

ZANIOLO G., MANNI L. and BURIGHEL P. (Department of Biology, University of Padua): *Test cells during embryogenesis in Botryllus schlosseri*.

In the ovoviviparous ascidian *Botryllus schlosseri* the full-grown oocyte is surrounded by the outer and inner follicle cells, the acellular vitelline coat and test cells, and is provided with its own vesicular oviduct (ZANIOLO *et al.*, *Can. J. Zool.*, 65, 1181-1190, 1987). At ovulation, the egg moves through the oviduct, while the outer follicle cells are discharged in the mantle. The inner follicle cells, together with the oviductal and atrial epithelium, form the cuplike « placenta » that hangs the embryo onto the atrial wall.

We have investigated the role of egg envelopes during embryogenesis, with particular attention to the differentiation and fate of test cells. Some observations on these cells during oogenesis have also been reported in a previous paper, in which the early appearance, progressive encasement in the oocyte, and the formation of electrodense granules in the test cells were studied (ZANIOLO *et al.*, *Acta Embryol. Morphol. Exper.*, 10, 229-236, 1989).

After ovulation and during embryogenesis, the test cells undergo modifications and interact with the embryo surface, showing a peculiar role in larval tunic formation.

During cleavage, test cells are discharged into the perivitelline space, and show a large, well-developed Golgi apparatus formed of many elongated and flattened cisternae; electrodense granules of different sizes are present in the test cell cytoplasm. The granules probably derive from the Golgi cisternae which characterize the test cells during embryogenesis: they are strongly electrodense, surrounded by the membrane, and seem to be progressively released from the test cells.

Starting from the tail bud stage, when the larval tunic appears around the embryo, many granules ejected from the test cells adhere to the outer border of the larval tunic.

In an advanced stage of development, the tunic shows the test cells at the periphery under the vitelline coat, while the granules seem to be dissolved on the tunic surface. In fact, there appears to be a strong relation between granule material and the electrodense material forming the cuticular layer of the tunic.

It is not currently clear whether the activity of test cells consists of a simple material contribution, or whether they form precursors interacting with the larval tunic matrix to form the cuticular layer.

This aspect is also present in the early tadpole stage, when the definitive tunic appears different from the larval tunic owing to the presence of many papillae on the outer border.

In the late tadpole stage, shortly before hatching, the test cells with scarce or no granules are scattered under the vitelline coat, outside the larval tunic and also outside the definitive tunic, which starts to be colonized by blood cells.