

# Article

# Benthic Fish Communities Associated with *Posidonia oceanica* Beds May Reveal the Fishing Impact and Effectiveness of Marine Protected Areas: Two Case Studies in the Southern Tyrrhenian Sea

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Abstract: The effectiveness of a Marine Protected Area (MPA) is strongly related to its zoning design, management, and surveillance, and fish communities are considered indicators of conservation effects. Posidonia oceanica beds and fish assemblages were studied here to evaluate the fishing impacts and the effectiveness of the Marine Protected Areas (MPAs) of Santa Maria di Castellabate (SMC) and Costa degli Infreschi e della Masseta (CIM) in the South Tyrrhenian Sea, Italy. P. oceanica characteristics were estimated counting and collecting shoots, while fish communities were investigated by visual census. Multivariate analyses were performed to detect differences in fish assemblages among the MPA protection levels and seagrass characteristics. Significant differences in fish communities were observed between the depth and protection levels. The latter are probably due to the zoning design at SMC, which does not allow a spillover effect, and to fishing activities, as showed by the ABC curve results. Trophic analyses suggested the use of longlines at SMC, due to the dominance of planktivorous fish species, and of trammel nets at CIM, due to the great heterogeneity of the fish trophic groups. Shoot density and leaf length were significantly correlated with fish communities. CCA analyses showed omnivorous species, mostly belonging to the Sparidae family, correlated with the highest shoot densities, with their feeding habits mostly directed to crustaceans, while macroand microinvertivores, mainly belonging to the Serranidae and Labridae families, correlated with the highest leaf lengths, where they sough shelter and feed. In conclusion, the data suggest that a zoning redesign at SMC and an increase in the surveillance of fishing activities at CIM might enhance the effectiveness of these MPAs, highlighting the role of fish community studies in identifying MPA efficiency issues and providing trustful guidelines for their management.

**Keywords:** marine protected areas; protection levels; posidonia oceanica; seagrass beds; phenological parameters; fish community; trophic analysis; zoning design; fishing activity

# 1. Introduction

Marine Protected Areas (MPAs) are management tools aimed at conserving biodiversity, concurrently with an increase in the local economies in a sustainable development scenario. Their success depends on management and enforcement, which are strongly influenced by their social acceptability [1]. Controversies about MPA effectiveness still



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2 of 19

exist, with MPAs being often exclusively considered as coercive tools established for conservation goals [2]. To smooth out these controversies, the Ecosystem-Based Management approach [3] points out the need to report, by means of investigations, the natural resources enforcement within MPAs [4], recognizable by reserve and spillover effects [5–10]. MPA success is primary determined by four issues: (i) engagement level of local populations, (ii) socioeconomic features of the institution areas, (iii) zoning design, and (iv) ecological factors [11]. Leaving aside the first two topics, which are not addressed in the present study, zoning design and ecological factors are the main issues over which researchers, decision makers, and stakeholders argue [1,12]. In particular, zoning design is one of the topics about which stakeholders are more concerned, because it directly influences local population economies. Too often, MPAs have been established only considering population needs or conservation goals, overlooking the meeting point between protection policies and stakeholders that, on the contrary, should be at the base of their establishment process [13,14]. Ecological factors are concretized in the list of priority habitats (Habitats directive 92/43/CEE, Barcelona Convention, SPA/RAC), recognized as those providing ecosystem goods and services for human health and the economy's well-being [15].

In the Mediterranean Sea, *Posidonia oceanica* (L.) Delile beds represent the most important and most-studied vegetated habitat [16,17], shaping extensive and extremely productive seagrass beds with high structural complexity [18,19]. *P. oceanica* leaves can reach over 1 m in height during summer and 40 cm in winter. Usually, shoot density is maximal in shallow waters, where it may attain more than 1000 shoots per m<sup>2</sup>, and decreases exponentially with depth [20,21]. Nevertheless, edaphic characteristics, such as strong long-shore currents and high water turbidity, can determine highest shoot densities in intermediate areas of the beds, where environmental characteristics are more suitable for their growth [22,23]. Beds can extend from shallow waters to 40 m depth, although in very clear water, they can occur in up to 50 m depth [24]. These habitats play a key role in carbon sequestration and stock, oxygenation of the oceans, prevention of coastal erosion, and provision of food and shelter to many invertebrate and fish species [25]. Indeed, seagrasses are recognized as settlement, spawning, nursery, and feeding areas for many species belonging to all trophic levels, from primary producers, epiphyting *P. oceanica* leaves and rhizomes, to top predators, finding prey during their different life stages [25].

Fish communities quickly respond to habitat alterations due to anthropic activities [26] and protection actions [7]. Spatial and temporal variations in their structure are influenced by both biological (e.g., predation, competition, larval dynamics, spawning, and recruitment rates) and environmental (e.g., depth, light, nutrient availability, habitat structure, bed complexity, and current) factors [27], as well as by anthropogenic disturbances (e.g., fisheries) [28,29]. Thus, for example, in bordering rock–algal/P. oceanica beds, the Labridae species was observed in higher abundances when compared to other seascape contexts; the zooplanktivorous Oblada melanurus (Linnaeus, 1758) is replaced by Spicara smaris (Linnaeus, 1758), and the species Chromis chromis (Linnaeus, 1758) is less abundant than in mosaic beds [30,31]. Nevertheless, *P. oceanica* beds are habitats for many fish species, also of commercial interest [32], forming a community structured according to their behaviour. In particular, four behaviours can be recognized: (i) open-water erratic species belonging to the planktivorous Alosidae and piscivorous Carangidae families, (ii) open-water sedentary species Chromis chromis (Pomacentridae) that live on P. oceanica beds, but eat plankton in the water column, far from the canopy, (iii) mesophilic nektobenthic species, primary belonging to the Sparidae family, that live near the canopy and are mainly composed of omnivorous individuals, and (iv) sedentary nektobenthic species, mainly belonging to the Searranidae and Labridae families, that live among the leaves and are mainly composed of macro- and microinvertivore individuals [26,33,34].

Although there are several studies on fish communities and their spatiotemporal variations in *P. oceanica* beds within Marine Protected Areas [35–39], there is a lack of information on the relationship between *P. oceanica* bed characteristics and fish community structures. The present study performed at the Santa Maria di Castellabate and Costa degli

Infreschi e della Masseta MPAs aims at (i) assessing the relationship between *P. oceanica* bed characteristics and the associated fish communities and (ii) evaluating fishing impacts and the effectiveness of MPAs as conservation tools through variations in fish assemblages at different MPA protection levels. Finally, possible advice to enhance conservation success and local economies are also provided.

#### 2. Materials and Methods

## 2.1. Study Area

The MPAs of Santa Maria di Castellabate (SMC) and Costa degli Infreschi e della Masseta (CIM) were established by the 21 October 2009 Ministerial Decree and stretch along the northern and southern parts, respectively, of Cilento Coast (South Tyrrhenian Sea), a vast mountain and coastal area in the Campania region (Italy) [40,41]. They are divided into three protection zones that regulate human activities: the fully protected (A) zone, where any activity that could cause damage or disturbance to the environment is forbidden; the general reserve (B) zone, where some activities are allowed, but regulated and authorized by the managing authority; and the partial reserve (C) zone, where fruition and sustainable use of the sea, even of moderate environmental impact, are allowed.

SMC (Figure 1a) includes 19 km of coastline. The "A" zone extends for 1.7 km<sup>2</sup>, and it is isolated at the northernmost part of the MPA; the "B" zone is divided in a small area (6.35 km<sup>2</sup>), surrounding the "A" zone, and in a very large area in front of Punta Licosa (21.35 km<sup>2</sup>); and the "C" zone includes beaches and deepest areas (37.89 km<sup>2</sup>). The SMC sea bottoms are characterized by lush macroalgal forests of Fucales in shallow waters [42] and coralligenous bioconstructions in deeper waters. The large acrochorus of Punta Licosa, that gently degrades more than 3 miles offshore, is completely covered by a vast *P. oceanica* bed, for more than 26 km<sup>2</sup>, and almost completely included within the "B" zone [43].



Figure 1. Marine Protected Areas of (a) Santa Maria di Castellabate and (b) Costa degli Infreschi e

della Masseta. Different protection levels (A: fully protected zone; B: general reserve zone; and C: partial reserve zone), *Posidonia oceanica* beds, and sampling stations at the seagrass upper limit and 15 m depth are shown.

CIM (Figure 2b) includes 13 km of coastline. The A zone extends for  $0.4 \text{ km}^2$ , and it is located at the MPA core; the B zone surrounds the A zone and extends for  $4.79 \text{ km}^2$ ; the C zone surrounds the B zone and extends for  $18.36 \text{ km}^2$ . Sea bottoms are characterized by photophilic algal and coralligenous habitats at the shallowest and deepest areas, respectively. The steep slope of the sea bottom allows for the settlement of *P. oceanica* beds (1.41 km<sup>2</sup>) for a few tens of metres offshore [44].



**Figure 2.** *Posidonia oceanica* bed characteristics of the Santa Maria di Castellabate (SMC) and Costa degli Infreschi e della Masseta (CIM) MPAs. (**a**) mean ( $\pm$ SD) shoot densities (n° shoots/m<sup>2</sup>) and leaf lengths (cm), divided in green and brown parts (%); (**b**) mean ( $\pm$ SD) leaf surfaces (m<sup>2</sup>/m<sup>2</sup>) and ratio of intact or broken (by mechanical or biological damage) apices (%).

#### 2.2. Sampling Activities and Data Collection

Sampling was carried out between September and October 2020 on *P. oceanica* beds at SMC and CIM. In particular, at each MPA, 4 stations were selected for each protection level ("A", "B", and "C") at two different depths: the seagrass bed upper limit (SL) and 15 m depth (15 m) (Figure 1). The present investigation aims at emphasizing differences on fish communities among protection levels and their relations with *P. oceanica* bed characteristics at each MPA.

*P. oceanica* beds were sampled following the standardized Marine Strategy Framework Directive protocol [45]. At each station, 6 randomly placed  $40 \times 40$  cm squares were considered to estimate the bed shoot density (shoots/m<sup>2</sup>). Moreover, 6 shoots were collected to estimate the following phenological parameters: average leaf number per shoot (considering only intermediate and adult leaves), leaf width, green and brown leaf lengths, leaf base length, percentage of intact or broken (by mechanical and biological factors) leaf apices, and total leaf surface.

Total leaf surface  $Ts (m^2)/m^2$  was calculated as in Equation (1):

$$Ts = \left(\overline{n^{\circ}L} * \overline{l} * \overline{w}\right) * n^{\circ}S \tag{1}$$

where  $n^{\circ}L, l, \overline{w}$  are the average number of leaves, average lengths, and average widths, respectively, and  $n^{\circ}S$  is the shoot density. Juvenile leaves were excluded because they presented neither photosynthetic nor brown parts.

At each station, fish communities were estimated on 125 m<sup>2</sup> by visual census technique replicated 3 times, as described by [5]. Species were classified according to their value as commercial (C), low commercial (LC), and noncommercial (NC), while biomasses were estimated using the population size–structure and length–weight relationships following [46].

Species richness (SR: total species number/125 m<sup>2</sup>), density (D: total abundance/125 m<sup>2</sup>), Shannon diversity index (H'), Pielou evenness index (J), and total biomass (B: total kg/125 m<sup>2</sup>) were also estimated.

Trophic group (TG) of each fish species was assessed following trophic level intervals (*TL*) defined as [47] pure herbivores ( $2 \le TL \le 2.1$ ); omnivores with a preference for vegetable materials ( $2.1 < TL \le 2.9$ ); omnivores with a preference for animals ( $2.9 < TL \le 3.7$ ); and carnivores with a preference for large decapods, cephalopods, and fish ( $3.7 < TL \le 4.5$ ). Finally, the mean sample trophic level  $\overline{TL}$  was also computed by Equation (2)

$$\overline{TL} = \frac{\sum_{i=1}^{n} Y_i TL_i}{\sum_{i=1}^{n} Y_i}$$
(2)

where  $Y_i$  is the biomass in 125 m<sup>2</sup> of species *I*;  $TL_i$  is the trophic level of the species *I*; and *n* is the total number of species in the sample [48,49].

#### 2.3. Data Analyses

Distance-based permutation multivariate analyses of variance (PERMANOVAs, [50]) were carried out on *P. oceanica* data, fish community densities, and biomasses to test for differences within each MPA and among protection levels and different depths. The experimental design involved the "protection level" factor (PL, fixed, three levels: A, B, and C zones), the "depth" factor (De, fixed, two levels: SL and 15 m), and the "station" factor (St, nested in PL and De, 12 levels). Before the analyses, P. oceanica data were standardized, and fish densities and biomasses were square-rooted to reduce the weight of very abundant species. Multivariate analyses were based on Bray-Curtis similarity, and each term was tested by 4999 random permutations [51,52]. Post hoc pairwise comparisons were conducted using the PERMANOVA t-statistic with 4999 permutations. Multivariate patterns were visualized through Canonical Analyses of Principal coordinates (CAP) of PLxDe elements [53]. ABC curves [54] were employed to compare abundance–biomass relationships of commercial species within each MPA protection level. Distance-based linear modelling (DistLM), using stepwise selection procedure and adjusted R<sup>2</sup> as selection criterion [55], was performed to find correlations among fish densities, fish biomasses, and P. oceanica bed characteristics, considered as independent variables. Three plot Canonical Correlation Analyses (CCAs, [56]) were also performed to assess correlation among PLxDe elements, trophic levels, and *P. oceanica* characteristics that significantly affect fish communities resulting from DistLM.

Finally, univariate PERMANOVAs based on Euclidean distance [57] were conducted on species richness, total density, diversity index, evenness, and total biomass of the whole assemblage. The experimental design for these analyses was the same described above for multivariate analyses.

#### 3. Results

# 3.1. Analysis of Posidonia oceanica Beds

In Table 1, the values of the physiographic parameters detected within MPAs and characterizing *P. oceanica* beds are reported.

At SMC, the shoot density was  $551.5 \pm 129.0 \text{ shoots/m}^2$  and the leaf length was  $39.5 \pm 17.18 \text{ cm}$  (brown part = 16.5%), with the highest values at the 15 m sites. At CIM, the shoot density was  $421.4 \pm 93.46 \text{ shoots/m}^2$ , with the highest values at the SL sites, and the leaf length was  $45.5 \pm 21.6 \text{ cm}$  (brown part = 16.0%), with the highest values at the 15 m sites (Figure 2a).

At SMC, the leaf surface was  $10.9 \pm 6.8 \text{ m}^2/\text{m}^2$ , with the highest values at the 15 m sites, and the mean percentages of the mechanical and biological broken apices were 17.5% and 2.9%, respectively. At CIM, the leaf surface was  $10.0 \pm 6.6 \text{ m}^2/\text{m}^2$ , with the highest values at the SL sites, and the mechanical and biological broken apices were 20.7% and 3.7%, respectively (Figure 2b).

<b>Table 1.</b> Mean values (±SD) of <i>P. oceanica</i> bed characteristics collected at the Santa Maria di Castella-
bate (SMC) and Costa degli Infreschi e della Masseta (CIM) MPAs. D: shoot density (shoots/m <sup>2</sup> ),
LL: leaf length (cm), Bp: brown leaf part, LS: leaf surface $(m^2/m^2)$ , Mec: apices showing mechanical
damage (%), Bio: apices showing biological damage (%).

			D	LL	Вр	LS	Mec	Bio
		А	$550.6\pm75.0$	$27.9 \pm 11.6$	16.60%	$7.1\pm3.7$	16.67%	4.17%
	SL	В	$599.5\pm131.0$	$42.9 \pm 18.7$	23.00%	$12.2\pm7.1$	20.25%	2.53%
SMC		С	$427.3\pm217.2$	$37.4 \pm 17.0$	11.60%	$7.5\pm5.3$	13.58%	6.17%
ome		А	$599.2 \pm 160.7$	$39.5\pm19.9$	16.40%	$12.3\pm9.0$	13.51%	0.11%
	15 m	В	$569.2\pm93.2$	$44.6 \pm 17.8$	15.80%	$13.8\pm8.3$	18.18%	2.27%
		С	$563.2\pm97.2$	$45.0\pm18.1$	15.90%	$12.6\pm7.5$	23.26%	2.33%
		А	$461.9 \pm 114.6$	$38.4 \pm 19.4$	16.00%	$9.81 \pm 6.79$	20.23%	3.37%
	SL	В	$539.4 \pm 85.9$	$46.9\pm19.1$	17.80%	$13.50\pm7.78$	15.91%	4.55%
CIM		С	$443.2\pm86.3$	$34.0\pm22.1$	20.70%	$7.87 \pm 5.95$	25.93%	3.71%
0111		А	$345.9\pm109.6$	$53.9\pm23.9$	14.70%	$9.41\pm 6.44$	20.48%	4.82%
	15 m	В	$384.2\pm94.8$	$55.9\pm22.9$	13.00%	$11.73\pm7.03$	23.86%	4.55%
		С	$353.6\pm69.6$	$44.2\pm22.2$	14.20%	$7.93 \pm 5.76$	17.86%	1.19%

# 3.2. Descriptive Analysis of Total Assemblages

A total of 36 species of fishes belonging to 11 families were detected at SMC and CIM. In Table 2, commercial value (CV) and trophic group (TG) of each species are reported.

**Table 2.** Species detected at the Santa Maria di Castellabate (SMC) and Costa degli Infreschi e della Masseta (CIM) MPAs. Families, commercial value (CV; NC: noncommercial; LC: low commercial; C: commercial), and trophic group (TG; Pisc: piscivorous fishes; Macro: macroinvertivores; Micro: microinvertivores; Omni: omnivorous species; Herb: herbivorous species; Plan: planktivorous fishes; Detr: detritivorous fishes) are also reported [48,58].

Family	Species	CV	TG	SMC	CIM
Apogonidae	Apogon imberbis (Linnaeus, 1758)	NC	Micro	х	х
Carangidae	Seriola dumerili (Risso, 1810)	С	Pisc	-	х
Alosidae	Sardina pilchardus (Walbaum, 1792)	LC	Plan	х	-
	Coris julis (Linnaeus, 1758)	NC	Micro	х	х
	Labrus merula Linnaeus, 1758	С	Micro	х	х
	Labrus viridis Linnaeus, 1758	LC	Micro	х	х
	Symphodus doderleini Jordan, 1890	NC	Micro	х	-
Labridae	Centrolabrus melanocercus (Risso, 1810)	NC	Micro	-	х
20011000	Symphodus ocellatus (Linnaeus, 1758)	NC	Micro	х	х
	Symphodus roissali (Risso, 1810)	NC	Micro	х	-
	Symphodus rostratus (Bloch, 1791)	NC	Micro	х	х
	Symphodus tinca (Linnaeus, 1758)	LC	Micro	х	х
	Thalassoma pavo (Linnaeus, 1758)	NC	Micro	х	х
Moronidae	Dicentrarchus labrax (Linnaeus, 1758)	С	Pisc	х	Х
Mugilidae	Mugil cephalus Linnaeus, 1758	С	Detr	-	х
	Mullus surmuletus Linnaeus, 1758	С	Detr	х	-
Mullidae	Mullus barbatus Linnaeus, 1758	С	Detr	х	х
Pomacentridae	Chromis chromis (Linnaeus, 1758)	NC	Planc	х	Х

Family	Species	CV	TG	SMC	CIM
	Serranus cabrilla (Linnaeus, 1758)	С	Macro	х	х
Serranidae	Serranus scriba (Linnaeus, 1758)	С	Macro	х	х
	Epinephelus marginatus (Lowe, 1834)	С	Pisc	х	-
	Boops boops (Linnaeus, 1758)	С	Planc	-	х
	Dentex dentex (Linnaeus, 1758)	С	Pisc	Х	х
	Diplodus annularis (Linnaeus, 1758)	С	Omni	х	х
	Diplodus puntazzo (Walbaum, 1792)	С	Omni	х	-
	Diplodus vulgaris (Geoffroy Saint-Hilaire, 1817)	С	Omni	х	х
Sparidae	Diplodus sargus (Linnaeus, 1758)	С	Omni	х	х
	Oblada melanurus (Linnaeus, 1758)	С	Planc	х	х
	Sarpa salpa (Linnaeus, 1758)	LC	Herb	х	х
	Sparus aurata Linnaeus, 1758	С	Omni	-	х
	Spicara maena (Linnaeus, 1758)	LC	Plan	х	х
	Spicara smaris (Linnaeus, 1758)	LC	Plan	х	х
	Spondyliosoma cantharus (Linnaeus, 1758)	С	Omni	х	х
Sphyraenidae	Sphyraena viridensis Cuvier, 1829	С	Pisc	-	x

## Table 2. Cont.

In Table 3, the values of the synecological indices detected within MPAs and characterizing fish communities are reported.

**Table 3.** Synecological indices (mean  $\pm$  SD) at the Santa Maria di Castellabate (SMC) and Costa degli Infreschi e della Masseta (CIM) MPAs. SR: species richness (species/125 m<sup>2</sup>); D: density (individuals/125 m<sup>2</sup>); H': Shannon diversity index; J: Pielou evenness index; B: total biomass (kg/125 m<sup>2</sup>).

MPA	De	PL	SR	D	H′	J	В
		А	$10.5\pm1.8$	$106.7\pm48.1$	$2.3\pm0.4$	$0.71\pm0.1$	$1.4\pm0.9$
	SL	В	$8.3\pm2.5$	$127.2\pm41.7$	$2.2\pm0.6$	$0.74\pm0.1$	$1.3\pm1.1$
		С	$7.2\pm2.9$	$95.3\pm56.2$	$1.7\pm0.6$	$0.59\pm0.2$	$1.3\pm0.1$
SMC		А	$7.8\pm2.0$	$92.5\pm56.0$	$2.0\pm0.2$	$0.68\pm0.1$	$0.9\pm0.4$
	15 m	В	$7.5\pm2.5$	$116.3\pm57.4$	$2.0\pm0.5$	$0.69\pm0.1$	$1.7\pm1.6$
		С	$7.0\pm0.9$	$122.7\pm54.8$	$2.0\pm0.5$	$0.69\pm0.2$	$2.5\pm1.4$
		А	$9.0\pm2.5$	$141.5\pm76.2$	$1.7\pm0.3$	$0.56\pm0.2$	$0.2\pm0.2$
	SL	В	$8.2\pm1.6$	$111.3\pm61.2$	$2.3\pm0.3$	$0.78\pm0.1$	$0.7\pm0.6$
CIM	С 7.2	$7.2\pm2.2$	$121.3\pm49.4$	$1.9\pm0.5$	$0.68\pm0.1$	$0.2\pm0.1$	
CIM		А	$7.2\pm1.9$	$66.5\pm46.0$	$2.1\pm0.4$	$0.76\pm0.1$	$0.01\pm0.0$
	15 m	В	$7.7\pm1.0$	$90.7\pm34.9$	$2.0\pm0.5$	$0.69\pm0.1$	$0.9\pm0.5$
		С	$5.8\pm0.7$	$99.0\pm40.7$	$1.8\pm0.3$	$0.71\pm0.1$	$0.1\pm0.1$

At SMC, the species richness was  $8.0 \pm 2.1$  species/125 m<sup>2</sup>, with the highest values at the SL sites, and the fish density was  $110.1 \pm 52.4$  individuals/125 m<sup>2</sup>, with the highest values at the 15 m sites. At CIM, the species richness was  $7.5 \pm 1.7$  species/125 m<sup>2</sup> and the fish density was  $105.0 \pm 51.4$  individuals/125 m<sup>2</sup>, with the highest values at the SL sites (Figure 3a).



**Figure 3.** Descriptive fish community analyses for each protection zone and depth of the Santa Maria di Castellabate (SMC) and Costa degli Infreschi e della Masseta (CIM) MPAs. Synecological indices: (a) species richness (SR) and density (D); (b) specific diversity (H') and evenness (J); (c) total biomass; and (d) percentage of trophic group frequencies and mean trophic level (TL).

At SMC, the Shannon diversity index was  $2.0 \pm 0.5$  and the Pielou evenness index was  $0.68 \pm 0.1$ , with the highest values at the SL sites. At CIM, the Shannon diversity index was  $2.0 \pm 0.4$  and the mean Pielou evenness index was  $0.69 \pm 0.1$ , with the highest values at the 15 m sites (Figure 3b).

At SMC, the fish biomass was  $1.5 \pm 0.9$ , with the highest values at the 15 m sites. At CIM, the fish biomass was  $0.9 \pm 0.3$ , with the highest values at the 15 m sites (Figure 3c).

At SMC, the dominant trophic group was the planktivorous one, and the corresponding average trophic level was  $3.47 \pm 0.17$  at SL and  $3.57 \pm 0.07$  at 15 m. At CIM, the trophic groups were very heterogeneous, with a nonevident dominant one; the average trophic level was  $3.04 \pm 0.17$  at SL and  $3.49 \pm 0.21$  at 15 m (Figure 3d).

No significant differences were detected using synecological parameters, except for biomasses. They were different between MPAs (p < 0.001). Within MPAs, at SMC, there was an interaction of PLxDe (p = 0.0084) due to differences between the C zone and the other ones at 15 m ( $p_{A-C} = 0.0028$  and  $p_{B-C} = 0.0476$ ) and between SL and 15 m (p = 0.0084) within the C zone. At CIM, differences were detected only for the factor PL (p = 0.006) between the B zone and the other ones ( $p_{A-B} = 0.015$  and  $p_{B-C} = 0.0412$ ).

#### 3.3. Multivariate Analyses on Fish Assemblages

At SMC, significant differences were detected for the PL, due to differences between the "A" and "C" zones, and the De factors. At CIM, significant differences were detected for the interaction of the PLxDe factors. In particular, differences were detected between SL and 15 m in the "A" and "C" zones; differences were also detected between the "A" and "B" zones at SL, while at 15 m, differences were detected between the "C" zone and the other ones for the density data and between the "A" zone and the other ones for the biomass data (Table 4). **Table 4.** PERMANOVA and pairwise results on fish densities and biomasses at the Santa Maria di Castellabate and Costa degli Infreschi e della Masseta MPAs; "A", "B", and "C" are the levels of the PL and SL factors, and 15 m are the levels of the De factor; pseudo-F and t values are reported; significant *p* values (<0.05) are in bold.

		SMC					CIM		
	Density	Data	Biomass	Data		Density Data		<b>Biomass Data</b>	
Factor	Pseudo-F	р	Pseudo-F	р	Factor	Pseudo-F	р	Pseudo-F	р
PL	1.89	0.021	1.88	0.018	PL	3.31	0.002	2.88	0.002
De	2.75	0.005	2.27	0.0182	De	2.77	0.004	2.86	0.006
PLxDe	0.92	0.5578	1.19	0.26	PLxDe	2.21	0.003	1.95	0.003
			Within	PL factor		W	ithin A leve	e <b>l</b>	
A-B	1.15	0.23	1.29	0.11	SL-15 m	1.69	0.005	1.82	0.002
A-C	1.71	0.003	1.56	0.006	Within B level				
B-C	1.24	0.16	1.28	0.11	SL-15 m	1.06	0.341	1.23	0.108
					Within C level			1	
					SL-15 m	1.71	0.009	1.28	0.134
						Wi	ithin SL leve	el	
					A–B	1.62	0.015	1.25	0.103
					A–C	1.11	0.322	0.93	0.532
					B-C	1.19	0.228	1.11	0.279
					Within 15 m level				
					A–B	1.41	0.071	2.01	0.002
					A–C	2.21	0.004	2.01	0.003
					В-С	1.91	0.004	1.38	0.051

At SMC, the CAP analysis on density data showed protection degrees and depth gradients (Figure 4a). Most of the "A" zone elements were distributed on the positive part of the CAP1 axis; the "B" zone elements were located around the 0 value, while the "C" zone elements were polarized on the negative part. Most of the SL elements were distributed on the positive part of the CAP2 axis, while most of the 15 m elements, relating to *Serranus scriba* (Linnaeus, 1758), *Coris julis* (Linnaeus, 1758), *S. smaris*, and *Symphodus tinca* (Linnaeus, 1758), were distributed on the negative part. A CAP analysis on biomass data (Figure 4b) did not highlight evident gradients, although most of the "A" and "B" zone elements were on the positive part. Most of the SL elements were distributed on the negative part of the CAP1 axis, while most of the "C" zone elements were distributed on the negative part of the CAP1 axis, while most of the "A" and "B" zone elements were distributed on the negative part of the CAP1 axis, while most of the "C" zone elements were distributed on the negative part of the CAP1 axis, while most of the "C" zone elements were on the positive part. Most of the SL elements were distributed on the positive part of the CAP2 axis, while the 15 m elements, relating to *C. julis, Spondyliosoma cantharus* (Linnaeus, 1758), *Diplodus sargus* (Linnaeus, 1758), *S. smaris*, and *Diplodus vulgaris* (Geoffroy Saint-Hilaire, 1817) were distributed on the negative part.

At CIM, the CAP analysis on density data showed protection levels and depth gradients (Figure 4c). "A" zone elements were polarized at the negative part of the CAP 1 axis; "B" zone elements were polarized around the 0 value, while "C" zone elements were distributed on the positive part. Most of the SL elements were distributed on the negative part of the CAP2 axis, relating to *D. sargus* and *D. vulgaris*. Most of the 15 m elements were distributed on the positive part of the CAP2 axis, relating to *S. cabrilla*, *C. julis*, *S. tinca*, and *S. smaris*. A CAP analysis on biomass data showed clear protection levels and depth gradients (Figure 4d). "A" zone elements were polarized on the negative part of the CAP 1 axis, while "B" and "C" zones elements were polarized on the positive part. SL elements were polarized on the negative part of the CAP2 axis, relating to *D. sargus* and *D. vulgaris*, while most of the 15 m elements were polarized on the positive part. SL elements *S. tinca*, *C. julis*, and *C. chromis*.



**Figure 4.** CAP analysis results on fish densities and biomasses at the Santa Maria di Castellabate (**a**,**b**) and Costa degli Infreschi e della Masseta MPAs (**c**,**d**). All density, biomass datasets, and fish species are correlated with the Pearson coefficient ( $\rho > 0.55$ ). Straight lines are vectors of species where the orientation and length are proportional to the most-correlated sites. Circles represent the confidence interval (95%).

At SMC, the ABC curves (Figure 5a) resulted in a negative W statistic in the "A" zone (W = -0.013) and a positive W statistic in the "B" (W = 0.113) and "C" zones (W = 0.038). At CIM, the ABC curves (Figure 5b) resulted in a negative W statistic in the "A" (W = -0.02) and "C" zones (W = -0.106) and a positive W statistic in the "B" zone (W = 0.07).

100

80

60

40

20

а

Cumulative Dominance%



 Image: species rank
 100
 100

 Species rank
 100
 Species rank

 Figure 5. Fish ABC curves at the (a) Santa Maria di Castellabate (SMC) and (b) Costa degli Infreschi e della Masseta (CIM) MPAs. Filled squares are abundances, and empty squares are biomasses. Red symbols refer to data detected within the "A" zone; yellow symbols refer to the "B" zone; and green

## 3.4. Relations between P. oceanica Bed Characteristics and Fish Communities

symbols refer to the "C" zone.

20

At SMC, shoot densities and total leaf lengths were the *P. oceanica* bed characteristics significantly affecting fish communities, while at CIM, fish communities were significantly affected by green and brown leaf lengths and shoot densities (Table 5).

<b>Tuble 5.</b> Characteristics of 1. becanical beas selected by district analyses and this	eme 2.
pseudo-F and $p$ values are reported. Significant $p$ values (<0.05) are in bold.	

	SM	C			CI	М	
Variable	Adj R <sup>2</sup>	Pseudo-F	р	Variable	Adj R <sup>2</sup>	Pseudo-F	p
	Dens	ities			Dens	ities	
Total length	$2.61  imes 10^{-2}$	1.9393	0.0476	Brown	$1.37  imes 10^{-2}$	1.4875	0.0465
Suface	$5.06  imes 10^{-2}$	1.8775	0.0638	Density	$2.58 imes10^{-2}$	1.4204	0.1806
	Bioma	isses		Green part	$2.70 \times 10^{-2}$	1.0397	0.0408
Total length	$1.83  imes 10^{-2}$	1.6513	0.094		Bioma	asses	
Suface	$5.11  imes 10^{-2}$	2.1769	0.285	Density	$6.10  imes 10^{-3}$	1.215	0.025
Density	$5.20  imes 10^{-2}$	1.0303	0.045	Green part	$1.51  imes 10^{-2}$	1.3104	0.0138
5				Suface	$2.46 \times 10^{-2}$	1.322	0.1894
				Brown	$3.21 \times 10^{-2}$	1.2478	0.2356

At SMC, the trophic groups strongly related to *P. oceanica* beds were macroinvertivores, microinvertivores, omnivorous, and planktivorous fishes. They are primary correlated with the 15 m sites that are, in turn, directly related to total leaf lengths and shoot densities (Figure 6a). At CIM, the trophic groups strongly related to *P. oceanica* beds were microinvertivores, omnivorous, planktivorous, and herbivores fishes. In particular, species were correlated with the 15 m sites, except for the omnivorous ones related to the SL sites of the B zone. The upper sites were correlated with shoot densities, while green and brown leaf lengths were mainly related to the 15 m sites (Figure 6b).



**Figure 6.** CCA analysis results on trophic group data at (**a**) Santa Maria di Castellabate and (**b**) Costa degli Infreschi e della Masseta MPAs. Pisc: piscivorous fishes; Macro: macroinvertivores; Micro: microinvertivores; Omni: omnivorous species; Herb: herbivorous species; Plank: planktivorous fishes; Detr: detritivorous fishes. Straight lines are vectors of *P. oceanica* bed characteristics, whose orientation and length are proportional to the most-correlated sites.

# 4. Discussion

MPAs are established to protect biodiversity and natural resources for future generations, stemming the decline of marine ecosystems and of the numerous goods and services they provide [59,60]. The urgent need to prove their effectiveness is mandatory in order to take prompt actions to enhance wildlife protection and local economy development. At the same time, it is necessary to know where possible actions, such as the zoning reshaping or the surveillance enhancing, should be implemented to increase the local economic yield (primarily in terms of production by fishing of target species), minimizing potential stakeholder objections. In this study, the effectiveness of the Santa Maria di Castellabate and Costa degli Infreschi e della Masseta MPAs was assessed to evaluate if new conservation actions are necessary. Moreover, the relationship between *P. oceanica* bed characteristics and fish assemblages was investigated to determine where zoning should be redesigned in order to enhance protection and local economies.

# 4.1. Posidonia oceanica Beds

*P. oceanica* beds can be classified as dense prairies [61] in an equilibrium state with environmental constraints [62,63]. In this study, the characteristics of *P. oceanica* beds were considered as environmental parameters structuring fish communities. Bed density ranged from ~350 to ~600 shoots/m<sup>2</sup>, with highest values at the 15 m sites of SMC and at the shallow sites of CIM.

Leaf length, both at SMC and CIM, was coherent with previous studies assessing that, and during autumn, average leaf length was ~30–50 cm [19,21], with larger lengths occurring at intermediate depths (e.g., 15 m). Higher lengths at the SMC and CIM intermediate depths corresponded to higher average leaf surfaces that provide a larger available substrate for sessile [64] and vagile [65] organisms. Indeed, leaves sustain many organisms that actively feed on them, eat their remains, or graze on their epiphytes. In turn, these organisms enter the food webs, transferring matter and energy up to the predators of the higher trophic levels [66].

The most frequent cause of broken leaf apices was mechanical damage rather than biological damage (i.e., grazing), also within the no-take (A) zone of both MPAs, where every human activity disturbing or damaging the environment should be forbidden. This result is presumably related to the anchoring activity carried out during summer [67].

#### 4.2. Fish Communities

Most evidence on the MPAs' effectiveness stems from quantitative assessments of fish populations [68], which rapidly and directly respond to protection instruments in terms of density, body size, and biomass [7]. Compared to other studies, the fish communities investigated here do not have a good status, as evident from the low density and biomass values [9,39,68]. Regarding the synecological parameters, differences in total biomass were detected between the two MPAs and among protection levels and depths. These differences were analysed in more detail considering the density and biomass of the different species, which also showed significant differences among protection levels and depths. Differences between depth levels are coherent with studies identifying different fish assemblages at different depths in *P. oceanica* beds [69,70]. Differences among protection levels could be related to two possible reasons: MPA zoning design and level of surveillance. In this framework, two well-known reviews written in the early 2000s reported the importance of the positioning of the fully protected (A) zone [2,71]. Indeed, due to its strict level of protection, the "A" zone is the most pristine zone of an MPA, providing refuge, feeding, and nursery areas for fish assemblages [4,12].

SMC is characterized by a fully protected (A) zone confined at the north edge of the MPA, surrounded by a small general reserve (B zone) and a large partial reserve (C zone) at the south. This zoning design does not allow possible spillover effects coming from the potential reserve effect [9]. Moreover, the "B" zone of Punta Licosa, which comprises most of the MPA area, includes the vast majority of the *P. oceanica* beds, with the most

abundant fish populations [31]. Fish assemblage size structure showed a dominance of small- or medium-sized individuals. This result is typical of *P. oceanica* beds for their role as a nursery area [26], although it is worth pointing out that the lack of large top predators might have also been caused by local fishing [58,72].

At CIM, the fully protected "A" zone is located at the MPA core, surrounded by the general reserve (B zone), which is in turn surrounded by the partial reserve (C zone). Hence, CIM zoning design should allow reserve and spillover effects, i.e., set of MPA positive effects that occur within and outside the fully protected zone [9]. However, the significant interaction of the PLxDe factors, a typical result of these effects [7,69], is not consistent with the very low synecological and biomass values. Weak reserve and spillover effects can be found at the SL sites, as suggested by the biomass CAP analysis, characterized by higher species richness and densities, although with small size classes [38]. Conversely, the reserve and spillover effects completely disappear at the 15 m depth sites, where the very low biomass values suggest a significant negative impact, probably of anthropic origin, on fish communities [73–77]. These findings were also confirmed by the ABC curves. Indeed, the positive W statistic in the B zone may be due to the convergence of incoming individuals from the fully protected "A" zone to the general reserve (B zone) during the adult phase, and the consequent concentration of fishing harvesting here and within the partial reserve (C zone).

In undisturbed communities, the presence of large individuals of the target species results in higher biomasses, whose curves lie completely above the abundance ones, determining a positive W statistic. Conversely, in a highly disturbed community, mainly characterized by small individuals, the abundance curves lie above the biomass ones due to the presence of anthropic impacts, determining a negative W statistic [78–80]. SMC showed a negative W statistic only in the fully protected "A" zone, suggesting that despite the strictly protection level, this zone is in an area impacted by anthropic activities, hampering the reserve effect. The latter, on the contrary, is more evident in the other MPA zones (B and C), as showed by the positive W statistic, suggesting that the fully protected "A" and partial reserve "C" zones are subjected to exploitation activities, possibly fishing.

These results are coherent with the trophic analyses that (i) confirm the fishing activities in both MPAs due to the almost complete absence of piscivorous species [58,81] and (ii) suggest the employment of specific and different fishing gear in both MPAs. The selectiveness of fishing gear affects the biomass and trophic structure of fish communities [82–86]. At SMC, the dominance of planktivorous species, such as *Oblada melanurus* (Linneus 1758), *S. smaris*, and *Sardina pilchardus* (Walbaum, 1792), probably depends on the use of selective gear, such as longlines, that act on the trophic structure removing fish predators [80]. At CIM, trophic structure heterogeneity coupled with very low biomasses suggests a larger use of nonselective fishing gear, such as trammel nets [87].

#### 4.3. Relationship between P. oceanica Beds and Fish Communities

The present study aims to enhance the knowledge on the relation between fish communities and *P. oceanica* bed characteristics, which can influence biodiversity protection and local economies. A clear space distribution of fish species was evident looking at the CAP and CCA analyses. In particular, the water column was dominated by erratic and sedentary fish species, such as *S. smaris* and *C. chromis*, both planktivorous species. Nektobenthic fish species were, as expected, more related to *P. oceanica* rhizomes and leaves, where they seek refuge and feed. In particular, sites characterized by the highest shoot densities, such as those at 15 m in SMC and at SL in CIM, were characterized by omnivores species, such as *D. vulgaris* and *D. sargus*, feeding on the most abundant invertebrates in the *Posidonia* canopy, i.e., decapods and amphipods [64,88]. Moreover, juvenile stages of the Sparidae species were commonly present among *P. oceanica* shoots, where they can easily find shelter and food, confirming the role of *P. oceanica* as a nursery habitat [38]. Sites characterized by the highest leaf lengths, such as those at 15 m in both SMC and CIM, were related to seagrass resident species [89], mainly belonging to the Labridae and Serranidae families, such as *C. julis, S. tinca, S. scriba*, and *S. cabrilla*, that find shelter and food within the canopy throughout their entire life cycle.

## 5. Conclusions

The present study has provided useful knowledge about the role of *P. oceanica* beds in structuring fish communities and in estimating the effectiveness of an MPA. Low fish densities pointed out some issues at both the Santa Maria di Castellabate and Costa degli Infreschi e della Masseta MPAs, probably due to inappropriate zoning design and legal or illegal fishing pressure. It might be essential to reconsider the SMC zoning design, moving the fully protected "A" zone to the wide P. oceanica beds of Punta Licosa, currently included in the "B" zone, in order to adequately protect this key feeding and nursery area, with possible consequent spillover effects in the next years. This new, fully protected (A) zone should be established within the area characterized by the highest *Posidonia* shoot densities, due to the evident correlations with the Sparidae species, such as D. sargus and D. vulgaris, that are among the most valuable commercial species. As suggested by [80], the results confirm that MPA fish assemblages are threatened also by relevant fishing pressure, primarily performed by longlines at SMC, for the lack of piscivorous species, and trammel nets at CIM, for the high heterogeneity of trophic groups and very low biomasses. Therefore, to enhance conservation and, concurrently, to ensure the survival of local economies, an overall better management and an increased surveillance would be desirable to allow the constant development and maintenance of fish assemblages. These findings highlight the urgent need to carry out up-to-date studies on fish communities at a regional scale to identify possible MPA management issues. Moreover, the idea of a new MPA concept is recently emerging, based on flexible and changeable zoning that considers anthropogenic threats at both global (climatic change) and local (fishery) scales. These studies would also provide useful guidelines to establish new potential MPAs or to enhance the effectiveness of those already existing.

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