



HISTOMORPHOLOGICAL STUDIES DURING VITELLOGENESIS IN AQUATIC BEETLE CYBISTER TRIPUNCTATUS OL. (COLEOPTERA: DYTISCIDAE)

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ABSTRACT:

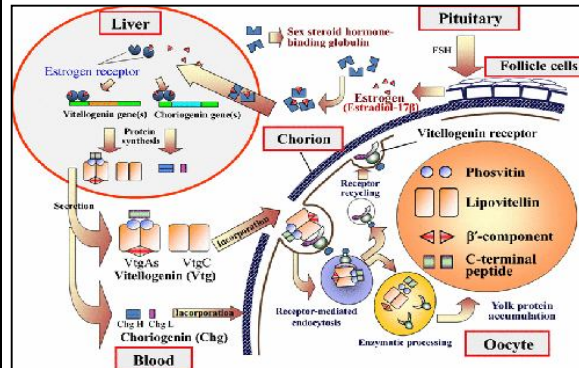
In the newly emerged females. The ovaries are small threadlike structures measuring about 20.00 ± 2.00 mg in weight. The follicle is filled with cytocysts and the oocytes are undifferentiated. In two-day-old females, differentiation of nurse cells and oocyte become distinct. Most of the region of follicle is occupied by the nurse cells, and the oocyte is very small, lying ventrally. The oocyte bears centrally placed large germinal vesicle. The cytoplasm of oocyte is granular.

KEYWORDS :

Vitellogenesis, Nurse cells.

INTRODUCTION:

The nurse cells are large and their nuclei are lobulated. The nurse cells measure about 96.10 ± 0.48 μ m in diameter. They discharge secretory material into the oocyte through the radial canals. The follicular



epithelium of the previtellogenic oocyte is composed of squamous epithelial cells. They possess large spherical nuclei at the centre measuring about 8.10 ± 0.5 μ m in diameter. The pre-vitellogenic oocyte grows up to $201 \pm \mu$ m in length. The transport of secretory material from the nurse cells to the previtellogenic oocytes is well evident. The previtellogenic oocytes are filled with the granular cytoplasmic inclusion. In 3-day-old females the pre-vitellogenic oocytes further grow up to about 255.0 ± 25 μ m in length and 20 ± 2 μ m in diameter respectively. Along with the oocyte, the follicular epithelial cells along with their

nuclei increase in size. The nucleoli in the nuclei of follicular cells are very prominent. The chromatin material of the nuclei of the nurse cells is dispersed and granulated. The previtellogenic oocytes are devoid of yolk bodies.

1. MATERIAL AND METHODS

1. MATERIAL
The present work is carried out on the aquatic beetle, *Cybister tripunctatus* OL.

1.1 CLASSIFICATION
(Richards and Davis 1977)

1.2 Systematic position of the aquatic beetle, *Cybister tripunctatus*

OL is given below.

- Class – Insecta
- Subclass – Pterygota
- Division – Endopterygota
- Order – Coleoptera
- Suborder – Adephaga
- Family – Dytiscidae
- Genus – *Cybister*
- Species – *tripunctatus* (OL)

CHARACTERS 1.2.1] In aquatic beetles *Cybister tripunctatus* OL, sexes are separate and sexual dimorphism is well marked as the forelegs of male beetles show presence of adhesive pads, while such structures are absent in the females.

1.2.2] It possesses filiform antennae.

1.2.3] Hind legs are notatorial, functioning as swimming organs, flattened and fringed with large hairs.

1.2.4] Larvae are with long sickle-shaped mandibles.

- 1.2.5] Last two abdominal segments along with abdominal lobes are fringed with hairs.
- 1.2.6] Elytra store air beneath them. It is the source of oxygen which is supplied to tracheal system by last two pairs of abdominal spiracles, during diving in deep water.

1.3 SELECTION

1.4 Aquatic beetle, *Cybister tripunctatus*. OL, is selected for the present work because of the following reasons-

- 1.3.1] It is easily available, and commonly found in local ponds in all seasons.
- 1.3.2] It can easily be collected by fishing nets or hand nets in ample quantity.
- 1.3.3] It can be acclimatized under the laboratory conditions for a long duration due to their sturdy nature and their quick adaptability to new environment.
- 1.3.4] It can be maintained by feeding small fishes and crustaceans.
- 1.3.5] It is of convenient size to handle and easy for experimentation.
- SOURCE The aquatic carnivorous beetles were collected from the ponds located Pavani, Dist. Bhandara (MS). The beetles were reared in laboratory throughout the year to carry out the present studies.

1.5 REARING

The larvae and adult beetles were kept in well aerated aquarium in the laboratory. The muddy water and small stones having crevices were kept in aquarium to maintain natural condition. The small fishes were kept as a food of the beetles. The stones were kept to provide place for egg laying. Some

times, they lay the eggs on the inner side of the wall of aquarium also. The aquarium was covered to prevent escaping of beetles from the aquarium. The fresh water was added for sufficient supply of oxygen. The larvae and beetles were acclimatized and reared in laboratory under normal condition of photoperiod 12L : 12 D and 24°C temperature. The mating occurred mostly during daytime. The mated female laid eggs in a capsule like case which hatched within 3-4 days depending upon environmental conditions. The food was supplied once every day. The first instar larvae underwent two moults. The well developed third instar larvae were transferred into another aquarium. The larvae lastly constructed the pupal chambers in a soil. Newly emerged adults of both sexes were separated and kept into individual glass jars. The date and time of emergence of the adult beetles were recorded.

2. METHODS

2.1 DISSECTION, FIXATION AND SECTIONING

The female reproductive organs dissected in insect Ringer's solution under stereoscopic binocular microscope. The organs were fixed in

OBSERVATION:

1. Oocyte Development and Vitellogenesis

The terminal oocyte undergo development periodically. Repeated cycles of oocyte development and subsequent cycles of oviposition occur in the adult female *Cybister tripunctatus*. Development of the terminal oocyte shows consecutive stages of vitellogenesis. During development the terminal follicles show remarkable changes in the oocyte shape, size, cytological structure, deposition of yolk material and formation of egg-membranes along with the cytomorphological

change in the Trophocytes and follicular epithelium. The entire process of vitellogenesis can be divided into following five stages:

- 1) Pre-vitellogenic;
- 2) Early-vitellogenic;
- 3) Mid-vitellogenic;
- 4) Late-vitellogenic and
- 5) Maturation stage.

1.1.1 The-Early vitellogenic stage

In the 4 day old beetles, the ovaries are gradually increased in size and measure about 97.00 ± 9.50 mg in weight. The terminal oocytes are encircled by a double layered follicular epithelium. The terminal oocytes become large and occupy almost half the portion of follicles. Rest of the portion of the follicle is occupied by a group of the nurse cells. The nurse cells are found to be large in size with well differentiated ring canals. The nuclei are lobulated enormously containing granular chromatin material. The transport of secretory material from nurse cells to oocyte occurs predominantly. Accumulation of fine granules is well-evident in the intrafollicular spaces formed within the follicular cells. The follicular cells are fully-packed with granular material. The follicular cells are tall and columnar in shape in the 5 day old beetles. The early vitellogenic oocytes measure about 290 ± 16 μm in length and 60.0 ± 4 μm in diameter while the nurse cells measure about 90.7 ± 6 μm in diameter. The nuclei of follicular cells measure about 7 ± 1 μm in diameter.

1.2.3 Mid-vitellogenic stage

In the 6 day old beetles, the ovaries become large and measure about 227.00 ± 13.00 mg in weight. It is found that the volume of oocyte increases greatly and subsequently, the nurse cells also attain the maximum size. At this stage, the nurse cells become active and the cytoplasmic material flows into the respective oocyte through the intercellular bridges. The terminal oocytes are filled with initially small dense spherical yolk bodies at the periphery.

They measure about 400 ± 23 μm in length and 295.5 ± 29 μm in diameter while the nurse cells are 97.0 ± 11 μm in diameter. The follicular cells are spherical in shape. The nuclei of follicular cells are measured about 11.0 ± 0.69 μm in diameter. In the 7th day old beetle, the terminal oocytes attain the maximum size i.e. 445 ± 22 μm in length and 342 ± 21 μm in diameter and are fully filled with yolk bodies. The nurse cells increase to 103 ± 9 μm in diameter and the nuclei of follicular cells to about $14 \pm$ standard error μm in diameter. At this stage the follicular epithelial cells are full of RNA contents.

1.1.2 The late- vitellogenic stage

In the 8 day old beetles, the ovaries increase in size and measure about 340.00 ± 20.00 mg in weight. The nurse cells undergo degeneration and are reduced in size greatly. The follicular epithelial cells become squamous and filled with large quantity of cytoplasmic inclusions. The yolk bodies occupy whole substance of the terminal oocytes. The follicular epithelial cells secrete globular chorion bodies in the form of fine membranous vesicles. The size of the late vitellogenic oocyte increases i.e. 663.5 ± 14.5 μm in diameter. The nurse cells decrease in size. The follicular nuclei measure about 15.2 ± 1.25 μm in diameter. The formation of vitelline membrane and the chorion is initiated.

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