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Multiple environmental descriptors to assess ecological status of sensitive habitats in the area affected by the Costa Concordia shipwreck (Giglio Island, Italy)

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The aim of the study was to evaluate the effectiveness of the application of multiple environmental descriptors through an asymmetrical sampling design to detect possible impacts related to the Costa Concordia event on the coastal marine environment. The Costa Concordia shipwreck occurred on a submerged rocky reef in the north-western Mediterranean Sea and the wreck was removed 2 years later. To achieve the proposed objective two main coastal ecosystems, the seagrass Posidonia oceanica *and coralligenous assemblages were studied using two ecological indices, PREI and ESCA, respectively. Both indices show a lower ecological quality in the disturbed sites compared with the control ones. Differences between the disturbed and control sites observed in both studied ecosystems would seem to indicate an increase of turbidity around the shipwreck as the most plausible cause of impact. The concurrent use of different ecological indices and asymmetrical sampling designs allowed detection of differences in ecological quality of the disturbed sites compared with the controls. This approach may represent an interesting tool to be employed in impact evaluation studies.*

Keywords: *Posidonia oceanica*, coralligenous assemblages, ecological status, environmental impact, Costa Concordia shipwreck, Mediterranean Sea

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INTRODUCTION

Marine coastal systems are affected by anthropogenic pressures worldwide and in many regions they have already been significantly altered (Thrush *et al.*, 2009). In this context the evaluation of the ecological status of marine ecosystems represents a main goal for the ecologists in order to plan monitoring programmes and impact assessments focused on environment conservation (Treweek, 1999). Different stressors often interact in the same area with consequent synergistic or antagonistic effects that may cause patterns of variability of natural marine systems that are difficult to interpret (Chapman *et al.*, 1995; Steinbeck *et al.*, 2005; Borja *et al.*, 2008; Gennaro & Piazzi, 2011). Thus the assessment of environmental quality is a complex ecological problem that needs the use of suitable ecological indicators and appropriate sampling designs (Benedetti-Cecchi, 2001; Martinez-Crego *et al.*, 2010).

European strategies currently adopted for assessing and improving the quality of marine and coastal waters (European Commission 2000, 2008) require the identification of suitable bioindicators to effectively reflect environmental changes (Martinez-Crego *et al.*, 2010). Biotic indices developed by using different ecosystem parameters may be able to condense information related to multiple environmental responses to human stressors (Birk *et al.*, 2012; Personnic *et al.*, 2014). Moreover, the concurrent use of multiple descriptors may allow evaluation of synergistic effects related to different sources of disturbance more effectively than surveys utilizing single descriptors or single communities (Borja *et al.*, 2009a, b; Bedini & Piazzi, 2012).

Another major problem in impact assessment studies concerns the sampling designs. Natural assemblages are highly variable in time and in space, and sampling designs have to be suitable to separate human-caused effects from patterns of natural, temporal and spatial variability (Underwood, 1992; Hewitt *et al.*, 2001). 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 consequences of human-induced changes (Underwood, 1991, 1992, 1994; Benedetti-Cecchi, 2001). However, 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 (Chapman *et al.*, 1995; Guidetti *et al.*, 2002; De Biasi *et al.*, 2016). This approach

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utilizes an asymmetrical sampling design with multiple reference sites, in order to separate the effects of impacts from the variability among sites (Terlizzi *et al*., 2005; Fraschetti *et al*., 2006; Benedetti-Cecchi & Osio, 2007; Martin *et al*., 2012).

In this context, multiple control sites must represent comparable habitats (in terms of biological assemblages, type and slope of the substrate and exposure of waves) to those occurring at the disturbed site; this requires that they are selected in the same geographic area as the disturbed site, but far enough away as to be outside the range of influence of the source of anthropogenic disturbance being examined (Terlizzi *et al*., 2005; Benedetti-Cecchi & Osio, 2007). Control sites should also occur on both sides of the impacted area, in order to avoid spatial segregation, although in some cases this is not possible (Terlizzi *et al*., 2005; Benedetti-Cecchi & Osio, 2007; Bacci *et al*., 2016).

The aim of the present study was to evaluate the effectiveness of the application of multiple environmental descriptors through an asymmetrical sampling design to detect possible impacts related to the Costa Concordia event on certain coastal marine habitats. The Costa Concordia shipwreck occurred on a submerged rocky reef near the Giglio Island (Tuscany Archipelago National Park, Italy) in January 2012 and the wreck was removed in July 2014, after the parbuckling and refloating activities that potentially further altered the surrounding environment. Several monitoring studies have been carried out in cases of ships sinking (Nikitik & Robinson, 2003; Dimitrakakis *et al*., 2014) but the Costa Concordia shipwreck represents an unusual case in the context of the Mediterranean Sea due to the proximity of the wreck to the coast and furthermore, can represent a useful example for the definition of impact assessment biomonitoring protocols in the event of accidents with limited influence in space and time. During the months following the ship disaster, besides the presence of the wreck which could be a source of polluting substances such as fuel and paint residues, or organic pollutants deriving from galley contents, new possible potential impacts may have been introduced, since activities for the removal of the wreckage started, with the installation of anchor fixed structures and a large traffic of workforce and equipment. Two of the main coastal ecosystems of interest in the presence of the were investigated: *Posidonia oceanica* (L.) Delile meadows and coralligenous habitat (calcareous structures edified by both macroalgae and sessile invertebrates, Ballesteros, 2006 and references therein). Both ecosystems are considered among the most relevant marine coastal habitats by international legislation and conventions (UN Barcelona Convention, 1976; European Commission, 1992) and suitable indicators of anthropogenic stress (Hong, 1983; Pergent *et al*., 1995; Montefalcone, 2009; Piazzi *et al*., 2012, 2016a, b). *Posidonia* meadows and coralligenous assemblages have been studied using two different ecological indices: PREI (Gobert *et al*., 2009) and ESCA index (Cecchi *et al*., 2014; Piazzi *et al*., 2015).

MATERIALS AND METHODS

Study site

The Costa Concordia ship collided with a submerged natural rocky reef close to Giglio Island (Tuscany, Italy), which is a Protected Area of the Tuscan Archipelago National Park and is characterized by species of high ecological and biological interest in accordance with the European Directives.

The Costa Concordia wreck lay on a seabed that goes from 18 m to more than 40 m depth, oriented NE, close to the coastline. The climate condition of the study area is complex because of the particular geographic location of the island that suffers the orographic effects of nearby Corsica and the continent. The worst storms are associated with winds of Sirocco (SE) and Ostro affecting mainly the southern and eastern side of the island.

The shipwreck lay over a granitic basement and *P. oceanica* meadow, while the deeper part of the seabed was characterized by coralligenous assemblages. The shipyard for the parbuckling and refloating activities was built all around the shipwreck (Figure 1).

The *P. oceanica* meadow was completely sealed under the wreck but it was still present near the bow and the stern from about 6 to 35 m depth. Coralligenous habitat occurred on the cliff below 30 m depth.

Sampling design and data collection

Coralligenous and *P. oceanica* sampling surveys were carried out in June and July 2015 in four sites: one Disturbed site (Dp *Posidonia* sampling site and Dc coralligenous sampling site) and three Control sites (C1p, C2p, C3p *Posidonia* sampling sites and C1c, C2c, C3c coralligenous sampling sites). Disturbed sites were chosen in relation to the vicinity of the shipyard (source of potential pollution), to the presence of 'still alive' *P. oceanica* meadow and of coralligenous assemblages, and where the presence of the shipyard allowed sampling dives. The proximity to Giglio Porto harbour does not affect the study because it does not represent a source of disturbance, since it is a small marina, mainly characterized by recreational traffic restricted to summertime. The control sites were randomly chosen among those with the same biological assemblages, waves exposure and geomorphological characteristics of the disturbed sites and located a few kilometres away on both sides (North and South) of the wreck (Figure 1). For *P. oceanica* meadows, in each sampling site three areas of about 400 m^2 at 15 m depth were randomly chosen. In each area five shoot density counts were performed in square frames of 0.16 m^2 (Panayotidis *et al*., 1981), highlighting a considerable error reduction when counts were performed in at least five quadrats (Bacci *et al*., 2015). Then six orthotropic shoots were sampled and stored at -20°C, pending laboratory examination. In addition, depth and type (Meinesz & Laurent, 1978; Pergent *et al*., 1995) of the lower limit of the meadow were assessed along a transect in front of the three areas. Biotic features of shoots were gathered according to Giraud (1979) and the shoot leaf surface area was calculated. Epiphytes were scratched with a razor blade and biomass of both leaves and epiphytes was evaluated as dry weight after 48 h at 60°C.

For coralligenous assemblages, in each sampling site two areas of about 100 m^2 were randomly chosen at 30–35 m depth where communities were developed, 100 m away from each other. In each area 15 photographic samples of 0.2 m^2 were obtained by a digital camera (Nikon Coolpix 6000sc). Organisms easily identified in photographic samples were considered as taxa, while those organisms displaying similar morphological features were assembled into morphological groups (Parravicini *et al*., 2010; Cecchi *et al*.,

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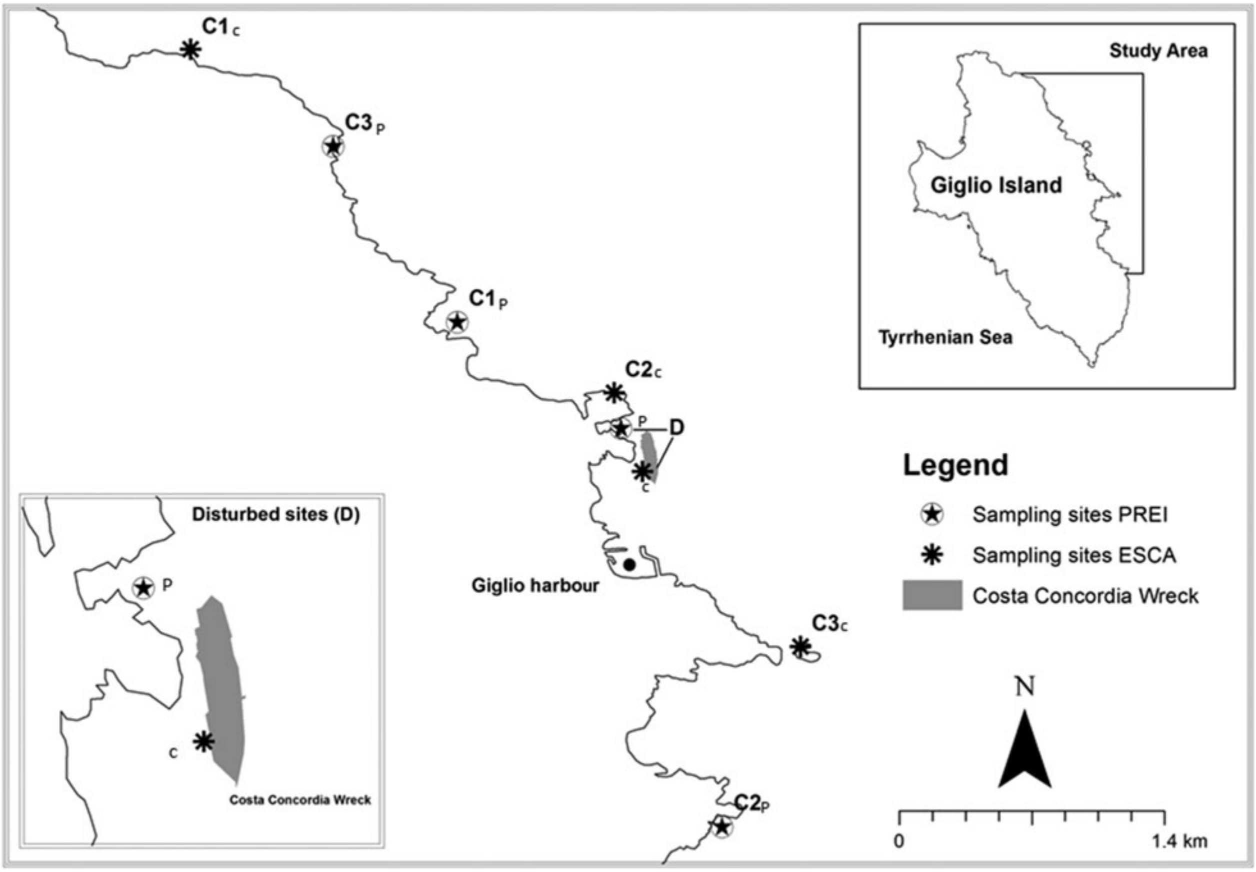

Fig. 1. Map of the study area, Dc, C1c, C2c, C3c coralligenous sampling stations; Dp, C1p, C2p, C3p *P. oceanica* sampling stations.

2014; Piazzi *et al*., 2014). The percentage cover of the main taxa/morphological groups was evaluated by ImageJ software. The sensitivity level value of each taxon/group refers to the average coverage of the taxon/group calculated among all samples of each site (Cecchi *et al*., 2014).

Ecological classification

Ecological classification of *P. oceanica* meadows was performed by PREI (*Posidonia oceanica* Rapid Easy Index, Gobert *et al*., 2009), the classification index adopted by Italy in the context of the Water Framework Directive 2000/60/CE (Italian Legislative Decree no. 152/2006). For the calculation of PREI physiographic and structural properties of the meadows were evaluated and analysed, as well as their functional and ecological features. The index includes the calculation of five descriptors: shoot density, shoot leaf surface area, E/L ratio (epiphytic biomass/leave biomass) sampled at 15 m depth; depth and type of the lower limit (progressive, erosive, sharp or regressive). Each of these metrics represents a partial component of the PREI, whose formulation integrates the components in an algorithm (for more details about PREI formulation, refer to Gobert *et al*., 2009). Reference values (referring to undisturbed conditions) have been factored into the PREI formula, in such a way that PREI already assumes the meaning of an Ecological Quality Ratio (EQR), by providing a measure of the 'distance' from those conditions considered to be 'natural'. In this regard, Reference Conditions (shoot density = 599 shoots m^{-2}; leaf surface area = 310 cm^2 $shoot^{-1}$; E/L = 0; lower limit depth = 38 m) have been modulated on the basis of the Italian national dataset (Bacci *et al*., 2013).

Ecological classification of coralligenous assemblages was performed through the ESCA index (Ecological Status of Coralligenous Assemblages, Cecchi *et al*., 2014; Piazzi *et al*., 2015). For the calculation of ESCA, three descriptors were used: (i) 'sensitivity level'(SL), based on the cover of different sensitive taxa; (ii) diversity of assemblages, expressed as 'α-diversity'; (iii) heterogeneity of assemblages, expressed as 'β-diversity'. For each study site, SL was calculated by adding all values of SL reported for all taxa/groups observed in each photographic sample (Cecchi *et al*., 2014); α-diversity was defined as the mean number of the main taxa/groups obtained in each photographic sample; β-diversity was evaluated as the mean distance of all photographic samples from centroids calculated by PERMDISP analysis (Primer 6 + PERMANOVA; Anderson, 2006; Anderson *et al*., 2006). ESCA was expressed as Ecological Quality Ratio (EQR), calculated as the mean of the three EQR_S obtained for the assemblage descriptors: $EQR = ((EQR_{SL} + EQR_{\alpha} + EQR_{\beta}) \times 3^{-1})$. Individual EQRs were calculated as the ratios between the values of SL, α-diversity and β-diversity, calculated for the study sites and the values obtained for the same descriptors in the Reference Conditions. Reference Conditions referred to Montecristo Island (Cecchi *et al*., 2014), a pristine site in the northern Tyrrhenian Sea.

The ecological quality status of *P. oceanica* meadows and coralligenous assemblages was then defined, according to European Directives, in the following five classes: high, good, moderate, poor and bad (European Commission, 2000; Gobert *et al*., 2009; Piazzi *et al*., 2015).

Data analysis

PERMANOVA analysis based on Euclidean distance of untransformed data was used as univariate test (Anderson *et al.*, 2008) in order to test any differences between disturbed and control sites for the considered multiple descriptors. Values of the PREI and ESCA indices were analysed through a one-way model PERMANOVA, with Site (4 levels) as fixed factor and partitioned into the contrast of Disturbed versus Controls (D *vs* C) and the variability among controls (Terlizzi *et al.*, 2005).

The main *P. oceanica* descriptors (shoot density, leaf surface and epiphyte/leaves biomass ratio) were analysed through a two-way model PERMANOVA, with Site (4 levels) as fixed factor and Area as random factor nested in Site. The mean square of factor Site was partitioned into two portions: the contrast of Disturbed versus Controls (D *vs* C) and the variability among controls. PERMANOVA multivariate analysis of variance (Anderson, 2001) based on Bray–Curtis resemblance matrix of untransformed data was performed to analyse the composition and structure of coralligenous assemblages.

For all statistical tests, *P* values were calculated using the Monte Carlo procedure when the number of permutations was not enough to do a test with reasonable power (Anderson & Robinson, 2003; Terlizzi *et al.*, 2005). Homogeneity of multivariate dispersions was verified with PERMDISP (Anderson, 2006) to test the robustness of PERMANOVA analysis with respect to sample dispersion (Anderson *et al.*, 2008).

Finally, the SIMPER test was used to evaluate the contribution of taxa/morphological groups that mostly contributed to significant effects on coralligenous assemblages.

Data analyses were performed using the PRIMER 6 + PERMANOVA software (Clarke & Gorley, 2006; Anderson *et al.*, 2008).

RESULTS

PREI values corresponded to High ecological status in the control sites and Good ecological status in the disturbed site, while ESCA values classified the disturbed site as in Poor ecological status and it varied between High and Good ecological status in the control sites (Table 1). With regard to the control sites, significant differences among sites were detected by PREI while no differences were highlighted by ESCA index (Table 2).

Table 1. EQR ESCA and PREI values ± SD. Dc, C1c, C2c, C3c coralligenous sampling stations; Dp, C1p, C2p, C3p *P. oceanica* sampling stations.

ESCA	EQR	Status class	PREI	EQR	Status class
Dc	0.27 ± 0.11	Poor	Dp	0.71 ± 0.03	Good
C1c	0.78 ± 0.03	Good	C1p	0.90 ± 0.01	High
C2c	0.80 ± 0.07	High	C2p	0.79 ± 0.03	High
C3c	0.78 ± 0.02	Good	C3p	0.86 ± 0.01	High

ESCA class boundaries High: ≥0.80, Good; 0.6–0.8, Moderate; 0.6–0.4, Poor: 0.4–0.2; Bad: <0.2.

PREI class boundaries High: 1–0.775, Good: 0.774–0.550, Moderate: 0.549–0.325; Poor: 0.324–0.1; Bad: <0.1.

Different types of lower limit were observed between the disturbed site (regressive) and the controls (progressive and sharp) (Table 3). *Posidonia oceanica* shoot density showed lower values in the disturbed site compared with controls, while no significant effects were observed for leaf surface and epiphyte/leaves biomass ratio (Table 4). Differences among controls were not significant for any descriptor investigated in the analysis (Table 4).

In the control sites, coralligenous assemblages were dominated by encrusting Corallinales and algal turf while erect Rhodophyta were also locally abundant; *Halimeda tuna*, *Flabellia petiolata*, *Peyssonnelia* spp., were widespread with low per cent cover (Table 5). Among the macro-invertebrates, Porifera, erect Bryozoa, *Eunicella cavolini* and locally *Paramuricea clavata* were the most abundant taxa/groups (Table 5).

The composition and structure of coralligenous assemblages differed significantly between the disturbed and control sites, while differences among controls were not significant (Table 6). The SIMPER test showed that differences were mostly related to a higher abundance of algal turfs in the disturbed site and a higher abundance of Udoteaceae (*Halimeda tuna* (J. Ellis & Solander) J.V. Lamouroux and *Flabellia petiolata* (Turra) Nizamuddin) and erect Rhodophyta and *Eunicella cavolini*, in the controls (Table 7). Both alpha and beta diversity showed lower values in the disturbed site compared with controls (Table 8).

DISCUSSION

Although with different responses in terms of ecological classification, both indices detected a lower ecological quality in the disturbed sites compared with the control ones. This finding attests the sensitivity of both *P. oceanica* and coralligenous habitats to human impacts and their suitability to be used as ecological indicators in cases of both diffuse and local pressure (Gobert *et al.*, 2009; Bacci *et al.*, 2013; Cecchi *et al.*, 2014).

In *P. oceanica* meadows, shoot density and the lower limit type were the most sensitive descriptors to the studied disturbance, confirming their effectiveness to be used in impact evaluation studies (Pergent *et al.*, 1995). Shoot leaf surface area, instead, did not differ among sites, reflecting previous results referring to the study area reported in Bacci *et al.* (2016). Epiphyte biomass also did not show differences between disturbed and control sites. However, significant differences in epiphytic community structure were detected in previous investigations (Bacci *et al.*, 2016). The structure of the epiphytic community of *P. oceanica* leaves, in fact, could be an indicator of multiple impacts while epiphyte biomass is more sensitive to strong nutrient enrichment (Piazzi *et al.*, 2016a).

In coralligenous habitats, high abundance of algal turf was consistent in the disturbed site, thus differentiating from the controls, where instead erect macroalgae and *Eunicella cavolini* (Koch, 1887) were dominant. Hence, although the aim of the study was not to identify or measure each temporal pressure acting on the disturbed site, the structure of coralligenous assemblages observed in this area seemed to indicate a kind of local impact, due to nutrient enrichment or sediment increasing, as reported in the literature (Balata *et al.*, 2005, 2007a, b; Piazzi *et al.*, 2011, 2012). Results of other studies

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Table 2. PERMANOVA on values of PREI and ESCA.

Source	df	PREI			ESCA		
		MS	Pseudo-F	P(MC)	MS	Pseudo-F	P(MC)
Site = S	3	0.022	14.615	**0.002**	0.1331	13.794	**0.011**
D *vs* C	1	0.045	13.945	**0.004**	0.3984	60.815	**0.002**
Among C	2	0.010	10.593	**0.012**	0.0004	0.08	0.926
Residual	4	0.001			0.0096		

D, disturbed site; C, control sites. Significant differences are in bold.

Table 3. Descriptors of *Posidonia oceanica* meadows (mean ± SD).

Descriptors	Dp	C1p	C2p	C3p
Shoot density (shoot m^{-2})	308.8 ± 67.3	414.5 ± 104.7	452.1 ± 116.1	411.6 ± 122.7
Shoot surface (cm^2 $shoot^{-1}$)	276.8 ± 88.6	358.5 ± 67.1	259.7 ± 64.1	299.2 ± 99.4
Lower limit depth (m)	28	32	32	28
Epiphytic biomass (mg $shoot^{-1}$)	152.1 ± 94	106.8 ± 56.3	75.1 ± 49.3	185.1 ± 202.4
Leave biomass (mg $shoot^{-1}$)	1545.4 ± 482.5	2060.5 ± 403.3	1063.4 ± 494.4	1400 ± 579.4
Lower limit type (λ)	Regressive	Progressive	Progressive	Sharp

Dp, disturbed site; C1p-2p-3p, control sites.

Table 4. PERMANOVA on *Posidonia oceanica* descriptors.

Source	df	Shoot density			Leaf surface			Epiphyte/leaves ratio		
		MS	Ps-F	P(MC)	MS	Ps-F	P(MC)	MS	Ps-F	P(MC)
Site = S	3	62,766	4.73	**0.041**	33,400	2.12	0.187	0.003	0.61	0.618
D *vs* Cs	1	173,060	10.40	**0.021**	11,345	0.73	0.398	0.001	0.24	0.625
Among Cs	2	7620	0.48	0.611	44,427	2.89	0.138	0.003	0.61	0.586
Area(S) = A(S)	8	13,251	1.24	0.316	15,704	3.03	0.008	0.004	2.61	**0.025**
A(S)(D)	2	5408	1.23	0.300	16,801	2.51	0.105	0.000	0.05	0.955
A(S)(C)	6	15,865	1.24	0.329	15,338	3.27	**0.004**	0.006	3.44	**0.021**
Residual	48	10,640			5179			0.002		

D, disturbed site; C, control sites. Significant differences are in bold.

carried out in the context of the Costa Concordia shipwreck excluded serious contamination events or increases in environmental pollution, also due to nutrient enrichment (Regoli *et al*., 2014). Conversely, significant increase of turbidity due to huge, although temporal, sediment release events was recorded along the entire water column (from the surface to 50 m depth) of the impacted area at different times during the Costa Concordia salvage activities (Casoli *et al*., 2017). Moreover, patches of debris and sediments were found to have affected both the shallower and deeper sea bottom, with consequent stress for coralligenous habitats (Casoli *et al*., 2017). Therefore it was reasonable to think that some of the negative effects observed in our study on coralligenous assemblages were linked to sediment and debris releases that occurred during the shipwreck removal activities. Turfs are mostly constituted by filamentous species that reproduce asexually and are well adapted to stressed environmental conditions thanks to their ability to quickly recover after disturbance (Airoldi, 2003; Balata *et al*., 2011). On the contrary, in stressed conditions, erect macroalgae and invertebrates reproducing sexually are damaged directly by physical stress, such as high sedimentation rates, and indirectly because they are outcompeted by turfs (Balata *et al*., 2011).

Both the shipwreck, and parbuckling and refloating activities may cause different kinds of impact and it is difficult to determine those that mostly have affected the disturbed site. However, also in accordance with Casoli *et al*. (2017), the increase of sedimentation and debris deposition, due to the leakage of fine particles of cement filling the grout bags on which the wreck was laid during the parbukling phase, could represent the main pressure determining differences observed between control and disturbed sites. To confirm this, both the increase of turf and the decrease of alpha and beta diversity in coralligenous assemblages, as well as the decrease of shoot density of *P. oceanica* meadows, can be related to high levels of sediment load (Manzanera *et al*., 1995; Terrados *et al*., 1998; Balata *et al*., 2005, 2011; Piazzi *et al*., 2012). An increase of sediment resuspension was observed by the authors in the area of the shipwreck (~1 km^2). The regressive lower limit at a high depth, with the presence of dead *matte*, may suggest recent damage, ascribable to the Costa Concordia event. As also discussed in Bacci *et al*. (2016), more concurrent causes may have led to the differences observed.

Past and present synergistic effects among factors, associated with the structure of the wreck itself, and those related to the removal yard, could have affected the ecological quality status of the area. In this regard, the wreck and the shipyard could have acted as a physical barrier to the natural hydrodynamics of the area, also changing the

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Table 5. The mean per cent cover of the taxa/groups characterizing coralligenous assemblages.

TAXA/GROUPS	Dc	C1c	C2c	C3c
Macroalgae				
Algal turf	62.61	6.21	44.57	9.70
Encrusting Corallinales	36.12	46.71	35.41	72.06
Peyssonnelia spp.	0.75	2.28	2.80	8.88
Erect Rhodophyta	0.07	32.23	8.92	1.02
Halimeda tuna (J. Ellis & Solander) J.V. Lamouroux	0.02	2.21	0.51	0.22
Flabellia petiolata (Turra) Nizamuddin	0.03	1.98	2.88	2.61
Palmophyllum crassum (Naccari) Rabenhorst	0.02	0.20	1.35	1.07
Pseudochlorodesmis furcellata (Zanardini) Børgesen	0.01	1.12	1.52	1.88
Zanardinia typum (Nardo) G. Furnari	0.01	0.06	0.03	0.08
Erect Ochrophyta	0.01	1.08	0.01	0.01
Dictyotales	0.01	0.01	0.28	0.03
Macro-invertebrates				
Porifera	0.11	2.04	1.25	1.43
Parazoanthus axinellae (Schmidt, 1862)	0.01	0.01	0.13	0.09
Leptopsammia pruvoti (Lacaze-Duthiers, 1897)	0.01	0.03	0.02	0.13
Corallium rubrum (Linnaeus, 1758)	0.01	0.09	0.01	0.01
Eunicella cavolini (Koch, 1887)	0.05	0.16	0.23	0.49
Paramuricea clavata (Risso, 1826)	0.01	2.05	0.01	0.01
Serpulids	0.13	0.02	0.05	0.05
Thin ramified Bryozoa	0.10	0.20	0.03	0.05
Erect Bryozoa	0.14	1.43	0.21	0.37
Encrusting Bryozoa	0.01	0.12	0.06	0.04

Table 6. PERMANOVA on species composition and abundance of coralligenous assemblages.

Source	df	MS	Ps-F	P(MC)
Site = S	3	28,370	3.9	**0.027**
D *vs* Cs	1	50,525	4.7	**0.031**
Among Cs	2	17,293	3.1	0.095
Area(S) = A(S)	4	7221	10.7	**0.001**
A(S)(D)	1	11,834	61.1	**0.001**
A(S)(C)	3	5684	6.8	**0.001**
Residual	112	672		

D, disturbed site; C, control sites. Significant differences are in bold.

Table 7. SIMPER test on coralligenous assemblages.

Taxa-groups	Disturbed Mean per cent cover	Controls Mean per cent cover	Contribution (%)
Algal turf	68.6	16.59	65.43
Erect Rhodophyta	0.06	14.29	13.79
Udoteaceae	0.04	3.45	6.4
Eunicella cavolinii	0.05	0.29	4.66

Table 8. PERMANOVA on alpha and beta diversity of coralligenous assemblages.

Source	df	Alpha diversity			Beta diversity		
		MS	Ps-F	P(MC)	MS	Ps-F	P(MC)
Site = S	3	0.14	6.90	**0.04**	0.21	29.05	**0.004**
D *vs* Cs	1	0.35	13.09	**0.01**	0.41	9.76	**0.028**
Among Cs	2	0.04	3.27	0.16	0.11	15.36	**0.030**
Residual	4	0.02			0.007		

D, disturbed site; C, control sites. Significant differences are in bold.

submerged landscape of the disturbed site with for example the shadow projected by the wreck on the seabed.

Both PREI and ESCA indices showed significant differences between disturbed and control sites, highlighting their effectiveness in detecting different kinds of human pressures. However, an important difference was detected between the response of ESCA and PREI, as ESCA scores differed sharply (i.e. Poor *vs* High/Good) between disturbed sites and controls compared with the PREI classification. All the three ESCA descriptors showed lower values in disturbed sites than in control ones, confirming the sensitivity of the ESCA index to stress induced by local impacts; on the contrary, the PREI descriptors showed variable responses to the same disturbance, thus appearing less sensitive to the impact of the Costa Concordia event on the meadow.

These findings highlighted the importance of testing the validity and applicability of biological indices in the context of situations and pressures that are different from those originally used for their development and calibration (Diaz *et al*., 2004; Borja *et al*., 2009b), as not all indicators adequately respond to different contexts.

Coralligenous assemblages are particularly sensitive to anthropogenic pressure acting on coastal areas, since they are constituted by organisms, both macroalgae and macro-invertebrates, adapted to spread in stable physical conditions, thus highly sensitive to most anthropogenic causes of stress and disturbance (Montefalcone *et al*., 2017). The lower effectiveness of PREI could be related to the type of pressure, which has produced a spatially limited direct damage on the meadow, especially at high bathymetry (lower limits). In addition, *P. oceanica* meadows, despite their effectiveness as indicators of water quality (Gobert *et al*., 2009; Lopez y Rojo *et al*., 2010), normally have higher times of responses and low resilience than macro-invertebrates and macroalgae (Balata *et al*., 2010) The better response of ESCA index could also be explained by the closer proximity of the disturbed site to the pressure.

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The different response of the two indices to the same pressure highlights the importance of using multiple biological descriptors in monitoring programmes and impact evaluation studies. This finding confirms previous studies, suggesting that the use of data obtained from different biological systems represents the most promising approach for assessing the ecological status of coastal waters (Martinez-Crego *et al.*, 2010; Bedini & Piazzi, 2012). Moreover, only some *P. oceanica* variables responded to changes in environmental conditions. Thus, the use of appropriate bioindicators should be coupled with that of appropriate descriptors, in order to assess the status of coastal waters.

In conclusion, ecological indices, usually employed in environmental monitoring programmes, could be used in synergy to describe marine ecosystem impacts due to local pressures. The concurrent use of different ecological indices and an asymmetrical sampling design is recommended to detect differences in ecological quality of the disturbed site compared with controls.

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REFERENCES

Airoldi L. (2003) The effects of sedimentation on rocky coastal assemblages. *Oceanography and Marine Biology: An Annual Review* 41, 161–203.

Anderson M.J. (2001) A new method for non-parametric multivariate analysis of variance. *Austral Ecology* 26, 32–46.

Anderson M.J. (2006) Distance-based test for homogeneity of multivariate dispersions. *Biometrics* 62, 245–253.

Anderson M.J., Ellingsen K.E. and McArdle B.H. (2006) Multivariate dispersion as a measure of beta diversity. *Ecology Letters* 9, 683–693.

Anderson M.J., Gorley R.N. and Clarke K.R. (2008) *PERMANOVA+ for PRIMER: guide to software and statistical methods.* Plymouth: PRIMER-E.

Anderson M.J. and Robinson J. (2003) Generalized discriminant analysis based on distance. *Australia and New Zealand Journal of Statistics* 45, 301–318.

Bacci T., Penna M., Rende S.F., Trabucco B., Gennaro P., Bertasi F., Marusso V., Grossi L. and Cicero A.M. (2016) Effects of Costa Concordia event on epiphytic assemblages and biotic features of *Posidonia oceanica* canopy. *Marine Pollution Bulletin* 109, 110–116.

Bacci T., Rende S.F., Penna M., Trabucco B., Montefalcone M., Cicero A. M. and Giovanardi F. (2013) A methodological approach to understand functional relationships between ecological indices and human-induced pressures: the case of the *Posidonia oceanica* meadows. *Journal of Environmental Management* 129, 540–547.

Bacci T., Rende S.F., Rocca D., Scalise S., Cappa P. and Scardi M. (2015) Optimizing *Posidonia oceanica* (L.) Delile shoot density: lessons learned from a shallow meadow. *Ecological Indicators* 58, 199–206.

Balata D., Piazzi L. and Benedetti-Cecchi L. (2007a) Sediment disturbance and loss of beta diversity on subtidal rocky reefs. *Ecology* 8, 2455–2461.

Balata D., Piazzi L., Cecchi E. and Cinelli F. (2005) Variability in Mediterranean coralligenous assemblages subject to local variation in turbidity and sediment deposits. *Marine Environmental Research* 60, 403–421.

Balata D., Piazzi L. and Cinelli F. (2007b) Increase of sedimentation in a subtidal system: effects on the structure and diversity of macroalgal assemblages. *Journal of Experimental Marine Biology and Ecology* 351, 73–82.

Balata D., Piazzi L., Nesti U., Bulleri F. and Bertocci I. (2010) Effects of enhanced loads of nutrients on epiphytes on leaves and rhizomes of *Posidonia oceanica. Journal of Sea Research* 63, 173–179.

Balata D., Piazzi L. and Rindi F. (2011) Testing a new classification of morphological functional groups of marine macroalgae for the detection or responses to disturbance. *Marine Biology* 158, 2459–2469.

Ballesteros E. (2006) Mediterranean coralligenous assemblages: a synthesis of present knowledge. *Oceanography and Marine Biology: An Annual Review* 44, 123–195.

Bedini R. and Piazzi L. (2012) Evaluation of the concurrent use of multiple descriptors to detect anthropogenic impacts in marine coastal systems. *Marine Biology Research* 8, 129–140.

Benedetti-Cecchi L. (2001) Beyond BACI: optimization of environmental sampling designs through monitoring and simulation. *Ecological Application* 11, 783–799.

Benedetti-Cecchi L. and Osio G.C. (2007) Replication and mitigation of effects of confounding variables in environmental impact assessment: effects of marinas on rocky shore assemblages. *Marine Ecology Progress Series* 334, 21–35.

Birk S., Bonne W., Borja A., Brucet S., Courrat A., Poikane S., Solimini A., van de Bund W., Zampoukas N. and Hering D. (2012) Three hundred ways to assess Europe's surface waters: an almost complete overview of biological methods to implement the Water Framework Directive. *Ecological Indicators* 18, 31–41.

Borja A., Bald J., Franco J., Larreta J., Muxika I., Revilla M., Rodríguez J.G., Solaun O., Uriarte A. and Valencia V. (2009a) Using multiple ecosystem components, in assessing ecological status in Spanish (Basque country) Atlantic marine waters. *Marine Pollution Bulletin* 59, 54–64.

Borja A., Bricker S.B., Dauer D.M., Demetriades D.N., Ferreira J.G., Forbes A.T., Hutching P., Jia X., Kenchington R., Marques J.C. and Zhu C. (2008) Overview of integrative tools and methods in assessing ecological integrity in estuarine and coastal systems worldwide. *Marine Pollution Bulletin* 56, 1519–1537.

Borja A., Ranasinghe A. and Weiseberg S.B. (2009b) Assessing ecological integrity in marine waters, using multiple indices and ecosystem components: challenges for the future. *Marine Pollution Bulletin* 59, 1–4.

Casoli E., Ventura D., Cutroneo L., Capello M., Jona-Lasinio G., Rinaldi R., Criscoli A., Belluscio A. and Ardizzone G.D. (2017) Assessment of the impact of salvaging the Costa Concordia wreck on the deep coralligenous habitats. *Ecological Indicators* 80, 124–134.

Cecchi E., Gennaro P., Piazzi L., Ricevuto E. and Serena F. (2014) Development of a new biotic index for ecological status assessment of Italian coastal waters based on coralligenous macroalgal assemblages. *European Journal of Phycology* 49, 298–312.

Chapman M.G., Underwood A.J. and Skilleter G.A. (1995) Variability at different spatial scales between a subtidal assemblage exposed to the discharge of sewage and two control assemblages. *Journal of Experimental Marine Biology and Ecology* 189, 103–122.

Clarke K.R. and Gorley R.N. (2006) *Primer v6: user manual/tutorial.* Plymouth: PRIMER-E.

De Biasi M., Pacciardi L. and Piazzi L. (2016) An asymmetrical sampling design as a tool for sustainability assessment of human activities in coastal systems: a fish farming study case. *Marine Biology Research* 12, 958–968. doi: 10.1080/17451000.2016.1225958.

Diaz R.J., Solan M. and Valente R.M. (2004) A review of approaches for classifying benthic habitats and evaluating habitat quality. *Journal of Environmental Management* 73, 165–181.

Dimitrakakis E., Hahladakis J. and Gidarakos E. (2014) The 'Sea Diamond' shipwreck: environmental impact assessment in the water column and sediments of the wreck area. *International Journal of Environmental Science and Technology* 11, 1421–1432.

European Commission (1992) Council Directive 92/43/EEC (Habitat Directive) of 21 May 1992 on the conservation of natural habitats and of wild fauna and flora. As amended by the Accession Act of Austria, Finland and Sweden. *Official Journal of the European Commission* L 1, 1.1, 135. EC.

European Commission (2000) Water Framework Directive 2000/60/EC of the European parliament and of the council, of 23 October 2000, establishing a framework for Community action in the field of water policy. *Official Journal of the European Commission, 22/12/2000*, L 327.

European Commission (2008) Marine Strategy Framework Directive 2008/56/EC of the European Parliament and of the Council, of 17 June 2008, establishing a framework for Community action in the field of marine environmental policy (Marine Strategy Framework Directive). *Official Journal of the European Commission, 25/6/2008*, L 164, 19.

Fraschetti S., Gambi C., Giangrande A., Terlizzi A. and Danovaro R. (2006) Structural and functional response of meiofauna rocky assemblages to sewage pollution. *Marine Pollution Bulletin* 52, 540–548.

Gennaro P. and Piazzi L. (2011) Synergism between two anthropic impacts: invasion of macroalga *Caulerpa racemosa* var. *cylindracea* and seawater nutrient enrichment. *Marine Ecology Progress Series* 427, 59–70.

Giraud C. (1979) Sur une méthode de mesure et de comptage des structures foliaires de *Posidonia oceanica* (Linnaeus) Delile. *Bullettin du Muséum d'Histoire Naturalle de Marseille* 39, 33–39.

Gobert S., Sartoretto S., Rico-Raimondino V., Andral B., Chery A., Lejeune P. and Boissery P. (2009) Assessment of the ecological status of Mediterranean French coastal waters as required by the Water Framework Directive using the *Posidonia oceanica* Rapid Easy Index (PREI). *Marine Pollution Bulletin* 58, 1727–1733.

Guidetti P., Fanelli G., Fraschetti S., Terlizzi A. and Boero F. (2002) Coastal fish indicate human-induced changes in the Mediterranean littoral. *Marine Environmental Research* 53, 77–94.

Hewitt J.E., Thrush S.F. and Cummings V.J. (2001) Assessing environmental impacts: effects of spatial and temporal variability at likely impact scales. *Ecological Application* 11, 1502–1516.

Hong J.S. (1983) Impact of pollution on the benthic community: environmental impact of the pollution on the benthic coralligenous community in the Gulf of Fos, north-western Mediterranean. *Bulletin of Korean Fishery Society* 16, 273–290.

Lopez y Rojo C., Casazza G., Pergent-Martini C. and Pergent G. (2010) A biotic index using the seagrass *Posidonia oceanica* (BiPo), to evaluate ecological status of coastal waters. *Ecological Indicators* 10, 380–389.

Manzanera M., Perez M. and Romero J. (1995) Seagrass mortality due to oversedimentation: an experimental approach. In *Proceedings of the Second International Conference on the Mediterranean Coastal Environment, MEDCOAST 95, 24–27 October 1995*. Taragona, Spain.

Martin C.J.B., Allen B.J. and Lowe C.G. (2012) Environmental impact assessment: detecting changes in fish community structure in response to disturbance with an asymmetric multivariate BACI sampling design. *Bulletin of Southern California Academy of Sciences* 11, 119–131.

Martinez-Crego B., Alcoverro T. and Romero J. (2010) Monitoring the quality of coastal waters at a large scale: bioindicators strengths and weakness. *Journal of Environmental Monitoring* 12, 1013–1028.

Meinesz A. and Laurent R. (1978) Cartographie et état de la limite inférieure de l'herbier de *Posidonia oceanica* dans les Alpes-Maritimes. Campagne Poséïdon 1976. *Botanica Marina* 21, 513–526.

Montefalcone M. (2009) Ecosystem health assessment using the Mediterranean seagrass *Posidonia oceanica*: a review. *Ecological Indicators* 9, 595–604.

Montefalcone M., Morri C., Bianchi C.N., Bavestrello G. and Piazzi L. (2017) The two facets of species sensitivity: stress and disturbance on coralligenous assemblages in space and time. *Marine Pollution Bulletin* 117, 229–238.

Nikitik C.C.S. and Robinson A.W. (2003) Patterns in benthic populations in the Milford Haven waterway following the 'Sea Empress' oil spill with special reference to amphipods. *Marine Pollution Bulletin* 46, 1125–1141.

Panayotidis P., Bouderesque C.F. and Marcot-Coqueugniot J. (1981) Microstructure de l'herbier à *Posidonia oceanica* (Linnaeus) Delile. *Botanica Marina* 24, 115–124.

Parravicini V., Micheli F., Montefalcone M., Villa E., Morri C. and Bianchi C.N. (2010) Rapid assessment of benthic communities: a comparison between two visual sampling techniques. *Journal of Experimental Marine Biology and Ecology* 395, 21–29.

Pergent G., Pergent-Martini C. and Boudouresque C.F. (1995) Utilisation de l'herbier à *Posidonia oceanica* comme indicateur biologique de la qualité du milieu littoral en Méditerranée: état de connaissances. *Mésogée* 54, 3–29.

Personnic S., Boudouresque C.F., Astruch P., Ballesteros E., Blouet S., Bellan-Santini D., Bonhomme P., Thibault-Botha D., Feunteun E., Harmelin-Vivien M., Pergent G., Pergent-Martini C., Pastor J., Poggiale J.C., Renaud F., Thibaut T. and Ruitton S. (2014) An ecosystem-based approach to assess the status of a Mediterranean ecosystem, the *Posidonia oceanica* seagrass meadow. *PLoS ONE* 9, e98994.

Piazzi L., Balata D. and Ceccherelli G. (2016a) Epiphyte assemblages of the Mediterranean seagrass *Posidonia oceanica*: an overview. *Marine Ecology* 37, 3–41.

Piazzi L., Cecchi E., Serena F., Guala I., Canovas Molina A., Gatti G., Morri C., Bianchi C.N. and Montefalcone M. (2014) Visual and photographic methods to estimate the quality of coralligenous reefs under different human pressures. In Bouafif C., Langar H. and Ouerghi A. (eds) *Proceedings of the second Mediterranean Symposium on the Conservation of Coralligenous and other Calcareous Bio-Concretions, Portorož, Slovenia, 29–30 October 2014*. UNEP/MAP-RAC/SPA, RAC/SPA publ, Tunis, pp. 135–140.

Piazzi L., Gennaro P. and Balata D. (2011) Effects of nutrient enrichment on macroalgal coralligenous assemblages. *Marine Pollution Bulletin* 62, 1830–1835.

Piazzi L., Gennaro P. and Balata D. (2012) Threats to macroalgal coralligenous assemblages in the Mediterranean Sea. *Marine Pollution Bulletin* 64, 2623–2629.

Piazzi L., Gennaro P., Cecchi E. and Serena F. (2015) Improvement of the ESCA index for the evaluation of ecological quality of coralligenous habitat under the European Framework Directives. *Mediterranean Marine Science* 16, 419–426.

Piazzi L., La Manna G., Cecchi E., Serena F. and Ceccherelli G. (2016b) Protection changes the relevancy of scales of variability in coralligenous assemblages. *Estuarine, Coastal and Shelf Science* 175, 62–69.

Regoli F., Pellegrini D., Cicero A.M., Benedetti M., Gorbi S., Fattorini D., D'Errico G., Di Carlo M., Nardi A., Gaion A., Scuderi A., Giuliani S., Romanelli G., Berto D., Trabucco B., Guidi P., Bernardeschi M., Scarcelli V. and Frenzilli G. (2014) A multidisciplinary weight of evidence approach for environmental risk assessment at the Costa Concordia wreck: integrative indices from Mussel Watch. *Marine Environmental Research* 96, 92–104.

Steinbeck J.R., Schiel D.R. and Foster M.S. (2005) Detecting long-term change in complex communities: a case study from the rocky intertidal zone. *Ecological Applications* 15, 1813–1832.

Terlizzi A., Benedetti-Cecchi L., Bevilacqua S., Fraschetti S., Guidetti P. and Anderson M.J. (2005) Multivariate and univariate asymmetrical analyses in environmental impact assessment: a case study of Mediterranean subtidal sessile assemblages. *Marine Ecology Progress Series* 289, 27–42.

Terrados J., Duarte C.M., Fortes M.D., Borum J., Agawin N.S.R., Bach S., Thampanya U., Kamp-Nielsen L., Kenworthy W.J., Geertz-Hansen O. and Vermaat J. (1998) Changes in community structure and biomass of seagrass communities along gradients of siltation in SE Asia. *Estuarine Coastal and Shelf Science* 46, 757–768.

Thrush S.F., Hewitt J.E., Dayton P.K., Coco G., Lohrer A.M., Norkko A., Norkko J. and Chiantore M. (2009) Forecasting the limits of resilience: integrating empirical research with theory. *Proceedings of the Royal Society* B 276, 3209–3217.

Treweek J. (1999) *Ecological impact assessment*. Oxford: Blackwell Science.

Underwood A.J. (1991) Beyond BACI: experimental designs for detecting human environmental impacts on temporal variations in natural populations. *Australian Journal of Marine and Freshwater Resources* 42, 569–587.

Underwood A.J. (1992) Beyond BACI: the detection of environmental impacts on populations in the real, but variable, world. *Journal of Experimental Marine Biology and Ecology* 161, 145–178.

Underwood A.J. (1994) On beyond BACI: sampling designs that might reliably detect environmental disturbances. *Ecological Application* 4, 3–15.

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United Nations (1976) Barcelona Convention for the Protection of the Mediterranean Sea against Pollution. 1102 UNTS 27.

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