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^{15}N ENRICHMENT IN FOULING COMMUNITIES INFLUENCED BY ORGANIC WASTE DERIVING FROM FISH FARMS

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ABSTRACT

The nitrogen stable isotopic composition of fouling communities colonizing hard substrates at the fish farms and at unpolluted reference locations in N Adriatic (Piran, Slovenia), E Mediterranean (Crete, Greece) and in the Red Sea (Eilat, Israel) was studied. An overall enrichment of the fouling community in 15-N at the fish farms has been detected at two of the three locations, indicating the incorporation of farm-derived particulate nitrogen into the organisms tissues.

Key words: fish farm, stable isotope, nitrogen, biofilters, fouling organisms

ARRICCHIMENTO IN ^{15}N IN COMUNITÀ DI FOULING INFLUENZATE DA RIFIUTI ORGANICI PROVENIENTI DA ALLEVAMENTI DI PESCI

SINTESI

L'articolo riporta lo studio della composizione isotopica stabile di azoto in comunità di fouling colonizzanti substrati solidi di allevamenti di pesci e in siti di controllo non inquinati, in Adriatico settentrionale (Pirano, Slovenia), Mediterraneo orientale (Creta, Grecia) e Mar Rosso (Eilat, Israele). Un arricchimento complessivo in 15-N delle comunità di fouling negli allevamenti di pesci è stato riscontrato in due delle tre postazioni, indicando l'incorporazione del particellato di azoto derivante dagli allevamenti nei tessuti degli organismi.

Parole chiave: allevamenti di pesci, isotopo stabile, azoto, biofiltri, organismi di fouling

INTRODUCTION

Fish farms release large amounts of soluble inorganic and particulate organic matter (POM) into the marine environment (McGhie *et al.*, 2000). There is, therefore, a large interest in reducing the effluent load to the surrounding waters in order to ease the environmental impact of mariculture. In order to capture and remove effluents released from fish farms, removable hard substrates were deployed near a fish farm, in order to be colonised by filtering organisms.

Stable isotopes of nitrogen were successfully used to trace the dispersion of organic debris and the sources of sedimentary organic matter in marine sediments and in aquaculture systems (Ye *et al.*, 1991; McGhie *et al.*, 2000; Mazzola & Sara, 2001). In organisms, the nitrogen stable isotopic composition ($\delta^{15}N$) of tissues provides a powerful tool for determination of respective sources of nutrition for consumers and trophic relationships among organisms. In the trophic network among animals, $\delta^{15}N$ of their tissues systematically increase on average by 3.4‰ per trophic level (enrichment ranging from 1.3 to 5.3‰; Minagawa & Wada, 1984).

The aim of the present study was to examine the influence of particulate organic nitrogen waste deriving from fish farms on the nitrogen stable isotope composition of fouling communities colonising hard substrates (hereafter referred to as biofilters) adjacent to the fish farm.

MATERIALS AND METHODS

Three study sites were selected: Crete (E Mediterranean), Eilat (Red Sea) and Piran (N Adriatic). Samples of particulate organic matter and fouling communities colonising the biofilters were taken at the fish farms and at reference locations, which had not been affected by aquaculture.

Particulate organic matter (POM) was collected in sediment traps seasonally from autumn 2001 to spring 2003. Samples were filtered through GF/C glass fibre filters and dried in oven overnight at 50 °C.

Samples consisting of entire fouling community on randomly selected biofilters (one per array) were removed from the substrate, dried, pulverised by grinding to particle size below 100μm and subsequently analysed for nitrogen isotope composition.

Nitrogen stable isotope composition ($\delta^{15}N$) was determined using a continuous flow isotope ratio mass spectrometer Europa 20-20 with ANCA SL preparation module (PDZ Europa Ltd., U.K.). Samples were packed in tin capsules, combusted in elemental analyser and gases produced were separated in a GC column. The results are expressed as relative δ values in ‰, *i.e.* the difference in parts per mil of the isotopic ratios $^{15}N/^{14}N$ from those of atmospheric nitrogen. Laboratory working standards (urea and ammonium sulphate solutions with 100 μg/μL N) were calibrated versus IAEA N-1 and IAEA N-2 $(NH_4)_2SO_4$ with $\delta^{15}N$ +0.4 and +20.3‰, respectively. Untreated GF/C filters and empty tin capsules were used as blanks. Samples were analysed in triplicate to assess the precision and the reproducibility of the analyses. Standard deviation of the measurement determined on multiple analyses of working standards was generally ≤ 0.2‰, while standard deviation of the samples was generally better than ≤ 0.3‰.

RESULTS AND DISCUSSION

Average $\delta^{15}N$ values of particulate organic matter and fouling communities at reference locations and fish farms in Crete, Eilat and Piran are listed in Table 1.

POM was enriched in ^{15}N at the fish farms compared to the reference locations at all sites, as a consequence of the presence of ^{15}N enriched organic debris originating from the cages (faeces with average $\delta^{15}N$ between 7 and 10.6‰, remains of fish food with $\delta^{15}N$ values between 7.6 and 9.7‰). An exceptionally high enrichment in ^{15}N in POM was detected in Crete, where the large difference cannot be attributed only to the presence of organic waste from the farm, but is partially related to the seasonally dependent differences in nitrogen isotopic composition of plankton, where $\delta^{15}N$ values can change by several per mil related to the temperature-dependent nitrogen cycling (Minagawa & Wada, 1984; Sigleo & Macko, 1985; Nakatsuka *et al.*, 1992; Montoya, 1994; Altabet *et al.*, 1999). The high average $\delta^{15}N$ value is a consequence of extreme enrichment of POM in ^{15}N in July 2001, when $\delta^{15}N$ values of 11-13 ‰ were determined. Later on the $\delta^{15}N$ ranged from 7.2 to 8.6 ‰, yielding a seasonal average of 9.2 ‰.

Tab. 1: Average $\delta^{15}N$ values of particulate organic matter and fouling communities at reference locations and fish farms in Crete, Eilat and Piran.
Tab. 1: Povprečne vrednosti $\delta^{15}N$ suspendiranih organskih snovi in združbe obrasti na referenčnih lokalitetah in v ribogojnicah na Kreti, v Eilatu in Piranu.

Location	$\delta^{15}N$ of POM [‰]	$\delta^{15}N$ of fouling community [‰]
CRETE		
Reference	4.2	4.5
Fish farm	10.8 (9.2*)	6.3
EILAT		
Reference	3.6 (3.8*)	3.6
Fish farm	4.8 (4.9*)	5.7
PIRAN		
Reference	5.3 (4.7*)	6.7
Fish farm	5.8 (5.7*)	5.7

*seasonal averages

Fouling communities at reference locations were enriched in ^{15}N relative to the POM at all three sites, where the enrichment in Eilat was within the analytical error of the measurements. An enrichment of fouling communities in ^{15}N at the fish farms relative to the reference locations was observed in Crete and Eilat, while in Piran the opposite occurred. The relation of $\delta^{15}N$ of POM to $\delta^{15}N$ of fouling communities at the fish farms, however, seems controversial.

The organisms are enriched in ^{15}N relative to their food sources by at least 1.3‰, depending on the species (Minagawa & Wada, 1984), therefore the observed enrichment can be used for an overall estimation of the sources of nutrition of fouling organisms. Where the organisms are enriched in ^{15}N relative to the POM, at least part of their nutrition derives from this source. Where the enrichment is very small or even negative, it is obvious that the particulate organic matter does not contribute to the average organisms' diet. Since the colonising communities are composed of different species, only average particulate nitrogen consumption of the whole community colonising the analysed biofilter can be detected.

^{15}N-enrichment of organisms at the fish farms relative to those at reference locations indicates that some of the incorporated nitrogen derives from the fish farms, however, its fractions depend mostly on the composition of the colonising community, *i.e.* relative abundance of non-selective suspension feeders, which are able to ingest and retain nitrogen from particles of various sizes. In Crete, the fouling community at the fish farm is clearly enriched in ^{15}N relative to the reference location, however, depleted in ^{15}N relative to the POM. That means that suspension feeders present only a minor fraction of the whole community. Similar situation was observed in Piran.

Figure 1 shows the enrichment of fouling communities relative to the POM at the fish farms and at reference locations. Different enrichments between POM and fouling communities at the fish farms and at reference locations show that the compositions of fouling communities at both locations were obviously different and highly variable from biofilter to biofilter, as well as seasonally, which can be seen from the large range of measured $\delta^{15}N$ values. Therefore they cannot be directly compared to each other. However, the overall enrichment of the organisms at the fish farms relative to the reference locations in Crete and Eilat shows that they are enriched in ^{15}N, *i.e.* at least part of their food is represented by ^{15}N enriched POM deriving from the cages.

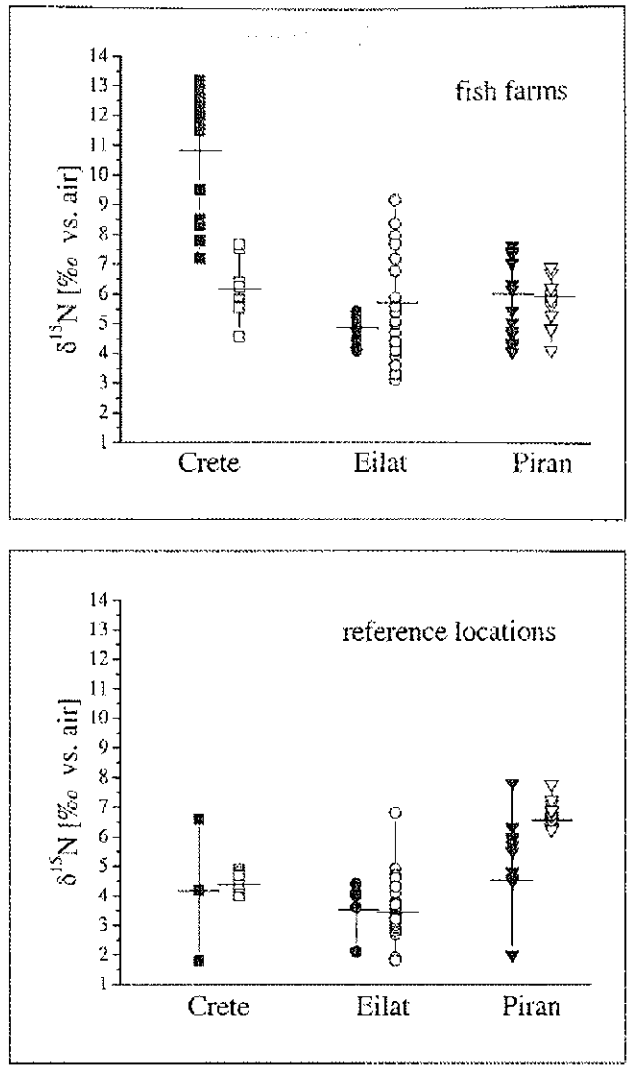

Fig. 1: $\delta^{15}N$ of particulate organic matter and fouling communities at the fish farms and at reference locations; solid symbols = POM, open symbols = fouling community. Horizontal bars represent average values of all measurements.
Sl. 1: $\delta^{15}N$ suspendiranih organskih snovi in združba obrasti v ribogojnicah in referenčnih lokalitetah; polni simboli = POM, odprti simboli = združba obrasti. Horizontalni stolpiči ponazarjajo povprečne vrednosti vseh meritev.

IZOTOPSKA SESTAVA DUŠIKA V MORSKIH ORGANIZMIH V VPLIVNEM OBMOČJU RIBJIH FARM

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POVZETEK

Analizirali smo izotopsko sestavo dušika v obrasti na umetnih substratih, lociranih v neposredni bližini ribjih farm in na neonesnaženih referenčnih lokacijah v severnem Jadranu (Piran, Slovenija), v vzhodnem Sredozemlju (Kreta, Grčija) in v Rdečem morju (Eilat, Izrael). Na dveh od treh lokacij smo izmerili povečano vsebnost $\delta^{15}N$ v organizmih ob ribjih farmah, kar je posledica vgrajevanja izotopsko težjega dušika iz suspendirane snovi, ki se sprošča v okolico iz ribjih kletk.

Ključne besede: ribja farma, stabilni izotop, dušik, umetni substrat, organizmi obrasti

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