USING DIVER-OPERATED STEREO-VIDEO TO MONITOR JUVENILE FISH ASSEMBLAGES IN MEDITERRANEAN COASTAL HABITATS FORMED BY MACROPHYTES

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Abstract – Temperate coastal ecosystems dominated by macrophytes are diverse, ranging from macroalgal forests to seagrass meadows. They provide numerous ecosystem services and are highly valuable as nursery areas for fish. However, these habitats are under increasing pressure due to anthropogenic actions.

In the last decades, the use of Diver Operated Stereo-Video (stereo-DOV) systems has become popular for making accurate, precise, and repeatable measurements of fish lengths. The objective of our study is to develop an efficient video-based methodology for studying the patterns of recruitment of temperate littoral fishes at various temporal and spatial scales. We aim to evaluate our methodology compared to other non-invasive techniques used for the study of juvenile fish assemblages.

In this paper, we propose a combination of habitat mapping and stereo-DOV transect methodologies and test them in two case studies to investigate the link between juvenile fish and macrophytes, considering various scales of heterogeneity of littoral habitats. Data on species composition, abundance, and mean total length of the juvenile assemblages and canopy height from each habitat can be obtained and geo-referenced in the sampling sites, mapping their distribution in space and time to understand recruitment patterns.

Some benefits of using stereo-DOV transects and habitat mapping for monitoring fish assemblages in comparison with traditional non-invasive methods used for fish counts, are a) videos are permanent, they can be reanalyzed; b) divers do not have to be experts in fish and macrophyte identification; c) inter-observer variability can be controlled through quality checks; d) high accuracy and precision in fish count and length measurements; and e) image data can be used for developments of artificial intelligence (AI). However, there are also some disadvantages: a) cameras have a limited field of view; b) video processing is time-consuming; and c) experts are needed for fish identification during image processing.

In conclusion, the combination of stereo-DOV and habitat mapping is an effective methodology for monitoring fish recruitment patterns and habitats at different scales. It can be also advantageous for evaluating temporal changes in the habitat structure, and therefore, in the ecosystem functions provided for the fish community.

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Introduction

Mediterranean shallow coastal ecosystems are diverse, ranging from rocky to sedimentary substrates, usually dominated by the presence of structurally complex marine macrophytes such as macroalgae (e.g., *Cystoseira* sp., *Halopteris* sp., or *Dictyopteris* sp.,) or seagrass (e.g., *Posidonia oceanica*). Macroalgal forests and seagrass meadows provide numerous ecosystem services and have been proved to be key nursery habitats, essential for the life cycle maintenance of many coastal fishes in the Mediterranean Sea [3] [13]. Several studies suggest a positive relationship between macrophyte three-dimensional structural complexity and its key role in fish recruitment, by providing increased potential prey and shelter from predators [4] [10] [14] [18].

However, these habitats have suffered a severe regression in the last decades due to an increasing pressure created by anthropogenic factors such as eutrophication, mechanical disturbances, climate change, or the introduction of invasive species, among others [4] [5]. Habitat transformation may have consequences on the recruitment of coastal fish assemblages [2]. Detailed data on the factors affecting the abundance of juvenile fish and their spatio-temporal patterns (e.g., habitat complexity, temperature, or depth) is needed for understanding the dynamics of these communities.

In the last decades, the use of Diver Operated stereo-Video (stereo-DOV) systems has become popular for making accurate, precise, and repeatable measurements of fish abundance and lengths [9] [15]. This method gives permanent information on species and related habitats than can be reanalyzed as many times as necessary. It also allows making precise measures of fish density and biomass. These properties are key to understanding the long-term evolution of population dynamics in an environment subjected to multiple stressors such as the infralittoral strip. While different sampling methodologies of stereo-video have been used to study adult fish communities [17], this method has been rarely used for evaluating the spatio-temporal dynamics of fish recruitment in shallow habitats of different complexity in temperate seas.

The objective of our study is to develop an efficient video-based methodology for studying the patterns of recruitment of temperate littoral fishes at various temporal and spatial scales. In this paper, we describe and evaluate our sampling methodology and explore some of the preliminary data collected in two case studies: a restored *P. oceanica* meadow and rocky reefs covered by macroalgae.

Materials and Methods

Stereo-DOV censuses and habitat mapping

Samplings for juvenile fish and associated habitats were performed 50 cm above the seabed by SCUBA divers operating a low-cost calibrated, un-baited, stereo-DOV system (Fig. 1). Associated GPS track data was taken from the surface by a snorkeling diver, shadowing the stereo-DOV position to attain a more precise geo-location measure of each fish or macrophyte measured. The system consisted of two GoPro HERO7 Black cameras (GoPro, Colorado USA) in waterproof housings, set to record on the wide field of view mode at 30 frames per second with a 1440-pixel resolution to optimize video quality and battery consumption. Previous trials with differing inter-camera distances and angles were carried out to determine the field of vision that would provide an accurate estimate of juvenile fish and macrophyte sizes. As a result of them, cameras were mounted separated from each other by 34 cm and with a 6° inward inclination angle which gives a wide field of view of 1.5 m. The stereo-DOV systems were calibrated in a pool before the sampling, and the processing of the calibrating videos was performed with CAL SeaGIS software (www.seagis.com.au).



Figure 1 – SCUBA diver operating the stereo-DOV system.

We considered juveniles all individuals of each species recorded smaller than onethird of the adult maximum total length (TL) [1] [6] [16]. Data on every fish that was observed on each transect was obtained from the continuous analysis of the stereo-pairs of videos with SeaGIS EventMeasure software Version 3.22 (specific software to count and identify fish) (Fig. 2). Species composition, abundances, and mean TL of the juvenile fish assemblages, as well as habitat structural parameters, were also obtained from the videos. To ensure accuracy and precision, fish total lengths were only measured when individuals had their bodies straight and were located within the field of view of both cameras. Only fish situated less than 7 m away from the camera were considered for the analysis. Fish individuals that did not follow those rules were discarded and were just identified and counted but not measured. Each fish observed and identified (at species level when possible) was georeferenced in the sampling site with QGIS software Version 3.18.2 (QGIS Association. http://www.qgis.org), mapping the fish distribution in space and time to understand the habitat use of juvenile fish in a defined area.

Two case studies (CS) were carried out for testing the versatility of this methodology: CS-A) recovery of the nursery function in a shallow restored *P. oceanica* meadow and comparison with the adjacent meadows (Pollença Bay; NE Mallorca); and CS-B) seasonal variation of juvenile fish assemblages from three shallow rocky reefs, dominated by the presence of macroalgal forests (SW Mallorca) (Fig. 3). Habitat types from both CS sites were identified and mapped before the beginning of the sampling period to obtain a context of the seascape characteristics (e.g., habitat heterogeneity, boulder sizes and distribution, vegetation cover) on a broader spatial scale.



Figure 2 – SEAGIS EventMeasure screen capture of: a) TL measurement of *Symphodus tinca* and *Chromis chromis* individuals in a *Posidonia oceanica* patch located in a rocky reef; b) height measurement of various *Caulerpa prolifera* blades to calculate mean canopy height.



Figure 3 – Sites and habitats sampled (Mallorca, Balearic Islands, Spain).

Maps of the area of CS-A were created using QGIS, applying the Voronoi algorithm to the habitat information obtained from the videos taken using a Towed Stereo Video (TSV) system operated 50 cm above the seabed along 20 transects of 400 m separated by 25 m during the years 2018, 2019 and 2020. This algorithm is based on shortest distance constraints and generates a polygon layer containing the inputs points [19]. Then, a simplification of the layer was performed to smooth the geometries of the generated polygons. Due to the impossibility to use TSV because of the abrupt relief of the landscape of the CS-B, maps of the reefs were created by using sequential time-lapse underwater images of the seafloor, taken from the surface in the summer of 2021 with a GoPro HERO7 Black camera, along 10 transects of 270 m in "Cala Egos", 7 transects of 310 m in "Cala Llamp" and 7 transects of 200 m in "Es Total". Images were taken early in the morning to avoid the shade of the boat which hindered the process of analyzing the images for habitat mapping. Mosaics from the images were created with Microsoft Image Composite Editor software. With the habitat information obtained from the mosaics, a polygon layer was created with QGIS.

Case study A

We sampled a sedimentary bottom, between 4 to 8 m in depth, covered by P. oceanica at different vegetative development stages and structural complexities, quantified as shoot density and size, and canopy height. Between March 2018 and March 2020, 12 800 P. oceanica fragments were replanted in a 2-ha area over a dead matte substratum, about 200 m North from an extensive living meadow. The sampling sites included an undisturbed meadow (inner and edge sections, structurally complex), the restored meadow (with transplanted rhizome fragments, structurally simple), and a dead matte meadow now colonized by the seagrass Cymodocea nodosa and the macroalgae Caulerpa prolifera (structurally simple) (Pollença Bay; NE Mallorca) (Fig. 3). These meadows are being sampled eventually during the summer season, starting from August 2021. On each sampling date, 7 transects of 7 minutes were performed in the three study habitats. Preliminary trials and analysis demonstrated that the number and duration of transects were sufficient to obtain a representative sample of the fish community of the study area with at least 90 % of the species present in each transect area. Transects were orientated in parallel to each other, separated by 10-20 m. Due to the habitat characteristics, depth among transects was constant in each area (4-5 m in the restored area; 6-8 m in the dead matte and the undisturbed meadow). Transects were not necessarily parallel to the coastline. Seawater temperature data were recorded on every sampling location and date.

Case study B

We sampled three rocky reefs of variable structural complexities, quantified as morphotype diversity [14] and canopy height: "Es Total" and "Cala Egos" as structurally simple habitats (1 km between sites), and "Cala Llamp" as structurally complex habitat (3-4 km to the other sites) (SW Mallorca) (Fig. 3tab). These reefs were sampled monthly, starting from July 2021. The same protocol as in the CS-A was followed (7 transects of 7 minutes by zone). However, in this case, the transects were performed in parallel to the coast following the depth profile of the rocky reef. The lower limit of the rocky reef, where sedimentary habitats commenced, was found at 7 meters at all three sites, therefore the depth of transects ranged from 7 m to 0 m. Transect dives were carried out at one-meter-depth intervals starting

from the 7-6 m isobaths towards the shallowest transect at 1-0 m depth. Seawater temperature data were also recorded on every sampling location and date.

Results

Stereo-DOV censuses and habitat mapping

For creating the map of the CS-A sampling site, the 20 transects carried out using TSV information (seagrass meadows) covered an area of 191000 m² (Fig. 4). In the CS-B, the area covered with time-lapse images (rocky reefs) was 14000 m² on "C. Egos" (10 transects), 6150 m² on "Es Total" (7 transects) and 9900 m² on "C. Llamp" (7 transects) (Fig. 4). The mapping fieldwork took approximately 15 minutes per transect in both CS-A (TSV technique) and CS-B (time-lapse technique). The time required for posterior image processing was lower for TSV information (1 h/transect) than for the time-lapse images (3 h/transect). The optimal conditions to take the TSV and time-lapse images and create the maps require good weather (low wind velocity and absence of waves) and good underwater visibility (>15 m).



Figure 4 - Maps obtained for: a) CS-A study area; b) CS-B "C. Egos" area.

The stereo-DOV sampling covered an area of 139.5 $m^2 \pm 11.2 m^2$ per 7-minute transect in CS-A zone and 158.4 $m^2 \pm 21.7 m^2$ in CS-B zone. The total study area covered per sampling site in 1 hour of diving was approximately 976.5 m^2 in CS-A and 1108.6 m^2 in CS-B. A team of 4 divers was able to sample 3 sites per day, covering an area of 2929.5 m^2 in 3 h of effective diving in CS-A and 3325.8 m^2 in CS-B. Posterior video processing with SeaGIS EventMeasure took approximately 2 h per transect, including fish and macrophytes identification and measurement after training. The optimal conditions for the sampling require luminosity, good underwater visibility, and the absence of waves.

The stereo-DOV system was able to deliver high precision measurements for the juvenile fish density (in this case number of individuals by area) and fish total lengths, as well as habitat structural parameters such as macrophyte canopy heights or boulder sizes, among others.

To date, videos from August 2021 have been analyzed for the three habitats included in CS-A study, and videos from July and August 2021 have been analyzed for the three areas of study of CS-B. In this preliminary analysis, the stereo-DOV system was able to detect juveniles with a minimum TL of 1.1 cm belonging to 18 species of 6 families (Table 1).

Family	Species	Rocky reefs			Seagrass meadows		
		Cala	Es	Cala	Undis-	Re-	Dead
		Llamp	Total	Egos	turbed	stored	matte
Atherinidae	Atherina sp.		35-39	37-39			
Labridae	Coris julis	21-66	24-70	18-67	50-67		
	Symphodus		67.9				
	mediterraneus						
	S. ocellatus		31-65	25-59			
	S. roissali	34-66	22-65	28-70			
	S. rostratus		64-67		67		
	S. tinca	30-70	17-69	19-67	42-69	22-58	
	Thalassoma pavo	51.2	46-60	47-59			
Mullidae	Mullus surmuletus	71-76	40-82	36-93	51	47	
Pomacentridae	Chromis chromis	18-36	12-39	11-36			
Serranidae	Serranus scriba	85		80-99			
Sparidae	Diplodus		67-79	39-75	12-79		
	annularis						
	D. puntazzo			48.2			
	D. sargus sargus	62.7	58-98	82-99			97
	D. vulgaris	57-96	51-98	44-99	44-99		
	Oblada melanura	33-75	15-99	21-92			89-99
	Sarpa salpa	78-97	47-99	39-65			
	Spondyliosoma				44-69		
	cantharus						

Table 1 – Range of TL (mm) of the juvenile fish found during the sampling.

Case study A

In the videos analyzed from the seagrass meadows (August 2021), a total of 9 species were observed (2 in the restored zone, 2 in the dead matte, and 7 in the undisturbed meadow) (Table 1). The abundance of juvenile fish in the restored meadow and the dead matte area was very low. Higher abundances were found in the natural *P. oceanica* meadow, indicating the relevance of mature and undisturbed meadows for fish recruitment.

Case study B

In the videos analyzed from the macroalgal forests (July and August 2021), 17 species were observed (11 in "C. Llamp", 15 in "Es Total", and 15 in "C. Egos") (Table 1). The abundance of juveniles was similar between the three areas in both months analyzed. Differences in the depth preference for recruitment are yet to be analyzed.

Discussion

According to the preliminary results of our study, we can consider that the video-based methodology proposed here is appropriate to identify and count recently settled juveniles (TL>1.1 cm) of a great number of species that recruit in temperate shallow areas [8]. It also provides valuable information about the composition, cover, and height of the macrophytes present in the study area. However, it presents some limitations such as the restricted area recorded by the cameras. Habitat mapping allowed us to obtain the spatial information that is lacking in the videos.

Shallow macroalgal forests and seagrass meadows are both important nursery areas for many littoral fishes, including commercial species, as previously reported by other studies [2] [7]. Although some studies have already determined the microhabitat use and seasonality of littoral juvenile fishes in the north-western Mediterranean [8] [11], with this new approach we will be able to provide updated, integrated, and precise data on the settlement patterns at various spatial scales, including a broader seascape context. Moreover, with this methodology, we will be able to provide precise information on temporal patterns for most littoral species inhabiting NW Mediterranean waters depending on environmental factors such as depth, temperature, or habitat structural complexity (e.g. changes in macroalgal or seagrass canopy height through the year).

The versatility of stereo-DOV allows using it in a wide variety of temperate littoral nursery habitats in comparison with other stereo-video techniques. Some other video-based techniques used for studying adult fish communities, such as TSV or Remotely Operated Vehicles (ROV), present a similar adult fish detectability potential as stereo-DOV [15], but may not be appropriate for studying juveniles, as they need to be operated higher above the substrate, which may result in a poor capacity to observe small fish. Moreover, remote underwater stereo-video (RUV) is also not appropriate for the study of juvenile fish due to their low density and patchy recruitment patterns [8], which would require a great number of replicates. Transects of stereo-DOV covering bigger areas are more adequate for identifying juvenile initial recruitment patterns (TL 1-2cm) and subsequent dispersion and habitat changes with growth. Also, stereo-DOV can orientate towards fish within the transect providing higher quality images for better identification and size determination.

Moreover, the combination of stereo-DOV (and SeaGIS EventMeasure) and habitat mapping presents several benefits compared to traditional methodologies used for the study of juvenile fish communities (e.g. UVC). Some of the benefits are: a) videos are permanent, so they can be reanalyzed as many times as necessary; b) divers do not have to be experts in fish and macrophyte identification, as pictures can be consulted with experts if verification is needed; c) inter-observer variability can be controlled through quality checks; d) accurate and precise counts and measurements of fish and macrophytes lengths can be made independently of the diver experience; e) the observation of fish can be geo-localized on an individual level in the particular habitats they are, while with other methods fish are usually integrated over the whole transect despite the different micro-habitats present along the transects; f) a context on the seascape is obtained from the maps, allowing to estimate spatial variables that may be essential for recruitment; g) habitat complexity and heterogeneity can be measured in an easy way directly from the maps and the video images, decreasing fieldwork efforts (e.g. macrophyte % cover or canopy height); h) videos can be revaluated and the attained image data can serve as a data source for artificial intelligence (AI) applications that aim to automate the image analysis (identification of species and quantification); and i) stereo-videos allow to make precise measures of fish density and biomass, not possible to obtain with other videobased methods such as non-stereo or baited cameras.

However, there are also some disadvantages compared to other methodologies: a) cameras have a limited field of view; b) cryptic species are difficult to observe with this methodology; c) video processing is time-consuming; and d) experts are needed for fish identification during image processing. Further efforts are needed to reduce the time cost of processing the video imagery through the automation of identification and measurement of fish [17].

Conclusion

Using a combination of the stereo-DOV and habitat mapping is an effective methodology for monitoring the spatio-temporal patterns of recruitment and habitats of preference for juvenile fish of temperate coastal areas at different scales. The methodology can be also advantageous for obtaining long data series and evaluating possible temporal changes in the habitat structure, and therefore, in the ecosystem functions provided for the fish community.

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