

INVESTIGATIONS ON THE PROTOZOAN FISH PARASITES OF THE ST. ANDREW'S REGION.—MARJORIE F. ELLIS, B.A., Zoological Laboratory, Dalhousie University, Halifax, N.S.

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ABSTRACT.

The following myxosporidian parasites were found in marine fishes of the St. Andrew's, N. B., Canada, region: *Ceratomyxa acadensis*; *Myxidium bergense*; *Leptotheca agilis* (new to the region); *Chloromyxum leydigi* (new to the region); *Ichthyophonus hoferi* (new to North America); *Goussia gadi*. The structure and habitat of these forms is described.

The Sporozoa are a group of endoparasites found in almost all sea fishes. In some cases they form conspicuous skin and muscular lesions, but more often they are free-living in the gall bladder and bile ducts of the host.

The trophozoites are generally amoeboid in form and motion. Some forms are attached to the epithelial lining of the cavity they inhabit. Within the class Sporozoa the Myxosporidia show great variation in size, form, structure, and mode of spore formation.

The spores are the agents for the infection of new hosts. Since the spore has a well defined structure it is usually the chief means of identification. Where the spores are not obtainable identification is difficult and sometimes impossible.

In describing the reproduction of individuals and the infection of new hosts, Davis¹ uses the terms multiplicative reproduction and propagative reproduction. The former refers to the increase of the parasites within the host; the latter to the means by which it may infect a new host.

It is still uncertain exactly how multiplicative reproduction takes place in free living forms. Doflein² maintains that it is entirely the result of division of the plasmodia. Cohn³ describes multiplicative reproduction taking place by exogenous budding. Davis¹ has given reasons for believing that the process as observed by Cohn is abnormal and he observed endogenous budding in the Myxosporidian, *Sinuolinea dimorpha*. In

1. Davis—*Bull. U. S. Bur. Fish.*, **35**, 203 (1916).

2. Doflein—*Lehrbuch der Protozoenkunde* 4th Ed. Fischer, Jena. 1916.

3. Cohn—Cited by Davis.

tissue parasites multiplicative reproduction takes place by multiple division.

Propagative reproduction is by means of spores. In the case of free-living forms the spores pass out with the faeces, but in tissue parasites they usually reach the exterior only after the death and disintegration of the host tissue. Following this the spores depend on chance for the infection of new hosts. Infection, as far as can be ascertained, takes place through the digestive tract. The sporoplasm is extruded from the spore in the intestine of the new host and by moving along the lymphatic ducts, or by entering the blood stream, it finally reaches the body organ it is destined to inhabit.

The investigations reported here were carried on at the Atlantic Biological Station, St. Andrew's, N. B., during the summer of 1928. I am very grateful to the Biological Board of Canada for the courtesies extended to me and for the facilities provided for my work at St. Andrew's; also to Professor J. N. Gowanloch for his assistance.

Search for external and intra-muscular parasites, the presence of which would be shown by conspicuous dermal and muscular lesions, was carried on, as well as the study of the internal parasites infecting the gall bladder. Both fresh and stained preparations were studied. The material for the latter was fixed in Bouin's picro-formal solution and stained with Heidenhain's Iron Alum Haematoxylin and Orange G.

The following is a list of the species of fish examined:

- 68 flounder (*Pseudopleuronectes americanus*, Walbaum)
11 gall bladders, 6 infected.
- 33 cod (*Gadus calarias*, Linn.)
1 gall bladder, no infection.
1 swim bladder, infected.
- 8 tomcod (*Microgadus tomcod*, Walbaum)
8 gall bladders, no infection.
- 38 hake (*Urophycis tenuis*, Mitchill)
12 gall bladders, all infected.
- 2 silver hake (*Merluccius bilinearis*, Mitchill)
2 gall bladders, no infection.

- 15 haddock (*Melanogrammus aeglefinus*, Linn.)
11 gall bladders, all infected.
- 16 eel pout (*Zoarces anguillaris*, Peck)
3 gall bladders, 1 infected.
- 1 halibut (*Hippoglossus hippoglossus*, Linn.)
1 gall bladder, no infection.
- 8 sea raven (*Hemitripterus americanus*, Gmelin)
3 gall bladders, 1 infected.
- 13 mummichog (*Fundulus heteroclitus*, Linn.)
13 gall bladders, no infection.
- 6 rock eels (*Pholis gunnellus*, Linn.)
6 gall and 6 urinary bladders, no infection.
- 6 ghost fish (*Cryptacanthodes maculatus*, Storer)
6 gall bladders, 6 infected.
- 8 herring (*Clupea harengus*, Linn.)
8 gall bladders, 3 infected.
- 13 smelt (*Osmerus mordax*, Mitchill)
13 gall bladders, no infection.
- 5 barndoor skates (*Raja stabuliformis*, Garman)
2 gall bladders, no infection.
- 22 skate (*Raja erinacia*, Mitchill)
22 gall bladders, 13 infected.

Negative results followed an examination of blood smears from the following species in a search for protozoan blood parasites.—

Melanogrammus aeglefinus, Linn; *Zoarces anguillaris*, Peck; *Raja diaphenes*, Mitchill; *Raja stabuliformis*, Garman; *Raja scabrata*, Garman; *Gadus calarias*, Linn; *Squalus acanthias*, Linn; *Urophycis tenuis*, Mitchill; *Pseudopleuronectes americanus* Walbaum.

LIST OF THE MYXOSPORIDIA FOUND.

1. *Ceratomyxa Acadiensis*, Mavor.

Trophozoite.—Club shaped with long slender tail. In fresh preparations ectoplasm distinguishable from endoplasm at anterior end. Trophozoites occur in clumps and attached to the epithelial wall of the gall bladder. Free swimming in-

dividuals move with rapid amoeboid motion. Length of vegetative trophozoites up to 40μ , width $5-10\mu$. Spore.—Bivalve, spindle shaped. Valves greatly elongated with very long slender lateral filaments whose total length can only be seen under favourable conditions. Polar capsules almost spherical and lie close to the sutural line. Width of spore $5-7\mu$. Length of spore, excluding lateral filaments $40-50\mu$. Diameter of polar capsules $3-4\mu$. Tri-radiate spores are of frequent occurrence. *Habitat*.—Gall bladders of the following fish: *Pseudopleuronectes americanus*, *Zoarces anguillaris*, *Urophycis tenuis*. Examination of fresh material from *Cryptacanthodes maculatus* and *Clupea harengus* revealed large numbers of parasites tentatively as *Ceacatomyxa acadensis*.

2. *Myxidium Bergense*, Auerbach.

Trophozoite.—Rounded or longish. Occurs as free moving forms or attached to the epithelial lining of the gall bladder of the host. Ectoplasm clearly differentiated from endoplasm. The pseudopodia may be lobose, or fine and filiform for the attachment of the trophozoite. Diameter of trophozoite $30-50\mu$. Polysporous and monosporous. Spore.—Cylindrical. Slightly bent in the form of an "s". Capsules situated at the ends. Spore has a transparent envelope appearing under the microscope as a bright ring. Length of spore $16-19\mu$, width 8μ , length of polar capsules $4-5\mu$, width $2-3\mu$. *Habitat*.—Gall bladders of *Urophycis tenuis* and *Melanogrammus aeglefinus*.

3. *Leptotheca Agilis*, Thelohan.

Trophozoite.—Club shaped, terminated in a point at posterior end. Posterior end sometimes divided into several lobes. Anterior pseudopodia are long, filiform, hyaline, and difficult to see. Trophozoites swim rapidly. Length of trophozoite $50-80\mu$, width $15-20\mu$. Disporous. Spore.—Slightly elongated, sutural line distinct and at right angles to the longitudinal axis. Polar capsules distinct and placed on either side of the sutural line. Sporoplasm distinct, finely granular and nearly filling

the valves. Length of spore 11-12 μ , width 6-7 μ , diameter of polar capsules 2-3 μ . *Habitat*.—Gall bladder of *Raja erinacea*.

4. *Chloromyxum Leydigi*, *Leydigi*.

Trophozoite.—Elongated or oval in form when first placed on the slide, but soon became spherical in shape. Ectoplasm clearly distinguishable from endoplasm in young individuals which move with slow amoeboid motion. Endoplasm characterized by yellow or greenish globules. In sporulating individuals the differentiation between ectoplasm and endoplasm disappears and the movements of the trophozoites cease. Spore formation was observed to take place in an individual within three hours after removal from the host. Clear globules first appeared in the endoplasm within each of which a single spore developed. Diameter of trophozoite 15-18 μ . Polysporous. *Spore*.—Ovoid and having a spore case characterized by large striae which give the posterior border of the shell a dentate appearance. Four polar capsules situated at the anterior end. Length of spore 10-12 μ , width 8-10 μ . Length of polar capsules 5 μ , width 2 μ . *Habitat*.—Abundant in the gall bladder of *Raja erinacea*.

5. *Goussia Gadi*, *Fiebiger*.

This parasite was found in the swim bladder of an emaciated cod, *Gadus calarias*. When opened the swim bladder was found to be filled with a white creamy mass. Microscopic examination showed many spores arranged in groups of four within a membrane, probably the remains of a host cell. Each spore contained two valves and the sporoplasm was very distinct. The form is reported by Mavor⁴ from the air-bladder of *Melanogrammus aeglefinus*. Length of spore 12 μ , width of spore 7 μ .

6. *Unidentified Ceratomyxa*.

Trophozoites of this form were found in the gall bladder of *Hemitripterus americanus*. Although somewhat larger they

4. Mavor—*Contrib. Can. Biol.* 1911-1914
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closely resemble the trophozoites of *Ceratomyxa acadensis*. No spores were present so identification of the species was impossible.

One of the forms found, *Ichthyophonus hoferi* is, so far as can be ascertained, a new record for North America. *Chloromyxum leydigi* and *Leptotheca agilis* have not previously been reported from the St. Andrew's region.

In the course of the investigations carried on at St. Andrew's approximately 500 fish were examined and no instances of external lesions were found. This confirms the work of Mavor⁴ in this region, and stands in contrast to the condition at Woods Hole, where such cases are fairly frequent in the same species of fish that were examined at St. Andrew's.

EXPLANATION OF FIGURES.

Ceratomyxa acadensis.

- Fig. 1. Group of trophozoites drawn from stained smear. X950.
- Fig. 2. Spore without lateral filaments. X950.

Myxidium bergense.

- Fig. 3. Attached trophozoite drawn from section of wall of gall bladder of haddock. X950.
- Fig. 4. Unattached trophozoite drawn from section. X950.
- Fig. 5. Trophozoite showing filiform pseudopodia for attachment. X950.
- Fig. 6. Spore of *Myxidium bergense*. X950.
- Fig. 7. Unidentified trophozoite of *Ceratomyxa* from the gall bladder of *Hemirhamphus americanus*. X950.

Leptotheca agilis.

- Figs. 8. and 10. Trophozoites. X950.
- Fig. 9. Spores. X950.

Chloromyxum leydigi.

- Fig. 11. Sporulating trophozoite. X 440.
- Fig. 12. Young trophozoite. X950.
- Fig. 13. Trophozoite showing pseudopodia. X 440.
- Fig. 15. Spore. X 950.
- Fig. 16. Trophozoite showing clear spheres in which the spores develop. X950.



