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ALEX index enables detection of alien macroalgae invasions across habitats within a marine protected area

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ABSTRACT

A modified version of the ALien Biotic IndEX (ALEX) has been recently proposed to evaluate biological invasions in macroalgal assemblages. ALEX was applied in a Marine Protected Area where a recreational-fishing port is present testing the following hypotheses: ALEX increases with the distance from the port, it changes between the two directions off the port and it changes among three different habitats: *Cystoseira* beds, algal turf and dead matte of the seagrass *Posidonia oceanica*. A total of 78 native macroalgal taxa and 4 introduced species were found, the Chlorophyta *Caulerpa cylindracea* and the Rhodophyta *Apoglossum gregarium*, *Acrothamnion preissii* and *Womersleyella setacea*. All study sites were in high quality status highlighting that the assemblages investigated were at an early stage of NIS invasion. However, ALEX detected different values among conditions and habitats within the MPA, suggesting a local dynamics of NIS spread and different resistance to invasion of the investigated habitats.

1. Introduction

Biological invasions are considered a main global change and among the most severe threats to biodiversity worldwide (Pyšek and Richardson, 2010; Simberloff et al., 2013; Katsanevakis et al., 2014). Because of the ecological and social consequences of invasions, the assessment of not indigenous species (NIS) establishment and spread is considered a priority aiming to control the invasions (Anderson, 2007). Marine coastal areas are particularly exposed to invasion as many human activities, such as shipping, fishing and aquaculture, can drive introductions and spread of Non Indigenous Species (NIS) (Piazzi and Cinelli, 2003). In this contest, Marine Protected Areas (MPAs) can represent a useful tool to limit the spread of NIS (Ardura et al., 2016). In fact, the establishment of MPAs can reduce human activities acting as vectors of introduction and can increase the ecosystem resistance due to the maintenance of biodiversity and natural regulating processes (Mumby et al., 2011; Giakoumi and Pey, 2017). On the other hand, the effective role of MPAs to avoid NIS invasion is controversial, as some human activities (shipping, tourism, *etc.*) are allowed and/or implemented within the MPAs (Byers, 2005; Klinger et al., 2006; Burfeind et al., 2013).

The detection of NIS at sea is commonly more difficult than on land and invasions are generally not detected in advance impeding the success of eventual control programs. Thus, the assessment of suitable methods to early detect the presence of NIS and the level of invasion is a main goal for ecologists (Hewitt and Martin, 2001; Olenin et al., 2007). In this context, the European Marine Strategy Framework Directive (MSFD, EC, 2008) emphasises the priority to monitor abundance and the state of introduced species and environmental impact of invasive NIS, in order to avoid that biological invasions adversely alter the Good Environmental Status of coastal ecosystems (Borja et al., 2010, 2013). To achieve these objectives, the optimization of methods suitable to NIS assessment is crucial. The most part of surveys on NIS assessment are carried out within port areas (Campbell et al., 2007), as most NIS are not able to survive in inside the ports while they can widely spread throughout the adjacent waters (Piazzi and Cinelli, 2003). Thus, the survey of NIS inside the port and the spread of NIS in natural ecosystems starting from a source of introduction are two distinct problems that should be achieved through different methods.

A major problem in the monitoring and impact assessment concerns the sampling designs. The difficulty lies on the possibility to separate human-caused effects from patterns of natural variability (Underwood, 1993; Hewitt et al., 2001). Several methods have been proposed to evaluate the presence and spread of NIS in marine systems (Hewitt and Martin, 2001; Hoedt et al., 2001; Olenin et al., 2007; Otero et al., 2013), although no protocol has ever been universally accepted.

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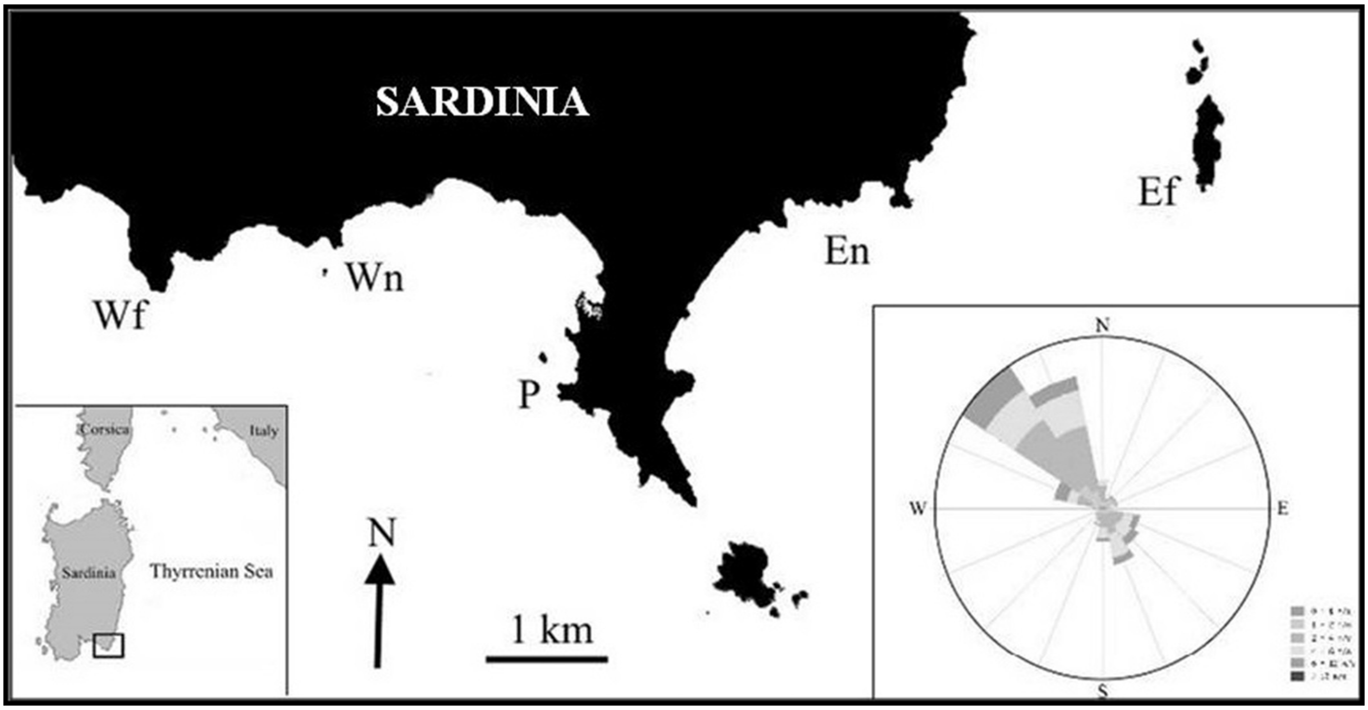

Fig. 1. Map of the study area with sampling sites and wind direction and intensity. P = port, En = East-near, Ef = East-far, Wn = West-near, Wf = West-far.

Beyond-BACI (Before/After-Control/Impact) designs comparing disturbed and control sites before and after the disturbance are considered the most suitable methods to evaluate effects of human-induced changes (Underwood, 1994; Benedetti-Cecchi, 2001), but, in the absence of 'before' data, post-impact studies have been widely used to detect differences between impacted and reference sites through ACI (After-Control/Impact) designs (Terlizzi et al., 2005; Fraschetti et al., 2006; Benedetti-Cecchi and Osio, 2007; De Biasi et al., 2016).

A further issue is the selection of suitable descriptors of ecological quality. The European strategies currently adopted for conserving the quality of coastal water (Water Framework Directive, EC, 2000; Marine Strategy Framework Directive, EC, 2008) require the identification of biotic indices suitable to identify environmental changes (Martinez-Crego et al., 2010). The ALien Biotic IndEX (ALEX) has been recently proposed to evaluate biological invasions in soft-bottom macro-invertebrate assemblages (Cinar and Bakir, 2014), and subsequently a modified version has been applied to Mediterranean macroalgal assemblages inhabiting hard bottoms (Piazzi et al., 2015). ALEX has been tested along gradients of invasions of five introduced macroalgae by using available datasets (Piazzi et al., 2015), but it has not been employed in the field. The invasion of alien macroalgae is considered a main threat for coastal ecosystems especially for the Mediterranean Sea that harbours the greatest concentration of introduced macroalgae of the world (Ribera and Boudouresque, 1995; Boudouresque and Verlaque, 2002; Klein et al., 2005).

The aim of the present study was to evaluate the effectiveness of ALEX to detect small-scale differences at early stages of NIS spread. To achieve this objective, ALEX was applied on three different habitats (a canopy-forming algae, algal turfs and degraded seagrass) within a Marine Protected Area where a recreational-fishing port is present. A comparison between a site next to the port and four reference sites distributed at two distances, but different directions, was done. The rationale is based on the expectation that the port is a source site of NIS to spread around. The following hypotheses were tested: ALEX increases with the distance from the port, it changes between the two directions off the port, and it changes among habitats. This latter derives from evidence that, in marine environments, the establishment and the spread of some of the most widespread non-native seaweeds are influenced by the characteristics of the receiving habitat (Arenas et al., 2006; Britton-Simmons, 2006).

2. Material and methods

The study was done within a Marine Protected Area (Villasimius - Capo Carbonara MPA, South-Eastern Sardinia, Fig. 1), where a recreational-fishing port is present. The dominant winds in the area are North-West (Servizio Mareografico Nazionale, Italian National Institute for Environmental Protection and Research, Fig. 1). Samplings were carried out in July 2016. A total of five sites were selected in the MPA: just outside the port, two near and two far control sites (about 2 km and 4 km from the port, respectively, Fig. 1). All sites were completely exposed to South winds and partially sheltered by the dominant North-West wind. In all the sites NIS were not visually detected through SCUBA surveys. In each site three habitats were sampled at about 6 m of depth: *Cystoseira* beds (rocky bottom colonized by canopy-forming seaweeds of the genus *Cystoseira,* mostly *C. brachycarpa* J. Agardh and secondly *C. compressa* (Esper) Gerloff & Nizamuddin and *C. crinita* Duby), algal turf (rocky bottom colonized by algal species < 10 cm high) and dead matte (dead matte of the seagrass *Posidonia oceanica* (L.) Delile colonized by algal turfs). In each site and habitat, two areas 10s of m apart were randomly selected and three samples were collected in each area for a total of 90 samples. Each sample consisted of all macroalgae within 400 cm^2 collected by scraping the bottom with a hammer and a chisel (Boudouresque, 1971). The material was conserved in formalin seawater and at the laboratory all taxa were identified and their abundance was expressed as percentage cover of the sample surface.

Environmental factors acting locally can select different assemblages modifying the resistance to NIS invasion. In order to attribute to the mere distance and direction from the port the cause for eventual different levels of NIS invasion, the homogeneity of the structure of the native assemblage for each habitat across sites was investigated. At this aim, the structure of the macroalgal assemblage of each habitat was analysed by a 2-way Permutational Analysis of Variances (PERMANOVA, Anderson, 2001) with the factor Condition (Port, East-near, East-far, West-near, and West-far) as fixed and the factor Site (2

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