Commun. Fac. Sci. Univ. Ank. Serie C
V. 6. pp. 199-204 (1988)

SURFACE ULTRASTRUCTURE OF THE LARVAL HEMOCYTES OF TURNIP MOTH AGROTIS SEGETUM DENIS AND SHIFF. LEPIDOPTERA: NOCTUIDAE

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ABSTRACT

Hemocytes of *Agrotis segetum* larvae were studied by scanning electron microscope. In this study, five types of hemocytes have been recognized: Prohemocyte, plasmatocyte, granulocyte, spherulocyte and oenocytoid. The prohemocytes are small, round cells and have less number of cyloplasmic projections. The plasmatocytes show a spindle shape and have several small processes. The granulocytes are round in shape and posses numberous projections. The sperulocytes are characterized by spherul like inclusions. The oenocytoids are also round but slightly compressed on surfaces.

INTRODUCTION

The lepidopterous insect blood cells are grouped into six main classes by the autors (JONES, 1962; GUPTA, 1979; RATCLIFFE, and ROWLEY, 1979; ARNOLD, 1982; ROWLEY, and RATCLIFFE, 1981): Prohemocytes, plasmatocytes, granulocytes, spherule cells, adipohemocytes and oenocytoids.

There are very few works done on the surface structure of hemocytes of insects (AKAI, and SATO, 1976; AKAI, and SATO, 1979; OLSON, and CARLSON, 1974).

The circulating hemocytes of *Agrotis ipsilon* and *A. segetum* were classified into five types according to their structures by light and electron microscobic observations (AYVALI, 1987; KAROL et. al., 1987).

It is very clear that the study of the hemocytes structure by different way of observations should provide more details. The scanning electron microscobic observation has been chosen as one of the techniques for this purpose.

MATERIAL AND METHODS

The larvae were reared in a culture room at 25–27°C. For observations of hemocytes with the scanning electron microscope, a hind leg of larva was cut. The drops of hemolymph were collected in the buffered % 2.5 glutaraldehide in a centrifuge tube and fixed for two hours in cold condition. After the first fixation, the cells were centrifuged at 600 rpm for five minutes and then pellet was washed in several changes of sucrose solution (buffered at pH: 7.2). The pellet was postfixed with 1 % osmium tetroxide in graded aceton series and then transferred to aceton in critical–point drying apparatus. Liquid CO_2 flushed the acetone from the cells. Following the critical–point drying, the specimens were coated with gold in a coating unite. (SALSBURY, and CLARKE, 1967). After then the coated samples were observed with scanning electron microscobic attachment of JEOL 100 CX II at 20kV.

RESULTS

When the centrifuged blood cells were observed in the scanning electron microscope, their surface structures were seen very clear at low magnification (Fig. 1). The larval hemocyte types were easily identified by our earlier knowledges.

The prohemocytes are usually round in shape and small, 4–5 μm in diameter. There are a few but thiny projections on the cell surface (Fig. 2).

The plasmatocytes are very typical and spindle–shaped. They are clearly distinguishable from other hemocytes by their typical shapes. The plasmatocytes are mostly spindle shaped. Their surface have several small projections (Fig. 3,4). The number of projection should probably be related to their hemocytic activities.

The granular hemocytes are clearly distinquishable by their numerous cytoplasmic processes (Fig. 3). This type of cells are mostly spherical in shape.

The spherule cells have characteristic surface structures. They are usually spherical and have numerous spherules which appear as bulbous swelling on the cell surface (Fig. 5). These spherular inclusions formed ondulated surface pattern for this type of hemocytes.

The oenocytoids are the largest and disc–shaped cells (Fig. 6). They have mostly smooth surfaces. Some of them may posses a few

and small projections. From lateral and dorsal view, these hemocytes are slightly biconcave, but hemispherically rimmed in the central region.

DISCUSSION

In this study, using the scanning electron microscope, the surface of ultrastructures of hemocytes of *A. segetum* were studied. In the circulating hemolymph of this species, five types of hemocytes were observed. For the identification, the earlier studies were utilized that was carried out by the transmission electron microscope.

The scanning electron microscope studies supported the classification and the identification of the hemocytes made by Akai and Sato (1976). In general, surface ultrastructure of hemocytes of *A. segetum* clearly resemble to that of *B. mori* with the exception of oenocytoids which are different from that of the oenocytoids of *B. mori* studied by Akai and Sato (1976).

Akai and Sato (1979) had studied the hemocytes of only one species from the five insect orders. When we compared the hemocytes of *A. segetum* with other insect blood cells, there are considerably differences and similarities among them.

The surface of the cells named by Akai and Sato (1979) as prohemocytes of larval *B. mori* (Lepidoptera), adult *Panesthia angustipennis* (Dictyoptera) and *Locusta migratoria* (Orthoptera) are very similiar in shape and size with *A. segetum.*

The so called plasmatocytes of *B. mori*, L. *migratoria* and *Holotrichia kiotoensis* (Coleoptera) are also similiar by their spindle shapes with the plasmatocytes of *A. segetum.* The plasmatocytes of *L. migratoria* have very smooth surface and swollen in shape, while in *B. mori* and *A. segetum* they have rough surface and leaf-like in shape. The plasmatocytes of *P. angustipennis* and *H. kiotoensis* are very different in shape from other three insect hemocytes. The plasmatocytes of *P. angustipennis* and *H. kiotoensis* are round and flat whereas the others are spindle shaped as was shown by Akai and Sato (1979).

The cell surface of granulocytes has the most similiar structure in these five insect species as well as in *A. segetum* being spherical shape and having numerous cytoplasmic processes. The spherule cells of

B. mori and *A. segetum* appear very similiar in shape. These cell types of two species are different from spherulocytes of *P. angustipennis* and *H. kiotoensis* of Akai and Sato (1976).

The oenocytoids of *A. segetum* are dissimilar with the same cell types of *B. mori* and *L. migratoria*. The oenocytoids of *B. mori* are spherical and those of *L. migratoria* are leaf–shaped, while the ones of *A. segetum* are slightly compressed in shape and have less numerous cytoplasmic processes.

The electron micrographs of the study of Olson and Carlson (1974) were not sufficient to use for the hemocyte comparison for this work.

Finally, we must point out that the hemocytes of *B. mori* and *A. segetum* have very close relation in the ultrastructural cell surface excluding the oenocytoids. In addition to the recognizable hemocytes, there some cells the identifications of which are very difficult to make. These unidentified cells might be the intermediate types. This kind of cells has also been encountered in TEM as well as in Phase–Contrast microscope by Ayvalı (1987), Shapiro (1979).

ACKNOWLEDGEMENT

We are grateful to Prof. Dr. Sevinç KAROL for her encouragement to carry out this work in The Unite of Electron Microscope as well as her kind help for the evaluation of our results. We are also, thankful to Doç. Dr. Zekiye SULUDERE for her assistance with the critical point drying apparatus.

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FIGURES

Fig. 1. The low magnification of the pellet of hemocytes. Pl. plasmatocytes, Pr. prohemocyte. Gr. granulocyte, s. spherul cell, o. Oenocytoid. X 2000.

Fig. 2. Prohemocyte (Pr), Oenocytoid (O), Granulocyte (Gr) and Spherul cell. X 5000.

Fig. 3. Granulocyte. X 5000

Fig. 4. Plasmatocyte. X 5000

Fig. 5. Sherule cell. X 5000.

Fig. 6. Oenocytoid. X 5000.