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Effects of fish farming waste to sedimentary and particulate organic matter in a southern Mediterranean area (Gulf of Castellammare, Sicily): a multiple stable isotope study ($\delta^{13}\text{C}$ and $\delta^{15}\text{N}$)

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Abstract

Carbon and nitrogen stable isotope analysis was used to investigate the dispersion area of waste material coming from fish farming activities in the western Mediterranean. Tests were conducted to see if uneaten feed and faecal material isotopic signals, originating from fish farms, could be detected in particulate organic matter (POM) and sedimentary organic matter (SOM). The detectable dispersion distance (from under cages as far as 1000 m) of cage-derived organic material was also examined. To do this, carbon ($\delta^{13}\text{C}$) and nitrogen ($\delta^{15}\text{N}$) composition in POM and SOM collected around the cages, in some control areas and in the waste material, was measured. Mean POM $\delta^{13}\text{C}$ was $-22.9 \pm 0.2\text{‰}$, while SOM $\delta^{13}\text{C}$ was $-22.1 \pm 0.1\text{‰}$, and did not show significant differences along a distance gradient. Mean POM $\delta^{15}\text{N}$ was $3.9 \pm 1.0\text{‰}$, while SOM $\delta^{15}\text{N}$ was $3.4 \pm 1.3\text{‰}$, showing significant differences between ^{15}N -enriched sites positioned near the cages and ^{15}N -depleted sites positioned at about 1000 m from the cages. The mixing model applied to each reservoir (POM and SOM) as targets showed an incidence of autochthonous carbon (mostly phytoplankton in the particulate and sand microflora in the sediments) of about 24% in POM and of about 19% in SOM. Terrigenous carbon, which represented 37% in POM and 33% in SOM, increased in moving from sites nearby cages to more distant sites. Farming waste carbon represented 39% in POM and 48% in SOM. The inputs of autochthonous N represented about 24% in POM and about 18% in SOM, and with terrigenous N (representing 62% and 70%, respectively, in POM and SOM) showed higher contribution than cage-derived nitrogen. On average, farming waste nitrogen in POM was 15%, while it was 11% in SOM. $\delta^{15}\text{N}_{\text{POM}}$ showed a significant difference between cage sites and sites positioned at about 300 m. The latter was similar to sites located at 1000 m from the

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cages. Results indicate that in a Mediterranean oligotrophic area, with a bottom about 25 m deep and a mean current speed of $10\text{--}12\text{ cm s}^{-1}$, the influence of carbon and nitrogen from farming waste can be isotopically detected both in the particulate matter and the sediments in a wide area around fish farming cages. Sediments around the cages have been observed to be organic-enriched at about 1,000 m from cages. Dispersion of cage waste by hydrodynamic advection, consumption and defecation by wild fish, and resuspension from the bottom currents were invoked as three combined factors to explain the greater impact area found in this study than has been previously reported in the literature.

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1. Introduction

In the last decade, the increasing number of fish culture industries in Mediterranean areas has begun to create serious environmental problems due to the impact caused by fish farming waste. Many countries, especially Italy and Greece, have designated several areas for fish farms without adequate policy limitations. In particular, the Sicilian fish culture industry is becoming very important, with the highest production of the European sea bass (*Dicentrarchus labrax*) and the gilt-head sea bream (*Sparus aurata*) in the western Mediterranean (FAO, 2002).

Very little information has been published on farming waste impact in the Mediterranean. Many cases, however, have been observed but not yet documented, inducing a number of researchers to address this problem. The output from aquaculture operations is primarily composed of suspended solids, originating from uneaten feed and faecal material (Holmer, 1991; Iwama, 1991), which are considered to highly impact and damage the natural environment (Troell and Norberg, 1998; Naylor et al., 2000). Many papers have shown how fish farm biodeposition products can negatively affect sediment chemistry, and community dynamics of marine seagrasses, benthic macrofauna, meiofauna, and benthic bacteria (Holmer, 1991; Findlay and Watling, 1997; Hargrave et al., 1997; Pergent et al., 1999; Pearson and Black, 2000; La Rosa et al., 2001; Mirto et al., 2002). Moreover, many papers have suggested the occurrence of a spatial gradient of waste dispersion, able to degrade the natural environment within a maximum radius of about 100 m from the cages (Holmer, 1991; Pearson and Black, 2000). Such a dispersion distance is a function of local current speed, water depth, and total output from the farms. This last factor appears to be important in determining the real impact on the adjacent environment (Iwama, 1991). Biochemical tracers, such as stable isotopes and, in particular, the carbon isotope, have rarely been used (Hansen et al., 1991; Ye et al., 1991; McGhie et al., 2000; Sutherland et al., 2001). Nevertheless, according to Hobson (1999), the isotopic segregation ability of biochemical tracers can be a powerful tool, above all, in tracing organic matter in several fields of ecological research, and other authors have recommended their use in pollution studies (Macko and Ostrom, 1994). In the last two decades, carbon and nitrogen isotopes have been used extensively to trace different types of matter: marine vs. freshwater (Fry and Sherr, 1984), aquatic vs. terrestrial (Thornton and McManus, 1994), and polluted vs.

unpolluted (Kidd et al., 1999; Waldron et al., 2001; Burford et al., 2002). Rarely, however, have these isotopes been used to trace fish farm organic vs. natural matter, even though aquaculture represents a fundamental industry that exploits all aquatic environments and is one of first modalities of food supply for worldwide coastal populations.

This is the one among a few studies to report on this topic. We hypothesised that uneaten feed and faecal material isotopic signals, which originate from fish farms, could be detected in particulate organic matter (POM) and sedimentary organic matter (SOM). We also tested the detectable range (the so-called “dispersion distance” of waste from their emission focus) of cage-derived organic material.

2. Materials and methods

2.1. Study area and sample collection

The study was carried out between May and June 2001 off the northern coast of Sicily, in the Gulf of Castellammare (latitude 38° 02 31 N; longitude 12° 55 28 E), an area well studied for its fish and shellfish farming potentiality (Sarà and Mazzola, 1997; Sarà et al., 1998; Mazzola et al., 1999). The area is seasonally constrained by terrigenous–continental inputs, particularly abundant in spring and early summer (Sarà et al., 1998), which originate from nearby streams.

In late spring 2000, four submersible cages (Floatex, Italy; volume = 1000 m⁻³) were positioned in the western part of the Gulf (latitude 38° 02 60 N; longitude 12° 55 60 E) and moored on the bottom at a depth of about 30 m, 2 km off the coast. In that area, sediments are generally unvegetated with low-frequency patches of the Mediterranean seagrass, *Cymodocea nodosa*. The hydrodynamic regime of the cage area (Table 1) is characterised by a dominant current, with an average speed of 12 ± 7.5 cm s⁻¹, coming from the third and fourth quadrants (along a west–east axis) for most of the year (CEOM, 2002).

The cages were filled in July 2000 with 53,550 specimens of *D. labrax* (initial total length of 160.9 ± 17.0 mm and total weight of 50.6 ± 16.3 g) and 69,500 specimens of *S. aurata* (initial total length of 129.9 ± 15.9 mm and total weight of 33.6 ± 12.9 g). The total initial transplanted biomass was 5,047 kg over four cages. Farming was carried out until summer 2001, when the total biomass reached was about 33,800 kg. Throughout the farming period, the average food conversion ratio was estimated to be about 2.5 (CEOM, 2002). The total supplied feed (different types of commercial feed produced by BioMar, France and Hendrix, Italy) was about 80 tons.

Table 1

Frequency of current velocities (%) between 5 cm s⁻¹ and equal to or higher than 20 cm s⁻¹ measured in the study area (average current velocity = 12.0 ± 7.5 cm s⁻¹)

	5 cm s ⁻¹	10 cm s ⁻¹	15 cm s ⁻¹	>20 cm s ⁻¹
Subsurface (– 5 m)	23.0	44.0	25.0	8.0
Mean depth (– 15 m)	22.3	27.4	23.2	27.1
Bottom (– 30 m)	20.0	24.0	23.0	33.0

Sample collection was carried out in late spring 2001 (May 3 and June 21, 2001) over six sites positioned downstream at different distances from the cages: two sites within a radius of 50 m (hereafter referred to as under cages), two sites within a radius of 300 m, and the other two at about 1,000 m from the edge of the cage system. To collect data with the highest rate of organic waste deposition from the cages, all sampling sites were located east of the cages, downstream, along the west–east main axis of the water current.

Sampling was carried out in late spring–early summer, based on the assumption that primary production (phytoplankton and microbenthic algae) and terrigenous–continental isotopic signals are at their maximum magnitude during this period, at Mediterranean latitudes (Margalef, 1985). Thus, if a significant dominance of the farming organic waste signal is detected in the particulate and sedimentary organic matter isotopic composition, such dominance might remain constant, or be amplified, in the other seasons when primary production and terrigenous–continental inputs are lower. Moreover, spatial variability is considered more evident and important than temporal variability in this type of system (Smith, 1996; sensu Hargreaves, 1998).

To compare results, we chose published data (Sarà et al., 2002) on an external control site positioned off the Egadi Islands in the western Mediterranean, at a depth of 15 m. At this site, there is no farm waste influence, and the authors observed that main isotopic signals were due to primary production and terrigenous–continental inputs, which originate from a nearby stream. $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ mean spring–summer values in the POM were $-19.9 \pm 1.9\text{‰}$ and $3.9 \pm 1.7\text{‰}$, respectively, while in the SOM, they were $-16.0 \pm 1.7\text{‰}$ and $2.3 \pm 0.9\text{‰}$, respectively, for carbon and nitrogen.

Two water samples were collected, each time at both sites, at a depth of about 12 m using a 5-l Niskin bottle (General Oceanic, USA), and two replicates of hand cores were taken from two quadrates (20 cm length, 400 cm² surface area) by SCUBA divers.

Water and sediment samples were brought back to the laboratory and processed within a few hours. Seawater (4 l) was filtered through precombusted fibreglass filters (Whatman GF/F) (450 °C, 4 h), while the top 0–1.5 cm layer of each core was immediately frozen at -20 °C and stored until analysed.

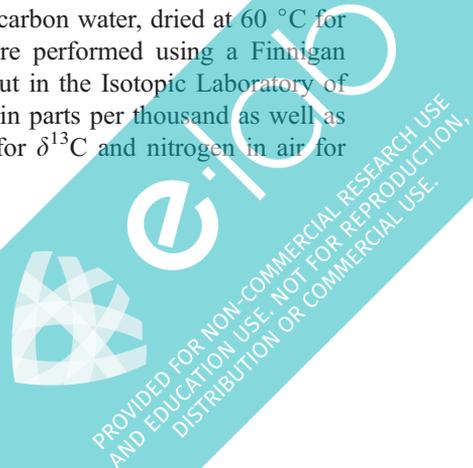
At the same time, about 10 specimens of each farmed fish species were sampled by means of a net and after an overnight evacuation, their faeces were collected using pipettes.

Water (POM) and sediments (SOM), as well as fish faeces, were analysed for each species to measure their carbon and nitrogen isotope ratios.

All samples were acidified in 2 N HCl, rinsed in free-carbon water, dried at 60 °C for at least 24 h, and grounded. The isotopic analyses were performed using a Finnigan Delta-Plus isotope ratio mass spectrometer and carried out in the Isotopic Laboratory of Geokarst (Trieste, Italy). Isotopic values were expressed in parts per thousand as well as deviations from standards (Peedee belemnite limestone for $\delta^{13}\text{C}$ and nitrogen in air for $\delta^{15}\text{N}$):

$$\delta^{13}\text{C} \text{ or } \delta^{15}\text{N} = [(R_{\text{sample}}/R_{\text{standard}}) - 1] \times 10^3,$$

$$\text{where } R = {}^{13}\text{C}/{}^{12}\text{C} \text{ or } {}^{15}\text{N}/{}^{14}\text{N}$$



2.2. Statistical analyses and elaboration

A three-way analysis of variance (ANOVA) was used (mixed design; Underwood, 1997) to test the hypothesis that carbon and nitrogen isotope compositions of POM and SOM organic matter varied as a function of the distance gradient from the focus of the cages. One factor was treated as fixed and orthogonal: distance from the focus of the cages (under cage, about 300 m and about 1000 m from the focus = three levels; DIST). Sites (two for each distance) were treated as random (two levels; SITE) and nested in DIST, while sampling dates (May 3 and June 21 = two levels; TIME) were also treated as a random factor and nested in SITE. Two replicates were randomly collected at each site. Heterogeneity of variances was tested using Cochran's test prior to the ANOVA and the appropriate means were compared using Student–Newman–Keuls (SNK) tests (Underwood, 1997).

Seasonal mixing equations (Phillips and Gregg, 2003) were also performed with POM and SOM as targets. Reference signatures in the model were for POM autochthonous (mostly phytoplankton, Martinotti et al., 1997): $\delta^{13}\text{C} = -19.0\text{‰}$ and $\delta^{15}\text{N} = 6.5\text{‰}$; for SOM autochthonous (mostly sand microflora; Martinotti et al., 1997): $\delta^{13}\text{C} = -15.0\text{‰}$ and $\delta^{15}\text{N} = 6.5\text{‰}$; for terrigenous–continental (Martinotti et al., 1997; Riera, 1997): $\delta^{13}\text{C} = -26.4\text{‰}$ and $\delta^{15}\text{N} = 1.6\text{‰}$, and the isotopic mean values measured in the present study for cage waste. The GMAV (1997) statistical package (University of Sydney, Australia) was used to perform ANOVA, while IsoSource software (Phillips and Gregg, 2003) was used to run mixing equations.

3. Results

Carbon and nitrogen isotopic values from organic waste sources are summarised in Table 2. Pellet feed had a carbon isotopic average value of $-22.8 \pm 1.0\text{‰}$, while nitrogen averaged at $8.3 \pm 1.9\text{‰}$. Farmed fish faeces had an average of $-21.0 \pm 0.6\text{‰}$ and $10.7 \pm 0.9\text{‰}$, respectively, for $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$. Throughout the sampling period, POM carbon isotopic composition was, on average, $-22.9 \pm 0.22\text{‰}$, ranging between -21.9‰ and -24.0‰ , while SOM was, on average, $-22.1 \pm 0.11\text{‰}$, ranging between -23.5‰ and -21.6‰ (Table 3). POM and SOM $\delta^{13}\text{C}$ values did not show

Table 2

Carbon and nitrogen isotopic signatures of the main organic matter sources measured throughout the sampling periods

Organic matrix	<i>n</i>	$\delta^{13}\text{C}$	\pm S.E.	$\delta^{15}\text{N}$	\pm S.E.
Feed 1	8	-23.9	0.1	6.5	0.3
Feed 2	10	-22.6	0.2	7.8	0.1
Feed 3	9	-21.7	0.2	10.7	0.3
Ejection (DI)	6	-21.1	0.9	10.9	1.2
Ejection (Sau)	6	-20.9	0.2	10.4	0.8

n = Number of observations; $\delta^{13}\text{C}$, ‰ = carbon isotopic values; $\delta^{15}\text{N}$, ‰ = nitrogen isotopic values; \pm S.E. = standard errors for means; DI = *D. labrax*; Sau = *S. aurata*.

Table 3
 Statistics of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ measured in the present study as a function of a spatial gradient downstream

Distance	Water column (POM)		Sediment (SOM)	
	Mean	\pm S.E.	Mean	\pm S.E.
$\delta^{13}\text{C}$				
Under cage	-22.7	0.3	-22.1	0.0
300 m	-23.0	0.2	-22.0	0.0
1000 m	-23.1	0.1	-22.2	0.2
Study area	-22.9	0.2	-22.1	0.1
$\delta^{15}\text{N}$				
Under cage	5.1	0.4	4.9	0.1
300 m	3.2	0.1	3.1	0.0
1000 m	3.6	0.1	2.2	0.2
Study area	3.9	1.0	3.4	1.4

Under cage = within 50 m from the edge of cages; 300 m = sites at about 300 m from the edge of cages; 1000 m = sites at about 1,000 m from the edge of cages; POM = particulate organic matter; SOM = sedimentary organic matter; \pm S.E. = standard errors of the mean.

significant differences (ANOVA, $p > 0.05$; Table 4) along a distance gradient. In the study area, the mean value of POM nitrogen isotopic composition was 3.9 ± 1.01 ‰ (Table 3), ranging between 3.0‰ and 7.4‰. $\delta^{15}\text{N}$ values showed significant differences (ANOVA,

Table 4
 Analysis of variance on $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ of POM and SOM performed to verify possible differences between increasing distances from cage emission focus

Source of variation	df	Water column (POM)			Sediments (SOM)		
		MS	F	P	MS	F	P
$\delta^{13}\text{C}$							
Distance = DIST	2	0.37	6.40	NS	0.08	1.85	NS
Site = Si	1	0.00	0.00	NS	0.02	0.18	NS
Time (Si)	2	0.17	0.45	NS	0.10	1.18	NS
DIST \times Si	2	0.06	0.09	NS	0.04	0.27	NS
DIST \times Time (Si)	4	0.67	1.80	NS	0.17	1.93	NS
Residual	12	0.37			0.09		
Cochran's C				NS			NS ^a
$\delta^{15}\text{N}$							
Distance = DIST	2	8.14	1028.26	***	15.01	123.34	**
Site = Si	1	0.00	0.00	NS	0.11	1.44	NS
Time (Si)	2	0.60	0.95	NS	0.07	0.54	NS
DIST \times Si	2	0.01	0.01	NS	0.12	0.91	NS
DIST \times Time (Si)	4	0.61	0.97	NS	0.13	0.98	NS
Residual	12	0.63			0.14		
Cochran's C				NS			NSa

* $P \leq 0.05$.

NS = not significant difference ($P > 0.05$).

^a Cochran's test significant and data transformed to natural logarithms.

** $P \leq 0.01$.

*** $P \leq 0.001$.



$p < 0.05$; Table 4) between ^{15}N -enriched sites, positioned near the cages (under cages; Table 3), and ^{15}N -depleted sites, positioned at more of 1000 m from the cages. SOM $\delta^{15}\text{N}$ values ranged between 1.6 ‰ and 5.3 ‰ (average 3.4 ± 1.3 ‰). In some cases, enriched values were recorded near the cages (4.9 ± 0.12 ‰), while more depleted values were found far away from the cages (2.2 ± 0.17 ‰).

The single-isotope three-source mixing model, applied to each reservoir (POM and SOM) as target, allowed us to investigate the percentage contribution of carbon and nitrogen coming from terrigenous, farming waste (feed + ejection), and autochthonous inputs. The incidence of terrigenous carbon was, on average, 37.4% in POM (upper and lower 95% confidence interval = 21% and 54%, respectively) and 33.3% in SOM (upper and lower 95% confidence interval = 3% and 63%, respectively), and generally increased, moving from sites nearby cages to more distant sites (Fig. 1). Carbon from autochthonous inputs (mostly phytoplankton in POM and mostly sand microflora in SOM) contributed to about 24% in POM (upper and lower 95% confidence interval = 0% and 48%, respectively) and about 19% in SOM (upper and lower 95% confidence interval = 0% and 38%, respectively). Apart from an insignificant peak at 300-m sites (29% and 19% in POM and SOM, respectively), autotrophic inputs generally decreased, moving away from the cages (Fig. 1). Carbon originating from farming facilities represented 38.9% in the POM (upper and lower 95% confidence interval = 0% and 79%, respectively) and 47.9% for SOM (upper and lower 95% confidence interval = 0% and 97%, respectively). Also regarding

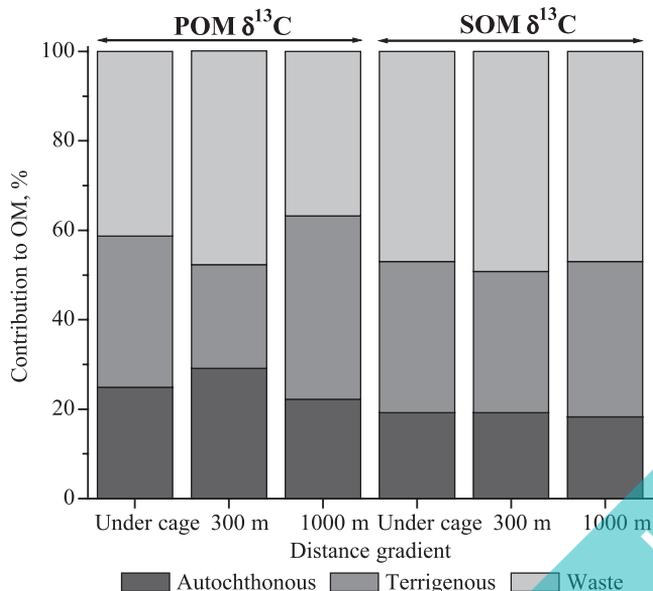


Fig. 1. Percentage incidence (%) of each main source (autochthonous, terrigenous, and farming wastes) able to constrain the carbon isotopic signatures of the POM and SOM in the study area as a function of increasing distance from the cage emission focus estimated using the mixing isotope model (see the text for literature reference and details).

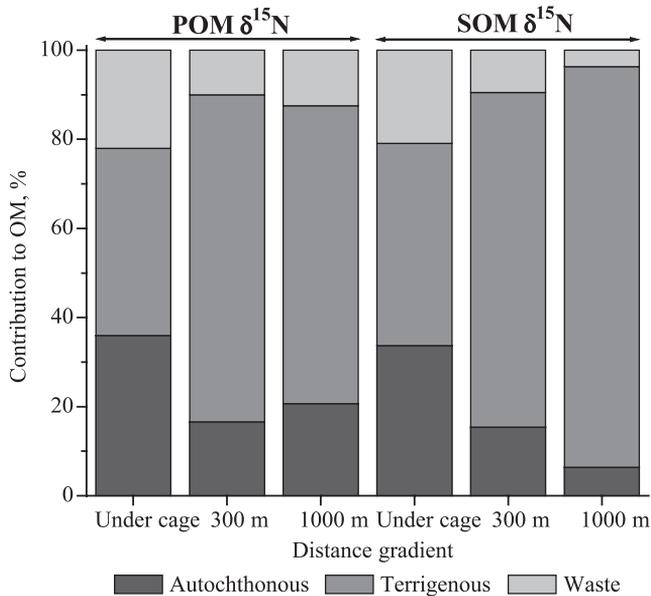


Fig. 2. Percentage incidence (%) of each main source (autochthonous, terrigenous, and farming wastes) able to constrain the nitrogen isotopic signatures of the POM and SOM in the study area as a function of increasing distance from the cage emission focus estimated using the mixing isotope model (see the text for literature reference and details).

farming waste input, a peak was quite evident around the 300-m sites and remained constant, above all in the SOM, until the 1000-m sites.

The input of terrigenous N was higher than farming waste nitrogen, representing 60% in POM (upper and lower 95% confidence interval = 52% and 72%, respectively) and 70% in SOM (upper and lower 95% confidence interval = 62% and 80%, respectively). This input increased (Fig. 2), moving from nearby cages to the more distant sites. Autochthonous nitrogen represented about 23.5% in the POM (upper and lower 95% confidence interval = 2% and 46%, respectively) and about 18.4% in the SOM (upper and lower 95% confidence interval = 0% and 37%, respectively), showing (Fig. 2), more remarkably in the SOM, a decreasing contribution gradient moving toward the farthest sites. Nitrogen from the cages contributed 14.5% in the POM (upper and lower 95% confidence interval = 0% and 30%, respectively) and 11.4% in the SOM (upper and lower 95% confidence interval = 0% and 24%, respectively), and it was higher in the sites around the cages (Fig. 2), representing 22% in the POM and about 21% in the SOM. In the sites furthest from the cages, nitrogen represented not more than 12% in POM and only 3% in SOM.

4. Discussion

Stable carbon and nitrogen isotopes as organic matter tracers emerge as extremely powerful and relatively simple tools to investigate the origin of fish farm-derived organic

matter in coastal marine systems (Hansen et al., 1991). In the portion of the Gulf of Castellammare affected by fish farming waste, $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ of particulate and sedimentary organic matter have adequately described the distribution pattern of organic enrichment as a function of a distance gradient of fish farming organic loading of the sediment.

To our knowledge, there are only a few studies in the current literature (Hansen et al., 1991; Ye et al., 1991; Sutherland et al., 2001) that used stable isotopes to detect the impact of fish farming waste on the adjacent environment.

The present study indicates that in a Mediterranean oligotrophic area, with a bottom about 25 m deep and a mean current speed of $10\text{--}12\text{ cm s}^{-1}$, the influence of carbon and nitrogen from farm waste could be detected in both the particulate and the sediments in a wide area around the fish cages. Sediments around the cages have been observed to be organic-enriched, and they represented a better “memory of disturbance” of system processes than the water column. This confirms what was stated by Ye et al. (1991), as well as what was reported in other studies carried out using other descriptors (Pearson and Black, 2000).

The POM collected in the area around the farms was constrained by waste carbon (for about 40%) with cage-derived C remarkably affecting the water column. By contrast, terrigenous–continental-derived and autochthonous N obscured the cage-derived N because, in the POM, they represented over than 80% of the total nitrogen. A similar pattern was found for the SOM, as farming waste carbon represented almost 50% of the total carbon, while only about 11% of the total nitrogen. In this case, a nitrogen gradient was also detected, moving away from the cages toward the more distant sites. Indeed, along this gradient, the influence on the water column of the nitrogen waste isotopic signal decreased both in the POM and SOM as a function of the distance.

Accordingly, apart from a few observations stating that there is no link between fish farm loading and benthic impact (Ritz et al., 1989), there is much evidence that farm loading affects the surrounding environment through farm emissions. Such observations have been obtained from the analysis of benthic macrofauna, since it has been observed that as the distance from the cages increases, pollutant-tolerant species appear, often in very high numbers and developing a peak in total fauna biomass (Holmer, 1991). At the same time, other papers have demonstrated that fish farm biodeposition products affect water column and sediment chemistry, seagrass, meiofauna, and benthic bacteria, by comparing impacted and unaffected areas (Findlay and Watling, 1997; Hargrave et al., 1997; Pergent et al., 1999; Pearson and Black, 2000; La Rosa et al., 2001; Mirto et al., 2002; Alongi et al., 2003).

Overall, a pattern of influence caused by farm waste has been described in the literature (Brown et al., 1987; Gowen and Bradbury, 1987; Holmer, 1991; Ye et al., 1991), but there is, however, no clear, well-tested evidence of the influence from cage waste over 100 m from the focus of cage emissions (Pearson and Black, 2000; Molina Domínguez et al., 2001), apart from rare examples (Martinotti et al., 1997; Pergent et al., 1999; Cromey et al., 2002). Thus, results from the present study partially counteract this general view, even though we are aware that the comparison of different hydrodynamic systems might not be valid (Silvert and Cromey, 2000). Results presented

here indicate that farming C-derived material may be isotopically detected in an area of many hectares around the cages, along the axis of main water currents and up to several hundreds of meters from the cages. Such a dispersion area is greater than that suggested (only 1.5–2 times the area of the farm itself) by the still-influential paper by Gowen and Bradbury (1987).

In our study area, the carbon signal almost obscured the signal originating from terrigenous and autochthonous inputs, suggesting that consumption and degradation rates are lower than the accumulation rate of organic material. This might be so, even if organic carbon dispersed from the farms is usually composed of highly labile compounds (Pearson and Black, 2000) and hence, it might be rapidly taken up by local consumers (nectobenthic and benthic organisms). We further validate the hypothesis of a constraining role of cage-derived carbon in the farming area by comparing our data with isotopic composition of a similar offshore external control area (Table 5), which is certainly not impacted by C-derived farming organic material.

Nitrogen did not show a dispersion pattern around the cages similar to that of carbon. Nitrogen isotopic signatures were overlapped by terrigenous signatures within a few hundred meters from the focus of the cage emissions, which produced results similar to those reported in literature (see Pearson and Black, 2000 for review) confirming, *a latere*, results obtained worldwide about the nitrogen dynamics in farm-impacted systems (Kaspar et al., 1985; Holmer, 1991; Iwama, 1991; Hargrave et al., 1997).

Cage-derived organic N contributed both to about 20% in the POM and SOM nitrogen isotopic composition, at sites close to the cages (within 300 m). But at sites located further than 300 m from the cages, cage-derived organic N decreased significantly to less than 10% on the average (3.7% in SOM). Thus, it seems that nitrogen is consumed faster than carbon, and that it also has a faster degradation turnover in the water column than in the sediments (Mann, 1988).

The present findings are important to explain the different behaviours between C and N and the greater spatial persistence of cage-derived C. We can consider a “quantitative hypothesis,” which is based on the assumption that there will be more supplied feed dispersed in the environment (as uneaten food), at the current speed (with peak also of 25 cm s⁻¹) found in the study area, than that dispersed in other farms with lower current speed. The total food conversion ratio for *D. labrax* and *S. aurata*, estimated in other areas of the Mediterranean, is about 1.5–1.6 (Mazzola et al., 1999; Molina Domínguez et al., 2001) and is about the same for salmon in Nordic Sea (Enell, 1995). In contrast, in the

Table 5

Percentage incidence of each main carbon and nitrogen source (terrigenous and autochthonous) able to constrain the carbon and nitrogen isotopic signatures of the POM and SOM in a Mediterranean control area estimated using the mixing isotope model (see the text for literature reference and details)

	$\delta^{13}\text{C}$ POM	$\delta^{13}\text{C}$ SOM	$\delta^{15}\text{N}$ POM	$\delta^{15}\text{N}$ SOM
Reservoir	-21.8	-16.0	3.9	2.3
% Terrigenous (A)	91.0	17.0	62.0	94.0
% Autochthonous (B)	9.0	83.0	38.0	6.0
Range % A	72–100	76–91	44–80	76–100
Range % B	0–28	9–24	20–56	0–24

studied farm, the estimated food conversion ratio was about 2.5 (CEOM, 2002). That higher value implied a larger loss of supplied feed during feeding operations and this could explain the dominance of the carbon signal in the POM and SOM in comparison to terrigenous background isotopic signals.

However, we found a discrepancy between isotopic findings coming from the present study and hydrodynamic calculations of waste dispersion reported in the recent literature (Cromeey et al., 1999, 2002). Uneaten feed is likely to have a settling velocity within a range of 2–14 cm s⁻¹, depending on the pellet diameter and type. Although there are few data on Mediterranean sea bass and sea bream settling food and faecal waste in the current literature, there are plenty of data on salmonids and the abovementioned range could be useful (C. Cromeey, personal communication). Thus, according to Cromeey et al. (1999), for example, an uneaten feed particle settling at a speed of 2 cm s⁻¹ to a sampling depth of 12 m would be advected to a distance of not more than 150–200 m from the cages with a current speed of 25 cm s⁻¹. In order to explain the above discrepancy (between the present isotope findings highlighting an influence of cage wastes at more than 300 m from the cages and beyond, and literature calculations highlighting the influence of cage wastes at not more than 100–150 m) and the greater spatial persistence of C-signal with respect the N-signal, we suggest two hypotheses.

Firstly, we hypothesise a role played by wild and escaped fish outside the cages, attracted in large densities around farms (functioning as attracting devices), which consume the waste particles dispersed in the environment around the cage, depositing them elsewhere by defecation. Thus, food pellets being eaten around the cages would be deposited at a distance *via* wild fish faeces (i.e., a wild fish mechanism delaying and enlarging the deposition of cage-derived material). Fish around the cages eat waste food pellets along the water column, which is likely to indirectly slow down the settling speed of waste particles. Thus, settling particles are N-impoverished by wild fish assimilation and C-enriched by fish faecal loss. In addition, the presence of this “pelagic consumer barrier” (*sensu* Pearson and Black, 2000 and *sensu* Fig. 3) around the cages should increase the permanency time of waste particles in the water column, allowing water currents to move these impoverished particles far from the cages. These particles are significantly signed by cage-derived carbon but poor in cage-derived nitrogen.

Moreover, the above situation allows us to state that the main components of cage-derived waste (uneaten and undigested food plus faecal material), apart from respective lability degree and settling properties, are impoverished of their nitrogen compounds by consumers (firstly fish, invertebrates, and bacteria), with a time lag lower than carbon. Such a process leads to an impoverishment of N, increasing the C:N ratio values of waste material. As a main consequence, we found organic material with isotopic composition significantly signed by cage-derived carbon in the sites more distant from the cages, thus explaining the lower persistence of cage-derived N in the surrounding of farms and its spatial gradient. As other authors have demonstrated, the biota (fish, macroalgae, and benthic invertebrates) can play a very important role in the moderation of hydrodynamics by enhancing the process of sedimentation and altering stability/erodibility of the sediments (Widdows et al., 1998).

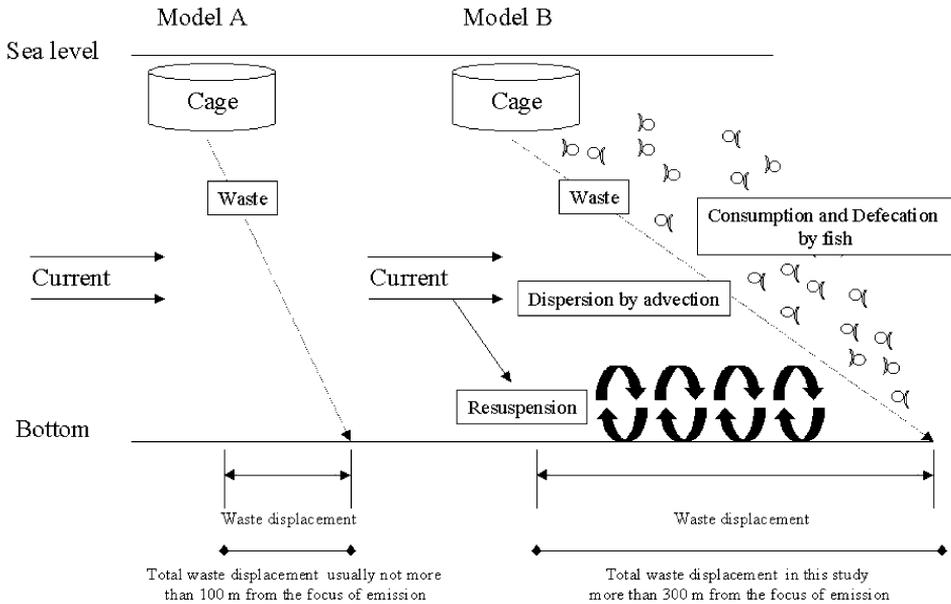


Fig. 3. Representative diagram of the hypothesised role of fish and resuspension from sediments in the amplification of waste material dispersion coming from cages. Model A = model according to [Silvert and Cromey \(2000\)](#) without the fish role and resuspension. Model B = the [Silvert and Cromey \(2000\)](#) modified model showing the presence of fish and further resuspension movements enlarging the displacement of waste material (diagrams not to scale).

We can combine this hypothesis with the recently hypothesised role ([Cromey et al., 2002](#)) played by resuspension movements from the bottom regarding the dispersion around the cages of waste material. The resuspension effect can act as prime factor in dispersing the waste material in a greater area. Previous papers dealing with the study area have also pointed out that resuspension can affect the feeding behaviour and the performance of cultivated mussels ([Sarà et al., 1998](#); see also current data in [Table 1](#)).

In conclusion, we hypothesise that ([Fig. 3](#)): (a) organic waste material coming from the cages is advected to a certain distance depending on its settling properties and local current intensity; (b) during the settling along the water column, it is consumed by wild organisms around cages, indirectly slowing down its settling speed; (c) waste being eaten around the cages is deposited at a distance via wild fish faeces; and (d) upon reaching the bottom (with an altered composition due to consumption by wild organisms, N-impoverished particles by predation, and C-enriched by fish faecal loss), resuspension movements, due to bottom currents, further enlarge the dispersion area of the waste. The result of these combined effects will presumably be the greater area of dispersion we found by using stable isotopes than what has been previously reported by other authors.

Nevertheless, further studies are needed to test both the role of wild fish around the cages and resuspension movements as amplifying factors of the dispersion of farm-derived waste materials.

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