VOLUME 9 ISSUE 1 2023

ISSN 2454 – 3055

INTERNATIONAL JOURNAL OF ZOOLOGICAL INVESTIGATIONS

Forum for Biological and Environmental Sciences

Published by Saran Publications, India

International Journal of Zoological Investigations Vol. 9, No. 1, 148-153 (2023)

International Journal of Zoological Investigations
Contents available at Journals Home Page: www.ijzi.net
Editor-in-Chief: Prof. Ajai Kumar Srivastav
Published by: Saran Publications, Gorakhpur, India

ISSN: 2454-3055

Electroantennogram Responses of *Chrysoperla carnea* Stephens Towards Seven Saturated Hydrocarbons

Singh Surabhi[1], Kapinder[2] , Paul Biswajeet[3] and Kumar Archna[1]*

[1]Biological Control Laboratory, Amity Institute of Biotechnology, Amity University, NOIDA 201313, Uttar Pradesh, India
[2]Department of Zoology, University of Allahabad, Prayagraj 211002, Uttar Pradesh, India
[3]Division of Entomology ICAR-IARI, New Delhi-110012

**Corresponding Author*

Received: 16th December, 2022; **Accepted:** 7th January, 2023; **Published online:** 24th January, 2023

https://doi.org/10.33745/ijzi.2023.v09i01.016

Abstract: Volatile cues, especially straight-chain hydrocarbons, play a vital role in governing the foraging pattern of natural enemies. Therefore, the impacts of various concentrations of seven crude hydrocarbons on the foraging potential of *Chrysoperla carnea* Stephens male and female population through Electroantennogram (EAG) were investigated. The male and female *C. carnea* showed its characteristics EAGs responses to seven straight-chain saturated hydrocarbons viz., Hexadecane ($C_{16}H_{34}$), Heptadecane ($C_{17}H_{36}$), Octadecane ($C_{18}H_{38}$), Nonadecane ($C_{19}H_{40}$), Tricosane ($C_{23}H_{48}$), Tetracosane ($C_{24}H_{50}$) and Pentacosane ($C_{25}H_{52}$)**.** The EAG responses of female antennae were higher as compared to male antennae for all tested compounds. An Electroantennogram study of *C. carnea* provided evidence that the antennal receptors were differentially sensitive to these compounds. These observations suggested that straight-chain saturated hydrocarbons can be utilized to augment the foraging behavior potential of natural enemies in Integrated Pest Management (IPM) programs.

Keywords: *Solanum tuberosum*, Integrated pest management, Biological control, *Chrysoperla carnea,* Electroantennogram, Saturated hydrocarbons, Plant volatile components

Citation: Singh Surabhi, Kapinder, Paul Biswajeet and Kumar Archna: Electroantennogram responses of *Chrysoperla carnea* Stephens towards seven saturated hydrocarbons. Intern. J. Zool. Invest. 9(1): 148-153, 2023.

https://doi.org/10.33745/ijzi.2023.v09i01.016

Introduction

Solanum tuberosum L. (Potato) is one of the crucial commercial solanaceous food crops (Scott and Suarez, 2011). It gets heavily infested by lepidopterous pests which eventually hamper the productivity of this crop (Malakar and Tingey, 2006). *Phthorimaea operculella* Zeller (Potato Tuber Moth) is a key pest of potato tubers in storage and in field. The economic damage caused by *P. operculella* is quite significant. In India, the major portion of damage is reported from Uttar Pradesh, Uttaranchal, Maharashtra, Bihar and Punjab. To overcome from pest infestation problem, chemical pesticides are used as one of the measures of control. But through this, satisfactory control could not be achieved due to the boring habit of *P. operculella* larvae. A better level of success had been achieved by the use of natural enemies. *Chrysoperla carnea* Stephens is one of the major predators against potato tuber moth and other sucking pests. In nature, crop ecosystem are influenced by blend of chemical cues in form of potential signaling molecules; which are responsible for governing entire tri-trophic interaction mechanism (Archna *et al*., 2009; Fatouros *et al.,* 2010; Zayeem and Kumar, 2012). Natural enemies' exhibit varied foraging behaviour in response to sensitization by these signaling molecules (Kumar *et al*., 2012). These signaling molecules consist of different chemical compounds and majority of these are hydrocarbons in nature (Kumar *et al*., 2018a). Thus, use of attractant or repellent volatile compounds offers a promising approach and sustainable control measure.

Thus, in the present study impact of three concentrations *(*1000 mg/l, 100 mg/l and 10 mg/l) of seven straight-chain saturated hydrocarbons viz., Hexadecane ($C_{16}H_{34}$), Heptadecane ($C_{17}H_{36}$), Octadecane ($C_{18}H_{38}$), Nonadecane ($C_{19}H_{40}$), Tricosane ($C_{23}H_{48}$), Tetracosane ($C_{24}H_{50}$) and Pentacosane ($C_{25}H_{52}$) towards olfactory receptors of male and female *C. Carnea* was analysed using Electroantennogram (EAG)

Materials and Methods

Rearing of the predator C. carnea:

The nucleus culture of predator *C. carnea* (NBAII-MP-CHR-01) was obtained from the National Bureau of Agricultural Insect Resources (NBAIR), Bengaluru, Karnataka, India. For further multiplication of culture, UV-treated *Corcyra cephalonica* Stainton eggs were provided to larvae till their pupation. The cocoons were separated and kept for emergence in a new glass jar. Larvae were reared on the sterilized eggs of *C. cephalonica* under similar conditions. The freshly emerged adults were used for EAG studies.

Preparation of saturated hydrocarbons concentrations:

Seven straight-chain saturated hydrocarbons viz., Hexadecane ($C_{16}H_{34}$), Heptadecane ($C_{17}H_{36}$), Octadecane ($C_{18}H_{38}$), Nonadecane ($C_{19}H_{40}$), Tricosane ($C_{23}H_{48}$), Tetracosane ($C_{24}H_{50}$) and Pentacosane ($C_{25}H_{52}$) were selected based on previous data and procured from Sigma-Aldrich for the preparation of samples. Three different concentrations viz., 1000 mg/l; 100 mg/l and 10 mg/l were prepared for each selected hydrocarbon by adding an appropriate quantity of distilled HPLC grade hexane.

Electroantennography:

To know the exact response of signaling molecules on behaviour of natural enemies Electroantennography (EAG) study was conducted. In this, EAG response of *C. carnea* (from both male and female population) were recorded towards three concentrations (1000 mg/l, 100 mg/l and 10 mg/l) of seven pure hydrocarbons. For this, EAG machine (M/s Syntech, Germany) was used. The antennae of predator were cut from the selected adult of *C. carnea* by using micro-scissors. The distal segment of antennae was clipped in 0.1 M electrolyte solution which was helpful in smooth electrical conduction between two electrodes. After this, the basal part of antenna was connected to different electrodes and the tip of antenna was fixed with recording electrode by applying electrical conductivity gel (Parker, Spectra 360). The ideal electrical conductivity of antennae between electrodes was standardized by stabilizing the base line having minimum fluctuations. Response of insect antennae towards various targeted chemical cues was recorded. Air flow maintained over antennal preparation throughout the experimental time was 500 ml/min. Hexane was utilized to normalize the test stimulation. Each recording session was started on application of control stimulus followed by 10 mg/ml doses of different test stimulus. Data recording was performed and analysed by using software, EAG

2000 (version 2.7c, Syntech, Germany) (Seenivasagan *et al*., 2011; Kumar *et al*., 2018a).

Statistical Analysis:

To evaluate comparative antennal sensitivity of male and female *C. carnea* towards different hydrocarbon concentrations tested, the data were tabulated and subjected to Two-way ANOVA (Analysis of Variance) (Kumar *et al*., 2018a).

Results

On the basis of interaction study of different concentrations of seven crude saturated hydrocarbons and *C. carnea*, selected hydrocarbons viz., Hexadecane, Heptadecane, Octadecane, Nonadecane, Tricosane, Tetracosane and Pentacosane were opted for EAG studies. In both sexes of *C. carnea*, antennae responded significantly to most of the tested compounds. In general, the EAG response of female antennae was higher as compared to male antennae in all tested compounds. In female, the EAG responses varied from 0.20±0.01 (Nonadecane at 1000 mg/l) to 2.35± 0.06 (Tricosane at 100 mg/l) (Table 1) and in males from 0.39±0.05 (Noadecane at 10 mg/l) to 2.68±0.09 (Hexadecane at 100 mg/l) (Table 2). The largest peak amplitudes in female antennae were obtained from Tricosane (2.34 mV) followed by Pentacosane (2.16 mV) and Heptadecane (2.14 mV). Similar to female antennae, male antennae also evoked higher response to Hexadecane (2.68 mV) and Octadecane (2.04 mV). In female antennae, Nonadecane at 10 mg/L evoked non-significant response while Heptadecane and Nonadecane at 1000 mg/l and Pentacosane at 10 mg/l generated non-significant response in males (Figs. 1, 2). Statistically, significant differences in EAG response between both sexes were observed from Hexadecane Octadecane, Tricosane and Tetracosane.

Discussion

EAG study revealed that the largest peak amplitudes in female antennae of *C. carnea* were obtained from Tricosane and male antennae of *C. carnea* evoked a response in Hexadecane. The study is discussed in support of the following studies. Bakthavatsalam *et al*. (2000) reported that adults of *C. carnea* showed good electroantennogram response to hexane extracts of flowers and bolls of Cotton infested by *Helicoverpa armigera.* Verheggen *et al*. (2008) advocated that plant green leaf volatiles enhanced the location selection of the oviposition site by predator *Episyrphus balteatus*. They also notified that the highest antennal responses for *E. balteatus* were elicited by six and nine carbon green leaf alcohols and aldehydes [i.e., (Z)-3-hexenol, (E)-2-hexenol, (E)-2-hexenal, and hexanal]. Hanumantharaya *et al*. (2010) reported that a higher electroantennogram response was recorded in mated females than mated males of *C. carnea* towards kairomone substance of Cotton leaf and boll extract. Seenivasagan and Paul (2011) reported that in EAG studies also, saturated hydrocarbons elicited differential EAG responses in the antennal response of *C. plutellae* females. Tricosane and Hexacosane elicited increased EAG response compared to control stimulus. Long-chain hydrocarbons C27, C28 and C29 also elicited a good response. The sensitivity of the antenna was 4-5 folds for C25, C14, C24, C15 and C30, while the short-chain hydrocarbons elicited 2-3 fold increased EAG responses. Kumar *et al*. (2012) reported that *Trichogramma chilonis* exhibit enhanced parasitism for Pentacosane whereas *T. brasiliensis* exhibit favorable responses towards Tricosane and Pentacosane. The largest peak amplitude by antennae of male and female, *Dysdercus cingulatus* during EAG responses were recorded by Heneicosane, Hentriacontane, Tricosane and Hexatricontane. These hydrocarbons were found to evoke significantly different antennal responses in *Dysdercus cingulatus* (Kumar *et al*., 2018b). Trang and Dey (2010) advocated that the maximum amplitude of EAG response for both male and female *Apanteles angaleti* was recorded for the extract obtained from the damaged bolls followed by extracts from leaves undamaged buds, damaged buds and undamaged bolls of Cotton variety LD 327, respectively. Electroantennograms (EAGs)

Table 1: Impact of saturated hydrocarbons on *C. carnea* females through electroantennography

Concentration→ Hydrocarbons↓	**1000 mg/l**			**100 mg/l**			**10 mg/l**		
	Source	**Control**	**Difference**	**Source**	**Control**	**Difference**	**Source**	**Control**	**Difference**
H1	0.33* ±0.01	0.01	0.32	0.37* ±0.02	0.07	0.30	2.09* ±0.04	0.02	2.07
H2	0.28* ±0.01	0.15	0.13	2.04* ±0.09	0.02	2.02	2.17* ±0.19	0.03	2.14
H3	0.32* ±0.01	0.01	0.31	2.08* ±0.20	0.00	2.08	0.60* ±0.01	0.03	0.57
H4	0.20* ±0.01	0.09	0.11	2.23* ±0.09	0.06	1.17	2.10* ±0.06	0.02	2.08
H5	0.27* ±0.02	0.00	0.27	2.35* ±0.06	0.01	2.34	1.92* ±0.04	0.01	1.91
H6	1.95* ±0.02	0.01	1.94	1.77* ±0.03	0.01	1.76	1.95* ±0.07	0.13	1.82
H7	0.95* ±0.05	0.01	0.94	2.16* ±0.05	0.00	2.16	0.29 ±0.04	0.07	0.22
Mean	0.61* ±0.13	0.04	0.57	1.89 ±0.07	0.02	1.87	1.58 ±0.05	0.04	1.54
Source of variations		Calculated F-ratio	p-value		probability at 5%	Standard error Mean	Standard error deviation		CD at 5%
Hydrocarbons (A)		8.27	0.004**		1.97	0.14	0.19		0.39
Concentration (B)		1.56	0.1615						
A*B		21.54	0.0001***						

H1: Hexadecane, H2: Heptadecane, H3: Octadecane, H4: Nonadecane, H5: Tricosane, H6: Tetracosane, H7: Pentacosane;
Values after ± indicates Standard Error; Values of source is mean of ten trials, Value for control is for one trial

Table 2: Impact of saturated hydrocarbons on *C. carnea* males through Electroantennography

Concentration → Hydrocarbons↓	**1000 mg/l**			**100 mg/l**			**10 mg/l**		
	Source	**Control**	**Difference**	**Source**	**Control**	**Difference**	**Source**	**Control**	**Difference**
H1	1.17* ±0.06	0.01	1.16	2.68* ±0.09	0.00	2.68	0.94* ±0.03	0.00	0.94
H2	1.82* ±0.04	0.84	0.98	1.21* ±0.04	0.09	1.12	0.73* ±0.01	0.03	0.70
H3	1.67* ±0.10	0.02	1.65	2.06* ±0.06	0.02	2.04	0.69* ±0.01	0.04	0.65
H4	1.64* ±0.11	0.03	1.61	1.53* ±0.04	0.13	1.4	0.39* ±0.05	0.19	0.20
H5	1.42* ±0.07	0.03	1.39	1.95* ±0.07	0.14	1.81	1.13* ±0.02	0.02	1.11
H6	0.88* ±0.09	0.17 ±0.17	0.71	1.58* ±0.03	0.00	1.58	0.60* ±0.01	0.03	0.57
H7	1.73* ±0.01	0.17	1.56	1.61* ±0.02	0.01	1.60	0.53* ±0.02	0.00	0.53
Mean	1.47 ±0.06	1.27	0.20	1.80 ±0.05	0.05	1.75	0.71 ±0.15	0.04	0.67
Source of variations		Calculated F- ratio	p-value		probability at 5%	Standard error Mean	Standard error deviation		CD at 5%
Hydrocarbons (A)		11.84	0.0000		1.97	0.16	0.24		0.47
Concentration (B)		6.46	0.0000						
A*B		29.63	0.0000						

H1: Hexadecane, H2: Heptadecane, H3: Octadecane, H4: Nonadecane, H5: Tricosane, H6: Tetracosane, H7: Pentacosane;
Values after ± indicates Standard Error; Values of source is mean of ten trials, Value for control is for one trial

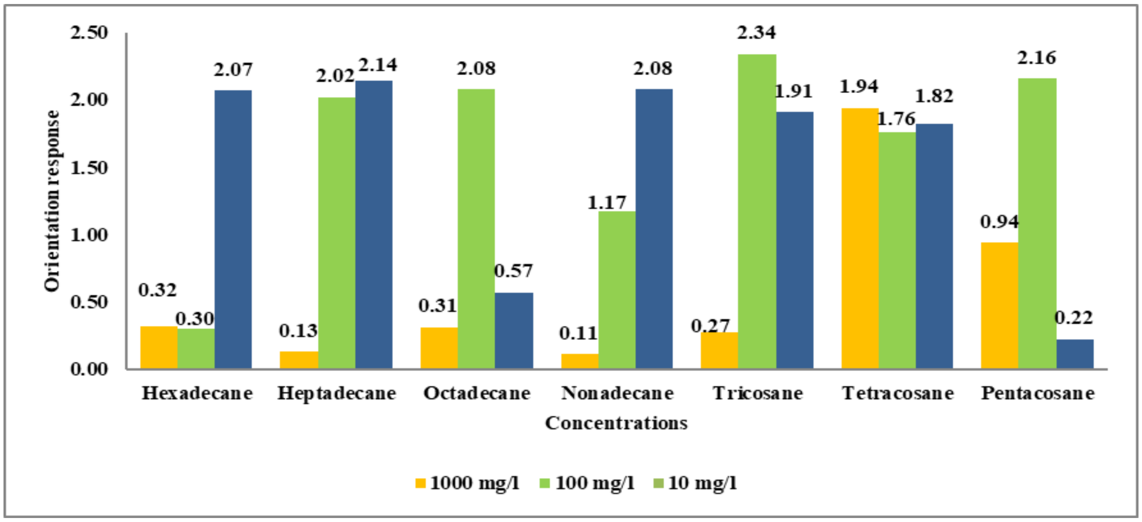

Fig. 1: Validation of stimulation activity of *C. carnea* females by saturated hydrocarbons through EAG.

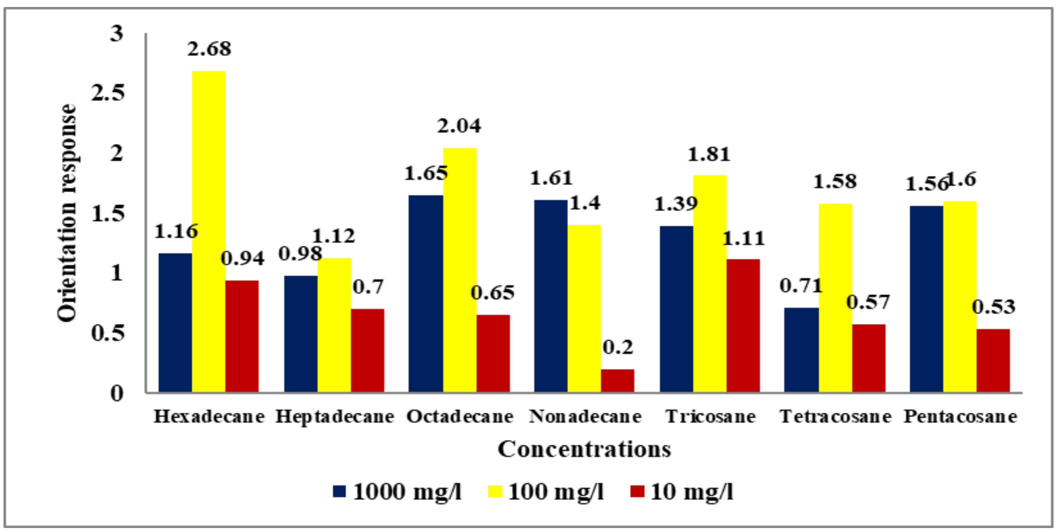

Fig. 2: Validation of stimulation activity of *C. carnea* males by saturated hydrocarbons through EAG.

response from unmated, laboratory-reared male and female *Helicoverpa armigera* adults was recorded by Chen *et al*. (2008) towards a range of plant volatile components. The greatest EAG responses of all plant volatiles tested were elicited by monoenic C-6 alcohol and aldehyde. They are constituents of the "general green-leaf odor" that emanates from most plants. Pinho *et al*. (2009) reported 88 volatile and semi-volatile components from flowers, leaves and stems of *Catharanthus roseus* L. Trang and Dey (2013) reported that large EAG responses by braconoid wasp *Chelonus blackburni* on volatile plant compounds of Cotton variety Pusa 8-6 was particularly found among green leaf extract followed by *Earias vitella* damaged bud, damaged boll, undamaged bud and undamaged boll, respectively. The findings of the present study which reflected the interaction of compounds of different chemical natures in varied numbers and concentrations is in accordance with previous research work.

Acknowledgements

Authors are thankful to National Bureau of Agricultural Insect Resources (NBAIR), Bangalore for providing the culture of natural enemies.

References

Archna, Singh AK, Paul AVN and Jain A. (2009) Synomonal effect of nine varieties and one culture ofRice on *Trichogramma japonicum* Ashmead and *Trichogramma chilonis* (Ishii) (Hymenoptera: Trichogrammatidae). Acta Entomol Sinica 52: 656-664.

Bakthavatsalam N, Singh SP, Tandon PL, Chaudhary M and Preethi S. (2000) Synomone mediated behavioural responses of *Chrysoperla carnea* (Stephens)(Neuroptera: Chrysopidae) to Cotton infested by *Helicoverpa armigera* (Hübner) (Lepidoptera: Noctuidae) J Biol Cont. 14(2): 1-6.

Chen X, Hou Z, Zhang Y, Yan F and Zhang G. (2008) Olfactory responses of Cotton bollworm, *Helicoverpa armigera* to sex pheromone and plant volatile components. Insect Sci. 4(2): 159-172.

Hanumantharaya L, Basavana GK, Krishna NL and Kulkarni KA. (2010) Electroantennogram responses of *Chrysoperla carnea* (Stephens) and *Helicoverpa armigera* (Hubner) to volatiles of different cultivars of Cotton. Karnataka J Agric Sci. 23: 123-126.

Huigens ME, Woelke JB, Pashalidou FG, Bukovinszky T, Smid HM and Fatouros NE. (2010) Chemical espionage on species-specific butterfly anti-aphrodisiacs by hitchhiking *Trichogramma* wasps. Behavioral Ecol. 21(3): 470-478.

Kumar A, Zayeem A and Kanameni S. (2012) Synomonal effect of cole crops on individual and associative learning behavior of *Cotesia plutellae*. Int J Biol Pharm Allied Sci. 1: 285-298.

Kumar A, Kanameni S, Arora P and Zaeem A. (2018a) Formulation of crucifer plant volatiles and assessment of their impact on parasitic efficiency of *Trichogramma chilonis*. Plant Arch. 18(2): 2077-2082.

Kumar J, Paul B and Nebapure SM. (2018b) Electrophysiological response of *Dysdercus cingulatus* (Fabricius) (Hemiptera: Pyrrhocoridae) to cotton plant extracts and other host related compounds. Proc Nat Acad Sci, India Sec B: Biol Sci. 89(3): 793-800.

Malakar KR and Tingey WM. (2006) Aspects of tuber resistance in hybrid potatoes to potato tuber worm. Entomologia Experimen Applicata 120(2): 131-137.

Mathur S, Zayeem A, Kanameni S, Tibrewal M, Wadhwa N, Arora P and Kumar A. (2012) Effect of various concentration of Octacosane, Pentacosane and Tricosane on Foraging Behavior of Trichogrammatids. Int J Scient Res Publications 2(6): 1-4.

Pinho PG, Gonçalves RF, Valentão P, Pereira DM, Seabra RM, Andrade PB and Sottomayor M. (2009) Volatile composition of *Catharanthus roseus* (L.) G. Don using solid-phase microextraction and gas chromatography/mass spectrometry. J Pharma Biomed Analysis 49(3): 674-685.

Scott GJ and Suarez V. (2011) Growth rates for potato in India and their implications for industry. Potato J. 38: 100-112.

Seenivasagan T and Paul AVN. (2011) Gas-chromatography and Electroantennogram analysis of saturated hydrocarbons of cruciferous host plants and host larval body extracts of *Plutella xylostella* for behavioural manipulation of *Cotesia plutellae*. Indian J Exp Biol. 49: 375-386.

Singh S, Paul B and Kumar A. (2019) Impact of volatile cues on foraging response of egg larval parasitoid, *Chelonus blackburni* Cameron. Indian J Agric Res. 53: 196-201.

Trang TTK and Dey D. (2010) Electroantennogram responses of *Apanteles angaleti* Muesebeck, a parasitoid of *Pectinophora gossypiella* to synomonal compounds of Cotton. Omon Rice 17: 71-79.

Trang TTK and Dey D. (2013) Electroantennogram responses of *Chelonus blackburni* Cameron, egg-larval parasitoid of spotted boll worm *Earias vitella* to infochemicals on Cotton ecosystem. Omon Rice 19: 118-130.

Verheggen FJ, Arnaud L, Bartram S, Gohy M and Haubruge E. (2008) Aphid and plant volatiles induce oviposition in an Aphidophagous Hoverfly. J Chem Ecol. 34: 301-307.

Zayeem A and Kumar A. (2012) Impact of Synomones emanating from three cole crop varieties on foraging behaviour of *Trichogramma brasiliensis* Ashmead (Hymenoptera: Trichogrammatidae). Ann Plant Prot Sci. 20: 481-482.