On a New Species of *Ichthyophthirius* found in Marine Fishes

(Contribution to the biological study of the diseases and parasites of fish in Japan. No. 2)

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# 日本産魚類の疾病と寄生虫の生物学的研究(2)

海水魚に見出された Ichthyophthirius の1新種について

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Of the parasitic ciliate, *Ichthyophthirius* found in the gill and skin of fish only one species *I. multifiliis* Fouquet, 1876<sup>1,2)</sup> is known up to the present. *Chromatophagus parasiticus* Kerbert, 1884<sup>3)</sup> and *I. cryptosomus* Zacharias, 1892<sup>4)</sup> are regarded as synonyms of *I. multifiliis*.

Similar parasites were found by several authors<sup>3,5,6,7)</sup> in marine fishes, too, but they were neither observed in detail nor recorded as different species. Schaeperclaus<sup>5)</sup> regarded the latter as *I. multifiliis*, recognizing however as the exception to the fact that it has somewhat longer cilia than the former.

In 1936 the author <sup>8,9)</sup> found a similar parasite in various marine fishes kept in the aquarium of the Institute for Fisheries of the Tokyo Imperial University in Aiti Prefecture, Japan, in a severe diseased condition; e. g. The White-Spot Disease in Marine Fishes.

As the author previously reported, this marine parasitic ciliate resembles the freshwater form, *I. multifiliis* in general appearance, but it differs distinctly from the latter not only in the habit obligatorily adapted to sea water and marine fishes, but also conspicuously in the shape of the nucleus.

As the parasite seems to have been hitherto unrecorded, the author gives a description of it under the name of *Ichthyophthirius marinus*.

### Ichthyophthirius marinus nov. spec.

Shape: The ciliate is egg-shaped, somewhat slender in young specimen and more round in mature. Carefully observed in the specimen swimming freely in water, the ciliate shows a contour in a lateral view resembling the seed of a persimmon, directing its cytostome foreward, which is at the anterior and more pointed end of the body. In this state the body is slightly slender posteriorly. In contrast with it the young specimen parasitic in the skin of fish has a slender anterior part, which is adapted to boring the host's tissue. Furthermore, the youngest specimen just left from cyst after multiplication provides almost the exact shape.

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In the host' tissue, in epidermis, or between epidermis and cutis, the young parasite moves actively showing various appearances; round, ovoid, long-ovoid, sometimes bent in a U-shape and irregular.

The shape of the transverse section of the body is almost round.

Size: The size of the 60 living parasites in the host' tissue were measured. The largest measured 452  $\mu$  in length and 360  $\mu$  in breadth, the smallest measured 66  $\mu$  in length and 34  $\mu$  in breadth. On the other hand, the youngest specimen just left from the cyst measured 65  $\mu$  in length and 35  $\mu$  in breadth (under a water-temperature of 20—23°C).



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Cilia: The body surface of the ciliate is fine streaked longitudinally. The short  $(7-8\,\mu)$  fine cilia are ordered in these streaks, which are 30-40 in number in a lateral view both in the young and the old.

Plasma: Ecto- and endoplasm are comparatively distinct; in living specimen the former appears clear and is thickest in the anterior end, especially in the part of mouth opening; the latter occupies the most parts of the content of the body and is filled with numerous granules, small food particles and contracting vacuoles. In many specimens the melanin-mass taken from the host's melanophores are found.

Nuclei: In most cases 5—6 small granules like micronuclei strongly stained with haematoxylin are recognized. Some of them may originate from the nuclei of the host's tissue cells, but the exact characteristics are not yet known in detail.

The macronucleus is located in the posterior half of the body and is so constituted that 4 (rarely 5-8) elliptical swellings are linked together in a U-form with a combining string. The macronucleus, both swellings and string, is strongly stained with haematoxylin. On an average the length of the elliptical swellings is somewhat larger than one fifth of the body length in the young specimens and somewhat smaller in the old ones. In the living specimen it is somewhat hard to observe macronucleus in Ichthyophthirius marinus, because of the thick protoplasma and the complicated structure of it, while it is always easily recognizable in multifiliis, as a distinct horse-shoe shape. In the living condition when the ciliate is slightly pushed between two glass plates (cover- and slide glasses) and the thick protoplasma is pushed out from the body to a certain degree, the macronucleus of I. marinus is easily observed as a light, clear figure with the above mentioned complicated shape. The ciliate was then cut  $10\,\mu$  thick and stained with Heidenhain's ironhaematoxylin. In the preparate the macronucleus showed the structure consisted of many achromatic bladders, in each of which a minute chromatic granule is contained. In the formation of the cyst the macronucleus often shows the shape of a long string, and the bladders and the chromatic granules at the same time extend along the string conspicuously.

Mouth: The mouth opening lies near the anterior end of the body, and is provided with a comparatively large field of the peristome and the short cytopharynx. Sometimes a more or less long food vacuole exists, which treads deep into the body.

Multiplication: The multiplication occurs regularly after the full-grown ciliate falls out the epidermis of the host fish. This can happen however after reaching quite different sizes of the body when the host died. The fallen out ciliate sinks to the bottom, adheres to the suitable substance and forms a cyst. Within the cyst it multiplies by division and a large number of minute young are produced in a similar way already known in the fresh water species, *I. multifiliis*. In the marine species the membrane of the cyst which is formed by the lapping of about 10 layers of very thin lamallae shows a far thicker and tougher appearance than the fresh water ones. When cut and stained with Delafield's haematoxylin and eosin about 6 outer layers of the completed cyst are somewhat thicker and retain more colar of eosin than the inner layers. The cyst is nearly round and measures  $90-400~\mu$  in diameter, average  $200-300~\mu$ .

Though the cytological study of the multiplication has not yet been carried out, it seems to me that the division in the cyst is somewhat uncommon. Through the first division the ciliate seems to divide in one small and in one large cell. Several following divisions occur only in the small cell, from which about 16 small cells arise. After that the large cell follows the division. After some hours (about 24 hours in a few examples in the dishes in the laboratory under an air temperature of 20-23C°) the cyst is filled with small cells measuring  $85\,\mu$  in length and  $70\,\mu$  in breadth on an average. The divisions continue further, and then 50-70 small cells can be seen in the external appearance of the cyst (in a cyst, of which the diameter measures  $200-300\,\mu$ ) These small cells measure about  $40\,\mu$  in length and about  $30\,\mu$  in breadth on an average. These cells always move lively. In such a way the young are formed. When the young slip out from the cyst its wall is not broken up and remains over a year in an unchanged appearance. How the young slip out from the cyst is not yet exactly known.

The young measures  $65\,\mu$  in length,  $35\,\mu$  in breadth and resembles the youngest parasite found on fish in the shape of macronucleus, in length of cilia  $(7-8\,\mu)$  and in some other common shapes. The number of the young formed in the cyst differs conspicuously according to the size of the cyst, which is related to the size of the ciliate left the skin of the fish. In a cyst which measured  $200-300\,\mu$  in diameter the number of young reached 100 or more under the same condition in a dish in the laboratory.

The above mentioned process of multiplication of this pathogenic ciliate is almost the same in principle to that of *I. multifiliis*. The division of the parasite in the skin of the host fish does not usually occur.

Location: The parasite situates mostly between epidermis and cutis, and rarely in the loose tissue of the scale-pocket after the inflamation.

Host: Many species of marine fishes.

Pathological: The ciliate parasitizes in the gill and the skin of fish, creeps about between epidermis and cutis, and causes an acute inflamation over the entire surface of the body. Suffocation caused by reducing the respiratory surface in the gills is the commonest result.

Distribution: This ciliate probably is world-wide. It was found in the adjacent waters around Japan, from Hokkaido to Kyūshū.

## 抄 録

淡水性白点病の病原繊毛虫 Ichthyophthirius multifiliis Fouquer, 1876 に酷似した繊毛虫が鹹水性白点病の場合に認められることはすでに周知の事実である。両者の種の異同については、すでに四竈 (1937, 1938) が大核の形態の著しい差異を指摘したが、口部の形態その他について更に精査の必要を認めたので、新種としての発表を行なわなかった。その後 Schaeperclaus, W. (1954) は両者を同種と見做したが、これは精密な観察を欠き、確証がない。殊に日本産のものも含めているが、これは明らかに誤りであろう。微細構造に関してなお研究すべきものを多く残しているが、これが未知の独立種であることはほぼ誤りないと考えられるので、ここに上述のごとく新種としての記載を行なった。 I. multifiliis との著しい差は淡水と鹹水という棲息水域の差、従っていうま

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でもなく罹病魚種の差,虫の形態では淡水種では大核が馬蹄状ないし三日月状であるのに対し,鹹水種は四連念珠状であること,さらに胞嚢の膜が淡水種では極めて薄いのに対し、鹹水種では極めて厚いことなどである。

本種は海産であることに因んで *Ichthyophthirium marinus* と命名された。その分布は日本では鹿児島県桜島から北海道小樽におよぶが、Kerbert 1884、Buschkiel 1936、Schaeperclaus 1954などの研究に徴するに、恐らく世界的に広いものと考えられる。

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