

LIPASE ACTIVITY DURING LARVAL DEVELOPMENT OF AN INSECT, *LEUCINODES ORBONALIS* (GUENEE)

RAMESH. M. GEJAGE AND MANISHA R. GEJAGE*
KUSUMATAI RAJARAMBAPU PATIL KANYA MAHAVIDYALAYA,
ISLAMPUR (SANGLI)-415 409, INDIA.
VISHWASRAO NAIK, MAHAVIDHYALYA, SHIRALA
(SANGLI)-415 408, INDIA*.
(e-mail : rameshgejage@yahoo.com)

The brinjal fruit borer, *Leucinodes orbonalis* (Guenee) is an important pest of eggplant in South Asia. Lipase activity during larval development of an insect, *L. orbonalis* have been studied. The larval lipase revealed optimum pH 7.8, incubation time 30 minutes, temperature 37°C, enzyme concentration 1% and substrate concentration 5%. The gradual increase in larval lipase activity was observed from 1-day old larvae to 8-day old larvae and gradual decrease from 8-day old larvae to 11-day old larvae. The maximum lipase activity was observed in 8- day old larvae. The physiological role of lipase during larval development of *L. orbonalis* (Guenee) has been reported in present paper.

Key words : Triacylglycerol lipase, insect, larval development, *L. orbonalis* (Guenee).

INTRODUCTION

Brinjal shoot and fruit borer, *Leucinodes orbonalis* (Guenee) is the most destructive pest of brinjal. The young larvae bore into tender shoots near the growing points into flower buds. The immature fruits are also damaged, which finally leads to the economic loss to the farmers. The larval developmental stage is of 11 days in August and September. The triacylglycerol lipase is an enzyme which is responsible for hydrolysis of triglyceride. Lipolytic enzymes are indispensable for the biological turnover of lipids. They are required as digestive enzymes in the transfer of lipid from one organism to another, that is from plant to animal and from animal to animal. Within the organisms, they are instrumental in the deposition and mobilization of the fat. They are also involved in the metabolism of intracellular lipids Pol & Salunkhe (2001).

Many attempts have been made to investigate lipase activity in few insect species (Price, 1975; Radha Pant & Sharma, 1978; Male & Story, 1981; Pistillo *et al.*, 1998; Pol & Sawant, 1999; Arreguin *et al.*, 2000; Pol & Sakate, 2001; Pol & Gejage, 2002; Pol & Salunkhe, 2002a & b; Ponnuvel *et al.*, 2003; Gejage & Awate, 2009; Horne *et al.*, 2009). The information on the larval development of *L. orbonalis* (Guenee) is rather scanty. In the present investigation, an attempt has been taken to evaluate lipase activity during larval development of *L. orbonalis* (Guenee) which is mainly concerned with release of energy for their active life and structural components of larval growth.

MATERIALS AND METHODS

The culture of *L. orbonalis* (Guenee) was maintained in the laboratory on the natural food of brinjal fruit as per the method suggested by Pol & Gejage (2002). The larval developmental stages from 1-day to 11-day larvae were taken for study of lipolytic activity. For the enzyme preparation the larvae were isolated and repeated cleaned with distilled water, weighed and homogenized in the cold double distilled water using a

ground glass mortar and pestle. The homogenate were diluted with cold double distilled water so as to get 1% (wt/vol) concentration. Such homogenate were used for the assay of lipolytic activity. The lipase was assayed by the method of Hayase & Tapple, 1970. The assay system contained 0.25 ml of 5% substrate dispersed in gum acacia; 1.0 ml of 0.1 M tris-maleate buffer pH 7.8 and 0.25 ml of 1% (wt/vol) enzyme solution in a total volume of 1.5 ml. The incubations were carried out in a Shaker with a continuous shaking for 30 minutes in glass stoppered vessels at 37°C. The colour was developed by the addition of 1ml of 0.5% solution of mixture of diphynyl carbazone and diphynylcarbaid (5:95 w/w) in methanol. At the end of the incubation the liberated fatty acids were measured colorimetrically (Itaya, 1977).

RESULTS AND DISCUSSION

Larval developmental period of *L. orbonalis* (Guenee) was 11 days. Lipase activity during larval development of *L. orbonalis* (Guenee) is shown in figure 1. The larval fat body lipase revealed optimum pH 7.8, incubation time 30 minutes, temperature 37 °C, enzyme concentration 1% and substrate concentration 5%. The gradual increase in larval lipase activity was observed from 1-day old larvae to 8-day old larvae and gradual decrease from 8-day old larvae to 11-day old larvae. The maximum lipase activity was observed in 8-day old larvae.

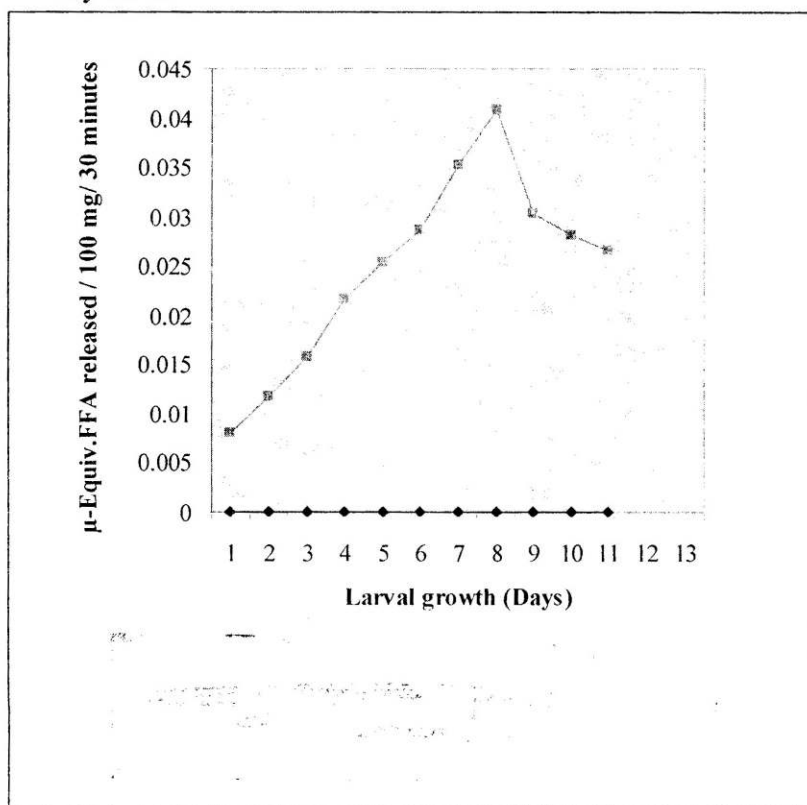


Fig. 1 : Lipase activity during development of *L. orbonalis* (Guenee).

In 7-day blowfly larvae lipase activity in fat body was optimal over the pH range 7.5 to 8.0 Price (1975). Male & Story (1981) were studied enzyme activities and isozyme

composition of triglyceride, diglyceride and monoglyceride lipases in *Periplaneta americana*, *Locusta migratoria* and *Polia adjuncta*. Pol & Sawant (1990) were studied lipolytic activity profile during larval growth of *Chrysomya rufifacies*. The fat body of *Manduca sexta* had a pH optimum 7.9 (Arrese & Wells, 1994). The lipase activity in fat body of *Chrysomya rufifacies* during larval growth and metamorphosis was maximal at the broad pH range 8.5 to 9.0 (Pol & Sawant, 1997). *Drosophila melanogaster* lipase homologs : a gene family with tissue and developmental specific expression were studied by Pistillo *et al.* (1998). Lipase activity was maximum in the 4-day larvae of blowfly *Chrysomya rufifacies* (Pol & Sawant, 1999). Purification and properties of a lipase from *Cephaloleia presignis* (Coleoptera : Chrysomelidae) were studied by Arreguin *et al.* (2000). The larval fat body lipase of *Chilo partellus* showed maximum activity at pH 8.0 by Sakate & Pol (2002).

Ponnuvel *et al.* (2003) isolated lipase from the silkworm *Bombax mori* shows antiviral activity against Nucleopolyhedrovirus. The main triglyceride-lipase (TG-lipase) from the fat body of *Manduca sexta* has been identified as the homolog of *Drosophila melanogaster* CG855 (Arrese *et al.*, 2006). The fat body lipase of *L. orbonalis* (Guenee) showed maximum activity at pH 7.9 by Gejage & Awate (2009).

Lipases have key roles in insect lipid acquisition, storage and mobilization and are also fundamental to many physiological processes under pinning insect reproduction, development, defense from pathogens and oxidative stress and pheromones signalling Horne *et al.* (2009). The hydrolysis of triglycerides by larval fat body homogenate indicates the presence of triacylglycerol lipase (EC 3.1.1.3) in the larval fat body homogenate of *L. orbonalis* (Guenee). Similar observations were reported in larval blowfly, *Chrysomya rufifacies* by Pol & Sawant (1999); in larval armyworm, *Mythimna separata* by Pol & Salunkhe (2001); in larvae of *Chilo partellus* by Pol & Sakate (2001) and in larval fat body lipase of *L. orbonalis* (Guenee) by Gejage & Awate (2009).

In the present work larval development of *L. orbonalis* (Guenee) showed maximum lipase activity at pH 7.8. This indicates the larval lipase is maximally active at an alkaline pH. The gradual increase in larval lipase activity was observed from 1-day old larvae to 8-day old larvae indicates that during this early feeding period of larval development of *L. orbonalis* (Guenee), the larvae are most active and the energy required for their active life may be supplied by triacylglycerol catabolism and supply of structural components to the developing larvae. Gradual decrease from 8-day old larvae to 11-day old larvae suggested the later feeding period of larval development was slow as compared to early active feeding period and accumulation of lipid which utilized during metamorphosis. The maximum enzyme activity observed in 8-day larval development indicated most feeding larval stage required more energy and structural components for larval growth. The present findings are in good agreement with Price (1975); Radha Pant & Sharma (1978); Pol & Sawant (1999); Pol & Sakate (2001); Pol & Salunkhe (2002a); Arrese *et al.* (2006); Gejage & Awate (2009) and Horne *et al.* (2009).

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