

**Otolith chemistry, stomach contents and stable isotope analysis
of a snapper (*Pagrus auratus*) caught in the Waikato River
at Ngaruawahia**

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by

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Summary

Analyses of stomach contents, stable isotopes and otolith microchemistry were carried out in order to ascertain the length of freshwater residence of a snapper (*Pagrus auratus*) caught in the Waikato River at Ngaruawahia in a net set for grey mullet (*Mugil cephalus*). Results of all analyses suggest that the snapper had spent all of its life in a marine environment with no evidence of freshwater residence in the otolith. Stable isotope analyses ($\delta^{15}\text{N} = 17.0\text{‰}$, $\delta^{13}\text{C} = -17.1\text{‰}$) indicated an entirely marine diet, and the stomach contents (two New Zealand screwshells, *Maoricolpus roseus*, and a hermit crab, *Pagurus novizelandiae*), suggested that the fish had not fed while in freshwater. However, this does not preclude the possibility that the snapper quickly travelled up the river, without eating, and was caught very soon after.

The finding of a small shark, probably a school shark, (*Galeorhinus galeus*), on 19 February 2005 on a beach of the Waikato River in Hamilton city provides other residents that marine fish to periodically travel up the Waikato River.

Introduction

On the 26 June 2009, Rodney Pointer caught a 1.039-kg, 365-mm fork length snapper (Sparidae: *Pagrus auratus*; Figure 1) 3 km downstream of Ngaruawahia Point (37.63784°S, 175.15316°E, NZ map grid 6374933 S, 2714101 E; Figure 2) while fishing for grey mullet (*Mugil cephalus*) with a mullet net. A number of mullet were captured in the net with the snapper, together with a substantial amount of oxygen weed (*Lagarosiphon major*) a common aquatic plant in the Waikato River (Figure 3). To ascertain the length of freshwater residence, we analysed stomach contents, stable isotopes and otolith microchemistry.

The snapper is found in marine and estuarine environments. Estuarine snapper occupy small home ranges within soft-sediment environments, and make small-scale movements over hundreds of metres (Hartill et al. 2003). Therefore, a migration to Ngaruawahia, approximately 94 km upstream from the sea, is unusual.



Figure 1. Snapper caught in the Waikato River, 3 km downstream of Ngaruawahia Point (37.63784N, 175.15316E). Photo: J. Blair.

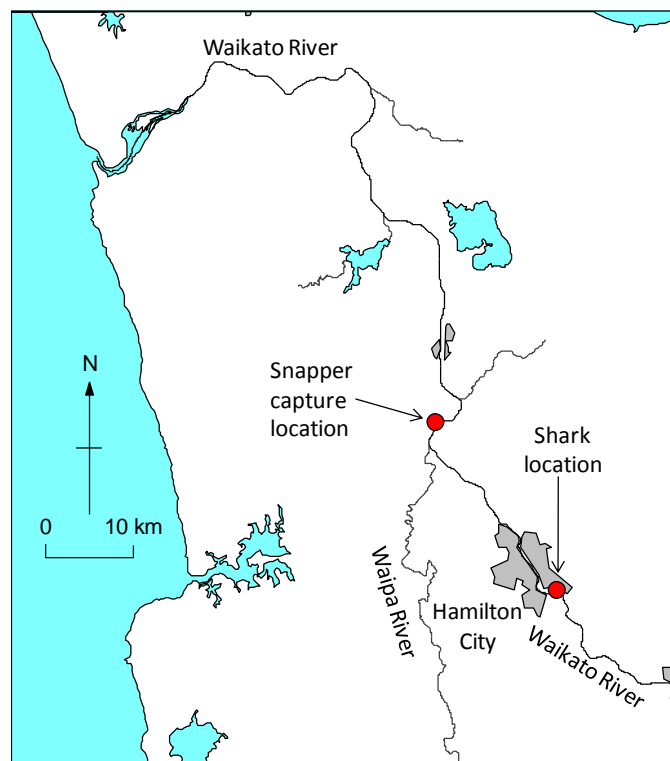


Figure 2. Location of the sites of capture of the snapper in the Waikato River, and the shark discovery on a Hamilton City beach.



Figure 3. Net with grey mullet and the snapper shortly after hauling near Ngaruawahia. Note the large amounts of the freshwater oxygen weed *Lagarosiphon major* that was also trapped in the net. Photo: Rodney Pointer.

Methods

Stomach contents

The snapper was thawed in a bath of warm water, weighed and measured. The gut was dissected from the fish and the contents of the entire digestive tract examined. Individual organisms were weighed and identified.

Stable isotope analysis

A small piece of dorsal white muscle (1 cm³) was removed from the snapper and dried on a piece of aluminium foil at 50° C for 24 h. The sample was then ground to a powder using a coffee grinder and sieved to remove any large pieces. Ratios of ¹³C/¹²C and ¹⁵N/¹⁴N in the sample were analysed at the Waikato Stable Isotope Unit using a Europa Scientific continuous flow 20/20 mass spectrometer with a triple ion-collector and ANCA SL inlet system. Results were calibrated using Australia National University (ANU) cane sucrose for ¹³C/¹²C, and N₂ in air for ¹⁵N/¹⁴N.

The ratios of ¹³C/¹²C and ¹⁵N/¹⁴N are reported as the relative difference per mil (‰) and are calculated using Equation 1, where X=¹³C or ¹⁵N, and R = ¹³C/¹²C or ¹⁵N/¹⁴N.

$$\delta X = 1000 \left[\frac{R_{\text{sample}}}{R_{\text{standard}}} - 1 \right] \quad (1).$$

Otolith chemistry

The saggital otoliths were removed and rinsed in household bleach and Milli-Q water. Otoliths were then mounted in a transparent epoxy resin. One of the mounted otoliths was cut into 0.6 mm sections using a Buehler IsoMet ® low-speed saw (Lake Bluff, Illinois) and polished using 400-2000 grit waterproof silicon carbide paper until the nucleus was clearly visible. The section was mounted on a microscope slide and stored in a plastic bag until ablation.

Otolith trace elements were analysed at the University of Waikato using laser ablation inductively coupled plasma mass spectrometry (LA-ICP-MS) using methods described in Blair (2009). Otoliths were ablated in a sealed chamber using a New Wave Research UP-213 Laser Ablation System (Fremont, CA) with a 213 nm neodymium yttrium aluminium garnet (Nd-YAG) laser. Ablated material was carried using a mixture of helium and argon gas to a Perkin Elmer DRCII ELAN 6000 inductively coupled mass spectrometer (Waltham, MA). Isotopes analysed were magnesium (^{25}Mg), aluminium (^{27}Al), calcium (^{42}Ca and ^{43}Ca), manganese (^{55}Mn), copper (^{65}Cu), zinc (^{66}Zn), nickel (^{62}Ni), rubidium (^{85}Rb), strontium (^{88}Sr) and barium (^{137}Ba). NIST SRM (National Institute of Standards and Technology Standard Reference Material) 612 was used as a calibration standard for all analyses. Background element concentrations were measured for 60 s prior to each ablation by analysing a gas blank (firing the laser with the shutter closed).

Element concentrations across the snapper otolith were measured using a line scan. Settings used were $10\ \mu\text{m s}^{-1}$ scanning speed, 5 Hz repetition rate, 60% output and $60\ \mu\text{m}$ spot size. The line was pre-ablated at $20\ \mu\text{m s}^{-1}$ in order to remove any possible surface contamination. To process the line scan, the mean isotopic counts from the first 60 seconds of analysis (taken as a background reading without firing the laser) were subtracted from isotopic counts taken during the line scan. Results are presented as counts of the isotope (^{88}Sr or ^{137}Ba) to ^{43}Ca .

Results

Snapper

The stomach contained two New Zealand screwshells (*Maoricolpus roseus*), weighing a total of 0.7 g, and hermit crab (*Pagurus novizelandiae*), weighing 0.54 g. The crab was in a more digested state, and was out of its shell.

Stable isotope values of the white muscle reflected marine residence. The percentage of N in the dried stable isotope sample was 13.5%, and the $\delta^{15}\text{N}$ value was 17.0‰. The percentage of C was 44.3%, and the $\delta^{13}\text{C}$ value was -17.1 ‰.

Otolith microchemistry also reflected marine or estuarine residence. Ratios of $^{88}\text{Sr}:^{43}\text{Ca}$ and $^{137}\text{Ba}:^{43}\text{Ca}$ in the snapper's sagittal otolith increased from the nucleus to the edge (Figure 4). The ratio of $^{88}\text{Sr}:^{43}\text{Ca}$ was higher than the ratio of $^{137}\text{Ba}:^{43}\text{Ca}$ over most of the scanned otolith surface (Figure 2). Higher barium counts at 190-250, 450-550, and 50-950 μm distance from the nucleus suggest short periods of possible estuarine residence.

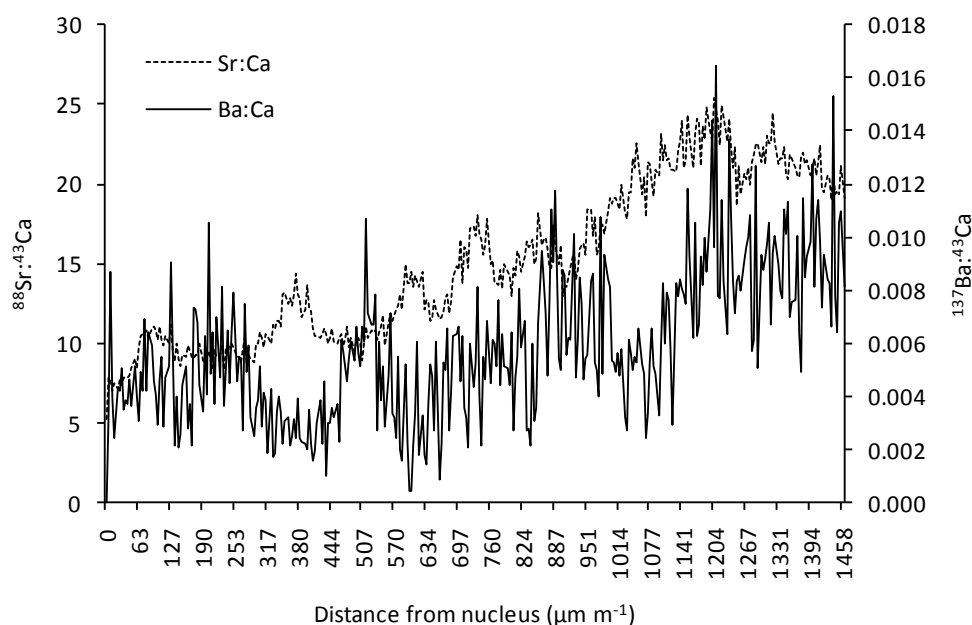


Figure 4. Ratios of $^{88}\text{Sr}:^{43}\text{Ca}$ and $^{137}\text{Ba}:^{43}\text{Ca}$ measured using a line scan from the nucleus to the edge of the snapper otolith.

Shark found in the Waikato River

On 19 February 2005, a shark about 60 cm long (Fig. 5), most likely a school shark (*Galeorhinus galeus*), was found on a beach of the Waikato River at Malcolm Street, Hamilton

City, in the suburb of Hillcrest (37.806656°S, 175.315469°E, NZ map grid 6394026 S, 2700274.39 E) by Brennan Mahoney, University of Waikato student. This site is 122 km upstream from the sea (Fig. 2). Because the shark was not formally identified, we cannot be certain of its species, but the tail conformation and widespread occurrence of school sharks in harbours and estuaries strongly suggests this identity.



Figure 5. Probable school shark, *Galeorhinus galeus*, about 60 cm long, found on a beach of the Waikato River on 19 February 2005. Photo: Brennan Mahoney.

Discussion

The organisms found in the snapper's stomach are marine or estuarine species (e.g., Powell 1979), suggesting that the snapper did not feed while in fresh water. Stable isotopes of C and N can be used to trace migrations of animals moving between areas with distinct food webs (Hobson 1999). The high $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ of the snapper tissue suggest a marine origin (Hicks et al. 2005).

Otolith microchemistry supports the marine residence of this snapper. The line scan across the snapper otolith showed that the ratio of $^{88}\text{Sr}:^{43}\text{Ca}$ was higher than the ratio of $^{137}\text{Ba}:^{43}\text{Ca}$ across the otolith and at the otolith edge. This is indicative of marine or estuarine residence, as Sr:Ca ratios tend to be lower in fish living in fresh water and Ba:Ca ratios higher. If the snapper had been residing in fresh water for a significant amount of time, a drop in the Sr:Ca ratio and a rise in the Ba:Ca ratio could be expected. A typical otolith a fish that has moved between the sea and freshwater is shown in Fig. 6. This torrentfish otolith demonstrates the characteristic change of Sr and Ba concentrations between the periods of marine and freshwater residence.

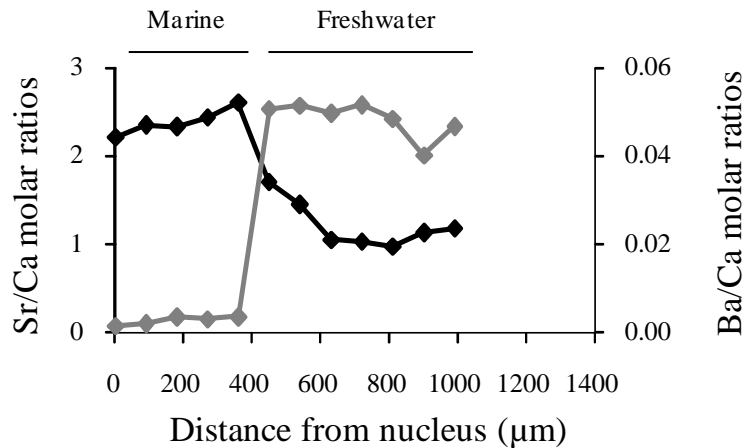


Figure 6. Sr and Ba concentrations in an otolith from a torrentfish from the Mangauika Stream. Marine residence from 0 to about 375 µm from the nucleus as shown by high Sr concentrations (black line) and low Ba concentrations (grey line). Freshwater residence from 400-1000 µm. Source: Tana (2009).

Conclusion

Stomach contents, stable isotopes and otolith microchemistry results all suggest that the snapper caught at Ngaruawahia spent its life in a marine or estuarine environment and had not been residing in freshwater for a significant length of time when it was captured. However, this does not preclude the possibility that the snapper quickly travelled up the river, without eating, and was caught immediately. In this case, we would not expect to see any evidence of freshwater residence in the otoliths or flesh.

Acknowledgements

We thank Rodney Pointon, Ngaruawahia, and Brennan Mahoney, Hamilton, for specimens and photographs of the fish referred to this report.

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