Microanatomical alterations in the gut of an marine polychaete (*Eulalia viridis*, Errantia: Phyllodocidae) during the digestive process

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The marine annelid Eulalia viridis is an opportunist scavenger, sharing characteristics of both detrivore and predator. However, definite data on feeding habits and digestive physiology are absent. In marine invertebrates, the digestive epithelia suffer complex changes during digestion. Still, such changes are far better described in the molluscan digestive gland, especially in cephalopods [1,2], than for polychaetes, regardless of a few descriptions of the digestive tract [3]. The main objective of this work was to identify microscopical changes in the gut epithelia of E. viridis along the digestion. Specimens (collected from SW Portugal), were fixed with glutaraldehyde and embedded in paraffin. Sections (5 µm thick) were stained with a tetrachrome stain for fibres and polysaccharides, some being post-fixed in osmium tetroxide for lipids [1]. At early stages of digestion, the epithelial cells contained many Alcian Blue (AB)-positive bodies, indicating active secretion of mucosubstances (Fig. 1A). As digestion proceeds, AB-positive bodies become scarcer as agglomerates of Periodic Acid-Shiff's (PAS)-positive substances increase in size and quantity, primarily near the basal of the epithelium, indicating reserve of glycogen-like sugars (Fig. 1B). Also, the rough endoplasmic reticulum proliferates, probably due to increased enzyme production [2] (Fig 1C). Excretory vacuoles (stained yellow by Picric Acid) were formed during digestion, being densely packed in specialized cells. Some excretory cells contained vacuoles with a AB-positive membrane (Fig. 1C) These were formed at a later stage and indicated two distinct types of excretory cells. During the final stages of digestion, membraneenclosed corpuscles were formed in the apical region of the digestive cells and released into the lumen of the gut (Fig. 1D). These structures probably share similar functions to cephalopod brown bodies [1,2]. Small lipidic droplets were noticed only in advanced digestion stages (Fig. 1D). Green pigments (for which the coloration of *E.viridis* is owed) were heterogeneous in size and colour density. The pigments were seemingly more concentrated in a more advanced digestion phase. Pigments were found closer to the lumen or to the epithelium depending on early or later digestion stages, respectively.

To summarize, the gut epithelia of *E. viridis* is formed mostly by digestive and excretory cells. The former undergo significant changes during digestive, beginning with secretion of mucosubstances, followed by synthesis of enzymes. The intracellular digestive process occurs in small digestive vacuoles containing amorphous materials that, in a posterior phase, may contain glycogen. However, lipids are scarce. There are no specialized sections of the gut, meaning that changes occur gradually, according to the presence of food items. Specific biochemical changes in epithelial cells need yet further investigation.

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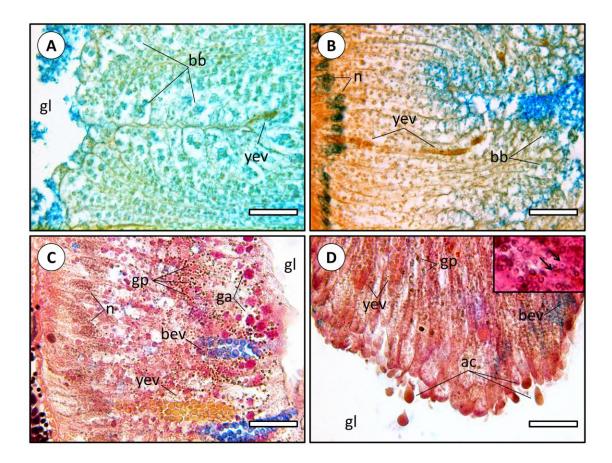


Figure 1. Morphological alterations during digestion phases in *E. viridis* intestinal epithelium (tetrachrome stain). **a** Early digestive phase. Mostly AB-reactive blue bodies (*bb*), indicating mucosubstances were being secreted into the lumen of the gut (*gl*). Few yellow (from Picric Acid) excretory vacuoles (*yev*), in specialized cells, are visible. **b** Subsequent(intermediate) phase. Blue bodies (*bb*) diminished in number and size and PAS-reactive glycogen aggregates (*ga*) began to appear. The nuclei of digestive cells (*n*), notorious near the basal lamina of the epithelium, were strongly stained by Weigert's Haematoxylin. Excretory vesicles (*yev*) were still scarce at this stage, albeit more organized in specialized cells.

- **c** Further along the digestion. The epithelium contained mainly PAS-reactive bodies (ga), with agglomerates of yellow excretory vacuoles, with (bev) or without (yev) a bluish, AB-positive membrane. Naturally coloured green pigments (gp) were notorious closer to the lumen gut.
- **d** Latter phase of digestion. The epithelium appeared more fragmented due to shedding of apical corpuscles (*ac*) being relased into the lumen. There was a higher predominance of *bev* over *yev*. Greenish pigments (*gp*) were observed closer to the epithelium. *Inset* Minute, blackish, lipidic droplets (arrows) were evidenced in osmium-treated samples, counterstained with Nuclear Fast Red. Scale bars: 25 μm.