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Morphological and Structural Observation on the Rectum of *Bolua turkiyae* Ünal, 1999 (Orthoptera, Tettigoniidae)

Damla AMUTKAN MUTLU*[1], Irmak POLAT[1], Zekiye SULUDERE[1]

Abstract

This study presents the morphology and structure of the rectum in *Bolua turkiyae* Ünal, 1999 (Orthoptera, Tettigoniidae) using light and scanning electron microscopy. The digestive system in insects is split up three parts as the foregut, the midgut and the hindgut. The rectum is last part of the hindgut. The results show that the rectum of *B. turkiyae* consists of 6 rectal pads, in similar to rectal pads found in others Orthopteran species. The rectum contains the muscle tissue with connective tissue, epithelial layer with columnar cells, and the cuticular intima from the outside to inside. There is microvillus on the apical side of the columnar cells that serves in water and ion reabsorption.

Keywords: Rectal pads, hindgut, histology, electron microscope

1. INTRODUCTION

The Orthoptera order is split up two suborders as Caelifera in which the species have small antenna and Ensifera in which the species have long antenna [1, 2]. Orthopterans have an incomplete metamorphosis [3]. Most species lay their eggs in the ground or on the parts of plants. The eggs hatch and the young nymphs occur. The young nymphs become the adult individuals after almost 5 moulting stages [3].

Many species in Orthoptera order are considered pests of crops, which these species are known for wiping fields out in a day [3]. Many adult and nymph individuals can cause major agricultural damage by virtue of feeding on weeds, grasses or crops [3, 4]. Therefore, the ortopteran species have pretty economic importance. Knowing the biology of these species is of great importance for the steps to be taken in the pest management.

Bolua turkiyae Ünal, 1999 is a species which belongs to Tettigoniidae family in Orthoptera order in Ensifera suborder. This species is an endemic species to Turkey [5]. Since this species was found in Bolu province for the first time, Ünal 1999 gave the name of this city to it. Type locality of the species is expressed as Bolu, Kastamonu, Balıkesir [5-7].

Many studies have been conducted on the phylogeny, taxonomy and systematics of this species [5-7]. According to our research, no studies on the biology of *B. turkiyae* have been found. Therefore, in the present study, we focused on to find out the morphology and histology of the

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rectum region located in the last part of the digestive canal. Major function of the rectum in especially terrestrial insects is the removal of water from the faeces and provides an absorptive role.

2. MATERIALS AND METHODS

2.1. Insect Collecting and Rearing

Approximately 10 live male and 10 live female adult species of *B. turkiyae* were captured in Hamidiye Village, Bolu Province in Turkey. After being caught, they were put in plastic containers as live.

2.2. Light Microscopy

Live individuals were anesthetized with ethyl acetate fume and the digestive system was dissected out in phosphate buffered saline under a stereomicroscope (Leica EZ4). After taking general photograph of the digestive system, the rectum was removed from the digestive system and prepared for light (LM) and scanning electron microscopy (SEM) studies.

The samples were fixed in Formaldehyde solution. Afterwards, the dehydration was performed in rising series of the alcohol and the samples were embedded in paraffin. the sections which were taken almost 6-7 micron thick were stained with Hematoxylin-Eosin (H&E) and Mallory trichrome staining. Finally, The stained sections were turned into permanent slides with entellan and observed using an Olympus BX51 light microscope (Japan) mounted with a digital camera Olympus E330 (Japan) and the photographs were taken.

2.3. Scanning Electron Microscopy

Dissected rectums were fixed in glutaraldehyde in phosphate-buffered saline. The fixed specimens were dehydrated through a rising ethanol series. Then the drying process was performed with a Polaron CPD 7501 Critical Point Dryer. The coated samples with gold (Polaron SC 502) were investigated with a JEOL JSM 6060 LV SEM at an accelerating voltage of 10 kV and the photographs were taken.

3. RESULTS

The digestive system of adult *B. turkiyae* mainly consists of three distinct regions. These are the foregut, the midgut, and the hindgut. The hindgut is split up regions as the ileum, the colon and the rectum. The rectum which is the subject of the study is the last part of the hindgut in *B. turkiyae.* The rectum is an organ which has ovoid form (Figures 1, 2).

The rectum of *B. turkiyae* has different layers from outside to inside. The outer surface of this organ is covered with the muscle tissue and connective tissue with prominent tracheal network (Figures 2-4). It is observed that it has 6 rectal pads in the cross sections of light microscope (Figure 3). The epithelial layer shows a monolayer sequence (Figures 3-7). The nucleus of each columnar cell is located in the center of the cell and has ovoid shape (Figures 4, 6). The apical side of the cells has a large amount of microvillus which plays the reabsorption function (Figures 5, 7). The epithelium has a cuticular intima layer that separates the lumen content with epithelial layer. This cuticular intima surrounds the epithelium on the luminal side (Figures 3-7).

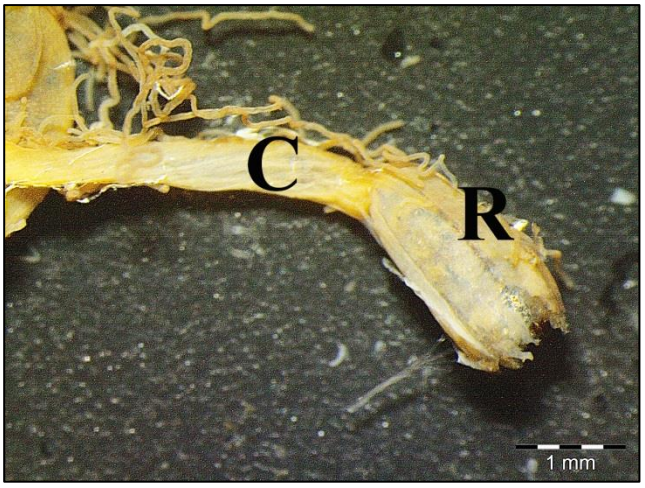

Figure 1 Stereomicroscope images of the hindgut of *B. turkiyae*. C: Colon, R: Rectum

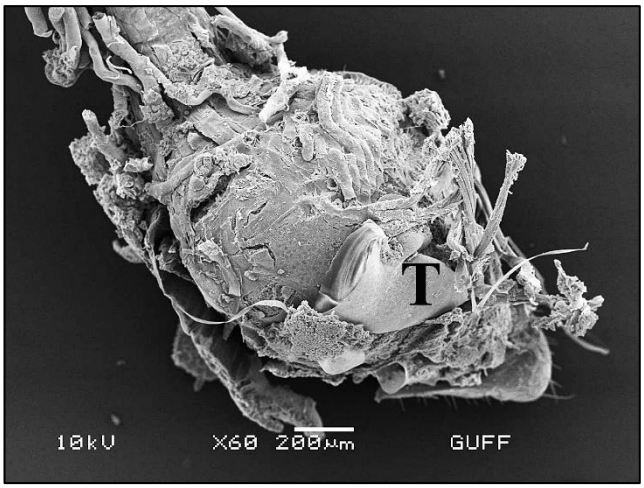

Figure 2 SEM micrograph of the general structure of the rectum of *B. turkiyae*. T: Trachea

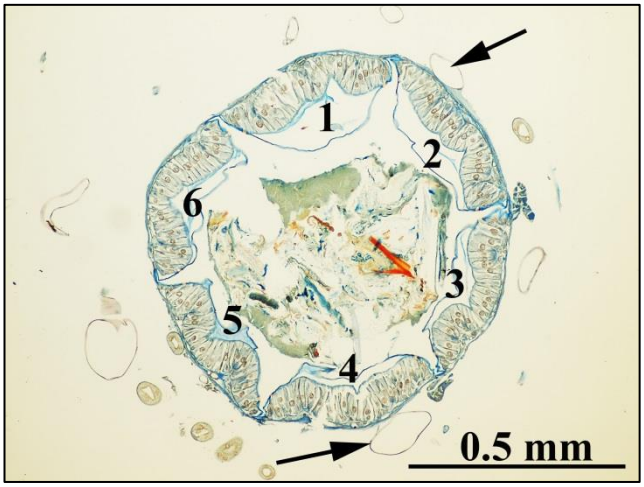

Figure 3 Light micrograph of the cross section of the rectum. →: Trachea (Mallory's trichrome staining)

Figure 4 Light micrograph of the cross section of the rectum. →: Trachea, E: Epithelial layer (H&E staining)

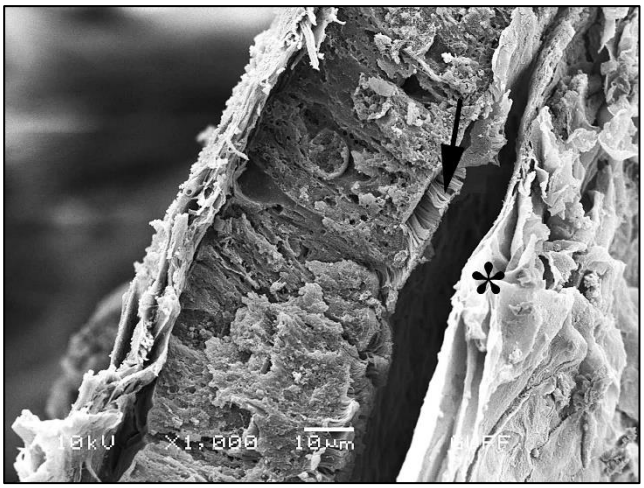

Figure 5 SEM micrograph of the cross section of the rectum. →: Microvillus, *: Cuticular intima layer

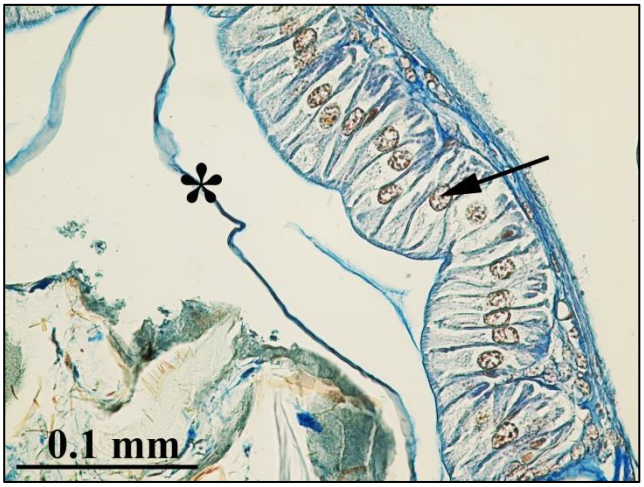

Figure 6 Light micrograph of the cross section of the rectum. →: Nucleus, *: Cuticular intima layer (Mallory's trichrome staining)

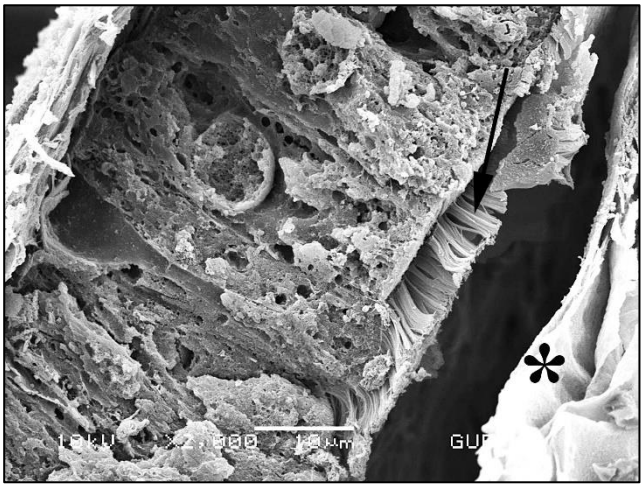

Figure 7 SEM micrograph of the cross section of the rectum. →: Microvillus, *: Cuticular intima layer

4. DISCUSSION

The rectal pads that are founded in some insect species are specialized structures of the rectum. These structures are appertaining to ion uptake and water removal to continue the body homeostasis [8, 9]. There is no rectal pad in rectum in some insect species such as *Pylaemenes mitratus* (Phasmid: Basillidae) [10] and *Karenia caelatata* (Hemiptera: Cicadidae) [11]. On the contrary, there is found the rectal pad in the rectum of *Streltzoviella insularis* (Lepidoptera: Cossidae) [12], *Cryptotermes brevis* (Blattodea, Kalotermitidae), *Cryptotermes cavifrons* (Blattodea, Kalotermitidae), *Coptotermes formosanus* (Blattodea, Rhinotermitidae), *Neotermes jouteli* (Blattodea, Kalotermitidae) [13], *Gryllus pennsylvanicus* (Orthoptera, Gryllidae) [14], and *Poecilimon cervus* (Orthoptera, Tettigoniidae) [15] similar to the rectum of *B. turkiyae*. The presence or not of rectal pads may be related to the insect's diet.

The rectal pads number can vary among the insect species. Although the number of the rectal pads is 6 in *G. pennsylvanicus* (Orthoptera, Gryllidae) [14], *Blattella germanica* (Orthoptera, Blattellidae) [16], and *Aedes aegypti* (Diptera, Culicidae) [17], this number is 4 in *Chrysoperla externa* (Neuroptera, Chrysopidae) [18]. The rectum of *B. turkiyae* has 6 rectal pads.

The epithelial cells have microvillus on the apical side of it. These microvilli help to reabsorption of the water, on the other hand it was indicated that the epithelial cells in the rectum of *Rhynchophorus phoenicis* (Coleoptera, Curculionidae) [19] and *P. cervus* (Orthoptera, Tettigoniidae) [15] do not have microvilli. It was remarked that the cells have either apical membrane infoldings or epithelial foldings. Contrary, in *Calliphora erythrocephala* (Diptera, Calliphoridae) [20] and *Melanoplus femurrubrum* (Orthoptera, Acrididae) [21] they have well developed microvilli in rectal cells.

In this paper, we focused to determine the morphology and structure of the rectum. Although the data we have obtained reveal some differences, they are mostly similar to the rectum structure of other insect species.

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The Declaration of Conflict of Interest

No conflict of interest has been declared by the authors.

Authors' Contribution

The authors contributed equally to the study.

The Declaration of Ethics Committee Approval

This study does not require ethics committee permission or any special permission.

The Declaration of Research and Publication Ethics

The authors of the paper declare that they comply with the scientific, ethical and quotation rules of SAUJS in all processes of the paper and that they do not make any falsification on the data collected. In addition, they declare that Sakarya University Journal of Science and its editorial board have no responsibility for any ethical violations that may be encountered, and that this study has not been evaluated in any academic publication environment other than Sakarya University Journal of Science.

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