1962 #6

PERSISTENCE OF TRANSCELLULAR SODIUM TRANSPORT BY AN EPITHELIAL CELL MEMBRANE IN THE ABSENCE OF OXIDATIVE PHOSPHORYLATION

N. S. Bricker, S. Klahr, H. Murdaugh, E. D. Robin, and P. S. Soteres, Washington University, St. Louis, Mo., The University of Alabama, Birmingham, Ala., and the University of Pittsburgh, Pittsburgh, Pa.

Active ion transport across cell membranes constitutes an essential endergonic biologic process which derives metabolic energy from high energy phosphate bonds (ATP). For epithelial cell systems transporting Na transcellularly (e.g. frog skin, toad bladder, renal tubule), this energy transformation is coupled to respiration. The availability of epithelial cells capable of anaerobic Na transport could facilitate investigation of energetics and mechanisms of active transport. The fresh water turtle urinary bladder has been found to possess this biologic attribute.

Isolated turtle bladders (Pseudemys scripta elegans) were suspended between symmetrical lucite chambers and bathed with modified Ringer's solution containing glucose as substrate. In 100% oxygen, spontaneous potential difference (P.D.) equaled 20 to 90 mV (serosa +) and short-circuit current (S.C.C.) 100 to 600 μ amps per 7 cm². In 100% nitrogen, P.D. and S.C.C. persisted for > 90 min., P.D. often > 50 per cent and S.C.C. > 25 per cent of control levels. In O₂, Na²² influx (mucosa → serosa) approximated S.C.C., and efflux was small. In N₂, Na²² influx exceeded S.C.C. by up to 100 per cent but efflux was increased proportionately. This apparent increase in passive Na conductance occurred despite an invariable increase in total resistance. In O₂, KCN (10⁻³ M) produced only modest decrements in P.D. and S.C.C. Na F (10⁻³ M) and monoiodoacetate (10⁻⁴ M) inhibited S.C.C. and P.D. in N₂. Dinitrophenol (10⁻⁴ M) inhibited in N₂ as well as O₂. It is especially significant that strophanthin (10⁻⁴ M) strongly inhibited in both O₂ and N₂.

These data indicate that energy requirements for sustained transcellular active Na transport could be met in the absence of molecular O_2 ; hence, energy transformation was not obligatorily coupled with respiration. Presumably, this metabolic energy arose from glycolysis and was delivered as ATP.

1962 #7

REGULATION OF ELECTROLYTE IN ELASMOBRANCH FISHES

J. W. Burger, Trinity College, Hartford, Conn.

Following the discovery in 1959 that the rectal gland of the spiny dogfish is a salt gland, the summers of 1960, 1961 were devoted to the physiology of this organ. The results have been published: Physiol. Zool. 35:205-217, 1962. During the summer of 1962 this work continued with special emphasis on long experiments with fish from which the rectal gland was removed. In addition to analyses for electrolyte in blood, urine, and rectal gland fluid, osmotic measurements were made. In general, the 1962 work was an expansion and confirmation of previously published shorter term experiments. While a miscellany of new facts were gathered, it is felt another sum-

mer's work is necessary to get the data into a comprehensive form.

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1962 #8

CLEAVAGE DELAY IN ULTRAVIOLET-IRRADIATED SAND DOLLAR ZYGOTES

J. S. Cook, New York University, New York, N. Y.

1. Ultraviolet (UV) irradiation after fertilization of sand dollar eggs delays first cleavage of the zygote. The delay can be partially reduced by treatment with visible light (photoreactiva-tion).

2. Late in the fertilization-to-first-cleavage (F-I) internal there is a so-called "refractory period," during which time UV treatment does not delay first cleavage, but does prolong the first-to-second (I-II) cleavage interval. Treatment with UV plus visible light at this time results in virtually complete photoreactivation, i.e., the cleavage intervals of treated zygotes are indistinguishable from those of unirradiated controls.

3. If zygotes are allowed to incorporate 5-bromodeoxyuridine (BUDR) into their DNA, partially replacing thymidine during the DNA-synthesis prior to first cleavage, the I-II interval shows hypersensitivity to UV irradiation in the "refractory period." Zygotes which have incorporated BUDR cannot be photoreactivated.

4. Zygotes which have been irradiated <u>while incorporating</u> BUDR prior to first cleavage show no apparent hypersensitivity or loss of photoreactivability of the F-I interval, but do show both hypersensitivity and loss of photoreactivability of the I-II interval.

5. A working hypothesis consistent with the data is that UV-induced cleavage delay results from interference with the primer-activity of DNA in subsequent DNA synthesis.

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1962 #9

FINE STRUCTURE OF SALT-REGULATING TISSUES

W. L. Doyle, The University of Chicago, Chicago, Ill.

- Holothuria. Preparations of the respiratory tree of the sea cucumber, <u>Cucumaria</u>, were made and shipped by air to Chicago for completion and preliminary examination in the electron microscope. On the basis of preliminary results, new evidence has been obtained relating to the function of certain amoebocytes in the formation of the extra cellular substances of the connective tissue.
- 2) <u>The so-called chloride cell of Fundulus</u>. Experiments were designed to clarify certain controversial aspects of the fine structure of the gill of Fundulus in relation to the state of adaptation of the organism to fresh and salt water respectively. Specimens were slowly adapted to fresh water under controlled conditions of temperature and salinity. Specimens

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