae were provided with fresh capsules once in 3-4 days. After pupation, the sex of the pupae of all three species was determined on the basis of morphological features seen on the ventral side of abdomen. In female pupae, a median shallow furrow is on the VIII sternum. A medium groove surrounded by pad-like circular area is seen on the midventral side of IX segment on male pupae. Male and female pupae were transferred to individual specimen tubes (25 x 100 mm, Borosil, Mumbai) for adult emergence and the mouth of the tube was covered with a black muslin cloth. Adults were fed with 10 % sucrose solution absorbed on a cotton pad. The required numbers of male and female moths for different experiments were taken from the culture. All experiments and mass rearing were carried out under ambient (27 ± 2°C, 60 - 70 % RH) conditions with natural photoperiod (12:12 L:D).

**Test compounds** : Six synthetic kairomonal attractants viz., benzaldehyde, benzyl acetate, p-anisaldehyde, phenyl acetaldehyde, 2-phenyl ethanol (Sigma-Aldrich, India) and methyl salicylate (HiMedia Laboratories Pvt. Ltd., Mumbai) were selected on the basis of a bibliographic search for compounds identified as attractants of lepidopteran moths.

**Electroantennography (EAG)** : Electrophysiological response of one-day-old male and female moths of S. litura, A. janata and C. punctiferalis selected six synthetic kairomones along with air as check was determined by electroantennographic system (Syntech, Hilversum, Netherlands) using completely randomized design. Test compounds were subjected to EAG by applying at two different concentrations (0.1 µl and 1 µl). Three replications with five adults each were observed. Antennal response to the compounds in negative milliVolts was recorded.

**Field tests** : Lures of three synthetic kairomonal attractants found promising in the EAG (benzaldehyde, phenyl acetaldehyde and 2-phenyl ethanol) were prepared in single and combinations. Yellow Silicone Septa (Sun Lure, Chennai, India) were used in the experiment. Septa were first ultrasonicated for 6 hr in hexane and then extracted three times with hexane for a total of 24 hr, and air-
Attraction of lepidopteran pests of castor to synthetic kairomones was carried out during the active period of occurrence of all the three lepidopteran pests (September to October, 2014) coinciding with vegetative and primary spike development stage of the crop. The traps were randomly distributed equidistantly with 50 m between them and placed 0.5 m above canopy height. Traps were monitored once a week from mid September to mid October (standard weeks from 38 to 41) and the lures were changed once in two weeks. The number of moths caught per week for each individual trap was recorded and were removed from the trap. Mean moth catches per trap per week of the 4-week trapping period was used for statistical analysis.

Data were analyzed statistically using AGRES statistical software. EAG data were subjected to analysis of variance (ANOVA) and differences in pairs of means between treatments were separated using least significant difference (LSD). For field experiment, square root transformed data was subjected to analysis of variance (ANOVA) and differences in pairs of means between treatments were separated using least significant difference (LSD).

Six synthetic kairomone loaded lures were evaluated using three trap designs viz., funnel trap (Fero-T®, PCI Pvt. Ltd., Hyderabad, India), sleeve trap (Phero-Sensor™, Pheromone Chemicals, Hyderabad, India) and water trap (Wota-T®, PCI Pvt. Ltd., Hyderabad, India) in factorial randomized block design with three replications. Traps with septa not containing any synthetic kairomones were used as control. The experiment was carried out during the active period of occurrence of all the three lepidopteran pests (September to October, 2014) coinciding with vegetative and primary spike development stage of the crop. The traps were randomly distributed equidistantly with 50 m between them and placed 0.5 m above canopy height. Traps were monitored once a week from mid September to mid October (standard weeks from 38 to 41) and the lures were changed once in two weeks. The number of *S. litura*, *A. janata* and *C. punctiferalis* moths caught per week for each individual trap was recorded and were removed from the trap. Mean moth catches per trap per week of the 4-week trapping period was used for statistical analysis.

**Statistical analysis**: Data were analyzed statistically using AGRES statistical software. EAG data were subjected to analysis of variance (ANOVA) and differences in pairs of means between treatments were separated using least significant difference (LSD). For field experiment, square root transformed data was subjected to analysis of variance (ANOVA) and differences in pairs of means between treatments were separated using least significant difference (LSD).

**Fig. 1A**: Electroantennographic response of tobacco caterpillar, *Spodoptera litura* to synthetic kairomone attractants. Bars marked by the same letter are not significantly different (LSD test; P=0.05)
subjected to analysis of variance. Original data is shown in the table. Following ANOVA, differences between datasets were determined using LSD at $P = 0.05$ at all instances.

**Results and Discussion**

The mean EAG response of one-day-old female and male moths of tobacco caterpillar, *S. littura* to synthetic kairomones at two concentrations (0.1 and 1 µl) is shown in Fig. 1A. In both the concentrations, significantly highest EAG response of female moths were elicited by phenyl acetaldehyde (-1.518 and -2.221 mV at a concentration of 0.1 and 1 µl, respectively) and 2-phenylethanol (-1.531 and -1.578 mV at 0.1 and 1 µl, respectively) as compared to other synthetic kairomones (-0.827 to -1.534 mV). The response of one-day-old male moths at lower concentration (0.1 µl) were very high to 2-phenylethanol (-1.779 mV) and phenyl acetaldehyde (-1.756 mV). At higher concentration (1 µl), the male response to phenyl acetaldehyde was found to be very high (-2.214 mV) but did not differ significantly with other compounds (-1.435 to -1.962 mV).

Synthetic kairomones at both the concentrations evoked significantly greater EAG responses in semilooper, *A. janata* (Fig. 1B). The highest EAG responses of one-day-old female moths were elicited by benzaldehyde (-2.573 and -3.336 mV at a concentration of 0.1 and 1 µl, respectively). It was followed by 2-phenylethanol (-2.160 and -2.867 mV at 0.1 and 1 µl, respectively) and phenyl acetaldehyde (-1.739 and -2.598 mV at 0.1 and 1 µl, respectively) exhibited significantly larger response as compared to other synthetic kairomones (-1.350 mV to -2.271 mV) and air check (-0.113 to -0.215 mV). Similarly, one-day-old male moths also showed significantly greater EAG responses to

![Graph showing EAG responses of one-day-old female and male moths of Achaea janata](image)

Fig. 1B: Electroantennographic response of semilooper, *Achaea janata* to synthetic kairomone attractants. Bars marked by same letter are not significantly different (LSD test; $P = 0.05$)
More than 16 species of lepidopteran insect pests are reported to damage castor in India. However, tobacco caterpillar, *S. litura* (Noctuidae: Lepidoptera); semilooper, *A. janata* (Noctuidae: Lepidoptera) and capsule borer, *C. punctiferalis* (Pyralidae: Lepidoptera) are major lepidopteran species of regular occurrence and causing considerable yield loss in castor (Lakshminarayana and Duraimurugan, 2014.). Male and female moth attraction to and feeding from blooming plants has been noted for several lepidopteran pest species (Guedot et al., 2008). Floral chemicals or synthetic kairomones have been experimentally demonstrated as attractants for trapping several lepidopteran species (Meagher and Landolt, 2010; Toth et al., 2010). In order to identify the synthetic kairomones or floral chemicals that elicit attraction from major lepidopteran pests of castor, a method of screening is required. This could be achieved by using electroantennography (EAG), a technique developed for the identification of lepidopterous sex pheromones and successfully adapted for use in identifying a wide range of kairomones. As a prelude, the current study was undertaken to identify promising synthetic kairomonal attractants for major lepidopteran pests of castor. EAG response of tobacco caterpillar, *S. litura* A. janata

For synthetic kairomones evaluated, there was a significant difference on the EAG response of capsule borer, *C. punctiferalis* in both the concentrations (Fig. 1C). It was observed that the response of one-day-old female moths was significantly very high to 2-phenylethanol (-2.563 and -3.152 mV at a concentration of 0.1 and 1 µl, respectively) and followed by phenyl acetaldehyde (-2.269 and -3.098 mV at 0.1 and 1 µl, respectively). It was followed by benzaldehyde (-2.152 and -3.098 mV at 0.1 and 1 µl, respectively) that showed larger response as compared to other synthetic kairomones (-1.063 mV to -1.893 mV). Highest EAG responses of one-day-old male moths were elicited by 2-phenylethanol (-2.475 and -2.903 mV at 0.1 and 1 µl, respectively), phenyl acetaldehyde (-2.381 and -2.839 mV at 0.1 and 1 µl, respectively) and benzaldehyde (-2.248 and -2.744 mV at 0.1 and 1 µl, respectively) as compared to other synthetic kairomones (-1.317 to-1.930 mV) (Fig. 1C).
Table 1: Mean number of pest moth species captures in different trap designs baited with synthetic kairomones singly and their combinations in castor ecosystem (Kharif, 2014)

<table>
<thead>
<tr>
<th>Synthetic Kairomones</th>
<th>No. of S. litura moths/trap/wk</th>
<th>No. of C. punctiferalis moths/trap/wk</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Funnel trap</td>
<td>Sleeve trap</td>
</tr>
<tr>
<td>benzaldehyde</td>
<td>0.08</td>
<td>0.00</td>
</tr>
<tr>
<td>Phenyl acetaldehyde</td>
<td>0.08</td>
<td>1.00</td>
</tr>
<tr>
<td>2-Phenyl ethanol</td>
<td>0.17</td>
<td>1.08</td>
</tr>
<tr>
<td>Benzaldehyde + Phenyl acetaldehyde</td>
<td>0.00</td>
<td>0.17</td>
</tr>
<tr>
<td>2-Phenyl ethanol + Phenyl acetaldehyde</td>
<td>3.58</td>
<td>2.83</td>
</tr>
<tr>
<td>Phenyl acetaldehyde + 2-Phenyl ethanol</td>
<td>(2.02)</td>
<td>(1.82)</td>
</tr>
<tr>
<td>Control</td>
<td>0.00</td>
<td>0.00</td>
</tr>
<tr>
<td>Mean</td>
<td>0.56</td>
<td>0.76</td>
</tr>
</tbody>
</table>

Figures in parentheses are square root transformed values; Different letters on the same column indicate significant differences (LSD test; P=0.05).

Monitoring of pest population is an essential component of integrated pest management. Lepidopteron pest population in agricultural crops are monitored by capturing males in traps baited with synthetically-produced female pheromones. However, sex pheromones are highly species-specific and attract only males (Burguiere et al., 2001). Floral chemicals or synthetic kairomones are attractive to adult moths of both sexes, and thus have advantages over sex pheromones that catch only males. Hence, these chemicals can be used to enhance monitoring tools for a wide range of insect pests, lure moths away from host plants.
Attraction of lepidopteran pests of castor to synthetic kairomones

or removal of insects from field by "attract-and-kill" system and control populations through mass trapping (Meagher, 2002; Landolt et al., 2011). In the present study, preliminary field trapping experiment with three synthetic kairomones (2-phenylethanol, benzaldehyde and phenyl acetaldehyde) single and in combinations revealed that the combination of phenyl acetaldehyde + 2-phenyl ethanol baited traps captured significantly higher moths of S. littura and C. punctiferalis as compared to these chemicals tested single or in combinations. The results are in consonance with Landolt et al. (2011) who found significantly higher moth catches of true armyworm, Mythimna unipuncta (Lepidoptera: Noctuidae) in traps baited with phenylacetaldehyde + 2-phenylethanole as compared to these chemicals used alone. This also corroborates with the reports of Meagher and Landolt (2008) who reported combination of phenylacetaldehyde + methyl salicylate trapped 76 % higher moth catches of soybean looper, Chrysodeixis includens (Lepidoptera: Noctuidae) as compared to phenylacetaldehyde alone. Based on these results, it is postulated that combination of phenylacetaldehyde + 2-phenylethanol has potential to use in monitoring or management of lepidopteran pests of castor by mass trapping or luring and killing approaches.

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References


