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ORGANIC MATTER DECOMPOSITION PATTERNS IN THE SOUTHERN ADRIATIC AND IONIAN SEAS: OBSERVATIONS FROM THE ADREX CRUISE (PERSEUS PROJECT)

PATTERNS DI DECOMPOSIZIONE DELLA MATERIA ORGANICA NEI MARI ADRIATICO MERIDIONALE E IONIO: OSSERVAZIONI DALLA CAMPAGNA ADREX (PROGETTO PERSEUS)

Abstract - The processes of organic matter decomposition via microbial enzymatic hydrolysis were studied in the southern Adriatic and Ionian Seas, areas characterized by ocean gyres and deep water formation, during the ADREX cruise (February 2014), to gain insights into the biogeochemical role of the microbial community. Proteolytic activity was predominant, followed by active decomposition of organic phosphates and polysaccharides. Dissolved enzymes contributed to organic matter recycling, especially at depth, suggesting the bioavailability of the pool of organic substrates. Correlations found between enzyme activity patterns and trophic variables suggested that microbial metabolism was driven by the organic substrates transported by water masses.

Keywords: organic matter, microbes, enzymes, Adriatic Sea, Ionian Sea.

Introduction - The Southern Adriatic and Ionian Seas represent model ecosystems to assess how microbial communities respond to environmental changes modulating organic matter decomposition. However, little attention has been devoted to the microbial processes involved in the deep pelagic ecosystem functioning (La Ferla *et al.*, 2005, 2010). Microbial enzymes such as Leucine-aminopeptidase (LAP), beta-glucosidase (GLU) and alkaline phosphatase (AP) drive the decomposition of proteins, polysaccharides and organic phosphates, respectively, playing a key role in water biogeochemistry, in terms of carbon (LAP and GLU), nitrogen (LAP) and phosphorus (AP) regeneration. While the prokaryotic abundance and activity in these pelagic ecosystems were previously studied (La Ferla *et al.*, 2005, 2010), the complex interplay between microbial metabolism and the organic matter pool has scarcely been addressed. In this study, the patterns of microbial decomposition of organic matter in the southern Adriatic and Ionian seas were examined in relation to the main physical and trophic parameters.

Materials and methods - During the ADREX cruise (February 2014) performed in the framework of the EU FP7 PERSEUS project (grant n. 287600), the enzymatic activities in the epi- (<100 m), meso- (100<depth<1000 m) and bathy- (>1000 m) pelagic layers were studied to evaluate the biogeochemical role of microbes. The sampling was carried out at 6 stations (3 on the western side of the considered basins and 3 on their eastern side, Fig. 1). A multiparametric probe SeaBird 911plus was used to measure Temperature (T), salinity (S) and dissolved oxygen (DO). Fluorimetric assays were performed onboard to measure total (T) and dissolved (D, <0.22 µm) leucine aminopeptidase (LAP), beta-glucosidase (GLU) and alkaline phosphatase (AP) enzymes; fluorogenic substrates were added to assess the potential

proteolytic, glycolytic and phosphatase activity, respectively. Trophic parameters such as Particulate Organic Carbon (POC) and Particulate Nitrogen (PN) were also determined via CHN elemental analyzer (Costech ECS4010). Nitrogen (N) and Phosphorus (P) nutrients (i.e., ammonia NH_4 , nitrite NO_2 , nitrate NO_3 , orthophosphate PO_4) were measured by conventional laboratory methods based on spectrophotometric analysis. Chlorophyll-*a* (Chl-*a*) as a proxy of phytoplankton biomass was determined by fluorimetric analysis (La Ferla *et al.*, 2005). Pearson correlations among microbial and environmental variables were computed by PAST v.4.0 software.

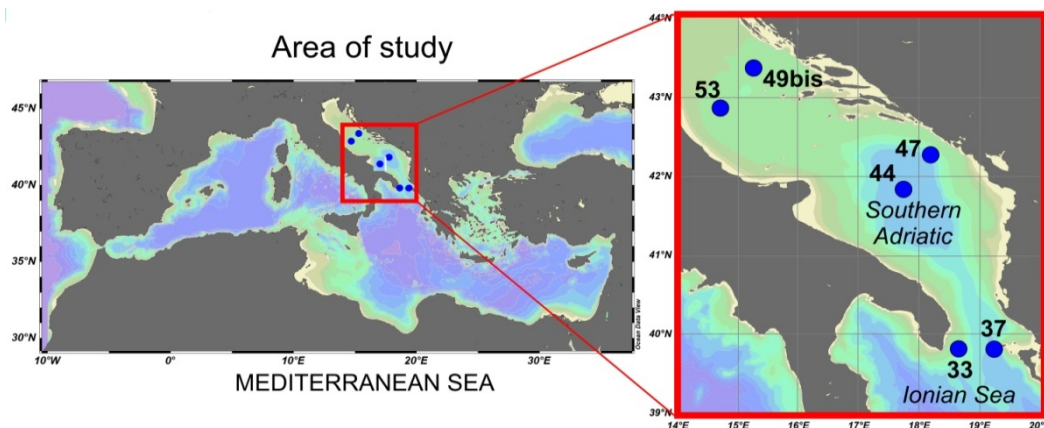
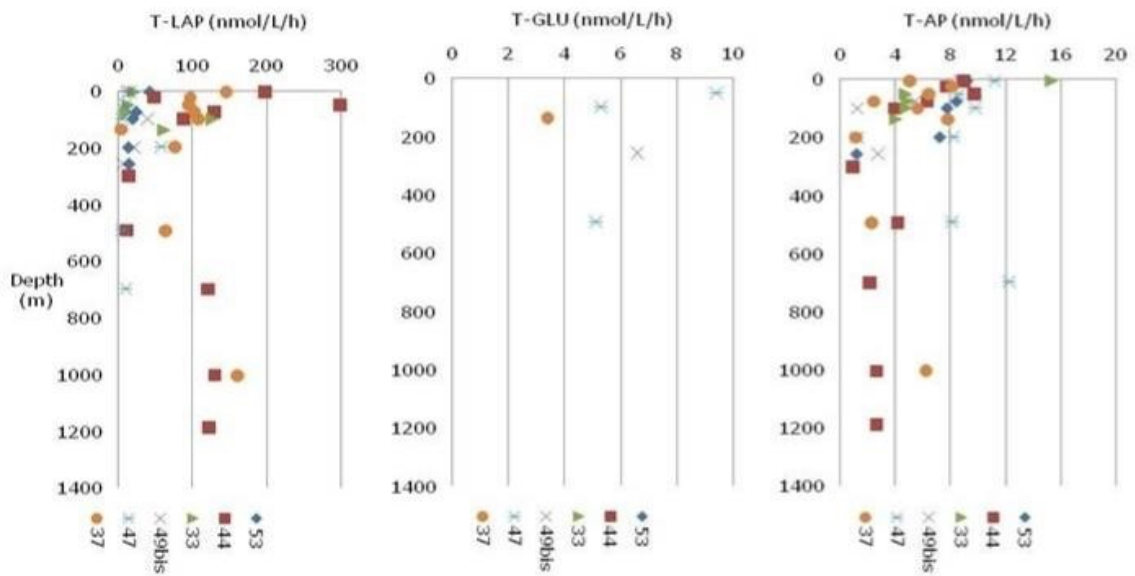


Fig. 1 - Location of the stations sampled in the southern Adriatic and Ionian Seas.

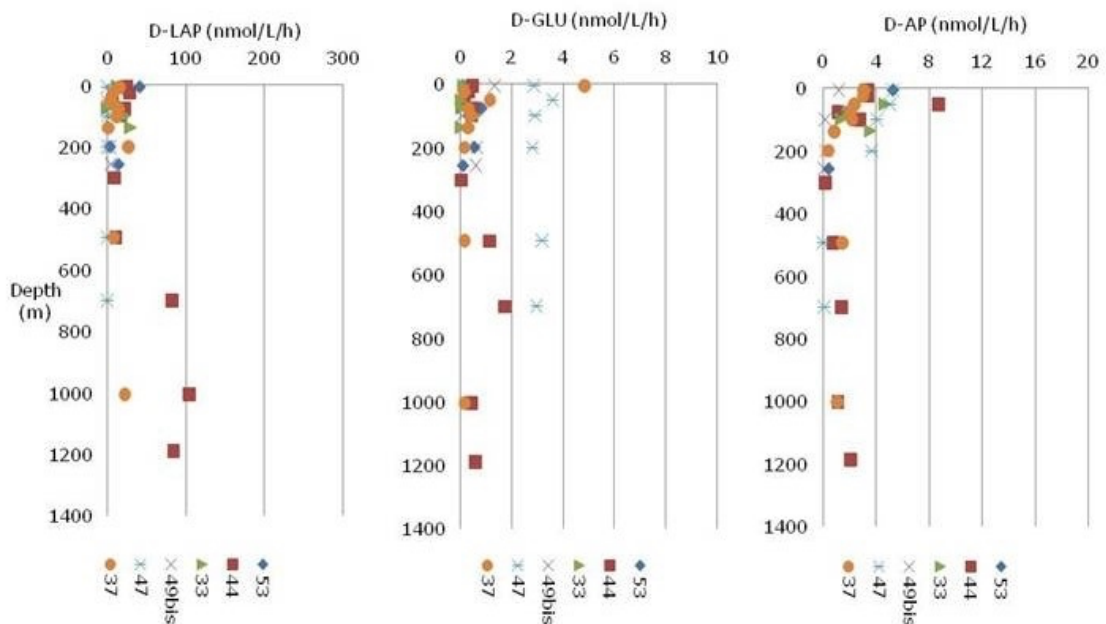
Posizione delle stazioni campionate nei mari Adriatico meridionale e Ionio.

Results – In the study period, mean (\pm st. dev.) T values varied between 12.1 ± 0.9 °C to 16.0 ± 1.0 °C; S from 38.5 ± 0.1 to 39.0 ± 0.1 (at western and eastern stations, respectively) and DO between 190.0 ± 17.5 $\mu\text{mol/L}$ and 249.7 ± 18.4 $\mu\text{mol/L}$. POC ranged from 0.3 to 3.6 $\mu\text{mol/L}$ and from 0.2 to 2.9 $\mu\text{mol/L}$ (at western and eastern side, respectively), PN from 0.04 to 0.37 $\mu\text{mol/L}$ and from 0.02 to 0.44 $\mu\text{mol/L}$ (at western and eastern side, respectively). NH_4 , NO_2 and PO_4 prevailed in the western stations, with ranges of 0.03-3.6 $\mu\text{mol/L}$, 0.01-0.3 $\mu\text{mol/L}$ and 0.01-0.2 $\mu\text{mol/L}$, respectively; NO_3 varied between 0.02 ± 0.2 and 6.4 ± 2.0 $\mu\text{mol/L}$, peaking in the eastern side. Total and dissolved enzymatic activity profiles are shown in Fig. 2. Proteins (T-LAP) were decomposed more effectively than phosphates (T-AP) and polysaccharides (T-GLU). Average T-LAP rates decreased from the epipelagic to deeper layers (from 84.9 ± 89.1 to 58.9 ± 53.9 nmol/L/h). High T-LAP rates measured at Stat. 44 in the southern Adriatic indicated abundance of proteins. D-LAP largely contributed to the total LAP activity in the bathypelagic layer of the western stations (D-/T-LAP = 74.3%, Tab. 1), suggesting the key role of dissolved enzymes at depth. T-GLU and T-AP (range: 1.6 ± 1.1 - 3.2 ± 2.3 nmol/L/h; 2.6 ± 0.01 - 7.6 ± 3.1 nmol/L/h, respectively) were high at Stat. 33 in the Ionian Sea; on the eastern side, both prevailed at Stat. 47 in the southern Adriatic Sea.

Converting POC and PON pools and enzymatic data from molar to $\mu\text{g/L}$ units, particulate organic matter with high C/N molar ratio (22.5, referable to detritus) was found at the bathypelagic layer in the western side (Tab. 2). LAP/GLU ratios increased from epi- to the bathypelagic layer, probably related with new deep waters' formation.



(a)



(b)

Fig. 2 - Integrated and depth-normalized activity rates of (a) total (T-LAP, T-GLU and T-AP) and (b) dissolved (D-LAP, D-GLU and D-AP)

Tassi di attività enzimatiche (a) totali (T-LAP, T-GLU e T-AP) e (b) disciolte (D-LAP, D-GLU e D-AP) integrati e normalizzati rispetto alla profondità.

On the Western side, T-AP correlated positively with T and DO ($r=0.57$, $p<0.01$; $r=0.52$, $p<0.05$, respectively) and negatively with PO_4 ($r=0.59$; $p<0.01$), suggesting active phosphorus regeneration. D-AP correlated also positively with T-LAP ($r=0.50$, $p<0.05$), suggesting synergy between C and P cycles. On the eastern side, PN was negatively related with NO_3 ($r=0.58$, $p<0.01$). T-AP correlated positively with D-GLU and T-GLU ($r=0.569$ and 0.70 , $p<0.01$, respectively), confirming links between polysaccharides' and phosphates' decomposition.

Tab. 1 - Ratios of dissolved (D) to total (T) leucine aminopeptidase (LAP), glucosidase (GLU) and alkaline phosphatase (AP) in the three layers (epi-, meso- and bathy-pelagic) expressed as percentage.
Rapporti degli enzimi leucin aminopeptidasica (LAP), glucosidasica (GLU) e fosfatasica (AP) riferibili ad enzima disciolto (D) rispetto all' attività enzimatica totale (T) nei tre strati (epi-, meso- e bati-pelagico), espressi in percentuale.

		D-/T-LAP (%)	D-/T-GLU (%)	D-/T-AP (%)
Epipelagic	West	20.9	11.9	44.1
	East	13.0	51.2	41.5
Mesopelagic	West	62.0	24.05	34.1
	East	17.6	50.8	15.7
Bathypelagic	West	74.3	31.5	61.8
	East	13.8	45.8	17.5

Tab. 2 - Organic Carbon and Nitrogen pool, C/N and LAP/GLU ratios and amounts of Carbon and Phosphorus mobilized by LAP+GLU activities and AP activity respectively, with the percentage of POC decomposed by LAP+GLU.
Pools di Carbonio ed Azoto Organico, rapporti C/N e LAP/GLU e quantità di Carbonio e Fosforo mobilizzati dalle attività LAP+GLU ed AP, rispettivamente, con la percentuale di POC decomposto da LAP+GLU.

West vs East	POC ($\mu\text{gC/L}$)	PN ($\mu\text{gN/L}$)	C/N molar Ratio	LAP/GLU	Mobiliz. C ($\mu\text{gC/m}^2/\text{day}$)	% decompos. POC	Mobiliz. P ($\mu\text{gP/m}^2/\text{day}$)
Epipelagic	28.5 vs 25.2	4.5 vs 4.0	7.7 vs 7.4	31.5 vs 18.2	1.6 vs 1.1	70.0 vs 53.0	0.06 vs 0.05
Mesopelagic	16.3 vs 10.5	1.9 vs 1.5	10.6 vs 9.7	15.9 vs 11.5	0.1 vs 0.08	8.9 vs 12.1	0.004 vs 0.006
Bathypelagic	23.9 vs 7.4	1.4 vs 0.2	22.5 vs 35.7	80.2 vs. 469.1	1.2 vs 4.2	58.5 vs 68.3	0.01 vs 0.07

Conclusions - Microbial community was able to hydrolyze proteins also at the bathypelagic layer, where dissolved enzymes contributed mostly to organic matter decomposition. Proteins were mostly utilized in the upper layers at the western side, in the Southern Adriatic, and at depths in the eastern side; GLU and AP were more active on the eastern side in the Southern Adriatic. Enzymatic rates measured in this study were in the same magnitude order as those reported by Azzaro et al. (2012) during the winter period. The observed distribution patterns and the relationships with physical-chemical variables suggested that organic matter decomposition was mostly driven by the water masses' circulation. Microbial metabolic activities were suitable descriptors of the biogeochemical fluxes in the studied area and of the role of microbial community in the changing Mediterranean scenarios.

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