Age Validation of Pacific Cod (Gadus macrocephalus) Using Stable Oxygen Isotopes (δ¹⁸O)

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THEORY
1. The δ¹⁸O in otoliths is a function of temperature, an inverse relationship showing seasonal cycles may exist.
2. Microsampling of an otolith from pre first year to the outer edge, with multiple samples across any one year’s growth, can follow seasonal changes throughout the life of the fish.
3. HYPOTHESIS: The number of δ¹⁸O peaks should equal the estimated age (count of translucent zones), and can be used as an AGE VALIDATION. (Fig. 1)

METHODS
1. Pacific cod were collected in the eastern Bering Sea (Fig. 2). Specimens with estimated ages up to 5 years were randomly chosen.
2. Otoliths, aged 2 to 5 years old, were sampled using a micromilling system (Fig 3). Up to 42 sequential microsamples were milled from the center to the edge of each otolith (Fig 1a).
3. Each microsample was analyzed for δ¹⁸O by mass spectrometry, and results were plotted to demonstrate δ¹⁸O changes during the life of the fish.
4. To confirm the relationship between δ¹⁸O and temperature, otoliths aged 1 year old were microsampled and analyzed for δ¹⁸O on the outer edge (reflecting capture temperature).

RESULTS
1. For specimens aged 2 to 5 years old (n = 9), most had same number of δ¹⁸O peaks as estimated age (Fig. 4).
   - Most translucent zones interpreted as annual (and counted) were associated with δ¹⁸O peaks, indicating a correct age estimate.
   - Non-annual translucent zones (not counted) were not associated with δ¹⁸O peaks (specimen 52, 120, and 287), also indicating a correct age estimate.
   - Otolith interpretation and correlation to δ¹⁸O peaks is difficult in older specimens due to compressed growth and reduced sampling resolution in later years (specimen 287).
2. As expected, relationship between Pacific cod otolith aragonite (δ¹⁸O) and bottom temperature showed an inverse, statistically significant linear relationship (Fig. 5; r²=0.74, p<0.1)

CONCLUSIONS
1. The δ¹⁸O signatures can provide guidance on interpretation of visual translucent zone counts.
2. Therefore, the use of δ¹⁸O is a promising age validation method. Future work will use δ¹⁸O to estimate possible ageing bias for incorporation into assessment models.
3. Water temperature (°C) appears to be the primary factor controlling the variation in the δ¹⁸O signatures of Pacific cod otoliths.

Figure 1. Pacific cod otolith cross section with translucent growth zones presumed as annual in blue. Translucent zones not interpreted as annual (not counted) in red with a question mark.

Figure 2. Pacific cod capture locations in the eastern Bering Sea.

Figure 3. Micromilling system, with close up of milling bit.

Figure 4. Example of results to date, one specimen for each age 2 to 5 years. Otolith images are overlaid with δ¹⁸O (in ‰ VPDB) results.

Figure 5. In 1 year old specimens, δ¹⁸O (in ‰ VPDB) from otolith edges is compared to bottom temperatures (°C) at capture.

δ¹⁸O = -2.71 - 0.219°C
r² = 0.74