

MICROPUNCTURE STUDIES ON THE NEPHRIDIA OF THE LEECH

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The leech, *Hirudo medicinalis* is able to osmoregulate in fresh water. When starving it maintains an osmotic concentration of the hemolymph around 225 mOsm/kg H₂O. The integuments are permeable to water (as shown previously, Schmidt-Nielsen and Pagel, Bull. MDIBL 8:61-62, 1968), and it therefore must take up water by osmosis. The excretion of water takes place through 17 pairs of nephridia. There is some question in the literature as to whether the nephridia are open to the coelomic fluid through a nephrostome or closed. Each nephridium opens into a bladder, measuring 2-3 mm in diameter when full.

The present study was undertaken to investigate the nephridial function in fed and unfed leeches. The leech was stretched out with pins on a cork plate under paraffin oil. An incision was made along the middle of the dorsal side. The integument was spread out and fastened with pins. The nephridia were carefully exposed by peeling away the intestine. When exposed the blood vessels were still pulsating. The direction of flow in the nephridium was studied by the injection of colored oil or lysamine green. The diagram shows the arrangement and direction of flow in a typical nephridium (Figure 1). Samples of urine were obtained from the bladder and nephridial samples were obtained along the nephridium. The puncture site could easily be identified.

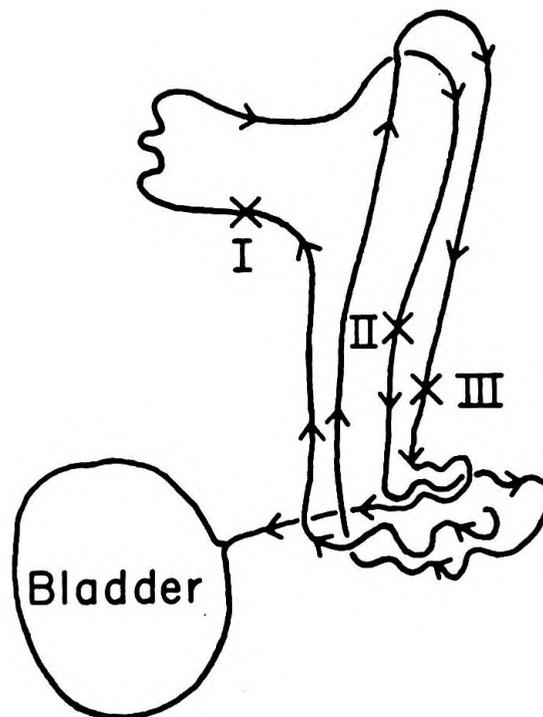


Figure 1. Schematic drawing of a nephridium. The direction of flow of the fluid is shown by arrows. The sites I, II, and III correspond to the puncture sites presented in Table 1.

As can be seen from Table 1, the urine osmolality in starving leeches was very low (27 mOsm/kg H₂O). The osmolality along the nephridium decreased continuously in the direction of the flow. It seemed that all parts of the nephridium contributed to the dilution of the urine. In leeches that were fed the osmotic concentration decreased only slightly along the entire nephridium and was only 27% lower in urine than in hemolymph. Thus, the resorption of solute along the nephridium appears to be regulated according to the need for electrolyte conservation.

Table 1

HEMOLYMPH, NEPHRIDIAL AND URINE OSMOLALITY mOsm/kg H₂O
IN STARVED AND FED LEECHES

| | Hemolymph | I | II | III | Urine |
|---------|--------------|--------------|-------------|--------------|-------------|
| Starved | (6) 224 ± 9 | (5) 182 ± 9 | (7) 150 ± 7 | (2) 70 ± 5 | (8) 27 ± 7 |
| Fed | (3) 282 ± 16 | (3) 262 ± 17 | (3) 224 ± 8 | (3) 189 ± 10 | (3) 182 ± 2 |

Preliminary studies with ¹⁴C inulin showed an inulin urine/hemolymph concentration ratio of unity. This would indicate that the urine is formed by filtration rather than secretion.

Further studies will include the determination of Na and K concentrations in the nephridial fluid.

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LIGHT AND ELECTRON MICROSCOPY OF REGENERATING Tubularia

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Complete understanding of the process of regeneration depends partly upon detailed descriptions of the morphological changes which occur. Descriptions of regeneration in the marine coelenterate Tubularia and other hydroids are of special value for several reasons. Hydroids are relatively simple in structure. Their tissue layers possess well-differentiated cell types. They undergo complex structural changes during regeneration. They are small (1 mm in diameter) and almost the entire cross section of a specimen can be seen in a low power electron micrograph. Unlike most other hydroids, which contract when they are cut, Tubularia is covered by a stiff perisarc which prevents contraction and the resulting alteration of relationships. The purpose of this work is to prepare a detailed description, at the ultrastructural level, of mouth and tentacle regeneration from isolated stems of Tubularia.

The summer was spent in preparing and embedding specimens of Tubularia in various stages of regeneration. These specimens will be sectioned for study with the electron microscope and for correlative study with the light microscope. Special attention will be given to the following stages: immediate tissue damage, healing, distal cell migration, tentacle formation and final emergence of the regenerate.