#### FRESH- AND BRACKISH-WATER OSTRACODA ON MT. DESERT ISLAND

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During the latter part of August, 1940, collections of Ostracoda were made in numerous fresh-water and brackish ponds on Mt. Desert Island. Since the material has not been worked over thoroughly as yet, few definite statements can be made concerning the species. A brief summary of the preliminary survey will be given, however.

Twenty species have been found so far, ten of which belong in the family Cytheridae, ten in the family Cypridae. Of the former group three species were found in fresh-water ponds, one each in Long Pond, lower Breakneck Pond and upper Breakneck Pond. Five different species came from a brackish pond somewhat north of Somesville. Two others came from a small brackish pool that occurred in the very spongy turf only a few feet away from the larger pond.

Of the Cypridae, two species belong to the genus Cypridopsis. One of the species was found in Somes Pond, Beaver Pond, Ripple Pond and Long Pond. The other was collected in Beaver Pond, a

small Pond in Somesville and upper Breakneck Pond.

Three species of the genus Cyclocypris were collected: one in Somes Pond and Lower Hadlock; another form in Lower Hadlock also; a third form, appearing to be Cyclocypris ovum (Jurine) in Lower Hadlock, Ripple Pond, and Lower Breakneck Pond.

A member of the genus Cyclocypria\* was collected in Somes Pond. Two Cypria were found, one in Beaver Pond, and another, Cypria elegantula (Lilljeborg) in a marshy spot near the junction of Highway 198 and the Eagle Lake road.

A Candona was collected in Beaver Pond, lower Breakneck Pond and Somes Pond. Possibly another species was present in the pond

at Somesville and in Long Pond.

Other forms will doubtless be brought to light with the further study of the material collected.

## THE PRESENT STATUS OF THE SYSTEMIC FUNGUS DISEASE IN HERRING OF THE GULF OF MAINE

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During the summers of 1939 and 1940 the author with the technical assistance of Allan Friedlich, Joseph Seronde and Mary Streeter conducted an incidence survey and investigation of the nature of a fungus disease in herring that assumed epidemic proportions and did great financial damage to the sardine industry of the State of Maine in 1930 and 1931 (Daniel, 1933 and 1933a; Fish, 1934). This study

\* A genus described by the author in a paper now in press: Fresh-water Ostracoda from Washington and other Western Localities. University of Washington Publications in Biology.

was fostered by the State of Maine Commissioner of Sea and Shore Fisheries, Arthur R. Greenleaf.

Over 6000 herring taken from the Mount Desert Island region were examined for disease. The average incidence of infection was found to be 7.5% in the herring taken inshore and 1.5% in those from the offshore islands. A detailed study of herring taken throughout a single season from a single weir in the Mount Desert Island Narrows showed that the infective incidence differed widely (5 to 30%) between different schools of fish. Many small alewives (sawbellies or kyaks) taken with the herring exhibited a similar course of the disease and had about the same incidence of infestation. No correlation was found between the age of the herring and the incidence of infection in those fish over 3 inches in length and under 12 inches.

The most commonly encountered stage of the parasite in the tissues of the host is the so-called 'resting stage'. This is usually a round multinucleate cyst having up to several hundred nuclei, and may be from 5 to over 150 micra in diameter. All of the tissues of the herring except the brain have disclosed the presence of this stage with the highest infestation found in the liver, heart and lateral wall musculature.

Any fish haboring the parasite was classified as diseased in this survey. In some instances the presence of the fungus could only be definitely determined by microscopic examination of the tissues, but usually yellowish spots on the viscera, especially liver, disclosed the presence of the parasitic organism. In advanced cases of the disease suppurative ulcers break through the skin and make pin-point holes in the silvery layer of the epidermis, thereby producing the 'pepper-spot' condition common to fishermen. Other characteristics of the advanced disease are a sigmoid curvature in the long axis of the body and the presence of pus sacs under the skin which vary in size from ½ inch to several inches in length.

The first host reaction to the parasitic organism is the invasion of monocytes which is closely followed by the deposition of epitheloid tissue which completely encircles the cyst. Finally this in turn is surrounded by a well-defined layer of connective tissue. The 'resting stage' consists of the organism and necrotic remains of the monocytes and epitheloid tissue surrounded by this dense wall of connective tissue.

The connective tissue can be penetrated, however, by the active parasite which can then reproduce and stimulate the formation of new layers of host tissue around the entire mass. This process can be repeated again and again with the parasite successfully breaking through and again being surrounded until the yellowish macroscopic visceral spots are produced, which are large masses of parasitic cysts and necrotic host tissue walled in by a connective tissue capsule.

The fungus seems to affect the herring simply by the destruction and replacement of normal host tissue rather than by the elaboration of any toxic substance. When the fish are badly diseased their movements are very feeble and they tend to remain almost stationary several feet below the surface of the water. This can be understood inasmuch as under these conditions the musculature has been almost completely replaced by fungus cysts. The heart has been observed in some instances to be made up of 9 parts fungus to 1 part cardiac muscle, and the liver almost totally destroyed and composed simply of hundreds of cysts and the jellied remains of necrotic tissue. Yet in even these instances the herring can still remain alive, the heart can beat feebly and the fish can make slow progress through the water. This probably would not occur if the damage done by the fungus were primarily due to the elaboration of a toxin.

The effect of the disease on the number of white corpuscles in the blood is striking. While no actual blood counts were made, estimations of the number of leucocytes in the microscopic fields were made on diseased blood which indicated that their number had increased up

to 50 times that of normal blood.

Reproduction of the fungus in the host is by a simple cleavage of the intracellular material which may occur within the cyst or within hyphal extensions that are sometimes several times as long as the diameter of the original cyst. The resultant daughter cysts are multinucleate and vary tremendously in size. Reproduction by 'fragmentation' as observed by Daniel (1933) was not encountered.

The fungus was successfully cultivated in 4 different media and in sea water. Growth and reproduction of the organism was followed for 6 weeks in these media and the representative stages photographed. Typically, growth at sea water temperature would begin after 8 to 10 days. This would be exhibited in two forms, either by the formation of blunt club-shaped projections which immediately involved the intracellular material, or by the formation of narrow elongate projections which were clear in unstained material and into which the intracellular material did not penetrate until the tube was well-formed. Frequently more than one of these club-shaped projections or aseptate hyphae would form from a single cyst. Either of these type projections could branch after being formed. The clubshaped projections would usually constrict to form daughter cysts before they were as long as twice the diameter of the parent cyst. The hyphae, however, were often observed 20 to 30 times as long as the diameter of the parent cyst. In old cultures the growth was simply a mass of these entangled hyphae twined about an occasional parent

Reproduction in culture, as in the host tissue, occurred by a simple cleavage of the intracellular material after it had merged into either the club-shaped projections or into hyphae. This cleavage in culture, unlike that in tissue was never seen to occur within the parent cyst. The cleavage in the club-shaped projections was simply a constriction at the tip. Cleavage in the hyphae was of two kinds. A series of constrictions sometimes resulted in daughter cysts arranged one after another down the length of the hypha, or multiple constriction could occur resulting in smaller and more numerous cysts

closely packed within the hypha.

Plehn and Mulsow (1911) cultured the systemic parasite found in European trout and observed a growth quite unlike that encoun-

tered in this study. It was characterized by the formation of many (3 to 12) hypha-like tubes, each of which achieved a length about equal to the diameter of the parent cyst and then rounded up to form conidia-like bodies. They believed the organism to be a Phycomycete resembling the Chytridiales. We also believe that the organism in the herring is a Phycomycete, subdivision Archimycetes, and probably very close to the Chytridiales. It is interesting to note that in these lower fungi conidia may or may not be formed depending upon environmental conditions. No zoospores have been observed in culture or in tissue, but it is entirely possible that the conditions under which this parasite has been studied are not favorable to the formation of this stage in the life cycle. Inasmuch as many of the known species of the Archimycetes are parasites that attack freshwater and marine algae, it is not unlikely that this gap in our knowledge of the life history of the parasite may be filled by more information concerning possible parasitic organisms encountered in the food of the herring.

This type of systemic parasitic fungus has been repeatedly described in European marine and freshwater fish (for review see Fish, 1934). Widespread epidemics of this sort in the herring have been reported in Western Atlantic waters in 1898, 1914 and 1930. These spacings of 16 year intervals indicate that, if the disease is really of a cyclical nature, one would expect the next occurrence of a widespread

epidemic in the middle nineteen-forties.

#### REFERENCES

#### THE GLUCOSE REABSORPTIVE PROCESS IN THE FROG RENAL TUBULE; EVIDENCE FOR GLOMERULAR FUNCTIONAL INTERMITTENCY IN THE NORMAL INTACT ANIMALS\*

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One hundred and fifty simultaneous inulin and glucose renal clearance determinations were made in this series of experiments on 15 bullfrogs with the glucose plasma concentrations ranging from 41 to 1650 mg. per cent, and the urine flows from 1 to 35 ml./kg./hr. The inulin clearance (occasionally creatinine clearance) was used to measure the rate of glomerular filtration, and the physiological and biochemical procedures were the same as those employed in an earlier study (Forster, 1938).

The amount of glucose reabsorbed by the renal tubule cells  $(T_G)$  was found to be influenced both by the simultaneous glucose plasma

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