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THE CIRCULATION OF THE CLAMWORM *Nereis virens*

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Since the publication by Federighi (Proc. Nat. Acad. Sci. U.S. 13:639, 1927) it has been commonly accepted that capillaries of the clamworm are contractile. Similarly, capillaries in frogs and mammals have been thought to contract, but it has been demonstrated beyond doubt during the last decade that this is not the case. With this in mind, it was decided to investigate the vascular system of the clamworm with light and electron microscopy in order to decide whether or not structures exist in relation to the capillary wall which might be responsible for this unique capacity of capillaries in the clamworm.

Clamworms were collected in Emery cove and subsequently kept for at least 48 hours in a mud-free tank with running sea water. The worms were anesthetized with MS 222, stretched out and pinned down. The entire worm was fixed with glutaraldehyde, and osmic acid and subsequently, smaller segments were dehydrated and embedded in Epon.

The preliminary studies of this material with light microscopy have concentrated on the ventral and dorsal vessels, of which only the dorsal is contractile. By serial sectioning, the vascular connections between the gut, the lateral and dorsal muscles, the dorsal capillary network, and the parapodia have been analyzed. Electron microscopy of these vessels is now in progress.

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CHANGES IN ULTRAVIOLET SENSITIVITY OF CLEAVAGE DELAY DURING THE CELL CYCLE IN ZYGOTES OF *Echinarachnius parma*

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The ultraviolet (U.V.) $L.D._{50}$ during the first cleavage cycle increases exponentially (Bull. M.D.I.B.L. 6:33, 1966), whereas, the cleavage delay produced with identical doses of U.V. decreases during this time. A specific study was therefore undertaken to study the changes in sensitivity of subsequent cell divisions and to relate this to cyclobutane type dimer production.

Fertilized eggs were exposed to identical non-lethal doses of U.V. at varying times after fertilization and the variations in cleavage delay were scored and related to the time of the cell cycle that the exposure was made. The U.V. source was a 2537 Å lamp (G.E.-G15-T8) placed about 20 cm from the eggs. The eggs were gently stirred during the time of exposure. Under such circumstances the first cleavage delay that is produced decreases so that at about 55-60 minutes post-fertilization non-lethal doses cannot affect a delay. The second cleavage-sensitive period occurs during this first cleavage insensitive time but becomes completely refractory to U.V. about 15 minutes post-division. The same pattern is noted for the third cleavage. Thus, the sensitivity of cleavage delay is decreasing during the first DNA synthetic cycle (S) but has its peak during the second and third S periods.

Thymine dimers were not detected when zygotes were irradiated even at 100% lethal dose levels. However, they were found when E. parma DNA was exposed in vitro. This unexpected finding cannot be precisely explained at present but the following are possibilities. (1) Thymine dimers may not be formed in vivo. (2) Relative to the biological activity manifested, dimer activity may be too low to be detectable with present methods. (3) Repair and/or preparation methods may monomerize formed dimers. It is hoped that future experiments will distinguish between these possibilities.

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1967 #35

THE ENERGETICS OF Na^+ AND K^+ TRANSPORT IN THE DOGFISH RED CELL (Squalus acanthias)

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The relationship between energy generation and the work involved in the active transport of Na^+ and K^+ has been extensively investigated in the mammalian erythrocyte. Simple thermodynamic considerations permit a calculation of the minimal work required for these processes. No comparable studies are available for the red cells of nucleated O_2 consuming poikilothermic species. Previous work in this laboratory has indicated the desirability of such studies. In particular, the presence of a very high rate of ouabain-sensitive Na^+ efflux suggested that the energetics of this cell with respect to cation transport would be of particular interest.

In the present studies, measurements of K^+ influx have been performed on dogfish erythrocytes so that the net minimal work required to transport both Na^+ and K^+ could be estimated. K^+ influxes were measured at 13°C and 23°C under control circumstances and in the presence of 10^{-4}M ouabain. Despite the high rate of Na^+ transport total K^+ influx at 13° averaged approximately 1.6 mEq/Kg red cells/hr and at 23° averaged about 4.3 mEq/Kg red cells/hr. The ouabain sensitive fraction of these fluxes averaged 1.0 mEq/Kg red cells/hr and 2.1 mEq/Kg red cells/hr respectively. The rate of "active" Na^+ efflux (ouabain-sensitive) has previously been shown to be 8.1 mEq/Kg red cells/hr at 13° and 13.9 mEq/Kg red cells/hr at 30° (Bull. M.D.I. B.L. 5-2:39, 1965). Lactate production averaged approximately 1 mM lactate/Kg red cells/hr. Thus approximately a minimum of 60% of total anaerobic energy generation is used for Na^+ and K^+ transport. This value is substantially the same in the shark erythrocyte as in the mammalian erythrocyte. The apparent coupling of Na^+ to K^+ transport in this cell is substantially different from that found in human red cells. It appears that this cell is an unusually appropriate model for study of energetics in a unit with a high transport rate and for studies of coupled Na^+ and K^+ active transport.