

perimental conditions used, cysteine would have been converted to cystine. No definite cystine peak was observed, but several *Homarus* and *Cancer* chromatographs showed double peaks in the cystine-valine area. The amino acids mentioned were present at lower concentrations than in cockroach blood. Quantities in *Homarus* and *Cancer* blood were roughly 5 to 10-fold, in *Limulus* blood 15 to 50-fold smaller than those previously found in *Periplaneta americana*. *Homarus* yielded relatively large amounts of proline. *Homarus* and *Cancer* blood contained large amounts of serine, glycine, and alanine. All three species showed presence of a substance which moved at a rate characteristic of taurine. In *Cancer irroratus*, it gave by far the largest peak of all.

A complete quantitative analysis of the data remains to be done. Frozen or lyophilized serum samples will be tried out as additives to media for insect cell cultures.

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I. Sialic Acid In Marine Organisms And Its Relationship To Influenza Virus.

II. Effect Of Viruses And Viral Nucleic Acids On Embryonic Development Of Echinoderms.

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I. Aqueous extracts of various marine organisms were hydrolyzed either with dilute HCl or with a concentrate of Asian influenza virus. Free sialic acid (SA) was assayed by a modification of Aminoff's thiobarbituric acid test (*Virology* 7:355, 1959). As a further check on the presence of SA-containing substrate, unhydrolyzed extracts were tested for their ability to inhibit the agglutination of red blood cells by heat-inactivated influenza virus (WS strain). Such indicator virus was found to agglutinate the RBC of hagfish, dogfish, herring gull, and harbor seal. Dogfish RBC were routinely used for hemagglutination inhibition (HI) tests. Members of the following genera were tested: *Pollachius*, *Anguilla*, *Lophius*, *Squalus*, *Myxine*, *Amphioxus*, *Halocynthia*, *Asterias*, *Chirodota*, *Nereis*, *Polinices*, *Baccinum*, *Omnastrephes*, *Mytilus*, *Lineus*, and *Cyanea*. Extracts of all the chordates had HI activity and yielded free SA on hydrolysis. Extracts of the two echinoderms could not be tested for HI because they were hemolytic. They did, however, yield small amounts of SA. None of the other invertebrates contained HI activity or hydrolyzable SA.

II. Many animal and plant viruses are capable of multiplying in cells of insects which serve as vectors. Therefore, it does not seem too far-fetched to suppose that some of these viruses may also be able to infect cells of marine species. Preliminary studies were undertaken to see whether interference with normal embryonic development of the sand dollar

(*Echinarachnius parma*) might be used as an indicator of viral activity. The method described by Karnofsky and Basch (*J. Biophys. and Biochem. Cytol.* 7:61, 1960) had to be modified because viral and control materials alike were inhibitory upon prolonged contact. These non-specific effects could be eliminated by repeated washing of embryos at different developmental stages. Refinement of the washing procedure will be necessary before further work on possible specific effects can be done.

III. Preliminary experiments on *in vitro* cultivation of trypsin-dispersed dogfish embryo cells gave encouraging results.

The Pulse Wave In *Squalus Acanthus*

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It has long been known that the pulse generated by the ventricle and bulbus arteriosus of the dogfish passes through the gills and appears in the dorsal aorta. This does not seem compatible with our knowledge of the damping effect of a capillary bed. How can a pulse pass the gills? Is there something unusual about the capillary network of the gill or is this phenomenon to be explained in some other way? Our studies have involved (1) pressure records from the ventral and dorsal aorta, (2) histologic study of normal and injected gills, and (3) direct observations of gill capillaries in the living dogfish.

Pressure records with a Lilly capacitance manometer confirm previous observations. In passing through the gill circulation, the pulse wave is reduced, usually being about $\frac{2}{3}$ or $\frac{1}{2}$ of its original size. In the dorsal aorta, the surviving pulse is modulated by small undulations, 4-5 times the pulse rate, synchronous with the respiratory movements of the gills. Histologic studies of the gills disclose that the capillary network of the lamellae appears to have a relatively large lumen, probably in order to accommodate the nucleated red blood cells of the dogfish which are considerably larger than the non-nucleated cells of mammals. A structure of special interest is the large arterial "sinus" which extends the length of each filament at its base. The lumen of this "sinus" is highly tortuous because it is encroached upon or traversed by many trabeculae which look like smooth muscle cells with an endothelial covering. Blood from the afferent gill arteries enters this sinus through defects in the medial coat and thence is distributed to the capillaries of each lamella. This tortuous swamp-like sinus, lying as it does between the artery and the gill capillaries, would seem to be ideally suited to damp out any pulse wave.

Direct observation of the gill circulation during life has been possible on several occasions. No convincing pulse wave was observed but the respiratory movements, always more rapid than the pulse rate, make observations difficult. Injection studies with India ink disclose no shunts between afferent and efferent gill arteries.