Comparative Functional Anatomy of the Accessory Respiratory Organ in Neritopsine Gastropods

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ABSTRACT

The mantle cavity in the molluscs is used mainly for respiration. The floor of the mantle cavity, the accessory respiratory organ is thought to play a role in respiration when the nerites are exposed to the atmosphere. The study seeks to examine the structure and function of this accessory respiratory organ in nerites through light microscopy and scanning electron microscopy using histological methods. Specimens from Nerita undata, Nerita lineata and Nerita planospira were investigated. Various staining methods were applied to the sections. The organ consists of three tissues layer: a surface epithelium layer, connective tissues layer and a layer of muscle coat. Two types of tubules were observed in N. undata and N. lineata, namely Type 1 and Type 2 tubules. Dorsally, the epithelium appeared to be folded and comprised of numerous openings and slits. Ventrally, the surface is smooth except that is consists of random appearance of openings. The accessory respiratory organ holds some resemblance to the frog skin. Therefore, implication on the function of the organ is made based on the open model of the amphibian’s gaseous exchange.

INTRODUCTION

It is known that the mantle cavity plays an important role in the molluscs especially in respiration. On examining the mantle cavity, Fretter (1965) observed that the floor of the mantle cavity, the accessory respiratory surface, together with the roof is used as the lung when the neritids is out of water. Therefore studies are conducted on the structure of the accessory respiratory organ in Nerita through light microscope and scanning electron microscope.

MATERIALS AND METHODS

Specimens were collected from the coast and mangroves of St John’s Island and from Fort Road Monsoon drain. After preserving in 4% formaldehyde in seawater, histological methods were carried out and sections were stained with Mayer’s haematoxylin and eosin; rapid phosphotungstic acid-hematoxylin (PTAH); Periodic Acid-Schiff reaction (PAS) and Alcian Blue-PAS before viewing under light microscope. The surface was studied both under the light microscope with and without staining in toluidine blue, and through scanning electron microscope.

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RESULTS

Gross anatomy
The accessory respiratory organ lies dorsal to the buccal mass and ventral to the gills and mantle skirt. The organ with a dimension that average to 0.8 cm in length x 0.7 cm in width x 30 µm in thickness, spreads over the floor of the mantle cavity. It can be distinguished from surrounding tissues through the coloration.

Microanatomy
Light microscopy
The structure of the accessory respiratory organ consists of the surface epithelium layer, the connective tissues layer and the muscle coat layer whereas tissues surrounding the organ have an epithelium layer and a muscle coat layer (figure 1). Structure of the organ in *N. lineata* and *N. undata* was as shown in Figure 1A. The muscle coat layer was thicker in *N. planospira* and the middle layer, the connective tissue was thinner compared to the other two species. The structure was haematoxylin and eosin positive; PTAH positive; PAS positive and Alcian Blue-PAS positive.

![Figure 1](image)

Figure 1. *Nerita undata*. General structure of the transverse section of the floor of the mantle cavity: (A) the accessory respiratory organ, and (B) tissues immediately adjacent to the accessory respiratory organ. The tissues are stained with PAS-Alcian blue. Scale bar = 5 µm. cb, cutaneous blood space; ct, connective tissues; ep, epithelium; lm, longitudinal muscles; om, oblique muscles; sl, subepithelial longitudinal muscles; tm, transverse muscles

Two types of vascular tubules was observed from the surface of the accessory respiratory organ. The first type (Type 1) appeared to originate from the left of the accessory respiratory organ from a single large blood vessel, while the second type (Type 2) were free-standing tubules that formed a network between the spaces created by the
first. The openings, whose occurrence on the surface appear random, were only associated with Type 2 tubules.

Scanning electron microscopy

The dorsal surface of the accessory organ in *N. lineata* and *N. undata* showed that the surface is not smooth; the epithelium appeared to be extensively folded and comprised of numerous openings and slits. In *N. planospira*, the surface was bulged. On contrary, the ventral surface in *N. lineata* and *N. undata* was smooth with random appearance of holes but this is not observed in *N. planospira*.

**DISCUSSION**

The structures of the floor of the mantle cavity of the three species of *Nerita* observed in this study generally concur with Fretter’s (1965). However, two distinct types of tubules observed were not referred to by Fretter (1965). Fretter (1965) only mentioned that the floor of the mantle cavity receives blood supply through the few vessels from the anterior aorta. Based on the ventral surface from SEM and light microscopy, it is suggested that blood is supplied to the mantle cavity floor through Type 1 tubules and distributed to the rest of the mantle cavity floor through the branching of the main vein. The blood is then returned to the general circulation through Type 2 tubules and drained into the body cavity through numerous openings through the muscle layer at the base of the accessory respiratory organ. In paraffin sections of the accessory respiratory organ, traces of vessels can be seen in the cutaneous blood space (cb) corresponding to the presence of Type 1 and Type 2 tubules.

Based on light microscopy observations of the accessory respiratory organ, the principal structures comprise the epithelium, smooth muscle, connective tissue which is formed from elastin fibers, and spherules. Smooth muscle works to force blood away from the organ (Fretter, 1965) and in the formation of elastic fibers (Ross, 1971). Elastin fibers provide elasticity and flexibility for contraction and bending of the muscles. Spherules in connective tissues contain carbohydrates and proteins. It is involved in the production of food reserves in times of need (Fretter, 1965). This could also suggest a reason for closeness of the spherules to the muscles. The food reserves and the oxygen from the environment provide immediate sources for energy formation so that the muscles are able to work without a need to wait for transport of substances from other parts of tissue.

*N. planospira* shows some structure differences from *N. lineata* and *N. undata* although both *N. planospira* and *N. lineata* are found in the same mangrove habitat. Phylogenetic could explain these differences in the structure of *N. planospira*. *N. planospira* appeared to have a basal position in the phylogenetic reconstruction than both *N. lineata* and *N. undata* suggested by molecular analysis (Frey and Vermeij, 2008). Hence, this gives rise to the closer relationships of *N. lineata* and *N. undata* to the freshwater molluscs and indirectly to terrestrial molluscs compared to *N. planospira* in that *N. lineata* and *N. undata* have better developed lungs. One part of the mantle cavity in freshwater molluscs possesses gills and another portion of it is turned into a gas-filled lung (Covich, 2000). Ruthensteiner’s (1997) work showed that the lung in the ellobiid *Ovatella myosotis* is homologous to the mantle cavity in prosobranchs and
opisthobranchs. This forms a continuum on development of gills to gills and lungs to lungs from marine to terrestrial environment. Therefore, the differences seen.

The nerites live in an ecological environment similar to that of the amphibians and the structure of the skin holds some resemblance to the accessory respiratory organ. The skin of the frog consists of an epidermis composed of several layers of epithelial cells and an underlying dermis containing blood vessels, glands and connective tissues. The nerites and frogs are similar in that the blood vessels in both organisms are in close proximity to the medium for gaseous exchange. However, frogs lack the muscle coat that is present in nerites. In amphibians, gaseous exchange takes place in the subepidermal capillary network (open model) (Piiper and Scheid, 1991). *Nerita* could also respire in the same way as the open model in amphibians. During low-tide when the nerites are exposed to the environment, gaseous exchange occurs between the accessory respiratory organ and the space in mantle cavity where oxygen supplies through the pneumostome.

This study does not investigate on the roof of the mantle and the circulatory system. More work is needed to confirm the possibility of the function. Therefore, future research can be done on this area.

REFERENCES