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BULLETIN
OF
THE BINGHAM OCEANOGRAPHIC COLLECTION
PEABODY MUSEUM OF NATURAL HISTORY
YALE UNIVERSITY

VOLUME IX, ARTICLES 4 AND 5

STUDIES ON THE MARINE RESOURCES OF
SOUTHERN NEW ENGLAND

IV THE BIOLOGY AND ECONOMIC IMPORTANCE
OF THE OCEAN POUT, *MACROZOARCES*
AMERICANUS (BLOCH AND SCHNEIDER)

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V. PARASITES AND DISEASES OF THE OCEAN
POUT, *MACROZOARCES AMERICANUS*

BY ROSS F. NIGRELLI
New York Zoological Society

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ABSTRACT

Protozoan parasites recorded from the ocean pout (*Macrozoarces americanus*) are: *Sarcocystis* sp. Fantham and Porter (1943), (Sarcosporidia); *Ceratomyxa acadensis* Mavor (1914), (Myxosporidia); *Chloromyxum clupeiidae* Hahn (1918), (Myxosporidia); and *Plistophora macrozoarcidis* sp. nov. (Microsporidia). *Sarcocystis* and *Ceratomyxum* were not found in this study. *M. americanus* is a new host record for *C. clupeiidae*, which, together with *P. macrozoarcidis*, is found in the flesh. The latter induces large tumor-like growths affecting the marketability of the fish. The morphology, cytology, life history, pathogenesis and toxicity of *P. macrozoarcidis* are discussed.

The helminthic parasites are: *Cryptocotyle lingua* (Creplin, 1825), (Trematoda); *Bothrimonus intermedius* Cooper (1918a), (Cestoda); *Porrocaecum decipiens* (?) (Krabbe, 1878), (Nematoda); *Contracaecum macrozoarcium* sp. nov., (Nematoda); and *Echinorhynchus gadi* Zoega, 1776 (Acanthocephala). All the parasites except *B. intermedius* were found in the present survey. The larval parasites *C. lingua* and *P. decipiens* (?) affect the marketability. *C. lingua* may possibly infect humans since they have been observed naturally and experimentally in mammals; they are metacercarial and are found encysted predominantly in the skin of ocean pout, although other larval trematodes, which may or may not be the larvae of *C. lingua*, were present in the flesh. *M. americanus* is also a new host record for *C. lingua*. The adults of *P. decipiens* have been reported from the intestine of marine mammals; whether or not they can infect land-living mammals has not been determined. *Contracaecum macrozoarcium* sp. nov. is found in the intestine of ocean pout; a detailed description is given.

A new species of ichthyobdellid leech, *Platybdella buccalis* sp. nov., found in the mouth of a single specimen, is described.

A fibro-epithelial growth, predominantly fibrous in nature and present on the snout of one ocean pout, is reported and the probable causes of its growth are discussed. No copepods have been recorded for this host, although they have been reported in other members of the family (Zoarcidae).

I. PROTOZOAN PARASITES, WITH SPECIAL REFERENCE TO *PLISTOPHORA MACROZOARCIDIS*, SP. NOV., AN INTRAMUSCULAR MICROSPORIDIAN

The sporozoan parasites described in this paper and reported by other investigators, except for *Sarcocystis* sp., belong to the order Cnidosporidia. All species discussed are listed in Table I. The most important parasite encountered, for reasons reported elsewhere in these studies (pp. 132-170), is the microsporidian *Plistophora macrozoarcidis* sp. nov., which induces large tumor-like lesions of the muscle. The parasite has been reported recently by Fischthal (1944) and Sandholzer, Nostrand and Young (1945) as a species of *Ichthyosporidium* on the basis of a tentative identification made by the author of this paper. This contribution deals with a description of *P. macrozoarcidis*, together with a description of each of the other protozoan parasites recorded from North Atlantic ocean pout.

TABLE I. PROTOZOAN PARASITES OF *Macrozoarces americanus*

Parasite	Author	Year	Infected Organ	Locality
PHYLUM: Protozoa				
CLASS: Sporozoa				
SUBCLASS: Acnidosporidia				
ORDER: Sarcosporidia				
SPECIES: <i>Sarcocystis</i> sp.	Fantham & Porter	1943	Muscle	Canada
SUBCLASS: Cnidosporidia				
ORDER: Myxosporidia				
FAMILY: Ceratomyxidae				
SPECIES: <i>Ceratomyxa acadensis</i>	Mavor	1914	Gall bladder	Canada
FAMILY: Chloromyxidae				
SPECIES: <i>Chloromyxum clupeiidae</i>	Hahn	Present Paper	Body muscle	North Atlantic
ORDER: Microsporidia				
FAMILY: Nosematidae				
SPECIES: <i>Plistophora macrozoarcidis</i>	Nigrelli	Present Paper	Body muscle	North Atlantic

1. ACNIDOSPORIDIA

Sarcocystis sp.

Sarcosporidia are parasites of reptiles, birds and especially mammals. Recently, Fantham and Porter (1943) recorded such sporozoans from Canadian fishes. The organism found in *Macrozoarces americanus* was not specifically designated. The parasites were found in small numbers in smears made of soft muscle taken from a fish which was bought in a shop in Montreal. They report that Miescher's tubes, about one mm. long, were present. The spores measured up to 15 microns in length, although most of them were between 6 and 10 microns, with a maximum breadth of 3.5 microns. Metachromatic granules were present at the more pointed end, and a distinct nucleus was seen in some of the spores.

No such spores were encountered in our material. Structures resembling Miescher's tubes were common, but these were interpreted as "cylinders" of muscle fibers containing developing trophozoites of *Plistophora macrozoarcidis*.

2. CNIDOSPORIDIA

Ceratomyxa acadensis Mavor, 1914

Mavor (1914, 1916) reported the myxosporidian, *Ceratomyxa acadensis*, from the gall bladder of Canadian ocean pout and other

marine fishes. Members of the genus *Ceratomyxa* are characterized by lateral prolongation of the spore; shell-valves conical and hollow, attached on bases; sporoplasm usually not filling intraporal cavity. Mavor reported that the vegetative form of *C. acadensis* was polymorphic and disporous in sporogony. The spores were wide, short and slightly compressed dorso-ventrally, with very long fine lateral filaments. The polar capsules were spherical, but the polar filaments were not visible in the fresh unextruded state. The spores measured 40–50 microns in breadth; diameter along the sutural line 7–8 microns; diameter of the polar capsule 3–4 microns; length of the polar filaments 70 microns; length of the lateral extensions of the spore case 250–300 microns.

Chloromyxum clupeiidae Hahn, 1918

(Plate I A, B, C)

Members of the genus *Chloromyxum* are myxosporidians possessing four polar capsules. The majority of species are coelozoic, occurring principally in the gall bladder of marine and fresh water fishes. However, a few have been reported as histozoic, particularly invading muscle tissue. The form found in *Macrozoarces* resembles in many respects *C. clupeiidae* Hahn (1918) from *Clupea harengus* and other marine fishes. It also shows certain similarities to *C. funduli* Hahn (1915), a species infecting *Fundulus heteroclitus*. This is not surprising since it is very difficult to make a distinction between *C. clupeiidae* and *C. funduli*. It should be pointed out that *C. clupeiidae* was seen and recorded by Linton (1901), who turned over the material to Tyzzer for further study. Kudo (1919) had the opportunity to examine the slides prepared by Tyzzer and a comparison of Kudo's description of *C. clupeiidae* with the form present in *Macrozoarces* shows close agreement.

The *Chloromyxum* from *M. americanus* are intramuscular parasites producing a hyalin degeneration of the muscle fibers involved. However, the degeneration is not as extensive as that caused by *Plistophora*, a microsporidian also infecting muscle fibers of the ocean pout. A mixed infection of both types of cnidosporidians often occurs; the two types cannot be distinguished from each other by the candling method suggested by Fischthal (1944). However, with the aid of a hand lens, and after a certain amount of experience, one may be able to distinguish the two types of infection, although final diagnosis must be made

microscopically. The nodules produced by *Chloromyxum* are whitish and invariably small, measuring about 0.3 mm. in diameter, and may reach a length of approximately 2 mm. On the other hand, the trophozoic masses of *Plistophora* are often yellowish in tint and the organisms are more malignant, showing greater invasive characteristics. A section through one of the *Chloromyxum* infected nodules is shown in Plate I A. This figure shows the parasites in a late stage of development in which most of the sporozoans are in the pansporoblastic phase of the cycle. This is an interesting feature, indicating that a single infection is involved. Other muscle fibers may show earlier or later stages of development. A smear of mature spores is shown in Plate I C. Plate I B is from a smear showing immature spores with capsulogenous and sporoplasmic nuclei.

The details in the early stages of development were not studied in this species, although multinucleate trophozoites occasionally were seen. The majority of the material studied was in various stages of sporogony. This species is monosporoblastic. The mature spores were studied from iron-haematoxylin stained smears shown in Plate I C. As may be seen from this figure, the spores when viewed from the top (anterior end) are somewhat quadrilateral in form. The sides and basal parts of spore, however, are round. The spore case is exceptionally thin and not distinct from the sporoplasm. The latter stains intensely with haematoxylin, and is uninucleate. The polar capsules are pyriform with capsulogenous nuclei often persisting. The filaments were indistinct and none were seen extruded. The spores measure about five microns in length and seven microns across the anterior end.

Plistophora macrozoarcidis sp. nov.

(Plates II-V; Text Figures A, B)

Plistophora macrozoarcidis is a microsporidian, the sporont of which develops into a variable number of sporoblasts, each of which becomes a spore. More than 16 sporoblasts may be formed in the process of sporogony. Other species have been reported from fish (Kudo, 1924, Dofflein, 1928 and Bond, 1937, 1938). The type species is *Plistophora typicalis* Gurley (1893) from the muscle of *Cottus bubalis*, *C. scorpius*, *Blennius pholis* and *Gasterosteus pungitius*.

P. macrozoarcidis is an intramuscular parasite inducing large tumor-like masses (Plate II A, B), measuring in some instances up to eight

or more centimeters. They are found more often in the deeper body muscle and are especially abundant in the regions of the vertebral column. The growths have never been seen in the ulcerative condition, *i. e.*, broken through the skin, as may occur in some of the nodule-producing myxosporidians (Nigrelli and Smith, 1938). It is not known how the spores are liberated. They may be set free in the process of being eaten by predators (although it does not seem likely that this fish is as subject to predation as many others), or they may be liberated only after the death of the host and its subsequent decomposition.

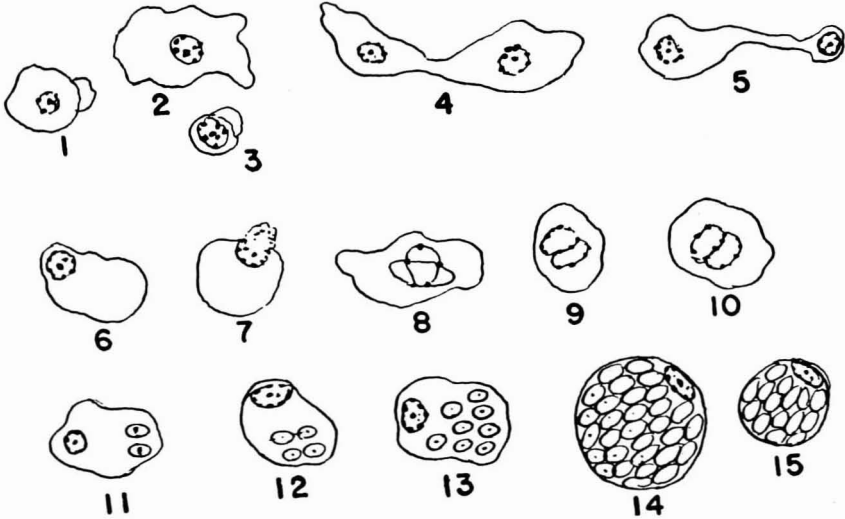
The larger tumor-like masses can sometimes be recognized by bulges on the body. The lesions are usually exposed by filleting and with the aid of a hand lens the smaller trophozoites may be seen scattered here and there in the muscle fibers. When the larger masses are cut, a pus-like exudation often occurs. Microscopical examination of this substance shows numerous spores and granular debris.

Life History. When the parasite was first examined, the organism was tentatively identified as an haplosporidial form and it was indicated that it might be a species of *Ichthyosporidium*. This conclusion was reached because of the large size of the tumor-like masses as well as the fact that the spores did not shoot out filaments when subjected to techniques usually employed for this purpose. However, after studies of sectioned and stained preparations it was established that the parasite was a microsporidian. A restudy of fresh spores verified this interpretation. *Mature spores* were recognized eventually and polar filament extrusion was successfully accomplished.

The stages in the life cycle of most species of *Plistophora* are not clearly understood, although they have been assumed to be similar to those reported for other microsporidians. Schizonts and pansporoblasts are easily recognized in sectioned and stained preparations, but details leading to the formation of these bodies have not been described heretofore for species of *Plistophora* from fishes.

It is assumed that the portal of entry is through the intestinal mucosa; uninucleate trophozoites (planonts) emerging from the spore pass through the gut wall and migrate to the muscle tissue of the body. These amoebulae invade the fibers, penetrating the sarcolemma. Here they undergo reproduction by binary or multiple fission producing more uninucleate forms now referred to as schizonts (Plate III A;

Text-Fig. A 1-5). The schizonts in turn invade adjacent regions of the muscle syncytia and continue the process of schizogony. These early stages are manifest macroscopically as minute whitish cylinders, the smallest of them measuring about 0.5 mm. in length, lying end to end along the long axis of the fibers (Plate II A). The presence of numerous such bodies scattered here and there indicate that a multiple infection may be involved. The schizonts vary considerably in size.



Text Figure A. Stages in the development of *Plistophora macrozoarcidis* ($\times 800$). 1-5 Schizonts; 6-10 Development of the pansporoblasts (6 Nucleus at the periphery of the cell; 7 Throwing off of chromatin; 8 Division to form binucleate stage; 9, 10 Fusion of nuclei to form the synkaryon); 11-15 Sporogenesis.

The nucleus is deeply basophilic, but chromatin granules can be differentiated, particularly in active dividing forms. There is no evidence of true mitosis.

Eventually each schizont undergoes a series of nuclear changes involving (1) a throwing off of chromatin material (Text Fig. A 6, 7), (2) a nuclear division which produces a binucleate cell (Text Fig. A 8), (3) a nuclear fusion to form the synkaryon (Text Fig. A 9, 10) and subsequently the pansporoblast. The latter, by repeated nuclear division, gives rise to a variable number of sporoblasts, each of which develops into a spore. Pansporoblasts in various stages

of sporogony are shown in: Text Fig. A 11-15; Plates III B, C; IV A, B; V A. The pansporoblast increases in size and the sporoblastic nuclei are developed successively, first two, then four, eight, sixteen, *etc.* However, the transformation of the sporoblast into the spore could not be followed. Eventually, the entire pansporoblast is filled with spores surrounded by a fairly resistant membrane, forming a "cyst" which measures from 15-30 microns in diameter (Text Fig. A 14, 15; Plate IV A); it will be noted also from this figure that the cysts occupy practically the entire mass of muscle bundles, and that although hyalinization has set in the fibers are intact. The spores in such cysts are immature, being for the most part still in the pansporoblastic stage containing developing sporonts. Macroscopically this stage is manifest as larger white bodies. However, such pansporoblasts continue to grow at the expense of the host tissue. Plate IV B shows how extensive the degeneration of the host tissue may be in this late stage of development. It is at this stage that the mature spores are found, and the tissue containing them is brownish and more or less granular in texture.

An interesting phenomenon noted, and one which may be new in the cycle of these microsporidians, is a division of the mature pansporoblasts as shown in Plate V A. The significance of this process is not understood.

A large number of spores, at this stage of development, are free, and many of them are highly refractive, indicating the absence of any internal structure. Among these spores, spore cases, and pansporoblasts may be found uninucleate cells typical of schizonts. Some of these are in the process of division. It is believed that these are the result of sporulation, and that auto-infection is taking place. The developing schizonts spread out along the interfibrous pathways and infect adjacent areas of the muscle to start the cycle again.

*The Spore.*¹ As stated above, mature spores are found in lesions containing the hard granular material. By mature spores is meant those spores which can be made to release their polar filaments. When bits of the brown granular substance are placed in sea water whether or not pressure is applied numerous spores are released which

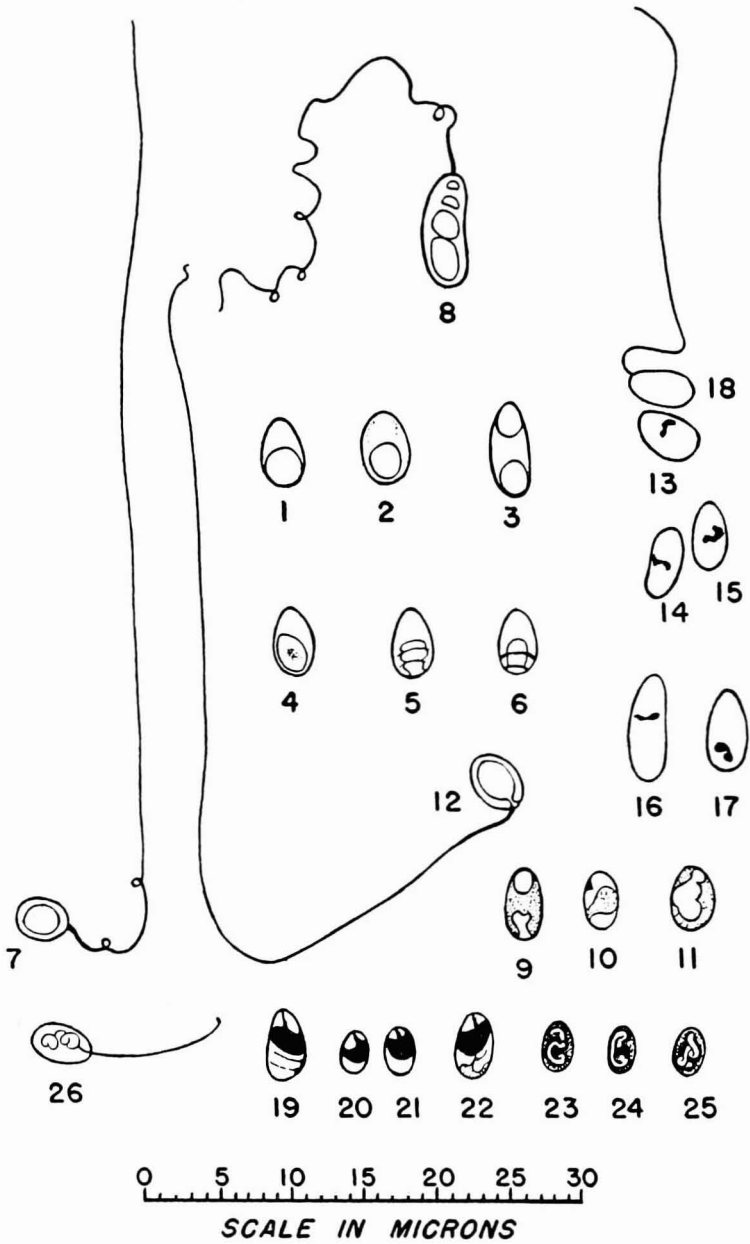
¹ We wish to express our thanks to Grace E. Pickford of the Bingham Oceanographic Laboratory, Yale University, for her studies on the spores which are included in this section.

can be seen to undergo violent discharge of the filaments. The soft white bodies release spherical cysts containing developing spores which are similar to those found in the brown bodies, but evidently these are not mature since they cannot be induced to extrude the threads.

Unstained live spores, examined under immersion lens, are variable in size but usually oval in shape (Plate V B; Text Fig. B). They measure from 3.5–5.5 microns in length. A few sausage-shaped spores were encountered which measured up to eight microns in length. This dimorphic nature of microsporidian spores is not unusual, but its significance is not definitely known. The spores have been regarded by some as micro- and macrospores, indicating a sexual phase. Others believe that the difference in size is due to the number of spores developed by the sporont; the smaller the number the larger the size (Kudo, 1924, 1930). In the present case, pansporoblasts with varying numbers of sporonts do occur and may account for the size variations found. However, although two distinct kinds of spores were found, no differences in the internal structures were noted. The larger spores also could be made to release their polar filaments under proper conditions.

The spore capsule is uniform in thickness and highly refractive. It is smooth and apparently structureless. Insofar as could be determined, it is composed of a single piece, as is found in most microsporidians (Monocnidea). The unstained, undischarged spore shows a clear space (vacuole) at the wider posterior end (Text Fig. B 1, 2). In some cases, both ends show these clear areas (Text Fig. B 3). Careful examination of the vacuole demonstrates the presence of a spiral structure, which is even more evident in spores placed in distilled water (Text Fig. B 5, 6). These observations indicate that the clear area is the polar capsule. It should be pointed out that the sporoplasm is indistinct in the living spore.

The extrusion of the polar filaments is a violent process. As the threads are "shot" out, the spores apparently rebound and spin. They may spin away from the filaments which become attached to other spores or entangled in debris. After the discharge the vacuole seems to occupy the entire spore case (Text Fig. B 12); a varying amount of material remains at the narrower (anterior) end, at the point of the discharge. Whether or not this material represents the sporoplasm could not be determined. Complete or partial discharge of spores may take place in distilled or salt water and the process is not



TEXT FIGURE B. (Explanation on opposite page.)

accelerated or induced in refractory material by the addition of hydrochloric acid, acetic acid or iodine placed under the coverslip. The iodine stains the filaments which are already discharged, but it will not cause the extrusion. This is not surprising, since it is well known that different species of microsporidians react differently to the various chemicals usually employed for this purpose. Threads stained with iodine are thicker at the basal than at the distal ends (Text Fig. B 7, 8, 12) and measure about 70 microns in length. It is the opinion of most investigators that the polar filaments in microsporidians are uniform in thickness throughout the entire length, except in the genus *Mrazekia*. In this group, however, the structure of the spores and filaments are different, for the thickened basal end (manubrium) is quite distinct from the finer distal part. Whether or not the filament in *P. macrozoarcidis* is tubular was not determined.

As mentioned above, no sporoplasm was seen to emerge after the discharge of the filament, although a blob of refractive material could be demonstrated in the iodine-stained preparations at the point where the filament was extruded. This may or may not be the sporoplasm. Spores stained with aceto-carmin or aceto-orcein show a faintly pink-staining mass near the middle of the spore; this is usually absent following the discharge (Text Fig. B 13-17). This would indicate that if the mass is the sporoplasm, it was discharged at the time of extrusion of the polar filament.

Smears of the material containing spores were fixed in Schaudinn's with acetic acid and stained with iron-haematoxylin. Some were counterstained with carbol-fuchsin. Eosin was found to be unsatisfactory as a counterstain for these spores. In such fixed and stained spores, the undischarged ones (Text Fig. B 19-22) show a central,

Explanation of Text Figure B. (Drawings by Grace E. Pickford.) 1, 2 Untreated, unexploded spores as they appear when first examined in sea water or distilled water; typical appearance with one "vacuole" at wide end. 3 Same, with two "vacuoles." 4 Same, showing faint structure within "vacuole"; ill-defined cloud often seen. 5, 6 Same after a few minutes in distilled water, with definite spiral structure in "vacuole." 7 Typical appearance of a discharged spore; thread slightly thicker proximally, but otherwise no structure observed. 8 Same, partial discharge of a large spore. 9-11 Undischarged spores after treatment with iodine. 12 Discharged spore showing aperture after iodine. 13-17 Undischarged spores after prolonged treatment with aceto-carmin; nucleus stained pink. 18 Typical discharged spore, after aceto-carmin; no nucleus. 19-22 Undischarged spores shown with Schaudinn-acetic, iron haematoxylin. Deeply stained central band of sporoplasm and slender thread or channel leading to apex. Occasional faint structure in "vacuole." 23-25 Undischarged spores, treated as above, sometimes show a highly refractive coiled tube inside. 26 Partly discharged spore, from fixed stained smear. No sporoplasm or nuclear material remains. Sporoplasm never seen in fully discharged spores.

darkly-staining mass with a line of similarly stained material leading towards the anterior end. The contents in the posterior part of the spore were often vague, but in some instances it was suggestive of a spiral (Text Fig. B 19). In certain spores the filaments were highly refractive, appearing as if filled with air (Text Fig. B 23-25).

The stained discharged spores never appear to contain the sporoplasm (Text Fig. B 26). When the appearance is to the contrary it is due either to the folding of the wall causing refractive indentations or to incomplete extrusion of the filament. In the latter, parts of the spiral thread are seen tightly coiled within the spore case, while a correspondingly shorter portion may be extruded.

Such observations of the stained spores suggest that the sporoplasm may be "shot" out before the thread, or simultaneously with it. If it moved out afterwards, by its own activity, one should find an occasional spore in which the planont could still be seen. On the other hand, if the planont is shot out one would expect to find amoeboid forms among the debris in the smear. It is difficult to say whether or not certain small objects seen in these preparations may be the released planonts.

Discharged spores with open anterior ends are seen rarely, but never when the thread remains connected; they are probably formed when the filament is torn away.

Comparisons. *Plistophora* have been reported from several species of marine fishes. However, it is very difficult to make comparisons since most of the investigators failed to report any detailed information which would distinguish their species from those previously recorded. Insofar as is known, *P. macrozoarcidis* differs from those species recorded from the muscle of marine fishes in the following ways: (1) size and shape of the pansporoblast, (2) size and shape of the spore, (3) length of the polar filament, (4) factors which will induce polar filament extrusion, (5) host, (6) size of the lesion induced by the parasite, and (7) geographical distribution.

Pathogenesis. Cnidosporidians are parasites of invertebrates and cold-blooded vertebrates. The host reactions to these Protozoa are manifest in many ways, from the production of simple cysts to hyperplastic growths of the tissues involved. In most cases, however, the reaction is merely a process involving tissue destruction. The para-

sites grow at the expense of the host. This action is especially evident in the case of infection with *P. macrozoarcidis*. Wherever the parasite infiltrates there is a complete hyalinization and destruction of muscle, eventually only granular debris remaining. Phagocytic activity of host cells is quite evident. Occasionally the host lays down a protective layer of fibrous connective tissue and this at times may be so extensive as to appear hyperplastic (Plate V C). There is very little inflammatory reaction involved.

Although many foci of infection may occur in any one fish in relatively distant parts of the body indicating a multiple infection, it appears that sporulation *in situ* is possible and that the planonts may migrate along the intermuscular or interfibrous pathways and set up new centers of schizogony and sporogony. Whether or not the organisms will grow and reproduce at freezing temperatures, as indicated by Sandholzer, Nostrand and Young (1945), was not determined by us. These investigators reported that microsporidian lesions increase in size and number at temperatures as low as -13° C., and that non-infected fillets placed in contact with infected ones became parasitized at this low temperature. It is obvious that these organisms are capable of growth and reproduction at comparatively low temperatures under conditions of the normal winter habitat of ocean pout, but if the experiments of Sandholzer, *et al.*, prove to be true, then these protozoans must have protoplasm that is very different from that in ordinary plants and animals, and even from that found in some psychrophilic bacteria which can grow and reproduce at 0° C.

Toxicity. The question as to whether or not cnidosporidians elaborate toxic substances has never been settled. Kudo (1924) refers to this problem only slightly. In our studies, glycerine extract or saline suspension of spores injected intraperitoneally into mice and rats demonstrated no toxic effects. Spores fed to mongrel dogs passed through the gastro-intestinal tract seemingly unchanged, with no visible effects to the animals other than the production of increased amounts of mucus in the stools. Sandholzer, *et al.*, reported no ill effects to cats and pigs. The results of post-mortem examination of cats and pigs sacrificed following the feeding of infected ocean pout material was summarized for these workers by Dr. D. R. Coburn, Veterinarian in charge of the Patuxent Wildlife Disease Research Laboratory, as follows: "Post-mortem examinations and histological

study of representative tissues did not reveal the presence of any pathological change attributable to the fish feeding." However, these experiments do not preclude the possibility that the organisms or the fish proteins denaturalized by the enzymatic action of the parasites may be toxic to humans.

SUMMARY

1. Four species of sporozoans are reported from ocean pout (*Macrozoarces americanus*) from the North Atlantic. Three species occur in the muscle and one in the gall bladder.

2. With the exception of *Sarcocystis* sp. reported by Fantham and Porter (1943), all the organisms belong to the subclass Cnidosporidia. Of the myxosporidians, *Ceratomyxa acadensis* Mavor (1914) is recorded from the gall bladder, while *Chloromyxum clupeidae* Hahn (1918) is reported for the first time from the muscle of the ocean pout.

3. A new species of intramuscular microsporidian, *Plistophora macrozoarcidis*, is described. Macroscopically the parasites are recognized as white or buff to brown colored bodies lying along the long axis of the muscle fibers. In some fish these infections involve large numbers of fibers which in certain instances collectively give the appearance of a massive tumor-like growth. Microscopically, the smaller white bodies contain developing schizonts; the larger white bodies are usually filled with schizonts and immature pansporoblasts; the buff or brown bodies contain mature pansporoblasts filled with ripe spores.

4. The life-history of *P. macrozoarcidis* seems to be as follows. The trophozoites penetrate the sarcolemma and undergo reproduction by binary or multiple fission producing numerous uninucleate schizonts; the schizonts in turn invade adjacent regions of the muscle syncytia and continue the process of schizogony. Eventually, each schizont undergoes a series of nuclear changes which involve the throwing off of chromatin material, a nuclear division resulting in a binucleate cell, a nuclear fusion to form the synkaryon and subsequently the pansporoblast. The pansporoblast increases in size and the sporoblastic nuclei are developed successively until sixteen or more such bodies are formed. However, the transformation of the sporoblast into spores could not be followed. It is assumed that the portal of entry is through the gastro-intestinal tract and the planont released from the spore makes its way to the body muscle. There is some evidence that auto-infection may take place.

5. A detailed description of the spore and polar filament extrusion is also reported. The latter is accomplished in sea or distilled water even when no pressure is applied.
6. The pathogenesis and toxicity of these organisms are discussed.

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II. ON *CONTRACAECUM MACROZOARCIIUM*, SP. NOV., WITH SPECIAL REFERENCE TO *PORROCAECUM DECIPIENS* (?) (KRABBE, 1878) AND OTHER HELMINTHIC PARASITES

An outstanding feature of the helminthic fauna of *Macrozoarces americanus* is the paucity of the trematodes. It is possible that they have been overlooked, but an intensive search in 25 specimens of ocean pout in this laboratory failed to uncover a single fluke other than the larval form of *Cryptocotyle lingua*. Further, insofar as is known, no trematodes have been reported by other investigators who have had the opportunity to examine ocean pout for parasites. The indication is, then, that these fish must be especially resistant to infections by trematodes, even by those numerous species that apparently show no host specificity. A similar condition was reported by Markowski (1939) in his studies on the helminthic forms of *Zoarces viviparus* from the Baltic. This investigator reported one species of fluke (*Gyrodactylus medius* Katherina, 1894) from the gills of this host. The internal helminths of *Z. viviparus* consist of: three species of cestodes (*Caryophyllaeus* sp., juv., from the gut; *Triaenophorus lucii* from liver and body cavity; *Bothriocephalus* sp. from the gut); four species of nematodes (*Cystidicola skrjabini* from the gut; *Raphidascaris* sp., larvae, from liver and gut; *Contracaecum aduncum* from gut and body cavity; *Contracaecum* sp., larvae, from the gut); five species of acanthocephalans (*Corynosoma strumosum* from the body cavity and *Echinorhynchus gadi*, *E. salmonis*, *Pomphorhynchus proteus* and *Neoechinorhynchus rutili* from the intestine).

Table II lists the helminthic parasites found in the American ocean pout. The metacercarial form and the larval nematode found in the flesh are especially important since they might affect both the marketability and edibility of the fish.

He stated further,

As the larva, now termed a metacercaria, grows larger the cyst increases in size . . . The metacercaria enlarges faster than the cyst and bends on itself to form a J- or U-shaped larva. Meanwhile, the mesenchymatous tissue of the fish has formed a strong connective-tissue capsule around the primary cyst which cannot increase further in size. Consequently, the larva, which fills the cyst completely, can grow no larger. It may persist in this condition for long periods of time, perhaps years.

However, metacercarial cysts are also of common occurrence in the *flesh* of the ocean pout. Whether or not they are the larvae of *Cryptocotyle* is still uncertain. The fact that the latter are present in the skin would lead one to assume that they may be the same. The ocean pout is a soft bodied form and it is possible that the parasites, if they are *C. lingua*, may migrate deep into the flesh before they become encysted. To establish this with certainty, the worms first must be liberated from the cysts, and detailed observations must be made on the excretory system and its pattern determined and compared with that found in previously described larvae of *Cryptocotyle*. Liberating the worms from the cysts is not a simple process since the host membranes forming part of the covering are tough, and the worms are often injured in the process of dissection. Stunkard (1930) obtained excellent material for study by subjecting the cysts to artificial gastric and intestinal enzymes.

The metacercarial cysts in the flesh of *Macrozoarces americanus* are minute, light or dark in appearance. They measure about 0.1–0.2 mm. in diameter. Sectioned and stained preparations show the presence of an encysted worm. The oral sucker is clearly visible and there is no indication of an acetabulum, the absence of which is typical of the metacercariae of *C. lingua*.

The capsule is especially thick and in some at least devoid of any pigmentation in contrast to the cysts seen in the skin, which are invariably surrounded by host melanophores or melanin-bearing cells which are massed around the encapsulated organisms. Such a condition has never been reported, insofar as is known by us, in flesh-inhabiting trematode larvae, as for example in the case of *Clinostomum marginatum* (found in the flesh of many species of fresh-water fishes). Hunter (1941) attempts to account for such differences by postulating that deposition of melanin in cells surrounding the cysts of the metacercariae may result through the action of a chromogen which is

brought in by the parasite, the host cells possessing the necessary enzyme for this reaction. However, parasites like *Clinostomum* may lack the necessary chromogen. At any rate, it is our belief that the presence or absence of pigmentation is one of location. In most instances, the larvae of *Cryptocotyle* encyst in the corium of the host tissue where melanophores are of great abundance; the melanated cells around the cysts may be melanophores or they may be massed macrophages heavily laden with melanin granules. The presence of the latter at sites of infection in fish is a common host reaction. In the case of *Clinostomum* larvae and some metacercarial cysts in the flesh of the ocean pout, the lack of pigmentation may be attributed simply to the absence of these melanin-bearing cells.

If the metacercariae found in the flesh prove to be different from those found in the skin of the ocean pout, then its life history also must be established. It is very likely that it would follow the same pattern reported for *C. lingua*, the cycle of which was determined by Stunkard (1930). The adult of this species normally becomes sexually mature in the intestine of terns and sea-gulls. The parasite belongs to the family Heterophyidae, about which Stunkard stated:

So far as known, all members of the family have a common life history, and the cercariae when they escape from the snail encyst in fishes, being transferred passively to their vertebrate hosts when the infected fish are eaten. One peculiarity of this group of trematodes consists in the fact that they have little specificity in regard to their final hosts . . . All species, insofar as they have been investigated, may develop to maturity in the common laboratory animals, cats, dogs, rats, rabbits, guinea-pigs, etc. Four have been proved experimentally infective for man and presumably all are potential human parasites.

It should be pointed out that *C. lingua* has been reported as occurring naturally in certain fish-eating mammals. Stunkard and Willey (1929) established experimental infection in kittens and mice. Willey and Stunkard (1942) reported similar results for dogs, showing that the pathology resulting from this infection is manifest by a denudation of the epithelium of the mucosa, production of large amounts of mucus, sloughing of tissue, hyperemia, and even a hyperplasia of the tissue elements involved.

2. CESTODA

Bothrimonus intermedius Cooper (1918a)

This is a pseudophyllidean tapeworm first described by Cooper (1918a) from the intestine of the flounder, *Pseudopleuronectes americanus* and redescribed by him in 1918b. The same investigator (1921) reported the parasite from *Macrozoarces americanus*. On the basis of this identification it was also recorded by Clemens and Clemens (1921). The parasite was not found in fish studied in this survey.

3. NEMATODA

Nematodes found in ocean pout belong to the order Ascaroidea, family Heterocheilidae, subfamily Anisakinae. Such worms are characterized as being stout in body and possessing a head with three large lobes or lips; alimentary canal with post-oesophageal ventriculus, and/or oesophageal or intestinal diverticula; cuticle not provided with spines or raised structures (Yorke and Maplestone, 1926). The generic and specific distinctions are based on the nature of the digestive tract (Baylis, 1921) and the lip structures.

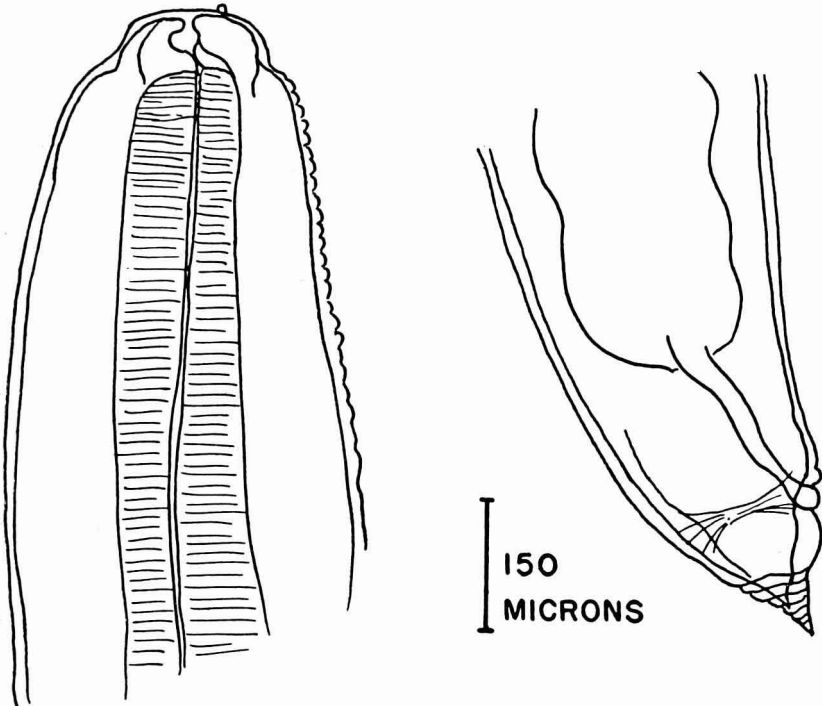
Porrocaecum decipiens (?) (Krabbe, 1878)

Plate VI, Text Figure C

Many species of larval nematodes have been reported from marine fishes of the American coast of the North Atlantic (Linton, 1901). The descriptions in most instances are incomplete and, consequently, it is very difficult to classify the worms. Earlier workers described the larval nematodes either under the name "Agamonema," which has no generic value, or under the name *Ascaris*, a term which is now used to designate those worms that infect land living mammals. This confusion was noted by Baylis (1916) and by Walton (1927). The latter worker pointed out that "The group *Agamonema capsularia* (*Ascaris capsularia* Rud. 1802 *sen. str.*) has been the recipient of many encapsulated larval Ascarids, usually from aquatic hosts, and undoubtedly of several different species." A similar opinion was held by Baylis (1916), but after a careful study of encapsulated worms from several fish species, he defined the limits which encompassed the parasites that could strictly be called *Ascaris capsularia*. In his report, Baylis indicated that *A. capsularia* was the larval form of *A. decipiens* Krabbe, sexually mature ascaroids found in the intestine of

marine mammals. Kahl (1939) is of the same opinion. However, it should be noted that the evidence for this conclusion is circumstantial and as yet no experimental data has been offered to substantiate this interpretation. Baylis (1921) later allocated the sexually mature *Ascaris decipiens* to the genus *Porrocaecum* Railliet and Henry.

The larval nematodes embedded in the flesh of the ocean pout are



Text Figure C. Anterior and posterior parts of *Porrocaecum decipiens* (?) Krabbe.

in all probability *Porrocaecum decipiens*. A species of nematode infesting the flesh of Canadian ocean pout was reported by Clemens (1920). The parasite was examined by the late Maurice C. Hall, who considered it a species of *Kathleena*, and it was later reported as such by Clemens and Clemens (1921). Hall further suggested that the worm might be a new species. Whether or not we are dealing with the same worm has not been determined, but it is our belief that the two are identical. However, there are certain differences between our specimens and *A. capsularia* described and figured by Baylis (1916),

especially in the nature of the lip structures and the posterior end. The similarities in the size and relationships of the several parts of the intestinal tract are striking even though Baylis' description is a composite one, *i. e.*, it is derived from specimens taken from different hosts. The absolute and relative size of these structures (oesophagus, ventriculus and caecum) has been and still is used for the diagnostic characters. We agree with Baylis (1916) and Dollfus and Desportes (1945) that the caecum is a highly variable organ and its size has no species value. In the case of the oesophagus and ventriculus, Baylis shows that the length of these structures is correlated to a certain degree with the length of the worms. The ventriculus (posterior oesophagus) is less plastic, attaining a maximum length (1.2 mm.) in worms of larger size (28 mm. or 38 mm.). He concludes from this that the ventriculus "does not continue to increase in size in proportion to the growth of the individual." In general, our measurements of the worms from the ocean pout fall within the expected variation given by Baylis for *Ascaris capsularia*. Our size range is slightly narrower, extending from 11-25 mm. in length and from 1.5-2.3 mm. in width. The cuticle is smooth, the anterior end is more or less round, and a boring tooth, typical of the larval ascaroids of fishes, is present. The lip is typically trilobed, but it should be noted that although the lip structures of these larval worms may be used to distinguish one form from another, they cannot be used for referring the parasites to any one adult species. The oesophagus averages in size about 2 x 0.2 mm.; ventriculus 1.3 x 0.3 mm. The caecum is highly variable measuring from 0.51 to 1.2 mm. This variation is not correlated with the size of the nematodes, since even small caecae may be found in larger worms.

The giant excretory cell characteristic of the forms designated as *Ascaris capsularia* and *Porrocaecum decipiens* was not observed, although the photograph of the worm (Plate VI) shows what may be its duct ending between the two rudimentary ventro-lateral lips.

The anal opening lies about 0.15 mm. from the posterior extremity, the latter terminating as a small spike-like tip that appears to be retractile.

If the form found in the flesh of the ocean pout is the same as *A. capsularia* and therefore the larval stage of *Porrocaecum decipiens*, then the adults may be expected to occur in the intestinal tracts of marine mammals, as indicated above. Whether or not the worms can

infect land inhabiting mammals, including man, has never been determined. Feeding experiments should be carried out to establish this point since it has been suspected that other nematodes of fishes do infect these forms.

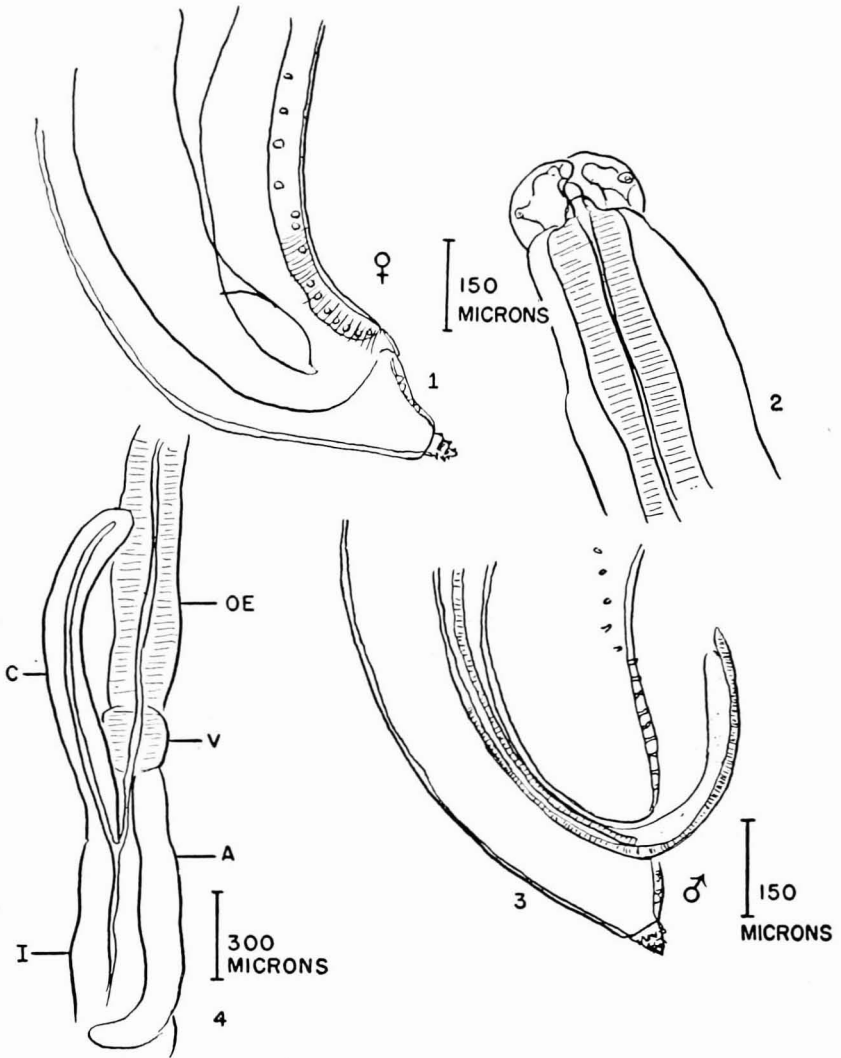
Contracaecum macrozoarcium sp. nov.

Text Figure D

Ascaroids in the stomach of ocean pout belong to the genus *Contracaecum* Railliet and Henry, 1912 (= *Kathleena* Leiper and Atkinson, 1914) (see Yorke and Maplestone, 1926). They are partly characterized by the presence of an oesophageal appendix and an intestinal caecum. Again similar worms were noted by Clemens and Clemens (1921); these were also identified by Hall as a species of *Kathleena*. Our studies show that at least one of the worms in our collection, *Contracaecum macrozoarcium*, is new. These are stout reddish worms found in comparatively large numbers in the gastro-intestinal tract of ocean pout. Both immature and mature males and females were found. The males are smaller, measuring 15–28 mm. in length and about 0.25–0.70 mm. in width. The females measure from 26–45 mm. in length and from 0.5–1.5 mm. in width. The uteri of the larger females are filled with thin shelled eggs.

Measurements of other structures are given in Table III. In both sexes the cuticular covering of the body is finely striated. The head end is truncate with typical lip structures, the nature of which can best be determined by examining Text Fig. D. The tail in both sexes is retractile and the cuticular covering at the tip is raised in minute spines, much like that reported by Chandler (1943) for *Contracaecum habena* (Linton), a common ascaroid from the toadfish, *Opsanus tau*. The spicules of the male *C. macrozoarcium* are sub-equal and the extended portions measure 0.31 and 0.39 mm. in length. There are about 12–14 pairs of pre-anal papillae and about 4 pairs of post-anal ones.

Several worms were found which, on the basis of differences in measurements of the several parts of the digestive tract, appear to be distinct. These measurements are given in Table III. The worms are small compared to *Contracaecum macrozoarcium* and they may be stages in the development of this species, since one larval form with boring tooth on the lip, and an immature male and female specimen,



Text Figure D. *Contracaecum macrozoarcium* sp. nov. 1, 2 Posterior and anterior end of female. 3 Posterior end of male showing spicules. 4 Digestive system: *Oe*, oesophagus; *V*, ventriculus; *A*, appendix; *I*, intestine; *C*, intestinal caecum.

TABLE III. COMPARATIVE MEASUREMENTS OF INTESTINAL FORMS OF *Contracaecum* IN *Macrozoarces americanus*

Structure	<i>C. macrozoarcium</i>		<i>Contracaecum</i> sp.		<i>Contracaecum</i> sp.
	Male	Female	Immature Male	Immature Female	Larval
No. of Worms	4	6	1	1	1
Length	15-28 mm.	26-45	12	20	10
Width	0.25-0.70	0.50-1.30	0.27	0.31	0.25
Oesophagus		5.30-6.30	2.20	2.40	1.40
Ventriculus	0.14-0.16 x 0.24	0.26	0.20	0.19	0.13
Appendix		1.05-1.12	0.84	0.18	0.63
Caecum		1.40-1.58	0.71	0.78	0.68

are represented in the series. All have the characteristic spiny tip and the lips show the same arrangement of the latero-ventral structures with the interlocking lobes. The writer is uncertain whether or not any taxonomic value can be given to the measurements of the other structures of these worms. The extreme variability of the caecum, as mentioned previously, certainly eliminates measurement of this structure as a diagnostic character. However, it may be summarized that the sexually mature ascaroids described here as *Contracaecum macrozoarcium* differ from other forms in each or all of the following respects: (1) both the caecum and the appendix measure one mm. or more in length, but are not necessarily equal in size, (2) the ventriculus is sub-globular, and (3) male spicules are sub-equal.

4. ACANTHOCEPHALA

Echinorhynchus gadi Zoega, 1776 (Plate VII)

Echinorhynchus gadi is a common parasite of marine fishes of the North Atlantic. Linton (1933) points out that this species has been recorded, either under the name *E. gadi* or its synonym, *E. acus* (Rudolphi) from at least 54 species of fishes from the Woods Hole region. In the large list of hosts he included the ocean pout, *Macrozoarces americanus*. He showed, also, that although the worm has been found in many teleosts, "the specific hosts in which it occurs frequently and in considerable numbers are limited to perhaps ten or twelve." *M. americanus* was not included in this latter list, perhaps because only a few specimens were examined. In the present studies, a large number of ocean pout from several areas along the North Atlantic were examined for their internal parasites. The incidence

of infection is very high and the number of *E. gadi* found was always large. One specimen examined by the writer yielded over 100 of these spiny-headed worms. Linton (1933) recorded a total of 74 from six specimens examined in April.

The generic characters for *Echinorhynchus* were defined by Van Cleave (1919). In 1925 he called attention to the great diversity of form and size in *E. gadi*. Linton (1933) showed, following studies on many specimens taken at random from different hosts, that the variations may extend "to details of structure, such as relative lengths of proboscis, receptacle, lemnisci, number of vertical rows of hooks on the proboscis, number of hooks in a row, and in some degree, to the size of the hooks." To these variations should be added body color, which Linton (1889, 1901) had noted in his earlier descriptions of this parasite under the name *E. acus*. The color of these worms may be ivory, red or orange. Among our specimens white and red were the only colors noted. Both types may occur in the same host, or each color variety may be found in different fish. The cause of this color variation is not known but is probably due to exogenous factors associated with the saprozoic habits of these organisms.

In the ocean pout examined in this survey both immature and mature worms were found. They measure from 10 to 50 mm. or more in length. Mature gravid females usually measure from 20 to 50 mm., while those under 20 mm. are either males or immature individuals. The worms contract and become strongly coiled when placed in preserving fluid. The measurements for the various structures of the parasite vary considerably but are within the range of variation established for these highly plastic organisms. Van Cleave (1925) believed that the variability in body form and size may be influenced by the host species parasitized. This may be so, but the same variation may occur within the same host.

The proboscis measures about one mm. in length in both sexes. The proboscis sheath is slightly longer. The hooks are alternate in arrangement, restricted to the proboscis. There are from 10 to 12 longitudinal rows of these stout recurved hooks, with from 12 to 14 to each row. Except for the basal hooks, which are smaller, they are uniform in size. The neck is short and is free of hooks. The lemnisci extend up to or slightly beyond the posterior level of the proboscis receptacle. The males have well developed copulatory bursa. The gravid females have many embryos in the body cavity which measure

on the average about 80 microns in length and about 10 microns in width.

Echinorhynchus gadi has also been reported from other Zoarcidae. Nicoll (1907) reported the parasite from *Zoarces viviparus* from the British coast under the synonym *E. acus*, and Markowski (1939) recorded it under its correct name from the same host from the Baltic.

SUMMARY

The helminthic parasites of the ocean pout (*Macrozoarces americanus*) consist of the following forms. (a) Trematoda: metacercariae of *Cryptocotyle lingua* found in the skin and fins; metacercarial forms found in the flesh which may or may not be the same. (b) Cestoda: *Bothrimonus intermedius* Cooper from the intestine. (c) Nematoda: *Porrocaecum decipiens* (?) (Krabbe) encapsulated in the flesh and *Contraecum macrozoarcium* sp. nov. from the intestine. (d) Acanthocephala: *Echinorhynchus gadi* Zoega from the intestine. All the forms listed above, with the exception of the tapeworm, were recovered in the present survey. Insofar as is known, *Macrozoarces americanus* is a new host record for *Cryptocotyle lingua* and *Porrocaecum decipiens* (?). These two parasites and the metacercarial form found in the flesh are important economically since they might affect the marketability and edibility of the fish. *Cryptocotyle lingua* is a potential human parasite.

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III. *PLATYBDELLA BUCCALIS*, SP. NOV., AN ICHTHYOBDELLID LEECH FROM THE MOUTH

The genus *Platybdella* was defined by Malm (1863) as follows:

Corpus modice depressum, longiusculum, ante medium constrictione distincta, diaphanum, non in globulum se involvens. Acetabulum oris evidenter discretum. Acetabulum anale oblique affixum, valde discretum. Annuli parum perspicui.

However, it has been determined by Leigh-Sharpe (1916, 1933), Johansson (1929) and Herter (1936) that the genus *Platybdella* as defined above included forms which have since been allocated to other genera.

The leeches found in the ocean pout are platybdellid forms as defined by these later investigators and belong to a group partially characterized by the presence of four eye spots. A comparison of these related forms shows that *P. buccalis* is different in certain respects and is considered a new species for the reasons which are given below.

Platybdella buccalis sp. nov.

(Plate VIII)

These ichthyobdellid leeches were found in the mouth of a single specimen of *Macrozoarces americanus*. They are pinkish in color; body cylindrical or only slightly flattened. Four worms were found measuring 15, 18, 23, and 35 mm. in length. Superficially the body appears smooth, but on closer examination many fine segmentation markings are evident, the exact number of which could not be determined. The primary annuli are deeper than the secondary ones. The anterior sucker is cupuliform and slightly indented on the dorsal border. It measures about 0.5 mm. in width or slightly narrower than the body proper in this region. Four eye spots, semilunar in shape and brownish in color, are present on the dorsal surface of this sucker.

They are closer together on the vertical axis than on the horizontal. There is no sharp demarcation between the anterior sucker and the rest of the body. Similarly, no sharp distinction can be made between the preclitellar, clitellar and abdominal regions of the body, although these regions can be established by careful examination. The body is narrow at the anterior end, widens slightly throughout the extent of the abdomen and then narrows again at the posterior end. The posterior end is not as wide as the region just behind the anterior sucker. The posterior sucker is terminal, comparatively large, also cupiliform, with its border entirely round and smooth. It is sharply separated from the body and measures from 1 to 1.5 mm. in diameter.

The details of the internal organ systems were not determined. The pharynx is provided with a protrusible proboscis, which is one of the distinguishing characters of the order Rhynchobdellae to which these leeches belong. The anal opening lies a short distance anterior to the region where the body and the posterior sucker join. The region around the anal opening is surrounded by numerous large cocoon gland cells which contain granules (secretory?) that stain deeply with para-carmine. According to Leigh-Sharpe (1916) and others, the glands extend from the end of the clitellum to the anus, forming a continuous layer underneath the subdermal muscle. The efferent ducts open to the surface at the region of the clitellum. They "do not, however, run forward independently one from another without arrangement, but are collected into close bundles which, as well in number as in position, are, in most cases, constant in the same species." Although the reproductive glands could be seen, their number and arrangement, as well as the positions of the external openings of their ducts, were not determined.

Platybdella buccalis is characterized, then, as follows. Anterior sucker smaller than posterior sucker, slightly smaller than the body in the anterior region; body more or less smooth, annulations not distinct, no special markings, excrescence, respiratory vesicles or other structures visible; color pinkish; preclitellar, clitellar and abdomen not especially distinct from one another; posterior sucker about twice the width of the anterior sucker, terminal, sharply separated from the body proper; pharynx with a short protrusible proboscis; anus a short distance from the posterior end of the body, surrounded by large gland cells.

Although several species of *Platybdella* were described by Malm

(1863), it has since been shown by the investigators cited above that the forms which rightfully belong to this genus could be reduced to two species: *P. anarrhichae* and *P. quadrioculata*. However, the former is characterized by the absence of eyes. It is commonly found on *Anarrhichas lupus*, and Herter (1936) reported it from gills of *Lycodes pallidus* (Family Zoarcidae) (see also Moore, 1898). *Platybdella quadrioculata* occurs on *Labrus maculatus*, and insofar as is known, it has never been rediscovered since it was originally reported by Malm. According to this investigator this leech is characterized as follows:

Ocelli 4, in series duas longitudinales, antrorsum convergentes, dispositi. Corpus unicolor, postice albo-punctatum. Acetabulum anale fere duplo latius quam acet. oris et 2/3 latitudinis mediae abdominis; concolor.

In some respects *P. buccalis* fits this description, but so do other species which he described and which have since been relegated to other genera (e. g., *P. scorpii* is a synonym of *Janusion scorpii* of Leigh-Sharpe, 1933). In comparing figures, *P. quadrioculata* differs from *P. buccalis* as follows: (1) in general appearance of the body; in *P. quadrioculata* the body is narrow at the anterior end and gradually widens towards the posterior extremity; (2) the oral sucker is more sharply demarcated from the body, and the eye spots are differently spaced; and (3) presence of the white spots. However, again by inspection, the main distinction is based on the size, shape and placement of the anterior and posterior suckers.

A species was described by Moore (1938) from the gill region of *Trematomus hansonii* as *Platybdella levigata* (Harding) (= *Cryobdella levigata*, Harding, 1922). However, this form differs from both *P. quadrioculata* and *P. buccalis* in that the caudal sucker has "a very finely serrated margin of about 160 teeth, the peripheral terminations of as many delicate radiating muscle bands, which are crossed by numerous circular and reticular bands." Other features peculiar to *P. levigata* are as follows: specimens have posterior suckers reversed, i. e., sharply bent dorsad; nuchal constriction deep; primary annuli are much deeper than secondary annuli. However, the last mentioned is also a character of *P. buccalis*.

Abranchus (= *Platybdella*) *sexoculatus* (Malm, 1863) was reported by Johansson (1929) as occurring on *Zoarces viviparus*. As the specific name indicates, the leech possesses six eyes.

SUMMARY

Platybdella buccalis sp. nov., an ichthyobdellid leech from the mouth of an ocean pout (*Macrozoarces americanus*), is described and compared with *P. quadrioculata* Malm (1863) and other related species.

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IV. ON A FIBRO-EPITHELIAL GROWTH ON THE SNOUT

(Plates X A, B)

Neoplastic growths in fishes are not uncommon. For a partial review of the literature the reader is referred to the reports of Schamberg and Lucké (1922), Lucké (1940) and Lucké and Schlumberger (1942). Nigrelli (1938) has outlined the possible causes for fish tumors and hyperplasia.

Certain growths are peculiar to fishes, but many of them are similar to neoplasms found in warm-blooded vertebrates. In general they are usually slow growing and benign; a few have invasive characteristics but rarely are they capable of true metastases. Although any part of the body may be involved in these growths, the skin seems to be most commonly affected. Certain of these skin growths are predominantly epithelial in character, while others are chiefly fibrous or composed of cellular elements other than those derived from the epithelium.

The present contribution deals with a fibro-epithelial growth in which the fibrous tissues predominate.

Gross Description. The growth is reddish and rugose in appearance, but some parts are more or less smooth. There is a slight similarity to certain papillary growths found in other vertebrates. It is spongy in texture and extends from the interorbital region, at the level of the middle of the eyes, to a little beyond and overhanging the dorsal border of the maxillary. The conjunctiva of the anterior surface of the left eye is slightly involved. The denser part of the growth is found on the middle of the snout, overgrowing the external nares. The neoplasm extends laterally on both sides to the suborbital regions. The dorsal and posterior parts of the growth are black, superficially resembling a melanosis.

Histological Description. Microscopically the growth is fibro-epithelial in character, in some respects not unlike the fibro-epithelial growths described by Smith and Coates (1938) on the marine turtle, *Chelonia mydas*. The epithelium varies in thickness from two or three to several layers of cells. Keratin formation is not present. Prickle cells showing intercellular bridges are evident but not especially predominant. The cellular elements, including the mucus cells and those of the sensory organs in the epithelium appear normal in structure, arrangement and distribution. There is no evidence of cytoplasmic or intranuclear inclusion bodies in cells stained for this purpose.

The major portion of the growth consists mainly of intermingling bands of fibrous tissue of varying density. In most regions of the growth the two areas, epithelial and fibrous, are distinctly separated from one another by the corial tissue elements or by an especially thickened band of fibrous tissue as shown in Plate X A. This figure also shows the variation seen in the thickness of the epithelial part of the growth together with the arrangement and distribution of the corial melanophores. Occasionally, the corial elements of this growth show invasive tendencies as seen in Plate X B. Here the neat pattern of the epithelial cells is no longer present. The corial cells, together with the melanophores, infiltrate among the epithelial elements, penetrating the basement membrane and eventually destroying and replacing these cells. A certain amount of infiltration also occurs among the muscle fibers; some of the latter are hyalin in appearance. Many of these features have also been described in the melanomas in *Platypoecilus maculatus-Xiphophorus hellerii* hybrids by Gordon and Smith (1938).

The blood supply is not especially rich. Certain sections show a delicate network of vessels, the walls of which vary as to thickness. No round cells or other special elements associated with an inflammatory reaction were present.

Discussion. It is evident from the above description that the corium is the primary seat of the fibrous part of the growth found on the ocean pout. It is further evident that the cellular elements arising from this region have invasive characteristics. A somewhat similar condition was also noted by Nigrelli and Gordon (1944) for a melanotic tumor on the skin of a silverside (*Menidia*).

The causative factors responsible for this growth were not determined. It is the opinion of the writer that the neoplasm was in all probability due to trauma. This laboratory has several records (Nigrelli, 1938, 1943), the details of which are as yet unpublished, that certain neoplasms appearing in the head and snout of fishes kept in captivity may be induced by injury resulting from hitting the walls of aquaria.

However, the possibility that the fibro-epithelial growth may be due, either directly or indirectly, to one of the several parasites recorded from the skin and flesh of the ocean pout cannot be excluded. Both protozoan and helminthic forms are of frequent occurrence. It is of interest, however, that copepoda have not been found on this fish. Since they have been recorded from a related host species (*Clavella pinguis* on *Lycopodes frigidus*) by Wilson (1932), it is highly probable that similar crustacean parasites may be found on ocean pout (*Macrozoarces americanus*), but as yet they have not come to our attention.

The host response to the various parasites occurring on the skin or in the flesh of the ocean pout is often manifest by the elaboration of fibrous tissues in the attempts to wall off the invading organisms. This reaction of the host to foreign agents is quite evident at times throughout the body of the fish. Beside metacercarial cysts, numerous bodies, some fatty in nature and others composed of granular debris, in all probability the remains of parasites and host cells, are frequently present and always surrounded with a capsule of fibrous tissue. Therefore, it is altogether possible that any of these agents may have provided the stimulus for the fibro-epithelial growth described above.

SUMMARY

A fibro-epithelial growth, predominantly fibrous, from the snout of an ocean pout (*Macrozoarces americanus*), is described and the probable causes of its growth are discussed.

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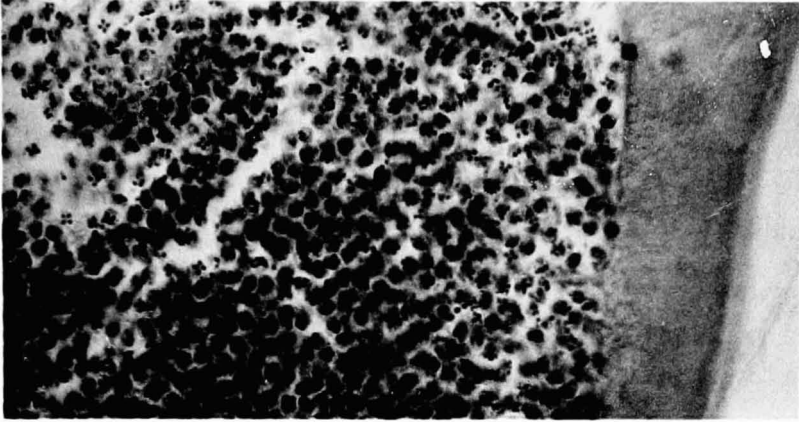
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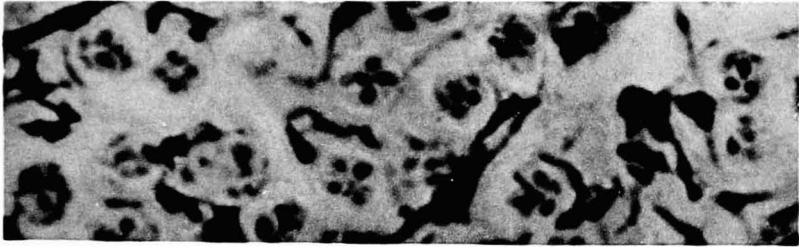
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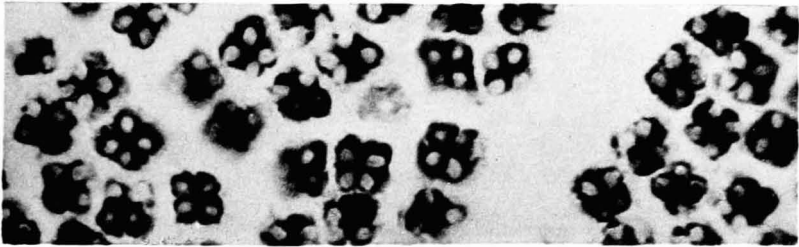
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A



B



C

Plate I. *Chloromyxum clupeiidae* Hahn. A. Photo-micrograph of section through a muscle bundle showing organisms in stages of sporogony. Iron-haematoxylin. About 600X. B. Smear made directly from nodule showing immature spores with capsulogenous and sporoplasmic nuclei. Iron-haematoxylin. About 900X. C. Stained smear showing typical *Chloromyxum* spores with four polar capsules at the anterior end. Iron-haematoxylin. About 1200X.



A



B

Plate II. *Plistophora macrozoarcidis* sp. nov. (Photographs by S. C. Dunton, New York Zoological Society) A. Photograph of fillet of ocean pout (*Macrozoarces americanus*) showing typical lesions resulting from infection with the microsporidian *Plistophora macrozoarcidis*. Note the minute trophozoitic masses (T.M.) lying along the long axis of the muscle bundles. These masses contain developing schizonts. Immature spores and pansporoblasts may be found in the larger lesions. Reduced $\frac{1}{2}$. B. Enlargement of lesion shown above. It is in the dark granular masses seen in this picture that the mature spores are found. About 2X.

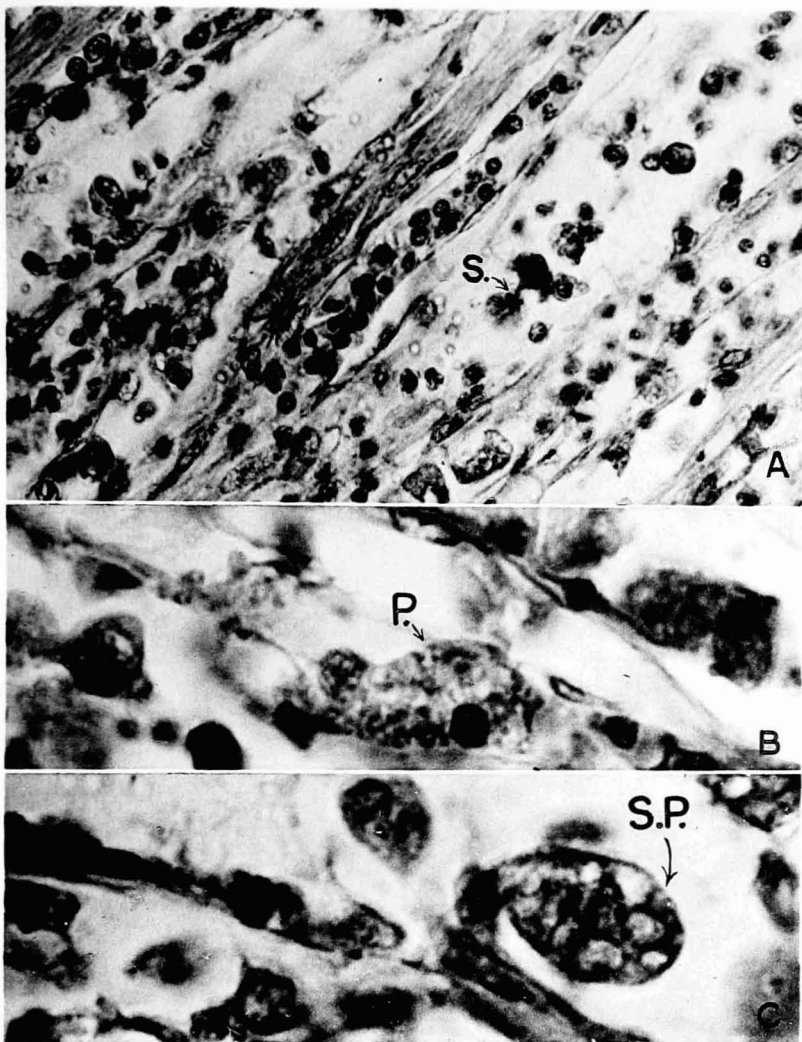
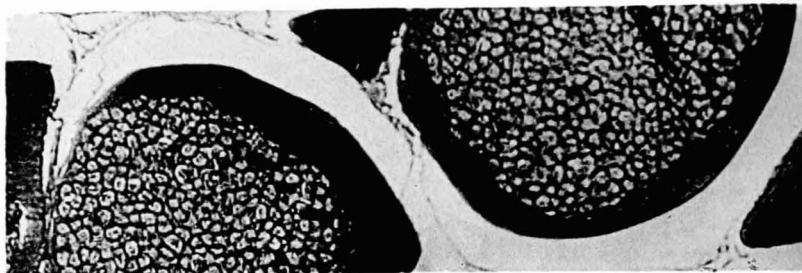


Plate III. *Plistophora macrozoarcidis* sp. nov. (Photographs by S. C. Dunton, New York Zoological Society) A. Schizonts in various stages of development. The cell in "S" is dividing. Delafield's haematoxylin-eosin. About 1500X. B. Pansporoblasts; many of them in the uninucleate stage. Note the cell marked "P". Delafield's haematoxylin-eosin. About 800X. C. Pansporoblasts in the process of sporogony. Some cells have two and others four sporoblasts in the process of being transformed into spores. Note cells marked "S.P." Delafield's haematoxylin-eosin. About 1500X.

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A

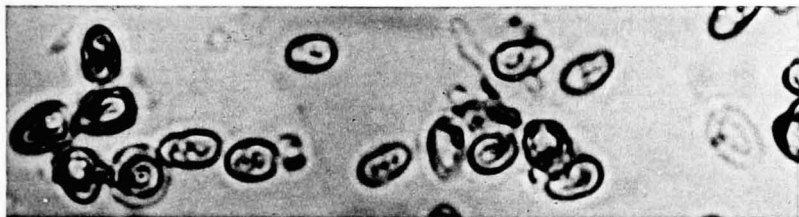


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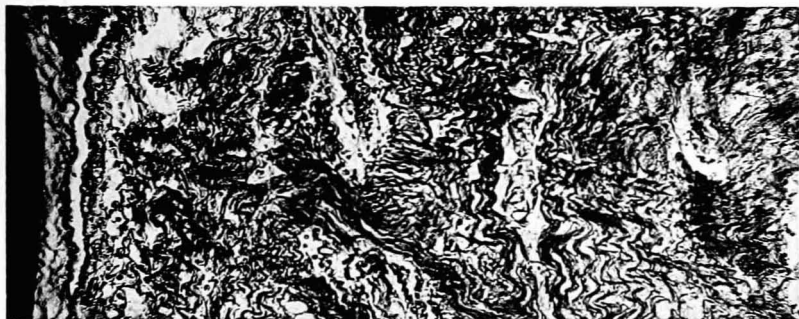
Plate IV. *Plistophora macrozoarcidis* sp. nov. (Photographs by S. C. Dunton, New York Zoological Society) A. Cross section of infected muscle bundles of ocean pout showing "cysts" containing immature spores. Each round body represents a cyst similar to that represented in Text Figure A, 8. Note the hyalin nature of the muscle fibers. These fibers, however, are still intact. Harris' haematoxylin-eosin. About 500X. B. Pansporoblasts in late stage of sporogony. The spores are mature and can be induced to release their polar filaments at this stage. Note the extent of the degeneration of the muscle tissue. Mallory's triple stain. About 500X.



A



B



C

Plate V. *Plistophora macrozoarcidis* sp. nov. (Photographs by S. C. Dunton, New York Zoological Society) A. Mature pansporoblasts dividing. The significance of this division is not known. Some of these spores are released and free among the tissue debris and pansporoblasts. Also among these elements may be found uninucleate cells typical of the schizonts shown in Figure 3. There is some evidence that auto-infection may take place. Delafield's haematoxylin-eosin. About 800X. B. Fresh, unstained spores. Note the variation in size and form. About 1200X. C. Mallory's stained section showing the extent of the development of connective tissue around the developing microsporidians. About 500X.

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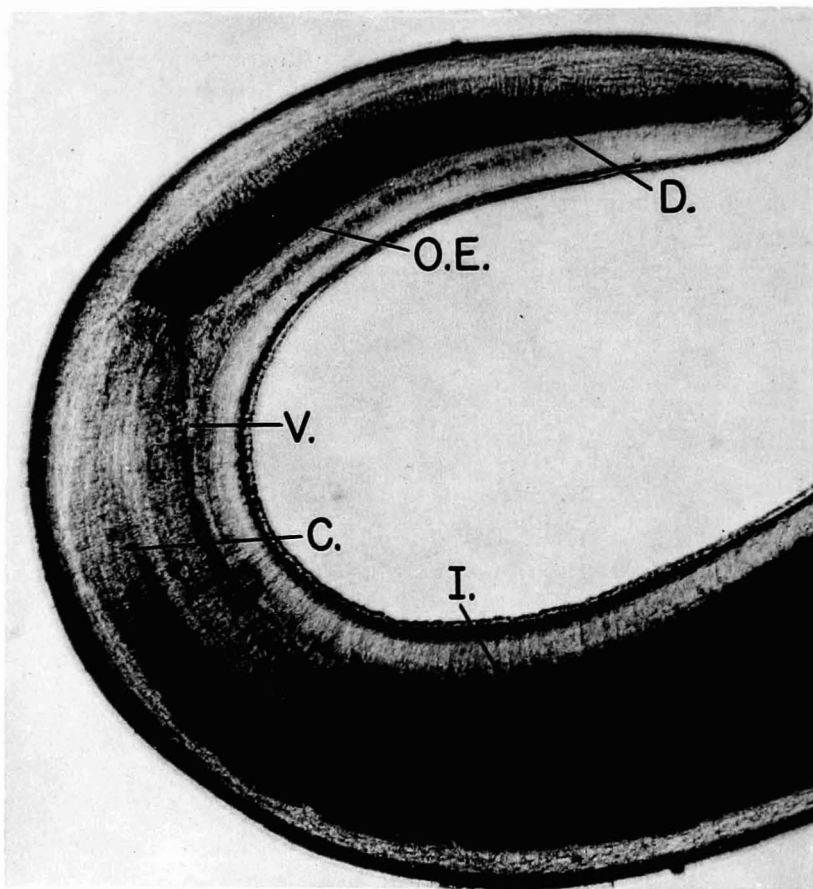


Plate VI. *Porrocaecum decipiens* (?). *Oe.*, oesophagus; *D.*, duct of excretory gland; *V.*, ventriculus, *C.*, intestinal caeca; *I.*, intestine. About 20X.

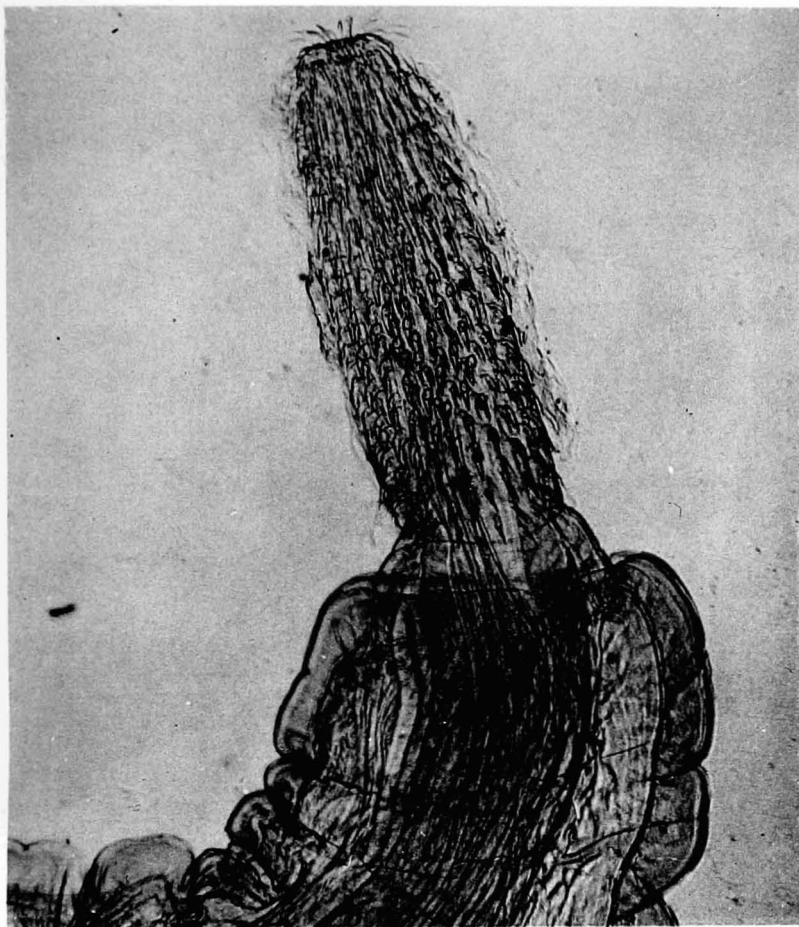


Plate VII. *Echinorhynchus gadi* Zoega. Acanthocephalan from the intestine of *Macrozoarces americanus*. Proboscis fully extruded. Stained with Paracarmin. About 75X.

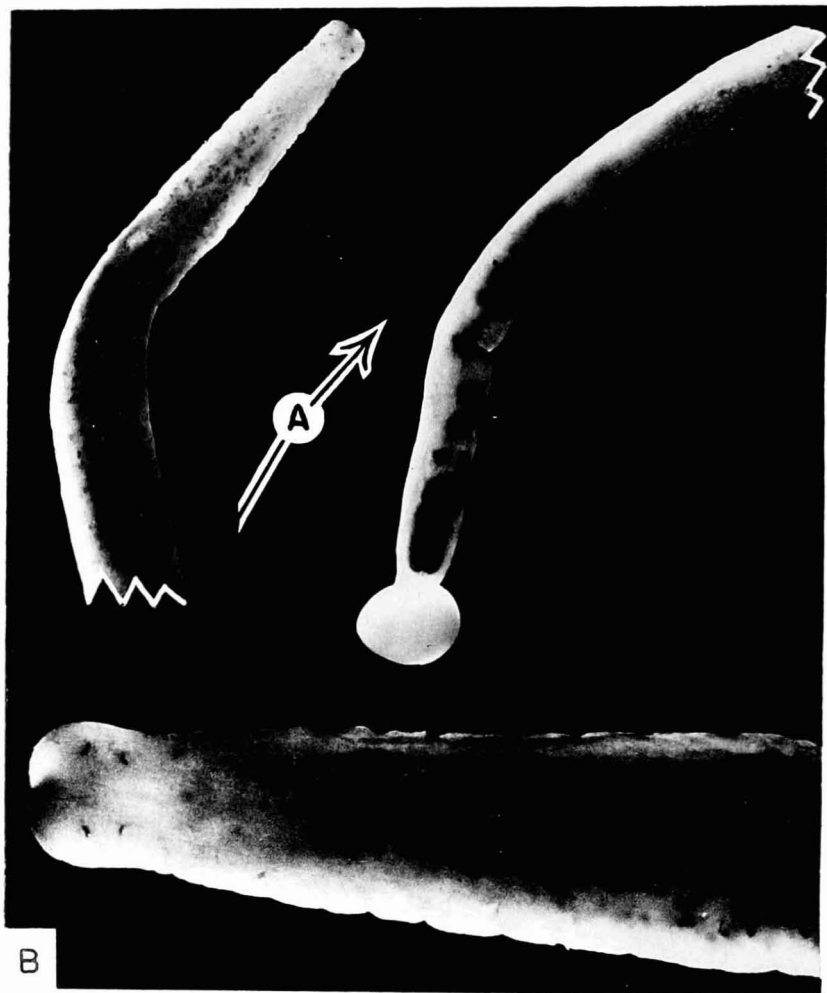
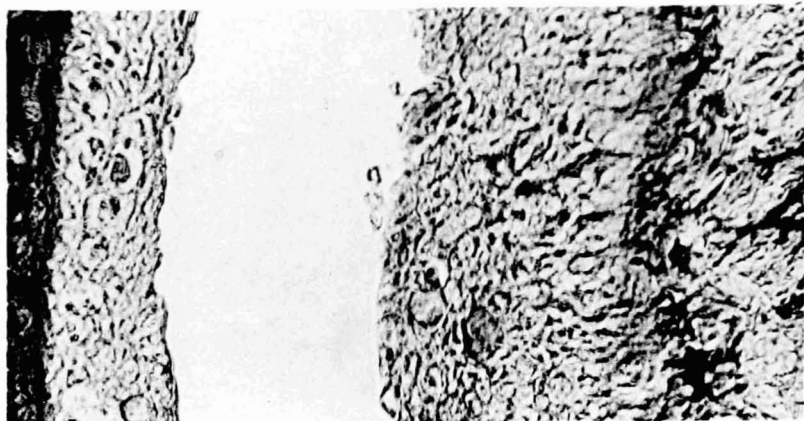


Plate VIII. (Photographs by S. C. Dunton, New York Zoological Society) A. *Platybdella buccalis* sp. nov. 6X. B. Anterior end enlarged to show details of ocelli. Note arrangement of eye spots, oral sucker and primary annuli. 9X.

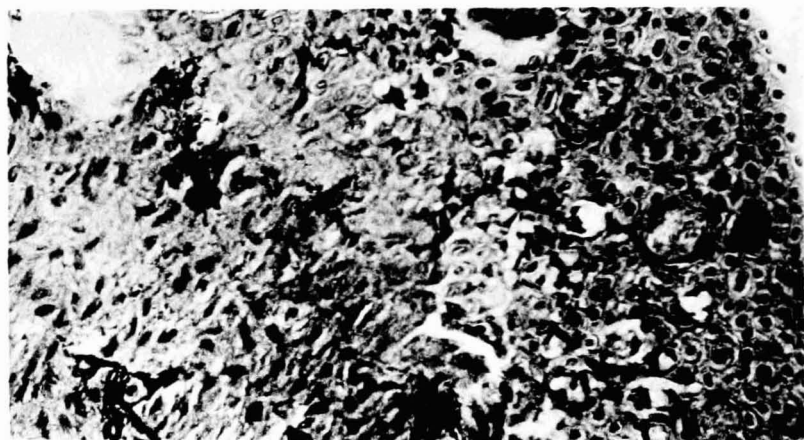


Plate IX. (Photograph by S. C. Dunton, New York Zoological Society)
Photograph of the head of an ocean pout, *Macrozoarces americanus*, showing fibro-epithelial growth. About natural size.

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details of
IX.



A



B

Plate X. *A.* Photomicrograph of a Mallory's stained section of fibro-epithelial growth. Note the variation in thickness of the epithelium, the corial melanophores and the exceptionally thickened band of fibrous tissue. About 950X. *B.* Section showing the corial elements invading the epithelium. Harris' haematoxylin-eosin. About 950X.