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Formation of pseudo-bands on the otoliths of walleye pollock *Gadus* chalcogrammus juveniles at high water-temperature

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Introduction

Walleye pollock Gadus chalcogrammus is an important commercial species in waters around northern Japan. Stock assessments of this species often rely on an age-dependent model (e.g., Yamashita et al., 2017[1]), thus accurate age determination is an important factor in their management. Counts of translucent annual bands in the otoliths are generically used as an indicator of fish age, including in walleye pollock (reviewed in Nishimura, 1993[2]). These bands are normally formed in winter, but similar pseudo-bands (translucent zones) occasionally appear in other seasons, and especially during the juvenile phase. Because a misreading of pseudo-bands could hinder precise determinations of age in this species, we examined sagittal otoliths for the formation of a translucent zone among juveniles kept in two different water-temperature regimes in the laboratory.

Materials and methods

Fertilized eggs of walleye pollock were collected from a broodstock tank at the Akkeshi Laboratory of the Hokkaido National Fisheries Research Institute, in January 2016, and the hatched-out larvae were reared to the juvenile stage. Two 4 000-L tanks were prepared, and 1 500 juveniles were added to each tank on June 9. The juveniles were fed to satiation with formula feed (Otohime C2 and Otohime EP1, Marubeni Nisshin Feed Co., Japan) until October 27; meanwhile, the two tanks were exposed to different water-temperature regimes: 1) natural temperature fluctuations (NT tank, not artificially controlled), and 2) controlled temperature (CT tank, maintained at 12°C from July 21 onwards). At 1- or 2-week intervals, approximately 20 juveniles were sampled from each tank to observe their body growth and otolith development.

The body length (mm standard length) and weight (total g without internal organs) were measured, and the



condition factor was calculated using body weight/body length [3]. Both the right and left sagittal otoliths of sampled fish were measured for length along the longest dimension and mean values were calculated for the sample date; otoliths were then embedded in epoxy resin to be cut into transverse sections for observations of the appearance of translucent zones.

Results and discussion

The water temperature (Fig. 1) in the NT tank was 9.5° C on June 9 (when the tanks were first stocked), then it increased to 19.8° C on September 8, and decreased to 12.0° C by October 27 (the end of the experiment). The water temperature in the CT tank was 9.2° C on June 9, 12.1° C on July 21, and ranged between 10.1 and 13.3° C after July 21. The survival rates of fish in the NT and CT tanks at the end of the experiment were 0.11 and 0.10, respectively, but the difference was not significant.



Fig. 1. Water temperatures in the two experimental tanks: the solid line indicates the tank with natural temperature fluctuations (NT) and the broken line indicates the tank with controlled temperature (CT).

From August 25, the juveniles reared in the CT tank steadily attained a greater length and weight as compared with fish reared in the NT tank (Fig. 2A, B). In contrast, the condition factors of the fish reared in the different tanks (Fig. 2C) did not significantly differ. The water temperature in the NT and CT tanks differed by at most ~8°C, but the survival rate and condition factor of the fish reared according to the two water-temperature regimes did not significantly differ

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during the experiment. These results indicate that warmer temperatures may be a limiting factor for growth but not for health conditions in artificially reared walleye pollock fed to satiation.



Fig. 2. The growth of juvenile walleye pollock (mean \pm SD) at each sampling day; closed circles indicate fish from the NT tank and open circles indicate fish from the CT tank: (A) body length; (B) body weight; (C) condition factor; (D) sagittal otolith length.

The length of the sagittal otoliths increased with body growth (Fig. 2D). From August 25 onwards, the otoliths of juveniles in the CT tank were slightly longer than those of fish reared in the NT tank. As of July 21, a translucent zone in the otoliths was not observed in fish reared in either tank (Fig. 3A), but was observed by October 27 (the end of the experiment) only in otoliths of fish reared in the NT tank (Fig. 3B); likewise, the transverse sections did not reveal a translucent zone in the otoliths of fish sampled from either tank on July 21 (Fig. 4A). On September 15, a translucent zone was clearly evident at the lateral edge of the otoliths of some individuals sampled from the NT tank (Fig. 4B). By October 27 (the end of the experiment), an opaque band



Fig. 3. Pairs of sagittal otoliths of juvenile walleye pollock reared in either the NT tank (left column) or the CT tank (right column), and sampled on (A) July 21 or (B) October 27. Arrowheads indicate the translucent zone evident in NT fish only. Bar = 1 mm.



Fig. 4. Transverse sections of the sagittal otolith of juvenile walleye pollock reared in a tank with naturally fluctuating water temperatures (NT: left column) or controlled water temperature (CT: right column), and sampled on (A) July 21, (B) September 15, or (C) October 27. Arrowheads indicate translucent zones. Bar = 1 mm.

was again clearly visible outside the translucent zone in the otoliths of individuals sampled from the NT tank (Fig. 4C).

Observations of the otoliths revealed that a translucent zone appeared only in juveniles reared under naturally fluctuating water temperatures (NT tank). Timing of the formation of a translucent zone coincided with a period of warmer temperatures, whereas the opaque band appeared again when the water temperature decreased. These observations suggest that pseudo-bands may be formed under conditions of warmer water temperatures.

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