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# Isotopic signatures of otoliths and the stock structure of canary rockfish along the Washington and Oregon coast

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#### ABSTRACT

Canary rockfish are one of the commercially important rockfish species along the US Pacific coast. Yet little is known about their life history and stock structure. In this study 120 canary rockfish otoliths were collected from waters off the Washington and Oregon coast and subjected to stable O and C isotope ( $\delta^{18}$ O and  $\delta^{13}$ C) analyses. One powder sample was taken from the nucleus of each otolith, and the other from the 5th annual ring. Data from otolith nuclei can provide information on the natal sources and spawning stock separations, while data from age-1 to age-5 may indicate changes in fish habitat. Overall the  $\delta^{18}$ O values in otoliths of canary rockfish ranged from -0.2% to +1.7%, whereas  $\delta^{13}$ C values of the same samples ranged from -5.4% to -1.4%. The isotopic data and correlation of  $\delta^{18}$ O versus  $\delta^{13}$ C did not show clear separation between Washington and Oregon samples, similar to those for a previous study on yelloweye rockfish from the same region. These results suggest that canary rockfish may belong to a single spawning stock or population along the Washington and Oregon coast.

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## 1. Introduction

Canary rockfish (Sebastes pinniger) are distributed from the western Gulf of Alaska to northern Baja California, and are one of the commercially important rockfish species along the United States Pacific coast (Hart, 1973; Stewart, 2007). The fishery is currently managed as a single population (see-Pacific Fishery Management Council website, http://www.pcouncil.org), because little is known about the life history of canary rockfish and there are few published references regarding the likely stock structure of the species (e.g., Gomez-Uchida et al., 2003; Stewart, 2007). Adult canary rockfish are believed to spawn in winter and mostly dwell on or near the bottom of near shore high-relief rock formations, with peak abundance found at less than 100 m (Love et al., 2002). In commercial landings, canary rockfish are often associated with yelloweye rockfish (Sebastes ruberrimus). Both canary and yelloweye rockfish are considered to be long-lived, late to mature, and slow growing species, and their recruitments are highly variable and strongly influenced by environmental factors such as ocean climate (Wallace et al., 2006; Stewart, 2007). Given their affinity for hard bottom reliefs, home range and longevity (cf. Tolimieri et al., 2009), it is possible that canary rockfish may form local or discrete populations along the Washington and Oregon coast.

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Stable O and C isotope ratios ( $\delta^{18}$ O and  $\delta^{13}$ C) in otoliths have been proved useful in discriminating fish stock structure (e.g., Nelson et al., 1989; Stephenson et al., 2001) and a recent study indicated that many US Pacific coast groundfish can be separated into two or more spawning stocks or subpopulations (Gao et al., 2010). In particular, the correlation of  $\delta^{18}$ O versus  $\delta^{13}$ C values in otoliths can depict the integration of the surrounding environment (from  $\delta^{18}$ O) with food conditions (from  $\delta^{13}$ C) that an individual fish encountered. If little genetic differentiation exists or no genetic data are available between populations (Grant et al., 1987), the isotopic methods of otoliths might provide an alternative solution for stock identification. Therefore, it is of importance to examine available isotopic data to determine whether or not local aggregations of canary rockfish can be identified as subpopulations or as separate stock units.

The objectives of this manuscript are twofold. First, 120 canary rockfish otoliths were collected from waters off the Washington and Oregon coast, and the nucleus of each otolith and the 5th annual ring analyzed for C and O stable isotope composition. Data from otolith nuclei can provide information on the natal sources and spawning stock differences, while isotopic data from age-1 to age-5 may indicate changes in fish diet and habitat. Second, the annuli of five randomly-selected otolith samples for both canary and yelloweye rockfish were analyzed to examine the early life history and behavior for the adult stage. Because the two rockfish species are commonly associated with each other and the tagging experiments have very low recovery rates, these natural isotopic

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signatures of otoliths and the comparison with yelloweye rockfish could provide valuable information on canary rockfish.

## 2. Methods

Sagittal otoliths of 120 canary rockfish were collected from Cape Blanco of Oregon State to Cape Flattery of Washington State in 2005, by Oregon Department of Fish and Wildlife (ODFW) and Makah Fisheries Management (MFM), respectively (Fig. 1). The Oregon otolith samples were aged by the ODFW ageing group, using a break-and-burn method (Beamish and Chilton, 1982). The Washington otolith samples were divided into two parts: the right piece of an otolith pair was used in this study, whereas the left piece was stored by Washington Department of Fish and Wildlife for records. In addition, isotopic data from a previous study on yelloweye rockfish, in which 200 otolith samples of yelloweye rockfish were collected from the similar coast-wide areas as canary

rockfish (cf. Fig. 1) and processed with the same methodology (Gao et al., 2010), were used for comparison purposes.

Otolith samples were first cleaned in an ultrasonic water bath for about 15 min, rinsed with ethanol, and air dried at room temperature. The methods for resin embedding and sectioning have been reported elsewhere (Gao and Beamish, 1999). Each otolith section was polished using an ECOMET 3 variable speed grinder-polisher with 320–1200 grit self-adhesive paper. At the final stage, an AP-Cloth was applied for surface polishing with 0.3  $\mu m$  Alumina powders.

Two aragonite powder samples were collected from each otolith section: one sample was taken from the nucleus of canary rockfish otoliths, and the other taken from the 5th annual rings. Five randomly-selected otolith sections were also sampled with annuli from age-1 to age-12 for both canary and yelloweye rockfish. Microsampling was conducted by using the Dremel method described by Gao (1999). At least 50 µg of aragonite powder

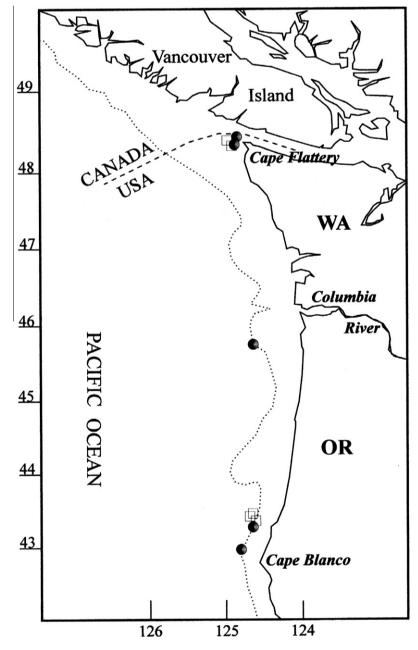


Fig. 1. Location map showing the sampling sites in this study. The solid circles were major otolith collections for canary rockfish in 2005; whereas the open squares were major yelloweye rockfish sampling from a previous otolith study (Gao et al., 2010).

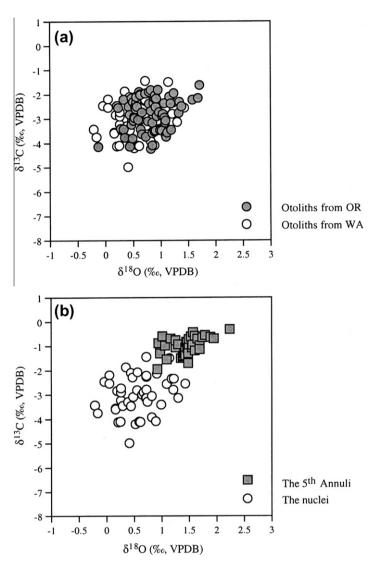


Fig. 2. The correlation of  $\delta^{18}$ O versus  $\delta^{13}$ C in canary rockfish otoliths showed: (a) no separations in otolith nuclei between Oregon (solid circle) and Washington (open circle) samples; and (b) isotopic shifts from the age-1 (open circles) to age-5 (solid squares) otoliths in Washington samples only.

material were extracted from the otolith surface for stable isotope analysis. Once a sample was taken, the powder was carefully tapped onto aluminum foil and placed into a metal cup. The otolith section and the sampling bit were subsequently cleaned using an Aero-Duster gas.

Analysis of powder samples of canary rockfish otoliths was performed in the Stable Isotope Laboratory at the University of Michigan Ann Arbor, using a Finnigan MAT Kiel carbonate preparation device directly coupled to the inlet of a Finnigan MAT 251 triple-collector gas-ratio mass spectrometer. All the measurements were reported in the standard  $\delta$  notation (%):  $\delta^{18}O = \{[(^{18}O/^{16}O)_A/(^{18}O/^{16}O)_S] - 1\} \times 1000$ , for instance, where A is aragonite sample and S is an international standard (VPDB, Vienna Peedee belemnite). Calibration of isotopic enrichments to VPDB standard is based on daily analysis of NBS-19 (National Bureau of Standards) powdered carbonate and the analytical precision is better than 0.08% for both  $\delta^{18}O$  and  $\delta^{13}C$  values.

# 3. Results and discussion

The  $\delta^{18}O$  values from the nucleus of canary rockfish otoliths ranged from -0.2% to +1.7%, while  $\delta^{13}C$  values of the same otoliths ranged from -5.4% to -1.4%. There were no isotopic separa-

tions between Oregon and Washington samples (Fig. 2a). From age-1 to age-5 otolith rings, the isotopic composition of Washington samples showed a clear shift in  $\delta^{13}$ C (Fig. 2b), indicating a significant dietary change (DeNiro and Epstein, 1978) over the period of juvenile growth. Compared with a previous otolith study on yelloweve rockfish from the same region (Gao et al., 2010), the isotopic data of canary rockfish showed a similar  $\delta^{18}$ O range, but distinctly higher  $\delta^{13}$ C values than those of yelloweve rockfish (Fig. 3). The similarity in  $\delta^{18}O$  and difference in  $\delta^{13}C$  suggest that the two rockfish species may be associated with similar bottom reliefs; but canary rockfish have different diet sources or consume much higher trophic-level food. In commercial catches canary rockfish are often found to be associated with yelloweye rockfish and the recent stock assessments in the Puget Sound of Washington State list both as "distinct population segments" (Dr. Theresa Tsou, Washington Department of Fish and Wildlife, pers. comm.). Therefore, if previous results on stock structure of yelloweye rockfish were applicable to canary rockfish (cf. Gao et al., 2010), the inseparable isotopic correlation in  $\delta^{18}O$  versus  $\delta^{13}C$  from the present study indicated that canary rockfish may belong to a single spawning stock or population coast-wide. Otoliths from the California coast were not included, obviously, because canary rockfish are less prevalent in sampling from the commercial fisheries in California State.

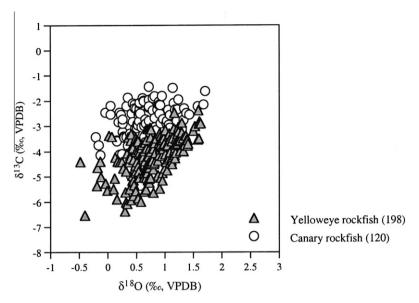


Fig. 3. The comparison between canary and yelloweye rockfish otoliths showed a similar  $\delta^{18}$ O range, but the  $\delta^{13}$ C values of canary rockfish were distinctly higher than those of yelloweye rockfish.

Table 1
Summary of stable isotopic data of otoliths sampled from nuclei (age of 0–3 month), the 5th annual rings (age-5), and the adult stage (>age-7) between canary and yelloweye rockfish.

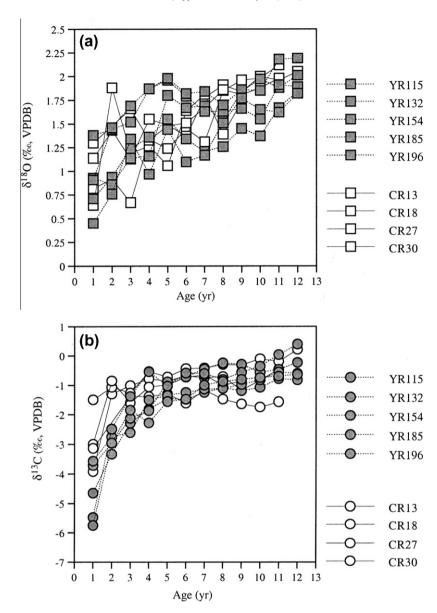
	N	δ <sup>13</sup> C (‰)	Mean	SD	$\delta^{18}$ O (‰)	Mean	SD
(Nuclei):							
Canary	120	−5.36 to −1.44	-2.90	0.75	-0.21 to 1.71	0.71	0.38
Yelloweye	198	-6.54 to $-2.38$	-4.21	0.78	-0.48 to 1.63	0.79	0.40
(The 5th annuli):							
Canary	83	-2.65 to 0.22	-1.09	0.57	0.67 to 2.24	1.51	0.34
Yelloweye	199	-3.15 to $-0.15$	-1.46	0.51	0.45 to 2.20	1.55	0.32
(The adult stage):							
Canary	23	-2.19 to 0.22	-1.04	0.70	1.12 to 2.12	1.80	0.28
Yelloweye	25	-1.19 to 0.40	-0.63	0.37	1.26 to 2.19	1.74	0.23

Statistical tests (i.e. Kolmogorov-Smirnov and Shapiro-Wilk tests) on otolith data of canary rockfish indicated that both  $\delta^{13}$ C and  $\delta^{18}$ O values were normally distributed. The isotopic data from otolith nuclei of canary rockfish showed significant differences between Washington and Oregon samples in  $\delta^{18}$ O (p = 0.002) but not in  $\delta^{13}C$  (p = 0.40). Based on microsampling, data from otolith nuclei represent a growth period of the initial 0-3 month after canary rockfish spawning. Thus the significant differences in  $\delta^{18}$ O may suggest a bottom habitat change or a surface water difference (Gao and Beamish, 1999). Direct observations about the early life history of canary rockfish are not available; however, juvenile canary rockfish have been reported to disperse widely and pelagic larvae are found in the upper water column for 3-4 months after spawning (Love et al., 2002). Due to isotopic fractionation by evaporation of water from oceans, the <sup>18</sup>O/<sup>16</sup>O ratios of seawater may show a latitude effect which reflects the temperatures of condensation (Craig and Gordon, 1965; Gat, 1981) and thus affect the  $\delta^{18}$ O values of canary rockfish otoliths. Therefore, the significant differences in  $\delta^{18}O$  values of otolith nuclei may help to interpret the latitude effect for <sup>18</sup>O/<sup>16</sup>O distributions in waters off the Washington and Oregon coast, while the correlated  $\delta^{13}C$  values showed no diet shifts for the canary rockfish larvae.

Nevertheless, the  $\delta^{13}C$  data of otoliths were especially informative between canary and yelloweye rockfish (Table 1). For comparison purposes, the annuli of five randomly-selected otoliths for the two rockfish species were analyzed. It appeared that the sampled

canary rockfish had more variations especially in the first one or two years, with the stable values after age-7 or 8 (Fig. 4). Overall both canary and yelloweye rockfish showed a consistent increase in  $\delta^{18}$ O and  $\delta^{13}$ C values and a two-stage life history: the initial low isotope values corresponding to the juvenile life while the stable higher isotope values corresponding to the adult stage. The transition of  $\delta^{13}$ C at age-7 or 8 (Fig. 4b) may be the period of sexual maturity (Schwarcz et al., 1998) for canary rockfish, which agrees well with the report from biological observations and gonad examinations (Yamanaka and Kronlund, 1997). A major question may be the age differences between canary and yelloweye rockfish otoliths. However, from this study it was known that more than 80% of Oregon otoliths within the range of 10–15 years (Fig. 5) such that it was possible to sample the main life stage of canary rockfish as was done for yelloweye rockfish.

It is important to note that the isotopic data and interpretation of a single population are supported by commercial trawl landings (Stewart, 2007) and agree with the current Pacific Fishery Management Council management plan. There are few biogeographic boundaries applicable to canary rockfish, so the current stock assessment that used the US and Canada border as the northern boundary for the species is based primarily on consistency with the management needs (Stewart, 2007). Limited tagging experiments found that 40% of the canary rockfish recovered moved more than 100 km, and that the fastest swimmers also moved to much greater depths than the shallow reefs at which they had been



**Fig. 4.** The annual isotopic variations of five randomly-selected otoliths between canary and yelloweye rockfish in  $\delta^{18}O(a)$  and  $\delta^{13}C(b)$ . Because of the laboratory failure, only four canary rockfish were presented here.

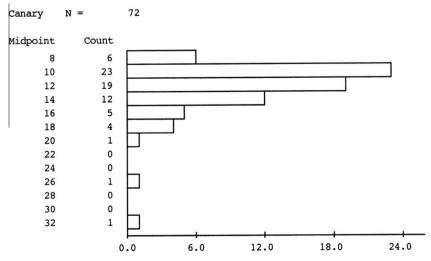


Fig. 5. The age component of canary rockfish in this study, showing about 80% of Oregon otolith samples within the age range of 10–15 years.

tagged (Stanley et al., 2005). The isotopic data in this study appear consistent with these biological observations: canary rockfish and yelloweye rockfish may locate in similar water depth between near-shore and offshore, but adult canary rockfish (10–15 years old) are generally more mobile than yelloweye rockfish.

#### 4. Conclusions

Based on isotopic data of otoliths and the correlation of  $\delta^{18}$ O versus  $\delta^{13}$ C, it is concluded that canary rockfish may belong to a single spawning stock or population along the Washington and Oregon coast, which is consistent with the stock structure of yelloweye rockfish as previously reported. Canary rockfish appear to be more mobile than yelloweye rockfish, either in the juvenile stage for higher trophic-level food or as adults occurring offshore. Because of limited genetic information available on canary rockfish, isotopic signatures of otoliths may provide supplemental data on stock structure for this data-poor species.

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