

PULMONARY DIFFUSING CAPACITY FOR CARBON MONOXIDE IN THE HARBOR SEAL, *Phoca vitulina*

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This laboratory has been involved in a systematic study of the adaptations which permit prolonged diving in the seal. As a part of this study, measurements have been performed of the steady state pulmonary diffusing capacity for carbon monoxide D_{LCO} . A priori, an increased D_L might be expected to enhance the ability of this animal to supply oxygen to tissues following the completion of a dive.

These measurements were accomplished by a modification of the method of Fillay substituting end tidal CO_2 tension (measured by an infrared CO_2 analyzer) for arterial CO_2 tension. A total of 13 measurements were performed on 8 animals. Mean D_{LCO} averaged 0.91 ml/min/mm Hg. On the basis of weight, the D_{LCO} amounted to 0.34 ml/min/mm Hg/kg. The comparable figure in normal human subjects is 0.25. Thus, an increased D_L does not appear to constitute a significant adaptive mechanism.

TISSUE CULTURE AND VIRUSES OF MARINE ORGANISMS

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Various tissues and organs from *Fundulus* and from dogfish embryos, pups, and adults were tested for their ability to grow in tissue culture media. All explants were minced with scissors and single cell suspensions made by trypsin digestion according to established techniques. The following is a list of tissues or organs tested.

Fundulus: gill, fin, and cell-laden fluid from the abdomen of a diseased fish.

Dogfish: whole embryos; pup spleen and pancreas; adult oviduct, pancreas, and kidney.

Only the dogfish embryo cells attached readily to the substrate and spread out to form a mono-layer. Such mono-layers were maintained in a state of gradual decline for as long as four months (August through early December) after which they were lost due to bacterial contamination. During this time subcultures were attempted. The cells were transferred to new containers after trypsinization of the original mono-layers and were found to re-attach themselves. Mitosis was not observed in any cultures.

A variety of media was tested, the best being as follows: Earle's saline with NaCl increased to 0.26 M, Eagle's vitamins and amino acid mixture, 10% human serum, 10% fetal calf serum, 5% whole egg ultrafiltrate, and appropriate concentrations of penicillin, streptomycin, and fungizone.

Other additives, tested at several concentrations, such as dogfish embryo extract, lactalbumin hydrolysate, urea, and dogfish serum, were found to be of no benefit or actually inhibitory.

The agents (presumed to be viruses) of infectious pancreatic necrosis of trout and lymphocystis disease of the Centrarchid fishes were obtained from Kenneth Wolf. Initial attempts to

pass these viruses in embryo dogfish tissue cultures were unsuccessful.

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EARLY EMBRYONIC DEVELOPMENT IN Fucus

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Although classic material for the study of early embryological development in plants, the brown alga, *Fucus*, has been little studied with respect to nuclear and cytoplasmic cytology or cytoplasmic events associated with fertilization, polarization and embryonic cleavage. We have explored appropriate technical procedures for fixation and staining of the unfertilized egg, the zygote and early embryonic stages for use in studies ranging from macroscopic morphology to ultrastructure. Effective fixation was achieved with 10% formalin in sea water or 2-1/2% glutaraldehyde in sea water. A number of routine nuclear stains proved ineffective; acetocarmine, alum cochineal, azure B, and toluidine blue all proved of some value in staining nuclear, cytoplasmic or wall structures in squash preparations. Some success in obtaining adequate fixation for electron microscopy was achieved. We gained sufficient experience in collecting reproductive plants and obtaining gamete release and fertilization in vitro to allow preliminary experiments on two specific problems: a) the distribution of nuclear and cytoplasmic structures within the zygote associated with the polarization of the embryo and the determination of unequal cytoplasmic division at the first cell division, and b) the use of raised temperature in the production of abnormal eggs which should make possible a study of the role of the nucleus in development and the importance of the nuclear-cytoplasmic ratio in normal embryogenesis.

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EFFECT OF POLYURIDYLIC ACID ON DEVELOPMENT AND PROTEIN SYNTHESIS OF INTACT SAND DOLLAR EMBRYOS

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Previous studies on polyuridylic acid (polyU) in subcellular systems suggest that if messenger RNAs can cross the cell membrane, they should redirect the pattern of protein synthesis of intact cells. This study describes the effect of polyU on the pattern of development and protein synthesis by intact sand dollar embryos (*Echinarchnius parma*). Embryos incubated in sea water containing 500 to 1000 $\mu\text{g/ml}$ of polyU developed normally to the early blastula stage. Thereafter the central cavity became filled with cells producing a multicellular mass which underwent subsequent opacification and degeneration. External cell layers did not develop ciliation. We have

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