



Research Article

Electrophysiological response of *Tetrastichus schoenobii* Ferriere (Hymenoptera: Eulophidae), an egg parasitoid of rice stem borer, *Scirpophaga incertulas* (Walker) (Lepidoptera: Crambidae) to the extracts of plants collected from rice ecosystem

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**ABSTRACT:** *Tetrastichus schoenobii* Ferriere is an efficient egg parasitoid on rice yellow stem borer, *Scirpophaga incertulas* (Walker). The electrophysiological response of females of *T. schoenobii* to the extracts of plants collected from rice fields was studied under laboratory conditions. The highest response was noticed to the host plant of the pest, *S. incertulas* and honey solution. Response was also noticed to the flowers of *Hyptis suaveolens*, a common weed in rice fields, mostly by female parasitoids to the flowers of the non-crop vegetation.

**KEY WORDS:** *Tetrastichus schoenobii*, *Scirpophaga incertulas*, electrophysiology, *Hyptis suaveolens*

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INTRODUCTION

Rice is infested by several species of pests and among them, the yellow stem borer, *Scirpophaga incertulas* (Walker), is considered as a serious pest throughout the Orient (Dale, 1994). It is regarded as monophagous with exclusive host specificity to rice. However, Zaheerdeen and Prakasa Rao (1983a,b) have shown that wild rice species such as *Oryza rufipogon* Grift, *O. nivara* Sharma et Shastry, *O. latifolia* Desv. and *O. glaberrima* (Steud) and a grass weed, *Leptochloa panicoides* (J. Presl.) act as potential alternate host plants for *S. incertulas*. Before the introduction of chemical pesticides, natural enemies played a significant role in maintaining the pest populations of rice at lower levels (Yasumatsu, 1967). Stem borers are parasitized by around 100 species of parasitoids (Nickel, 1964) and although all juvenile stages are attacked, eggs suffer highest parasitism (Ressig *et al.*, 1986). Three groups of egg parasitoids, namely, *Telenomus* spp., *Tetrastichus* sp. and *Trichogramma* spp. dominate the parasitoid complex (Ooi and Shepard, 1994). In India, five species of egg parasitoids have been recorded on the eggs of *S. incertulas* from different regions, viz., *Telenomus dignus* (Gah.), *T. dignoides* Nixon, *T. rowani* (Gah.), *Tetrastichus schoenobii* and *Trichogramma japonicum* Ashmead

(Hikim, 1979; Rai and Gowda, 1977). *T. schoenobii* is endoparasitic on stem borer eggs, but later instars of the parasitoid become predacious on other host eggs within the egg mass (Rothschild, 1970; Kim and Heinrichs, 1985). Each developing instar of *T. schoenobii* is capable of destroying 3-4 eggs before completing its development and therefore this species is considered as the best among the egg parasitoids. *T. schoenobii* is also considered to be a more predominant parasitoid than *Telenomus rowani* (Islam, 1991). The role of *T. schoenobii* as an efficient parasitoid was studied from several parts of India (Ram *et al.*, 1996, 1997; Pandya *et al.*, 1995) and in some geographical areas parasitization was observed up to 95% (Kim and Heinrichs, 1985; Kumar *et al.*, 2008).

Besides rice borer, *T. schoenobii* parasitizes borers of corn, sugarcane, wheat and many other grass species in South East Asia and India and a total of 10 stem borer hosts have been reported for *T. schoenobii* (Khan *et al.*, 1991). The detailed biology of this species was studied by several authors (Bhuiyan and Sufian, 1986). Hymenopteran parasitoids visit plants not only in search of hosts but also for feeding and mating (Jervis *et al.*, 1993). Volatiles of undamaged plants are important for

the location of flowers as food sources (Wäckers, 2004) and for the location of the prey (Elzen *et al.*, 1983; Benrey *et al.*, 1991). The behaviour of parasitoids is mediated by volatiles emanating from host plants as well as host insects. While host plant volatiles are known to act as long range attractants, host kairomones act as short range attractants. Studies on the behaviour of *T. schoenobii* are limited (Qiu *et al.*, 1984; Ding *et al.*, 1991; Bhuiyan and Sufian 1986; Zhang and Zhang, 1985). He (1984) reported eight kinds of sensilla on the antenna of *T. schoenobii* through scanning electron microscopy studies and *sensilla placodeum* Type I is involved in the oviposition behaviour of females. The assemblage of vegetation in a crop ecosystem may have profound influence on the behaviour of the host insects and their natural enemies. Several weed species are observed along the bunds and adjoining fields of rice which may have an influence on the behaviour of the parasitoids of stem borer. The behaviour of *T. schoenobii* to plant cues is not known till date. The present studies were conducted to understand the influence of the plant volatiles on the electrophysiological response of *T. schoenobii* and these studies are important to understand whether *T. schoenobii* utilizes plants for food or shelter.

## MATERIAL AND METHODS

### Cultures of *T. schoenobii*

Field collected egg masses of *S. incertulas* were brought to the laboratory and kept in small plastic vials (10 cm length and 2 cm dia.) for the emergence of adult parasitoids at  $26\pm 2^{\circ}\text{C}$ ,  $70\pm 5\%$  RH and a 12 hour photophase. After emergence, the adults of *T. schoenobii* were separated and reared in vials in groups along with 50% honey.

### Preparation of plant extracts

A survey was conducted to find out the weed flora of rice ecosystem in and around Mandya, Karnataka. All the plants found within and adjoining rice fields and harvested fields adjacent to rice fields were collected in polythene bags, brought to the laboratory and identified. After identification, one gram (fresh weight) from each plant material was weighed and extracted with 100ml of hexane (Analytical reagent grade, 95% purity, SD Fine Chemicals, Mumbai) in a rotary shaker at a speed of 100 rpm for 60 minutes. The extract was filtered through a Whatman's filter paper and the final solution was made up to 100 ml to represent 1% (weight/volume) concentration.

### Details of plant species studied

Plant species	Location from where collected
<i>Alternanthera ficoidea</i> (L.)	Mandya
<i>Asclepias curassavica</i> L.	Mandya
<i>Aristida</i> sp.	Channapatna
<i>Calotropis gigantea</i> (L.)	Mandya
<i>Cassia tora</i> L.	Mandya
<i>Cleome viscosa</i> L.	Channapatna
Grass	Mandya
<i>Hyptis suaveolens</i> (L.)	Mandya
<i>Malvastrum coromandelianum</i> (L.)	Channapatna
<i>Melia dubia</i> Cav.	Channapatna
<i>Morus alba</i> (L.)	Mandya
<i>Oryza sativa</i> L.	Mandya/Channapatna
<i>Panicum setigerum</i> (Roth.)	Mandya
<i>Parthenium hysterophorus</i> L.	Channapatna
<i>Spilanthes acmella</i> (Murri.)	Mandya
<i>Tephrosia purpurea</i> (L.)	Mandya

### Electroantennogram studies

Electroantennogram (EAG) studies were conducted in a Syntech® electroantennogram consisting of EAG amplifier (model AM-02) and Stimulus Controller (model CS-05) and IDAC (Intelligent Data Acquisition Controller) card of type AM 02 supplied by Syntech, Netherlands along with the Syntech EAG recording system for Windows version 2.6 (1997) EAG software supplied by Syntech, Netherlands. Two-day-old females of *T. schoenobii* were selected for the electroantennogram studies. The antennae of the adults were excised from the head carefully with minimal disturbance and the tip was trimmed and fixed between the two capillary electrodes filled with electroconductivity gel (Dia med make manufactured by, Sakthi Mediwin India, Puducherry), forming a continuous electrical circuit. Pure air filtered through activated charcoal granules was blown on the antennal preparation @ 720 ml per minute and a stimulus was given @ 720ml per minute through the stimulus controller. For the delivery of odours a non-adsorbent, odourless polystyrene tube (6mm outer diameter and 4 mm inner diameter) was used which was connected to the continuous flow and stimulus air flow delivery knobs by appropriate Y-tube connector with the Synthech stimulus controller unit (Model CS 05) on one hand and to the stainless steel stimulus delivery unit (type ST-02, Syntech, Germany) on the other hand. The tip of the

stimulus delivery unit was placed around 0.5mm distance from the antennal preparation. The stimulus was given for a period of 0.4 seconds and the recordings were done for a period of five seconds. An amount of 0.1 ml of the extract from the plants (containing 10ug of the plant material) was impregnated in a small strip of 75mm length and 1.5mm breadth Whatmann filter paper and presented as a stimulus to the antenna. Diluted honey (50%) was used as the control stimulus as diluted honey elicits good response in most of the parasitoids we have studied. The potential difference between the antennal tip and the base on stimulation with the odours was recorded through the EAG software® supplied by Syntech. Three replications were maintained for each treatment. The maximum amplitude recorded for different treatments in different replications was analysed through Duncan's Multiple Range Test.

## RESULTS AND DISCUSSION

This is the first electronantennogram study on any of the *Tetrastichus* spp. The lowest response of electrical conductivity was recorded to the air, indicating lower response due to mechanical receptors. The highest response of antenna was observed for honey, rice leaf extract and flowers of *Hyptis suaveolens* (Table 1). The response to honey indicates that the parasitoid utilizes the honey from plant sources for its survival. The least response was noticed to most of the leaf extracts of different plants including the monocot plants. Response to the leaves of rice indicated that *T. schoenobii* responded to the volatiles of the plants. Since the parasitoid is an obligate parasitoid of rice stem borer, obviously its response should have been tuned to the volatiles of rice. Strikingly important is the response of the antenna to the extracts of *Hyptis*

Table 1. Electroantennogram response of *T. schoenobii* to the extracts of some plants

Name of the treatment	Mean electrical response (in negative volts)	Standard error
Air	0.036 <sup>a</sup>	0.036
Honey	3.129 <sup>de</sup>	0.016
Leaf extract		
<i>Parthenium hysterophorus</i>	0.998 <sup>bc</sup>	0.360
Grass	1.018 <sup>bc</sup>	0.131
<i>Calotropis gigantea</i>	1.343 <sup>c</sup>	0.256
<i>Asclepias curassavica</i>	1.344 <sup>c</sup>	0.534
Mulberry	1.421 <sup>c</sup>	0.263
<i>Panicum setigerum</i>	1.604 <sup>c</sup>	0.246
<i>Cleome viscosa</i>	1.418 <sup>c</sup>	0.340
<i>Hyptis suaveolens</i>	1.675 <sup>c</sup>	0.233
<i>Melia dubia</i>	1.947 <sup>cd</sup>	0.424
<i>Tephrosia purpurea</i>	1.697 <sup>cd</sup>	0.059
<i>Alternanthera ficoidea</i>	1.970 <sup>cd</sup>	0.053
<i>Cassia tora</i>	1.852 <sup>cd</sup>	0.112
<i>Spilanthes acmella</i>	1.811 <sup>cd</sup>	0.306
<i>Asistida</i> sp.	1.667 <sup>cd</sup>	0.137
<i>Malvastrum coromandelianum</i>	2.128 <sup>cde</sup>	0.391
<i>Oryza sativa</i>	3.327 <sup>e</sup>	0.301
Flower extracts		
<i>Calotropis gigantea</i>	1.659 <sup>cd</sup>	0.375
<i>Hyptis suaveolens</i>	2.769 <sup>cde</sup>	0.295

Values with the same alphabets are not significantly different from each other by DMRT.

*suaveolens* flowers. *T. schoenobii* is reported to parasitise other borers such as *Chilo auricilius* Dudgeon, *C. partellus* (Swinhoe), *C. polychrysus* (Meyrick), *C. suppressalis* Walker, *Scirpophaga innotata* (Walker), *S. nivella* (Fabricius) and *Sesamia inferens* (Walker) apart from *S. incertulas* (Khan *et al.*, 1991). However, *H. suaveolens* is not known to be a host plant for any of the hosts of *T. schoenobii*. Floral compounds such as 1,8, cineole,  $\beta$ -caryophyllene with least amounts of  $\alpha$ -copaene,  $\alpha$ -phyllandrene,  $\beta$ -elemene and eugenol were identified earlier from the flowers of *H. suaveolens* (Peerzada, 1997). The EAG response of *T. schoenobii* may be attributed to one and or all the above volatiles, as the role of some these volatiles as an attractant or repellent for several insect species was well established earlier (Elzen *et al.*, 1984; Colazza *et al.*, 2004.; Elfekih and Abderrabba, 2008; FengQin *et al.*, 2008). *T. schoenobii* was also known to respond to the extracts from the anal tuft of female moths (Ding *et al.*, 1981). Mucopolysaccharides from salivary glands and calcium oxalate of oothecae of cockroaches act as kairomones for the closely related species, *T. hagenowii*, a parasitoid on the oothecae of cockroaches (Vinson and Piper, 1986).

This study gives an indication that adults of *T. schoenobii* probably utilize the honeydew of other plants for their survival. However, it is difficult to predict the role of *H. suaveolens* as an attractant or repellent based on EAG studies. Behavioural assays are necessary to confirm the behaviour of *T. schoenobii* to the volatiles of *H. suaveolens*. It is quite feasible that *T. schoenobii* utilizes the florets of rice and flowers of other plant species for their survival and to increase their fitness, which needs to be further studied in the context of using them as ecofeast plants.

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#### REFERENCES

Benrey, B., Denno, R. F. and Kaiser, L. 1991. The influence of plant species on attraction and host acceptance in *Cotesia glomerata* (Hymenoptera: Braconidae). *Journal of Insect Behaviour*, **10**: 619-630.

Bhuiyuan, B. A. and Sufian, M. A. 1985. Biological studies of *Tetrastichus schoenobii* Ferriere (Hymenoptera: Tetra-

stichidae), an egg parasite of yellow rice borer, *Scirpophaga incertulas* Walker. *Bangladesh Journal of Zoology*, **14**: 75-82.

- Colazza, S., McElfresh, J. S. and Millar, J. G. 2004. Identification of volatile synomones induced by *Nezara viridula* feeding and oviposition on bean spp. that attract the egg parasitoid, *Trissolcus basalis*. *Journal of Chemical Ecology*, **30**: 945-964.
- Dale, D. 1994. Insect pests of the rice plant – their biology and ecology, pp. 363-485. In: Heinrichs, E. A. (Ed.) *Biology and management of rice pests*. Wiley Eastern Limited, New Delhi.
- Ding, D. C., Qiu, H. G., Du, J. W., Fu, W. J. and He, L. F. 1981. Studies on the kairomone influencing oviposition behaviour of *Tetrastichus schoenobii* Ferriere: source and extraction. *Acta Entomologica Sinica*, **24**: 262-267.
- Elfekih, S. and Abderrabba, M. A. 2008. Effect of *Mytus communis* essential oil in the Mediterranean fruit fly behaviour. *Biosciences, Biotechnology Research Asia*. **5**: 537-540.
- Elzen, G. W., Williams, H. J. M and Vinson, S. B. 1983. Response by the parasitoid *Campoletis sonorensis* (Hymenoptera: Ichneumonidae) to chemicals (synomones) in plants- Implications for host habitat location. *Environmental Entomology*, **12**: 1873-1877.
- Elzen, G. W., Williams, H. J. and Vinson, S. B. 1984. Isolation and identification of cotton synomones mediating searching behaviour by parasitoid, *Campoletis sonorensis*. *Journal of Chemical Ecology*, **10**: 1251-1264.
- Feng-Qin, C., Waa-Xue, I., Zhong-Nan, F., Fang-Hao, W. and Li-Sheng, C. 2008. Behavioural responses of *Bemisia tabaci* B-biotype to three host plants and their volatiles. *Acta Entomologica Sinica*, **51**: 830-838.
- He, L.F. 1984. Studies on the sensilla of *Tetrastichus* Ferriere (Hymenoptera: Eulophidae). *Contributions from Shanghai Institute of Entomology*, **4**: 71-75.
- Hikim, I. S. 1979. Egg parasites of the yellow stem borer in West Bengal. *International Rice Research Newsletter*, **4**: 19.
- Isalm, Z. 1991. Parasitic efficiencies of two egg parasitoids of the rice yellow stem borer *Scirpophaga incertulas* (Lepidoptera: Pyralidae). *Bangladesh Journal of Entomology*, **1**: 51-57.
- Jervis, M. A., Kidd, N. A. C., Fitton, M. G., Huddleston, T. and Dawah, H. A. 1993. Flower visiting by hymenopterous parasitoids. *Journal of Natural History*, **27**: 67-103.
- Khan, Z. R., Litsinger, J. A., Barrion, A. T., Villanueva, F. F. D., Fernandes M. J. and Taylo, L. D. 1991. *World Bibliography of rice stem borers 1790-1990*. IRRI Los Banos, Philippines and ICIPE, Kenya, Nairobi. 424 pp.

- Kim, H. S. and Heinrichs, E. A. 1985. Parasitization of yellow stem borer (YST) *Scirpophaga incertulas* eggs. *International Rice Research Newsletter*, **10**: 14.
- Kumar, S., Khan, M. A., Kumar, A. and Sharma, K. 2008. Biodiversity of natural enemies in paddy ecosystem and their seasonal dominance. *Annals of Plant Protection Sciences*, **16**: 381–383.
- Nickel, J. L. 1964. *Biological control of Rice Stem Borers: a feasibility study*. Technical Bulletin. 2. International Rice Research Institution, Las Banos, The Philippines.
- Ooi, P. A. C. and Shepard, B. M. 1994. Predators and parasitoids of rice insect pests, pp. 585-612. In: Heinrichs, E. A. (Ed.) *Biology and Management of Rice Pests*. Wiley Eastern Limited, New Delhi.
- Pandya, H. V., Shah, A. H., Patel, C. B., Purohit, M. S. and Rai, A. B. 1995. Study on egg parasitism of rice yellow stem borer in Gujarat. *Gujarat Agricultural University Research Journal*. **21**: 197–199.
- Peerzada, N. 1997. Chemical composition of the essential oil of *Hyptis suaveolens*. *Molecules*, **2**: 165–168.
- Qiu, H. G., Ding, D. C. and Qiu, Z. L. 1984. Observations on the mating behaviour of *Tetrastichus schoenobii* Ferriere. *Contributions from Shanghai Institute of Entomology*, **4**: 77–84.
- Rai, P. S. and Gowda, G. 1977. Parasitization of *Tryporyza incertulas* egg masses in Karnataka, India. *International Rice Commission Newsletter*, **26**: 35–36.
- Ram, S., Patnaik, N. C., Sahoo, S., Mohapatra, A. K. B., Samal, K. C. and Mehta, S. 1996. Seasonal activities of egg parasites of *Scirpophaga incertulas* and extent of parasitization in the Eastern Ghat Highland Zone of Orissa. *Indian Journal of Entomology*, **58**: 387–389.
- Ram, S., Panaik, M. C., Sahoo, S., Mohapatra, A. K. B., Samal, K. C. and Mehta, S. 1997. Seasonal activities of egg parasites of *Scirpophaga incertulas* in the Eastern Ghat Highland Zone of Orissa. *Indian Journal of Entomology*, **59**: 329–331.
- Ressig, W. H., Heinrichs, E. A., Litsinger, J. A., Moody, K., Feiedler, Z., Mew, T. W. and Barrion, A. T. 1986. *Illustrated guide to Integrated Pest Management in Rice in Tropical Asia*. International Rice Research Institute, Los Barlos, Philippines. 411 pp.
- Rothschild, G. N. L. 1970. Parasites of rice stem borers in Sarawak (Malaysian, Borneo). *BioControl*, **15**: 21–25.
- Vinson, S. B. and Piper, G. L. 1986. Source and characterization of host recognition kairomones of *Tetrastichus hagenowii*, a parasitoid of cockroach eggs. *Physiological Entomology*, **11**: 459–468.
- Wackers, F. L. 2004. Assessing the suitability of flowering herbs as parasitoid food sources. Flower attractiveness and nectar accessibility. *Biological Control*, **29**: 307–314.
- Yasumatsu, K. 1967. The possible control of rice stem borers by use of natural enemies, pp. 431-441. In: *Major insect pests of the rice plant*. John Hopkins Press, Baltimore, USA.
- Zaheruddeen, S. M. and Prakasha Rao, P. S. 1983a. Record of some wild species of rice as potential hosts of yellow rice borers. *Science and Culture*, **49**: 207–208.
- Zaheruddeen, S. M. and Prakasha Rao, P. S. 1983b. *Leptochloa panicoides* Wight, an occasional host of the yellow rice borer, *Scirpophaga incertulas* Walker. *International Rice Research Newsletter*, **8**: 17–18.
- Zhang, G. L. and Zhang, G. H. 1985. Observations on the *Tetrastichus* sp. natural enemies of insects. *Kunchong Tiandi*, **7**: 201–203.