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SPATIAL AND TEMPORAL VARIATIONS IN MICROBIAL ACTIVITY IN THE MEDITERRANEAN SEA

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INTRODUCTION

The cycle of organic substances in the sea is strongly influenced by metabolic processes carried out by microorganisms within the microbial loop. In particular, heterotrophic bacteria are responsible for hydrolysis of a considerable fraction of the photosynthetic production both in particulate form (POM) and dissolved form (DOM) through their ectoenzymatic activity (EEA). The production of ectoenzymes is closely correlated to the availability of polymeric substrates or the lack of directly utilizable monomers. Hydrolysis products, transported to the inside of the cell, are in part used for the production of bacterial biomass (BCP, Bacterial Carbon Production). The coupling of hydrolysis and uptake processes increases the utilization efficiency of DOM by aquatic bacteria (Chrost, 1993).

Thereafter, the produced biomass is oxidised during respiration, with the release of CO_2 and nutrients in the aquatic systems in a newly assimilable form by primary producers. Furthermore, the respiration rates along the water column provide an index of the export of phytoplanktonic photosynthetic products towards the oceanic depths and as a consequence the oceans constitute a reservoir of CO_2 in the deepest layers.

Organic matter decomposition by means of ectoenzymatic activity and oxidation by means of respiration are the processes by which the input and output of C through the bacterial cells occur.

Microbial enzymatic and respiratory activities in the Mediterranean have been studied in the last years. The Mediterranean basin is, from a biogeochemical point of view, strongly conditioned by the morphology of the straits, which subdivide it into different sub-basins with peculiar characteristics affecting the trophic levels in the system. In this paper an overview of recent studies on the hydrolytic and respiratory activities observed in areas with different trophic conditions is reported.

MATERIALS AND METHODS

Enzymatic microbial activity (EEA) on the labile component of particulate and dissolved organic matter was determined by the hydrolysis of the fluorogenic substrate Leucina-7amino-4methylcoumarin and spectrofluorometric measurements. The utilisation of this substrate was proposed as a valid indicator of the protease activity in aquatic environments (Hoppe *et al.*, 1988, Zaccone and Caruso, 1996).

Respiratory rates (R) and the consequent metabolic production of CO_2 were evaluated by the ETS assay (Electron Transport System) which exploits the reduction of tetrazolium salts in formazan (Packard and Williams, 1981). The conversion factors of ETS to CO_2 for the euphotic and aphotic zones are reported in La Ferla *et al.* (1996) and Azzaro *et al.* (in press), respectively.

RESULTS AND DISCUSSION

Spatial variations

In the Mediterranean the average levels of protease activity ranged between fairly reduced levels in sites characterised by moderate trophism as in the Egadi Islands, to higher levels as in the Straits of Messina, reaching their maximum in the eutrophic waters in the Northern Adriatic (Fig.1).

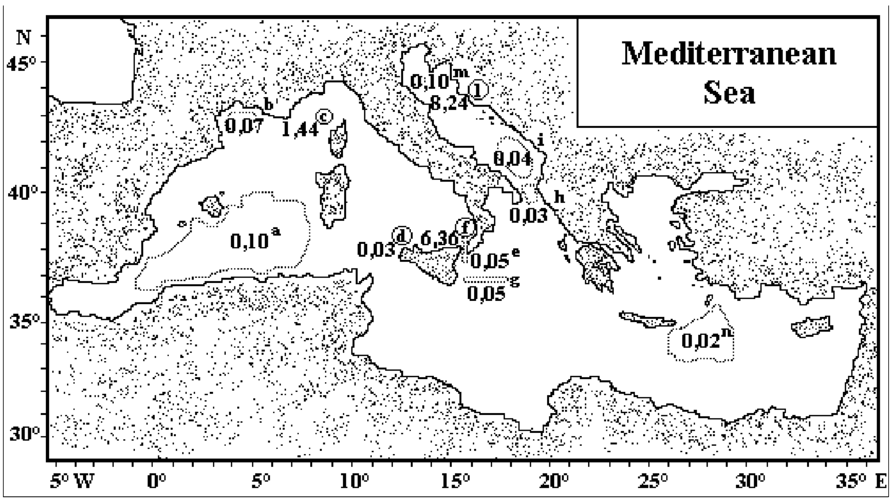

Fig. 1 Distribution of ectoenzymatic (in the circled letters) and respiratory activities (both expressed as µg C h^{-1} dm^{-3}) in the superficial waters of the Mediterranean Sea. (a: Martinez et al., 1990; b: Agustì and Cruzado, 1992; c: Karner and Rassoulzadegan, 1995; d: Caruso et al., 1988; e: Data Report,1997; f: Caruso, unpublished data; g: La Ferla et al., 1996; h: La Ferla et al., in press; i: La Ferla et al., 1996; l and m: Prisma-2, unpublished data; n: Azzaro, 1997).

In the area around the Egadi Islands, Atlantic waters flow in the picnocline layer and carry out trophic resources towards the southern part where a greater protease activity can be observed in the surface layers (0.048 µg C h^{-1} dm^{-3}). Moving from the South to the North, protease activity levels shift to the deeper layers in relation to the sinking of picnocline (0.013 µg C h^{-1} dm^{-3} in the photic zone and in bottom layer 0.055 µg C h^{-1} dm^{-3}), following a trend which is related to the distribution of particulate organic nitrogen content (PON) (Caruso *et al.*, 1998).

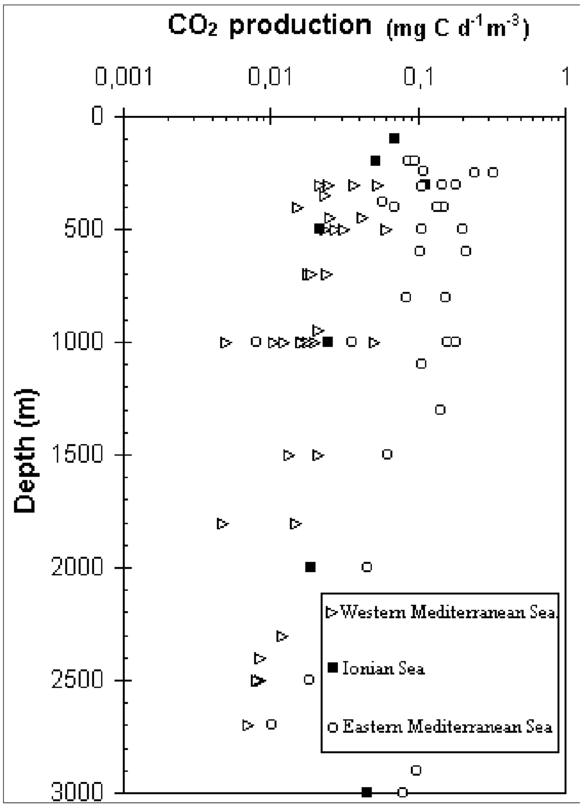

Fig. 2 Distribution of the CO_2 production rates along the water column (from 100 to 3000 m) in the Western Mediterranean (Christensen et al., 1989), in the Ionian Sea and in the Eastern Mediterranean (Azzaro, 1997).

A general picture of the Mediterranean Sea deriving from mean respiration data has revealed a different scenario for the euphotic and aphotic zone. Infact, in the surface layer (0-200m) a decreasing respiratory activity from the West to the East has been observed, as in the Northern Adriatic towards the Otranto Channel (Fig. 1). In opposite the respiratory rates and than the CO_2 production rates, in layers below 200 m, proved to be higher in the Eastern Mediterranean, probably because of the winter convection processes which entrain organic substances in the deep waters (Fig.2).

Integrated data of CO_2 production in the Mediterranean Sea (derived from the equation mg C d^{-1} m^{-3} = 2.467 $z^{-0.631}$) compared with those found in the Pacific, Indian and Atlantic Oceans are reported in Table 1.

TABLE 1.
Shallow respiratory rates (integrated from 200 to 1200 m), deep respiratory rates (integrated from 1500 to 2500 m) and deep/shallow ratios (expressed as percentage) estimated in the Mediterranean Sea and in the Indian, Pacific and Atlantic Oceans (from Azzaro, 1997).

Area	$_x$Rdz: Shallow (200-1200 m) mgC d^{-1} m^{-2}	$_x$Rdz: Deep (1500-200 m) mg C d^{-1} m^{-2}	$_x$Rdz: Deep / $_x$Rdz: Shallow %	Reference
Mediterranean Sea	44.2	20.6	46.6	Azzaro 1999
Indian Ocean	105.1	22.9	21.8	Naqvi at al. 1996
Pacific Ocean	140.4	29.6	21.1	Packard et al. 1998
Atlantic Ocean	50.4	7.5	14.9	"

The Deep zone/Shallow zone ratio, espressed as a percentage, shows just how important is the oxidation taking place in the deeper layers of the Mediterranean Sea with respect to others seas.

According to Christensen *et al.* (1989), who had already determined a shallow zone/deep zone ratio of 45.4 in the Western Mediterranean, such evidence may be imputed to the trapped DOC in waters which sinks during the formation of deep and intermediate waters, in addition to the normal particulate flow which sinks down from surface waters.

Temporal variations

Temporal variations in respiratory activity were found in the surface layers of the Otranto Channel during two surveys carried out in February and August 1994, with higher values in the euphotic layers during the summer (February: 0.41 ± 0.21; August: 0.74 ± 0.76 µl O_2 h^{-1} l^{-1}). In the deeper layers winter and summer respiratory rates were comparable (February: 0.39 ± 0.18; August: 0.43 ± 0.16 µl O_2 h^{-1} l^{-1}) (La Ferla *et al.* in stampa).

Results of microbial enzymatic activity assay obtained in the Northern Adriatic in different seasonal periods are strongly affected by the peculiar hydrological conditions of this basin. In fact, the Northern Adriatic water circulation determines variations on a seasonal scale, with re-mixing in winter and a stratification period in the other seasons (Artegiani *et al.*, 1997a; 1997b). The intense input of organic matter from rivers constitutes an anthropic supply, which determines a decreasing level of microbial protease activity moving from the coast toward off-shore areas. This trend can be observed in the summer months in the surface and sub-surface layers. Protease activity also shows a decreasing trend from the surface to greater depths (Fig.3).

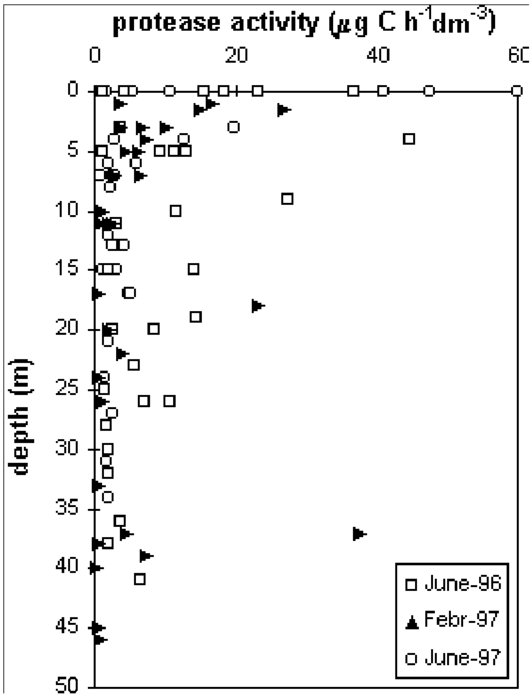

Fig. 3 Distribution of Protease activity in Northern Adriatic during the three surveys.

During winter average protease activity (6.18 µg C h^{-1} dm^{-3}) shows lower values than in summer (9.69 µg C h^{-1} dm^{-3}). As observed in the area of the Egadi Islands, the break down of the thermocline affects the activity distribution; during this season, the re-mixing condition determines a rapid sinking of organic matter in some stations influenced by Po river runoff, and therefore a higher activity in the deeper rather than surface layers. The variability in the area seems greater at depth (Variation Coefficient = 136) than at the surface (V.C. = 93).

Greater temporal variations were observed in respiratory rates in the Northern Adriatic (range 0.05- 11.51 µl O_2 h^{-1} dm^{-3}). These too, were related to the different hydrological conditions governed by water circulation and river input. In particular, high values were found in June 1996 and 1997 (mean values: 1.84 ± 1.55 and 3.31 ± 2.79 µl O_2 h^{-1} dm^{-3}, respectively), with increasing values over the two years. Lower values were recorded in the winter (mean value: 0.34 ± 0.21µl O_2 h^{-1} dm^{-3}).

On seasonal scale, different correlations showed that during summer the hydrolytic and respiratory processes are well coupled both each other and with other parameters (Table 2). During winter the processes are uncoupled with each other and showed different behaviour.

TABLE 2.
Significant correlation coefficients between EEA and R versus other parameters in Adriatic Sea (' : P < 0.05; " : P< 0.01; n = 30). .

	June 96	Febr 97	June 97		June 96	Febr 97	June 97
EEA versus				R versus			
R	0.39'		0.55''	EEA	0.39'		0.55''
DOC	0.47'	0.81"	0.68"	*DOC*	0.39"		
BCP	0.75"	0.47"	0.99"	*BCP*			0.53"
T° C	0.50"		0.61	*T° C*	0.44'		0.47"
S	-0.75"	-0.42'	-0.94"	S	-0.58'		-0.65"
POC	0.55"		0.94"	*POC*			0.74"
PON	0.56"		0.93"	*PON*			0.77"

On yearly scale the hydrolysis of organic matter by enzymes and respiration increased from 1996 to 1997, with a preferential attack on the dissolved fraction (DOC) in June '96 and on the particulate matter (POC) in June '97. This would suggest that hydrolytic and respiratory capacities are to be imputed to the quality of substrates present in the environment.

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