ELSEVIER

Aquaculture 192 (2001) 361–379

Aquaculture

www.elsevier.nl/locate/aqua-online

The effect of fish farming organic waste on food availability for bivalve molluscs (Gaeta Gulf, Central Tyrrhenian, MED): stable carbon isotopic analysis

A. Mazzola, G. Sarà *

Marine Biology and Sea Resources Laboratory, Department of Animal Biology, University of Palermo, Via Archirafi, 18, I-90123 Palermo, Italy

Received 27 March 2000; received in revised form 26 June 2000; accepted 26 June 2000

Abstract

Stable carbon isotope ($\delta^{13}C$) analysis was used in a fish-farming impacted Mediterranean area (the Gulf of Gaeta, Central Tyrrhenian) to determine the predominant carbon sources available to bivalve molluscs cultivated around fish cages. Whether the organic matter generated by fish farming was taken up by the bivalve molluscs was also investigated. Stable carbon isotope values were measured in the particulate organic carbon (POC) of samples from potential organic matter sources such as fish-pelleted feed, mollusc faecal waste and bivalve flesh. The sources of organic matter affecting the study area water column and benthic communities appeared to be terrigenous-continental, autochthonous (phytoplankton) and anthropogenic inputs due mainly to fish-farming and bivalve mollusc activities. The POC was dominated by organic waste isotopic signatures, while the bivalve mixed diet was composed of organic matter with different isotopic signatures (phytoplankton, waste material from the bivalves themselves and surplus uneaten pelletted feed). Organic waste appears to be the dominant trophic resource in the deeper-cultivated clam diet, while phytoplankton organic carbon plays a more important role in the diet of the mussel. We propose that bivalve organic matter uptake may play an effective role in reducing the environmental impact of fish organic waste. The organic matter produced by bivalves (faecal material) under these hydrodynamic conditions (low current velocities) can be recycled through the filtration activities of the bivalves themselves, together with most of the organic matter produced by fish-farming activities (uneaten feed and faecal material). Bivalve cultivation around

* Corresponding author. Tel.: +39-091-6230127; fax: +39-91-6230144.
E-mail address: gsara@mbox.unipa.it (G. Sarà).

0044-8486/01/$ - see front matter
PII: S0044-8486(00)00463-4

cages may reduce the environmental impact of organic waste from fish-farming activities and increase the profitability of fish culture activities.

Keywords: Environmental impact; Organic waste; Food availability; Bivalve mollusc; $\delta^{13}C$; MED

1. Introduction

In the Mediterranean, as in the rest of the world, it is common practice to couple fish farming with shellfish cultivation, particularly of bivalve molluscs. Intensive cultivation in open-sea fish cages produces considerable amounts of particulate organic matter (POM) in the form of suspended detritus, which is mainly composed of uneaten feed and faecal material (Ye et al., 1991). This organic waste disperses in the water column and may be a food source for bivalve molluscs such as mussels, oysters and clams. Bivalve filter feeders are essentially generalist consumers (Dame, 1996) of POM and it has been demonstrated that they can exploit organic matter from several sources (autochthonous, terrigenous natural allochthonous or anthropogenic) as a function of its availability (Langdon and Newell, 1990; Stirling and Okumus, 1995; Riera and Richard, 1997; Sarà et al., 1998; in press). Therefore, for bivalves, which filter seston (Dame, 1996; Wildish and Kristmanson, 1997) in fish-farming impacted areas organic waste (uneaten pelletted feed, fish and bivalve faeces) represents a potential food source. The direct use of the waste organic matter by bivalves could reduce its impact on the environment (Shpigel and Blaylock, 1991; Shpigel et al., 1991, 1993b), increasing the cultivated edible biomass and, potentially, the profitability of farming activities (Jones and Iwama, 1991; Shpigel et al., 1993b; Léfebvre et al., 2000). Several authors have demonstrated that organic carbon originating from uneaten feed and faecal material produced by cultivated fishes represents a source of available food for shellfish. Shellfish may thus act as biological filters, being environmental cleaner (Neori et al., 1998, 2000; Shpigel et al., 1991, 1993a,b; Shpigel and Neori, 1996). However, to our knowledge, none of these authors have tested this hypothesis using the natural stable carbon isotope tracer. Consequently, we have designed a study involving the use of the stable carbon isotope composition ($\delta^{13}C$, ‰) of organic matter sources channelled into food webs (Peterson et al., 1985) and in particular through bivalves cultivated around fish cages. At present, this analytical method is considered one of the best tools for identifying the original food sources of benthic consumers (Riera and Richard, 1996; Peterson et al., 1985; Peterson and Fry, 1987; Fry, 1988; Kreeger et al., 1988; Michener and Shell, 1994; Hwey-Lian et al., 2000). The stable carbon isotopic composition is a characteristic of every organic matter source and the average $\delta^{13}C$ of consumers relates to that of their food (De Niro and Epstein, 1978) through well-known metabolic isotopic fractionation processes. Harrigan et al. (1989) have defined metabolic fractionation as the difference between the isotopic composition of a consumer and that of the food source. Thus, the carbon isotope ratios of animals reflect those of their food resources plus a slight enrichment (the average for marine invertebrates is about 1–1.5‰) (De Niro and Epstein, 1978; Fry and Sherr, 1984).

The aims of the present study were (i) to identify the main sources of organic matter in a Mediterranean-sheltered fish-farming area; (ii) to investigate whether the organic matter generated by fish farming is taken up by bivalve molluscs, by analysing their somatic carbon isotopic composition and whether it represents an effective food resource for them; and (iii) to investigate whether faecal material produced by bivalves cultivated near cages re-enters the bivalve food chain or whether it represents a further, unutilised organic matter load for the near-the-cage environment.

2. Materials and methods

2.1. The study area

Sampling was carried out seasonally in the Gulf of Gaeta (Mediterranean Sea; LAT N 51°14′21″; LONG E 13°35′12″), an area, which is sheltered from dominant winds (data source: Forecast Weather Airforce Service, Gaeta Meteorological Station reported in Lá Rosa, 1999). The study area is characterised by low current velocities with an average yearly range of 1–5 cm s^{-1} (Lá Rosa, 1999). No significant bottom current (as measured by current meter) was present (Lá Rosa, 1999). The area is seasonally constrained by terrigenous-continental inputs, which originate from two nearby streams (at about 15 km distance). The sand–muddy sediments of the Gulf are generally unvegetated and no primary macro-producers (such as algae or phanerogames) were present. The depth of the sampling sites was about 10 m.

Intensive fish cultivation (*Dicentrarchus labrax* and *Sparus aurata*) represents an important commercial activity (250 t $year^{-1}$) in the Gulf of Gaeta, which is also the most important area in the central Tyrrhenian for the commercial production of bivalve molluscs. Mussels (*Mytilus galloprovincialis*) and clams (*Tapes* sp.) are cultivated in long lines around cages and in suspended baskets, with an annual production of approximately 400 t $year^{-1}$.

2.2. Sampling, sample preparation and analytical technique

The study was designed to investigate the carbon isotopic composition of POM, of the principal OM sources and the relative isotopic signature of the two bivalve molluscs throughout 1 year. The stable carbon analysis of the POM was carried out seasonally from March 1997 to February 1998 at two depths (sub-surface and bottom [10 m]) in two sampling stations inside the Gulf. Both sites are constrained by anthropogenic inputs (i.e. fish organic waste). The first station (hereafter called Cage station) was positioned near the fish cultivation cages, while the second (hereafter called Shellfish station) was between the suspended long lines (about 200 m from the cages). Two stations were taken as controls: the first, called Internal Control (I-CTRL), was positioned inside the Gulf at about 1 km from the cages and shellfish plants at the same depth (this station was not considered in the ANOVA design [see below] as only one site was sampled). This station was representative of the area but without organic waste inputs. The second station was called External Control (E-CTRL). In this case, data were extrapolated from

a data set taken from a monthly environmental study carried out at the same depth in another unvegetated Mediterranean area outside the Gulf. This area has a low frequency of terrigenous and no anthropogenic inputs of organic matter (Mazzola et al., 1999c; Sarà et al., 1999). In the present study, it was assumed that the POM signatures determined in this station at one depth only (5 m; area depth 10 m) were representative of a trophic situation in which organic matter was affected by autochthonous production (i.e. phytoplankton) (Mazzola et al., 1999c; Sarà et al., 1999) in the absence of other extreme carbon signals such as phanerogames or constant terrigenous-continental inputs.

Water samples were collected monthly at two depths (surface and 10 m [i.e. bottom]) from the sampling sites using Niskin bottles. The samples were then immediately screened through a 200-μm mesh net to eliminate larger zooplankton and debris. They were then filtered under moderate vacuum and within 2 h of collection onto pre-washed, precombusted (450°, 4 h) and pre-weighed Whatman GF/F filters (0.70 μm nominal pore size) and then stored frozen until laboratory analysis.

Particulate organic carbon (POC, mg C l^{-1}), its isotopic composition (δ^{13}C,‰), the elemental C/N ratio of the particulate matter, the biopolymeric particulate organic carbon (BPC, mg C l^{-1}; Fabiano and Pusceddu, 1998) and phytoplankton carbon calculated from suspended chlorophyll-*a* (C-CHLa, mg C l^{-1}) concentrations were determined. The CO_2 produced by combustion of the tin capsule containing the sample in a Carlo Erba (Italy) elemental analyser (model EA1110) was analyzed in a mass spectrometer (Delta Plus, Finnigan MAT). The results expressed in the usual δ units (‰) were reported against the PDB international standard (Craig, 1953) and the reproducibility of the δ^{13}C determination was ± 0.2‰.

The biopolymeric fraction of the POC was determined as the sum of the carbon equivalents of the three main biochemical components of POM: particulate carbohydrates, particulate proteins and particulate lipids. Particulate carbohydrate concentrations (CHO, μg l^{-1}) were measured according to Dubois et al. (1956) and reported as glucose equivalents. Particulate proteins (PRT, μg l^{-1}) were determined according to Hartree (1972) and reported as bovine serum albumin (BSA) equivalents. Particulate lipid concentrations (LIP, μg l^{-1}), measured according to Bligh and Dyer (1959) and Marsh and Weinstein (1959), were reported as tripalmitin equivalents. Carbohydrates, proteins and lipids were converted into carbon equivalents using the coefficients 0.40, 0.49 and 0.75, respectively. CHLa (μg l^{-1}) was determined according to Lorenzen and Jeffrey (1980). Phytoplankton carbon (C-CHLa) was calculated by converting CHLa concentrations to carbon content (C-CHLA) using a conversion factor of 52 (Nival et al., 1972).

Analysis of the isotopic composition of the bivalves and their faecal material and pseudofaeces was carried out in two seasons (spring 1997 and winter 1998). We manually sampled adult specimens of *M. galloprovincialis* (sample size = 25; average length 45 ± 0.5 mm) cultivated on suspended long lines (rope height was about 3 m from the surface) near the fish cages and adults of *Tapes* sp. (sample size = 25; average length 20 ± 0.3 mm) cultivated in baskets suspended 1 m over the sea bottom (at about 9 m depth). The sampled organisms (pooled $n = 7$) were cleaned of epibionts and kept alive overnight in filtered sampling site water (GF/F, 0.7 μm) to allow evacuation of their gut contents (Riera, 1997). Bivalve flesh (after dissection from the shell), faeces

and pseudofaeces (the latter for mussels only) were collected separately using pipettes, ground using a mortar and pestle, acidified with 1 M HCl and stored frozen (−80°C) until laboratory carbon isotope analysis (as described above).

Samples of different types of commercial feed of four different sizes (RD 97/15 [size 2] BioMar [France] (P1), Perla Plus [size 3.0] Hendrix [Italy] (P2), RD 97/15 [size 4.5] BioMar [France] (P3) and Aqualife [size 6.0] BioMar [France] (P4) commonly used to feed fish in cages were also treated as described above (grinding, acidification and freezing). These samples were then analysed for their stable carbon isotopic composition as above. As the diet of the cultivated fish was mono-specific (pellet only), no isotopic analysis was carried out on the fish faeces because its isotopic value and variability could be predicted to undergo a slight enrichment (~ 1‰) with respect to the food source (viz. pellet; Ye et al., 1991).

The isotopic signatures of the other two potential organic matter sources (terrigenous input from streams and mixed phytoplankton) were taken from the current literature (Fry and Sherr, 1984; Dauby, 1989; Jennings et al., 1997; Riera, 1997; Sarà et al., 1999). To assess the contribution of the different carbon sources entering the POM and available to molluscs, the mixing model was used (Fry and Sherr, 1984; Dauby, 1989).

2.3. Statistical analyses

To test the hypothesis that POM features varied as a function of time and space and were constrained by organic waste, a four-way ANOVA was used (mixed design; Underwood, 1997). Three factors were treated as fixed and orthogonal: station (cages and bivalves = two levels; STAT), season (spring, summer, autumn and winter = four levels; SEAS), and depth (surface and 10 m = two levels; DEP). Sites (two for each station) were treated as random (two levels; SITE) and nested in STAT, SEAS and DEP. Two replicates were effected randomly at each site. In addition, a further ANOVA (three-way) was performed in order to verify whether the patterns observed in the Gaeta Gulf were similar to those of the External control site. In this case, two factors were treated as fixed and orthogonal: area (External control and Gaeta [averaged data between cages and bivalves] = two levels; AREA) and season (spring, summer, autumn and winter = four levels; SEAS). Sites (two for each area) were treated as random (two levels; SITE) and nested in AREA and SEAS. In all analyses, the heterogeneity of variances was tested using Cochran's test prior to the analysis of variance and the appropriate means compared using Student–Newman–Keuls (SNK) tests (Underwood, 1997). The GMAV (1997) statistical package (University of Sydney, Australia) was used to perform ANOVA, while other statistics were assessed using STATISTICA (Statsoft) statistical package.

3. Results

3.1. The isotopic composition and C/N elemental ratio of the major organic carbon sources in the study area

The carbon isotopic composition of the main carbon sources as determined in the study area near the fish and shellfish culture locations are summarised in Table 1. The

Table 1
Carbon isotopic signatures of the main organic matter sources and consumers in the study area. (Obs = number of observations; $\delta^{13}C$, ‰ = carbon isotopic values; C/N = carbon by nitrogen ratios; ±s.e. = standard errors for means)

Substratum	Obs (n)	$\delta^{13}C$	±s.e.	C/N	±s.e.
Feed 1	3	−21.1	0.1	20.2	0.1
Feed 2	3	−23.1	0.2	13.6	0.2
Feed 3	3	−22.8	0.1	17.2	0.1
Feed 4	3	−23.2	0.1	14.8	0.1
Average feed	12	−22.6	1.0	16.4	2.9
Mussel	5	−20.5	1.2	16.3	1.2
Faeces	5	−23.3	0.1	27.4	0.1
Pseudofaeces	5	−23.1	0.1	28.2	0.1
Clam	5	−21.4	0.7	14.5	0.7
Faeces	5	−23.2	0.1	34.1	0.1
POM around cages (Stat 1)	32	−22.7	0.7	22.3	0.7
POM around bivalves (Stat 2)	32	−22.7	0.8	22.4	0.8
POM internal control	10	−22.6	0.9	22.5	0.9
POM external control	8	−21.3	2.5	5.9	2.5

first source of potential POM isotopic variability was the fish feed, with an isotopic composition, which ranged between −21.1‰ and −23.2‰ (average -22.6 ± 1.0‰). The elemental C/N ratio of the feed averaged 16.4 ± 2.9 and varied with respect to the relative isotopic composition, following a significant linear relationship described as follows: $\delta^{13}C = -27.6 + 0.31$ C/N ($n = 12$; $r = 0.91$; $P < 0.05$). Another potential source of POM variability was faecal material from the cultivated bivalves. The average isotopic signature of the mussel faecal material was -23.3 ± 0.1‰ and elemental C/N 27.4 ± 0.1, while that of the clams averaged -23.2 ± 0.1‰ and the elemental C/N 34.1 ± 0.1. Since the clams did not produce pseudofaecal material in either sampling period, the isotopic composition was determined only on the mussel pseudofaeces, which was on average -23.1 ± 0.1‰ and the elemental ratio 28.2 ± 0.1.

3.2. Changes in the features of POC

The statistics of the variables in the study area are reported in Table 2A. Seasonality seemed to be the main effect on the features of suspended organic matter in the study area. ANOVA demonstrated that, apart from POC, $\delta^{13}C$ and the C/N ratio, BPC and C-CHLa varied primarily as a function of time (see Tables 3 and 4; Fig. 1). The isotopic composition of organic carbon was significantly enriched in ^{13}C in summer and winter, while as shown in Fig. 2, $\delta^{13}C$ was more depleted in spring and autumn. No significant differences were observed as a function of station and interaction terms.

Elemental C/N varied with the station, as that measured in the cage sites was higher (23.2 ± 0.41) than that in the bivalve sites (21.7 ± 0.8). A seasonal effect was also evident for the C/N ratio, as it was significantly higher in summer and autumn than in

Table 2
Statistics of the variables measured in this study. (A) The seasonal averaged data set on which ANOVA was performed is summarised in Table 3; (B) The overall averaged data set on which ANOVA (E-CTRL vs. ALL Gaeta) was performed is summarised in Table 4. (I-CTRL = internal control and E-CTRL = external control; All Gaeta = averaged data between cages and bivalves). (δ^{13}C, ‰ = carbon isotopic values; POC, mg C l^{-1} = particulate organic carbon; C/N = elemental carbon by nitrogen ratio; BPC, mg C l^{-1} = biopolymeric organic carbon; C-CHLa, mg C l^{-1} = chlorophyll-*a* carbon)

	δ^{13}C				POC				C/N				BPC				C-CHLa			
	Mean	±s.e.	Min	Max	Mean	±s.e.	Min	Max	Mean	±s.e.	Min	Max	Mean	±s.e.	Min	Max	Mean	±s.e.	Min	Max
(A)																				
Spring	−23.53	0.62	−25.00	−22.96	0.40	0.27	0.19	1.22	22.57	2.28	18.55	26.81	0.14	0.06	0.04	0.33	0.04	0.02	0.00	0.08
Summer	−22.59	1.50	−24.97	−19.81	0.49	0.52	0.13	1.98	24.34	2.10	21.59	29.48	0.07	0.03	0.04	0.12	0.04	0.04	0.00	0.18
Autumn	−23.03	0.48	−23.78	−22.28	0.24	0.11	0.10	0.40	23.81	1.85	19.96	25.82	0.08	0.03	0.03	0.13	0.06	0.03	0.02	0.11
Winter	−22.45	0.30	−23.04	−22.00	0.30	0.11	0.12	0.53	20.49	1.34	18.14	22.40	0.21	0.08	0.09	0.41	0.10	0.05	0.03	0.20
Study year	−22.90	0.94	−25.00	−19.81	0.36	0.31	0.10	1.98	22.79	2.43	18.14	29.48	0.13	0.09	0.03	0.41	0.06	0.04	0.00	0.20
(B)																				
Cages	−22.90	0.91	−24.57	−19.81	0.32	0.25	0.10	1.36	23.05	2.50	19.14	29.48	0.13	0.09	0.03	0.37	0.06	0.05	0.00	0.18
Bivalves	−22.90	0.99	−25.00	−21.16	0.40	0.37	0.00	1.98	22.49	2.38	18.14	26.81	0.12	0.09	0.04	0.41	0.05	0.04	0.00	0.20
I-CTRL	−22.91	0.82	−24.64	−21.42	0.35	0.37	0.00	1.31	23.57	2.93	18.41	29.33	0.13	0.09	0.03	0.34	0.06	0.05	0.00	0.17
ALL Gaeta	−22.90	0.95	−24.79	−20.49	0.36	0.31	0.05	1.67	22.77	2.44	18.64	28.15	0.13	0.09	0.04	0.39	0.06	0.05	0.00	0.19
E-CTRL	−21.34	2.47	−25.10	−16.40	2.45	0.32	1.90	2.80	5.90	1.87	1.29	7.88	0.21	0.10	0.08	0.37	0.02	0.01	0.00	0.04

Table 3
Analysis of variance on all measured variables performed to verify differences between cage and bivalve stations, seasons and depth and interaction between these main effects. ($\delta^{13}C$ = carbon isotopic values; POC = particulate organic carbon; C/N = elemental carbon by nitrogen ratio; BPC = biopolymeric organic carbon; C-CHLa = chlorophyll-*a* carbon) (§ = data transformed to natural logarithms) [Ns = non-significant difference ($P > 0.05$)]

Source of variation	Df	$\delta^{13}C$			POC			C:N			BPC			C-CHLa		
		MS	F	*P*	MS	F	*P*	MS	F	*P*	MS	F	*P*	MS	F	*P*
Station = STAT	1	0.0	0.0	Ns	1.1	65.7	*	39.7	13.9	*	0.0	0.0	Ns	2006	3.6	Ns
Season = SEAS	3	3.8	12.7	***	0.7	1.7	Ns	56.5	7.0	**	0.1	42.8	***	12073	19.4	***
Depth = DEP	1	2.7	8.4	Ns	0.2	0.5	Ns	68.2	5.5	Ns	0.1	8.9	Ns	11222	4.4	Ns
Site (STAT × SEAS × DEP)	16	0.3	0.3	Ns	0.4	1.6	Ns	8.1	0.8	Ns	0.0	0.2	Ns	623.9	0.4	Ns
STAT × SEAS	3	0.3	1.0	Ns	0.0	0.0	Ns	2.9	0.4	Ns	0.0	0.8	Ns	552.8	0.9	Ns
STAT × DEP	1	0.0	0.0	Ns	0.3	0.4	Ns	1.2	0.1	Ns	0.0	0.0	Ns	307.2	0.6	Ns
SEAS × DEP	3	0.4	1.1	Ns	0.4	0.9	Ns	12.4	1.5	Ns	0.0	5.5	**	2578	1.2	Ns
STAT × SEAS × DEP	3	0.6	2.1	Ns	0.9	2.1	Ns	13.7	1.7	Ns	0.0	1.9	Ns	496.7	0.8	Ns
Residual	32	1.0			0.3			10.4			0.0			1611		
Cochran's C				Ns			Ns(§)			Ns(§)			Ns			Ns

* $P \leq 0.05$.
** $P \leq 0.01$.
*** $P \leq 0.001$.

Table 4
Analysis of variance on all variables measured. ($\delta^{13}C$ = carbon isotopic values; POC = particulate organic carbon; C/N = elemental carbon by nitrogen ratio; BPC = biopolymeric organic carbon; C-CHLa = chlorophyll-*a* carbon) (§ = Data transformed to natural logarithms) [Ns = non-significant difference ($P > 0.05$)]

Source of variation	Df	$\delta^{13}C$			POC			C:N			BPC			C-CHLa		
		MS	F	*P*	MS	F	*P*	MS	F	*P*	MS	F	*P*	MS	F	*P*
Area = AR	1	65.3	38.4	***	65.8	172.6	***	2334.2	379.1	***	2.2	4.5	Ns	0.0	10.9	**
Season = SEAS	3	9.8	5.8	*	1.4	3.7	Ns	1.1	0.2	Ns	0.3	0.6	Ns	0.0	0.7	Ns
Site (AR x SEAS)	8	1.7	0.7	Ns	0.4	0.9	Ns	6.2	2.3	Ns	0.5	2.2	Ns	0.0	6.6	***
AR x SEAS	3	7.9	4.6	*	1.6	4.1	*	1.6	0.3	Ns	0.7	1.3	Ns	0.0	0.6	Ns
Residual	16	2.4			0.4			2.7			0.2			0.0		
Cochran's C				Ns			Ns§			Ns			Ns§			Ns§

* $P \leq 0.05$.
** $P \leq 0.01$.
*** $P \leq 0.001$.

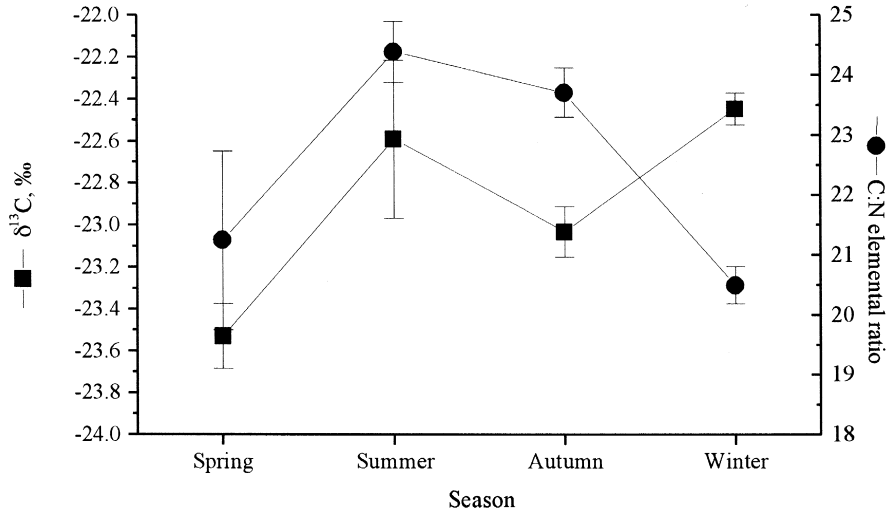

Fig. 1. Seasonal averaged trend of POC stable isotopic composition (δ^{13}C) and elemental C/N ratio in the Gulf of Gaeta. Standard errors are reported.

spring and winter (Fig. 2). BPC values were similar in each station studied, while changes were observed with the interaction between seasons and depths. Indeed, BPC values were more variable at the surface than at deeper sites, showing significant peaks in BPC concentration in winter (0.25 ± 0.05 mg C l^{-1}) and spring (0.20 ± 0.03 mg C

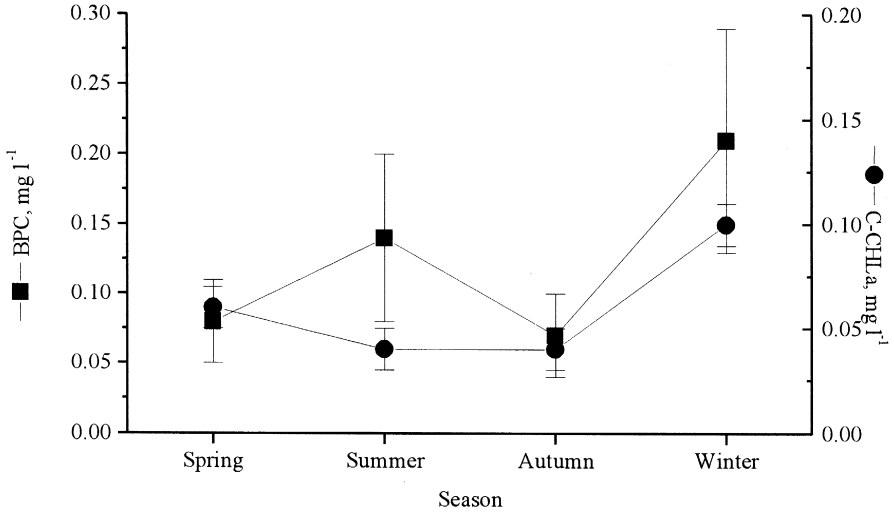

Fig. 2. Seasonal averaged trend of biopolymeric organic carbon (BPC) and chlorophyll-*a* carbon (C-CHLa) in the Gulf of Gaeta. Standard errors are reported.

l^{-1}) and the minimum in summer (0.07 ± 0.01 g C l^{-1}). This trend was not followed at 10 m depth, where concentrations were lower (0.15 ± 0.01 mg C l^{-1}) than at the surface depth (0.08 ± 0.01 mg C l^{-1}). At 10 m depth, winter concentrations were significantly higher (0.16 ± 0.02 g C l^{-1}) than in other seasons, which did not show differences between them (0.06 ± 0.01 g C l^{-1}). POC varied with space (stations), with higher concentrations (0.41 ± 0.06 mg C l^{-1}) around bivalves than cages (0.31 ± 0.04 mg C l^{-1}), but did not vary as a function of season or depth.

Suspended carbon CHLa showed a seasonality effect only (Table 4), with a significant peak of 0.96 ± 0.13 mg C l^{-1} in winter, while values measured in the other seasons were more similar and no significant differences were detected between them. This trend was followed in both stations and depths. Similar values were also obtained in the Internal control station, which showed the same patterns overall as the cage and bivalve stations (Table 2a). In contrast, ANOVA performed on the comparison matrix (External control vs. Gaeta data) showed large differences for most of the variables measured, which were basically maintained in all seasons (see Table 2b for averaged values and Table 4 for ANOVA). The y-intercept calculated from linear regressions between $\delta^{13}C$ and POC was a useful tool for identifying the main origin in the POC, as revealed by its isotopic composition in the POC. The intercept calculated from the POC–$\delta^{13}C$ relationship was -22.9 (the entire equation was $\delta^{13}C = -22.9 + 0.07 \times POC$; $n = 84$; $P = 0.05$), while that from the BPC–$\delta^{13}C$ relationship was -23.1 (the entire equation was $\delta^{13}C = -23.1 + 0.02 \times BPC$; $n = 84$; $P = 0.05$). The results of the mixing equations (Table 5) indicate that the mean relative contribution of organic wastes (fish feed +

Table 5
Percentage incidence of each main carbon source constraining the seasonal carbon isotopic signatures of the POC in the study area, estimated using the mixing isotope model (Fry and Sherr, 1984; Dauby, 1989). Reference signatures were: −21.4‰ for phytoplankton (Dauby, 1989; Jennings et al., 1997), −26.0‰ for terrigenous-continental carbon (Riera, 1997) and the measured isotopic value of −23.0‰ for organic waste (averaged value of rejection and uneaten pelletted food). ($\delta^{13}C$ = POC isotopic value measured seasonally in this study; Terrig C = percentage value of terrigenous carbon; Waste C = percentage value of waste carbon; Phyto C = percentage value of phytoplankton carbon)

Season	Depth	$\delta^{13}C$	%Terrig C	%Waste C	%Phyto C
Spring	0	−23.5	32.0	30.0	37.0
Summer	0	−22.1	10.0	75.0	15.0
Autumn	0	−22.8	17.0	46.0	37.0
Winter	0	−22.4	12.0	60.0	28.0
		Mean	17.8	52.8	29.3
		±s.d.	9.9	19.2	10.4
Spring	10	−23.6	22.0	30.0	48.0
Summer	10	−23.1	20.0	32.0	48.0
Autumn	10	−23.2	22.0	30.0	48.0
Winter	10	−22.6	15.0	54.0	31.0
		Mean	19.8	36.5	43.8
		±s.d.	3.3	11.7	8.5
Annual means of		Mean	18.8	44.6	36.5
water column		±s.d.	6.9	17.1	11.7

bivalve faeces + mussel faeces) to POC was markedly higher (45%) than that of phytoplankton (36%) and terrigenous inputs (19%). Phytoplankton carbon contributed more at deeper sites (44%) than at the surface (30%). The terrigenous contribution was similar at both depths (about 18%), while organic waste was much more abundant at the surface (53% at the surface vs. 36% near the bottom).

3.3. Somatic and biodeposition isotopic composition of bivalves

There were no marked differences in the somatic isotopic composition of mussels throughout both sampling periods (spring and winter). The average $\delta^{13}C$ was $-20.5 \pm 1.0‰$ and the elemental C/N ratio 16.3 ± 1.1. The isotopic composition of the mussel biodeposition material (faeces + pseudofaeces; Table 1) was more depleted than the somatic composition and the C/N ratio was much higher (27.8 ± 0.1). The somatic composition of the clams was more depleted ($-21.4 \pm 0.7‰$) than that of the mussels and the clam faeces was more depleted ($-23.2 \pm 0.1‰$) than its somatic signature. In addition, the C/N ratio of faecal material was higher (34.1 ± 0.1) than that of the mussels (approximately 27.5 ± 0.1).

4. Discussion

4.1. The organic carbon sources in the study area as revealed by $\delta^{13}C$ analysis

The Gulf of Gaeta is a typical Mediterranean area constrained by several types of suspended organic matter, which may be included in the diet of cultivated filter feeders molluscs (Fig. 3). The sources of organic matter affecting the study area water column and benthic communities appeared to be terrigenous-continental, autochthonous (phytoplankton) and anthropogenic inputs due mainly to fish-farming and bivalve mollusc activities (Favaloro, 1999). In this context, identifying the relative contribution of each organic matter source and in particular that originating from terrigenous inputs and primary production was not a straightforward matter. Firstly, the role of terrigenous-continental input was difficult to evaluate because, at these latitudes, these inputs are characterised by their low frequency and are closely correlated with seasonal rain (spring and autumn). Although on a yearly basis the terrigenous POC contribution does not exceed an average of 18–20% of the total carbon inputs (Table 5), the isotopic mixing model results substantiate that terrigenous POC may play an important role in spring and autumn, constraining the POC isotopic signatures during these periods. On the other hand, the C/N values in the study area were quite high (and significantly different from the external control), particularly in correspondence with the main terrigenous inputs from streams. This suggests that refractory terrigenous organic matter was present seasonally in the water column. It is likely that in spring and autumn mainly the terrigenous detritus-attached bacteria represent a potential spot trophic source for benthic organisms (Langdon and Newell, 1990; Riera, 1997; Lá Rosa, 1999; Sarà et al., 1999).

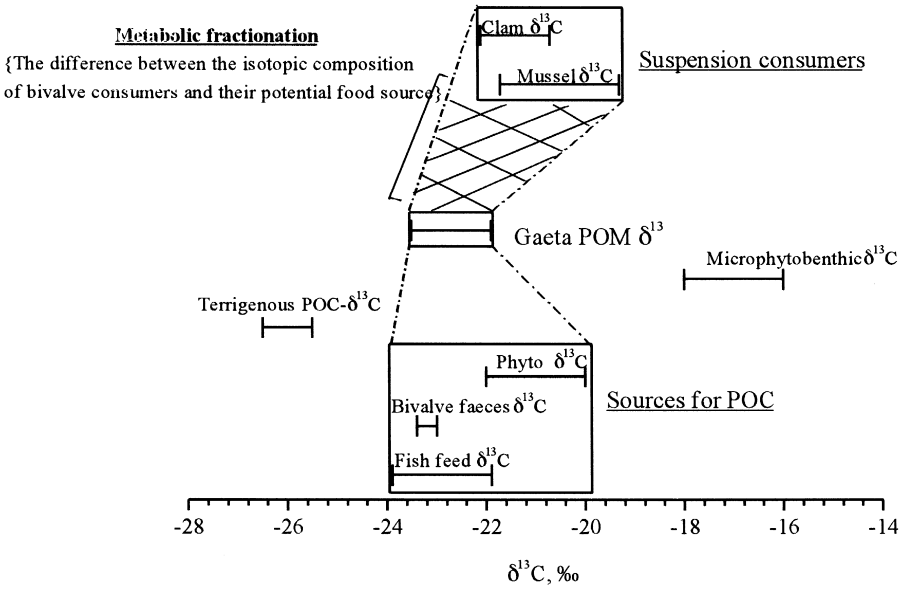

Fig. 3. Schematic diagram of ultimate carbon sources for bivalves in the Gulf of Gaeta. The height of each item along the *y*-axis is arbitrary.

Secondly, the difficulty of isolating phytoplankton accurately (Sarà, unpublished observation) in the present study prevents a definitive assessment of its relative contribution to the Gaeta POC and its role in the food web. However, at these Tyrrhenian latitudes, primary production is generally scant and the waters oligotrophic, with CHLa concentration below 1 $\mu g\ l^{-1}$, rarely exceeding the mesotrophic level (Margalef, 1985; Lá Rosa, 1999; Sarà et al., 1998). The low CHLa concentrations measured in the study area fit well into this Tyrrhenian pattern and the isotopic signal in the POC may reflect a relative phytoplankton contribution, which rarely exceeded 40–50% of the total (Table 5). The comparison with the External control station confirms that phytoplankton represents a secondary trophic input with respect to the other inputs (see C/N values; Table 2). The allochthonous nutrient inputs represent increasing factors of primary production indicating, in some localised situations and periods, an enhancement in the contribution of phytoplankton organic carbon to the filter feeder diet (Dame, 1996; Wildish and Kristmanson, 1997). Indeed, the phytoplankton organic matter is highly labile with respect to that coming from streams (terrigenous), which is mainly composed of highly refractory lignocellulosic compounds (Valiela, 1984; Mann, 1988; Langdon and Newell, 1990).

It has been hypothesised that in the study area, fish-farming waste represents a primary source of organic matter, which is diluted in the water column and potentially available to the suspension feeders cultivated around the fish cages. A similar suggestion has been put forward for other fish cultivation systems in the Mediterranean (Shpigel et al., 1993b), the Atlantic (Stirling and Okumus, 1995; Léfebvre et al., 2000) and the Pacific (Jones and Iwama, 1991). The mixing of carbon from these sources was

quantitatively constant throughout the year, while phytoplankton and terrigenous-continental organic carbons followed seasonal trends (Table 5). The POC isotopic signature in the Gulf was mainly dominated by organic waste signals, as demonstrated by the results of the mixing isotope equations (Fig. 3). This fact could substantiate the hypothesis that organic waste represents the most important source of organic matter in the Gaeta water column available to filter feeders. The mean intercept value from the POC–δ^{13}C relationship (−22.9‰) suggests that the isotopic signatures of POC may effectively reflect the averaged signature of the different organic wastes (mean carbon isotopic value of biodeposits = −23.2‰; mean carbon isotopic value of fish feed = −22.6‰; mean value of the two = −22.9‰). Instead, the intercept from the BPC–δ^{13}C relationships (−23.1‰) leads us to propose that the POC labile fraction (BPC) may originate primarily from a mixed detritus in which the isotopic signal of bivalve faecal material dominates.

4.2. The role of the different carbon sources in the diet of cultivated bivalves

Throughout the study period, the bivalve flesh δ^{13}C [−20.5‰ and 21.4‰ for mussel and clams, respectively] reflected the isotopic signature of organic waste plus a slight enrichment. More precisely, we envisage a mixed diet composed of organic matter with different isotopic signatures (Fig. 3) (phytoplankton [∼ −21.0‰; Dauby, 1989; Jennings et al., 1997], biodeposition material from the bivalves themselves [−23.2‰] and uneaten fish feed [−22.6‰]; mean isotopic signature of this mixing = −22.3‰). In such a mixed diet, organic waste appears to be the dominant organic source, remaining constant throughout the year (see also Table 5). If we apply the mixing model equations to the bivalve isotopic composition (Harrigan et al., 1989), the results confirm the dominant role of faecal and uneaten feed in the diet of the study area bivalves. Fish feed carbon may represent the main trophic resource in the deeper-cultivated clam diet, accounting for about 80% of the diet, biodeposition carbon the secondary source (4–20%) and, lastly, the phytoplankton carbon providing 0–15% of the diet.

In contrast, phytoplankton organic carbon may play a more important role in the diet of the mussel, although its role seems to vary seasonally (see the large standard deviation of the mean somatic isotopic composition of mussels). Phytoplankton carbon makes a contribution to the mussel diet, which varies seasonally from 5% to 100%, fish feed carbon contributes on average 50% and biodeposition carbon between 3% and 40%. These differences between the two bivalves may be explained by a species-specific response, which reflects their different food preferences (Dame, 1996) and depths of cultivation. These results lead us to exclude ^{13}C depleted terrigenous carbon as a trophic resource for bivalves. In addition, the mean fractionation schemes (0.9‰ and 1.8‰ for clams and mussels, respectively; mean fractionation value for both bivalves = 1.3‰) of the Gaeta bivalves with respect to their food sources fall well into the fractionation ranges reported for other bivalves (Peterson et al., 1985; Riera, 1997; Riera and Richard, 1997; Hwey-Lian et al., 2000) and described widely for other marine invertebrates (Dauby, 1989; Harrigan et al., 1989). Nevertheless, the highest isotopic fractionation measured in the mussels (about 2‰) was too high to be explained only by the metabolic ^{13}C enrichment occurring during the assimilation of food (∼ 1‰). Thus, we hypothesise

the presence of an additional carbon source, which is more enriched in ^{13}C and seasonally available to these organisms. Indeed, it has been observed that in many coastal areas the more enriched carbon from microphytobentic algae (about −16/−18‰; Couch, 1989) can also actively contribute to the diet of some filter feeders (e.g. *Crassostrea gigas*, Riera, 1997). However, we would exclude that benthic diatoms "normally" enter the diet of suspended mussels because throughout most of the study period the bottom current velocity of the study area ($^{*}u$; Ward et al., 1984; Smaal and Haas, 1997) was below the resuspension threshold (Sarà, unpublished data). Further investigation is needed to resolve and fully understand this apparent discrepancy. However, such an isotopic picture substantiates well the hypothesis that under the hydrodynamic conditions of the study area organic waste represents the main trophic source of cultivated near-the-cage bivalves. The results of the POC seasonal mixing model (Table 5) lead us to suppose that the contribution of each source varied as a function of the season and that phytoplankton carbon played a primary role in the diet, above all during spring and autumn. In addition, the intercept of the BPC–$\delta^{13}C$ relationships (−23.2‰) suggests that most of the labile fraction in the POC available to bivalves may originate from bivalve faecal detritus and very likely also from fish faeces. It has widely been demonstrated that, apart from phytoplankton carbon, the mucopolysaccharides contained in the pseudofaecal capsules abundantly produced by mussels (Dame, 1996) also represent a rich organic matter source, which is highly labile and available to suspension feeders. Assuming that fish somatic composition was similar or slightly enriched with respect to the fish feed (Ye et al., 1991), metabolic processes lead fish faeces to assume isotopic values, which are slightly depleted (about −23‰ in this study). Consequently, it is also likely that fish faeces represent a further depleted carbon source, which would justify the isotopic composition of bivalves.

4.3. Concluding remarks: environmental impact amplifiers or reducers?

Although our experiments were not designed to specifically test whether bivalves cultivated around fish cages play a role (either positive or negative) in the recycling of organic matter, the results of our isotopic analysis seem to substantiate such a hypothesis. Many authors have suggested that that bivalves cultivated in fish-farming areas may represent a biological filter (Shpigel and Blaylock, 1991) and that detrital waste from intensive fish farming can contribute to bivalve growth (Jones and Iwama, 1991; Stirling and Okumus, 1995; Léfebvre et al., 2000). By using the information from the carbon isotopic analysis of sources and consumers, we can tentatively estimate the relative contribution of the main components of the organic waste originating from the cultivation. In the study area 250 t of fish (*D. labrax* and *S. aurata*) are cultivated and if we adopt a conversion factor of 1.6 (Favaloro, 1999; Mazzola et al., 1999a), about 500 t of feed per year may enter the cultivation system. About 85–90% of feed should be transformed into fish biomass (Favaloro, 1999; Lopiano, 2000), while the remainder (approximately 50 t of surplus feed) should disperse in the water column around the cages. In addition, from field and laboratory experiments (Lopiano, 2000), it has been estimated that cultivated species produce on average approximately 0.4 mg g^{-1} wet-OM

die^{-1} of faeces. Consequently, on a yearly basis, the faeces dispersed in the water column should not exceed 5 t.

We can also estimate the mussel biodeposition input to the water column, taking into consideration that about 400 t of bivalves are cultivated yearly in the study area. If we consider a mean production rate of faecal and pseudofaecal material of 3 mg h^{-1} g of mussel (Widdows et al., 1979; Dame, 1996), about 1–3 t of organic matter could disperse yearly in the environment around the cages. Thus, we can hypothesise that approximately 55–58 t of organic matter originating from waste (surplus feed + faecal material from mussels + fish faeces) may disperse in the water column around the cages per year and potentially accumulate in the sediments. Consequently, we can say that suspension feeders are able to exploit suspended organic matter from waste (which our isotopic analysis suggests may represent overall the ultimate trophic resource in the diet of cultivated bivalves). About 86% of this matter is represented by surplus feed from fish cultivation, about 9% bivalve faecal and pseudofaecal material and about 5% fish ejection.

If we assume that (i) the waste organic matter components have a similar degree of availability with respect to the bivalve nutritional needs, (ii) bivalve somatic isotopic signatures ultimately reflected that of organic waste and (iii) mean isotopic signatures of sediments range between $-18‰$ and $-20.5‰$ (Sarà, unpublished data), then it is feasible to propose that bivalve organic matter uptake may play an effective role in reducing the environmental impact of fish organic waste. It seems that the organic matter produced by bivalves under these hydrodynamic conditions (low current velocities) can on the whole be recycled through the filtration activities of the bivalves themselves, together with most of the organic matter produced by fish farming activities (uneaten feed and faeces). However, further information is needed on the role of sedimentation and on the quantity of organic matter, which accumulate in sediments, as Mazzola et al. (1999b) observed changes in sediments beneath fish cages, which determined modifications in the total meiofaunal density.

In conclusion, we believe that in oligo-mesotrophic waters (such as most of the Mediterranean and several areas at the same latitudes) and fairly calm waters (with low current velocities; < 10 cm s^{-1}) bivalve cultivation around cages may produce at least two economic benefits: (i) it may reduce the environmental impact of organic waste from fish-farming activities, with the bivalves functioning as "recyclers" of allochthonous organic matter and contributing to "environmentally clean" aquaculture (Shpigel et al., 1993a) and; (ii) it may increase the profitability of fish cultivation, producing new edible biomass in coastal areas (Sarà and Mazzola, 1997; Sarà et al., 1998; Mazzola et al., 1999a) where these activities are seldom practised.

Acknowledgements

The authors would like to thank two anonymous referees for their indispensable help in improving the original version of the manuscript. This work was funded by the Ministero Politiche Agricole (MiPA, Italy) and Ministero dell'Universitá Ricerca Scientifica e Tecnologica (MURST, Italy).

References

Bligh, E.G., Dyer, W., 1959. A rapid methods for total lipid extraction and purification. Can. J. Biochem. Physiol. 37, 911–917.

Couch, C.A., 1989. Carbon and nitrogen stable isotopes of meiobenthos and their food resources. Estuarine Coastal Shelf Sci. 28, 433–441.

Craig, H., 1953. The geochemistry of the stable carbon isotopes. Geochim. Cosmochim. Acta 3, 53–92.

Dame, R.F., 1996. Ecology of Marine Bivalves. An Ecosystem Approach. CRC Press, Boca Raton, FL, p. 254.

Dauby, P., 1989. The stable isotope ratios in benthic food webs of the Gulf of Calvi, Corsica. Cont. Shelf Res. 9, 181–195.

De Niro, M.J., Epstein, S., 1978. Influence of diet on the distribution of carbon isotopes in animals. Geochim. Cosmochim. Acta 42, 495–506.

Dubois, M., Gilles, K.A., Hamilton, J.K., Rebers, P.A., Smith, F., 1956. Colorimetric method for determination of sugars and related substances. Anal. Chem. 28, 350–356.

Fabiano, M., Pusceddu, A., 1998. Total and hydrolizable particulate organic matter (carbohydrates, proteins and lipids) at a coastal station in Terra Nova Bay, Ross Sea, Antarctica. Pol. Biol. 19, 125–132.

Favaloro, E., 1999. Analisi morfometrica e studio delle anomalie scheletriche in *Diplodus puntazzo* (Cetti, 1977 Pisces: Sparidae): prove comparate tra diverse tipologie di allevamento. PhD Dissertation, University of Messina, Italy, p. 102.

Fry, B., 1988. Food web structure on Georges Bank from stable C, N and S isotopic compositions. Limnol. Oceanogr. 33, 1182–1190.

Fry, B., Sherr, E.B., 1984. $\delta^{13}C$ measurements ad indicators of carbon flow in marine and freshwater ecosystems. Contrib. Mar. Sci. 27, 13–47, Univ. Texas.

Harrigan, P., Zieman, J.C., Macko, S.A., 1989. The base of nutritional support for the grey snapper (*Lutjanus griseus*): an evaluation based on a combined stomach content and stable isotope analysis. Bull. Mar. Sci. 44, 65–77.

Hartree, E.F., 1972. Determination of proteins: a modification of the Lowry method that gives linear photometric response. Anal. Biochem. 48, 422–427.

Hwey-Lian, H., Wen-Yuan, K., Chang-Po, C., Pi-Jen, L., 2000. Detrital flows through the feeding pathway of the oyster (*Crassostrea gigas*) in a tropical shallow lagoon: $\delta^{13}C$ signals. Mar. Biol. 136, 677–684.

Jennings, S., Renones, O., Morales-Nin, B., Polunin, N.V.C., Moranta, J., Coll, J., 1997. Spatial variation in the ^{15}N and ^{13}C stable isotope composition of plants, invertebrates and fishes on Mediterranean reefs: implications for the study of trophic pathways. Mar. Ecol.: Prog. Ser. 146, 109–116.

Jones, T.O., Iwama, G.K., 1991. Polyculture of the Pacific oyster, *Crassostrea gigas* (Thumberg), with chinook salmon, *Oncorhynchus tshawytscha*. Aquaculture 92, 313–322.

Kreeger, D.A., Langdon, C.J., Newell, R.I.E., 1988. Utilization of refractory cellulosic carbon derived from *Spartina alterniflora* by the ribbed mussel *Geukensia demissa*. Mar. Ecol.: Prog. Ser. 42, 171–179.

Lá Rosa, T., 1999. Comunit microbiche in aree interessate da impianti di maricoltura. PhD Dissertation, University of Messina, Italy, p. 168.

Langdon, C.J., Newell, R.J.E., 1990. Utilization of detritus and bacteria as food sources by two bivalve suspension-feeders, the oyster *Crassostrea virginica* and the mussel *Geukensia demissa*. Mar. Ecol.: Prog. Ser. 58, 229–310.

Léfebvre, S., Barill, L., Clerc, M., 2000. Pacific oyster (*Crassostrea gigas*) feeding responses to a fish-farm effluent. Aquaculture 187, 185–198.

Lopiano, L., 2000. Aspetti di bioenergetica della crescita di *Diplodus puntazzo* (Cetti, 1777 Pisces: Sparidae). PhD Dissertation, University of Palermo, Italy, p. 202.

Lorenzen, C., Jeffrey, J., 1980. Determination of chlorophyll in sea water. UNESCO Techn. Papers Mar. Sci. 35, 1–20.

Mann, K.H., 1988. Production and use of detritus in various freshwater, estuarine and coastal marine cosystems. Limnol. Oceanogr. 33, 910–930.

Margalef, R., 1985. Western Mediterranean. Pergamon, Oxford UK, p. 363.

Marsh, J.B., Weinstein, W.J., 1959. A simple charring method for determination of lipids. J. Lipid Res. 7, 574–576.

Mazzola, A., Favaloro, E., Sar, G., 1999a. Comparative growth of *Diplodus puntazzo* (Cetti, 1777) in different condition of cultivations (Pisces: Sparidae) (southern Tyrrhenian sea). Aquacult. Res. 30, 773–780.

Mazzola, A., Mirto, S., Danovaro, R., 1999b. Initial fish-farm impact on meiofaunal assemblages in coastal sediments of the Western Mediterranean. Mar. Pollut. Bull. 38, 1126–1133.

Mazzola, A., Sarà, G., Venezia, F., Caruso, M., Catalano, D., Hauser, S., 1999c. Origin and distribution of suspended organic matter as inferred from carbon isotope composition in a Mediterranean semi-enclosed marine system. Chem. Ecol. 16, 215–238.

Michener, R., Shell, D.M., 1994. Stable isotope ratios as tracers in marine aquatic food webs. In: Lajtha, K., Michener, R.H. (Eds.), Stable Isotopes in Ecology and Environmental Sciences. Blackwell, Oxford, UK, pp. 138–157.

Neori, A., Ragg, N.L.C., Shpigel, M., 1998. The integrated culture of seaweed, abalone, fish and clams in modular intensive land-based systems: performance and nitrogen partitioning within an abalone (*Haliotis tuberculata*) and macroalgae culture system. Aquacult. Eng. 17, 215–239.

Neori, A., Shpigel, M., Ben-Ezra, D., 2000. A sustainable integrated system for culture of fish, seaweed and abalone. Aquaculture 186, 279–291.

Nival, P., Charra, R., Malara, G., Boucher, D., 1972. La matière organique particulaire de la Méditerranée occidentale en mars 1970. Mission Mediprod du "Jean Charchot". Ann. Ist. Oceanogr. 48, 141–156.

Peterson, B.J., Fry, B., 1987. Stable isotopes in ecosystem studies. Annu. Rev. Ecol. Syst. 18, 293–320.

Peterson, B.J., Howarth, R.W., Garritt, R.H., 1985. Multiple stable isotope used to trace the flow of organic matter in estuarine food webs. Science 227, 1361–1363.

Riera, P., 1997. δ^{15}N of organic matter sources and benthic invertebrates along an estuarine gradient in Marennes-Olron Bay (France): implications for the study of trophic structure. Mar. Ecol.: Prog. Ser. 166, 143–150.

Riera, P., Richard, P., 1996. Isotopic determination of food sources of *Crassostrea gigas* along a trophic gradient in the estuarine bay of Marennes-Olron. Estuarine Coastal Shelf Sci. 42, 347–360.

Riera, P., Richard, P., 1997. Temporal variation of δ^{13}C in particulate organic matter and oyster *Crassostrea gigas* in Marennes-Olron Bay (France): effect of freshwater inflow. Mar. Ecol.: Prog. Ser. 147, 105–115.

Sarà, G., Mazzola, A., 1997. Effects of trophic and environmental conditions on the *Crassostrea gigas* growth in culture. Aquaculture 153, 81–91.

Sarà, G., Manganaro, A., Cortese, G., Pusceddu, A., Mazzola, A., 1998. The relationship between food availability and growth in *Mytilus galloprovincialis* in the open sea (southern Mediterranean). Aquaculture 167, 1–15.

Sarà, G., Leonardi, M., Mazzola, A., 1999. Spatial and temporal changes of suspended matter in relation to wind and vegetation cover in a Mediterranean shallow coastal environment. Chem. Ecol. 16, 151–173.

Sarà, G., Romano, C., Caruso, M., Mazzola, A., in press. The new Lessepsian entry Brachidontes pharaonis (Fischer, P., 1870) (Bivalvia, Mytilidae) in the western Mediterranean: a physiological analysis under varying natural conditions. J. Shellfish. Res.

Shpigel, M., Blaylock, R.A., 1991. The pacific oyster, *Crassostrea gigas*, as a biological filter for a marine fish aquaculture pond. Aquaculture 92, 187–197.

Shpigel, M., Neori, A., 1996. The integrated culture of seaweed, abalone, fish and clams in modular intensive land-based systems: I. Proportions of size and projected revenues. Aquacult. Eng. 15, 313–326.

Shpigel, M., Neori, A., Gordin, H., 1991. Oyster and clam production in the outflow water of marine fish pond in Israel. EAS Spec. Publ. 14, 295.

Shpigel, M., Lee, J., Soohoo, B., Friedman, R., Gordin, H., 1993a. The use of outflow water from fish ponds as a food source for Pacific oyster *Crassostrea gigas* Thumberg. Aquacult. Fish. Manage. 24, 529–543.

Shpigel, M., Neori, A., Popper, D.M., Gordin, H., 1993b. A proposed model for environmentally clean land-based culture of fish, bivalves and seaweeds. Aquaculture 117, 115–128.

Smaal, A.C., Haas, H.A., 1997. Seston dynamics and food availability on mussel and cockles beds. Estuarine Coastal Shelf Sci. 3, 290–309.

Stirling, A., Okumus, I., 1995. Growth and production of mussels (*Mytilus edulis* L.) suspended at salmon cages and shellfish farms in two Scottish sea lochs. Aquaculture 134, 193–210.

Underwood, A.J., 1997. Experiments in Ecology. Their Logical Design and Interpretation Using Analysis of Variance. Cambridge Univ. Press, UK, p. 504.

Valiela, I., 1984. Marine Ecological Processes. Springer-Verlag, New York, p. 546.

Ward, L.G., Kemp, W.M., Boynton, W.R., 1984. The influence of waves and seagrass communities on suspended particulate in a estuarine embayment. Mar. Geol. 59, 85–103.

Widdows, J., Fieth, P., Worrall, C.M., 1979. Relationship between seston, available food and feeding activity in the common mussel *Mytilus edulis*. Mar. Biol. 50, 195–207.

Wildish, D., Kristmanson, D., 1997. Benthic Suspension Feeders and Flow. Cambridge Univ. Press, UK, p. 409.

Ye, L., Ritz, D.A., Fenton, G.E., Lewis, M.E., 1991. Tracing the influence on sediments of organic waste from a salmonid farm using stable isotope analysis. J. Exp. Mar. Biol. Ecol. 145, 161–174.