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Role of Fenoxycarb, a Juvenile Hormone Analogue, on the Developmental Stages of Rice-Moth, *Corcyra cephalonica* Staint. (Lepidoptera: Pyralidae)

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Abstract: The fourth instar larvae of *Corcyra cephalonica* were exposed to seven concentrations of fenoxycarb i.e. 0.001, 0.005, 0.01, 0.05, 0.10, 0.50 and 1.00 ppm and its insecticidal activities were evaluated. The higher concentrations of this compound severely disrupted the metamorphosis of *C. cephalonica*. The significant differences in larval mortality, pupation, pupal mortality and adult emergence in comparison to their control were observed. At 0.05 and 1.00 ppm concentrations of fenoxycarb there was 100% suppression of adult emergence. Thus, fenoxycarb at these higher concentrations behaves as insecticide that severely hampers the normal growth, development and metamorphosis of *C. cephalonica.* So, this juvenile hormone analogue may be used for successful control of this pest in particular and lepidopterous pests in general.

Keywords: *Corcyra cephalonica*, Fenoxycarb, LD_{50}, metamorphosis

Introduction

Post-harvest losses in India amount to 12-16 million metric tons of food grains each year, an amount that the World Bank estimates could feed one-third of India's poor population (Nagpal and Kumar, 2012) . The post-harvest losses of food grains have been estimated to the extent of about 10% of the total production of which losses during storage alone are estimated to be 6.5%. The insects alone damage 2.5% of the food grains in storage. Seventy five per cent of total grain loss occurs at farm level. Majority of such losses occur by inadequate storage (22%), drying of crops (15%), transportation (12%) and threshing (10%) while rest of the losses occur by scattering among several other activities (Grain Depot Fund Prospectus, 2011).

Storage loss of food grains at the level of Government and its agencies such as Food Corporation of India, Central and State Warehousing Corporations and State Civil Supplies Departments/ Corporations have been reduced to the minimum. However, 60-70% of the total production are retained by the farmers for their own food, cattle feed, seed etc. and they generally store their grains in traditional storage structures where maximum loss occur that require intensive care of pest management.

Control of insect pests is a puzzling problem since many decades. *Corcyra cephalonica* Stainton, commonly known as rice moth, is a severe pest of stored cereals and cereal products in Asia, Africa, Europe, North America and other tropical and subtropical regions of the world. This moth was first identified and reported by Stainton (1866), who named it *Melissoblaptes cephalonica.* Later, Ragonot (1885) gave it the generic name *Corcyra.* The only recognized species of this genus is *cephalonica.* Ayyar (1919) made the first record of *Corcyra cephalonica.* This moth is believed to be of eastern origin but has become a cosmopolitan species. Its larval stages cause serious damage to rice, gram, sorghum, maize, groundnut, cotton seeds, peanuts, linseeds, raisins, nutmeg, currants, chocolates, army biscuits and milled products (Ayyar, 1919, 1934; Munro and Thompson, 1929; Richards and Herford, 1930; Noyes, 1930; Atwal, 1976 and Piltz, 1977).

Modern insecticide research started almost 65 years ago with the chlorinated hydrocarbons, followed shortly by the organophosphates, methylcarbamates, botanicals and synthetic pyrethroids (Mojaver and Bandani, 2010). The use of these conventional organic insecticides to control insect pests has given rise to problems of the proliferation of resistance and accumulation of residue in the environment with adverse ecological effects (Hoffmann and Lorenz, 1997). In the search for safer insecticide technologies, i.e. more selective mode of action and reduced risks for non-target organisms and the environment, progress has been made in the last two decades with the development of natural and synthetic compounds capable of interfering with the processes of growth, development and metamorphosis of the target insects (Smet *et al*., 1989, 1991; Oberlander *et al*., 1997).

Usually, the control measures in stores are based on fumigation with chemicals like hydrogen phosphate. Residues and insect resistance are reasons for potentially limiting the use of fumigation with chemicals in the near future (World Metrological Organisation, 1994). In such condition, there is a need for new alternatives to traditional insecticides used in stored product pest management (Arthur, 1996; Arthur and Phillips, 2003). In this regard, the insect growth regulators (IGRs) (Fox, 1990), which mimic insect's hormone and regulate the insect population through the disruption of moulting and metamorphosis (Williams, 1956; Oberlander *et al*., 1997) have captured the interest of stored product entomologists. The first use of IGRs against stored product pests was reported by Thomas and Bhatnagar-Thomas (1968). The term IGR was designed by Staal (1975) to describe a class of bio-rational compounds. Through selectivity of action, these compounds appear to fit the requirements for "Third Generation Pesticides" (Williams, 1967) that disrupt the normal development of several species of insects (Henrick *et al*., 1973). They are highly effective against various insects attacking stored products and other pests that have become resistant to organic insecticides. Meanwhile, all these compounds are less toxic to mammals and non-target organisms because of their non-toxic effect and their quick disintegrating abilities (Carter, 1975; Staal, 1975; Zurflueh, 1976; Oberlander *et al*., 1978, 1997; Ishaaya *et al*., 1987; Ishaaya and Horowtz, 1998; Kostyukovsky *et al*., 2000; Parthasarathy *et al*., 2012).

IGRs generally control insects either through regulation of metamorphosis or interference with reproduction (Riddiford and Truman, 1978).: The search to apply knowledge of JHs (juvenile hormone) to the development of effective insecticides has since been limited to the area of JH

analogues (Matolcsy *et al*., 1988). Compounds that mimic the action of natural juvenile hormones are called as juvenile hormone analogues (JHAs) or said to be active mimics of JHs (Matolcsy *et al*., 1988). JHAs can function as agonists or antagonists or a mixture of both with natural JHs (Kramer and Stall, 1981). They interfere with important biochemical mechanisms such as the secretion and transportation of natural JHs from the secretory site to the target site, degradation, excretion and feedback control (Retnakaran *et al*., 1985). They act at the genetic level and are associated with transcription of mRNA (Coudron *et al*., 1981). Hence, their biological effects are very complex, and vary from one analogue to another.

One of the JHA, methoprene was found to be effective against the ontogeny of stored product pests like red flour beetle, *Tribolium castaneum* (Parthasarathy and Palli, 2009; Wijayaratne *et al*., 2012); *C. cephalonica* (Deb and Chakravorty, 1985; Tripathi and Tiwari, 2014; Tripathi, 2014; Singh and Tiwari, 2014, 2014a, 2014b; 2015) and almond moth, *E. cautella* (Chandra and Tiwari, 2013). Pyriproxyfen also affected the developmental stages of *O. surinamensis*, *T. castaneum* and cigarettle beetle, *Lasioderma serricorne* (Arthur *et al*., 2009); *P. interpunctella* (Arthur *et al*., 2009; Ghasemi *et al*.,2010); sun pest, *Eurygaster integriceps* (Mojaver and Bandani, 2010) and *T. confusum* (Loni *et al*., 2011). Similarly, hydroprene also influenced the developmental stages of *C. cephalonica* (Deb and Chakravorty, 1982; Bhargava and Devaraj Urs, 1992, 1993); *T. casteneum* (Arthur *et al*., 2009; Parthsarthy and Palli, 2009); *O. surinamensis* and *P. interpunctella* (Arthur *et al*., 2009);

Although, sufficient knowledge exist on the effect of certain insecticidal agents (organochlorines, organophosphates, synthetic pyrethroids and few natural plant products) influencing ontogeny as well as haemolymph, fat body and gonadial biochemistry of *C. cephalonica* (Tiwari, 1987; Tiwari and Bhatt, 1987, 1988, 1989, 1992, 1994a, 1994b, 1994c, 1994d, 1994e, 1996, 1999a,1999b, 2000a, 2000b; Tiwari and Tripathi, 2001, 2003, 2005, 2006; Pathak and Tiwari, 2010a, 2010b; Shukla and Tiwari, 2011a, 2011b, 2011c; Pathak and Tiwari, 2012; Shukla and Tiwari, 2011a, 2011b, 2011c) and a few IGRs influencing ontogeny, reproductive potential and egg and gonadial biochemistry of this pest (Tripathi and Tiwari, 2013, 2014, Tripathi 2014; Tripathi and Tiwari, 2015; Singh and Tiwari, 2014a, 2014b; Singh, 2014; Singh and Tiwari, 2015) but scientific contribution of fenoxycarb influencing developemental stages of rice moth, *C. cephalonica* is still wanting. The acquisition of such knowledge in this area becomes essential for a comprehensive appreciation of the physiological and ecological relationship that exists between this pest and its host material (stored cereals and cereal commodities). This knowledge in turn, is likely to generate new insights into divising ways and means for controlling *C. cephalonica,* by disrupting its moulting and metamorphosis so that evolution of a new generation of this pest for the eventual establishment on stored cereals and cereal products can be considerably restricted. Hence, as an objective of such programme the present work for the first time, has been designed and conducted to examine into the impact of a juvenile hormone analogue (JHA) i.e. fenoxycarb on the ontogeny of rice moth, *C. cephalonica*.

Materials and Methods

Corcyra cephalonica Stainton adults were obtained from laboratory stock culture maintained on normal dietary medium composed of coarsely ground jowar (*Sorghum vulgare*) mixed with 5% (w/w) powdered yeast inside large glass containers (150 mm diameter, 200 mm height) at temperature 26 ± 1^0C, relative humidity

(R.H.) 93 ± 5% and a light regime of 12 h light and 12 h dark. Such a standard culture was maintained throughout the year.

From the above culture whenever needed, newly emerged males and females were transferred to oviposition glass chambers (35 mm diameter, 200 mm height). Since *C. cephalonica* individuals do not feed during their adult stage, no food was provided to them during their confinement in these vessels. Eggs laid by the females were collected and then placed in glass chambers (consisting of 250 ml beakers) with the help of zero number camel hair brush for hatching.

Fenoxycarb ethyl[2-(4-phenoxy-phenoxy)-ethyl]carbamate, molecular formula- $C_{17}H_{19}NO_4$, a non terpenoid juvenile hormone analogue, P-686N, Lot-20071 used in the experiment, was obtained from AccuStandard, New Haven, CT 06513, USA.

Different concentrations of fenoxycarb, in dietary media, were preapared. For this purpose, a stock solution of known concentration of JHA was prepared by dissolving it in acetone and then adjusted via serial dilutions to achieve its required concentrations. Then, required volume of different concentrations of fenoxycarb was thoroughly mixed with the required quantity of normal food (roughly ground jowar mixed with 5% w/w yeast powder) to get different desired concentrations i.e. 0.001, 0.005, 0.01, 0.05, 0.10, 0.50 and 1.00 ppm of fenoxycarb in dietary media. This treated food was then air dried at room temperature to eliminate completely the acetone. For control purposes, the normal food was thoroughly mixed with a required volume of acetone similar to that of treated food and then air dried in the same way.

To evaluate the toxicity of fenoxycarb, when exposed to fourth instar larvae, on the developemental stages of *C. cephalonica,* freshly hatched larvae were allowed to feed on normal dietary medium (kept inside 250 ml beakers) for exactly 20 days. On 21st day, 25 fourth instar larvae were transferred to each similar rearing chambers (250 ml beakers) containing 50 g of dietary medium mixed and treated separately with seven different concentrations i.e. 0.001, 0.005, 0.01, 0.05, 0.10, 0.50 and 1.00 ppm of fenoxycarb. 25 larvae were also kept on normal food treated with acetone alone, serving as control. Experiments were kept at the temperature, relative humidity and photophase, as mentioned earlier.

On the completion of life-cycle, per cent adult emergence and pupal mortality were observed and on that basis per cent pupation and per cent larval mortality were calculated. The developmental course and external morphology of larvae, pupae and adults were also observed. Adult mortality was also noted up to 24 h of emergence. The corrected total mortality was calculated by Abbott's formula (1925), as:

$$\text{Corrected total mortality} = \frac{100 \times \%\text{ experimental mortality} - \%\text{ control mortality}}{100 - \%\text{ control mortality}}$$

After completion of life-cycle, the remaining food was weighed to calculate the amount of fenoxycarb consumed per larva (µg/larva) at each concentration of fenoxycarb.

Experiments were replicated six times and values have been expressed as mean ± SEM. Student's t-test was applied to determine the significant differences between the treated groups and their control (Finney,1952). LD_{50} values (µg/larva), 95% confidence limits (lower and upper confidence limits) of LD_{50}, slope values and heterogeneity of fenoxycarb were calculated by Polo Plus, Probit and Logit Analysis, Version: 2.0, LeOra Software based on probit analysis (Finney, 1959).

LD_{50} value (Table 1) was calculated against the insecticide consumed by larvae (µg/larva) and not against the ppm concentration of fenoxycarb.

Results

Results presented in table 2 reveal that a significant larval mortality was obtained with the increase of fenoxycarb concentration in the diet. In case of control larvae, larval mortality was recorded to be 1.33 ± 0.84% but at 0.001 ppm concentration of fenoxycarb larval mortality was observed to be 2.67 ± 0.84% which increased to 85.00 ± 1.71% at 1.00 ppm concentration of fenoxycarb. As the fenoxycarb concentration increases a significant reduction in pupation and a significant enhancement in pupal mortality did occur. In case of control larval groups, 98.67 ± 0.84% pupation was observed that decreased to 97.33 ± 0.84% at 0.001 ppm concentration of fenoxycarb but this value was reduced to 15.00 ± 1.71% at 1.00 ppm concentration of fenoxycarb. Under same conditions, per cent pupal mortality was found to be 2.03 ± 0.89% in control larvae, which was increased to 6.84 ± 0.92% at 0.001 ppm and 100% at 0.50 and 1.00 ppm concentrations of fenoxycarb. It is noticeable that at 0.50 and 1.00 ppm concentrations of fenoxycarb pupation took place but all the pupae get perished and hence none of the adults emerged at these two concentrations of fenoxycarb. A significant reduction in adult emergence was recorded following increased concentrations of JHA. In control larval groups, 96.67 ± 0.67% adult emergence was recorded that decreased to 90.67 ± 1.69% and 40.00 ± 2.07% at 0.001 and 0.10 ppm concentrations of fenoxycarb, respectively.

Table 1: LD_{10}, LD_{50} and LD_{90} values, Confidence limits (LCL and UCL) of LD_{10}, LD_{50} and LD_{90}, Slope Values and Heterogeneity of fenoxycarb to the IV instar larvae of rice moth, *C. cephalonica*

Insecticide	Instar	Effective doses (µg/ larva)		95% Confidence limits of LD_{50}		Slope Values	Heterog-eniety
				LCL	UCL		
Fenoxycarb	IV	LD_{10} LD_{50} LD_{90}	0.009 0.068 0.518	0.006 0.012 0.343	0.054 0.088 0.895	1.455 ± 0.089	1.67

LCL = Lower Confidence Limit; UCL = Upper Confidence Limit

Table 2: Effect of fenoxycarb on the ontogeny of rice moth, *C. cephalonica* exposed as fourth instar larvae

Fenoxycarb concentra-tion (ppm)	Fenoxycarb consumed* (µg/larva)	% Larval mortality#	% Pupation#	% Pupal mortality#	% Adult emergence#	% Adult mortality#	% Total mortality#	Corrected total mortality**
Control	0.0000	1.33 ± 0.84	98.67 ± 0.84	2.03 ± 0.89	96.67 ± 0.67	-	3.33 ± 0.67	-
0.001	0.0001	2.67 ± 0.84	97.33 ± 0.84	6.84 ± 0.92[b]	90.67 ± 1.69[b]	-	8.67 ± 1.23	5.52 ± 1.35
0.005	0.0804	4.67 ± 1.23[c]	95.33 ± 1.23[c]	8.39 ± 1.29[b]	87.33 ± 1.90[b]	-	12.00 ± 1.79	8.97 ± 1.85
0.01	0.0181	17.33 ± 1.98[a]	82.67 ± 1.98[a]	21.77 ± 3.66[a]	64.67 ± 3.49[a]	-	35.33 ± 3.49	33.11 ± 3.61
0.05	0.0102	21.33 ± 2.23[a]	78.67 ± 2.23[a]	26.27 ± 2.15[a]	58.00 ± 2.68[a]	14.93 ± 0.74	50.67 ± 3.68	48.97 ± 2.31
0.10	0.0214	32.67 ± 3.17[a]	67.33 ± 3.17[a]	42.59 ± 2.63[a]	40.00 ± 2.07[a]	21.24 ± 2.32	68.40 ± 1.23	67.31 ± 1.77
0.50	0.1160	53.33 ± 2.67[a]	46.67 ± 2.67[a]	100[a]	-	-	100	100
1.00	0.2560	85.00 ± 1.71[a]	15.00 ± 1.71[a]	100[a]	-	-	100	100

Values are expressed as mean ± SEM of six replicates.
*Known weight of treated diet was given to each set of 25 larvae. After completion of life-cycle, the remaining food was reweighed to calculate the amount of fenoxycarb consumed per larva (µg/larva) at each concentration of fenoxycarb.
a, b and c Significantly different $p < 0.001$, $p < 0.01$ and $p < 0.05$ respectively compared with control when t- test was applied.
Total mortality includes larval mortality, pupal mortality and adult mortality.

** Calculated by Abbott's formula (1925)

Discussion

In the present investigation, fenoxycarb exposed to fourth instar larvae of *C. cephalonica* caused a significant enhancement in larval and pupal mortality and a significant reduction in pupation and adult emergence. Larval mortality which was 2.67+ 0.84% at 0.001ppm concentration of fenoxycarb increased to 85.00+1.71% at its 1.00 ppm concentration. In first instar larval treatment of *C. cephalonica* the tenure of fenoxycarb exposure was maximum, which revealed maximum toxicity to the insect (Singh and Tiwari, 2015), but in consecutive instars i.e. second instar (Singh and Tiwari, 2014a) and third instar (Singh and Tiwari, 2014b) the tenure of exposure gradually decreased that resulted in decreased order of toxicity in second and third instars. In this study, the fourth instar larvae exposed to fenoxycarb revealed comparatively poor toxicity and this may be possibly due to reduced tenure of exposure of fenoxycarb to the fourth instar larvae. Thind and Edwards (1986) reported that fenoxycarb at 1 and 5 ppm concentrations caused 100% larval mortality of both

insecticide susceptible and resistant strains of *T. castaneum.* Fenoxycarb, when exposed to last larval instar of *Ephestia kuehniella* at 0.1, 1 and 10 µg/ml concentrations caused extreme enhancement in its larval mortality (Moreno *et al*., 1992). In a similar way, fourth instar larvae of *C. cephalonica* treated with methoprene at doses 1.0 and 0.5µg/larva caused 100% larval mortality by 42 and 90 days respectively, and such larval treatment resulted in the formation of supernumerary larva (Ambika and Abraham, 1982). In addition, 10 and 100 µg of methoprene caused 11.00 and 16.67% larval mortality, respectively in the last larval instars of *C. cephalonica* (Deb and Chakravorty, 1985), and their larvae attained extraordinary giant size without undergoing extra moult. Similarly, Kostyukovsky *et al*. (2000) reported that 0.1, 0.5, 1, and 2 ppm of pyriproxyfen (a fenoxycarb derivative) caused 100% larval mortality in F_1 generation of insecticide susceptible and actellic resistant strain of *T. castaneum* when parental adults were allowed to lay eggs in treated food medium.

In holometabolous insects, the developmental switch between juvenile and adult forms depends on juvenile hormone (JH), a sesquiterpenoid produced by the corpora allata gland (Gilbert *et al*., 2000). The presence of JH in pre-final larval instars ensures that the next molt, promoted by ecdysteroids, produces another, only a larger larva (Nijhout, 1994). In the present investigation, larval mortality may be the result of competing of JHA with natural JH for binding to its receptors or to the JH carrier proteins, injuring the corpora allata cells, or interfering with JH biosynthesis (Leighton *et al*., 1981). Disturbed biometrical proportions of different parts of the body of affected larvae may also contribute to the ecdysial failure (Slama *et al*., 1974). Larval mortality however, may be also attributed to combination of secondary factors, which are not directly related to the hormonal ability of JHAs (Sehnal, 1983) but may also be related to the suffocation, bleeding and dessication due to imperfect excuviation, starvation due to morphogenetic effects, failure of vital homeostatic processes and metabolic impairment. Loss of normal activity due to JHA treatment can also be correlated with muscle paralysis (Mulder and Gijswijt, 1973), which could be the direct cause of larval mortality in any instar (Retnakaran and Wright, 1987; Soshkin, 1991).

In the present investigation, it was observed that due to increased larval mortality pupation decreased with increase in the fenoxycarb concentration. Up to 0.01 ppm concentration, pupation was not severely affected by the action of fenoxycarb but beyond this concentration pupation was severely affected in dose-dependent manner. The concentrations 0.50 and 1.00 ppm fenoxycarb are considered to be more toxic, as the rate of pupation was very low i.e 46.67± 2.67 and 15.00 ± 1.71 % respectively, in comparison to rest of the concentrations. Similar results have also been reported in case of *C. cephalonica* following exposure of methoprene (Deb and Chakravorty, 1985) and Ro 20-3600 (Bhargava and Devraj Urs, 1992), and in *T. castaneum* treated with methoprene (Kostyukovsky *et al*., 2000). Bhargava and Devraj Urs (1992) reported that exposure of Ro 20-3600, a juvenile hormone analogue, decreased pupation of *C. cephalonica* in dose-dependent manner when fourth or fifth instar larvae were treated. Decreased pupation with increase in concentration of fenoxycarb was also achieved by Moreno *et al*. (1992) in *E. kuehniella* and Liu and Chen (2001) in *Chrysoperla rufilabris*.

Pupation is normally reduced in JHA treated larvae either due to larval death during metamorphosis or inhibition of pupation due to prolonged juvenilization, when fed, injected or in contact with JHAs and/ or IGRs (Mondal and Parween, 2000).

Giant larvae produced by JHAs in *T. castaneum* also failed to pupate (Ishaaya and Yablonski, 1976). Pupation is found to be negatively correlated with the concentration of IGRs and positively correlated with the age of the treated larvae (Kramer *et al*., 1985; Mondal and Port, 1995; Parween, 1996).

Our finding regarding pupation in *C. cephalonica* following exposure to fenoxrcarb is in accordance with the results of Moreno *et al*. (1992) in case of *E. kuehniella* exposed to fenoxycarb; Liu and Chen (2001) in case of *Chrysoperla rufilabris* Ishaaya and Yablonski (1976) in case of *T. castaneum* treated with different JHAs; Deb and Chakravorty (1985) in case of *C. cephalonica* and Smet *et al*. (1989) in case of *T. confusum* exposed to methoprene; Bhargav and Devraj Urs (1992) in case of *C. cephalonica* treated with JHA (RO 20-3600) and Kostyukovsky *et al*. (2000) in case of *T. castaneum* exposed to methoprene**.**

The pupal mortality, in the present investigation, has also been recorded to increase with the increase in the concentration of fenoxycarb. Fenoxycarb at 1.00 ppm concentration caused very poor pupation, and all get perished. Increased pupal mortality, with increase in concentration of fenoxycarb was also reported by Moreno *et al*. (1992) in *E. kuehniella* and Liu and Chen (2001) in *C. rufilabris.* This result is supported by the the result of Parthasarathy and Palli (2009) indicated that 1 ppm concentration of methoprene blocked larval to pupal metamorphosis in 85% of larvae treated during the penultimate larval stage and more than 95% of larvae treated during the final instar larval stage in *T. Castaneum*. Exposure of C_{18} juvenile hormone has also been reported to increase pupal mortality in addition to the formation of pupal-adult intermediates in *T.castaneum* larvae (Edwards, 1976).

Fenoxycarb exposure to the larvae of *C. cephalonica*, in the present investigation, caused a significant dose-dependent reduction in adult emergence. The reduction in adult emergence is due to larval and pupal mortalities influenced by the fenoxycarb action. The adult emergence was recorded as 90.67 ± 1.69% at 0.001 ppm concentration of fenoxycarb that decreased to 40.00 ± 2.07 % at 0.10 ppm concentration of fenoxycarb. Due to 100% pupal mortality at 0.50 and 1.00 ppm concentrations of fenoxycarb, there was no adult emergence at both these concentrations of fenoxycarb. Similar to this result, Thind and Edwards (1986) have also reported that fenoxycarb even at its lower concentrations i.e. 0.001, 0.01and 0.1 ppm caused considerable reduction in adult emergence of insecticide susceptible and resistant strains of *T. castaneum, C. ferrugineus, O. surinamensis* and *R. dominica*. They further reported that 1 and 5 ppm concentrations of fenoxycarb caused 100% reduction of adult emergence of *T. castaneum, C. ferrugineus* and *O. surinamensis* whereas 93 and 100% reduction of adult emergence of *R. dominica* was observed at 1 and 5 ppm concentrations of fenoxycarb respectively. Our finding pertaining to adult emergence of *C. cephalonica* following exposure of fenoxycarb is in accordance with the results of the above workers as observed in case of *T. castaneum, C. ferrugineus, O. surinanamensis* and *R. dominica* treated with fenoxycarb (Thind and Edwards, 1986); *S. cerealella* exposed to fenoxycarb (Eisa, 1992); *P. interpunctella* (Ghasemi *et al*., 2010); *T. castaneum* (EI-barky *et al*., 2012) and *E. integriceps* (Mojaver and Bandani, 2010) treated with pyriproxyfen and *T. castaneum* and *T. confusum* exposed to methoprene (Tucker *et al*., 2014a, 2014b).

Earlier findings revealed that higher concentrations of fenoxycarb i.e. 0.01, 0.05,

0.10, 0.50 and 1 ppm in case of first and second instar and 0.05, 0.10, 0.50 and 1ppm in third instar larval treatment of *C. cephalonica* produced giant larvae, supernumerary larvae, larval-pupal intermediates and abnormal pupae (Singh andTiwari,2015,2014a,2014b). But, fourth instar larvae exposed to above concentrations of fenoxycarb, in the present investigation, revealed comparatively poor results. These larvae remained as larvae and after variable periods they stopped feeding and movement, turned black and eventually died. It also deserves mention that at 1.00 ppm concentration of fenoxycarb, in first instar exposed larvae, pupoids and adultoids did not form but extra moults occur resulting into the formation of giant larvae and supernumerary larvae which after certain life-span stopped feeding and movement, their body shrinked, body wall became loose, turned black and died (Singh and Tiwari,2015). We have considered those larvae as larval- pupal intermediate that were able to form cocoon but failed to form pupae inside cocoon. Similar observation has also been reported by Edwards (1976) in case of *Tribolium castaneum* larvae exposed to juvenoids for a longer period which frequently led to the production of giant larvae. Present finding was also supported by Dyte (1972) who observed that methoprene exposure for a tenure of 12 days produced giant larvae in *T. castaneum* that were heavier and darker than untreated larvae of the same age group. The effect was found to be similar in both susceptible (FSS II) and resistant (Kano and CTC 12) strains of the beetle, *T. castaneum* (Dyte, 1972).But, according to Hoppe (1976) exposure of Juvenoids such as Ro 10-3108 and Ro 20-3600 produced giant larvae in *T. castaneum* but not in *E. cautella*. These giant larvae were characterized by large head capsules and they completed development into morphologically normal appearing adults when transferred to untreated food.

It is thought that JHAs interact with DNA molecule and as a result, either the expression of the adult genes is inhibited (Williams, 1961), the larval genes are activated (Wigglesworth, 1961), or the replication of DNA in the larval part of the body is induced (Novak, 1967), which further results the retention of larval gene traps in the pupal stage or retention of pupal gene traps in the adults (Retnakaran *et al*., 1985), which may be plausible reason of observed abnormalities in pupae and adults form in the present investigation. JHAs also influence the endocrine physiology of the insect which may, in part, cause abnormal morphogenesis and is primarily seen during the larva-pupal transformation (Retnakaran *et al*., 1985). The degree of morphogenetic effects produced by JHAs differs with the mode of application, dose administered, species and age of the treated insects (Mondal and Parween, 2000).

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