

Egyptian Academic Journal of Biological Sciences **D. Histology & Histochemistry** ISSN 2090 - 0775 http://eajbsd.journals.ekb.eg



Ultrastructure of the Midgut Adult, Trachyderma philistina (Coleoptera: Tenebrionidae)

Nancy Taha Mohamed; Ahmed Salem El-Ebiarie and Waffa Essam Zoology & Entomology Department, Faculty of Science, Helwan University. E.Mail: <u>nancyt0000@yahoo.com</u> - <u>aselebiarie@hotmail.com</u>

ARTICLE INFO

Article History

ABSTRACT In the previous section of the Midgut adult Trachyderma

Received:3/10/2020 Accepted:5/12/2020 Keywords: Trachyderma

philistine. Ultrastructure, Coleoptera, Tenebrionidae

philistina the apical section of the microvilli appeared to be in the form of a plucked membrane, and slender and even microvilli. There were a lot of ribosomes and rough endoplasmic reticulum between these plates of microvilli. Vesicles secretory are released from the mid-gut part. The cytoplasm near the apical part of the cell produced a well-developed oval nucleus. A lot of free ribosomes and rough endoplasmic reticulum surround the nucleus. Near the apical part of the cell, mitochondria occurred. The basal section of the anterior midgut of adult beetle Trachyderma philistina showed a welldeveloped, thick basement membrane. A well-designed, welldeveloped brush border microvillus appeared in the middle of the midgut. Cytoplasm showed up with mitochondria and numerous tracheas. During cytoplasm, the rough endoplasmic reticulum occurred. A lot of vacuums in the secretory were apically observed. A well-developed oval and circular nucleus were found near the cell base and surrounded by numerous crude endoplasmic reticulum A welldeveloped muscle fiber appeared apically and basically in the basement part of the cell. A very small cell membrane appeared in the base part of the cell.

INTRODUCTION

Insects were shown to have great differences in their organ and process of digestion because of the variations in food consumption. To adapt an insect to its nutrient source, vital for growth, development, reproduction, and population maintenance, it is necessary to have a unique combination of behavioral, physiological and biological processes (Slansky, 1982). The intestinal length is usually linked to diet. The insects that use high protein in their diets are generally short intestines (Pradhan, 1939).

A number of authors including Talbot (1928), Miller (1961), Mukherji and Singh take an interest in morphology and histology of the food channel in various groups of coleopterans (1973). The most extensive and longest food channel diversion is midgut canal. The most extensive and longest food channel diversion is midgut. The endodermal origin of the midgut is therefore not chitinous. It is well known that most digestive enzymes are secreted in insects and are mainly digestive (Gilmour, 1961; Dadd, 1970; Wigglesworth, 1972 Coleoptera midgut is long and spiny in adults such as Trypodendron lineatum (Schneider & Rudinsky 1969) (Lopez Guerrero, 2002).

Citation: Egypt. Acad. J. Biolog. Sci. (D-Histology and histochemistry) Vol.12(2) pp45-57(2020)

The midgut is morphologically differentiated into broad anterior and narrow posterior regions of certain species of Meloidae. In some Cleridae midgut species, which are divided in anterior, middle, and postal regions, based on size and form differences, small papillas are also covered with the exterior projections. The butterflies have regenerative cells (Chapman, 1998). Insect midgut Histologically has three types of epithelial cells, as described by Shinoda (1930), including columnar, clover, and regenerative cells that show functional variation in different insects. (Lewis, 1926; Waterhouse, 1952 and Wigglesworth, 1965).

One of the biggest kites is the Tenebrionidae family, the dark beetles. The Latin Tenebrio family name means one who loves obscurity. It is of economic importance as it pests contains insect that are cosmopolitan in nature and most imperatively are associated with stored products.

The objective of this paper is to study the various parts of the adult stage of *Trachyderma philistina* by means of a transmission electron microscope to provide a structural framework for the physiological interpretations of the process in this section.

MATERIALS AND METHODS 1- Collection of Insects:

Trachyderma philistina are collected from Cairo and Siwa in Egypt and identified according to Zumpt (1965).

2- Dissection of Organs:

In the adult phosphaticbuffered [PBS: 10 mM Na2SO4, 145 mM NaCL(pH 7,2)], mid-guts of Trachyderma philistina were dissected with the help of the thin entomologic needles and transferred to a tube with a of PBS small volume with а microscope magnification of 4x. In the middle, the middle and the back portions were divided.

3-Transmission Electron Microscope Preparations:

In order to achieve primary fixation of pH 7.4 at 4 ° C with 2,5% glutaraldehyde for 24 hours, the mid-gut dissected material was transferred from the phosphate buffer. Then rinse with osmium tetroxide for a double post fixation at room temperature for 30 minutes. The solution was twice rinsed, and alcohol dehydrated for the phosphate buffer after fixation. They had a rising alcohol sequence to replace water with alcohol. Organ specimens were subsequently transferred into resin ratios from 1:3 for 24 hours to 1:1 for 24 hours and to 3:1 sequential for 24 hours. Followed by twice for 3 hours, treatment with pure resin. Every sample was then inserted into Epson resin and incubated for 24 hours at a temperature of 70 degrees C. Every sample was a semi-thin section ($\notin 0.5$ mc) with the glass knife on an ultramicrotome (Boeckler ®, USA). This was accompanied by 1% methylene blue bleaching and 1% azure II (1:1) for the view of light microscopes (Olympus®, Japan). The sections of ultrathin (90 nm) were stained by Uranvl acetate and plumage. The ZEISS EM 10electron microscope then examined (Germany).

RESULTS

In the anterior part of the Trachyderma midgut of adult philistina, the apical part showed microvilli in the form of membrane with folds (plate 1 a, b, c) and in other parts microvilli appeared slender and uniform (plate 2 a, b, c). Between these folds of microvilli appeared a lot of ribosomes and rough endoplasmic reticulum (plate 1 b). Secretory vesicles are discharged from the apical part of the anterior midgut (plate 1 c). well-developed oval nucleus А appeared in the cytoplasm near the apical part of the cell. The nucleus is surrounded by a lot of free ribosomes and rough endoplasmic reticulum (plate 1 a, b). Mitochondria appeared near the apical part of the cell (plate 1 c). A well-developed thick basement membrane appeared in the basal part of the anterior midgut of adult beetle Trachyderma philistina (plate 1 a). In the middle part of the midgut, a very thick well-developed brush border microvilli appeared in the form of papillae (plate 3 a, b, c, d). Mitochondria and numerous tracheae appeared. (plate 3 a, b, d). Rough endoplasmic reticulum appeared throughout the cytoplasm (plate 3 c, d, plate 4 b, c, d). Also, smooth endoplasmic reticulum appeared as a fingerprint (plate 3 d). A lot of secretory vacuoles were observed directed apically (plate 3.a, b, c, d). A well-developed oval and circular nucleus appeared near the basal part of the cell and surrounded by numerous rough endoplasmic reticulum (plate 4 b,c,d). A layer of regenerative cell appeared basally (plate 4 c, d). A well-developed muscle fiber appeared apically and basally (plate 3 c & plate 4 d). A very thin basement membrane appeared in the basal part of the cell (plate 4 b, c, d). A lot of dense secretory granules were observed near the basal part of the cell (plate 4 b, c). In the posterior part of the midgut, microvilli appeared in the form of membranes due to the presence of numerous organelles that are discharging into the lumen (plate 5 a, b, plate 6 a, d). Lipid spheres and aggregations of glycogen are present throughout the cell (plate 5 a, c, d). A thin basement membrane was observed.



Plate (1): Electron micrograph showing anterior midgut of adult stage of *Trachyderma philistina*:

A: Lumen (lu), basement membrane (bm), nucleus (nu), rough endoplasmic reticulum (rer) and free ribosome (arrow). Magnification (x 5000).

B: nucleus (nu), rough endoplasmic reticulum (arrow) and free ribosome (rb). Magnification (x 12000).

C: Lumen (lu), nucleus (nu), microvilia (mv) and secretory vesicle (sv). Magnification (x 6000).

D: Lumen (lu)), smooth endoplasmic reticulum (ser), microvilli (mv), secretory vesicle(sv), secretory product(sp) and dilated rough endoplasmic reticulum (drer). Magnification (x 3000).



Plate (2): Electron micrograph showing anterior midgut of adult stage of *Trachyderma philistina*:

A: Lumen (lu), microvilli (mv), dilated rough endoplasmic reticulum (drer). Magnification (x 3000).

B: Lumen (lu), microvilli (mv), rough endoplasmic reticulum (rer), apical part (arrow) and dense granules(dg). Magnification (x 25000).

C: Lumen (lu), microvilli (mv), rough endoplasmic reticulum (rer) and mitochondria (arrow). Magnification (x 15000).

D: Lumen (lu), rough enoplasmic reticulum (rer), mitochondria(arrow) and vesicle of rough enoplasmic reticulum (vrer). Magnification (x 15000).

49



Plate (3): Electron micrograph showing middle midgut of adult stage of *Trachyderma philistina*:

A: Lumen (lu), mitochondria (mt), microvilli (mv), trachea (tr) and secretory vesicle (sv). Magnification (x 12000).

B: Lumen (lu, mitochondria (mt), microvilli (mv), trachea (tr), secretory vesicle (sv) and muscle fiber (mf). Magnification (x 6000).

C: microvilli (mv), muscle fiber (mf), trachea (tr), secretory vesicle (sv) and rough endoplasmic reticulum (rer). Magnification (x 12000).

D: Lumen (lu), rough endoplasmic reticulum (rer) in form of finger print, mitochondria (mt), muscle fibre (mf), trachea (tr)(arrow), secretory vesicle (sv) and smooth endoplasmic reticulum (ser) in form of finger print.Magnification (x 15000).



Plate (4): Electron micrograph showing middle midgut of adult stage of *Trachyderma philistina*:

A: Lumen (lu), microvilia (mv), trachea (tr) and dilated rough endoplasmic reticulum (drer). Magnification (x 4000).

B: rough enoplasmic reticulum (rer), nucleus (nu), basement membrane (bm) and secretory granules (sg). Magnification (x 5000).

C: rough enoplasmic reticulum (rer), nucleus (arrow), basement membrane (bm) and secretory granules (sg). Magnification (x 4000).

D: rough enoplasmic reticulum (rer), layer of regenerative cells (arrow), basement membrane (bm) and muscle fiber(mf). Magnification (x 6000).



Plate (5): Electron micrograph showing posterior midgut of adult stage of *Trachyderma philistina*:

A: microvilli (mv), lipid sphere (lip) and apical membrane (arrow), basement membrane (bm). Magnification (x 8000).

B: microvilli (mv), apical membrane (arrow), basement membrane (bm) Magnification (x 8000).

C: basement membrane (bm), lipid sphere (lip), glycogen (gly) and basal membranes (arrow). Magnification (x 8000).

D: lipid sphere (lip) and glycogen (gly). Magnification (x 8000).



Plate (6): Electron micrograph showing posterior midgut of adult stage of *Trachyderma philistina*:

A: lipid sphere (lip), basement membrane (bm), and apical folds (arrow). Magnification (x 4000).

B: lipid sphere (lip), basement membrane (bm), basal membranes (arrow) and secretory vesicle (sv). Magnification (x 10000).

C: basement membrane (bm), lipid sphere (lip), secretory vesicle (sv) and basal membranes (arrow) and muscle fibre(mf). Magnification (x 6000).

D: lipid sphere (lip). Magnification (x 3000).

DISCUSSION

The apical part of the anterior midgut of adult stage of possessed Trachyderma philistina microvilli in the form of membrane. Between these folds of microvilli appeared a lot of ribosomes and rough endoplasmic reticulum, these organelles press on microvilli causing them to be in the form of folds in this part of the cell as previously mentioned by Nancy et al., (2015) and Abdel-Meguid et al. (2013). In others it seemed slender and uniform, like other insects such as Aedes gambiae Hyalophora cecropia and infestane Triatoma. The cells are more absorbed than other cells in this region (Anderson and Harvey 1966, Burgos and Gutierrez 1976, Hecker 1977, Lane et al., 1996). Secretary vesicles are dispelled by the apical part of the anterior midgut. An adult midgut epithelium is common when cytoplasmic vesicles and entire cells enter the lumens. The alternative to this cell phenomena is: 1) the apocrine and holocrine digestive enzymes secretion in the lumen, 2) the autophagic processes that extend the lifetime of the midgut cells (Snodgrass, 1935; Lehane, 1998). Lumens of every beetle type are contained in cytoplasmic vesicles budding from the luminal surface. The fullness of these vesicles is often linked with the presence, but not always, of large regenerative bags as well as the extrusion of whole midgut cells. Regenerative pouch reclused cells cannot substitute damaged or lost cells in short-lived beetles or feed as adults for damaged or lost cells (Nardi et al., 2012).

In the central part of the midgut of adult beetle *Trachyderma philistina*, a thick well developed brush border microvilli appeared which shows a higher absorption in the middle midgut than in the anterior midgut. Appeared mitochondria with numerous tracheae. This can be because energy is required to perform protein synthesis (Boonsriwong, et al., 2012). The cytoplasm was full of endoplasmic reticulum. A large number of proteins are synthesized inside the cells in both regions due to the raw endoplasmic reticulum. Golgi complex, lysosomes and other cytoplasmic vesicles could be used as membranes, for separation or as part of certain organells. (Nishiitsutsuji-Uwo and Yasuhisa 1981 Balogun 1969). A very well developed, oval and cyclical nucleus appeared near a basal area of the cells, with a large raw endoplasm. Such reticular an could indicate organization high protease synthesis, or a mechanism regulating RNA transportation from nucleus to cytoplasm, (Stäubli et al, 1966) (Reinhardt 1976). There appeared apically, and essentially, a welldeveloped Muscle Fibre. In the rear section of the midgut, many apical appeared membranes as apical protrusions. These membranes have been ruptured to release digestive organels. Inside the midgut of adult beetle Trachyderma philistina, peritrophic membrane has not been observed. In a review Terra (1990) notes that the digestive systems for can "change remarkably insects between larvae and holometabolous insect adults." This statement refers to the medium-sized beetle architecture that is examined as larvae and adults. Lumen of larva Tenebrio (1933 by 1990). The peritrophic Dehn. membranes are not contained in adult Beetles of the same species. These membranes are reported. Peritrophic membranes are nor, as in T's case, are universal features of insect midguts. In all stages of the development of a particular species, molitor is always present. In adult midgut sections, only two Chrysomelidae feeding members were shown to have peritrophic membranes. The peritrophic membranes of the both adult Chrysomelid beetles were on their whole surfaces. The peritrophic membrane likely has a protective barrier in the medium-gut epithelium apical surface of this species

which reduces the needs of the monocellular epithelial cells, which can degenerated continuously with be increased renewal bag densities in the medium-gut epithelium (Snodgrass, 1935; Malagoli et al., 2010). The lumens in Midgut with a high regenerative bag density were neither straightforward in their peritrophic membranes nor in adult beetle midguts, such as the anobiid S. paniceum and the bruchid Callosobruchus maculatus. Peritrophic membranes that line midgut insects are divisive, but presumably also increase the cell survival by protecting the luminous surfaces against the abrasive effects of the food they consume (Gullan and Cranston, 2010). The exception was peritrophic membranes, rather than the rule of adult beetles, and the beetle's regenerative density was low. The protective function of a peritrophic membrane or its substitution in regenerative pouches by dividing the cell divider offset all declines and damage to midgut cell populations. The density and presence of peritrophic membranes is adversely affected by regenerative sachets for beetles feeding adults. Both peritrophic membrane and renewable sacks may be missing or reduced significantly in adult bugs that seldom feed. (Nardi et al., 2012).

The location of longitudinal midgut muscles in relation to their proximodistal axes varies according to beetle's species. 1) adult beetles feed habitat, 2) presence of peritrophic membranes, and 3) expulse of whole midgut epithelial cells or fragments of these epithelial cells to midgut lumens are the product of their presence, size, and density. (Nardi *et al.*, 2012).

The cell contains numerous lipid spheres and glycogen aggregations. These are one of the most important intracellular sugar store areas due to glycogen granules observed in the midgut epithelia. These granules are used for the energy required for the different metabolism operations known in the intestinal epithelial cells in mammals (Novikoff 1983). Glycogen deposits often have an extreme absorption function in connection with cells. (Billingsley 1990).

REFERENCES

- Afaf A. Meguid Hanan H. Awad, Aziz H. Omar and Heba A.S.Elelimy (2013). Ultrastructure study on the mid-gut regions of the Milkweed Bug Spilostethus pandurus Scop. Asian Journal of Biological Sciences, 6(1):54-66,
- Anderson E. and Harvey W.R. (1996). Active transport by the *Cecropia* mid gut. II. Fine structure of mid gut epithelium. *Journal cell Biology*, 31:107-134.
- Balogun, R. A. 1969. Digestive enzymes of the alimentary canal of the larch bark beetle.Comparative.Biochemistr y. *Physiololog*, 29: 1267-1270.
- Billingsley, P. F. 1990. The midgut ultrastructure of hematophagous insects. Annuals Review of. Entomology, 35: 219.
- Boonsriwong, W., Kom, S. and Jimmy, K. (2007). Fine structures of the alimentary canal of the larval blow fly *Chrysomya megacephala. Parasitology Research*, 100: 561–574.
- Boonsriwong, W., K.L. Sukontason, T. Chaiwong, U.Chaisri R. . C.Vogtsberger, and K.Sukontason, (2012): Alimentary canal of the adult blow fly, Chrysomya megacephala (F.) (Diptera: Calliphoridae) - Part I: Ultra structure of Salivary glands. **Parasitology** Journal of Research, ID 382917, 7 pages.
- Burgos, M. H., and L. S. Gutierrez. 1976. The intestine of *Triatoma infestans.* Journal of Ultrastructure Research, 57:1-9.
- Chapman, R.F. 1998. A Textbook of the Insects: Structure and

Function IV Cambridge University Press U.K.

- Dadd, R.H. 1970. Digestion in insects in Chemical Zoology (Ed by Florrin m and Secheer B. T) Vol 5, Arthropoda Part! pp. 117-145Academic Press Newyork.
- Gilmour, D. 1961. The Biochemistry of Insects.Acad.Press. New York.Gongalves,
- Gullan PG, Cranston,PS. 2010. The Insects. An Outline of Entomology. Hoboken NJ: Wiley-Blackwell. p 584.
- Hecker, H. 1977. Structure and function of midgut epithelial cells in Culicidae mosquitoes (Insecta, Diptera). *Cell Tissue Research*, 184: 321-332.
- Lehane, M.J. (1998). The midgut. In FW Harrison, MLocke, eds.Microscopic anatomy of invertebrates.vol.11B, Insecta.New York:Wiley-Liss, 725-746.
- Lewis, H.C.1926 The alimentary canal of Passalus.OhioJour. Sci 26: 11 24.
- Lopez-Guerrero, Y. 2002. Anatomy and Histology of digestive system of *Cephalodesmes armige*r Westwood (Coleoptera, Scarabaeinae). *The Coleopterists Bulletin*, 56: 97-106.
- Malagoli D, Abdalla FC, Cao Y, Feng Q, Fujisaki K, Gregorc A, Matsuo T, Nezis IP, Papassideri IS, Sass M, Silva-Zacarin ECM, Tettamanti G, Umemiya-Shirafugi R. 2010. Autophagy and its physiological relevance in arthropods. *Current knowledge and perspectives. Autophagy*, 6:575–588.
- Miller, A. 1961 The mouth parts and digestive tract of adult dung beetles with reference to ingestion of Helminth eggs. *Journal Parasitology*, 47(5):735 –744.

- Mukherji S. P. and Singh, C.B. 1973. The structure of alimentary canal of *Sitophilus oryzae* Linn. (Curculionidae: Coleoptera). *Indian Journal of Zoology*, 12: (12) 94 – 102.
- Nancy Taha Mohamed, Mohamed Salah Mohamed and Doaa Hassan Abdel-Salam (2015). Ultrastructure of the mid-gut of the third instar larvae of Sarcophaga aegyptiaca (Diptera: Sarcophagidae). International Journal of Applied Biology and **Pharmaceutical** Technology Page: xx Available online at www.ijabpt.com.
- Nardi James B and Charles Mark Bee 2012. Regenerative Cells and the Architecture of Beetle Midgut Epithelia. *Journal of Morphology*, 273:1010–1020 (2012).
- Nishiitsutsuji-Uwo, J., and E. Yasuhisa. 1981. Gut endocrine cells in insects: the ultrastructure of the endocrine cells in the cockroach midgut. *Biomedical Research*, 2: 30-39.
- Novikoff, B. A. 1983. Estructura y dina'micacelular.Interamericana ,Me' xico, D. F.Pradhan, S. 1939. The alimentary canal and proepithelial regeneration in *Coccinella punctata* with a comparison of carnivorous and herbivorous Coccinellids.Quar. *Journal Microscopy, Science*. 81: 451 – 478.
- Reinhardt, C. A. (1976). Ultrastructural comparison of the midgut epithelia of fleas with different feeding behavior pattern (Xenopsylla cheopis, *Echidnophaga gallinacea*, *Tunga penetrans*, Siphonaptera, Pulicidae). *Acta Tropica*, 33 (2): 105-32
- Schneider, I. and Rudinsky, J.A.1969. Anatomical and histological changes in the internal organs of adult *Trypodendron*

lineatum, *Gnathotrichu sretusus* and G. Sulcatus (Coleoptera: Scolytidae). *Annuals of. Entomological society of America*, 62: 995–1003.

- Shinoda, O. 1930. Contribution to the knowledge of intestinal secretion in insects IV A. A preliminary note. Kyoto Imp. Univ. Anniversary Vol. Dedicated to Maumi Chikashige pp 9 – 24.
- Slansky, F.1982. Insect nutrition: An adaptation is perspective. *Florida Entomology*, 65: 45 71.
- Snodgras, R.E. 1935 Principles of Insect Morphology PP. 347 – 388 McGraw Hill New York.
- Silva, C.P, Silva J.R., Vasconcelos, F.F., Petrestski, M.D., Damatta, R.A., Ribeiro, A.F., & Stäubli, W., T.A Freyvogel, and J. Suter (1966). Structural Modification of the Endoplasmic Reticulum of Midgut Epithelial Cells of Mosquitoes in Relation to Blood Intake. Journal Microscopy, vol. 5, 189.
- Terra WR. 1990. Evolution of digestive systems of insects. Annu Rev Entomol 35:181-200. Von Dehn M. 1933. Untersuchungen u¨ ber die Bildung der Peritrophischen Membran bei den Insecten. Zeitschrift fu" r Zellforschung und Mikroskopische Anatomie 19: 79–105.

- Terra, W.R. 2004. Occurrence of Midgut Perimicrovillar Membrane in Paraneopteran Insect Orders with Comments on Their Function and Evolutionary Significance. *Arthropod Structure and Development*,33(2): 139-148.
- Talbot, M. 1928. The structure and digestive system in *Creophillus* maxillosus (Coleoptera). Ohio Journal of Science, 28: 261 – 266.
- Von Dehn M. 1933. Untersuchungen u["] ber die Bildung der Peritrophischen Membran bei den Insecten. Zeitschrift fu["] r Zellforschung und Mikroskopische Anatomie, 19:79–105.
- Wigglesworth, V.B. 1972. The principles of insect physiology 7th Ed. New York, N.Y. John Wiley and Sons. Inc. 827 p p.
- Waterhouse, D.F. 1952a. Studies on the digestion of wool by insects. IV. Absorption and elimination of metals by Lepidopterous larvae, with special reference to the Clothes Moth, Tineola *bisselliella* (Humm.). *Australian Journal of Scientific. Research*, B 5: 143 – 168.
- Wigglesworth, V.B. 1965. The principles of Insect physiology. Dutton, New york, PP 427 – 497
- Zumpt, F., (1965): Myiasis in man and animals in the old-world. Butter Worths, London, XV: 267 pp.

57