

range; the urine pH was the same or slightly more acid.

No effort was made to study the mechanism of nephridial action, viz. filtration, secretion, etc. The evidence indicates that the lobster's nephridia act as a 'kidney' differentially conserving some substances (glucose) while concentrating others (dyes, sulphate). Certain monovalent ions are regulated largely extrarenally. The hepato-pancreas while capable of absorbing a variety of materials placed in the stomach, and capable of the differential secretion of substances into the gut is not an excretory organ in that its products do not directly reach the outside of the animal. Phenol red is not eliminated by the anus (or the gills). Since the lobster and marine fish are both complex animals living in the same environment, a comparison of methods of solution of their homeostatic problems is of interest. The lobster apparently has simplified his problems by using a slightly modified form of sea water for an internal fluid instead of a dilute saline solution.

A Study of Pulmonary Epithelium of the Tadpole (*Rana catesbiana*) in Tissue Culture

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One of the basic problems in cellular physiology is to determine how specific alterations in metabolic functions effect growth of cell types of the same and different species. The purpose of this study was to determine the relationship of one of these metabolic functions, respiration, to the cellular activity of one particular cell type, pulmonary epithelium.

All cultures were prepared from pulmonary tissues of tadpole specimens of *Rana catesbiana*. Each lung was cut into small fragments approximately 1 mm cubed. The fragments were explanted in roller tubes and Carrel flasks. The medium was composed of equal parts of embryonic extract from 7-day chicks and cockerel plasma in which 1000 units of penicillin G was incorporated. To each culture was added 1 cc of supernatant consisting of 40% human ascitic fluid, 40% Tyrode's solution, and 20% embryonic extract. The cultures were grown at 25° - 26° C.

The first step was to determine the control activity of the tadpole's lung in vitro. Cellular migration could be divided into five stages: (1) During the first 48 hours there was an initial migration of macrophages. (2) During the next 5 days there was extensive fibroblastic migration. (3) Between the

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9th and 11th days the first pulmonary epithelial sheets could be distinguished. (4) Around the 15th day the ciliated columnar epithelium usually began its migration. Not only did ciliary activity persist, but definite ciliary patterns consisting of specialized channels were established. (5) Pigmented cells wandered from original explant during the third week in culture.

The two types of pulmonary epithelia of the tadpole appeared to have different metabolic needs for: (1) The ciliated epithelium had a longer latent period before any migratory activity than the pulmonary epithelium. (2) Aberrant nuclear forms such as multinuclear cells were seen only in the pulmonary epithelium. As such aberrant forms occurred only after 9 days in vitro, it is suggested that interference with normal mitosis was due to a metabolic deficiency of the environment such as partial anoxic conditions. These findings agree with previous work¹ in which the two types of pulmonary epithelia of the adult newt, *Triturus viridescens*, showed similar morphological activity in vitro.

The next step was to study the relationship of respiration to cell growth such as division and migration. Cultures of the tadpole's lung were subjected to various concentrations of HCN in modified roller tubes². Preliminary results indicated that the pulmonary epithelium could not withstand partial curtailment of aerobic respiration as well as the ciliated epithelium.

It is hoped that these observations can be used as a basis for further study in determining the interrelationships between cellular respiration and cell growth of epithelium in tissue culture.

References

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