

# Assessing marine environmental status through microphytobenthos assemblages colonizing the Autonomous Reef Monitoring Structures (ARMS) and their potential in coastal marine restoration

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## ABSTRACT

Microphytobenthos is potentially highly sensitive to environmental alterations, but has been rarely utilized in monitoring studies. Here we investigated the use of microphytobenthos colonizing Autonomous Reef Monitoring Structures (ARMS) to assess the marine environmental quality. We analysed microphytobenthic assemblages in terms of abundance, biomass and species composition on ARMS deployed in northern Adriatic Sea along a gradient of increasing impacts. We show that microphytobenthic variables changed significantly across sites, with lowest abundance and biodiversity in the highly impacted site. Moreover, the specific analysis of Diatoms revealed that genera like *Entomoneis* and *Cylindrotheca* could be used as indicators of nutrient enriched and stressed conditions. We provide evidence that the analysis of microphytobenthos colonizing artificial substrates could be used as a tool for detecting altered environmental characteristics. We also show that the ARMS, re-creating hot spots of microphytobenthic biodiversity, and protect them from grazing, could be potentially utilized to restore degraded hard substrates. Our result indicates that microphytobenthos can be easily incorporated in future monitoring and restoration programmes to assess and improve marine environmental health.

## 1. Introduction

Microphytobenthos (MPBs) is composed by unicellular eukaryotic algae (i.e., Bacillariophyceae, Chlorophyceae and Dinophyceae), and autotrophic prokaryotes (Azovsky et al., 2013). Microphytobenthos is composed by highly productive phototrophic taxa and represent one of the most important and widespread component of marine primary production at global scale (Underwood and Kromkamp, 1999). The ecological importance of the microphytobenthos assemblages in marine ecosystems is due to the: a) their role in benthic primary production; b) the relevance as food for grazers and deposit feeders (De Jonge and Van Beusekom, 1992; Defew et al., 2002; Miller et al., 1996); c) the effects on sediment properties (through the production of extracellular polymeric substances, EPS; Ubertaini et al., 2015; Underwood and Paterson, 2003), and d) the role on nutrient and oxygen fluxes across sediment-water interface.

The analysis of microphytobenthic assemblages can provide important insights into the assessment of marine environmental status and of the impact of global change (Dura et al., 2016; Kostecki and Janczak-Kostecka, 2012; Lobban and Jordan, 2010; Recasens et al., 2015; Rioual et al., 2007; Romero and Armand, 2010; Vegas-Vilarrúbia et al., 2013).

These microalgae are considered useful indicators of environmental alteration for their short life cycles and quick response to abiotic and biotic changes (Chen et al., 2016; Potapova and Charles, 2007; Potapova et al., 2005). Other studies showed that diatom assemblages are sensitive to changes in salinity and to water movement (Busse and Snoeijs, 2002, 2003; Ulanova and Snoeijs, 2006). Planktonic and periphytic diatoms are included in many water quality monitoring programmes worldwide (Kireta et al., 2012; El-Karim, 2014), and they are routinely utilized in freshwater systems, whereas applications to marine water are still limited (Cibic et al., 2012; B-Béres et al., 2016; Desrosiers et al., 2013). Diatom assemblages have been utilized also for the study of the effects of contamination by heavy metals, where this group showed also the presence of teratogenic forms in response to this kind of pollution (Rogelja et al., 2016; Siqueiros-Beltrones et al., 2014). Recent studies showed that the assemblage structure of benthic diatoms and other microscopic benthic components can be modified by altered environmental conditions and some taxa can be used as bio-indicator both at the level of genus and species (Desrosiers et al., 2013; Hill et al., 2001; Pusceddu et al., 2009).

Diatoms represent also a significant component of the epilithon (flora adhering to hard substrates, and this definition is often utilized at

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the place of microphytobenthos of hard substrates; Busse and Snoeij, 2002, 2003; Facca and Sfriso, 2007; Ulanova and Snoeij, 2006) and, together with bacteria and Cyanobacteria, contribute to the biofouling (Callow and Callow, 2002; Charpy et al., 2012; Molino and Wetherbee, 2008). The colonization of hard substrates by microphytobenthos is thus a crucially important step for the succession of non-colonized (new) hard substrates at sea. Altered ecological conditions can modify significantly these processes with consequences on the entire benthic assemblage structure of hard substrates (Cibic et al., 2012; Franzo et al., 2016).

Moreover, the importance of microphytobenthos in the colonization and succession on hard substrates makes this component important for the use of artificial substrates in the recovery of damaged habitat. In this regard, the deployment of Autonomous Reef Monitoring Structures (ARMS), which have been conceived to monitor marine biodiversity across regions, could be of particular interest, since their peculiar and multilayer structure can allow the colonization by a wide variety of organisms, with different demands in terms of available light and protect them against grazing (e.g., by sea urchins). Moreover, the use of ARMS reduces the costs related to field work and monitoring, can be used as a tool for restoration of degraded habitats, does not depend on the natural substrate characteristics (Mirto and Danovaro, 2004; Danovaro et al., 2016). Hurley et al. (2016) and Leray and Knowlton (2015), investigated the biodiversity of the ARMS, but the microphytobenthos has been so far neglected.

Here, we investigated, for the first time, the microphytobenthic assemblages colonizing the ARMS with a twofold objective: i) to determine the response of this component to a gradient of environmental conditions and thus its suitability in monitoring studies; ii) to evaluate the similarity between the microphytobenthos colonizing the ARMS and the natural assemblages colonizing the surrounding hard substrates in order to explore the suitability of ARMS in restoration actions.

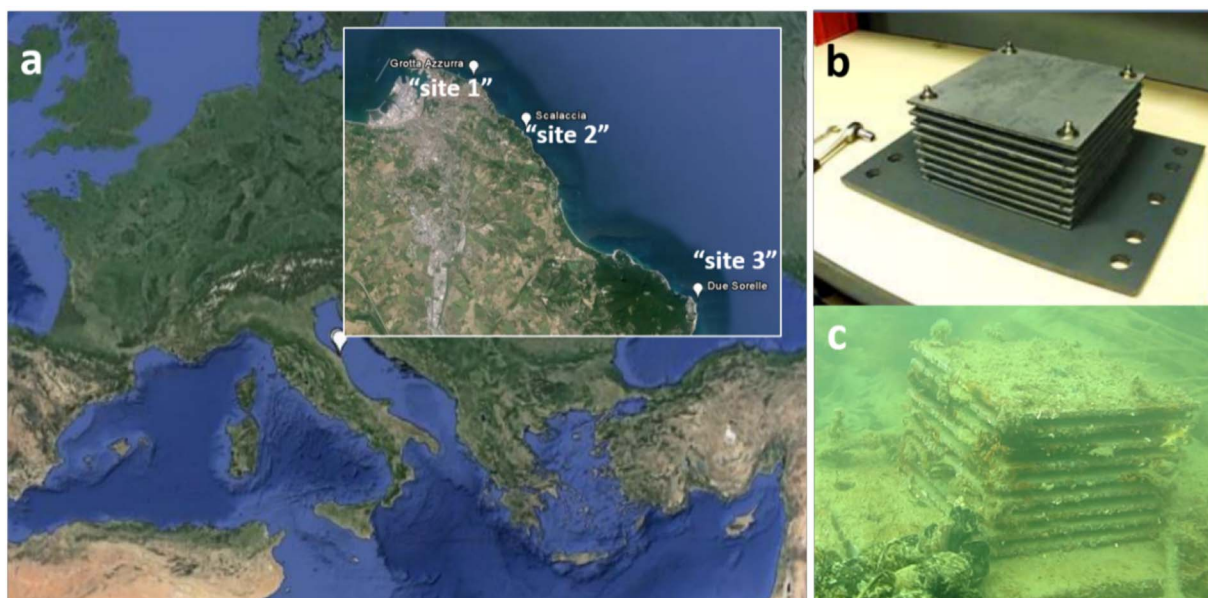
## 2. Materials and methods

### 2.1. Sampling and sites

Study areas were located in three coastal sites of the Northern Adriatic Sea (Fig. 1a), which is characterized by a strong gradient of environmental conditions at mesoscale (Giordani et al., 2002). The

selected area located between Ancona and the Conero promontory was characterized along a documented gradient environmental stress where the site 1 showed the highest values of nutrients (nitrogen and phosphorus) and lowest values of the environmental health index evaluated during the continuous monitoring conducted by the Regional Agency for the Protection of the Environment (A.R.P.A.M., 2013, 2014, 2015). The area investigated is known for the presence of relevant gradients and changes in environmental quality and previously utilized to test the suitability of artificial substrates to monitor meiofauna biodiversity and colonization (Mirto and Danovaro, 2004). Site 1 (Grotta Azzurra, 43°108 37.313 N, 013° 31.691E) is close to a rocky beach, and is characterized by the presence of artificial structures. The “urban beach” is subjected to intensive tourism and sewage discharge; moreover, it is close to Ancona commercial harbor thus it is defined “high impact” in the present study. Site 2 (La Scalaccia, 43° 36.291 N, 013° 33.102 E) is characterized by small rock platforms and artificial caves interested by intense activities, thought subjected to lower pressure than Site 1 and is considered as “moderate impact” in the present study. Finally, Site 3 (Due Sorelle, 43° 32.953 N, 013° 37.699 E) is located within the Conero Park area, it is thus protected, accessible only by sea and this far from any urban residential area; it is thus considered as “low impact” in the present study.

At each site, Autonomous Reef Monitoring Structures (ARMS) were placed at 7–9 water depths in 2 (site 1) or 3 point (sites 2 and 3) used as replicates. At each site replicated were located at approximately 2–5 m apart from each other. The ARMS, indeed, consist of 9 PVC (i.e., polyvinyl chloride) plates (225 mm × 225 mm × 6 mm) assembled on different layers to forms a single structure (Fig. 1b,c). The present experiment was conducted from June 2014 to July 2015. After 13 months, the ARMS were removed from the benthos according to the standardized procedure (Danovaro et al., 2016). Once recovered, the ARMS were brought in the laboratory, disassembled, scraped and preserved in ethyl alcohol 95% at 4 °C until subsequent analyses. In the present study we analysed half of the top surface of plate 1 (i.e., the most external PVC panel, directly exposed to light), plate 6 (i.e., in an intermediate position) and the plate 9 (i.e., in the most internal and thus almost completely obscure) for a total of 27 PVCs investigated.



**Fig. 1.** Sampling sites and artificial substrates in the Northern Adriatic Sea (Marche region). Reported are: a) the sites 1 (i.e., Grotta Azzurra) which has been considered “highly impacted”, the site 2 (i.e., La Scalaccia) considered here as “moderately impacted” and site 3 (i.e., Due Sorelle) considered as “low impact”; b) an image of the ARMS (Autonomous Reef Monitoring Structure) before deployment and c) an image of the same ARMS after deployment in the field.

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