

Prevalence of skin pseudotumors in three species of pleuronectids in Notsuke Bay, Hokkaido, northern Japan

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Abstract *Pleuronectes schrenki* has been reported to bear skin pseudotumors in Notsuke Bay, Hokkaido, northern Japan, but the past identification of the fish from this bay has remained uncertain. Thus, pleuronectids were collected from the bay and, after exactly identified, they were examined for the presence of skin pseudotumors. Three species of the genus *Pleuronectes* (*P. schrenki*, *P. obscurus*, and *P. pinnifasciatus*) were collected, and prevalence of skin pseudotumors varied between these species. The disease was most frequently found in *P. obscurus*, but was very rare in *P. schrenki* and *P. pinnifasciatus*, which suggests that the fish reported previously as *P. schrenki* from the bay were not the species but *P. obscurus*. In *P. obscurus*, prevalence of the disease was lower in larger fish (150–236mm in standard length [SL]) than in smaller fish (65–149 mm SL).

Key words: Notsuke Bay, *Pleuronectes obscurus*, *Pleuronectes pinnifasciatus*, *Pleuronectes schrenki*, pleuronectids, skin pseudotumors

INTRODUCTION

To date, seven species and a hybrid of the family Pleuronectidae (Actinopterygii: Pleuronectiformes) have been reported to bear skin pseudotumors in coastal waters of Hokkaido, northern Japan: they are *Pleuronectes schrenki* (Schmidt, 1904) (as *Limanda schrenki*); *Pleuronectes obscurus* Herzensteini, 1890 (as *Liopsetta obscura*); *Platichthys stellatus* (Pallas, 1787); *Verasper moseri* Jordan and Gilbert, 1898; *Hippoglossoides pinetorum* (Jordan and Starks, 1904) (as *Cleisthenes pinetorum herzensteini*); *Hippoglossoides dubius* Schmidt, 1904; *Kareius bicoloratus* (Basilewsky, 1855); and a hybrid of *P. stellatus* and *K. bicoloratus* (Awakura, 1974; Oishi *et al.*, 1976; Stich *et al.*, 1977; Yamazaki *et al.*, 1978a, 1978b; Shinkawa and Yamazaki, 1983; Katsura *et al.*, 1984; Fujimoto *et al.*, 1986; Kato *et al.*, 1990; Freeman, 2009; Freeman *et al.*, 2011; Nagasawa and Nishiuchi, 2012). The skin pseudotumors are histologically characterized by X-cells (Stich *et al.*, 1977; Yamazaki *et al.*, 1978a, 1978b; Katsura *et al.*, 1984; Fujimoto *et al.*, 1986; Kato *et al.*, 1990; Freeman, 2009; Freeman *et al.*, 2011) and classified as epidermal papillomas (Oishi *et al.*, 1976; Yamazaki *et al.*, 1978a, 1978b; Fujimoto *et al.*, 1986), X-cell pseudotumors (Kato *et al.*, 1990), epidermal pseudotumors (Freeman *et al.*, 2011), or skin pseudotumors (Nagasawa and Nishiuchi, 2012). According to Freeman *et al.* (2011), the X-cell parasite causing epidermal pseudotumors in *P. obscurus* from Notsuke Bay, Hokkaido, is the same agent to cause the same disease condition in *H. dubius* in other waters of Japan (see Miwa *et al.*, 2004).

Notsuke Bay is located in eastern Hokkaido and is known to have populations of pleuronectids

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bearing skin pseudotumors. Oishi *et al.* (1976) first reported the disease in *P. schrenki* (as *Limanda schrenki*) collected in Notsuke Bay (as Odaito Inlet), but there was a problem in fish identification. Because it was difficult to identify "*L. schrenki*" and "*Liopsetta* sp.," the authors did not separate these species from each other and reported them simply as "*L. schrenki*." They stated that their "*L. schrenki* may also include *Liopsetta* sp." Subsequently, Stich *et al.* (1977) found the disease in *P. schrenki* (as *L. schrenki* [sic]) from eastern Hokkaido including Notsuke Bay, and Yamazaki *et al.* (1978b) also collected the species (as *L. schrenki*) in the bay (as Odaito Bay), but all of these authors did not refer to the identification of the fishes examined. On the other hand, while Katsura *et al.* (1984) collected *P. schrenki* and *P. obscurus* (as *L. schrenki* and *L. obscura*, respectively) bearing skin pseudotumors in Notsuke Bay (as Odaito Bay) and adjacent waters, these authors showed prevalences of the disease only in *P. schrenki*. Most currently, Freeman *et al.* (2011) sampled *P. obscurus* affected by skin pseudotumors in Notsuke Bay. The indistinct fish identification found in the past papers raised a question which species of pleuronectid was virtually affected by skin pseudotumors in the region. The question appears important in terms of epidemiology and host specificity of the X-cell parasite. Thus, the present study was intended to exactly identify the fish affected by skin pseudotumors and to determine the prevalence of the disease among three species of pleuronectids caught in Notsuke Bay.

MATERIALS AND METHODS

Notsuke Bay (43°36'N, 145°13'E) is a small shallow (mostly < 2 m depth) cove with a surface area of over 3,500 ha, facing the Nemuro Strait connecting the North Pacific Ocean with the Sea of Okhotsk. Fishes were collected with a small trawl net operated from a chartered fishing vessel from May to December 1983 and from April to May 1984. No collection was made from January to March 1984 because of the ice cover of the bay. Fishes were fixed in 10% formalin immediately after capture and brought to the laboratory of the Hokkaido Fisheries Experimental Station, Kushiro, where they were identified, measured for standard length (SL, mm), and examined for the presence or absence of skin pseudotumors. Pleuronectids, in particular two morphologically similar species *P. schrenki* and *P. obscurus*, were carefully identified, using Matsubara (1963) and Ueno (1965), on the basis of various morphological characters, such as the shape of the pharyngeal teeth and the lateral line, and the presence or absence of a skin flap on the interopercle. When skin pseudotumors were found, their position was recorded. The scientific names of fishes used in this paper follow Nakabo (2013).

RESULTS

Three species of pleuronectids were collected in this study: *P. obscurus* (n=70, 65–236 [mean 153] mm SL), *P. schrenki* (n=52, 66–231 [140] mm SL), and *Pleuronectes pinnifasciatus* Kner, 1870 (n=49, 93–255 [183] mm SL). Skin pseudotumors were most frequently (n=12, 17.1%) found in *P. obscurus* (Fig. 1), but this disease was very rare in *P. schrenki* (n=1, 1.9%) and *P. pinnifasciatus* (n=1, 2.0%).

In *P. obscurus*, prevalence of skin pseudotumors was as high as 25.0% and 27.3% in fish of 65–99 mm SL (n=8) and 101–149 mm SL (n=22), respectively, but decreased to 9.4% in fish of 150–199 mm SL (n=32) and 12.5% in fish of 200–236 mm SL (n=8). The number of pseudotumors on an affected fish ranged from 1–7 (mostly 1, mean 2.2). Of the 26 pseudotumors found, 15 (57.7%) occurred on the fins, followed by the trunk (n=8, 30.8%) and others including the head and the caudal peduncle (n=3, 11.5%).

A single pseudotumor was found on the caudal fin of the eyed side in *P. schrenki* (91 mm SL) and also on the skin posterior to the pectoral fin of the blind side in *P. pinnifasciatus* (177 mm SL). This is



Fig. 1. A formalin-fixed specimen (abocular side) of *Pleuronectes obscurus*, 141 mm SL, bearing skin pseudotumors (arrowheads) on the head, the dorsal fin, and a skin region of its base. The fish was collected in Notsuke Bay on July 8, 1983. Scale bar: 30 mm.

the first finding of skin pseudotumor in *P. pinnifasciatus*.

DISCUSSION

There are several papers to report prevalence of skin pseudotumors in pleuronectids caught in Notsuke Bay. Oishi *et al.* (1976, table 1) found this disease in 55 (7.9%) of 699 pleuronectids taken from December 1974 to May 1975. While these authors used *L. schrenki* (= *P. schrenki*) as the scientific name of the fish examined, their fish identification was uncertain. Later, Katsura *et al.* (1984) reported that 12.3% of 349 *P. schrenki* from the bay were affected by the disease. The results in these studies definitely differ from those in the present study, which is presumably due to misidentification of the fishes examined in the previous studies. In other words, it is highly likely that the pleuronectids previously reported as *P. schrenki* from Notsuke Bay are not the species but *P. obscurus*. Most currently, Freeman *et al.* (2011) used *P. obscurus* as the scientific name of pleuronectid with skin pseudotumors from Notsuke Bay.

Prevalence of skin pseudotumors varied among the three species of pleuronectids in Notsuke Bay, where *P. obscurus* showed highest prevalence of the disease. Similar differences in prevalence among different pleuronectid species have been reported in Hokkaido (Katsura *et al.*, 1984) and North America (McArn *et al.*, 1968; McArn and Wellings, 1971; Stich *et al.*, 1976). It is difficult at the moment to explain what factors were responsible for the observed differences in prevalence between pleuronectid species, but one of the possible explanations is that susceptibility and response of pleuronectids to the X-cell parasite may vary among species.

Prevalence of skin pseudotumors decreased with increasing body size of *P. obscurus*. This trend is consistent with the results of other observations in Hokkaido (Oishi *et al.*, 1976; Kato *et al.*, 1980; Nagasawa and Nishiuchi, 2011) and North America (McArn and Wellings, 1971; Wellings *et al.*, 1976). Based on the field and laboratory studies of skin pseudotumors of *P. stellatus*, Campana (1983) suggested that such an age-related decrease in pseudotumor prevalence is due to mortality of affected fish. Kato *et al.* (1990) also stated that within the same age-groups, pseudotumor-bearing individuals of *P. stellatus* were much smaller and fed less abundantly than those without pseudotumors. Recently, Nagasawa and

Nishiuchi (2012) suggested that small individuals of the species may die due to the disease in a brackish-water lake, northern Japan. As the relationship between skin pseudotumors and size (age) of pleuronectids is yet poorly understood, we need more work to assess the impact of the disease on pleuronectids at both individual and population levels.

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REFERENCES

- Awakura, T., 1974. [On ganbe-karei]. *Fishes and Water*, (10): 48-50. [In Japanese].
- Campana, S. E., 1983. Mortality of starry flounders (*Platichthys stellatus*) with skin tumors. *Canadian Journal of Fisheries and Aquatic Sciences* **40**: 200-207.
- Freeman, M. A., 2009. X-cell parasites in the European dab *Limanda limanda* are related to other X-cell organisms: a discussion on the potential identity of this new group of parasites. *Parasitology* **136**: 967-980.
- Freeman, M. A., Eydal, M., Yoshimizu, M., Watanabe, K., Shin, A. P., Miura, K., Ogawa, K., 2011. Molecular identification and transmission studies of X-cell parasites from Atlantic cod *Gadus morpha* (Gadiformes: Gadidae) and the northern black flounder *Pseudopleuronectes obscurus* (Pleuronectiformes: Pleuronectidae). *Parasites & Vectors* **4**: 15. doi:10.1186/1756-3305-4-15.
- Fujimoto, Y., Madarame, H., Yoshida, H., Moriguchi, R., Kodama, H., Iizawa, H., 1986. Pathomorphological observations on epidermal papilloma of flatfish (*Liopseta obscura*). *Japanese Journal of Veterinary Research* **34**: 81-103.
- Kato, T., Masahito, P., Ishikawa, T., Minami, T., 1990. The X-cell pseudotumor found on the starry flounder, *Platichthys stellatus*, in the [sic] Lake Mokoto, the [sic] eastern Hokkaido. *Bulletin of the Hokkaido National Fisheries Research Institute*, No. 54: 33-49. [In Japanese with English abstract].
- Katsura, K., Yamazaki, F., Hamada, K., Oishi, K., Harada, T., Shinkawa, T., 1984. Geographic distribution and frequency of tumorous fishes collected from the coastal waters of Hokkaido, Japan. *Bulletin of the Japanese Society of Scientific Fisheries* **50**: 979-984.
- Matsubara, K., 1963. *Fish Morphology and Hierarchy. Second Edition*. Ishizaki Shoten, Tokyo: 1605 pp. 135 pls. [In Japanese].
- McArn, G. E., Chuinard, R. G., Miller, B. S., Brooks, R. E., Wellings, S. R., 1968. Pathology of skin tumors found on English sole and starry flounder from Puget Sound, Washington. *Journal of the National Cancer Institute* **41**: 229-242.
- McArn, G. E., Wellings, S. R., 1971. A comparison of skin tumors in three species of flounders. *Journal of the Fisheries Research Board of Canada* **28**: 1241-1251.
- Miwa, S., Nakayasu, C., Kamaishi, T., Yoshiura, Y., 2004. X-cells in fish pseudotumors are parasitic protozoans. *Diseases of Aquatic Organisms* **58**: 165-170.
- Nagasawa, K., Nishiuchi, S., 2012. Prevalence of skin pseudotumors in starry flounder (*Platichthys stellatus*) and pleuronectid hybrids in a brackish-water lake, Hokkaido, Japan. *Biosphere Science* **51**: 9-14.

- Nakabo, T., ed., 2013. *Fishes of Japan with Pictorial Keys to the Species. Third Edition*. Tokai University Press, Hadano: 2428 pp. [In Japanese].
- Oishi, K., Yamazaki, F., Harada, T., 1976. Epidermal papillomas of flatfish in the coastal waters of Hokkaido, Japan. *Journal of the Fisheries Research Board of Canada* **33**: 2011-2017.
- Shinkawa, T., Yamazaki, F., 1983. [On MDH isozymes specific to the epidermal papilloma of flatfish and Japanese goby]. *Fish Genetics and Breeding Science* (8): 30-33. [In Japanese].
- Stich, H. F., Acton, A. B., Forrester, C. R., 1976. Fish tumors and sublethal effects of pollutants. *Journal of the Fisheries Research Board of Canada* **33**: 1993-2001.
- Stich, H. F., Acton, A. B., Oishi, K., Yamazaki, F., Harada, T., Hibino, T., Moser, H. G., 1977. Systematic collaborative studies on neoplasms in marine animals as related to the environment. *Annals of the New York Academy of Sciences* **298**: 374-388.
- Ueno, T., 1965. [Fishes in coastal waters of Hokkaido. (2). Bastard halibuts, righteye flounders, and tong soles]. *Journal of Hokkaido Fisheries Experimental Station* **22**(2): 10-29. [In Japanese].
- Wellings, S. R., McCain, B. B., Miller, B. S., 1976. Epidermal papillomas in Pleuronectidae of Puget Sound, Washington. Review of the current status of the problem. *Progress in Experimental Tumor Research* **20**: 55-74.
- Yamazaki, F., Hibino, T., Oishi, K., Harada, T., Stich, H. F., Acton, A. B., 1978a. X-cell morphology in the epidermal papillomas of flatfish collected from coastal waters of Hokkaido, Japan. *Bulletin of the Japanese Society of Scientific Fisheries* **44**: 407-413.
- Yamazaki, F., Hibino, T., Oishi, K., Harada, T., 1978b. Morphological changes in the the X-cells in epidermal papillomas of the flatfish *Limanda schrenki*. *Bulletin of the Japanese Society of Scientific Fisheries* **44**: 723-727.

北海道野付湾産カレイ類3種における皮膚偽腫瘍の発生状況

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要 旨 北海道東部にある野付湾のクロガシラガレイ *Pleuronectes schrenki* には皮膚偽腫瘍と呼ばれる病変が見られることが知られているが、魚種の同定に問題があることが指摘されていた。そこで、野付湾からカレイ類を採集して種を正確に同定し、本病変の発生状況を調べた。その結果、本病変はクロガレイ *Pleuronectes obscurus* に高頻度に認められたが、クロガシラガレイには低頻度にしかな認められず、過去にクロガシラガレイと報告された種はクロガレイであると示唆された。病変の発生頻度は、クロガレイの魚体長の増加とともに低下した。また、病変はトウガレイ *Pleuronectes pinnifasciatus* にも極めて低頻度に認められた。トウガレイにおける本病変の確認は本報告が最初である。なお、前報 (Nagasawa and Nishiuchi, 2012) の和文要旨で、オシヨロガレイをヌマガレイとスナガレイの雑種と記したが、正しくはヌマガレイとイシガレイの雑種であり、ここで訂正する。

キーワード: カレイ類, クロガシラガレイ, クロガレイ, トウガレイ, 野付湾, 皮膚偽腫瘍