Journal of Sea Research 79 (2013) 20–26

Contents lists available at SciVerse ScienceDirect

Journal of Sea Research

journal homepage: www.elsevier.com/locate/seares

The effects of protection measures on fish assemblage in the Plemmirio marine reserve (Central Mediterranean Sea, Italy): A first assessment 5 years after its establishment

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ARTICLE INFO

Article history:
Received 6 April 2012
Received in revised form 23 December 2012
Accepted 28 January 2013
Available online 5 February 2013

Keywords:
MPA
Reserve
UVC
Fish assemblage
Mediterranean Sea
Protection

ABSTRACT

This 2-year study was aimed to investigate the early effects of protection measures on fish assemblage in the Plemmirio marine reserve and to evaluate its level of enforcement. Sampling was carried out by means of underwater visual census techniques in four sampling sites within the reserve boundaries and eight outside the reserve. Results showed significant inside/outside differences in the multivariate abundance of fish assemblage. These results were confirmed and exemplified by significant univariate differences between locations for total abundance, Species Richness and diversity of the fish assemblage; values of these metrics were higher inside the reserve than outside. Small fish size and species of low and medium fishing value did not display significant inside/outside differences in abundances whereas medium, large size fish and high value species showed abundances significantly higher inside the marine reserve. Protection effects were particularly evident for large specimens of high fishing value, most of which were exclusively found inside the reserve (*Diplodus puntazzo*, *Epinephelus costae*, *Mycteroperca rubra*, *Scorpaena scrofa*, *Spondyliosoma cantharus*, *Sciaena umbra* and *Epinephelus marginatus*). The present study provides evidence of a reserve effect on fish populations after only five years since its establishment. This is an extraordinary result likely due to the high level of enforcement observed inside the Plemmirio MPA.

1. Introduction

Marine protected areas (MPAs) refer to portions of the coastline and/or sea where human activities, especially fishing, are restricted or banned (Agardy et al., 2003). This form of spatial management has been advocated as a solution to many important and pressing problems within the marine environment (Dayton et al., 2000; Gell and Roberts, 2002), such as loss of marine biodiversity (Jackson et al., 2001), alteration of trophic structures (Babcock et al., 1999; Castilla, 1999; Jackson et al., 2001; Pauly et al., 1998, 2002), loss of habitat (Sumaila et al., 2000) and chronic over-fishing (Hutchings, 2000; Jackson et al., 2001; Pauly et al., 1998, 2002). At the same time, MPAs may bring social and economic benefits through enhanced tourism (Dayton et al., 2000; Gell and Roberts, 2002).

In particular, protection from fishing may directly restore populations of target fishes and indirectly drive whole communities towards an unfished state (Bevilacqua et al., 2006; Guidetti, 2006; Micheli et al., 2004; Sala et al., 1998; Shears and Babcock, 2002), especially in MPAs having no-take reserves (Dayton et al., 1995; McClanahan et al., 2007; Micheli et al., 2004) that are places where all forms of extraction, particularly fishing, are banned permanently (Dayton et al., 2000; Gell and Roberts, 2002; Roberts and Polunin, 1991).

The evaluation of these benefits, in terms of increase in density and size of target fish species (Claudet et al., 2006; Côté et al., 2001; Guidetti et al., 2008; Halpern, 2003; Micheli et al., 2004; Mosquera et al., 2000), can be useful to assess the ecological effectiveness of reserves (Guidetti et al., 2008).

MPAs are also predicted to benefit adjacent fisheries through two mechanisms: net emigration of adults and juveniles across borders, termed ‘spillover’ (Rowley, 1994), and export of pelagic eggs and larvae. Inside reserves, populations increase in size, individuals live longer, grow larger and develop an increased reproductive potential (Bohnsack, 1998). Enhanced production of eggs and larvae inside a reserve is predicted to lead to net export and increased settlement of juvenile animals outside the boundaries (Alcala et al., 2005; Gell and Roberts, 2002; Roberts and Polunin, 1991).

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http://dx.doi.org/10.1016/j.seares.2013.01.004

In the Mediterranean Sea there has been a rush, over the last 15 years, to establish MPAs and reserves (Juanes, 2001). In Italy, specifically, there are currently 30 MPAs formally established and these include one or more no-take/no-access zones (hereafter called 'reserves' in the text and formally defined as 'A zones' according to Italian law), surrounded by buffer zones (defined as 'B and C zones', where restrictions to human uses, including fishing, become progressively more lax) (Guidetti et al., 2008).

One of the youngest MPAs in Italy is the "Plemmirio", located on the eastern coast of Sicily and established in 2004. This MPA has been included in the SPAMI (Specially Protected Areas of Mediterranean Importance) list since 2008 (decision UNEP/MAP, Athens, 2007) due to its importance for conserving the components of biological diversity in the Mediterranean area. A special interest at the scientific, aesthetic, cultural and educational level has been recognized by UNEP and this represents an added value to the MPA. Because of its recent implementation, studies on fish assemblage of the Plemmirio MPA are sparse, therefore it is not known if this MPA meets its potential ecological objectives, nor if its protection occurs only "on paper" (i.e. ineffective enforcement). The present study, the first undertaken in the Plemmirio MPA, was aimed to assess the level of enforcement of this young reserve and the effects of protection on the fish assemblage. In order to do this, we hypothesized that community metrics were significantly higher inside the marine reserve than outside.

2. Materials and methods

2.1. Study sites

Located on the eastern coast of Sicily (Central Mediterranean Sea), the Plemmirio Marine Protected Area was established in 2004 and protects about 2400 ha of marine territory (Fig. 1). Its primary aim is to protect marine biodiversity, to favour social and economic activities linked to the sea, especially fisheries, and to promote public education and scientific research.

2.2. Visual censuses

Fish species and their abundance and size were recorded on standardized sheets by underwater visual censuses using SCUBA diving on rocky substrates by means of 25×5 m transects parallel to the coast (surveyed area $= 125$ m^2). Underwater visual census (UVC) monitoring techniques provide qualitative and quantitative surveys with a limited impact on the ecosystem, and are therefore particularly suited for marine reserves (Harmelin et al., 1995). Divers swam one way for 5–7 min along each transect, identifying and recording the number and size of the observed fishes. Fish density was estimated by counting single specimens to a maximum of ten individuals, whereas classes of abundance (11–30, 31–50, 51–100, 101–200, 201–500, >500 individuals) were used for larger schools (Guidetti et al., 2004). Fish size was assessed by classifying fishes within three size categories (i.e. small, medium, large) on the basis of the maximum total length attained by each species (Whitehead et al., 1984–1986). All fish seen were recorded but highly gregarious species (*Sardinella aurita*, *Spicara* spp., *Boops boops* and *Chromis chromis*) were excluded from the analyses. Early juvenile stages (settlers and recruits) were not taken into account. All surveys were done on the rocky bottoms because in coastal areas these are where fishing pressure is greatest (Francour, 1994), and where, if a reserve effect exists, it can be easily detected (Harmelin et al., 1995). Habitat structure is one of the factors to be invoked to explain the small-scale spatial variability of Mediterranean fish assemblages (García-Charton and Pérez-Ruzafa, 2001; García-Charton et al., 2000, 2004) and may mask the effect of protection if protected areas present simpler habitats than non-protected ones (García-Charton et al., 2004). Accordingly, all surveyed areas had similar substrata topographic complexity (rocky substrata with scattered boulders), benthic community and a gentle slope. Moreover, as fish distribution is depth dependent (Bell, 1983; Lipej et al., 2003), all transects were conducted at depths between 15 and 20 m.

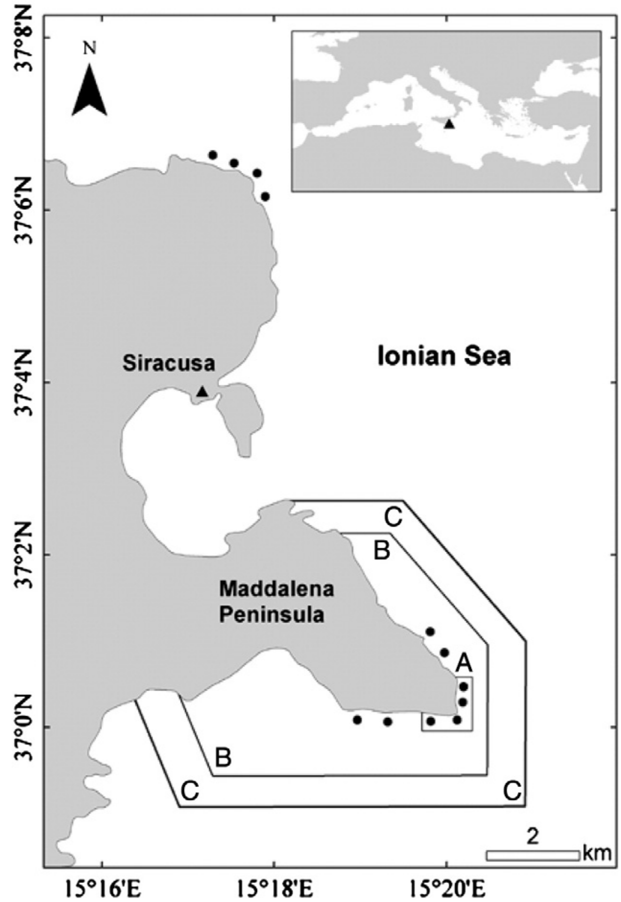

Fig. 1. The Plemmirio MPA (Sicily, Italy, Mediterranean). The sampling sites are circled: four sites were located inside the integral reserve (WR = zone A) and eight outside the reserve (OR; in this case sites were chosen both within the B zone and outside the MPA).

2.3. Sampling design

The sampling design included 3 factors:

(1) "Year" fixed and orthogonal with 2 levels: 2009 and 2010; surveys were conducted at the beginning of summer season in both years for a maximum period of 20 days;
(2) marine reserve "Status" fixed and orthogonal with 2 levels: inside the marine reserve (WR = zone A, where all fishing activities, commercial and recreational, are forbidden) and outside the reserve (OR, corresponding both to the B zone or buffer zone, where only some controlled fishing activities are allowed, and to areas outside the MPA); and
(3) "Site" random and nested within "Status" factor; UVCs were carried out in four sites within the reserve and in eight sites outside the reserve. In this case, sites were chosen both within the B zone and outside the MPA. Sites for each reserve status (WR and OR) were randomly selected from a group of sites identified during a preliminary study (Fig. 1). Three replicated transects ($n = 3$) were performed for each site, leading to a total of 72 observations in the data set. We excluded the C zone (where controlled fishing activities are allowed) from the sample design since this area is different from the other two (A and B) being mainly characterized by shallow sandy bottoms with *Posidonia oceanica* meadows.

2.4. Data analysis

We were interested in assessing whether the Plemmirio reserve was effective at restoring local fish assemblages. This evaluation was carried out at the fish assemblage, fish species and community metrics level. In order to do so, we used multivariate and univariate techniques that are suited for ecological data.

In order to assess how the fish assemblage responds to the effect of protection a three-way permutational analysis of Variance (PERMANOVA; Anderson, 2001; McArdle and Anderson, 2001) was employed using the software package PRIMER 6 with PERMANOVA+ add-on (Anderson et al., 2008). The analysis was based on Bray–Curtis dissimilarities (Bray and Curtis, 1957) calculated on log transformed data and each term of the analysis was tested using 4999 random permutations of appropriate units (Anderson and ter Braak, 2003). This permutation method is generally thought to be the best because it provides very large statistical power and the most accurate control of Type I error (Anderson and Legendre, 1999). Differences in biological responses across years and between inside and outside the reserve (termed WR/OR differences from this point onwards) were interpreted by pair-wise comparisons conducted on these interaction terms. Differences between sites over years in a given reserve status (i.e., a significant Year × Site (Status) interaction) do not interfere with the MPA effects. These differences could be due to small-scale variability in the assemblages of fish (Claudet et al., 2006). Analyses were conducted for sets of abundance indices calculated at several levels and for several components of the fish assemblage (1) abundance per species for the whole fish assemblage; (2) abundance per observed size group (small, medium and large) and (3) abundance for species groups based on the species fishing value. For fishing value, three groups of species were considered: species with low, medium and high fishing values, whereas unfished species (i.e. small species that are not catchable by local fishing gear) were excluded from the analyses.

A two-dimensional non-metric multidimensional scaling (nMDS) plot was generated on the basis of Gower similarity matrix of abundance data. Finally, the similarity percentage procedure SIMPER (Clarke, 1993) was used to identify the fish species mostly contributing to the differences between locations.

Furthermore, we were also interested in analysing the effect of the MPA on diversity metrics. This was addressed through univariate

Table 1
Mean species abundances and standard errors (±SE) per year (2009 and 2010) and reserve status (OR = outside the reserve and WR = inside the reserve). Fishing values groups corresponded to unfished species (U) and species with low (L), medium (M) or high (H) values.

Year	Taxa	2009						2010						Fishing value
Status		OR			WR			OR			WR			
Family		Mean		SE	Mean		SE	Mean		SE	Mean		SE	
Apogonidae	*Apogon imberbis*	13.88	±	4.58	27.17	±	4.06	8.67	±	2.66	5.67	±	1.00	U
Blenniidae	*Parablennius pilicornis*	0.13	±	0.07										U
	Parablennius rouxi	0.63	±	0.15	0.33	±	0.14							U
Centracanthidae	*Spicara* spp.	0.08	±	0.08	28.33	±	9.99				116.25	±	54.03	U
Clupeidae	*Sardinella aurita*				250.00	±	75.38							U
Gobiidae	*Gobius bucchichi*	0.29	±	0.14	0.17	±	0.11	0.25	±	0.09				U
	Gobius cruentatus	0.13	±	0.07				0.04	±	0.04				U
	Gobius geniporus	0.13	±	0.07				0.13	±	0.07				U
	Gobius vittatus							0.21	±	0.08				U
Labridae	*Coris julis*	44.08	±	4.34	72.17	±	9.36	26.67	±	3.67	56.08	±	5.91	U
	Labrus merula	0.04	±	0.04										M
	Labrus viridis	0.04	±	0.04				0.08	±	0.08				M
	Symphodus doderleini	0.75	±	0.17	0.92	±	0.45	0.75	±	0.16	1.00	±	0.39	U
	Symphodus mediterraneus	0.46	±	0.19	1.17	±	0.27	0.92	±	0.17	0.92	±	0.31	U
	Symphodus melanocercus										0.17	±	0.11	U
	Symphodus ocellatus	0.38	±	0.16	0.17	±	0.11	1.00	±	0.31	2.00	±	0.73	U
	Symphodus roissali	0.04	±	0.04	0.08	±	0.08							U
	Symphodus rostratus	0.79	±	0.31				0.63	±	0.29				U
	Symphodus tinca	2.54	±	0.81	2.25	±	0.52	1.33	±	0.33	2.83	±	0.58	M
	Thalassoma pavo	17.67	±	3.37	43.92	±	9.18	27.21	±	3.26	29.50	±	6.31	U
Mullidae	*Mullus surmuletus*	1.63	±	0.41	0.17	±	0.11	0.58	±	0.28	8.50	±	1.99	H
Muraenidae	*Muraena helena*	0.04	±	0.04	0.75	±	0.13	0.08	±	0.06	2.00	±	0.65	L
Pomacentridae	*Chromis chromis*	150.29	±	44.22	110.00	±	5.22	118.71	±	46.07	36.00	±	15.68	U
Scaridae	*Sparisoma cretense*	1.42	±	0.27	5.50	±	1.08	0.79	±	0.17	2.25	±	1.49	L
Sciaenidae	*Sciaena umbra*				0.50	±	0.50				10.00	±	5.22	H
Scorpaenidae	*Scorpaena maderensis*	1.17	±	0.37	0.25	±	0.13	0.71	±	0.16	0.33	±	0.19	M
	Scorpaena notata							0.13	±	0.07				M
	Scorpaena porcus	1.50	±	0.49	2.25	±	0.39	0.21	±	0.08	0.58	±	0.19	M
	Scorpaena scrofa				0.25	±	0.13				0.75	±	0.13	H
Serranidae	*Anthias anthias*				2.25	±	1.25				1.25	±	0.37	U
	Epinephelus costae				0.50	±	0.26				0.58	±	0.31	H
	Epinephelus marginatus				0.83	±	0.30				0.75	±	0.35	H
	Mycteroperca rubra				10.00	±	5.22				1.00	±	0.54	H
	Serranus cabrilla	2.58	±	0.38	1.50	±	0.38	3.33	±	0.35	4.00	±	0.37	L
	Serranus scriba	1.54	±	0.34	3.50	±	0.42	2.21	±	0.32	3.25	±	0.51	L
Sparidae	*Boops boops*	0.88	±	0.58	75.00	±	22.61	2.83	±	1.46	21.67	±	7.57	U
	Diplodus annularis							0.17	±	0.10	0.33	±	0.14	L
	Diplodus puntazzo	0.33	±	0.33	0.42	±	0.29							H
	Diplodus sargus	0.50	±	0.12	1.00	±	0.39	0.54	±	0.16	2.50	±	0.70	H
	Diplodus vulgaris	4.08	±	0.87	5.92	±	2.32	1.17	±	0.21	4.83	±	1.85	H
	Oblada melanura	0.33	±	0.13							0.25	±	0.25	M
	Pagrus pagrus	0.46	±	0.24										H
	Salpa salpa	3.33	±	2.31	0.17	±	0.17							L
	Spondyliosoma cantharus										0.33	±	0.14	H
Sphyraenidae	*Sphyraena viridensis*				125.00	±	65.28							M
Tripterygiidae	*Tripterygion delaisi*	0.50	±	0.15	0.25	±	0.13	0.42	±	0.10	0.25	±	0.13	U

Table 2
PERMANOVA analyzing the effect of factors Year, Status and Site on fish assemblage based on Bray–Curtis dissimilarities of log transformed data. OR = outside the reserve and WR = inside the reserve; *P<0.05; **P<0.01; ***P<0.001; n.s. = not significant.

Source of variation	df	SS	*F*	P	
Year	1	3917.40	2.24	0.03	*
Status	1	7895.10	2.66	0.014	*
Site (Status)	10	29,722.00	20.85	0.001	***
Year × Status	1	3265.10	1.87	0.099	n.s.
Year × Site (Status)	10	17,498.00	12.28	0.001	***
Res	48	6841.70			

analyses. We then modelled the overall fish abundance, Species Richness and the Shannon–Wiener diversity index (Magurran, 1988). The abundance index was calculated for the same fish categories used in the multivariate analyses and also for species with high fishing value considering only large fishes, because they usually respond more to protection (Mosquera et al., 2000). Analyses were conducted using permutation tests realised using the software package PRIMER 6 with PERMANOVA + add-on (Anderson et al., 2008) with 4999 random permutations. In the models, only abundance variables were log-transformed. Unlike multivariate analyses described above, we used a Euclidean distance in the univariate models.

2.5. Level of enforcement

We also were interested in assigning a level of enforcement. Categorizing enforcement in the reserve required obtaining information about (1) the frequency of illegal fishing within the reserve, and (2) the efficacy of the reserve personnel, the coast guard or other marine police forces in active surveillance against illegal activities (Guidetti et al., 2008). This information was directly collected by the researchers involved in the project, and/or gathered by questioning the reserve personnel. The relative enforcement categories were high (poaching very occasional if any, patrol very active and continuous), medium (illegal fishing occurring but limited by infrequent surveillance) and low (common illegal fishing and virtually nonexistent surveillance). Categorization was obtained by first assigning a score to surveillance and poaching for any single marine reserve in terms of percentage of days per year when there was active surveillance (<25, 25–75, >75%, corresponding to score values of 0, 1 and 2, respectively) and events of poaching (<25, 25–75, >75%, corresponding to scores of 2, 1 and 0, respectively). Then, the product of surveillance and poaching scores was calculated and the enforcement category assigned with 0 = low, 1–2 = medium and 4 = high enforcement.

Table 3
SIMPER of fish taxa contributing most (%) to dissimilarity between inside (WR) and outside (OW) reserve, and mean abundances.

Species	OR	WR	Contrib%	Cum.%
	Mean abund.	Mean abund.		
Coris julis	35.38	64.13	22.66	22.66
Thalassoma pavo	22.44	36.71	17.14	39.81
Sphyraena viridensis		62.5	13.62	53.43
Apogon imberbis	11.27	16.42	12.99	66.42
Diplodus vulgaris	2.63	5.38	4.06	70.47
Mullus surmuletus	1.1	4.33	3.79	74.27
Sciaena umbra		5.25	3.6	77.87
Sparisoma cretense	1.1	3.88	2.56	80.43
Symphodus tinca	1.94	2.54	1.77	82.2
Serranus scriba	1.88	3.38	1.66	83.86
Mycteroperca rubra		5.5	1.45	85.31
Serranus cabrilla	2.96	2.75	1.44	86.75
Anthias anthias		1.75	1.32	88.07
Diplodus sargus	0.52	1.75	1.31	89.38
Scorpaena porcus	0.85	1.42	1.28	90.66

3. Results

In Table 1, mean abundances and standard errors of each species are shown for factors "year" and "status". Overall 46 fish taxa belonging to 17 families were recorded in the study area; thirty-seven taxa were found inside the reserve and 36 outside. Twenty-seven species were in common to both locations (WR and OR) whereas ten were exclusive of the reserve and nine were observed only outside the reserve. In particular high fishing value species, such *Epinephelus costae*, *Epinephelus marginatus*, *Mycteroperca rubra*, *Sciaena umbra*, *Spondyliosoma cantharus* and *Scorpaena scrofa*, were observed exclusively inside the reserve (Table 1).

Table 4
Results of permutational univariate ANOVAs performed on fish abundances per size group (small, medium, large), fishing value group (low, medium, high) and on Species Richness (S) and Shannon Wiener (H′) indexes. *P<0.05; **P<0.01; ***P<0.001; n.s. = not significant.

Community metrics	Source of variation	df	SS	*F*	P	
All fishes	Year	1	1.868	2.459	0.140	n.s.
	Status	1	9.669	16.012	0.005	**
	Site (Status)	10	0.604	39.039	0.001	***
	Year × Status	1	0.457	0.602	0.441	n.s.
	Year × Site (Status)	10	0.760	49.124	0.001	***
	Res	48	0.015			
Fish size						
Small fishes	Year	1	5.845	4.151	0.076	n.s.
	Status	1	4.861	3.743	0.073	n.s.
	Site (Status)	10	12.986	18.189	0.001	***
	Year × Status	1	0.191	0.136	0.750	n.s.
	Year × Site (Status)	10	14.080	19.721	0.001	***
	Res	48	3.427			
Medium fishes	Year	1	2.240	3.632	0.088	n.s.
	Status	1	5.964	11.314	0.004	**
	Site (Status)	10	5.271	12.708	0.001	***
	Year × Status	1	0.515	0.835	0.372	n.s.
	Year × Site (Status)	10	6.168	14.871	0.001	***
	Res	48	1.991			
Large fishes	Year	1	0.041	0.041	0.840	n.s.
	Status	1	21.304	20.472	0.002	**
	Site (Status)	10	10.406	27.932	0.001	***
	Year × Status	1	1.936	1.913	0.205	n.s.
	Year × Site (Status)	10	10.120	27.164	0.001	***
	Res	48	1.788			
Fishing value						
Low value	Year	1	0.000	0.001	0.979	n.s.
	Status	1	4.774	5.697	0.058	n.s.
	Site (Status)	10	8.381	6.309	0.001	***
	Year × Status	1	0.011	0.025	0.892	n.s.
	Year × Site (Status)	10	4.572	3.441	0.004	**
	Res	48	6.376			
Medium value	Year	1	16.786	6.983	0.033	*
	Status	1	13.992	4.571	0.057	n.s.
	Site (Status)	10	30.610	14.045	0.001	***
	Year × Status	1	3.375	1.404	0.280	n.s.
	Year × Site (Status)	10	24.040	11.031	0.001	***
	Res	48	10.461			
High value	Year	1	0.005	0.002	0.967	n.s.
	Status	1	35.105	35.824	0.001	***
	Site (Status)	10	9.800	3.950	0.002	**
	Year × Status	1	10.393	4.789	0.056	n.s.
	Year × Site (Status)	10	21.702	8.747	0.001	***
	Res	48	11.910			
Diversity indexes						
Species Richness (S)	Year	1	0.001	0.007	0.943	n.s.
	Status	1	0.713	5.268	0.045	*
	Site (Status)	10	1.353	10.167	0.001	***
	Year × Status	1	0.159	1.107	0.328	n.s.
	Year × Site (Status)	10	1.436	10.795	0.001	***
	Res	48	0.639			
Shannon Wiener diversity (H′)	Year	1	0.102	0.956	0.359	n.s.
	Status	1	0.542	3.109	0.048	*
	Site (Status)	10	1.743	5.816	0.001	***
	Year × Status	1	0.345	3.232	0.099	n.s.
	Year × Site (Status)	10	1.068	3.564	0.002	**
	Res	48	1.439			

Table 5
Mean Species Richness (S) and Shannon Wiener index (H′) values together with mean abundances of size groups and fishing value groups, calculated per year (2009 and 2010) and location (OR = outside the reserve and WR = inside the reserve). The P values of pair-wise comparisons for factor Year and Status are also reported. * P<0.05; ** P<0.01; *** P<0.001; n.s. = not significant. SE = standard errors.

Factors	Time					Status				
Levels	2009		2010		P	OR		WR		P
Variables	Mean	SE	Mean	SE		Mean	SE	Mean	SE	
Abundance (N)	170.69	31.69	99.44	6.79	n.s	89.79	6.06	225.63	43.33	**
Species Richness (S)	12.58	0.55	12.22	0.51	n.s	11.50	0.44	14.21	0.53	*
Shannon Wiener diversity (H′)	1.55	0.04	1.58	0.05	n.s	1.51	0.04	1.69	0.05	*
Fish size										
Small	40	4.573	20.861	2.226	n.s	26.9	3.61	37.5	3.822	n.s
Medium	64.667	10.3	38.556	2.257	n.s	37.56	2.3	79.708	14.2	**
Large	65.806	19.37	40.028	4.109	n.s	25.17	1.36	108.42	26.5	**
Fishing value										
Low value	9.75	1.619	8.3333	0.734	n.s	7.75	1.23	11.625	0.8	n.s
Medium value	47	23.1	2.9722	0.322	*	4.042	0.36	66.875	34.14	n.s
High value	10.972	2.172	11.278	2.631	n.s	4.479	0.4	24.417	3.642	***

PERMANOVA on the total fish assemblage showed significant differences for each factor considered in the analysis except for the interaction factors Year × Status (Table 2).

SIMPER procedure identified some fish taxa as major contributors to the inside/outside dissimilarities. In particular, high densities of labrids *Coris julis* and *Thalassoma pavo* characterized the censuses carried out inside the marine reserve (Table 3).

PERMANOVA (Table 4) performed on the total fish abundance, confirming the result of multivariate analysis, showed significant WR/OR differences, with higher values within the reserve (Table 5). As regards fish size groups, there was not an effect of Year, whereas, an effect of Status was detected on medium and large specimens (P<0.01). However all three size groups showed higher mean abundances inside the marine reserve (Tables 4 and 5).

All three fishing value groups showed higher abundances inside the reserve than outside, though significant differences were found only for high value species (P<0.001; Tables 4 and 5). A similar trend was also observed for large fishes of these high value species, the mean abundances always being larger within the reserve (Fig. 2). Moreover, it is important to observe how seven of these high valuable species were exclusively recorded inside the reserve.

On average Species Richness (S) and Shannon Wiener (H′) values didn't significantly differ between 2009 and 2010, whereas results were significantly higher within the reserve (P<0.05; Table 5).

It is important to note that between sites differences and small-scale variability results were always very high for each variable considered both with univariate and multivariate statistical analyses. In fact the interaction term Year × Site (Status) was always highly significant (P<0.01; Tables 2 and 4).

During the two sampling years the Plemmirio MPA was characterized by an active surveillance carried out every day and by very few poaching events, thus, enforcement of this reserve is very high (LE = 4).

4. Discussion

The present study provides an extensive dataset on the coastal fish assemblage of Plemmirio MPA. Our data showed that Plemmirio MPA is characterized by a high level of enforcement and by a positive reserve effect, as seen also in other well-enforced Italian MPAs (Guidetti et al., 2008). Enforcement and compliance of an MPA are pre-requisites for the effective protection of fish populations (Guidetti and Sala, 2007; Guidetti et al., 2008), to facilitate spillover of adult fish (Roberts et al., 2001), maintain trophic structure (Sala et al., 1998) and promote socio-economic benefits (Holmund and Hammer, 1999). Therefore, the assessment of enforcement is important because the comparison of "reserve vs. fished" only makes sense if the MPA is well enforced. Thus, the scant information in many published studies about compliance and enforcement at the reserves investigated often makes the interpretation of results uncertain (Guidetti et al., 2008).

In general, our results showed significant inside/outside differences in the multivariate abundance of fish assemblages. *T. pavo* and *C. julis* were the species most responsible for these differences, being more abundant inside the reserve. The same results were also found in other Mediterranean MPAs (Bell, 1983; García-Rubies and Zabala, 1990; Harmelin, 1987; Harmelin et al., 1995; La Mesa and Vacchi, 1999). These results were confirmed by significant univariate

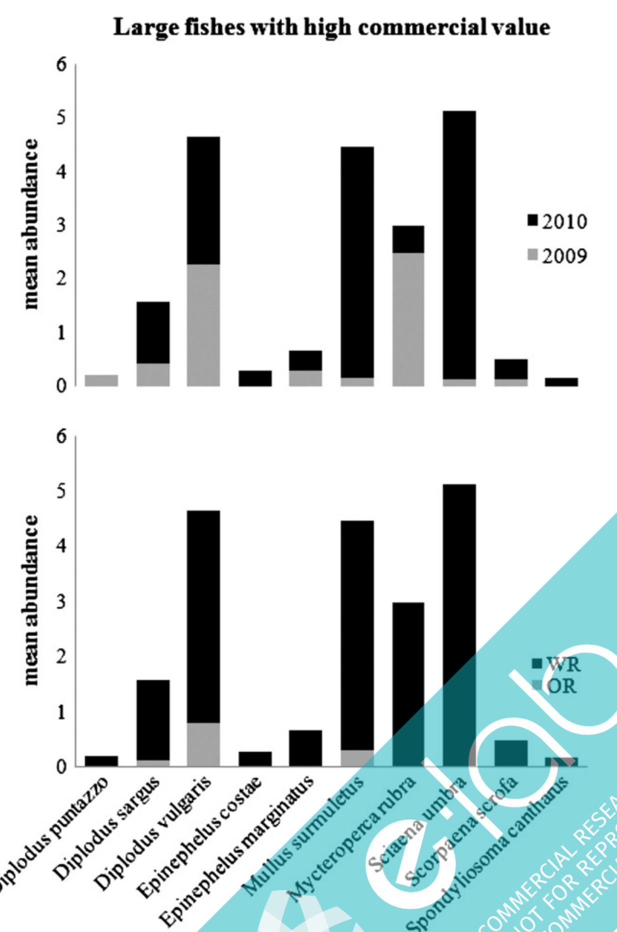

Fig. 2. Mean abundances of large fishes, belonging to high value species group, inside (WR) and outside (OR) the marine reserve.

inside/outside differences for total abundance, Species Richness and Shannon Wiener indexes of the fish assemblage: values of these metrics were higher, even if not always significantly, inside the reserve than outside. In our study, the groups of small size fish and species of low and medium fishing value did not display significant inside/outside differences in abundances whereas medium, large size fishes and high value species showed abundances significantly higher inside the marine reserve.

Protection effects were evident for large specimens of high fishing value, most of which were exclusively found inside the reserve (*D. puntazzo*, *E. costae*, *M. rubra*, *S. scrofa*, *S. cantharus*, *S. umbra*, *E. marginatus*,): it is worth noting that the last two species are included in annex III (list of species whose exploitation is regulated) of SPA/BIO Protocol (Protocol concerning Specially Protected Areas and Biological Diversity in the Mediterranean; CELEX-EUR Official Journal L 322, 14 December 1999, pp. 3–17) and in appendix III (list of protected fauna species) of the BERN Convention (Convention on the Conservation of European Wildlife and Natural Habitats; Bern, 19.09.1979) and were only found inside the MPA, as reported in other NW Mediterranean marine reserves (Francour, 1994; García-Rubies and Zabala, 1990; Harmelin et al., 1995); the dusky grouper, *E. marginatus*, is also included in the IUCN red list as endangered and at risk of dramatic reduction (see http://www.iucnredlist.org). The sighting of *Gobius vittatus* is also noteworthy due to its rarity in the Mediterranean Sea (La Mesa and Vacchi, 1999) and also the presence of certain thermophilic species such as *E. costae*, *Sparisoma cretense* and *Scorpaena maderensis* is geographically relevant (La Mesa and Vacchi, 1999).

Fish predators of sea urchins (*Diplodus sargus* and *Diplodus vulgaris*) also clearly responded to protection. In spite of this, the density of *Diplodus* fish was under the threshold (~12 adult individuals per 125 m^2) required to control sea urchin populations inside the reserve (Guidetti and Sala, 2007). This ecological threshold seems to be attainable only where enforcement is high or where local fishing pressure is not very strong (Guidetti et al., 2008). In the studied area, this threshold value was never reached probably because in the area there are more than 40 artisanal fishing boats that operate around the MPA.

Despite this, in general, at species level, mean fish abundances responded to MPA establishment through increasing abundances within the reserve. In accordance with Willis and Anderson (2003) and Claudet et al. (2006), many cryptic fish species censused outside the reserve were rarely recorded inside and when found were in lower densities. This might be explained by the effect of predators. In fact, protection can improve abundances or sizes, but target species are very often predator species and thus there will be higher predation pressure inside the MPA, leading to changes in the fish assemblage (Ashworth and Ormond, 2005; Francour, 1994; Pinnegar et al., 2000). Consequently, the increase in the number and size of predators inside the Plemmirio's MPA might have altered the trophic structure by increasing the pressure on prey.

Finally, the differences we found between sites could be due to small-scale variability in the assemblages of fish and not to an interaction with the MPA effects (Claudet et al., 2006). This is not surprising, given that the spatial scale of individual sites is not large compared to the high mobility of many fish species included in these surveys (Anderson and Millar, 2004). This result concurs with many studies of invertebrates and algae in intertidal and subtidal environments, which have also often found the greatest variability to occur at small spatial scales (e.g., Archambault and Bourget, 1996; Fowler-Walker and Connell, 2002; Menconi et al., 1999; Underwood and Chapman, 1996).

5. Conclusions

The present study provided evidence of a positive reserve effect on fish populations only five years after its establishment and this represents an important result, likely due to the high level of enforcement. Indeed, our study seems to be in contrast to the outcomes of other studies carried out in different Mediterranean MPAs which provided opposite results, even several years from their establishment (Dufour et al., 1995; Palmeri, 2004; Tunesi et al., 2006). It is worth nothing that some of these negative results could be linked to the MPA not having been in existence for a sufficiently long time to allow fish populations to recover (García-Charton et al., 2004), to problems with sampling design enhancing fish abundance (Fraschetti et al., 2002; García-Charton and Pérez-Ruzafa, 1999; García-Charton et al., 2000; Guidetti, 2002) or to a masking effect of habitat structure and depth (García-Charton et al., 2000, 2004).

Although no previous data are available regarding fish populations before this marine reserve was created, this study establishes a baseline from which further studies can be compared in the Plemmirio MPA and to assess the long-term effect of protection on fish assemblages.

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