VARIABILITY OF $^{13}C/^{12}C$ AND $^{15}N/^{14}N$ IN DIFFERENT MUSSEL TISSUES (*MYTILUS GALLOPROVINCIALIS*) : IMPLICATIONS FOR FOOD WEB STUDIES

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Abstract

Measurements of the stable isotope ratios of carbon and nitrogen were obtained for different mussel (*Mytilus galloprovincialis*) tissues from two locations in the Gulf of Trieste (Adriatic Sea). We found no significant differences in $\delta^{13}C$ and $\delta^{15}N$ between the two sites ($p > 0.5$) nor between two size classes ($p > 0.9$) of mussels. Differences were significant ($p < 0.05$) among different mussel tissues indicating that within the organism variability may be large enough to affect the conclusions drawn about trophic position of animals when based only on analysis of one particular tissue.

Key-words: bivalves, food webs, Adriatic Sea

Introduction

The use of stable isotope ratios as indicators of animal trophic position and food web relationships has become increasingly widespread, especially using combined measurements of $\delta^{13}C$ and $\delta^{15}N$ (1). The characteristic isotopic composition of each biogenic material depends on the isotopic ratios of diet (substrate), metabolic pathways and fractionating processes (2); consequently, stable isotope signatures of organisms may provide information of the trophic linkages within food webs (3). The stable isotopic ratios of C and N in consumers have been shown to reflect those assimilated with about 1-2 enrichment of ^{13}C and 3-4 enrichment of ^{15}N (4, 5). Generally, implicated organic matter pathways are based on bulk samples and/or the isotopic composition of whole animals and plants, despite variations within individuals and populations (6), and variability among different tissues (7). Moreover, significant spatial differences in isotopic compositions have been found within a few kilometres (8, 9).

In our previous studies we used C isotopic ratios to assess the food sources of the planktivore jellyfish *Pelagia noctiluca* in the northern Adriatic (10) and to elucidate the sources of sedimentary organic matter in the nearshore marine environment (11). The aim of the present study was: (1) to determine the C and N isotopic composition of the benthic filter feeding, mussel *Mytilus galloprovincialis*, (2) to determine mussel $\delta^{13}C$ and $\delta^{15}N$ at two different locations in order to assess spatial variations, and (3) to determine $\delta^{13}C$ and $\delta^{15}N$ of different mussel tissues in order to estimate how the selection of a particular tissue for isotopic analysis may affect the trophic positioning of animals.

Material and methods

Mussels (*Mytilus galloprovincialis*) were collected at two sites on the south-eastern coast of the Gulf of Trieste (Adriatic Sea), one in the bay of Piran (SEC) close to the Dragonja river outflow, and the second in the bay of Strunjan (STR) which is without significant freshwater input (Fig. 1). Sampling was carried out in June 1996 after the spring spawning period of mussels (12).

Individual mussels were pooled according to shell length in the two size classes: 40-60 mm and 60-90 mm, the former less than two years and the latter more than two years old (12). Different tissues (foot, gills, byssus, stomach) were carefully dissected for isotopic analysis. Stomachs were analysed together with their food content.

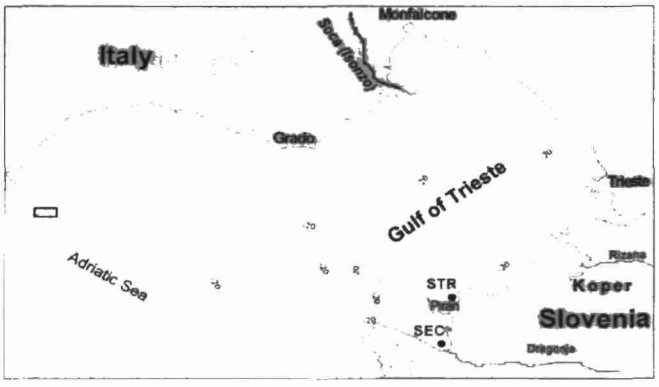

Fig. 1. Study area and sampling locations, sec and str.

To determine the stable isotope composition of nitrogen and carbon, freeze-dried samples of different tissues were ground and analysed using an Europa 20-20 Stable Isotope Analyser (Europa Scientific Ltd., UK) with an ANCA-NT preparation module for automatic combustion and separation of produced gases (13). Samples were treated with 1N HCl and washed with distilled water several times prior to measurement to eliminate residues of carbonates, which might be present in tissues in contact with the sediment. Stable isotope compositions of C and N were expressed as values in per mill deviation from a standard as follows:

$$\delta^{13}C \text{ or } \delta^{15}N = \delta^{13}C \text{ or } \delta^{15}N[‰] = \left(\frac{R_{sample}}{R_{standard}} - 1\right) x1000$$

where $R=^{13}C/^{12}C$ or $^{15}N/^{14}N$, respectively. PDB and atmospheric nitrogen were used as standards. Analytical error was 0.2 for both $\delta^{13}C$ and $\delta^{15}N$.

Results and Discussion

The $\delta^{13}C$ and $\delta^{15}N$ data obtained for mussels (*Mytilus galloprovincialis*) from both locations varied between - 22.63 and - 18.58‰, and between 4.18 and 5.69‰, respectively, with an overall mean for $\delta^{13}C$ of - 20.79 ± 1.21‰ (n = 46) and mean for $\delta^{15}N$ of 4.84 ± 0.44‰ (n = 46). These results are similar to or slightly lower than those reported for different benthic filter feeders in the literature (14, 4). The mean $\delta^{13}C$ of mussels (results from all analyses pooled) was around 2.5‰ higher than that of their presumed food - phytoplankton dominated suspended organic matter (- 23.3 ± 1.3‰; 11), consistent with the widespread recognition of C isotopic fractionation in consumers. Unfortunately, we have no data for $\delta^{15}N$ in suspended organic matter as source material.

When data for all tissue samples were combined, we did not find significant differences between the two sites ($p > 0.5$) in either $\delta^{13}C$ or $\delta^{15}N$, nor between the two size classes of mussels ($p > 0.9$). However, differences were significant ($p < 0.05$) among different mussel tissues (Tables 1, 2). The highest $\delta^{13}C$ or $\delta^{15}N$ was found for mussel byssus followed by gills, while other tissues contained less heavy isotopes of C and N.

Table 1. Mean $\delta^{13}C$ and $\delta^{15}N$ of different tissues at two sites (mean ± SD,‰, n=6)

	STR		SEC	
tissue	**$\delta^{13}C$**	**$\delta^{15}N$**	**$\delta^{13}C$**	**$\delta^{15}N$**
foot	- 22.0 ± 0.18	4.84 ± 0.23	-21.36 ± 0.86	4.75 ± 0.28
byssus	- 19.31 ± 0.88	5.17 ± 0.23	- 19.15 ± 0.74	5.16 ± 0.35
gills	- 20.49 ± 0.29	5.19 ± 0.31	- 20.33 ± 0.28	5.0 ± 0.42
stomach	- 22.0 ± 0.39	4.26 ± 0.07	- 21.4 ± 0.27	4.31 ± 0.16

Table 2. Results from multiple comparison test (Scheffe test) to detect differences between the $\delta^{13}C$ and $\delta^{15}N$ of different tissues (data for mussels from both sites pooled together, positioning of * denotes similarity and/or dissimilarity $\delta^{13}C$ and $\delta^{15}N$ of analyzed tissues)

	Homogeneous groups (95 % Confidence intervals)	
tissue	**$\delta^{13}C$**	**$\delta^{15}N$**
foot	*	*
stomach	*	*
gills	*	* *
byssus	*	*