Sediment biogeochemical differences in two pristine Mediterranean coastal lagoons (in Italy) characterized by different phanerogam dominance—A comparative approach

A. SIGNORINI*, G. MASSINI, G. MIGLIORE, M. TOSONI, C. VARRONE and G. IZZO
ENEA Cre Casaccia BIOTEC, Via Anguillarese 301, 00123 Rome, Italy

ABSTRACT

1. The aim of this study was to investigate functional differences in two Italian coastal lagoons (Caprolace and Fogliano, Tyrrhenian Sea) characterized by the dominance of two different seagrass species: *Cymodocea nodosa* (Ucria) Ascherson in the Caprolace lagoon and *Ruppia cirrhosa* (Petagna) Grande in the Fogliano lagoon.

2. A monitoring system was set up in both lagoons in order to (i) map the distribution of macrophytes, (ii) measure spatio–temporal changes in the biological parameters of the seagrasses, and (iii) measure spatio–temporal changes in biological and chemical–physical parameters in the water column and sediments.

3. The results showed that the relationships between macrophytes and aquatic and sediment compartments entail functional differences described by buffering systems that counteract the accumulation of free sulphide.

4. This study also suggests that *R. cirrhosa* and *C. nodosa* are indicators of different levels of ecological stability.

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KEY WORDS: coastal lagoon; *R. cirrhosa; C. nodosa*; dominant seagrasses; biogeochemical parameters

INTRODUCTION

Transitional ecosystems, such as lagoons and coastal lakes, are highly vulnerable to eutrophication, as a consequence of their position between land and sea, their shallowness and their relatively limited exchange with the open sea. These ecosystems are characterized by alternative stable ecological states (dynamic regimes) with structural and functional changes on short-, medium- and long-term scales. In particular, displacement of long life-cycle species such as phanerogams by species with short life cycles, such as Chlorophyceae, is a process that affects coastal areas and has been related to increasing eutrophication (Duarte, 1995; Nixon, 1995; Cloern, 2001). These shifts typically occur in a sudden and drastic way, and early-warning signals are difficult to obtain (Scheffer et al., 2001).

In shallow water ecosystems, the mineralization of organic matter (OM) occurs in sediments and at the water–sediment
interface. The quantity and quality of the organic matter supplied to the sediment control benthic nutrient regeneration and metabolism (Giordani et al., 1996; Gray et al., 2002). When large amounts of labile organic matter sink to the bottom, the sediments become anaerobic a few millimetres below the sediment surface. As the metabolism shifts from aerobic to anaerobic, bacterial sulphate reduction (BSR) becomes the dominant carbon oxidation pathway (Jørgensen, 1982).

One of the major causes of seagrass decline is the susceptibility of their roots to highly reducing sediment conditions, and particularly to the toxicity of free sulphide, a phytotoxin produced by sulphate-reducing bacteria (Bartoli et al., 1996; Gray et al., 2002; Goodman et al., 1994; Holmer and Bondgaard, 2001). Hemminga, 1998; Terrados et al., 1996; Heijs et al., 2000; Koch and Erskine, 2001; Borum et al., 2005).

Nevertheless, two mechanisms in the rhizosphere sediments counteract the accumulation of free sulphide. First, seagrasses transport oxygen from above (water column) via a lacunae system down to the below-ground tissues (sediment), thereby sustaining aerobic respiration in these organs. Part of this oxygen may be lost from the plant roots by diffusion to the sediment, where it can support both biotic and abiotic reoxidation of reduced compounds; this phenomenon is known as radial oxygen loss (ROL). Under anoxic conditions, the metabolism of below-ground tissues must rely on fermentation processes that may excrete toxic end-products such as ethanol (Smith et al., 1988). This alcohol, being a labile carbon source for sulphate-reducing bacteria, speeds up sulphide production (Izzo and Hull, 1990). Since sulphide has a negative impact on photosynthesis, aerobic metabolism, growth and nutrient uptake in rooted plants, it contributes to ‘self-accelerating eutrophication processes’ (Duarte, 1995; Goodman et al., 1995; Holmer and Bondgaard, 2001).

Second, the reaction with sedimentary iron constitutes a potential buffer mechanism against sulphide toxicity (Heijs and Gemerden, 2000; de Wit et al., 2001; Kanaya and Kikuchi, 2004; Azzoni et al., 2005). Ferrous and ferric iron are present in the sediment in both amorphous and crystalline form (Lovley and Phillips, 1987). The amorphous or poorly crystalline (labile) iron species were assumed to be readily available for fast reactions with sulphide, thus accounting for the actual sulphide-buffering capacity (Azzoni et al., 2005). In this pool, ferric iron consists mainly of Fe(III)-oxyhydroxides, while ferrous iron consists largely of AVS (acid volatile sulphide). Thus the saturation of the iron buffer can be estimated as a function of total labile iron (LFe) and the AVS to LFe ratio (Giordani et al., 1996; Azzoni et al., 2005). With AVS/LFe > 1, the sulphide produced by BSR activity is expected to be released.

The iron and sulphide interactions, favouring phosphate release to pore water and to the water column, contribute to ‘self-accelerating eutrophication processes’ (Bartoli et al., 1996; Azzoni et al., 2001).

The aim of this study was to investigate the relations between dominant vegetation, biogeochemical characteristics and buffer mechanisms in two Italian coastal lagoons, both located in the Circeo National Park and characterized by the dominance of two different seagrass species: Ruppia cirrhosa (Petagna) Grande in the Fogliano lagoon and Cymodocea nodosa (Ucria) in the Caprolace lagoon. Different mechanisms seem to be at work in the two lagoons to counteract the accumulation of free sulphide. In fact, in the last 20 years dystrophic crises have occurred in the Fogliano lagoon during warmer summers, but never in Caprolace.

**STUDY AREA**

The shallow Caprolace and Fogliano coastal lagoons are located on the Tyrrhenian coast in central Italy, about 100 km south of Rome (Figure 1). Since 1975 they have been part of the Circeo National Park, and in 1978, they were declared ‘Wetlands of International Importance’ according to the Ramsar Convention.

Caprolace covers an area of approximately 226 ha and has a mean depth of 1.3 m (max. 2.9 m). It is connected to the sea through the San Nicolò inlet and is influenced by a microtidal regime in which the maximum tidal amplitude is 21 cm. The theoretical water turnover time is about 90 days.

Fogliano is located a few kilometres north of Caprolace. It covers an area of about 408 ha, has a mean depth of 0.89 m (max. 2 m) and is connected to the sea through the Foce del Duca inlet. Its maximum tidal amplitude is 23 cm and the theoretical water turnover time is about 60 days.

In the early 1980s, because of the high nutrient loadings, all freshwater inputs to the two lagoons were closed resulting in hypersaline conditions nearly all year round.

Five (1–5) sampling stations were selected in the Caprolace lagoon, and four (6–9) in Fogliano lagoon, based on the dominant submerged vegetation at these locations as determined from mapping done in 2001 (unpublished data) (Figure 1; Table 1).

**MATERIALS AND METHODS**

**Macrophytobenthos sampling**

The distribution maps of the benthic vegetation were obtained through direct field observations, using an optical viewer. The data were recorded with a GPS (Global Positioning System) and displayed with a GIS (Global Information System).
Before the surveys, a number of inspections were made to evaluate difficulties and operating conditions. The map of the lagoon, obtained via aerial imagery, was divided into optimum square 10 × 10 m sectors by Arc View (ESRI). The central point of each square was used to create a road map, which was loaded on the GPS. In the field, an operator followed the route in a slow-moving boat (≈ 1 kt.) while another operator continually observed the bottom with an optical viewer, signalling the presence of species to the GPS operator. The collected data were organized in two tables:

- the first one showed the GPS coordinates and the time at each track point, continuously recorded at 2 s intervals;
- the second one showed the GPS coordinates and the corresponding species, based on the recorded time.

The second table was displayed on the lagoon map with ArcView (ESRI), which showed the track by coordinate points. Each point was assigned a specific symbol for the species found there (Figures 2 and 3). By interpolating the points marked by the same symbol, it was possible to create irregular polygons that defined the area occupied by each species.

For the seasonal biomass sampling (July and October 2004 and April 2005), three replicate 50 × 50 cm squares were taken at each station to plot mixed communities of macrophytes. Scuba-divers collected the macrophytes by hand, taking care to
pick up whole seagrass shoots. All biomass was immediately freed of sediment and taken in cool boxes to the laboratory for further treatment. Here the epiphytes were removed, the species were separated, and each sample was washed with tap water to remove excess salts and carefully dried with blotting paper. The seagrasses were then divided into above- and below-ground portions. Lastly, the macrophytes were weighed and oven-dried at 70°C until constant dry weight (d.w.) was reached.

### Water sampling and analysis

At stations 2, 5, 7 and 8, daily variations in water column temperature, salinity, pH and dissolved oxygen (DO) were measured with a multiprobe logger (Datasonde 3, Hydrolab, Austin, Texas, USA). Nutrients were analysed monthly at all stations. Water samples (1 L) were collected at 50 cm depth, taken to the laboratory in cool boxes, filtered through
a 0.22 μm cellulose acetate membrane (GSWP, Millipore Corp., Bedford, Massachusetts, USA) and stored at −18°C.

Nutrient concentrations were measured with a spectrophotometric technique, using the Flow Injection Analysis (FIA) instrument (Lachat Instruments, Milwaukee, Wisconsin, USA). In particular, orthophosphates were determined by ascorbic acid reduction, nitrites by the colorimetric method of sulphanilamide diazotisation, nitrates by a cadmium-reducing column and ammonia by the phenolate method (APHA, 1989).

The filters, stored at −18°C, were processed according to the Standard Methods (APHA, 1989) for chlorophyll a analysis.

**Sediment sampling and analysis**

Sediment samples were collected monthly from February 2004 to November 2004. At each station, 20 horizontal core samples were taken for analysis of the upper sediments (2.5 cm), using a 50 cm³ horizontal cover designed and built in ENEA Research Centre (Italian National Agency for New Technology, Energy and the Environment, Rome, Italy). The cores were immediately stored in anaerobic jars and taken in cool boxes to the field laboratory, where they were placed in an anaerobic glove-box for further treatment and handling. Nutrients and biochemical composition were determined seasonally, sulphate-reduction rates and porosity monthly, and AVS and iron every 2 months.

**Elemental and biochemical composition of organic matter**

A fraction of the sample cores was used for nutrient detection and determination of the biochemical composition of the organic matter. The samples were mixed and then frozen until tested. Before analysis, they were freeze-dried, sieved (at 2 mm) and milled.

Total carbon (TC) and total nitrogen (TN) were analysed with a Carlo Erba EA 1108 Elemental CHN Analyser (Carlo Erba, Milan, Italy). Inorganic carbon (IC) was determined by the Dietrich-Fruehling Calcimeter method (ISNP, 2000), then subtracted from TC to estimate the organic carbon (TOC) content (Weliky et al., 1983; Fichez et al., 1993; Newton et al., 1994; Wai Ting Tung and Tanner, 2003).

Total and inorganic phosphorous (TP and IP) were analysed by a colorimetric method, as described by Aspila et al. (1976), and organic phosphorous (OP) was determined by subtracting IP from TP.
Carbohydrates (CHO) were analysed according to Dubois et al. (1956), as optimized by Gerchacov and Hatcher (1972), and were expressed as glucose equivalents. Proteins (PRT) were analysed according to Hartree (1972), as modified by Rice (1982), and were expressed in terms of bovine serum albumin (BSA) equivalents. Lipids (LIP) were extracted according to Bligh and Dyer (1959) and analysed according to Marsh and Weinstein (1966), as modified by Danovaro (1996) for sediment samples and were expressed as tripalmitic-acid equivalents.

Sediment samples combusted at 500°C for 4 h and processed as described above were used as sediment blanks for all biochemical analyses (Pusceddu et al., 2003).

An estimate of the trophic availability of the sediment was obtained by assuming proteins, carbohydrates and lipids as the more labile compounds of the sedimentary OM (Danovaro et al., 1993). The three main biochemical classes were converted into carbon equivalents, using the conversion factors 0.45 gC g⁻¹, 0.50 gC g⁻¹ and 0.70 gC g⁻¹, respectively, according to the standards used (Fichez, 1991; Fabiano et al., 1995). The sum of converted carbohydrates, proteins and lipids was defined as the biopolymeric fraction of carbon (BPC, as per Fabiano and Danovaro, 1994), and taken to be a reliable estimate of the labile fraction of OM readily available to deposit-feeders.

The BPC/OC ratio, expressed as a percentage, was used as an index of ‘trophic quality,’ while the protein-to-carbohydrates ratio (PRT/CHO) was assumed as an estimate of OM ageing (Fabiano et al., 1997).

Chlorophyll a and pheopigments were analysed with the spectrophotometric methods described by Plante-Cuny (1974). Autotrophic organic carbon was determined by converting phytopigments (chlorophyll a and pheopigments) into carbon equivalents, using the conversion factor 0.40 gC g⁻¹ (Nival et al., 1976; Pusceddu et al., 1999).

Lastly, porosity was estimated from weight loss at 70°C in a known volume of sediment.

**Sulphate reduction, AVS, labile iron**

For each station, central parts (20 mL) of the cores were placed together in a N₂ glove-box under anaerobic conditions. BSR rates were estimated by the ³⁵SO₄ radiotracer technique (Rosser and Hamilton, 1983). The labile iron pool was determined by extracting 1 g of wet sediment with 100 cm³ of HCl 0.5 mol L⁻¹. After shaking for 12 h, then filtering, ferrous iron was measured in the supernatant according to the TPTZ method (Dougan and Wilson, 1973). Total extractable iron was determined with the same method, after ferric iron
reduction with hydroxylamine chlorhydrate (10 g 100 mL\(^{-1}\)). Labile ferric iron concentration was estimated as the difference between total and ferrous iron. The sediment’s chemical buffering capacity was estimated according to Azzoni et al. (2005), using the equation \(bL = [LFe(II) - AVS] + 1.5LFe(III)\).

AVS were determined by distillation under anoxic conditions (Fossing and Jorgensen, 1989). AVS, which released by the addition of \(H_2SO_4\) were removed through an AVS determination was made every 2 months. These determinations were made every 2 months.

**Statistics**

To explore the spatial patterns of the stations, a principal component analysis (PCA) was performed on the average annual data, using Statistica 6.0 software. The non-parametric Mann–Whitney U-test was performed with Statistica 6.0 software for the AVS parameter. The non-parametric Spearman correlation coefficient (R) was estimated for sulphate reduction activity, temperature, C/N and BPC/TOC ratios and for P-PO_4^{3-}, BSR and AVS.

**RESULTS**

**Benthic vegetation**

The distribution maps of submerged vegetation in the Fogliano and Caprolace lagoons are shown in Figure 1.

The Fogliano lagoon is characterized by \(R.\ cirrhosa\) (99% of the total surface), while \(C.\ nodosa\) occupies 5%. The \(C.\ nodosa\) bed grows at station 8, and in part of the area both seagrasses are present. There is little growth of green algae (Chlorophyceae); in particular, Ulvales are limited to the south, at station 9.

Caprolace, by contrast, is characterized by a continuous bed of \(C.\ nodosa\) (80% of the total surface) around almost the whole perimeter of the lagoon. In limited areas near the inlet, two other Spermatophyta are found: \(R.\ cirrhosa\), in a narrow strip, south of the mouth, and \(Zostera noltii\) Homem in a small area just off the mouth. The red seaweed \(Rytiphloea tintoria\) (Clemente) C. Agardh is also present (7%).

Biomass is a useful quantitative and qualitative indicator of the primary production of the whole ecosystem. The Fogliano lagoon shows higher mean total biomass than Caprolace (120 g d.w. m\(^{-2}\), whereas in the Caprolace lagoon they are fairly constant (41 g d.w. m\(^{-2}\), though with an increase in spring (55 g d.w. m\(^{-2}\) (Figure 2). The differences and the seasonal variations are closely related to the dominant seagrass species: in summer, \(R.\ cirrhosa\) (stations 6, 7 and 9) accumulates 2.5 times more biomass than does \(C.\ nodosa\) (stations 1, 2, 5 and 8), mainly due to growth in the above-ground portion (Figure 3). The ratio of above-ground to below-ground biomass in these seagrasses fluctuates during the year, but for \(C.\ nodosa\) it ranges from 0.4 to 2.9 whereas for \(R.\ cirrhosa\) it ranges from 0.8 to 9.0. Notably at station 7, where the range of \(R.\ cirrhosa\) was the widest, the above-ground to below-ground biomass ratio peaked in July, but after a dystrophic crisis in late summer, \(R.\ cirrhosa\) completely disappeared.

**Oxygen and salinity**

Daily variations in DO in the Caprolace and Fogliano lagoons (stations 2 and 5 in Caprolace and stations 7 and 8 in Fogliano) are shown in Figure 4. At all four stations, variations were always wider from April to August, and began to narrow in September. In late spring and in summer, hypoxic conditions were more marked at station 7 in Fogliano. At station 8, DO values were generally higher than 50%, except in August, when they fell to almost 20%. At stations 2 and 5 in Caprolace there were smaller fluctuations and hypoxic conditions were never reached.

The salinity values in Caprolace (stations 2 and 5) and Fogliano (stations 7 and 8) are shown in Figure 5. Both lagoons showed similar trends, with higher salinity measured during the dry season. The widest range was recorded at station 7 in Fogliano (28.47–48.9\(^%\)), while station 8, off the inlet, was more influenced by seawater (29.74–44.9\(^%\)). In the Caprolace lagoon, salinity values ranged between 32.4\(^%\) and 42.9\(^%\), with very similar values at both stations.

**Nutrients and chlorophyll a in the water column**

The nutrient analysis showed low concentrations of dissolved inorganic nitrogen (DIN) and orthophosphate in both lagoons, with mean values of about 80 \(\mu g L^{-1}\) and 7 \(\mu g L^{-1}\), respectively (Table 2). By contrast, in the Fogliano lagoon P-PO_4^{3-} was found to have a typically seasonal pattern, with a summer increase (Figure 6). In the Caprolace lagoon, N-NH_4^+ concentrations were relatively stable throughout the year, while in Fogliano the concentrations increased from late summer to the winter months (Figure 7).

Chlorophyll \(a\) concentration in the water column was higher in Caprolace than in Fogliano, with mean values of about 10 mg m\(^{-3}\) and 3 mg m\(^{-3}\), respectively. In Fogliano no clear

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trend was observed; Caprolace showed a clear seasonal pattern, with an increase in autumn (Figure 8).

**Elemental composition of sedimentary organic matter**

Sediment analysis revealed clear differences between the two coastal lagoons (Figures 9 and 10). The mean annual TC concentration in Caprolace was 143.25 mg C g\(^{-1}\) sed. d.w. (SE \(\pm\) 9.06), whereas in Fogliano it was only 80.1 mg C g\(^{-1}\) sed. d.w. (SE \(\pm\) 4.4). The mean TN value was 12.27 mg N g\(^{-1}\) d.w. (SE \(\pm\) 1.01) in Caprolace and 7.17 mg N g\(^{-1}\) d.w. (SE \(\pm\) 0.91) in Fogliano. No evident seasonal fluctuations were observed.

TOC differed in much the same way as TC, with a mean annual value of 132.98 mg C g\(^{-1}\) d.w. (SE \(\pm\) 10.67) in Caprolace and 70.91 mg C g\(^{-1}\) d.w. (SE \(\pm\) 4.97) in Fogliano. TOC thus...
accounted for a considerable fraction of the total carbon (about 90%).

The average C:N ratio was higher and more homogeneous in Caprolace (C:N = 12.77; SE = 0.07) than in Fogliano (C:N = 11.75; SE = 1.08), and decreased in the summer in both lagoons. By contrast, station 8, near the inlet in Fogliano, showed much higher values (ranging from 13.25 to 16.54) and a different temporal pattern, suggesting a different origin for the OM.

TP concentrations were 388 mg kg\(^{-1}\) d.w. (SE = 8.9) in Caprolace and 354 mg kg\(^{-1}\) d.w. (SE = 12.3) in Fogliano. Lastly, OP concentrations were higher in Caprolace (244 mg kg\(^{-1}\) d.w; SE = 9.6) than in Fogliano (186 mg kg\(^{-1}\) d.w; SE = 11.2).
Mean sediment porosity values ranged from 0.73 to 0.78 in Caprolace (annual average 0.76), and from 0.62 to 0.73 in Fogliano (annual average 0.68).

**Biochemical composition of sedimentary organic matter**

Concentrations of proteins, lipids and carbohydrates were always higher in Caprolace than in Fogliano (Figures 9 and 10). The average protein content during the study period was 45.61 mg g$^{-1}$ d.w. in Caprolace (SE ± 3.29) and 35.24 mg g$^{-1}$ d.w. (SE ± 1.40) in Fogliano. The seasonal trend showed an increase in late spring and a decrease in the colder months, particularly marked in Caprolace.

Moreover, carbohydrate concentrations in Caprolace were more than twice those in Fogliano, with an average of 46.58 mg g$^{-1}$ d.w. (SE ± 3.22) and 20.93 mg g$^{-1}$ d.w. (SE ± 1.68), respectively; no relevant temporal variations were observed.

Average annual lipid concentrations were 7.51 mg g$^{-1}$ d.w. (SE ± 0.30) in Caprolace and 5.22 mg g$^{-1}$ d.w. (SE ± 0.58) in Fogliano. The concentration at Fogliano decreased markedly between May and August, while Caprolace did not display any clear trend.

BPC concentrations showed a temporal trend similar to that of proteins, and averaged 46.61 mgC g$^{-1}$ d.w. in Caprolace (SE ± 2.82) and 29.55 mgC g$^{-1}$ d.w. (SE ± 1.37) in Fogliano.

In both lagoons, proteins were the dominant class of labile compounds, with yearly averages of 48% and 58% in Caprolace and Fogliano respectively, followed by carbohydrates (40% and 29% respectively) and lipids (12% and 13%). However, Caprolace’s sediments seemed much more influenced by the carbohydrate contribution than those of Fogliano, where proteins accounted for most of the biopolymeric fraction (Figure 11).

The mean PRT/CHO ratio in Caprolace was almost half that in Fogliano (Figure 12) with mean values of 0.98 (SE ± 0.05) and 1.71 (SE ± 0.12), respectively. The temporal pattern showed an increase in late spring-summer and a decrease in the colder months, especially in the Caprolace lagoon, as was the case with PRT and BPC.

The BPC/TOC ratio values indicate that in terms of the annual average, BPC accounted for a small fraction of TOC in
the Caprolace lagoon, with only 35.38% (SE ± 1.61), but constituted a larger pool in Fogliano, reaching 49.95% (SE ± 1.54) (Figure 13).

By contrast, autotrophic carbon in the two lagoons did not differ significantly: 7.6 mgC g\(^{-1}\) d.w. (SE ± 0.28) in Caprolace and 7.45 mgC g\(^{-1}\) d.w. (SE ± 0.7) in Fogliano. Seasonal variations showed an increase in spring, followed by a summer decrease. The lowest values were recorded at stations 3 (Caprolace) and 8 (Fogliano), both located near the lagoon inlets.

Lastly, the autotrophic carbon/BPC ratio indicated that autotrophic carbon accounted for a smaller fraction of the BPC in Caprolace (16.46%) than in Fogliano (25.16%), suggesting a larger ‘autotrophic contribution’ (Pusceddu et al., 1999) in the latter lagoon.

**Bacterial sulphate reduction (BSR), AVS and labile iron**

Average values and standard error or range of variation of the measured variables are reported in Table 3.

Annual variations in BSR activity, in relation to temperature, are shown in Figures 14 and 15. BSR rates had a typical seasonal pattern, peaking in summer and falling to a minimum in winter at all stations, as appears from the significant correlation between temperature and BSR (Spearman > 0.7, \(N=11\); \(P<0.01\) at all stations). There were no differences between the Caprolace and Fogliano lagoons, except at station 3 and station 8, both located near the inlets, where the annual averages were lower.

Nevertheless, AVS concentrations revealed significant differences between the two lagoons, with the values of Fogliano being twice those of Caprolace (Mann–Whitney test = 176; \(n=28\); \(P<0.01\)) (Figure 16). Particularly in Caprolace, AVS concentration showed little annual variation and the highest values were measured in summer, concomitantly with the highest sulphate-reduction activity. In Fogliano the minimum values occurred in April with a gradual increase until October.

In both lagoons, sediment pools of labile amorphous iron were dominated almost exclusively by ferrous iron during all the sampling periods, with concentrations of ferric iron below or close to the detection limits (0.5 \(\mu\)mol mL\(^{-1}\)). However, concentrations of labile ferrous iron were relatively constant over the year, with the values one order of magnitude higher in Fogliano. The AVS/LFe ratio was always < 1 in both lagoons and at all stations, suggesting that the sediment-buffering

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*Figure 9. Seasonal trends in the elemental composition and in the biochemical parameters of sedimentary matter in the Caprolace lagoon.*
capacity of labile amorphous iron to precipitate sulphide exceeded sulphide production. BSR showed a significant negative correlation with C/N and BPC/TOC (Spearman $R = -0.59; -0.50$, $N = 36; P < 0.01$). Significant positive correlation was found between P-PO$_4$ and both BSR and AVS in Fogliano lagoon (Spearman $R = 0.71; 0.70$, $N = 16; P < 0.01$).

**Multivariate comparison between systems**

The PCA on annual mean values showed a clear differentiation between the two lagoons, with the first two principal components explaining 73.47% of the total variance (Figure 17). All the stations in Caprolace (1 to 5) were clearly distinguished from those in Fogliano (6 to 9), as shown in the score plot. Moreover, the stations close to the inlets (3 and 8) turned out to be distinct from the others. The first axis, which by itself explains 53.15% of the total variance, most probably describes the quantity and availability of sedimentary OM, as can be observed on the loading plot. In the negative part are the Caprolace stations, which are positively correlated to porosity, TOC, TN, CHO, LIP, PRT, BPC and OP. In the positive part are the Fogliano stations, where the variables that appear to most affect this distribution are temperature, PRT/...
CHO, BPC/OC, autotrophic contribution and available Fe. Moreover, the parameters most affecting the Caprolace stations are negatively correlated with those affecting Fogliano.

The second component, an additional 20.32% of the variance, discriminates between Fogliano stations 6, 7 and 9, grouped together and dominated by *R. cirrhosa* and station 8, which lies just off the inlet and is dominated by *C. nodosa*. The variables that seem to explain this distribution are C/N, BSR, AVS, autotrophic C and orthophosphate; however, except for station 3, their influence on the distribution at the Caprolace stations (mainly along the first factor), is rather small.

Interestingly, nitrogen compounds (especially nitrites and ammonia) in the water column do not seem to be related to any other variables, and display the lowest loadings on the first two principal components.

**DISCUSSION**

The seasonal biomass and the variations in the above-ground to below-ground ratio were the most significant differences between the two phanerogams: the biomass of *C. nodosa* was subject to weaker seasonal fluctuations, with greater variations in the above-ground portion than *R. cirrhosa*. The ecological and morpho-physiological characteristics of the two species are well known. In *C. nodosa*, the biomass of the leaves is more variable than the below-ground structures, which consist of horizontally-growing elements of larger diameter (rhizomes) and thinner vertical roots that penetrate 30 cm down into the sediment (as measured by the authors of this study; Duarte *et al.*, 1998; Cancemi *et al.*, 2002; Agostini *et al.*, 2003). By contrast, the biomass of *R. cirrhosa* is more variable during the year and lies mainly in the above-ground portion. The root apparatus penetrates only a few centimetres into the sediments.

**Table 3. Mean annual concentration of LFe, AVS, BSR and βL (with relative SE) and annual AVS/LFe and βL range values**

<table>
<thead>
<tr>
<th>Station</th>
<th>LFe μmol ml⁻¹</th>
<th>AVS μmol ml⁻¹</th>
<th>BSR mmol SO₄ m⁻² d⁻¹</th>
<th>AVS/LFe (max and min)</th>
<th>βL μmol ml⁻¹ (average, max and min)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Caprolace 1</td>
<td>27.4 ± 0.9</td>
<td>15.8 ± 0.7</td>
<td>1.9 ± 0.4</td>
<td>0.43–0.76</td>
<td>11.8 ± 1.3</td>
</tr>
<tr>
<td>Caprolace 2</td>
<td>14.7 ± 0.7</td>
<td>9.3 ± 0.5</td>
<td>2.0 ± 0.4</td>
<td>0.45–0.72</td>
<td>5.4 ± 0.7</td>
</tr>
<tr>
<td>Caprolace 3</td>
<td>26.3 ± 3.1</td>
<td>10.6 ± 0.8</td>
<td>1.4 ± 0.3</td>
<td>0.35–0.53</td>
<td>17.3 ± 2</td>
</tr>
<tr>
<td>Caprolace 4</td>
<td>37.7 ± 1.4</td>
<td>16.4 ± 0.6</td>
<td>1.7 ± 0.4</td>
<td>0.37–0.49</td>
<td>21.3 ± 0.8</td>
</tr>
<tr>
<td>Caprolace 5</td>
<td>23.8 ± 1.1</td>
<td>16.8 ± 0.8</td>
<td>1.8 ± 0.3</td>
<td>0.56–0.84</td>
<td>7 ± 1.0</td>
</tr>
<tr>
<td>Fogliano 6</td>
<td>55.9 ± 3.0</td>
<td>26.7 ± 2.1</td>
<td>2.0 ± 0.4</td>
<td>0.27–0.67</td>
<td>29.1 ± 3.6</td>
</tr>
<tr>
<td>Fogliano 7</td>
<td>49.9 ± 2.5</td>
<td>26.8 ± 1.4</td>
<td>2.0 ± 0.5</td>
<td>0.34–0.65</td>
<td>23.3 ± 2.6</td>
</tr>
<tr>
<td>Fogliano 8</td>
<td>35 ± 1.5</td>
<td>12.1 ± 1.1</td>
<td>1.2 ± 0.3</td>
<td>0.17–0.55</td>
<td>22.8 ± 2.1</td>
</tr>
<tr>
<td>Fogliano 9</td>
<td>47.2 ± 1.5</td>
<td>21.8 ± 2.6</td>
<td>1.7 ± 0.4</td>
<td>0.05–0.72</td>
<td>24.7 ± 5.5</td>
</tr>
</tbody>
</table>

*Figure 12. Seasonal variation of PRT/CHO ratio in the two lagoons.*

*Figure 13. Seasonal variation of BPC/TOC ratio in the two lagoons.*
while the photosynthetic portion occupies the whole water column.

Parameters describing sedimentary OM likewise revealed a significant ecological difference between the two lagoons, as shown in the PCA (Figure 17). OM quality can be considered a key indicator for predicting the ability of a given lagoon to buffer dystrophic crises (Giordani et al., 1996). Recent studies have estimated the available fraction of sedimentary organic pools through the determination of the main biochemical classes of organic compounds (proteins, carbohydrates and lipids), which are assumed to be easier to mineralize and assimilate (Fichez, 1991; Dell’Anno et al., 2000): higher values of BPC/TOC ratio represent the labile fraction of the organic substance (Fabiano et al., 1997; Pusceddu et al., 1999; Cividanes et al., 2002), while higher values of PRT/CHO represent the fresh fraction of OM. In fact, proteins are known to be more rapidly mineralized (by bacteria) than carbohydrates (Newell and Field, 1983), hence higher PRT/CHO ratios indicate living OM or ‘newly-generated’ detritus (Danovaro et al., 1993). Therefore, according to the BPC/OC and PRT/CHO, OM in Caprolace seems to be less available than in Fogliano, and is characterized by a larger amount of aged organic matter (on average <1). Moreover, the PCA showed a negative correlation between the amount of OM and its quality. On the other hand, several studies have suggested that accumulation of the amino acid proline in the cytoplasm of Ruppia spp., as well as in other halophytes, may enhance their halotolerance (Marchioni Ortu and Ortu, 1988). High proline concentrations may act as a defence mechanism, to withstand and balance high saline concentrations in vacuolar fluids (Stewart and Lee, 1974; Cavalieri and Huang, 1979). Proline concentrations in Ruppia spp. have been shown to rise

Figure 14. Monthly bacterial sulphate reduction activity versus temperature at the Caprolace stations.

Figure 15. Monthly bacterial sulphate reduction activity versus temperature at the Fogliano stations.
as habitat salinity increases (Brock, 1981, 1982); in leaf tissues, total carbohydrates decrease and soluble carbohydrate levels increase (Murphy et al., 2003). This adaptation could explain the higher PRT/CHO ratio as well as the origin of the autotrophic contribution found in Fogliano.

Moreover, in this study a significant negative correlation was observed between BSR and both the C/N and BPC/TOC ratios, suggesting that these parameters could be considered representative of an available substrate for BSR.

An important result of this study is that, although sulphate-reduction activity and AVS/LFe ratios in the two lagoons were comparable, the significant differences in their AVS concentrations and trends suggest that another buffer mechanism counteracting the accumulation of free sulphide is operating in Caprolace. The daily oxygen variations, the minimum oxygen values as well as the above- to below-ground ratio provide information about this question. In the Cymodocea-dominated lagoon the resistance to sulphides is reasonably higher because sediment reoxidation, through radial oxygen loss from roots (ROL), can proceed down to a depth of several centimetres, thus representing an effective barrier against H₂S diffusion. This could explain the different AVS trend in the two lagoons, with an accumulation in Fogliano. Besides, the different OM quality and total biomass suggest different respiration rates with a lower oxygen demand of a Cymodocea-dominated system. Moreover, a higher sediment porosity in Caprolace favours a faster oxygen diffusion from the water column to sediments.

More particularly, the similar AVS trends observed in all stations of Caprolace, cannot be easily explained on the basis of the below-ground portion alone. However, considering that C. nodosa covers 80% of the lagoon bottom, it is possible to hypothesize that the entire lagoon is quite uniformly influenced by its presence, in term of sediments oxidation and respiration rate.

Sulphide resistance is an important threshold of the ecosystem, and has further implications as well. Nutrient immobilization and release mechanisms depend on the oxidation state of sediments, which in turn is modified by the build-up of reduced sulphur compounds. In the Caprolace and Fogliano lagoons, the mean annual values and PO₄³⁻ in the water column were comparable. However, the trends were different: in Fogliano a significant positive correlation was observed between PO₄³⁻ and both AVS and BSR suggesting that a great amount of phosphate was released from sediments to the water column.

In conclusion, the results of this study highlight the functional differences of two coastal lagoons characterized by the dominance of two different species of seagrass. The lagoons differ in terms of ecological stability, which is mainly related to difference in their sediment buffer capacity to counteract the accumulation of free sulphide. These results give indications on how to assess the quality state of a Mediterranean lagoon and how to address conservation strategies. In particular the ecological difference of these two lagoons suggests that Caprolace needs surveillance in terms of routine monitoring, while Fogliano needs actions aimed at reducing its trophic state.

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REFERENCES


Figure 17. Results of the principal component analysis. The biplot ordination diagram for annual means for station (a) and variables which explain the ordination (b) are reported. PC1 explains 53.15% of the total variance of data and PC2 20.32%.


IRSA-C.N.R. 1986. Metodi analitici per i fanghi. Quaderni dell’Istituto di Ricerca delle acque 64(3).


