Ultramorphology of digestive tract of *Anticarsia gemmatalis* (Hübner, 1818) (Lepidoptera: Noctuidae) at final larval development

Ultramorfologia do trato digestivo de *Anticarsia gemmatalis* (Hübner, 1818) (Lepidoptera: Noctuidae) no final do desenvolvimento larval

Sheila Michele Levy[1*]; Ângela Maria Ferreira Falleiros[2]; Flávio Moscardi[3]; Elisa Aparecida Gregório[4]; Luis Antônio Toledo[4]

Resumo

O trato digestivo dos insetos constitui uma importante barreira físico-química natural contra invasão de patógenos. Algumas larvas de lepidópteros são consideradas pragas agrícolas potenciais e sua biologia tem recebido muita atenção; no entanto, pouco se sabe sobre a morfologia do sistema digestivo. A análise morfológica do trato digestivo de *Anticarsia gemmatalis* em nível ultraestrutural é um método bastante eficaz para o estudo dos seus mecanismos de defesa. Os materiais foram fixados (solução de glutaraldeído 2,5%; 0.1M tampão fosfato, pH 7.3), pós-fixados (tetróxido de ósmio 1% no mesmo tampão), desidratados em ponto crítico, recobertos com ouro e analisados ao microscópio eletrônico de varredura 515-Philips. O trato digestivo de *A. gemmatalis* consiste de um tubo retilíneo de diâmetro e comprimento variável, subdividido em três regiões: intestino anterior formado pela cavidade bucal, faringe, esôfago e papo; o intestino médio que é a região mais longa do trato digestivo, sem aparente diferenciação morfológica ao longo do comprimento; e o intestino posterior que é diferenciado em piloro, íleo, cólon, e reto. Embora a morfologia geral do trato digestivo *de A. gemmatalis* seja bastante semelhante ao de outras espécies de Lepidoptera, o arranjo anatômico das camadas musculares do papo difere do descrito para larvas destes insetos.
Palavras-chave: Microscopia eletrônica de varredura, tubo digestivo, lagarta-da-soja

Abstract

The digestive tract of insects is an important natural, physical, and chemical defense barrier against pathogen invasion. Certain lepidopteran caterpillars are serious pests of agricultural crops and their biology has received much attention, but little is known about the larval noctuid gut. The morphological analysis of the digestive tract in *Anticarsia gemmatalis* under scanning electron microscopy (SEM) is a good model for studies about its defense mechanism. The material was fixed (2,5% glutaraldehyde solution; 0.1M-phosphate buffer, pH 7.3), post-fixed (1% osmium tetroxide in the same buffer), dried at critical point, gold coated and analyzed in a SEM 515-Philips. *A. gemmatalis* digestive tract consists of a straight duct of varying length and diameter, subdivided in three main regions: the foregut formed by the oral cavity, pharynx, esophagus, and crop; the midgut that is the largest portion of the digestive tract

1 Docente do Dept. de Histologia, Centro de Ciências Biológicas, Universidade Estadual de Londrina - UEL, Londrina, PR, Brazil E-mail: sheilalevy@uel.com.br.
2 Docentes do Dept. de Histologia, Centro de Ciências Biológicas, Universidade Estadual de Londrina - UEL, Londrina, PR, Brazil.
3 Pesquisador do Centro Nacional de Pesquisa da Soja, Embrapa, Londrina, PR, Brazil
4 Docentes do Dept. de Morfologia, Instituto de Biociências, Universidade Estadual Paulista- UNESP, Botucatu, SP, Brazil.
* Autor para correspondência

Recebido para publicação 26/06/07 Aprovado em 15/12/07

without noticeable morphological differentiation along its length; and the hindgut that is morphologically differentiated in pylorus, ileum, colon, and rectum. Although the general morphology of the *A. gemmatalis* digestive tract is quite similar to the other Lepidoptera species, the anatomical array of the crop muscular layers is quite different comparing with the description for other larval insect.
Key words: Scanning electron microscopy, digestive tube, velvet bean caterpillar

Introduction

The digestive tract of the insects constitutes the main natural and physical-chemical defense barrier against microorganism invasion that are ingested with the food. Three main regions constitute the digestive tract in insects: foregut, midgut and hindgut (CHAPMAN, 1998; EATON, 1988; TERRA; FERREIRA, 1994). The midgut is the largest portion of the digestive tract, playing a major role in the absorption of nutrients, chemical and biological insecticides (BARRETT et al., 1998; BILLINGSLEY; LEHANE, 1996; CRISTOFOLETTI; RIBEIRO; TERRA, 2001 SANTOS et al., 1984).

The pathogen's action has been used for the biological control of the insect that affects the agriculture crop, leading to an increase of interest for knowledge about its internal morphology. The larvae of *Anticarsia gemmatalis* (Hübner 1818) (Lepidoptera: Noctuidae) are considered one of the most serious pests of agricultural soybean crops, and they are known as velvet bean caterpillar (MOSCARDI; CARVALHO, 1993). Although there is an effective program for the biological control of *A. gemmatalis* using the nucleopolyhedrovirus that get into the insect through the digestive tract (FLIPSEN et al., 1995; MOSCARDI; CARVALHO 1993), the internal morphology of this insect is not well known.

Many studies have been described the structure of the digestive tube in Lepidoptera larvae, most of them emphasizing histological and ultrastructural aspects of the constituting cells at their different regions (ANDERSON; HARVEY, 1966; BILLINGSLEY; LEHANE, 1996; CAVALCANTE; CRUZ-LANDIM, 1999; JORDÃO et al., 1999; JUDY; GILBERT, 1970), but none of them related to anatomical aspects. This work provides the ultramorphological description of the digestive tract in *A. gemmatalis* larvae in order to improve the anatomical knowledge of this system, as well as to compare the results with other insect's orders.

Material and Methods

Anticarsia gemmatalis (Hübner) larvae were provided by the Laboratório Entomológico - Centro Nacional de Pesquisa da Soja (Embrapa Soja), Londrina, Paraná State, Brazil. The larvae were maintained in the laboratory with artificial diet (HOFFMANN-CAMPO; OLIVEIRA; MOSCARDI, 1985) under controlled temperature (25-27^{o}C), photoperiod (14h light/10h dark), and 80% relative humidity.

Larvae of the 5th and 6th instars (12-16 days old larvae) were dissected under stereomicroscope. For ultramorphological analyzes, the material was fixed in 2.5% glutaraldehyde in 0.1 M phosphate buffer (pH 7.3), post-fixed in 1% osmium tetroxide solution in the same buffer, dehydrated with alcohol and dried at the critical point (CPD 010, Balzers Union) in liquid CO_2. The materials were coated with gold in sputtering device (MED 010, Balzers Union), analyzed and photographed with SEM 515 (Phillips) scanning electron microscope.

Results and Discussion

The digestive tract in *A. gemmatalis* larvae is a relatively straight tube, with variable length and diameter where three main regions can be recognized: foregut, midgut and hindgut (Figs. 1B, 1C, 2A, 2B, 2C, 3A, 3B and 3C), as described for other phytophagous species (EATON, 1988; LEVY et al., 2004a, 2004b; JUDY; GILBERT, 1969; SMITH et al., 1969; MARANHÃO, 1978; MATHUR, 1972;).

The foregut is constituted by oral cavity, pharynx, esophagus and crop (Fig. 1). The buccal cavity opens externally in the mouth that is located among the mandibles, epipharynx and hypopharynx (Fig. 1A); two pairs of dorsal dilator muscles characterized morphologically the buccal cavity. The pharynx and the esophagus are a narrow duct showing well-developed circular, dorsal and ventral dilator muscles (Fig. 1B). Similar characteristic was described in other Lepidoptera (DRECKTRAH; KNIGHT; BRINDLEY, 1966; EATON, 1988; JUDY; GILBERT, 1969; MATHUR, 1972; STANDLEA; YONKE, 1968) Coleoptera (AREEKUL, 1957; VASQUES, 1988) and Diptera (BOULARD, 1969; HUNG; LIN; LEE, 2000; PATIL; GOVINDAN, 1984). According to Gillott (1995), the foregut anatomical structure can be correlated with the insects feeding habits: if they feed with solid material, like Lepidoptera, Blattaria and Orthoptera, variation in the diameter due to the circular and dilator muscles would be responsible for the food propulsion; in the sucking insects these muscles would be more developed and specialized to allowed this region to function as a sucking pump for the food.

The crop in *A. gemmatalis* larvae is a sack like structure surrounded by muscular layers (Figs. 1B and 1C). The outer circular muscular fibers are better visualized at the junction of the esophagus-crop as well as between the crop-midgut (Figs. 1B and 1C), with different anatomical array at the ventral and dorsal side of the insect. In the crop's dorsal region these circular fibers form a ring-like structure only at posterior and anterior boundary of the crop, in the middle region of the crop the anastomosing circular muscle fibers insert bilaterally on an outer group of longitudinal muscles (Fig. 1B). In the crop's ventral region the circular fibers are thin and sparsely arranged surrounding a network of larger longitudinal fibers (Fig. 1C). This particular aspect of this muscular arrangement has not been described in others Lepidoptera larvae.

The crop was considered as a simple dilatation of the posterior region of the esophagus (CHAPMAN, 1998). In insects with a slow digestive process, the crop dilatation could be related with its ability to store food (BRUSCA; BRUSCA, 1990; WIGGLESWORTH, 1984); besides, the beginning of the digestion would occur in this region due to the action of the digestive enzymes from the salivary glands as well of those regurgitated from the midgut (CHI et al., 1975; RICHARDS; DAVIES, 1994).

A constriction named cardiac sphincter exists between the end of the foregut and the beginning of the midgut. In *A. gemmatalis* this junction can be externally visualized by well-developed circular muscular fibers (Fig. 1C). Similar aspects were found in *Achaea janata* Linnaeus (MATHUR, 1966) *Heliothis zea* Boddie (STANDLEA; YONKE, 1968), *Hyalophora cecropia* Linnaeus (JUDY; GILBERT, 1969) and *Rhynchophorus palmarum* Linnaeus (SÁNCHEZ et al., 2000). According to Maranhão (1978) this structure functions as a reflux sphincter, not allowing the return of the food from the midgut to the foregut.

The midgut was the largest and the longest region of the digestive tract (Figs. 1C, 2A, 2B, 2C, 2D and 3A). According to Gillott (1995), the length and the diameter of the midgut may vary depending on the feeding habits of the insect species. The muscular layer of the midgut in *A. gemmatalis* is less developed than in the foregut (LEVY et al., 2004b). It is constituted by internal circular layer and an external longitudinal one (Figs. 2A, 2B and 2C). Similar findings were described for most of the insects (BINDER; BOWERS, 1994; CAETANO, 1988; CHI et al., 1975; HECKER, 1977; SMITH et al, 1969; WERNER; MOUTAIROU; WERNER, 1991). Unlike the visceral muscles of the vertebrate digestive system, the ones of insects are formed by striated fibers (RICHARDS; DAVIES, 1994; CHAPMAN, 1998).

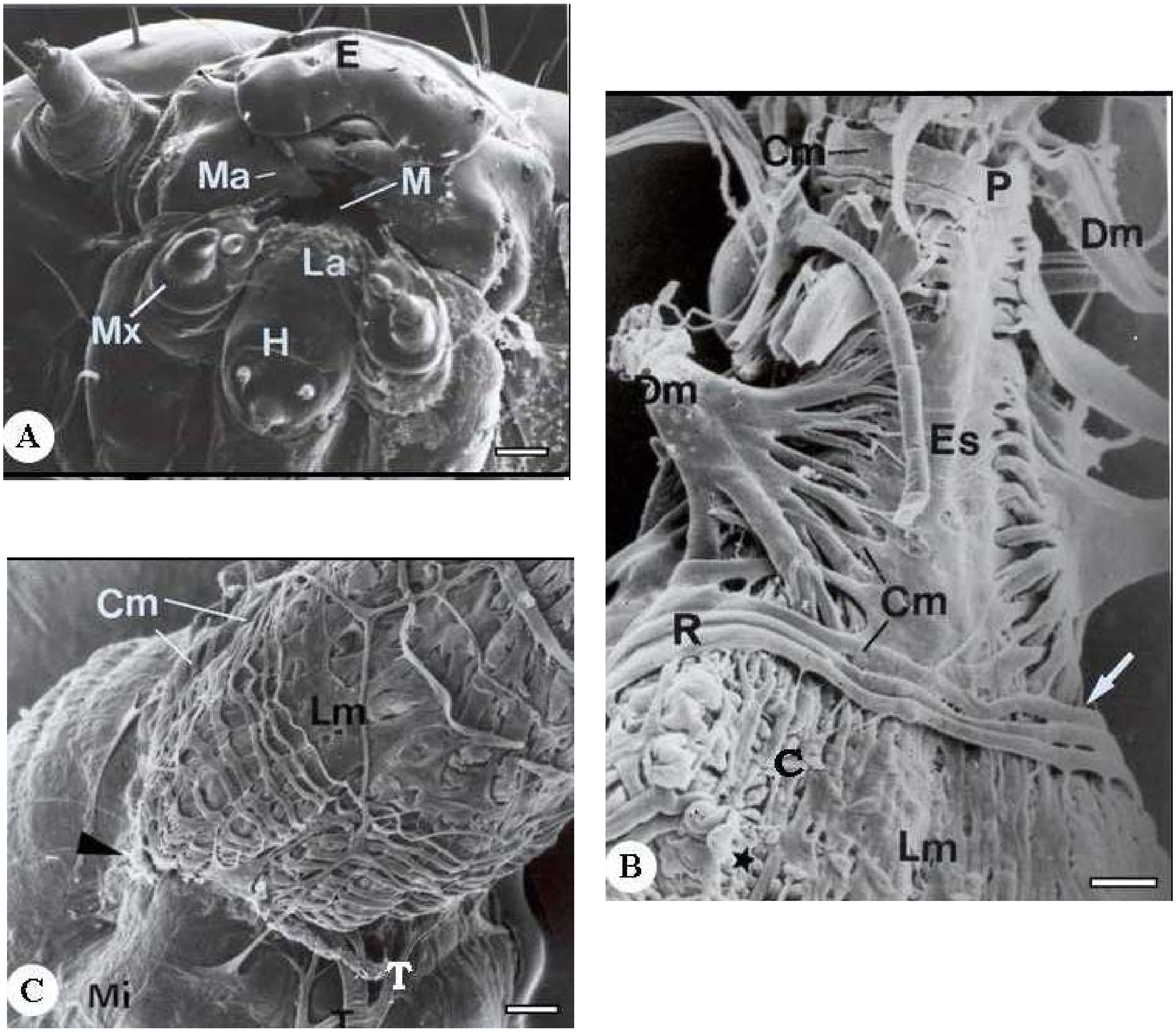

Figure 1. SEM micrograph of the *A. gemmatalis* foregut. A- Ventral view of the mouthparts: epipharynx (E), mandible (Ma), maxilla (Mx), labium (La), hypopharynx (H), mouth (M). Bar=50µm.. B- Dorsal view of the pharynx (P), esophagus (Es) and crop (C): dilator dorsal muscles (Dm), esophagus-crop limit (➔) with ring-like structure of outer circular muscles (Cm); network of circular muscle fibers (★) with the central longitudinal layer of muscle fibers (Lm). Barra=100µm. C- Ventral view of the crop: thin outer circular muscular fibers (Cm), large longitudinal fibers (Lm). Cardiac valve (▶), midgut (Mi), tracheae (T). Bar=100µm.

Figura 1. Elétron-micrografia do intestino anterior de *A. gemmatalis*. A- Vista ventral do aparelho bucal: epifaringe (E), mandibula (Ma), maxila (Mx), lábio (La), hipofaringe (H), boca (M). Barra=50µm. B- Vista dorsal da faringe (P), esôfago (Es) e papo (C): músculo dilatador dorsal (Dm), limite esofago-papo (➔) com fibras musculares circular (Cm) em forma de anel, rede de fibras musculares circular (★) e longitudinal (Lm). Barra=100µm. C- Vista ventral do papo: músculo circular externo (Cm), fibras musculares longitudinais (Lm). Válvula cárdia (▶), intestino médio (Mi) e traquéias (T). Barra=100µm.

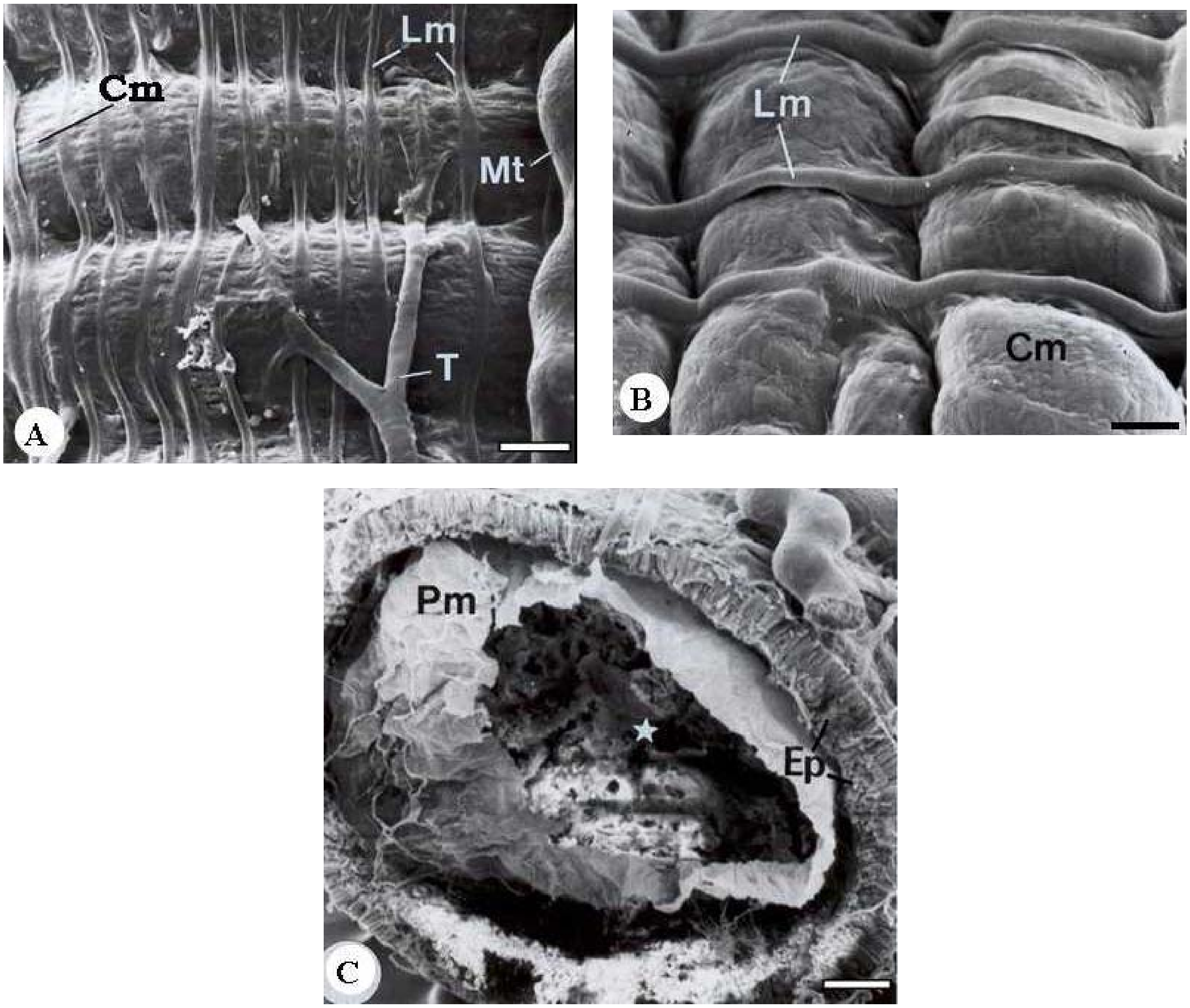

Figure 2. SEM micrograph of the A. *gemmatalis* midgut. A/B- External surface: longitudinal muscular layer (Lm), circular muscular layer (Cm), Malpighian tubules (Mt), tracheae (T). A- Bar=25µm; B- Bar=50µm. C- Cross section: peritrophic membrane (Pm), midgut epithelium (Ep), food bolus (★). C- Bar=100µm.

Figura 2. Elétron-micrografia do intestino médio de *A. gemmatalis*. A/B - Superfície externa: camada muscular longitudinal (Lm) e circular (Cm), túbulos de Malpighi (Mt), traquéias (T). A- Barra=25µm; B- Barra=50µm. C- Corte transversal: membrana peritrófica (Pm), epitélio do intestino médio (Ep), bolo alimentar (★). Barra=100µm.

The hindgut, the last and most complex region of the alimentary canal, shows a high structural variability among the different insect groups. In *A. gemmatalis* the hindgut is formed by the pylorus, ileum, colon, and rectum (Figs. 3A, 3B, 3C and 3D), as described for most Lepidoptera (CHI et al*.,* 1975; EATON, 1988; JUDY; GILBERT, 1969, 1970; LEVY et al., 2004b), but other insect orders, as Hymenoptera (CAETANO; OVERAL, 1984), Coleoptera (SÁNCHEZ et al., 2000; SHEEHAN; CRAWFORD; WIGLEY, 1982) and some Diptera (PATIL; GOVINDAN, 1984) may exhibit different morphological portions.

The pylorus of *A. gemmatalis* has two distinct regions: an anterior region with well defined circular muscle fibers surrounding the longitudinal muscular layer (Fig. 3A), and a posterior region, named pyloric valve, where the common ampulla of the Malpighian tubules are inserted (Figs. 3A and 3B). There are

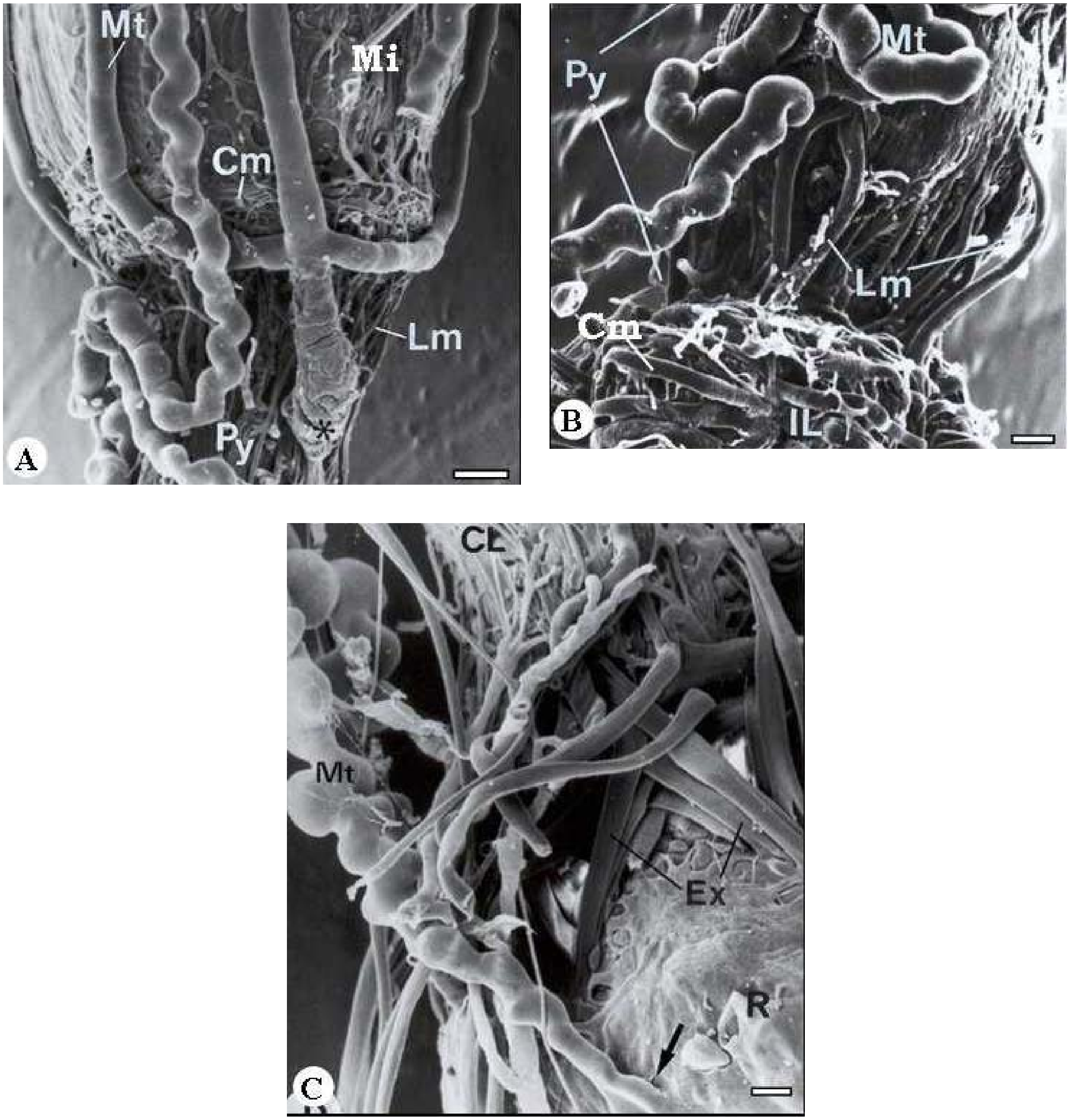

Figure 3. SEM micrograph of the A. gemmatalis hindgut. A- Midgut (Mi) and pylorus's (Py) limit: circular muscular layer (Cm); pyloric valve (Py); common Malpighian ampulla (*). Bar=100µm. B- Transition between the pylorus posterior region (Py) and ileum (IL). Bar=50µm. C- Transition between colon (CL) and rectum (R): extrinsic muscles fibers (Ex), distal ends of Malpighian tubules inserted to rectal wall (➔). Longitudinal (Lm) and circular muscular layer (Cm), Malpighian tubules (Mt). Bar=50µm.

Figura 3. Elétron-micrografia do intestino posterior de A. gemmatalis. A- Limite entre intestino médio (Mi) e piloro (Py): camadas de músculo circular (Cm), válvula pilórica (Py), ampola comum dos túbulos de Malpighi (*). Barra=100µm. B- Transição entre região posterior do piloro (Py) e íleo (IL). Barra=50m. C- Transição entre cólon (CL) e reto (R): músculos extrínsicos (Ex), porção distal dos túbulos de Malpighi inseridos na parede do reto (➔). Barra=50µm. Musculatura longitudinal (Lm) e circular (Cm), túbulos de Malpighi (Mt).

longitudinal muscles fibers inserted beneath the circular muscle of the pyloric anterior region and attached beneath the strong circular muscle of the posterior pyloric valve (Figs. 3A and 3B). According to Gillott, (1995), this circular muscle layer helps the pyloric valve contraction, while the prominent longitudinal muscle layer is responsible for the pyloric valve relaxes after the food movements toward to ileum.

The ileum was more dilated than pylorus and presented well-developed circular muscles surrounding the internal longitudinal muscles layers (Fig. 3B). In wood eating insects like termites, symbionts microorganisms are found into the ileum and they are supposed to help in the fermentation and the cellulose digestion (COSTA-LEONARDO, 1995; MORAES; CAMARGO; COSTA-LEONARDO, 1990). Although the colon is longest and narrowest than the ileum, both hindgut regions present similar muscular layer (Figs. 3C and 3D). In *H. zea*, the ileum and colon cannot be easily subdivided like in other Lepidoptera as they have the same diameter (CHAUTHANI; CALLAHAN, 1967).

The rectum is usually an enlarged and dilatable sack, surrounded by dorsal and lateral extrinsic muscles fibers (Fig. 3D) that act opening the rectal valve, facilitating the feces movement. The main characteristic of this region is the rectal complex that is constituted by the association of the distal ends of the Malpighian tubules which cross the rectal wall (Fig. 3D), forming the Lepidopteran characteristic cryptonephric excretory system (EATON, 1988; LEVY et al., 2004b). The cryptonephric complex conserves water by absorbing it actively from the feces either to the hemolymph or to the lumen of the Malpighian tubules. Besides, this system is very important for insects that live in dried habitats, which use this mechanism to control the water reabsorbing and ions loss (RICHARDS; DAVIES, 1994; RIGONI; TOMOTAKE; CONTE, 2004).

In conclusion, although the general morphology of the *A. gemmatalis* digestive tract is quite similar to other Lepidoptera species, the anatomical array of the crop muscular layers are quite different comparing with the description for other larval insect.

Acknowledgements

We thank: the Centro Nacional de Pesquisa da Soja (Embrapa Soja), Londrina, Paraná, for supplying *A. gemmatalis* larvae, and the Departamento de Morfologia, UNESP, Botucatu, São Paulo for helping in the procedure and analysis of the material. This work was supported by the Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq) and Fundação de Amparo a Pesquisa do Estado de São Paulo (FAPESP) (Proc: 00/12183-2).

Literature Cited

ANDERSON, E.; HARVEY, W. R. Active transport by the *Cecropia* midgut: II. Fine structure of the midgut epithelium. *Journal of Cell Biology*, New York, v. 31, n. 1, p. 107-134, 1966.

AREEKUL, S. The comparative internal larval anatomy of several genera of Scarabaeidae (Coleoptera). *Annals of the Entomological Society of America*, College Park, v. 50, p. 562-577, 1957.

BARRETT, J. W.; BROWNWRITHT, A. J.; PRIMAVERA, M. J.; RETNAKARAN, A.; PALLI, S. R. Concomitant primary infection of the midgut epithelial cells and the hemocytes of *Trichoplusia ni* by *Autographa californica* Nucleopolyhedrovirus. *Tissue and Cell*, Essex, v. 30, n. 6, p. 602-616, 1998.

BILLINGSLEY, P. F.; LEHANE, M. J. Structure and ultrastructure of the insect midgut. In: LEHANE, M. J.; BILLINGSLEY, P. F. (Ed.) *Biology of the insect midgut*. London: Chapman and Hall, 1996. p. 3-30.

BINDER, B. F.; BOWERS, W. S. Precocene II-induced changes in the anatomy of midgut goblet cells of last instar larvae of *Helicoverpa zea* (Boddie) (Lepidoptera: Noctuidae). *International Journal of Insect Morphology and Embryology*, New York, v. 23, n. 1, p. 27-34, 1994.

BOULARD, C. Anatomie et histologie du tube digestif de la larve *D'Hypoderma bovis* (Diptere: Oestriforme). *Annales de la Societe Entomologique de France*, Paris, v. 5, p. 371-387, 1969.

BRUSCA, R. C.; BRUSCA, G. J. (Ed.) *Invertebrates*. Massachusetts: Sinauer Associates Inc Publishers, 1990.

CAETANO, F. H. Anatomia, histologia e histoquímica do sistema digestivo e excretor de operárias de formigas (Hymenoptera, Formicidae). *Naturalia*, São Paulo, v. 13, p. 129-174, 1988.

CAETANO, F. H.; OVERAL, W. L. Estudos do trato digestivo de vespas (Hymenoptera, Vespidae) e estruturas excretoras associadas. I. morfologia. *Revista Brasileira de Entomologia*, São Paulo, v. 28, n. 4, p. 403-408, 1984.

CAVALCANTE, V. M.; CRUZ-LANDIM, C. Types of cells present in the midgut of the insects: a review. *Naturalia*, São Paulo, v. 24, p. 19-40, 1999.

CHAPMAN, R. F. *The insects*: structure and function. Harvard: Cambridge University Press, 1998.

CHAUTHANI, A. R.; CALLAHAN, P. S. Developmental morphology of the alimentary canal of *Heliothis zea* (Lepidoptera: Noctuidae). *Annals of the Entomological Society of America*, College Park, v. 60, p. 1136-1141, 1967.

CHI, C.; DREW, W. A.; YOUNG; J. H.; CURD, M. R. Comparative morphology and histology of the larval digestive system of two genera of noctuidae (Lepidoptera): *Heliothis* and *Spodoptera*. *Annals of the Entomological Society of America*, College Park, v. 68, p. 371-380, 1975.

COSTA-LEONARDO, A. M. Morphology of the digestive tube in the termite *Serritermes serrifer* (Isoptera, Serritermitidae). *Naturalia*, São Paulo, v. 20, p. 31-44, 1995.

CRISTOFOLETTI, P. T.; RIBEIRO, A. F.; TERRA, W. R. Apocrine secretion of amylase and exocytosis of trypsin along the midgut of *Tenebrio molitor* larvae. *Journal of Insect Physiology*, Oxford, v. 47, p. 143-155, 2001.

DRECKTRAH, H. G.; KNIGHT, K. L.; BRINDLEY, T. A. Morphological investigations of the internal anatomy of the fifth larval instar of the european corn borer. *Iowa State Journal of Science*, Ames, v. 40, p. 57-86, 1966.

EATON, J. L. (Ed). *Lepidopteran anatomy*. New York: Wiley-Interscience, 1988.

FLIPSEN, J. T. M.; MARTENS, J. W. M.; VAN OERS, M. M.; VLAK, J. M.; VAN LENT, W. M. Passage of *autographa californica* nuclear polyhedrosis virus through the midgut epithelium of *Spodoptera exigua* larvae. *Virology*, New York, n. 208, p. 328-335, 1995.

GILLOTT, C. (Ed.) *Entomology*. New York: Plenum Press, 1995.

HOFFMANN-CAMPO, C. B. H.; OLIVEIRA, E. B.; MOSCARDI, F. (Ed.) *Criação massal da lagarta da soja (Anticarsia gemmatalis)*. Londrina: EMBRAPA/CNPSo, 1985.

HECKER, H. Structure and function of midgut epithelial cells in culicidae mosquitoes (Insecta, Diptera). *Cell Tissue Research*, New York, v. 184, n. 3, p. 21-41, 1977.

HUNG, C. N.; LIN, T. L.; LEE, W. Y. Morphology and ultrastructure of the alimentary canal of oriental fruit fly, *Bactrocera dorsalis* (Hendel) (Diptera: Tephridae): the structure of the midgut. *Zoological Studies*, Taipei, v.39, n. 4, p. 387-394, 2000.

JORDÃO, B. P.; CAPELLA, A. N.; TERRA, W. R.; RIBEIRO, A. F.; FERREIRA, C. Nature of the anchors of membrane-bound aminopeptidase, amylase, and trypsin and secretory mechanisms in *Spodoptera frugiperda* (Lepidoptera) midgut cells. *Journal Insect Physiology*, Oxford, v. 45, p. 29-37, 1999.

JUDY, K. J.; GILBERT, L. I. Morphology of the alimentary canal during the metamorphosis of *Hyalophora cecropia* (Lepidoptera: Saturniidae). *Annals of the Entomological Society of America*, College Park, v. 62, p. 1438-1446, 1969.

______. Histology of the alimentary canal during the metamorphosis of *Hyalophora cecropia* (L). *Journal of Morphology*, New York, v. 131, n. 3, p. 277-300, 1970.

LEVY, S. M.; FALLEIROS, Â. M. F.; GREGÓRIO, E. A.; ARREBOLA, N. R.; TOLEDO, L. A. The larval midgut of *Anticarsia gemmatalis* (Hübner) (Lepidoptera: Noctuidae): light and electron microscopy studies of the epithelial cells. *Brazilian Journal of Biology*, São Carlos, v. 64, n. 3B, p. 1-6, 2004a.

LEVY, S. M.; FALLEIROS, Â. M. F.; MOSCARDI, F.; GREGÓRIO, E. A.; TOLEDO, L. A. Morphological study of the hindgut in larvae of *Anticarsia gemmatalis* Hübner (Lepidoptera:Noctuidae). *Neotropical Entomology*, Londrina, v. 33, n. 4, p. 427-431, 2004b.

MARANHÃO, Z. C. (Ed.) *Morfologia geral dos insetos*. São Paulo: Nobel, 1978.

MATHUR, L. M. L. Morphology of the alimentary canal and ciated glands of the mature larva of *Achaea janata* L. (Lepidoptera: Noctuidae). *Indian Journal of Entomology*, New Delhi, v. 28, p. 318-331, 1966.

______. Developmental changes in the alimentary canal of *Prodenia litura* Fabr. (Lepidoptera). *Journal of Natural History*, London, v. 6, n. 1, p. 39-46, 1972.

MORAES, L. D. V.; CAMARGO, R. S; COSTA-LEONARDO, A. M. Anatomy of the digestive tube of imagoes of *Cornitermes cumulans* and *Ruptitermes* sp. (Isoptera, Termitidae). *Anais da Sociedade Entomológica do Brasil*, Jaboticabal, v. 19, n. 1, p. 95-99, 1990.

MOSCARDI, F.; CARVALHO, R. C. Z. Consumo e utilização de soja por *Anticarsia gemmatalis* Hüb.

(Lepidoptera: Noctuidae) infectada, em diferentes estádios larvais, por seu vírus de poliedrose nuclear. *Anais da Sociedade Entomológica do Brasil*, Jaboticabal, v. 22, n. 2, p. 267-280, 1993.

PATIL, G. M.; GOVINDAN, R. Internal anatomy of silkworm Uzi-Fly, *Exorista sorbillans* (Wiedemann) (Diptera: Tachinidae). *Indian journal of sericulture*, Bombay, v. 23, p. 22-31, 1984.

RICHARDS, O. W.; DAVIES, R. G. *IMMS'general textbook of entomology*. London: Chapman and Hall, 1994.

RIGONI, G. M.; TOMOTAKE, M. E. M.; CONTE, H. Morphology of malpighian tubules of *Diatraea saccharalis* (F.) (Lepidoptera: Crambidae) at final larval development. *Cytologia*, Tokyo, v. 69, n. 1, p. 1-6, 2004.

SÁNCHEZ, P. A.; SÁNCHEZ, F.; CAETANO, F. H.; JAFFÉ, K. El tubo digestivo en adultos de *Rhynchophorus palmarum* (L.) (Coleoptera:Curculionidae): Morfología y ultraestrutura. *Boletin de Entomologia Venezolana*, Caracas, v. 15, n. 2, p. 195-216, 2000.

SANTOS, C. D.; RIBEIRO, A. F.; FERREIRA, C.; TERRA, W. R. The larval midgut of cassava hornworm (*Erinnyis ello*): Ultrastructure, fluid flux and the secretory activity in relation to the organization of digestion. *Cell Tissue Research*, New York, v. 237, p. 565-574, 1984.

SHEEHAN, C. M.; CRAWFORD, A. M.; WIGLEY, P. J. Anatomy and histology of the alimentary canal of the black beetle, *Heteronychus arator*. *New Zealand Journal of Zoology*, Wellington, n. 9, p. 381-386, 1982.

SMITH, D. S.; COMPHER, K.; JANNERS, M.; LIPTON, C.; WITTLE, L. W. Cellular organization and ferritin uptake in the mid-gut epithelium of a moth *Ephestia kühniella. Journal of Morphology*, New York, v. 127, p. 41-72, 1969.

STANDLEA, P. P.; YONKE, T. R. Clarification of the description of the digestive system of *Heliothis zea*. *Annals of the Entomological Society of America*, College Park, v. 61, p. 1478-1481, 1968.

TERRA, W. R.; FERREIRA, C. Insect digestive enzymes: properties, compartmentalization and function. *Comparative Biochemistry and Physiology*, New York, v. 109B, n. 1, p. 1-62, 1994.

VASQUES, A. M. Estudo morfológico e histológico do tubo digestivo de larvas de *Pyrearinus termitilluminans* costa, 1982 (Coleoptera, Elateridae, Pyrophorini). *Revista Brasileira de Entomologia*, São Paulo, v. 32, p. 253-258, 1988.

WERNER, K.; MOUTAIROU, K.; WERNER, G. Formation and structure of the surface coat in the midgut of a waterstider, *Gerris najas* Deg. (Heteroptera: Gerridae). *International Journal of Insect Morphology and Embriology*, New York, v. 20, n. 1-2, p. 69-77, 1991.

WIGGLESWORTH, V. B. *The principles of insect physiology*. 8.ed. London: Chapman and Hall, 1984.