

# Stable Isotopes of Estuarine Fish: Experimental Validations and Ecological Investigations

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Cover image: Chapman River, Kangaroo Island, November 2010.

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Ecological Investigations**

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## **Abstract**

Stable isotopes of carbon and nitrogen are commonly used in ecological research to determine food webs and trace anthropogenic inputs. These applications rely on understanding isotope signature differences between an animal and its food. When an animal consumes a food item, or changes diet, it does not instantaneously reflect the isotope ratios of that food item. The isotopic signature of animal tissue gradually approaches equilibrium with the isotopic signature of its food, as molecules are turned over and new food items are assimilated into tissues. Stable isotope ratios also change between food consumed and animal tissues that are commonly sampled. The difference in stable isotope ratios between an animal's tissue and the food it consumes is called discrimination. The rate of change, or tissue turnover, and discrimination of stable isotopes varies among and within animals, and with environmental factors. I investigated the effects of temperature and diet on these isotope parameters for two fish species and applied results to improve determination of autotrophic sources within estuaries.

I studied two common, omnivorous, estuarine fishes found in South Australia: black bream (*Acanthopagrus butcheri*) and yellow-eye mullet (*Aldrichetta forsteri*). Temperature and diet affected both tissue turnover rates and discrimination of carbon ( $\delta^{13}\text{C}$ ) and nitrogen ( $\delta^{15}\text{N}$ ) isotope ratios in fish muscle. Fish reared at warmer temperatures generally had faster tissue turnover rates and smaller discrimination factors than fish reared at cooler temperatures. However, temperature interacted with diet quality to affect  $\delta^{13}\text{C}$  discrimination. Fish fed diets with low C:N ratios had larger  $\delta^{13}\text{C}$  discrimination at warmer temperatures than at cooler temperatures. This may be caused by fish catabolising more protein for energy and therefore being able to store more lipids at cooler temperatures

than warmer temperatures. Fish fed diets with high C:N ratios were the opposite, with larger  $\delta^{13}\text{C}$  discrimination at cooler temperatures than at warmer temperatures.

Compound-specific  $\delta^{15}\text{N}$  analyses were performed on amino acids from experimental black bream muscle tissues to see if the change in  $\delta^{15}\text{N}$  of amino acids could explain the bulk change in  $\delta^{15}\text{N}$  of whole muscle tissue. Some amino acid  $\delta^{15}\text{N}$  results mirrored those of bulk  $\delta^{15}\text{N}$  analyses suggesting that they may be non-essential amino acids, although there was large variation among individual fish.

Wild fish commonly consume more than one dietary item, necessitating the use of mixing models to determine source contributions to diets. Omnivores consume animal and plant matter that can differ greatly in their elemental composition and this can affect the uptake of isotopic signatures from different food sources. I tested the importance of using elemental concentration in mixing models by combining two diets with different carbon and nitrogen concentrations and feeding them to yellow-eye mullet. I compared measured  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  of fish muscle with predicted values calculated with and without using elemental concentration. Using elemental concentration in mixing models improved estimates of predicted isotopic signatures.

The experimentally derived discrimination factors for black bream and yellow-eye mullet were used to investigate the relative importance of autotrophic sources to their diets in four estuaries in South Australia. Isotope signatures of carbon and nitrogen can also be used to investigate ecological niches of animals, as isotope signatures reflect what an animal has eaten from different habitats and environments. I expected the isotopic niches of black bream and yellow-eye

mullet to overlap, due to their shared environmental tolerances and feeding habits, as they are commonly found in the same estuaries. However, I found no overlap in isotopic niches between black bream and yellow-eye mullet. In some estuaries the autotrophic sources that black bream and yellow-eye mullet relied on were similar, however, in these estuaries fish appeared to be either feeding at different trophic levels or were likely not in competition with one another as they were caught in different areas within estuaries. The separate isotopic niches of black bream and yellow-eye mullet may be caused by habitat partitioning or interspecific competition within the estuaries studied.

I used  $\delta^{15}\text{N}$  of black bream muscle to trace anthropogenic inputs of nutrients across a range of estuaries and related nutrient concentrations of estuarine waters to black bream abundance and recruitment. Black bream abundance and recruitment showed subsidy-stress responses to nutrient concentrations of ammonia, oxidised nitrogen and orthophosphorus, with peaks in abundance and recruitment occurring at low concentrations. A positive linear relationship was found between ammonia concentration of estuarine waters and  $\delta^{15}\text{N}$  of black bream. This suggests that anthropogenic ammonia was being taken up into the food web, or directly by black bream, and affecting black bream abundance and recruitment.

In summary, I found environmental factors affected stable isotope signatures of fish muscle tissue. These results further show how important it is to quantify isotope parameters for individual species. Future research should focus on how to quantify influences on isotope signatures that cannot be determined in the field, such as ration intake, and how to account for these factors in field studies.

## **Declaration**

I, Alexandra Louise Bloomfield certify that this work contains no material which has been accepted for the award of any other degree or diploma in any university or other tertiary institution and, to the best of my knowledge and belief, contains no material previously published or written by another person, except where due reference has been made in the text.

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Signed:

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Archie „helping“ me study.