

UNIVERSIDADE DO ALGARVE

FACULDADE DE CIÊNCIA DO MAR E AMBIENTE

ALGARVE ARTIFICIAL REEFS FISH ASSEMBLAGES AND TROPIC ECOLOGY: IMPLICATIONS FOR THE LOCAL NEAR SHORE FISHERIES

(Tese para a obtenção do grau de doutor no ramo de Ciências e Tecnologias das Pescas,
especialidade Dinâmica de Populações)



FRANCISCO MIGUEL DE SOUSA LEITÃO

Orientador: Doutor Miguel Neves dos Santos

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Constituição do Júri:

Presidente: Doutor Rui Santos

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Faro
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*“New opinions are always suspected and usually opposed,
without any other reason but because they are not already
common”*

(Unknown author)

To Claudia and Maria
and to my mother, father and sisters

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Ictiofauna e Ecologia Trófica nos Recifes Artificiais do Algarve: Implicações para as Pescarias Locais

RESUMO

O presente trabalho teve como objectivo estudar: i) a colonização e a ictiofauna dos recifes artificiais (RA), ii) a contribuição dos RA para a ecologia trófica de *Diplodus sargus* e *Dicentrarchus labrax*, iii) o contributo dos RA para o aumento das pescarias litorais iv) a influência das comunidades bentónicas na estrutura populacional de sparídeos (*D. bellottii*; *D. sargus*; *D. vulgaris*). O processo de colonização foi rápido, devido ao deslocamento de peixes dos RA mais antigos para os novos RA e à atracção trófica. A dieta de *D. sargus* foi grandemente constituída por fauna bentónica, existente nos RA. O *D. labrax* alimenta-se de juvenis de peixes demersais, atraídos para o RA. Para os *Diplodus*, os RA servem como áreas de alimentação, de recrutamento, crescimento e desova/reprodução. Neste sentido, podem ser considerados como “habitats essenciais para peixe”. A composição do habitat bentónico condiciona a densidade de *Diplodus*. A contribuição dos RA para o aumento da pesca depende da adopção de medidas de gestão eficientes. Estas devem ser orientadas para a adopção de estratégias de pesca que tenham em consideração a biologia e ecologia das comunidades ictiológicas mas igualmente, dado a sua importância para os peixes, a protecção dos habitats.

Palavras-chave: Colonização; *Diplodus* spp.; *Dicentrarchus labrax*; recifes artificiais; pesca artesanal; habitats essenciais para peixes.

TITLE OF THE THESIS:

Algarve Artificial Reefs Fish Assemblages and Trophic Ecology: Implications for the Local Near Shore Fisheries

ABSTRACT

The purpose of this dissertation was to study: i) the colonisation and fish assemblages of artificial reefs (AR), ii) the contribution of ARs to the trophic ecology of *Diplodus sargus* and *Dicentrarchus labrax*, iii) the role of newly deployed ARs as fishery enhancing tools for near shore sparid fisheries, and iv) the influence of AR benthic habitat structure on sparid fish assemblage composition (*D. bellotii*; *D. sargus*; *D. vulgaris*). Fish colonisation was fast due to the migration of fish from nearby old AR to new deployed ARs and to trophic attraction. The diet of the *D. sargus* was directly linked to AR fauna. *D. labrax* feed on juveniles of demersal species attracted to ARs. ARs are expected to contribute to enhance fishery production through transfer of energy produced or attracted to AR to fish growth. For *Diplodus* spp. the artificial habitats served as feeding, recruitment, growth and spawning/mating areas, and can thus be considered essential fish habitat. The composition of the benthic artificial habitat affects *Diplodus* assemblage's densities. The success of ARs in enhancing the fishery depends on the adoption of efficient management measures. The latter should be goal oriented to the adaptation of fishing methods that take into consideration the biology and ecology of ARs fish assemblages but also, given their importance to fish, the conservation of habitats.

Key-words: Colonisation; *Diplodus* spp.; *Dicentrarchus labrax*; artificial reefs; artisanal fisheries; essential fish habitats.

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CHAPTER 1

GENERAL INTRODUCTION AND OBJECTIVES

ARTIFICIAL REEFS

The building of artificial reefs (AR) goes as far back as the 1790's in Japan when fisherman placed large wooden bamboo frames to increase catches (Pickering 1996). Artificial reefs were primarily made from materials of opportunity. Due to their use in Japan AR, these were primarily defined as “*based on the evident analogy with natural reefs, the artificial reefs are artificial reef structures deployed in the sea area, intended for fisheries productivity enhancement*” (Thierry 1990). Thousands of artificial reefs have been constructed around the world, both in salt and fresh water, since their first use in the enhancement of fisheries in Japan in the late 18th century (Stone *et al.* 1991). The major areas presenting this kind of activity include the Mediterranean and Caribbean Seas, South-eastern Asia, Japan, North America, Australia and some islands in the South Pacific (for a complete review see Jensen (2002) and references therein). While AR have been used for a number of reasons, their deployment is generally associated with specific purposes such as to balance the impact of fishery overexploitation, for ecological reasons or to achieve economic goals. Seaman (1995) outlines 5 main purposes of AR: commercial fishing, recreational fishing, SCUBA diving places, environmental restoration and management. Examples of large applications of AR are diverse: anti trawling devices to mitigate environment effects of trawling on seagrass meadows, e.g *Posidonia oceanica* (Ramos-Esplá *et al.* 2000) and macroalgae (Choi *et al.* 2002), to improve habitat and food limitations (Spanier *et al.* 1990), to provide new fishing grounds and increase fish (Santos and Monteiro 2007) and shellfish production (Bombace *et al.* 1995; Posey *et al.* 1999; Breitburg *et al.* 2000), as a biofiltering systems to clean up fish farms (<http://www.ugc.edu.hk/rgc/rgcnews10/Pages/2b%20Biofilter-E.html>), and to protect shorelines, among others. Thus, based on their use and their consequent effects, several definition of AR have been put forth, each one reflecting the

goal for which they have been deployed (FAO 1986; Thierry 1990; Polovina 1991; Seaman and Sprague 1991; EARRN 1998). Seaman (2000) provides a generalist definition of AR: *“An artificial reef is one or more objects of natural or human origin deployed purposefully on the sea floor to influence physical, biological, or socioeconomic processes related to living marine resources. Artificial reefs are defined physically by the design and arrangement of materials used in construction and functionally according to their purpose. Items used in reef construction add vertical profile to the benthic (sea floor) environment”*.

There has been a general shift worldwide towards using materials dedicated solely for the creation of artificial reefs, whilst low cost materials are no longer used to face the waste disposal problem (e.g. tyre reefs). This allows for better designs and more effective reefs. Nowadays, the expansion of this activity is related to the evolution of the structures and materials used. In fact, large scale concrete multi reef systems with structured organization have increased worldwide in the last 20 years. Many of the world’s largest reefs have been deployed as part of a national fisheries program, such as in Japan, where large steel and concrete frameworks have been carefully designed to withstand strong ocean currents (Seaman 2007). Concrete has been found to be very favourable for artificial reef construction. It does not degrade in seawater, can be made to have neutral pH, is easily moulded, not easily moved once in place, but is hard to transport to the deployment site. Concrete can be made to have a texture comparable to natural reefs and develops very similar communities as natural reefs (Pickering and Whitmarsh 1997). In order to protect and restore biological resources, implementation of new measures or adaptations of existing management measures have become urgent at both local and regional scales. Because most fishing activities, either commercial or recreational are practiced in specific and sometimes small inshore areas, AR have been

envisaged as potentially interesting tools to deal with the availability and accessibility of marine resources.

AR ECOLOGICAL FUNCTIONING

ARs are deployed to enhance/replicate rocky areas. In this context, ARs may be seen as contributing to environmental production enhancement. As far as AR is concerned, their productivity relies on the assumption that habitat is a limiting factor and that AR surfaces provide additional critical habitat which increases the environmental carrying capacity, and therefore the abundance and biomass of marine biota (Polovina 1994) through increased growth and survival of juveniles (Bohnsack 1989).

After man made structures are deployed on the seabed they become an integral part of the natural aquatic habitat. The presence of hard substrate allows the settlement of hard substrate benthic invertebrates larvae and marine plants that otherwise would not survive beyond their crucial post recruitment life-cycle stages. These groups of species in turn might increase local trophic efficiency (Bombace 1989), in former low productivity and nutrient poor sandy bottoms (Leewis *et al.* 1997; Steimle *et al.* 2002). As far as reef assemblages are concerned, it is assumed that habitat limitation is the primary factor in determining the specific composition of fish assemblages (Coll *et al.* 1998) by availability of food or shelter (Bohnsack 1989).

AR fish colonisation starts immediately after the immersion of structures, with the attraction of fish from other areas. The most basic mechanisms for fish attraction are instinctive orientation response to structure or current (taxes or kinesis) and thigmotropic response (contact with objects). If fish find reefs attractive they will aggregate on them. During early AR colonisation stages, the hypothesis of fish production is not sustainable because of the initial lack of food (e.g. invertebrate fauna).

Therefore, if ARs are deployed for creating new habitats for fish resources enhancement, then the initial balance (removal by fishing and addition by reef production) will be negative overall in terms of fish mass balance. Nevertheless, after benthic fauna cover AR structures, fish production may occur due to trophic transfer from macrofauna taxa to higher trophic levels. Increased habitat usually produces a local increase in fish abundance as carrying capacity increases with reef size and decreases recruitment limitations as large reef area attract more recruits (Grossman *et al.* 1997). The attraction of fish from natural to AR areas also means the opening of space in donator's areas. Consequently, the re-colonisation of donators areas is expected to contribute to the increase of overall fish biomass. The success of AR and natural areas colonisation will mainly depend on larvae supply that in coastal temperate rocky waters, independently of area (Almada and Faria 2004) have been shown not to be a limiting factor.

AR deployment is expected to affect nearby sandy communities in various ways (Donavaro *et al.* 2002): (a) by altering the hydrodynamic regime and physical and chemical characteristics of the substrate. For instance, fish species can induce nutrient production in the water column, excreting substantial amounts of ammonium, urea and depositing organic material, which is then incorporated into the reef food web. AR physical structure changes the sea bottom's currents that can promote recycling of benthic nutrients, enhancing nutritional sources from the bottom waters (Parrish 1989; Moffitt *et al.* 1989; Falcão *et al.* 2007); (b) by modifying the distribution and/or composition of the available food sources (Bulleri 2005); the concentration of fish in AR can reduce the local abundance of their food sources and consequently change animal and algae community structure near AR due to predation (Davis *et al.* 1982; Ambrose and Anderson 1990; Kurz 1995; Grossman *et al.* 1997; Williams and Polunin

2001; Einbinder *et al.* 2006) and consequently; (c) by altering the biological interactions (e.g hard substrata-sandy; hard substrate-water column) between different parts of food web. This process implies the increase of productivity and diversity on a local scale by increasing biotic and abiotic habitat complexity (Ambrose and Anderson 1990), promoting biological colonisation and species richness in many ecological niches and food webs (Relini *et al.* 1994; Snelgrove and Butman 1994).

If AR try to replicate natural reefs comparison must be done with rocky areas and include a synthesis of birth rates, mortality, immigration/emigration and growth to interpret fluctuations in biomass overtime (Carr and Hixon 1997). AR do in fact mimic some attributes of rocky areas: they act as nursery areas providing shelter from predation (Anderson *et al.* 1989; Carr and Hixon 1997) and from tidal currents (Nakamura 1985; Collins *et al.* 1991; Spanier 1997); provide habitat for recruitment of individuals that would otherwise be lost from the population; they reduce harvesting pressure on natural reefs (Stone *et al.* 1979; Harmelin and Bellan-Santini 1996; Santos *et al.* 2007); they act as reproduction and spawning locations for benthic invertebrates, for mollusc (cephalopod) and fish (see Pickering and Whitmarsh 1997); they contribute to the creation of new feeding areas for many marine fish (Leitão *et al.* 2007). Due to the attraction effect they can also serve purely as aggregating devices, whereby the behavioural preferences of fish result in aggregation on and around artificial reefs without any increase in biomass (Bohnsack 1989).

Several studies (Bohnsack 1994; Santos and Monteiro 1998) suggest that artificial reefs are just as good as or better than natural reefs at either supporting or attracting fish. However, according to Bohnsack (1994) it is difficult to determine why this might be the case. “For example, there is no way to discern (1) whether fishes that settle or that were attracted to the artificial reefs would have found suitable habitat if the

reefs were not present; (2) whether fishes had better survival or faster growth at artificial reefs than in natural habitats; (3) whether foraging success and food web efficiency was improved by artificial reefs; and (4) whether habitat vacated by fishes moving to artificial reefs was reoccupied so that total population size increased". This leads to the debate of attraction, and redistribution of existing individuals, versus production on artificial reefs. The attraction and production are not mutually exclusive and can be considered extremes along a gradient. While artificial reefs may merely attract and concentrate some fishes, they may promote the production of others. The species most likely to benefit in terms of increased biomass are habitat limited, demersal, philopatric, territorial and obligatory reef species (Bohnsack 1989). Most fishes probably lie somewhere between the two extremes (Bohnsack 1989). However, there is little value in discussing whether fish assemblages are attracted or produced at AR as probably these man-made structure can contribute to both. According to Osemberg *et al.* (2002) the challenge lies in defining an effective, and pluralistic, conceptual framework that allows us to quantify the production of a system, partition that production to the respective habitats (natural and artificial), and evaluate how much harvesting the aggregate system can likely sustain. According Brickhill *et al.* (2005) inappropriate experimental designs have prevented many studies from discriminating between the attraction-production processes. According to the latter authors to ultimately determine whether attraction and production is responsible for increased abundances on reefs, requires two experimental features: 1) control sites, both interspersed among artificial reefs and at reef and non-reef locations outside the test area and 2) incorporation of fish age and length data over time. Techniques such as otolith microchemistry, telemetry and stable isotope analysis can be used to help resolve feeding and movement mechanisms driving attraction and production.

FISHERIES AND AR OFF THE ALGARVE-BRIEF OVERVIEW

In the Algarve (Southern Portugal, Figure 1) the fishing fleet is divided into inshore (*local* or artisanal), coastal (*costeira*) and long-distance (*largo*) components. The local/artisanal fishery is socially and economically important for the local population that has increased along the Algarve over the last decades. In the Algarve the fishing effort of the artisanal fleet on many fisheries resources is very intense and a great number of fishermen are involved. The calmness of the sea during most of the year, favors fishing activity, particularly the use of fishing gears set close to the shore, along the continental shelf sandy bottom, contributing to this overfishing scenario. In the south coast of Portugal the local/artisanal fleet consists of 854 fishing boats (DGPA 2005). The artisanal fleet is mainly composed of simple open fibreglass boats that have licenses to use more than one gear (e.g. gill/trammel nets, traps/pots and longline, seine and dredges). Among the most used are monofilament and multifilament gill nets (740 licenses) and trammel nets (604 licenses), the longline (1504 licenses) and the tramps/pots (568 licenses).

Taking into consideration the socio-economic importance of coastal artisanal fishing in Algarve in 1990, the National Fisheries Institute (IPIMAR) started to deploy AR. These AR were purposefully constructed to enhance the fisheries and simultaneously were expected to contribute to the protection of juvenile fishes that migrate from the lagoon systems located along the south coast (Monteiro and Santos 2000). Results of scientific fishing surveys (gill net) carried out during the first four years after AR deployment showed that fishing yields (1.5-2.1 times) were higher in AR than in control areas (Santos *et al.* 1995). These results motivated the increase of the AR area in two posterior phases, the first in 1997-1998 and more recently in 2002-2003. Presently, seven artificial systems cover an area of 43.5 km², the largest area of this

type (20 748 modules) implemented in European waters over the last two decades (Figure 1).

The grounds for initiating this programme and for implementing it on the southern coast of Portugal included the presence of several highly productive lagoon and estuarine systems in this region, the relative scarcity of natural reefs (especially on the south-eastern part of this coast), the high fishing intensity offshore, and the need to provide alternative means that would minimise the effect of fishing in order to yield a sustainable management of this coastal region (Monteiro and Santos 2000). Recent studies have corroborated initial expectations regarding AR fishing enhancement with fishing yields surpassing by 1.8 to 2.6 times the ones in control areas and aggregating higher species abundance (Santos *et al.* 2007). Moreover, AR deployment did not change the composition of the fish assemblage caught by gill nets since the relative proportion of different functional groups (demersal, pelagic and benthic) remained stable. Therefore, AR seem to have contributed to enhancement and diversification the catches, thus contributing to improving local artisanal fisheries, which play a major role in these region (Santos *et al.* 2007). In addition, economic analysis shows that the fishing revenues in artificial reef areas are expected to surpass those in natural areas, showing AR capacity to increase fishing incomes in coastal fisheries (Whitmarsh *et al.* 2008). However, as expected, different stakeholders take somewhat different positions and attitudes towards AR impacts: usually scientists are the most optimistic, whereas fishermen take the most sceptic view (Ramos *et al.* 2007).

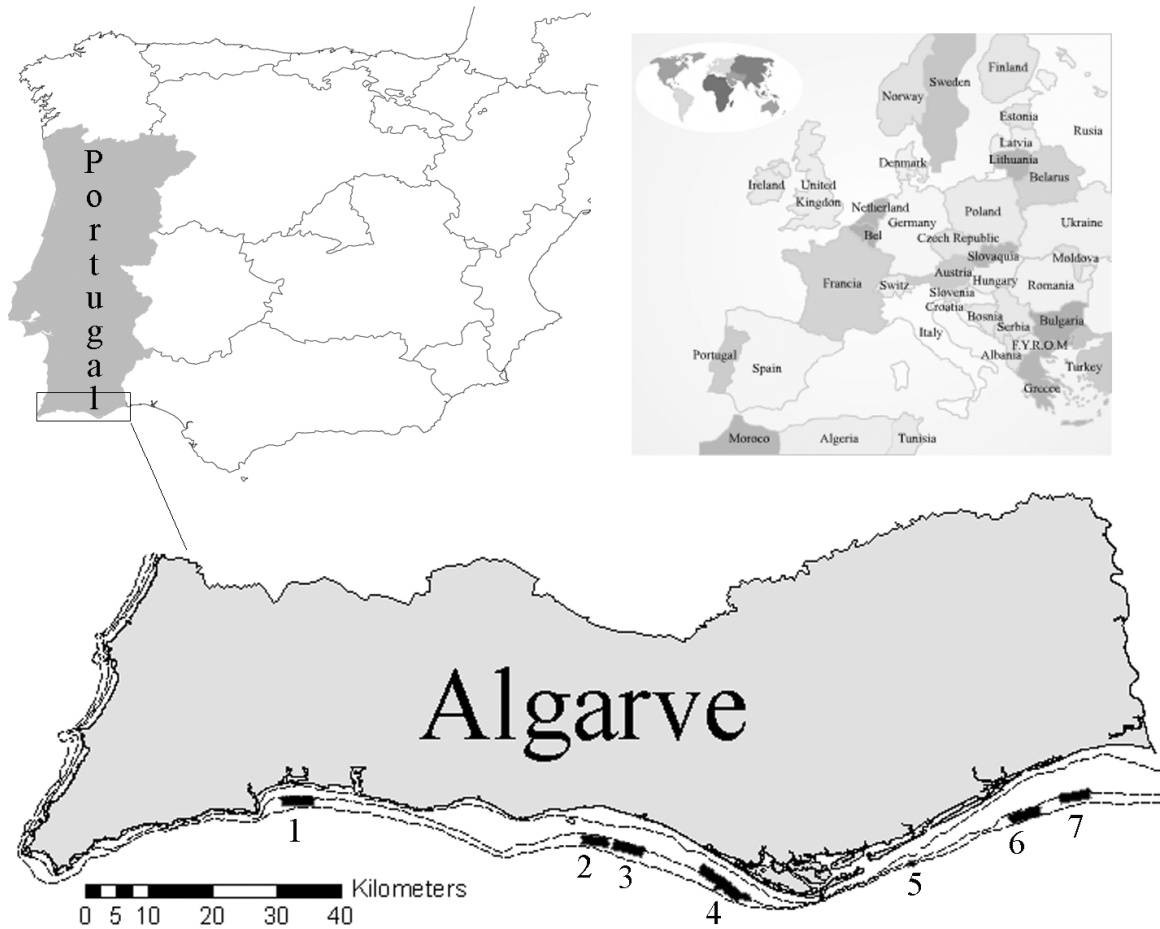


Figure 1. Map showing Algarve (South Portuguese Coast) artificial reef systems in relation to the 10, 20 and 30m isobaths. 1- Alvôr; 2- Oura; 3- Quarteira/Vilamoura; 4- Faro/Ancão; 5- Olhão; 6- Tavira; 7- Cacela.

Sparids species, and namely *Diplodus* spp. genera are economically important in Portugal at both national and regional scales. In fact, sparids, mainly *D. vulgaris* and *D. sargus* are particular important in the south coast of Portugal, being among the most important landed coastal species, ranked nine and ten of 40 commercial species in 2006. The catches of *D. vulgaris* and *D. sargus* in the Algarve have been in a steady state between 1997–2006, with average landings of 92 (coefficient of variation= 12.6%) and 337 (coefficient of variation= 19.3%) tones per year, respectively (DGPA 1997-2006). Experimental longline fishing trials in the Algarve showed that the dominant species in

terms of weight (29%) was the highly valuable *D. sargus* (Erzini *et al.* 1996). Moreover, *D. vulgaris* is also important in terms of artisanal fisheries being a common target for gill nets and longlines and accounting for 5.2 and 23.3% of the catches in number, respectively (Erzini *et al.* 1998, 2003). The bass, *Dicentrarchus labrax*, is also a very high value commercial species, with both values and catches increasing slightly from 1997 (33 tones) to 2000 (112 tones), remaining stable thereafter despite the slight decrease of the catches between 2001 and 2006 with average landings of 74 tones per year (coefficient of variation= 13.3%).

RATIONALE AND OBJECTIVES OF THE STUDY

As AR are considered to contribute to fishing enhancement, due to attraction and production of new biomass, they have been proposed as a tool to further contribute to the management of the heavily exploited near shore fisheries off the Algarve coast (Monteiro and Santos 2000; Santos *et al.* 2007). If AR in Algarve were deployed to enhance fish populations it is important to question their effectiveness, especially when the strong caveat regarding fish production has not been addressed. One of the mechanisms suggested that AR may increase fish biomass production by providing additional food (Bohnsack 1989). Therefore, for fish species that feed on food available on AR, man made structures that produce significant benthic biomass may be useful for increasing fishery resources, through the biomass gain by fish from AR. Thus, knowledge of the trophic ecology of reef assemblage is important for understanding the dependence of the species on the AR benthic production and for evaluating the importance of such structures for the maintenance of fish populations. This means that the effective use of AR to enhance fish populations along the Algarve coast requires the understanding of the ecological role of reefs in supporting exploited fish assemblages. The above necessarily requires the knowledge of the fish assemblages structures and

their dynamic (e.g. seasonal patterns). Although several studies on Algarve AR have focused on different aspects of the reef fisheries (Santos and Monteiro 1997, 1998; Santos *et al.*, 2005), ecology (Santos *et al.* 2002; Boaventura *et al.* 2006; Moura *et al.* 2007) and economics (Ramos *et al.* 2006a,b and 2007), knowledge of the role of AR as fish production areas, available exploitable biomass and demographic aspects, and fish-AR interactions, is still limited. The present work aims to contribute to the knowledge on AR, namely through the:

- Study of fish assemblages colonisation, characterization of abundance/biomass and size structure variation of reef assemblage patterns along time and the role of AR deployment on changing local fish assemblages.
- Evaluation of the contribution of AR to the trophic ecology of commercial species (*Diplodus sargus* and *Dicentrarchus labrax*) and to AR long term fish production.
- Evaluation of the potential of the most important *Diplodus spp.* schools to near shore fisheries.
- Understanding the importance of benthic fauna and flora on determining *Diplodus* reef associated assemblages.

This thesis is structured in chapters containing the compilation of articles (Chapter 2 to chapter 7) published or submitted for publication in the scope of the present study.

The specific objectives of these chapters were the following:

- Chapter 1 is the general introduction and objectives of the study, briefly reviewing the AR functioning, their role in relation to the local fisheries, specifically AR importance in the context of near shore fisheries in Algarve;

- Chapter 2 aimed to describe reef fish assemblage colonisation, to characterise reef species according to their habitat use (fidelity), density, biomass and size structure of reef assemblages and their patterns of variation (year and season);
- Chapter 3 and 4 described the importance of AR for the feeding of *D. sargus* and *D. labrax*, respectively and to fish production;
- Chapter 5 is a case study that describe the colonisation process, assesses the role of the AR in terms of juvenile recruitment and growth and as mating/spawning areas and evaluates the AR potential as near shore artisanal fishing grounds for three economically important fish species, *Diplodus bellottii*, *D. sargus* and *D. vulgaris*.
- In chapter 6 the effect of habitat structure across reef age on fish densities (*Diplodus bellottii*, *D. sargus* and *D. vulgaris*) was tested.
- The major results and conclusions obtained from the various chapters are discussed in chapter 7.

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CHAPTER 2

Fish assemblages and rapid colonisation after enlargement of an artificial reef off the Algarve coast (Southern Portugal)

Leitão F, Santos MN, Erzini K, Monteiro CC (2008) Fish assemblages and rapid colonisation after enlargement of an artificial reef off the Algarve coast (Southern Portugal). *Marine Ecology* 29: 435-448.

ABSTRACT

Artificial reefs (ARs) have been deployed in Algarve (Southern Portugal) coastal waters in order to contribute to the sustainability of local near shore fisheries. Herein, we describe the colonisation process of the recently deployed Faro/Ancão AR, assess time until the fish assemblage reaches stability and the seasonal patterns. In addition, we compare the results from the present study to those previously reported for an older AR. The fish assemblages were monitored monthly over a two year period by means of visual census. A rapid increase in fish colonisation occurred within the first 4 months. After this initial period the assemblage structure showed high similarity (> 73%). The high rate of colonisation of the AR was related to the maturity already achieved by the nearby 14 year old AR and with the fish migration from the Ria Formosa lagoon, a nearby nursery habitat. The reef fish assemblage structure showed a seasonal pattern, mainly associated with recruitment episodes of occasional demersal species (*Boops boops*, *Trachurus trachurus* and *Pagellus* spp.) in spring and summer. A total of 66% of the species found in AR are of commercial and recreational importance. The overall mean density and biomass were 2.8 ind/m³ and 207 g/m³. The occasional demersal species accounted for 42% of the fish density. The most important species in terms of biomass belong to the Sparidae family along with *Dicentrarchus labrax*. The fish assemblage of the new ARs showed higher mean number of species, diversity, density and biomass values than those reported for the older AR. This result was associated with enlargement of the AR area and to the fishing exploitation of the isolated, small and patchy old AR. Moreover, the high biomass values recorded in the new ARs were mainly due to the increased density of *D. labrax* after AR enlargement. The results of the present study are used to define guidelines for suitable management strategies for the AR areas that are exploited by the local commercial and recreational fisheries.

INTRODUCTION

Artificial reefs (AR) have multiple purposes, including the enhancement of fishery resources and the rehabilitation/mitigation of habitat loss (Bohnsack and Sutherland 1985; Monteiro and Santos 2000; Ramos-Esplá *et al.* 2000). In the Algarve, southern Portuguese coast, an AR program aiming at the sustainability of local near shore fisheries has been established since 1990.

The Algarve ARs have been shown to have an important role, serving as protection areas for juveniles that migrate from surrounding nursery areas (Santos *et al.* 2005), as feeding areas (Leitão *et al.* 2007), enhancing fishing yields (Santos and Monteiro 1997, 1998), and contributing to the rehabilitation of coastal areas through trophic chain pull-up (bottom-up production) (Falcão *et al.* 2007). As ARs are considered to increase fishing yields, through attraction and enhanced sustainability and to increase fish biomass due to benthic biomass production, they have been proposed as a further contribution to the management of the heavily exploited near shore fisheries off the Algarve coast. Currently the overall area of the Algarve ARs is 43.5km².

As reported by Bortone and Kimmel (1991) knowledge of different aspects of the reef fish assemblages is important for evaluating community responses to natural and artificial changes in the biotope. Several of these aspects have been studied by many authors: species composition and colonisation (Coll *et al.* 1998; Reñones *et al.* 1998; Relini *et al.* 1994); recruitment (García-Rubíes and Macpherson 1995; Harmelin-Vivien *et al.* 1995); trophic (Herrera *et al.* 2002; Leitão *et al.* 2007) and fisheries aspects (Solonsky 1985; Santos and Monteiro 1997, 1998).

Artificial reef fish assemblages have been studied mainly with artisanal fishing gear (D'Anna *et al.* 1994; Santos and Monteiro 1997, 1998). However, data collected by visual census (SCUBA) allows the efficient and precise spatio-temporal study of AR

fish assemblages (Bortone and Kimmel 1991). Most of the studies in the Mediterranean using visual census methods describe colonisation, the influence of seasonality, and the effects of AR structural complexity on fish assemblages (Bohnsack *et al.* 1991; Ody and Harmelin 1994; Relini *et al.* 1994; García-Rubíes and Macpherson 1995; Reñones *et al.* 1998; Jensen *et al.* 2000 and references therein; Ramos-Esplá *et al.* 2000).

Although several studies have focused on different aspects of Algarve AR fisheries (Santos and Monteiro 1997, 1998, 2007) and ecology (Santos *et al.* 2002; Boaventura *et al.* 2006; Moura *et al.* 2006, 2007), information regarding the fish colonisation process (Santos *et al.* 1996) and the structure of reef fish assemblage is still limited (Santos *et al.* 2005). Given the fishing potential of the AR, the knowledge on latter aspects is of considerable importance for defining suitable exploitation strategies and management for the AR areas.

The AR located off Faro was significantly enlarged in 2002. Recognizing the limited amount of work previously done, herein we describe the process of reef fish colonization of the newly deployed Faro/Ancão AR, in terms of the species composition, diversity, density, biomass and assemblage size structure (juveniles *vs.* adults). Moreover, we assess time until AR fish assemblage stability is reached and the existence of reef fish assemblage seasonal patterns. In additional, we compare the results from the present study to those previously observed in an older AR (pilot project) deployed in the same area more than a decade before. Finally, based on the present results, proposals regarding the exploitation strategy and management for the local ARs are discussed.

MATERIAL AND METHODS

Study area

A small (0.6 km²) artificial reef, consisting of 7 reef groups (RGs) was deployed off Faro (Algarve, Southern coast of Portugal) in 1990 and enlarged in 2002. The new ARs run parallel to the coast line in a NW to SE direction along the 20m isobath and were set on either side of the old ARs. The new reefs are identical to the old reefs in terms of organization and module type (Figure 1). At present the Faro/Ancão AR system consists of 52 RGs of small concrete cubes of 1.4m side length, covering an area of 12.2 km². Each RG consists of 3 reef sets (RS) of 35 units each. Each RS consists of a randomly deployed two-layer assemblage of units. These reefs were deployed on a sandy bottom, where natural rocky bottom is scarce, at depths ranging between 20 and 22 m, approximately 2.5 km from the shore (Figure 1).

Experimental design

The study started in early Autumn 2002, a month after the enlargement of the Faro/Ancão AR. Three RGs were randomly selected out of 8 RGs that were closest to the older reefs (between 400m and 600m). The RGs were sampled monthly, between October (2002) and September (2004), the exception being the 17th month (February 2004) due to rough sea conditions. The nearest natural rocky area is about 2 km from the sampled ARs. Given the distance from natural reef areas, we consider these ARs to be statistically independent from any rocky reef source. Given the distance between RGs (~400m), these were also considered to be independent sampling locations. Three independent random counts were made in each reef set.

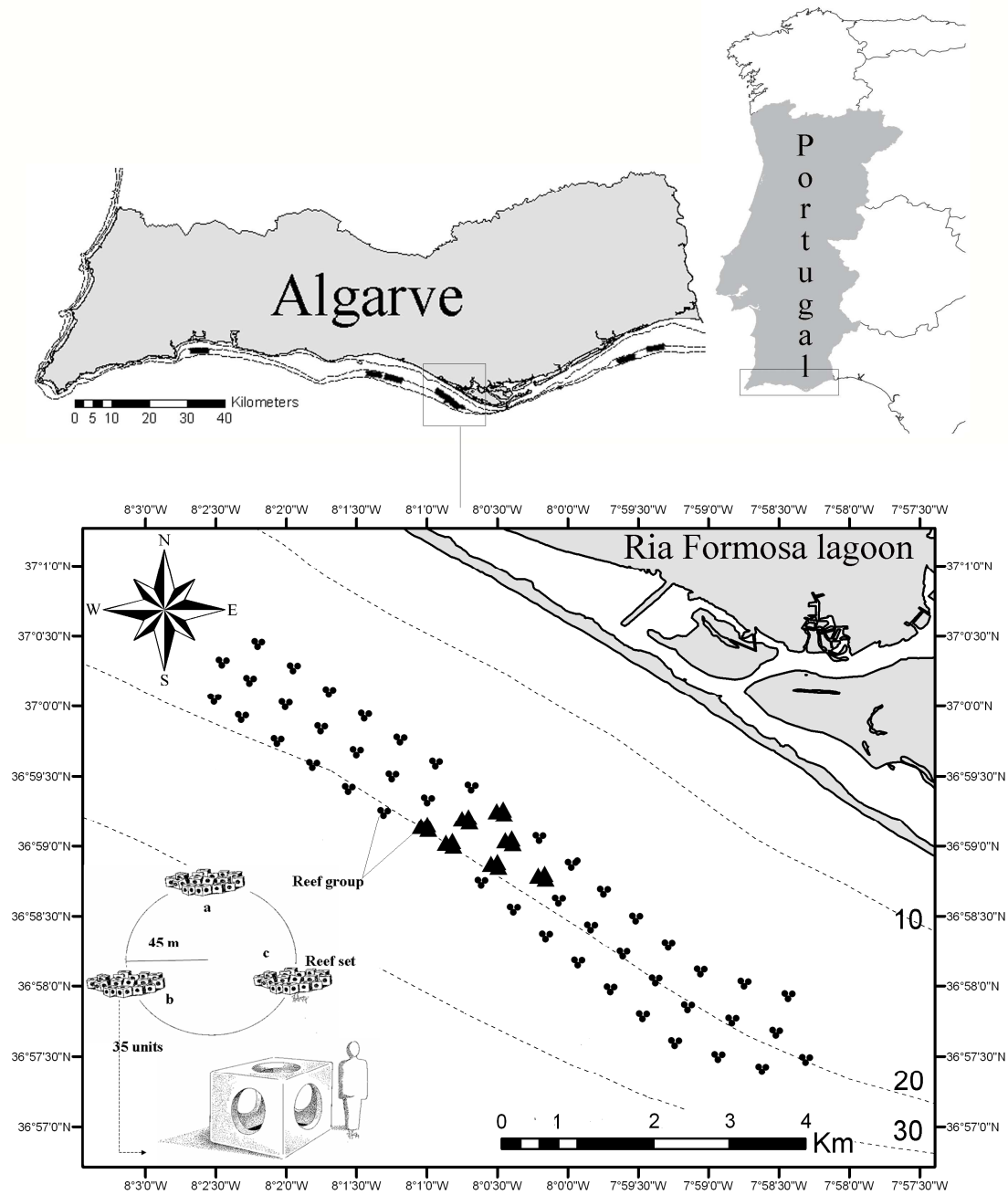


Figure 1. Map showing Algarve AR systems, reef organization and the location of the study area (Faro/Ancão AR system) in relation to the Ria Formosa lagoon and the 10, 20 and 30m isobaths. Triangles for old AR sets and circles for new AR sets.

Data collection

The fish assemblage was determined by visual census using the stationary point method developed by Bohnsack and Bannerot (1986) and further adapted by Santos *et al.* (2005) to the Algarve AR environment. This method is based on censuses taken at randomly selected points, where the single observer rotates around himself/herself within a cylinder of known volume (1.5 m^3) and counts the number and sizes of all fishes in a radius of 6m during six minutes (for details see Santos *et al.* 2005). All counts were made in the first week of each month and always in the same pre-selected RGs. In order to reduce bias due to daily migration (Santos *et al.* 2002) all visual census surveys were carried out between 10:00 and 12:00. Fish density was calculated as the number of fish per m^3 of water. The calculation of biomass densities (g/m^3) was made using: (1) the estimated fish length size, (2) the length-weight relationship, and (3) the estimated density of fish. Demographic structure was studied in terms of the juvenile *versus* adult specimens, based on the sizes at first maturity available in the literature.

Based on the work of Ody and Harmelin (1994) and Reñones *et al.* (1998), each fish species was assigned to one of four spatial categories: SC1, cryptic and sedentary species that occupy a stationary position inside the cement block or on the bottom around it; SC2, species with a medium home range, living in the proximity of the blocks; SC3, species with a relatively wide home range, moving throughout the reef area; and SC4, surface and mid-water fishes that show a swarming response to the reef. Fishes were also categorised as resident, transient or occasional accordingly to their resident status (RSt) (for details see Bayle-Sempere *et al.* 1994). Trophic level of the fish species (herbivores, omnivores and carnivores) was described from available data on feeding habits (Bell 1983; <http://www.fishbase.org>). Information about reef use (eg. refuge, shelter) of reef associated species was assessed visually.

Data analysis

For each month the mean number of species, Shannon–Wiener diversity ($H' = - \sum P_i \ln(P_i)$), density and biomass for overall reef fish assemblage, as well as for the different SCs and RSts categories were calculated, using RS mean values, obtained by averaging reef set data. Standard deviations (\pm SD), were calculated on a monthly basis using RSs data. The frequency of occurrence (% f.o) was calculated using monthly RG data. As monthly sampling was always conducted in the same RGs, replicates data may not be independent but RG (site) correlated in time (Underwood 1997). The studied “Time” factors were season and year. Monthly data were assigned to a season according to the follow criteria: Winter (January-March); Spring (April-June); Summer (July-September); Autumn (October-December). Seasonal (inter- and intra-annual seasons) and annual differences were assessed by means of two-way (site and “time”) repeated measures ANOVA (Zar 1996) using each RGs mean monthly value ($\alpha= 0.05$). Whenever normality (Kolmogorov-Smirnov test) and/or heterogeneity (Bartlett test) assumptions failed, data were $\ln(n+1)$ transformed to produce normality and homogeneity of variance. If the null hypothesis was rejected, the multiple comparison test of Tukey was performed (Zar 1996). The Statistica (v. 5.0) software package was used for all univariate analyses.

Monthly reef fish assemblage temporal evolution and assemblage stabilization was studied (RGs independently) by means of multivariate hierarchical cluster analyses. Cluster analysis applied the unweighted pair group average algorithm to the Bray-Curtis similarity matrix based on monthly reef group data, after standardization and square root transformation of the data. In order to cope with the repeated measures design of cross factors (RGs and time), seasonal and annual changes in reef fish assemblage structure were analysed through multivariate second-stage matrix ordination (Clarke *et*

al. 2006). Thus, a Bray-Curtis similarity matrix was built using each RSs monthly mean fish density data, after standardization and square root transformation of the data. Thereafter, differences between seasonal and annual reef fish assemblage structure were tested by the one-way ANOSIM permutation test. Non-Metric Multidimensional Scaling (MDS) was also used to investigate seasonal assemblage fish structure. The SIMPER routine was used to determine the species that most contributed to the assemblage structure. Moreover, the species that most contributed to explain changes between seasons were super-imposed in the MDS. The multivariate analysis was carried out using the Primer 5.1 software package (Clarke and Warwick 2001).

Comparison between the number of fish species, diversity, density and biomass previously reported by Santos *et al.* (2005) for a five year old AR deployed in 1990 (n=12 months), and those observed in the present study were done by means of the *t*-student independent (unpaired) sample test, using RGs monthly row data after verification of normality (Kolmogorov-Smirnov test) and homogeneity of variance (Levenes's test for equality of variance) assumptions. The Mann-Whitney U test (M-W) was used when the assumptions were not met (Zar 1996).

RESULTS

Assemblage composition

A total of 28 species belonging to 14 families were observed. The Sparidae family was the most represented, accounting for 38% of the species, followed by the Labridae family with 14% of the species. The most frequent taxa (> 90%) were the Blenniidae, *Diplodus vulgaris*, *D. bellottii*, *Coris julis*, *Scorpaena notata* and *Boops boops*. With the exception of *B. boops* and *S. notata* all the former species were seen feeding on the AR substrate. *Dicentrarchus labrax*, *Symphodus* sp. and *D. sargus* were also very frequent

(63% < f.o. <71%). It is worth noting that 19 species (66%) are commercially important, including some of the most frequently observed, such as *D. labrax*, *D. sargus* and *D. vulgaris*. The majority of the recorded fish species are carnivores (Table 1).

Resident species accounted for the highest number of fish *taxa*, with 13 species, followed by the transient group with 9 species and the occasional with 6 species (Table 1). Most resident species use the AR as a shelter. In contrast, transient and occasional species usually do not use these structures as a refuge (Table 1). High density schools of fishes that are attracted/aggregated by the AR, such as *B. boops*, *Pagellus* spp., *Trachurus trachurus* did not use these structures for shelter (Table 1). SC3 and SC4 were the most represented spatial categories with 10 (34%) and 8 (28%) species, respectively, with most of these species belong to the Sparidae family (Table 1).

Colonisation process

Number of species

By the 4th month 22 species (79%) had already recruited to the AR, and after 8 and 12 months, 23 (82%) and 25 (89%) of the species were recorded in the AR, respectively. In fact, during the 2nd year of the study period only three additional new species (*Scomber japonicus*, *Pagrus pagrus* and *Diplodus cervinus*) were recorded. A sharp increase in the mean number of species was observed during the first 4 months after AR deployment (Figure 2). Thereafter, the mean number of species remained almost constant, on the order of 13 (Table 2). The first species colonizing the ARs, in the 1st month after deployment, belong to the resident category, followed by those from the transient and occasional categories with 4, 3 and 1 species, respectively (Table 1). By the 4th month 10 resident species (76%), 7 transient (78%) and 6 (100%) occasional

species were already established on the AR. Nine months after the AR deployment all resident species had already been recorded.

The mean number of species was significantly higher for the resident fishes, followed by the transient and occasional (ANOVA: $F_{2, 66} = 110$; $P < 0.01$, Table 2). Significant differences were found between all RSt categories in terms of the number of species (ANOVA: $F_{2, 66} = 158$; $P < 0.01$). In the 1st year, the mean number of species in autumn, 9, was lower than in other seasons, 13 (ANOVA: $F_{3, 8} = 11.80$; $P < 0.01$, see Table 3). The mean number of species was high (ANOVA: $F_{3, 88} = 9.05$; $P = 0.15$, see Table 2) for SC3. During the first three months, the assemblage was dominated (52%) by surface and mid-water fish species (SC4).

Diversity

Monthly reef fish diversity ranged between 0.41 and 2.34 (Figure 2). During the first 6 months a sharp increase in diversity was recorded. After this period, diversity ranged between 1.32 and 2.34. In fact, after the first six months diversity decreased and remained below the mean until it increased again in late summer, in winter/early spring and spring/early summer (see Figure 2). In the 1st year differences in diversity (ANOVA: $F_{3, 8} = 6.06$ $P < 0.05$) were only observed between autumn, with the lowest diversity ($H' = 1.1$), and winter with the highest diversity ($H' = 2.15$) (Table 3).

Reef mean diversity was significantly higher for the resident species, followed by the transient and the occasional groups (ANOVA: $F_{2, 66} = 67.84$; $P < 0.01$) (Table 2). SC mean diversity was highest for SC3 and SC2 (Table 3). Differences in mean diversity were observed (ANOVA: $F_{3, 88} = 7.73$; $P < 0.01$) between SC1, with the lowest and SC3, with the highest diversity.

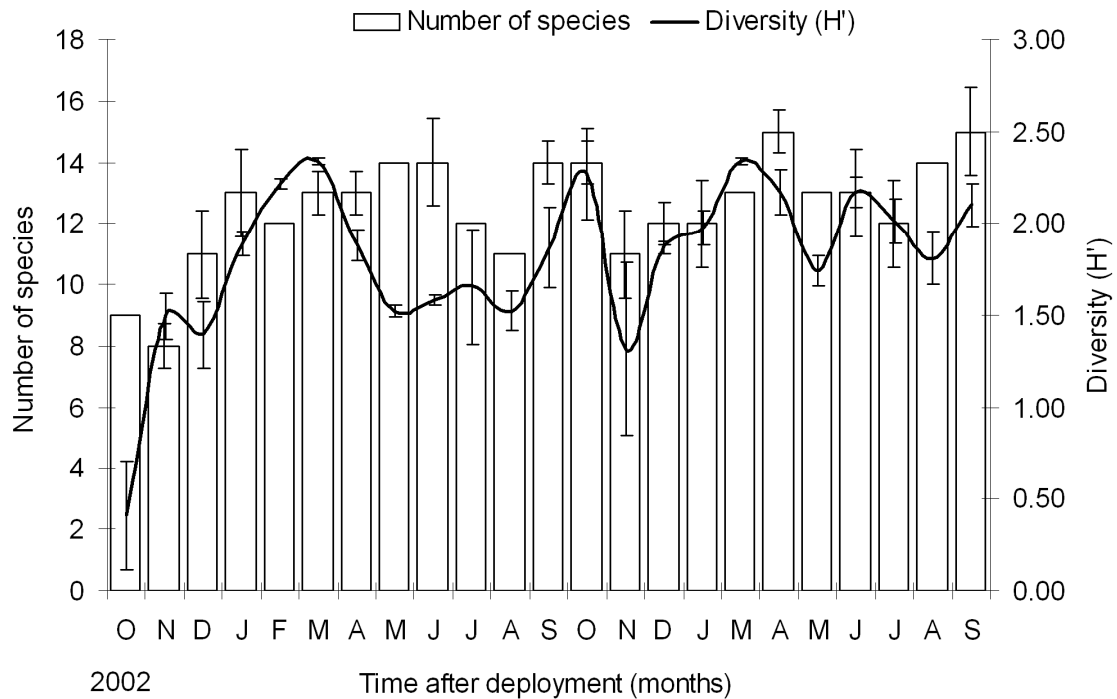


Figure 2. Mean (\pm standard deviation) monthly variation of the number of species and diversity.

Fish and biomass densities

The mean density of the overall reef fish assemblage ranged from 0.3 to 9.5 ind/m³ (Figure 3), with an average density of 2.8 ± 1.8 ind./m³. In decreasing order of numerical importance, the most important *taxa* were *B. boops* (29%), Blenniidae (19%), *D. vulgaris* (11%), *D. bellottii* (7%), *C. julis* (5%), *S. notata* (3%) and *D. sargus* (2%) (Table 1). Two overall fish density peaks were registered in May (2003) and April (2004), related to the recruitment to the AR of schools of YOY of demersal species such as *B. boops*, *T. trachurus*, and *Pagellus* spp. (Figure 3). Together, the latter species account for 1.2 ± 0.4 ind/m³, which corresponded to 42% of the total reef density. *T. trachurus* has a seasonal occurrence on the AR, being present only from March to June.

Table 1. List of the species recorded on the Faro/Ancão artificial reef: RSt - resident status: R – resident; T – transient; O – occasional. SC - spatial category (see text for definitions). F.O – frequency of occurrence (%). Month – month of first occurrence in AR. CV – commercial value: N – none; L – Low; H - high. Shelter – species that refuges in the AR. Feeding – species that feed on the AR substrata; Trophic structure – trophic behaviour of fish (see text for details); Juveniles (%) – percentage of juveniles; Length – mean total length (cm); Mean densities (n° ind/m³) and Mean biomass (g/m³) with standard deviation.

Family	Especies	RS	SC	F.O	Month	CV	Shelter	Feeding	Trophic structure	Juveniles (%)	Length (cm)	n.º ind./m ³	g./m ³
APOGONIDAE	<i>Apogon imberbis</i>	R	1	4.35	1	N	yes	Not observed	Carnivoros	-	10 ± 3.5	0.001 ± 0.004	0.019 ± 0.075
BALISTIDAE	<i>Balistes carolinensis</i>	O	3	2.17	9	L	No	No	Carnivoros	100	35 ± 0	0.001 ± 0.001	0.089 ± 0.43
BATRACHOIDIDAE	<i>Halobatrachus didactylus</i>	T	1	23.91	3	L	No	No	Omnivoros	-	27 ± 3.9	0.01 ± 0.018	3.656 ± 7.55
BLENNIIDAE	Blenidae	R	1	100.00	2	N	yes	yes	Carnivoros	-	5 ± 2	0.557 ± 1.092	0.486 ± 0.41
CALLYONINIDAE	<i>Callyonimus lyra</i>	O	1	13.04	2	N	No	Not observed	Carnivoros	-	7 ± 1.8	0.01 ± 0.028	0.052 ± 0.14
CARANGIDAE	<i>Trachurus trachurus</i>	O	4	39.13	6	L	No	yes	Carnivoros	100	12 ± 1.7	0.192 ± 0.473	2.318 ± 4.79
GADIDAE	<i>Trisopterus luscus</i>	R	3	23.91	9	H	yes	Not observed	Carnivoros	100	17 ± 4	0.017 ± 0.032	0.334 ± 0.6
LABRIDAE	<i>Coris julis</i>	R	2	95.65	2	N	yes	yes	Carnivoros	-	22 ± 2.7	0.143 ± 0.054	17.242 ± 9.7
	<i>Ctenolabrus rupestris</i>	R	2	26.09	7	N	yes	yes	Carnivoros	-	10 ± 0	0.018 ± 0.031	0.216 ± 0.38
	<i>Labrus merula</i>	R	2	2.17	8	N	yes	yes	Carnivoros	-	30 ±	0.001 ± 0.005	0.522 ± 2.5
	<i>Symphodus spp.</i>	T	2	63.04	3	N	yes	yes	Carnivoros	-	15 ± 4.2	0.047 ± 0.044	3.807 ± 4.91
MORONIDAE	<i>Dicentrarchus labrax</i>	T	4	71.74	5	H	No	yes	Carnivoros	75	29 ± 2.9	0.044 ± 0.033	118.284 ± 96.72
SCOMBRIDAE	<i>Scomber japonicus</i>	O	4	15.22	17	L	No	yes	Carnivoros	100	21 ± 2.5	0.058 ± 0.153	4.89 ± 15.05
SCORPAENIDAE	<i>Scorpaena notata</i>	R	1	95.65	1	L	No	Not observed	Carnivoros	-	14 ± 1.1	0.076 ± 0.032	0.404 ± 0.18
SERRANIDAE	<i>Serranus cabrilla</i>	R	2	45.65	5	N	yes	yes	Carnivoros	33	20 ± 6.7	0.024 ± 0.026	2.61 ± 3.51
	<i>Serranus hepatus</i>	T	2	28.26	1	N	yes	yes	Carnivoros	-	6 ± 2.2	0.013 ± 0.018	0.054 ± 0.1
SPARIDAE	<i>Boops boops</i>	O	4	95.65	2	L	No	Not observed	Omnivoros	100	7 ± 3.4	0.861 ± 0.705	2.31 ± 3.16
	<i>Diplodus annularis</i>	R	3	6.52	5	L	yes	yes	Carnivoros	17	16 ± 0.2	0.015 ± 0.042	1.129 ± 3.07
	<i>Diplodus bellottii</i>	R	3	84.78	1	H	yes	yes	Omnivoros	18	15 ± 1	0.196 ± 0.131	11.204 ± 8.46
	<i>Diplodus cervinus</i>	T	3	4.35	16	L	yes	yes	Carnivoros	-	28 ± 3.5	0.002 ± 0.007	0.866 ± 2.99
	<i>Diplodus puntazzo</i>	R	3	6.52	5	H	yes	yes	Carnivoros	-	25 ± 0	0.003 ± 0.009	0.707 ± 1.87
	<i>Diplodus sargus</i>	R	3	69.57	4	H	yes	yes	Omnivoros	8	24 ± 5	0.071 ± 0.073	18.519 ± 19.35
	<i>Diplodus vulgaris</i>	R	3	100.00	1	L	yes	yes	Omnivoros	92	11 ± 3.8	0.313 ± 0.135	11.248 ± 7.05
	<i>Pagellus acarne</i>	T	4	43.48	1	H	No	Not observed	Carnivoros	100	11 ± 4	0.113 ± 0.224	1.122 ± 1.76
	<i>Pagellus erythrinus</i>	T	4	17.39	1	H	No	Not observed	Carnivoros	48	14 ± 6.3	0.009 ± 0.021	0.772 ± 3.17
	<i>Pagrus pagrus</i>	O	3	4.35	18	H	yes	Not observed	Carnivoros	0	25 ±	0.005 ± 0.026	1.345 ± 6.45
	<i>Spondylisoma cantharus</i>	T	3	13.04	2	L	yes	yes	Omnivoros	96	12 ± 2.7	0.004 ± 0.01	0.097 ± 0.29
TRACHINIDAE	<i>Trachinus draco</i>	T	3	19.57	1	L	yes	Not observed	Carnivoros	-	27 ± 4.8	0.018 ± 0.021	2.532 ± 3.57

Table 2. Mean (\pm standard deviation) overall density (n. ind/m³), biomass (g/m³) and contribution of the number of species (maximum, mean) and diversity (H) according to the resident status (R – Resident; T – Transient; O - Occasional) and Spatial Categories (SC).

	ind/m ³	g/m ³	No. of species		H		
			Maximum	Mean	Maximum	Mean	
Resident status							
Reef	2.8 \pm 1.8	207 \pm 127	15	13 \pm 1.7	2.38	1.82 \pm 0.4	
R	1.4 \pm 1.2	66.3 \pm 34	9	7 \pm 0.7	1.92	1.55 \pm 0.3	
T	0.3 \pm 0.2	130.2 \pm 99	6	4 \pm 1.4	1.77	1.12 \pm 0.4	
O	1.1 \pm 1.0	11.4 \pm 21	3	2 \pm 1.1	0.91	0.37 \pm 0.3	
Spatial category							
SC1	0.65 \pm 1.1	4.6 \pm 8	5	3 \pm 0.8	1.26	0.67 \pm 0.3	
SC2	0.25 \pm 0.1	24.5 \pm 12	4	3 \pm 0.8	1.27	0.84 \pm 0.3	
SC3	0.63 \pm 0.3	47.7 \pm 30	6	4 \pm 1.0	1.42	0.98 \pm 0.3	
SC4	1.23 \pm 1.1	130.4 \pm 95	5	3 \pm 1.0	1.29	0.67 \pm 0.4	

Several peaks in fish biomass were recorded during the course of the study. Transient species, namely *D. labrax* that accounted for 118 \pm 96 g/m³, which corresponded to 90% of transient and 57% of the overall biomass, was the RSt category that contributed most to the biomass of the reef fish assemblage (Figure 3; Table 1). High biomass values were also recorded for the resident species *D. sargus* (9%), *C. julis* (8%), *D. vulgaris* (5%) and *D. bellotti* (5%) (Table 1). The overall mean fish assemblage biomass ranged between 14 and 506 g/m³, with a mean value of 207 \pm 127 g/m³. Economically important species account for a mean of 180 \pm 8.1 g/m³ (87%) of total reef fish biomass. The densities of both resident and occasional species were higher than transient species (ANOVA: $F_{2, 66} = 10.63$; $P < 0.01$, see Table 2). The contribution of transient species (63%) in terms of biomass was significantly higher than that for both resident (32%) and occasional (5%) species, due to *D. labrax* (ANOVA: $F_{2, 66} = 21.97$; $P < 0.01$, see Table 2).

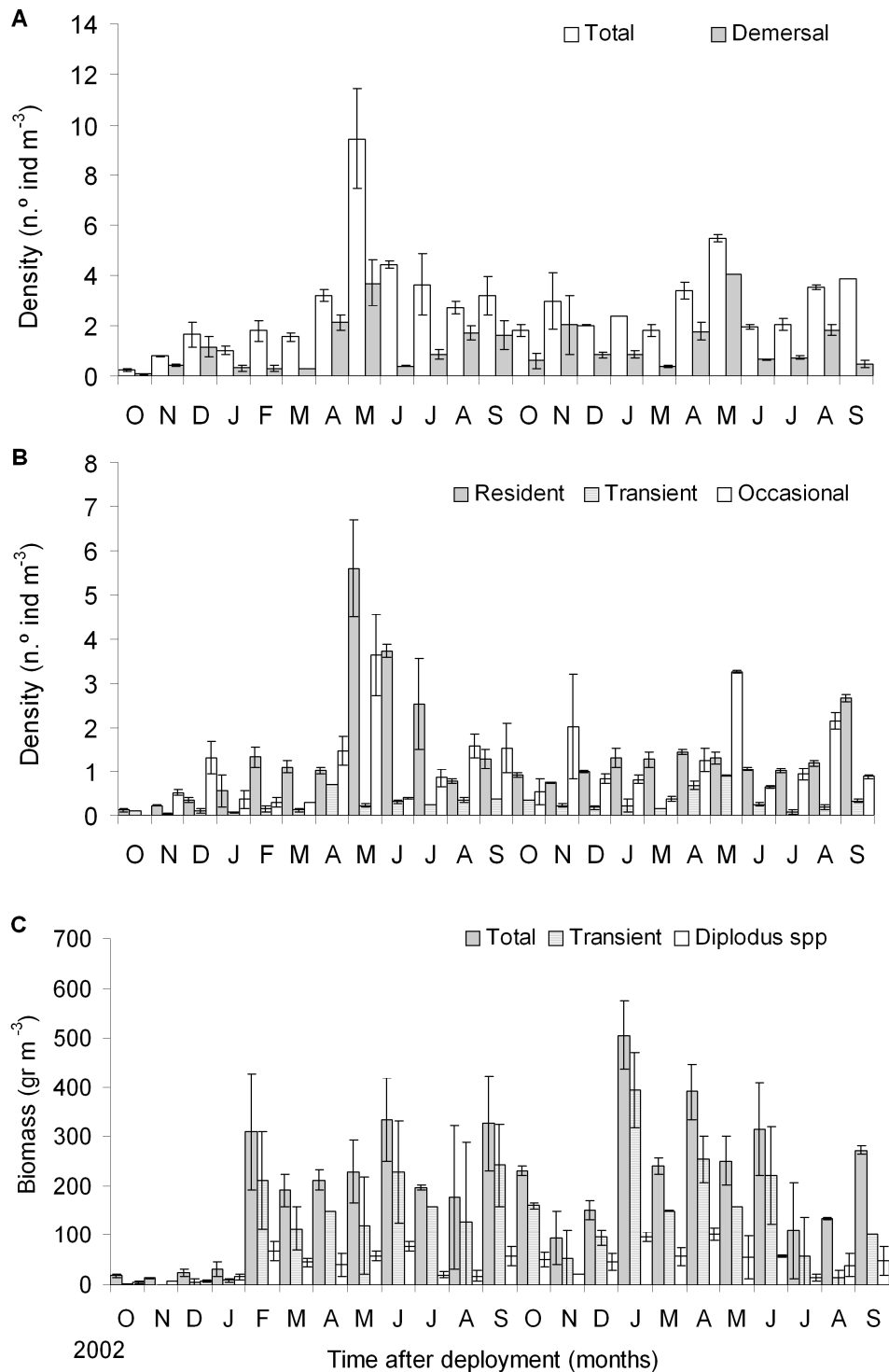


Figure 3. A: Total (\pm standard deviation) and mean demersal occasional (include *B. boops*, *T. trachurus* and *Pagellus* spp.) densities; B: Resident status category assemblage densities; C: Total, transient (90% *D. labrax*) and *Diplodus* spp. (*D. bellottii*, *D. sargus* and *D. vulgaris*) assemblage biomass.

The highest SC densities corresponded to SC4, due to both *B. boops* and *D. labrax*, followed by SC3, SC1 and SC2, respectively (Table 2). Significant differences (ANOVA: $F_{3, 88} = 6.96$; $P < 0.01$) in densities were found between SC4 and all the remaining SC (Tukey: $P < 0.01$). In relation to reef fish SC assemblage biomass, the highest figures corresponded to SC4 due to *D. labrax*, followed by SC3, SC2 and SC1, respectively (Table 2). Significant differences in terms of mean biomass were found between SC4 and the remaining SC categories (ANOVA: $F_{3, 88} = 27.05$; $P < 0.01$).

During the first year differences in terms of mean density were found between autumn and spring (ANOVA: $F_{3, 8} = 4.73$; $P < 0.05$). However, no other differences in mean density and in biomass were found between years, or between seasons within and between years. A significant RG effect was verified for density in 1st year seasons (Table 3).

Demographic structure

It was not possible to classify all specimens as juvenile or adult due to the lack of information regarding species size at first maturity, namely for non commercial species (Table 1). The monthly density and biomass of juveniles of commercial species were 1.58 ± 0.25 ind/m³ (56%) and 114 ± 25 g/m³ (55.1%), respectively.

Schools of young-of-the-year (YOY) of *B. boops* and *Pagellus* spp. (0⁺ and 1⁺) were regularly observed. The schools of *T. trachurus* and *Scomber japonicus* were composed of age 1⁺ specimens.

Benthic species such as *S. notata*, *Trachinus draco* and *Halobatrachus didactylus* were represented both by adults and juveniles. Rocky bottom associated reef fish species were mainly represented by adult specimens. These included *S. cabrilla*, *D. bellotti*, *D. annularis*, *D. sargus* and *P. pagrus* (Table 1).

Table 3. Results of the repeated measure ANOVA for N – number of species; H – Diversity; Density (ind/m³); Biomass (g/m³), and ANOSIM for total reef fish assemblage structure (C). A - Autumn; W – Winter; Sp - Spring; S – Summer. ns - not significant. * - significant differences were highlighted by the repeated measure ANOVA between reef groups. No significant differences were found for any variable with the repeated measure ANOVA for interaction between RGs x Season and/or year effects.

Factor	Fish assemblage				
	N	H	C	ind/m ³	g/m ³
Year	ns	ns	1 st vs. 2 nd	ns	ns
Season 1 st year	A vs. W/Sp/S	A vs. W	A vs. W/Sp/S W vs. Sp/S	A vs. Sp*	ns
Season 2 nd year	ns	ns	All seasons	ns	ns
Intra-seasonal	ns	ns	A vs. S	ns	ns

Assemblage structure

The structure of the fish assemblage in the first month after deployment (Cluster 1) differs from the remaining months, due to the low number of species and respective densities. Differences in similarity between cluster 2, which groups the 2nd to the 4th months after deployment, and the other clusters are due to the increasing numbers of *B. boops* and *D. vulgaris* after the 4th month. After this initial four months period, monthly samples are mixed, indicating that the assemblage has become more similar. For instance, clusters 3, 4, 5 and 6 have similarity of more than 73% (Figure 4).

During the study period intra-seasonal differences were found (ANOSIM_{1st year}: R=0.38, P<0.01; ANOSIM_{2nd year}: R=0.60, P<0.01) (Table 3). In the first year the

difference were particularly marked between autumn, with low number of species and densities, and the other seasons. Overall, seasonal differences were mainly associated (SIMPER) with the high recruitment episodes of demersal species in spring/summer (*B. boops*, *T. trachurus* and *P. acarne*) and Blenniidae in spring 2003 (Figure 4 and 5). Inter-annual seasonal differences were found between autumn and summer (Table 3). For the latter period these were mainly due to the high densities of *S. japonicus* in 2004, and low densities of *B. boops* and Blenniidae in 2003 (Figure 4). The contribution of the latter species to the dissimilarities was 17%, 10% and 10%, respectively. Differences between 1st and 2nd year fish assemblages (ANOSIM: $R=0.13$, $P<0.01$) were mainly due an increase in occasional category demersal species and resident species, accounting for 29% and 44% of the dissimilarity, respectively (Table 4).

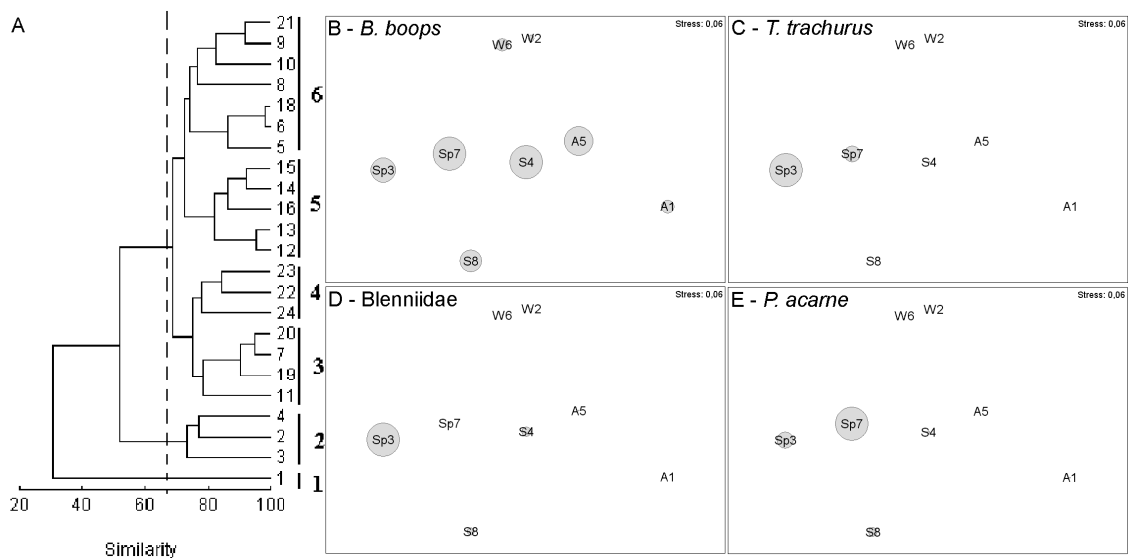


Figure 4. Cluster analysis (average) of overall reef fish monthly assemblage (A) and seasonal nMDS plots for mean *B. boops* (B), *T. trachurus* (C), Blenniidae (D), and *P. acarne* (E) densities. A1 and A5- Autumn 2002 and 2003; W2 and W6 – Winter 2003 and 2004; Sp3 and Sp7 – Spring 2003 and 2004; S4 and S8 – Summer 2003 and 2004.

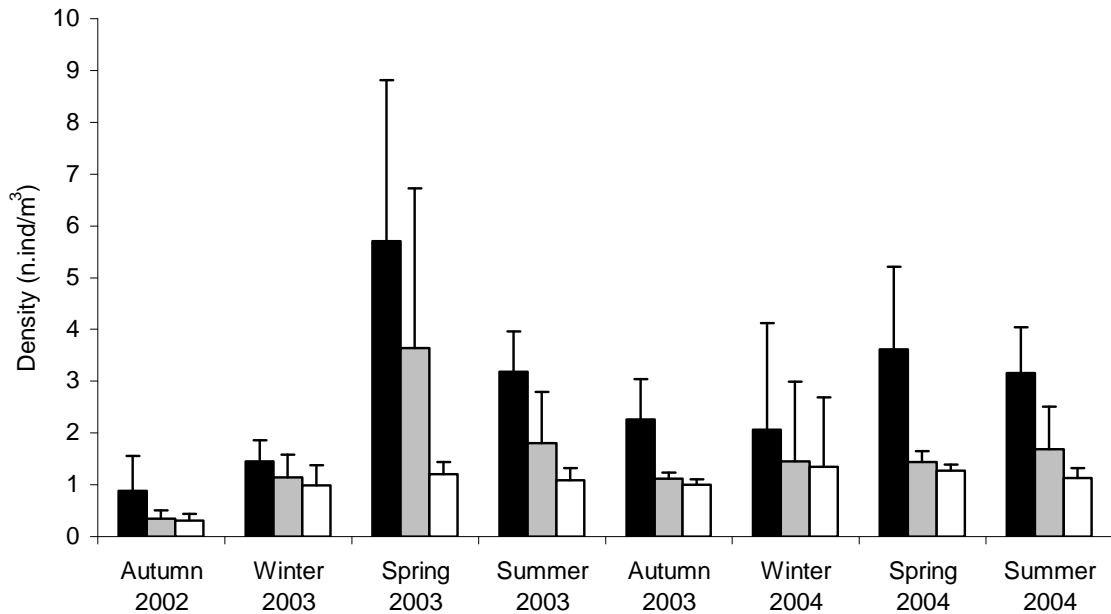


Figure 5. Mean (\pm standard deviation) seasonal overall assemblage density (black bars), mean assemblage density without occasional demersal species that include *B. boops*, *T. trachurus*, and *Pagellus* spp. (grey bars) and without occasional demersal species and Blenniidae (white bars).

Comparison with the older AR

Comparisons with an older mature AR were performed excluding the data of the first four months, as we considered that the new AR assemblage structure stabilised after this period. The monthly mean of species in old ARs (6.95 ± 2.41) was less than that of the new ARs (M-W_{Number}: $U=0.1$; $Z=4.62$, $P<0.01$). This is also true in the case of the mean fish diversity (1.49 ± 0.64 ANOVA: $F_{1,33}=4.57$; $P<0.05$), density (1.49 ± 0.73 ind/m³, M-W_{Number}: $U=25$; $Z=3.61$, $P<0.01$) and biomass (81.80 ± 42.85 g/m³, M-W_{Biomass}: $U=8$; $Z=4.30$, $P<0.01$).

Table 4. Species that contributed most to the differences (Density, n. ind/m³) found between the 1st and 2nd year. Diss. – Average dissimilarity; Contrib - contribution to dissimilarities found between years measured in terms of percentage.

Species	1 st year	2 nd year	Diss.	Contrib.
<i>Boops boops</i>	0.75	0.94	4.4	10.85
Blenniidae	0.84	0.24	3.41	8.4
<i>Diplodus bellottii</i>	0.15	0.26	3.35	8.25
<i>Trachurus trachurus</i>	0.25	0.13	2.94	7.25
<i>Pagellus acarne</i>	0.07	0.15	2.81	6.92
<i>Diplodus sargus</i>	0.06	0.08	1.77	4.37
<i>Scomber japonicus</i>	0.00	0.11	1.7	4.2
<i>Diplodus vulgaris</i>	0.28	0.35	1.69	4.16
<i>Symphodus</i> spp.	0.05	0.04	1.65	4.07
<i>Dicentrarchus labrax</i>	0.04	0.05	1.65	4.05
<i>Serranus cabrilla</i>	0.01	0.04	1.59	3.92
<i>Coris julis</i>	0.13	0.16	1.53	3.76
<i>Scorpaena notata</i>	0.06	0.09	1.38	3.4
<i>Serranus hepatus</i>	0.01	0.01	1.34	3.3
<i>Trachinus draco</i>	0.02	0.02	1.29	3.18
<i>Callyonimus lyra</i>	0.02	0	1.19	2.92
<i>Ctenolabrus rupestris</i>	0.01	0.03	1.07	2.64

DISCUSSION

The pattern of fish colonisation in the Faro/Ancão artificial reef displayed an initial rapid increase in terms of fish species and diversity, with stabilization of the fish assemblage structure occurring 4 months after AR deployment. A marked increase in the AR macrobenthic community cover was observed by the 3rd month (~50%), and by the 6th month the reef surface was totally colonized by Cirripedia, Serpulidae, Bryozoa and Ascidiacea (Boaventura *et al.* 2006). Thus, stabilization of the AR fish assemblage matches the rapid increase in macrobenthic colonisation. Invertebrate settlement occurred fast, which may have contributed to the rapid increase of reef associated species, as most of the fish species are carnivorous. On the other hand, resident species are dependent on invertebrates for shelter purposes (Golani and Diamant 1999),

especially small sized species and juveniles (D'Anna *et al.* 1994; García-Rubies and Macpherson 1995) or for food (Leitão *et al.* 2007). This fast rate of fish colonisation associated to benthic cover development in the months shortly after AR deployment was also observed in other studies in temperate waters (Solonsky 1985; Bohnsack *et al.* 1991; Relini *et al.* 1994).

Within less than one year after AR deployment the number of species, diversity and the assemblage structure had stabilised. Santos *et al.* (2005) postulated that the colonisation of a similar reef in the area would stabilize after five years. However, the latter study was carried out in an AR on sandy grounds, with little natural and non artificial hard substrata nearby and four years after reef deployment.

Extensive movements of rock fishes from the shore across sandy areas to newly settled ARs have been reported by different authors (Mathews 1985; Solonsky 1985). Although the movements of fish from natural reefs to ARs are well studied, movements between ARs are not. In the present study, the initial rapid fish colonisation may have originated from the adjacent older AR. In fact, most of the species recorded in this study were readily found on the nearby older AR (Santos *et al.* 1996). A notable example is the rapid appearance of adults of rocky bottom associated species such as *D. sargus*, *D. bellottii*, *C. julis* and *S. notata* that do not have difficulty in crossing open spaces (Coll *et al.* 1998). The colonisation pattern of these species and other rocky bottom species (e.g. *Labrus merula*, *Symphodus* spp.) characterised by wide horizontal movements, is reinforced by the fact that most of them recruit in very shallow areas and move towards deeper waters in response to ontogenic changes in habitat choice (García-Rubies and Macpherson 1995; Harmelin-Vivien *et al.* 1995). The fast rate of colonisation could also be associated with seasonality, an important factor affecting the quantitative species composition of temperate AR assemblages (Coll *et al.* 1998), and with the migration of

juveniles of several species from the nearby Ria Formosa nursery habitat. According to Monteiro *et al.* (1990) migration of sub-adults of a variety of species from the Ria Formosa loagoon (e.g. *D. vulgaris* and *D. bellottii*) occurs in late summer/autumn. Such a migration pattern coincides with the period (first trimester) after which increases in number of species, diversity, density and biomass were most noted.

During the first 8 months after reef deployment the number of species and the diversity were higher than those reported from the Western Mediterranean (Bayle-Sempere *et al.* 1994), Monterey Bay, California (Solonsky 1985) and Puget Sound, Washington (Laufle and Pauley 1985), but lower than those observed from the Ligurian Sea (Relini *et al.* 1994) and the Canary Islands (Herrera *et al.* 2002). Differences between the reef fish communities and colonisation patterns are certainly linked to AR characteristics and establishment areas (e.g. in trawl affected areas, see Bayle-Sempere *et al.* 1994). For instance, depth (Bell 1983), reef design and complexity (Solonsky 1985) and reef substrate (Chandler *et al.* 1985; Laufle and Pauley 1985) are factors that influence the establishment of reef fish assemblages. On the other hand, it is also generally assumed that greater complexity is associated with more complex assemblage structure, species richness, diversity and abundance. Charbonnel *et al.* (2000) found that the fish assemblage evolution of large, low complexity ARs decreases rapidly due to the vast undivided empty spaces that have no equivalence in relation to the natural environment. However, some species such as the sea breams and wrasses are particularly successful on all types of AR (Solonsky 1985; Bayle-Sempere *et al.* 1994; Relini *et al.* 1994; Herrera *et al.* 2002; Santos *et al.* 2005).

For other species, especially resident species, it is known that AR complexity/heterogeneity is a key factor (Harmelin 1987). The Algarve AR modules only have a simple large hole in each side which creates a large inner open space. This

might be limiting colonisation of resident piscivorous species such as conger and moray eels. According to Hixon and Beets (1993) for such species that prefer hole sizes near their body size, the lack of shelter of the appropriate size is a primary limiting factor. This may also be the case for groupers (*Serranus* spp.), which in the current study showed low frequency of occurrence and low densities. In addition, the small number of recruits in large volume ARs in temperate waters may be associated to enhanced predation by larger fishes in such artificial habitats (Bayle-Sempere *et al.* 2001).

Our results showed that reef fish assemblage structure varied seasonally. These results are in agreement with those reported by several authors for other ARs in temperate Mediterranean waters (D'Anna *et al.* 1994; Relini *et al.* 1994). Seasonal variations were mainly associated with the arrival of YOY schools (0^+) of *T. trachurus*, *B. boops* and *Pagellus* spp. during spring and summer. In fact, seasonal variation in Mediterranean (D'Anna *et al.* 1994; Charbonnel *et al.* 2000) and Canary Islands (Herrera *et al.* 2002) reef assemblages are generally associated to recruitment of high abundance schools of species such as *Sardinella aurita*, *Spicara* spp. *B. boops*, *Chromis chromis* and *Atherina presbyter*. It is worthy of note that juvenile demersal specimens have a swarm response to AR and probably use these structures for protection from currents and/or feeding. The shape and size of reefs influences the currents around and through the reef, with a shielded locale of little or no currents behind the reef attractive to fish as provides an area where they do not need to fight against constant current (Nakamura 1985). Despite the seasonal trends observed, overall densities and biomass did not vary seasonally. The lack of a seasonal trend in density is explained by the high standard deviation of the mean of the above mentioned occasional demersal species. In the case of biomass, the absence of a seasonal trend was due to the fact that the overall biomass was highly conditioned by a single species, *D. labrax*.

Differences were found between fish density and biomass, diversity and the mean number of species observed in the present study and that previously reported for a five-year old AR in the same area (Santos *et al.* 2005), based on data collected and analyzed with the same methodology. In the case of biomass, this was mainly due to *D. labrax*, a species that was rarely observed in the older ARs until 2002. One of the problems that may arise regarding the comparison between old and new ARs is the assumption of independency of data among years. In the present study no significant annual changes were recorded for ecological indexes, either in abundance or in biomass densities between years. Nevertheless, changes in reef fish assemblage structure were observed between seasons within the same year in both the new and in the older ARs (Santos *et al.* 2005). Moreover, the latter authors also reported that two similar ARs deployed at different sites (Faro and Olhão) had different fish assemblages but both exhibited similar seasonal variations within the year. In fact, ecological indices, and fish and biomass densities in the new Faro/Ancão ARs followed seasonal variation in the water temperature, as previously reported for the Algarve (Santos *et al.* 2005), the Mediterranean (D'Anna *et al.* 1995) and the Adriatic Sea (Fabi and Fiorentini 1994). Thus, it seems reasonable to assume that in the Algarve seasonal ARs changes are more important than annual changes. In addition, to the author's best knowledge, no changes in terms of biotic and abiotic conditions have been reported for the studied area. Therefore, we can conclude that differences between old and new ARs are not associated with inter-annual variability but are more likely due to other causes.

Increased habitat usually produces a local increase in fish abundance as carrying capacity increases with reef size and decreases recruitment limitations as large reef area attract more recruits (Grossman *et al.* 1997). Such an impact, together with the decrease in isolation of the old AR, has enhanced the opportunities for species that were

uncommon before, thus increasing mean diversity and species richness. Sayer *et al.* (2005) showed that provision of large-scale multi-reefs alters the numbers and types of fishes present in areas where they were previously largely absent. Density was also higher in newly deployed ARs compared to old ARs. This may be partly due to the fact that older reef are limited in area and separated from other reefs (Bohnsack 1989).

However, differences in density between ARs may also be related with fishing activity, which may significantly affect adult population size. The older AR comprises a relative small patchy area on a predominately sandy bottom and has been heavily fished since deployment. While the new reefs are also in an area where normal fishing activity takes place, they have greatly increased the hard substrate area available to the fishery, thereby reducing fishing pressure.

An important question concerns when to start exploiting in ARs. Fishing should not start immediately after deployment in order to guarantee reef assemblage stability. Based on the results of the present study, we suggest that fishing can start one year after deployment. However, increase in fishing effort should be progressive in order to avoid disruption in the natural evolution of reef fish assemblages. Fishing strategies should also take into consideration seasonal variation of temperate AR fish assemblages. For instance, YOY occasional demersal species with a swarming response must be protected during recruitment events by prohibition of the use of low selectivity gears such as purse seine nets or small mesh sized gill nets. Fishing effort and the impact of particular gears should also be carefully considered, especially for resident species that have high site fidelity may be more vulnerable to artisanal and recreational fisheries.

The real efficiency of ARs has been questioned as these may act merely as aggregation tools (Polovina 1991) leading to the attraction versus production debate (Brickhill *et al.* 2005). Attraction and production are not mutually exclusive and can be

considered extremes along a gradient (Osenberg *et al.* 2002). While ARs may merely attract and concentrate some fishes, they may promote the production of others. For most fishes, the situation probably lies somewhere between the two extremes (Bohnsack 1989). In this study several adult species (sparids, *D. labrax*, *C. julis*, *S. cabrilla*; *S. notata*) were found shortly after ARs deployment, clearly indicating attraction. For some of these species, greater susceptibility to fishing due to aggregation may contribute to decreasing overall total biomass.

However, biomass production through recruitment and fish growth (King 1995) can not be excluded. Brickhill *et al.* (2005) report that production will be more likely with the addition of more reefs, or more complex reefs. Results also showed that ARs act like nursery areas for YOY of economically important species. In addition, trophic attraction may favor those species that depend on ARs for feeding, consequently contributing to biomass production in the long run (Leitão *et al.* 2007, 2008). In the Algarve, ARs are considered to fulfill their goal in enhancing the local fisheries (Santos and Monteiro 2007). Nevertheless, we cannot conclude that fishing enhancement is mostly due to attraction or to production. In fact, resolving this issue would need a different and broader approach, as it should be evaluated within the ecological context of each fish species (species-specific), taking into account their relationship with the ARs structures.

Several studies have questioned the potential of ARs to fulfill their objectives and whether some of these could have been achieved more efficiently with a management tool other than ARs (Polovina 1991; Baine 2001; Brickhill *et al.* 2005). An assessment of reef performance indicated that only 50% of the cases studied met their objectives (Baine 2001). According Polovina (1989) ARs are popular as management options because they do not require reductions in fishing effort and they aggregate fish,

resulting in higher catches in the initial stages. Thus ARs may actually be detrimental to the fishery and the stock simply because they allow managers to delay making hard but necessary decisions, such as imposing size limits or reducing effort.

With the collapse of fisheries on a global scale (Watson and Pauly 2001), and with the high risk of extinction of several marine fish species due to human activities, it has become clear that traditional technical management tools (e.g. minimum sizes, closed seasons, catches limits, closed areas, effort or gears restrictions, etc) have often failed their objectives. Although this is one of the reasons why ARs and Marine Protected Areas (MPAs) have become popular, they should be considered as complimentary and not alternatives to the traditional fisheries management tools. Regardless of the objective of the implementation of an MPA, its success may hinge on proper location relative to critical habitats that support living marine resources and this may limit the potential of reef fish populations to increase in abundance (Sladek and Friedlander *et al.* 2004; Monaco *et al.* 2007). The lack of a suitable area with a full complement of habitats and structural complexity in traditional Algarve fishing grounds was the justification of the choice for ARs instead of MPAs. These have provided at least some of the necessary habitat requirement for future implementation of MPAs in the region.

Another possible solution to conserve and replenish marine resources is the system of Marine Life Conservation Districts (MLCDs) developed in Hawaii (Friedlander *et al.* 2007). These MLCDs vary in size, habitat quality, and management regimes, providing an excellent opportunity to test hypotheses concerning MPAs design and function using multiple discreet sampling units. This may also apply in the case of Algarve AR systems, providing a more specific and small scale evaluation and management of the different ARs areas.

Although Algarve ARs have proved to be a potentially useful fisheries management tool, and the need for fishing exploitation strategies is recognised (Santos and Monteiro 2007), there are to date no guidelines in effect for sustainable AR exploitation. However, it is also important to note that until 2002, the AR area was probably too small to justify patch specific AR management. Recognizing the potential of AR species, the knowledge of their demographic structure and seasonal events is of considerable importance for defining suitable exploitation strategies and management of the AR areas that are exploited by the local commercial and recreational fisheries.

The current study showed that the deployment of large-scale multi-reefs near to a small pilot AR contributed to a rapid colonisation and a change in the ecological indices (higher species richness and diversity) and an increase in terms of fish and biomass densities, compared to what was previously reported for the ARs fish assemblages. Such effects can be highly beneficial for local fisheries enhancement. This study also provides some guidelines for sustainable exploitation strategies. Nevertheless, as reported by Baine (2001), the success of ARs will depend on the quality of the prior planning and ongoing management. Thus, it is important to guarantee long-term monitoring of the AR fish assemblages since, as a result of the natural evolution of the fish assemblages and/or the effect of fishing, these can change over time. In fact, this will be essential for preparation of the best management measures aiming towards the sustainable exploitation of the AR resources.

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CHAPTER 3

Contribution of artificial reefs to the diet of the white sea bream (*Diplodus sargus*)

Leitão F, Santos MN, Monteiro CC (2007) Contribution of artificial reefs to the diet of the white sea-bream (*Diplodus sargus*). ICES Journal of Marine Science 64: 473-478.

ABSTRACT

An evaluation of the trophic relationship between *Diplodus sargus* and artificial reefs (ARs) in the Algarve (southern Portugal) is based on a comparison of stomach contents and the macrobenthic communities present at the AR and in surrounding sandy bottom areas. Only adult white sea bream were observed in the vicinity of the ARs. The percentage of items found in the stomach that were characteristic of AR hard substratum was high (67%). Although the diet contained a wide variety of items, namely reef algae, invertebrates (crustaceans, gastropods, and bivalves), and fish, *Balanus amphitrite* and *Gibbula* spp. contributed most to the diet. The diet of *D. sargus* was strongly associated with prey availability on the AR, so highlighting the importance of these artificial habitats to the species. It seems that these artificial feeding areas, owing to their extent and benthic production, are enhancing the local *D. sargus* stock and hence the fishery.

INTRODUCTION

Artificial reefs (ARs) can be part of a solution to some of the problems concerning coastal resources, ecosystems, and fisheries, and in many countries, these man-made structures are an important tool of management (Bohnsack and Sutherland 1985; Polovina 1994; Jensen 2002). The Algarve (southern Portugal) ARs were conceived with several objectives in mind, including the enhancement of fish populations and the improvement of nearshore fisheries (Monteiro and Santos 2000). Algarve ARs now cover 43.5 km² and are the largest artificial habitat (productive type) in European waters.

AR structures provide a hard substratum (HS) for the settlement of benthic prey, contributing to the creation of new feeding areas, and consequently increase trophic efficiency (Bombace 1989) on formerly less productive, sandy seabeds (Leewis *et al.* 1997). In terms of AR deployment, their productivity relies on the assumption that AR surfaces provide additional critical habitat which increases the environmental carrying capacity, and hence enhances the abundance and biomass of marine biota (Polovina 1994). Nevertheless, some doubts persist as to whether ARs contribute to the production of new fish biomass or attract fish from surrounding areas without actually increasing total biomass (Bohnsack and Sutherland 1985). Stomach content studies have been carried out aiming to clarify the importance of AR production as potential feeding areas for fish, especially AR-resident fish (Lindquist *et al.*, 1994). Some studies highlight the importance of AR habitats for fish foraging (Pike and Lindquist 1994; Relini *et al.* 2002), and others report that fish feed primarily on adjacent sandy seabeds (Lindquist *et al.* 1994; Pepe *et al.* 1996).

For some species, ARs can serve as spawning areas or as refuge rather than as feeding areas. For those species that feed at ARs, man-made structures that produce

significant benthic biomass may be useful to support fish biomass recovery. Therefore, knowledge of the trophic ecology of reef ichthyofauna is important in understanding the dependence of species on an AR's benthic production and in evaluating the importance of such structures for the maintenance of fish populations. This means that the use of ARs to enhance any fishery requires the understanding of the ecological role of reefs in supporting exploited fish assemblages. Studies on the feeding ecology and trophic interactions of ARs and fish are scarce (Pepe *et al.* 1998; Relini *et al.* 2002; Fabi *et al.* 2006), and none have been made in southern Portugal. This scarcity of information is notable, because addressing it may shed light on whether the biological production of Algarve ARs contributes to an increase in fish biomass and consequently to local fisheries enhancement.

Diplodus sargus (Linnaeus 1758), a species with a coastal rocky reef distribution ranging from the Mediterranean to the eastern Atlantic (from the Bay of Biscay to South Africa), is important commercially. It is also one of the most abundant and frequent species found at ARs (Santos *et al.* 2005). Although several aspects of its feeding ecology have been studied (Baldó and Drake 2002; Mariani *et al.* 2002; Figueiredo *et al.* 2005), little attention has been paid to its trophic relationship with ARs (Pepe *et al.* 1998). Here, our aim is to evaluate the contribution of Algarve ARs to the diet of *D. sargus*.

MATERIAL AND METHODS

Study site and sampling

The experiment was carried out in the Faro/Ancão AR system, located off Faro (southern Portugal) on a sandy seabed, at depths ranging from 16 to 37 m. The system was deployed in 1990 and enlarged in 2002. It occupies an area of 12.2 km² and has a

total of 174 reef sets, of which 156 are small modules. Each of the latter consists of 35 cubic concrete units (2.7 m^3 each). Data collection was carried out at four of the oldest reef groups, FP1, FP3, FP5, and FP7, which were deployed in 1990 at depths of 19-21 m. The distance from the AR to the nearest rocky site, a small natural reef, is about 2.3 km. All fish were caught at the AR by spearfishing between August 2002 to August 2004, on a monthly basis and with an effort of two to three fishing days per month. Spearfishing was carried out during the morning (08:00-11:00) by two divers. In all, 64 dives and 48 visual census surveys were carried out. Fish behaviour and size were recorded according to the stationary point count technique, as described by Santos *et al.* (2005).

Data analyses

Data on the Faro/Ancão AR benthic and ichthyofauna communities have been reported previously (Santos *et al.* 2005; Boaventura *et al.* 2006; Moura *et al.* 2006). However, in order to gather more data regarding AR communities, *in situ* observations were carried out focusing on large mobile macrobenthic invertebrates and seaweeds. To characterize the soft-bottom macrobenthic community, sandy seabed samples were collected every three months during the study period. The samples were collected both inside the reef modules and outside the reefs along a transect at increasing distances (0, 1, 5 and 20 m from the reef edge). Divers collected three 0.02 m^2 corer samples, to depths of 15 cm at each sampling site.

Fish were measured to the nearest millimetre below. The stomachs were cut off at the oesophagus and pylorus. All prey items were separated by *taxon*, counted and weighed to the nearest 0.01 g. Depending on the level of digestion, prey items were identified to species or to the lowest possible *taxon*. The colonial taxa, hydrozoans and

bryozoans, and plants were not counted, so the numerical value attributed to those prey items was 1. Barnacles were counted based on their opercular pair structures, *tergum*, and *scutum* plaques.

AR and sandy seabed communities were compared in terms of the total abundance, and diversity [Shannon–Wiener index: $H' (\log_2)$] through one-way ANOVA (F-test). Prior to ANOVA, tests for normality (Anderson–Darling test) and homogeneity of variance (Bartlett’s method) among treatments were carried out (Zar 1996). ANOSIM (Bray–Curtis similarity matrix) was used to compare AR and sandy macrobenthic fauna (fourth-root transformation of the number of individuals per m^2) (Clark and Warwick 1994). The mean number of items per stomach, the mean number of items and the percentage of items preyed on per stomach, by season, were also compared using ANOVA (Zar 1996). Seasonal diversity of *D. sargus* diet was also tested. All univariate statistical analyses were carried out using Statistica V6.0 software, with a significance level of a $\alpha = 0.05$.

Dietary composition was assessed through mean numerical percentage (%N), mean weight percentage (%W), and frequency of occurrence (%FO) of each prey *taxon*, following Hyslop (1980). To evaluate the diet of *D. sargus* we used: (i) the vacuity coefficient (Hureau, 1970), (ii) the feeding coefficient (Q) (Hureau, 1970), and (iii) the index of relative importance (IRI) (Pinkas *et al.* 1971). Sigurdsson and Astthorsson (1991) scales were used to measure stomach fullness and the state of prey digestion.

Because the units of measurement for soft seabed and AR species (number m^{-2} , and cover percentage in the case of colonial species) is different from that of stomach contents (number of items per stomach), data were transformed to presence/absence, in order to construct a Bray–Curtis similarity matrix for multidimensional scaling (MDS) analysis. Stomach contents and data on sandy seabed macrofauna (three samples per

trimester) were pooled by season. Species of both AR and soft seabed samples were then used to assess items' provenance. All multivariate analyses were performed using the statistical package PRIMER (Clark and Warwick 1994).

RESULTS

Diplodus sargus was present at a high FO (67%) at the AR. Speared specimens ranged in size between 20.6 and 53.6 cm (mean 35.43 ± 6.65 cm). In situ visual census revealed an absence of juveniles (<20 cm total length) at the AR.

Macrozoobenthic community of ARs and neighbouring sandy areas

The number of *taxa* identified over soft substrata (SB) was higher (196) than over hard substratum (HS) (154). The most important groups available for fish foraging were polychaetes (SB = 94; HS = 52), crustaceans (SB = 52; HS = 47), bivalves (SB = 26; HS = 9), and gastropods (SB = 12; HS = 13). Cirripedia, Serpulidae, Bryozoa, and Ascidiacea were the major taxonomical groups that colonized the AR. Among soft seabed species, the most abundant *taxa* were Nematoda (14%) and Polychaeta (*Pisione* spp. and *Glycera lapidum* accounted for 13.71% and 7.55%, respectively). Despite the high number of *taxa* found in the soft-bottom macrobenthic community, the mean abundance was significantly higher over the AR HS ($92\,785 \pm 74\,645$ m⁻²) than over SB ($16\,464 \pm 6567$ m⁻²) (ANOVA: P = 0.01). Colonial *taxa* such as hydrozoans, bryozoans, and barnacles were not considered for the estimation of HS abundance. However, there was no change in Shannon–Wiener's diversity index between HS (2.75 ± 0.32) and SB (2.87 ± 0.33) communities (ANOVA: P = 0.29). Statistical analysis showed differences between the macrobenthic structure of the soft seabed and AR communities (ANOSIM: R = 0.98; P = 0.01).

Diet

The number of empty stomachs (33 out of 107) was low, reflecting a low feeding coefficient (30%) for this species. All remaining stomachs (74) with contents were analysed: 11 in winter, 12 in spring, 25 in summer, and 26 in autumn. Most (80%) of the sampled stomachs were reasonably full (50–75%) or full (>75%). Most of the prey items found in the stomachs (85%) were not yet digested or just slightly digested, which allowed their identification.

The diet of *D. sargus* contains a wide variety of food items. In all, 14 *taxa* were identified (Table 1). The most important taxonomic groups contributing to *D. sargus* diet (Q and IRI) were Crustacea (*Balanus amphitrite*) and Gastropoda, with *Gibbula* spp. also important. Those *taxa* live at the AR surface. Sandy seabed species were also present in the stomachs (e.g. *Parvicardium scabrum* and *Cassidaria echinophora*). However, compared with the AR species, these species contributed less to the diet of *D. sargus*. Some algae that colonize the ARs, such as *Cystoseira usneoides* and *Rhodomenia holmesii*, were also recorded in fish stomachs. Other plants, which do not colonize the AR, such as *Zostera* spp. and *Ulva lactuca*, were also frequently present in the stomachs. Bryozoans and hydrozoans, commonly found at the AR, were also observed in the stomachs. The burrowing polychaete *Polydora hoplura* and tube-building polychaete *Serpula vermicularis*, which is found in AR hard substrata, were also being preyed upon. There were no significant seasonal differences in diet diversity of the diet (Shannon-Wiener mean diversity: $H' = 1.84 \pm 0.76$; ANOVA: $P = 0.89$).

Table 1. List of the species found in *D. sargus* stomachs. Plus signs indicate *taxa* found exclusively on AR hard substratum and minus signs *taxa* found exclusively on sandy seabed. %N, mean numerical percentage; %W, mean percentage by weight; %FO, frequency of occurrence; Q, feeding coefficient; IRI, index of relative importance.

		N%	W%	FO%	Q	IRI
Anthozoa	Actiniaria ⁺	0.72	0.36	4.05	0.26	4.38
Bivalvia	<i>Anomia ephippium</i> ⁺	0.96	0.11	2.7	0.1	2.89
	<i>Hiattela artica</i> ⁺	0.24	0.00	1.35	0.00	0.33
	<i>Lima lima</i> ⁺	0.24	0.05	1.35	0.01	0.39
	<i>Parvicardium scabrum</i> ⁻	0.24	0.00	1.35	0.00	0.33
	Unidentified	0.72	0.00	4.05	0.00	2.93
Bryozoa ⁺		1.69	0.6	12.16	1.01	27.82
Cephalopoda	<i>Sepia officinalis</i>	0.48	12.66	2.7	6.1	35.53
Crustacea	Amphipoda	0.96	0.00	2.7	0.00	2.61
	Isopoda	0.24	0.00	1.35	0.00	0.33
	Decapoda	0.24	1.12	1.35	0.27	1.84
	<i>Diogenes pugilator</i>	0.72	0.26	1.35	0.19	1.32
	Pagurus spp.	0.72	0.38	4.05	0.27	4.47
	<i>Necora puber</i> ⁺	1.2	1.61	6.76	1.93	18.99
	<i>Polybius henslowi</i>	0.96	1.38	5.41	1.33	12.69
	<i>Goneplax rhomboides</i> ⁺	1.2	2.37	6.76	2.86	24.18
	<i>Xanthus spp.</i> ⁺	0.24	0.05	1.35	0.01	0.4
	<i>Balanus amphitrite</i> ⁺	18.31	2.54	17.57	46.53	366.35
Echinodermata	Asteroidea	0.48	0.06	2.7	0.03	1.46
	Echinoidea	2.41	1.22	12.16	2.94	44.17
	Ophiuroidea	0.48	0.09	2.7	0.04	1.55
Porifera ⁺		0.48	1.49	2.7	0.72	5.32
Foraminifera ⁺		0.24	0.00	2.7	0.00	0.65

Table 1 (Cont.). List of the species found in *D. sargus* stomachs.

		N%	W%	FO%	Q	IRI
Gastropoda	<i>Cassidaria echinophora</i> ⁻	0.48	1.3	2.7	0.62	4.81
	<i>Cerithium vulgatum</i> ⁻	0.48	0.07	2.7	0.04	1.5
	<i>Gibbula spp.</i> ⁺	40.24	9.65	12.16	388.34	606.79
	<i>Nassarium spp.</i> ⁺	2.17	0.15	6.76	0.32	15.67
	<i>Ocinebrina edwardsi</i> ⁻	0.24	0.00	1.35	0.00	0.33
	Unidentified	2.65	0.38	12.16	1.01	36.88
Hydrozoa ⁺		0.24	0.00	1.35	0.00	0.33
Nematoda		0.24	0.00	1.35	0.00	0.33
Osteichthyes	<i>Scomber japonicus</i> ⁺	0.24	15.11	1.35	3.64	20.74
	Sparidea ⁺	0.24	1.26	1.35	0.3	2.03
	<i>Trachurus trachurus</i> ⁺	0.24	1.1	1.35	0.26	1.81
	Unidentified	0.24	3.85	1.35	0.93	5.53
Plantae	<i>Zostera marina</i>	3.61	12.53	20.27	45.29	327.25
	<i>Zostera noltii</i>	1.45	1.64	8.11	2.37	25.00
	<i>Ulva lactuca</i>	1.2	5.69	6.76	13.72	93.23
	<i>Cystoseira usneoides</i> ⁺	0.48	1.77	2.7	0.85	6.09
	<i>Rhodymenia holmesii</i> ⁺	2.89	3.13	16.22	9.05	97.63
	<i>Aphanocladia stichidosa</i> ⁺	0.48	0.24	2.7	0.12	1.96
Polychaeta	<i>Polydora hoplura</i> ⁺	0.24	0.00	1.35	0.00	0.33
	<i>Serpula vermicularis</i> ⁺	0.72	0.02	4.05	0.01	3.01
Non-identified items		5.78	5.34	32.43	30.91	360.91

Feeding dependence on ARs

Most *taxa* found in fish stomachs were also present at the AR. Of the 39 *taxa* identified in stomach contents, 19 were found exclusively at the AR, and just three from the sandy surrounding area.

Diplodus sargus feeding dependence on the AR varied seasonally (Figure 1a). The number of items found in the diet was greatest in summer (35 items). Prey items were fewest in spring (11). However, neither the mean number of items per stomach (ANOVA: $P = 0.15$), nor the mean number of items per stomach preyed on from the AR

(ANOVA: $P = 0.43$) differed between seasons. Results showed that a high percentage of items contributing to the diet of *D. sargus* belong to the AR HS community. These values were slightly higher during winter and spring (74% and 79%, respectively) than in summer and autumn (63% and 65%, respectively). The rest of the diet was items of indeterminate origin, such as echinoderms and species belonging to the sandy macrobenthic community. However, differences between the mean percentages of items present at the AR between seasons were not significant (ANOVA: $P = 0.16$).

The MDS analysis (Figure 1b) highlights the relationship between the AR macrobenthic community and *D. sargus* diet. Three groups are clearly observed. The diet items are grouped near the AR items, whereas soft-bottom items are isolated and distant from the other two groups.

DISCUSSION

Our study was carried out in order to assess the importance of the macrobenthic community of an AR to the diet of *D. sargus*. One of the most important theoretical questions arising from AR deployment is whether the contribution of the food available at ARs (macrobenthic community production) leads to an increase in the biomass of reef fish assemblages. The high FO of *D. sargus* denoted reef fidelity, confirming previous results (Santos *et al.* 2005), which categorized the species as resident. The absence of juveniles at the AR was not a surprise, because it is known that juveniles prefer shallow rocky areas near the coast and in the Ria Formosa coastal lagoon, an important nursery area for the species, with high juvenile abundance (Monteiro *et al.* 1990). Additionally, recently tagged, reared, sub-adult *D. sargus* released at the same AR have been recaptured in coastal shallow waters along the south coast and in the Ria Formosa lagoon (Santos *et al.* 2006).

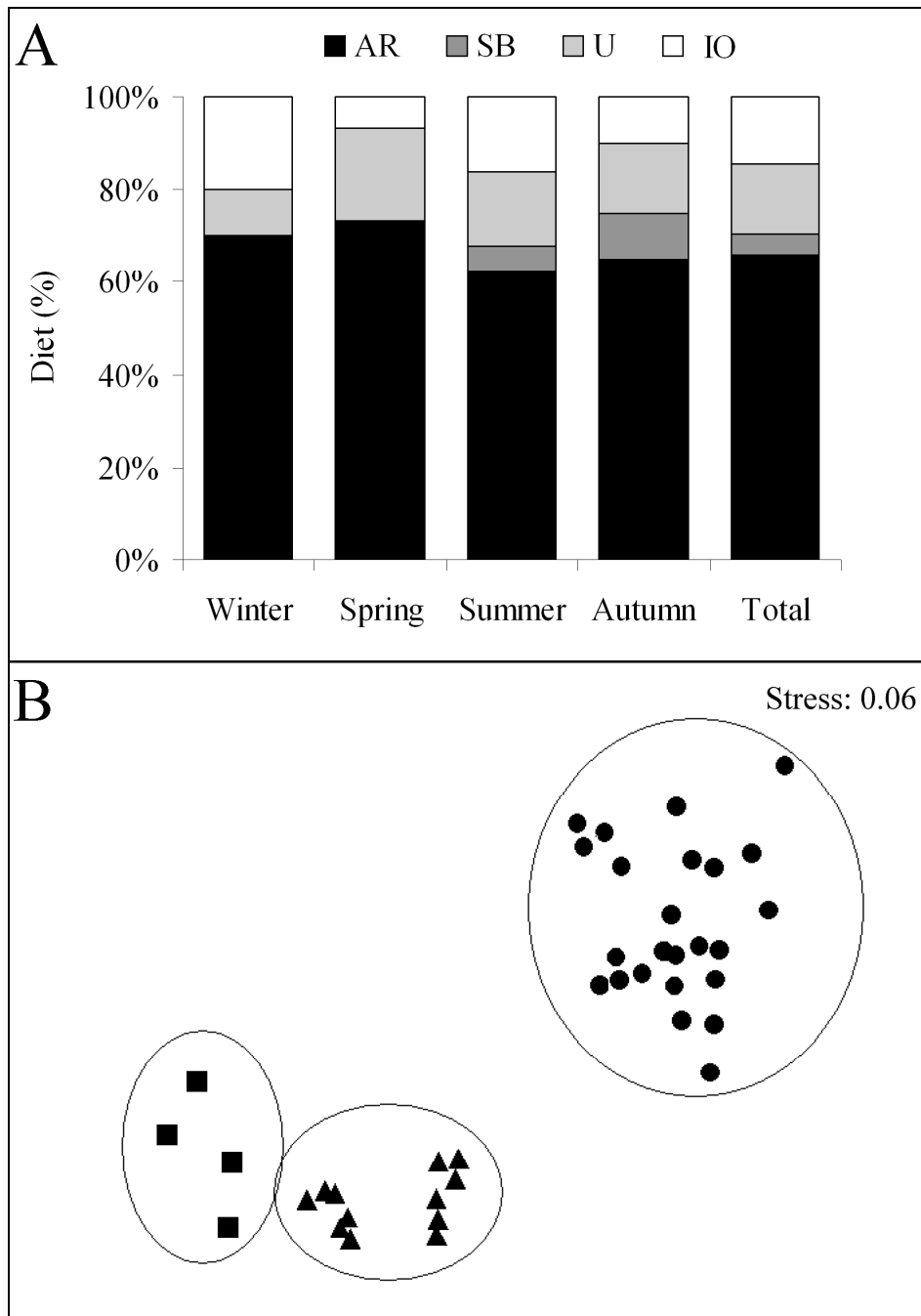


Figure 1. (a) Relative contribution of the different items (based on the number) found in *D. sargus* stomachs by season: AR, artificial reef species; SB, sandy seabed species; U, unidentified species; IO, species of indeterminate origin. (b) MDS ordination inferred from the macrobenthic communities from the AR and surrounding soft bottom, as well as from the diet of *D. sargus*. Filled circles - soft seabed species; triangles - artificial reef species; squares - diet.

Our results suggest that *D. sargus* benefits from ARs, using them as areas where food is plentiful. In fact, most of the prey we found belong to the AR benthic community. This highlights the importance for *D. sargus* of artificial habitats, which in this case cover an area of 12.21 km², at depths ranging from 17 to 24 m, in a 36 km² zone where natural reefs are scarce (occupying just 2.7 km²). Moreover, the stomach vacuity index was low, with the fullness and digestion levels showing that most stomachs were almost full and that feeding activity was recent. The AR's contribution to the diet of *D. sargus* was mainly through crustaceans, barnacles (*B. amphitrite*), and gastropods (*Gibbula* spp.), which were the dominant food items found in the stomachs. The presence of some polychaetes (*P. hoplura* and *S. vermicularis*) in the stomachs demonstrates that fish forage over hard substrata, because those species are common in artificial habitats (Boaventura *et al.* 2006). *Diplodus sargus* also feeds on algae which grow on AR hard substrata (e.g. *R. holmesii*), and on seaweeds (e.g. *Zostera noltii*) that are brought by currents and deposited on the AR blocks. Our results show that *D. sargus* takes a wide variety of prey, as reported previously by Figueiredo *et al.* (2005). Those authors classified *D. sargus* as a generalist, opportunistic, and remarkably omnivorous species. In contrast to our results, Pepe *et al.* (1998) showed that the diet of *D. sargus* was mainly sandy macrobenthic species of bivalves, gastropods, and echinoderms. However, the ARs of the area of that study (northwestern Sicily, Italy) are located close to a field of sea grass (*Cymodocea nodosa*), which was the main food item found in the fish stomachs. According to Pepe *et al.* (1998), the AR was more important as a refuge than as a feeding area. Nonetheless, Hueckel and Buckley (1987) showed that while fish may initially come to an AR for shelter or orientation, they soon become foragers on reef-produced items. *Diplodus sargus* is able to take advantage of the environments they colonize/inhabit. Therefore, as suggested by Baldó and Drake (2002), feeding on AR

macrofauna can be, among other reasons, a consequence of the available macrobenthic community rather than a feeding preference. The importance of ARs for fish diet has also been recorded in other studies (Donaldson and Clavijo 1994; Pike and Lindquist 1994). *Diplodus annularis* depends on AR fauna because the dominant prey items were crustaceans, amphipods, and decapods belonging to the AR community (Relini *et al.* 2002). Pepe *et al.* (1996) found that serpulid polychaetes were the most important prey of *D. vulgaris* foraging over AR HS.

According to Bohnsack's (1989) predictions, the biomass production and catches will increase as some function of the amount of AR material deployment. However, AR maturity and production is not immediate, and a lag before significant AR production and consequently fishery enhancement is to be expected. Hueckel and Buckley (1987) found that as an AR ages, food resources and predator populations associated with the reef also increase. When well-designed, located and constructed, with an adequate quantity of stable and durable substratum, man-made reefs can, in theory, be equally as productive as naturally occurring hard-substrata habitats, limited only by the lifespan of the materials utilized in their construction. Given the material used in the construction of the Algarve ARs, the structures in place could remain productive for several hundred years. Therefore, among the potential benefits of these man-made reefs, is enhancement of the availability of food for many years. Steimle *et al.* (2002) reported the importance of habitat AR value to enhance benthic productivity. This is also the case for the Algarve coast, for which it has been shown that ARs contribute to the increase of local biological production (Boaventura *et al.* 2006; Moura *et al.* 2006).

We have demonstrated that *D. sargus* uses the available biomass produced at the AR as food. Therefore, energy is transferred from the AR to the fish, and is used for fish

growth, movement, and reproduction. Because of *D. sargus* reef fidelity and the large size of the Algarve artificial habitats (covering more than 47 km²), it is reasonable to expect that these man-made structures will enhance the local fishery. Hopefully, this predicted enhancement of the *D. sargus* fishery will be confirmed in future through analysis of the evolution of the landings.

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CHAPTER 4

Effect of predation on artificial reef juvenile demersal fish species

Leitão F, Santos MN, Erzini K, Monteiro CC (2008) Effect of predation on artificial reef juvenile demersal fish species. *Marine Biology* 153: 1233-1244.

ABSTRACT

There is a concern that artificial reefs (AR) may act purely as fishing aggregation devices. Predators attracted to ARs can influence the distribution and abundance of prey fish species. Determining the role of predators in AR is important in advancing understanding of community interactions. This paper documents the effects of predation on fish assemblages of AR located near a coastal lagoon fish nursery. The *Dicentrarchus labrax* is a very opportunistic species preying on juveniles (0⁺ and 1⁺ age classes) of several demersal fish species on the ARs. Reef prey and sea bass abundance were negatively correlated. The mean numbers of prey per sea bass stomach increased with the increase of reef fish prey abundance, suggesting that predation has a significant influence, resulting in a decrease in prey abundance. Prey mortality (4 to 48%) of demersal reef fish associated species depends on bass density. Prey selection was related both with prey abundance and vulnerability. Results showed that *D. labrax* predation on AR-fish associated species can increase prey natural mortality. However, the role of bass predation on the ecological functioning of exploited ARs is not clear. There may be increases in local fishing yields due either to an increase in predator biomass through aggregation of sea bass attracted to ARs or to greater production. In contrast, predation on juveniles of economically important reef fish preys, especially the most frequent and abundant (*Boops boops*), can contribute to a decrease in recruitment to the fishery. Our results indicate that inter-specific interactions (predator-prey) are important in terms of conservation and management, as well as for the evaluation of the long-term effects of reef deployment. Thus, it is necessary to consider ecological interactions, such as predation, prior to the development and deployment of artificial habitats as a tool for rehabilitation.

INTRODUCTION

Populations of marine fishes are often characterized by dramatic fluctuations in abundance. However, the causes of such variations are difficult to measure and quantify due to the different scale effects that natural and anthropogenic factors may have on the ecosystem. Most of the studies at the ecological level to date have directly or indirectly concerned the question of whether reef fish assemblages are structured by competition or recruitment limitations (Hixon 1991). The debate concerning the dynamics of reef fish populations has centred on the relative contributions of density-independent factors acting during larval dispersal and density-dependent processes following the larval stage (Hixon *et al.* 2002; Hixon and Webster 2002). Although a growing number of field studies have documented density-dependent mortality in reef fish populations (Hixon and Webster 2002; Hixon and Jones 2005), very few studies have identified the actual mechanisms and conditions responsible for these patterns (Hixon and Carr 1997; Forrester and Steele 2000; Anderson 2001).

Abrams (1987) demonstrated that the interaction between prey and predator species could be positive, negative or neutral, depending on the population dynamics of the predator and prey species involved. The idea that fish predation can strongly affect the distribution and the abundance of prey species within a community is a central tenet of modern ecology (Hixon and Beets 1993). It is often hypothesised that predation can significantly reduce the abundance of juvenile fish attracted to reefs for refuge or feeding, and this may lead to a reduction of total biomass (Hixon 1991; Stewart and Jones 2001; Stewart and Connell 2002). However, evidence of predation and its direct effects is difficult to obtain in most systems.

Artificial reefs (AR) are deployed worldwide with a number of goals, including the mitigation of habitat (coral reefs) loss, enhancement of fish and bivalve catches

(Bohnsack and Sutherland 1985; Monteiro and Santos 2000), and habitat protection (Bayle-Sempere *et al.* 1994). Since ARs are very efficient aggregation tools (Bohnsack and Sutherland 1985; Polovina 1991), there is a concern that greater natural mortality of prey may result from high concentration of predators. Predation may play an important role in artificial habitats, with the structure of the food web controlled by the biodiversity within the system and/or by top predators (top-down control). Although predation was proposed as an important structuring factor in the 1970s (Hixon 1991), it has received little attention as far as AR studies are concerned. From both management and biological perspectives it is necessary to consider the secondary/indirect effects of predation on the environment, prior to the development of artificial habitats as a tool for rehabilitation.

Most fish predation studies in reefs have focused on predator-prey relationships between resident predator and prey species and the role of reefs in providing shelter from predation (Shulman 1985a,b; Hixon and Bettes 1993; Hixon and Jones 2005). However, few studies have examined the influence of transient predators on reef fish assemblages, mainly because of the difficulty in assessing predator pressure of this nature (Carr and Hixon 1995).

Dicentrarchus labrax Linnaeus (common sea bass), an important transient predator that had rarely been observed on the Faro AR (deployed in 1990) in the fifteen years following deployment (Santos *et al.* 1995a; Santos *et al.* 2005) has been recorded frequently and abundantly since the enlargement of the artificial reef system in 2002 (Leitão *et al.* 2008).

Given the lack of resident piscivores and the dominance of the sea bass in terms of biomass on the reefs (Leitão *et al.* 2008), it was hypothesized that this transient predator could have a significant impact on AR prey species. Predator density may lead

to increased consumption of prey as a result of more predators consuming more prey in total. It has been recognized that piscivorous predatory fish species are more likely to respond to larger aggregations of prey (Stewart and Jones 2001; Connell 2002). This may cause an increase in the proportional mortality of aggregated prey, since the predators may feed at a greater rate (Connell 2000; Connell 2002). Such feeding behaviour may not only destroy the protection afforded to fish in larger schools, but it may also cause greater per capita mortality in larger schools (density-dependent mortality), consequently resulting in the reduction of prey abundance (Connell 2000; Stewart and Jones 2001). More directly, inverse relationships have been noted between the local abundances of prey fish and resident piscivores in natural rocky and coral reef areas (Shulman *et al.* 1983; Shulman 1985a, b; Hixon and Beets 1989; Hixon and Beets 1993; Overholtzer-McLeod 2006; Johnson 2006).

This paper documents the effects of predation in structuring AR reef fish assemblages. We tested whether *D. labrax* (top predator) affects the structure of demersal artificial reef fish assemblages. Firstly, we evaluate the relationship between predator and prey abundances. Secondly, we quantify the number of prey consumed by the predator when prey numbers varied. Third, the influence of predator abundance on prey mortality was studied. Finally, the question of predator preferences was examined.

MATERIAL AND METHODS

Study area

In the south coast of Portugal (Algarve) multi-purpose ARs have been constructed and deployed since 1990. Their deployment aimed to increase the amount of hard bottom habitat, which is scarce on the southern grounds, to provide suitable habitat and protection for juvenile fish that annually migrate from the lagoon systems located along

the southern coast, to promote biodiversity and to increase fishing yields (Monteiro and Santos 2000).

The sampling site chosen for this study was the Faro/Ancão artificial reef system (Lat 36° 59.25'; Long 8° 00.43') 4km off the Ria Formosa lagoon, an important nursery area (160 km²) that supplies juvenile fishes to nearby coastal waters (Figure 1). Four AR groups were surveyed, specifically FP1_{a,b,c}, FP3_{a,b,c}, FP5_{a,b,c} and FP7_{a,b,c}, deployed in 1990 off Faro beach (Algarve, Southern Portugal) on clean sandy grounds (few natural rocky areas nearby) at depths of approximately 21-23m. These ARs, were selected since we assume that they have been fully colonized by reef fish assemblages (Santos *et al.* 2005). Given the distance between reef groups (~300m), these were considered to be independent sampling locations. Each artificial reef group comprises three reefs sets, with 35 concrete cubic units each, organized in a two-layer pyramid (Figure 1). Each reef set occupies an area of approximately 132m² corresponding to 529m³ in water volume (Santos 1997).

Methodology

The understanding of the trophic relationship between the *D.labrax* and the AR fishes necessarily requires knowledge of changes in predator and prey abundance within the AR and the diet of the predator.

To characterise predator-prey assemblage abundance changes, a long-term study (from August 2003 to September 2004) using visual censuses was carried out on a monthly basis. Data were recorded by scuba divers who documented size, density and behaviour (feeding and site fidelity of the species). The stationary point count technique as described by Santos *et al.* (2005) was used. The data were recorded with an underwater writing pad, always by the same diver.

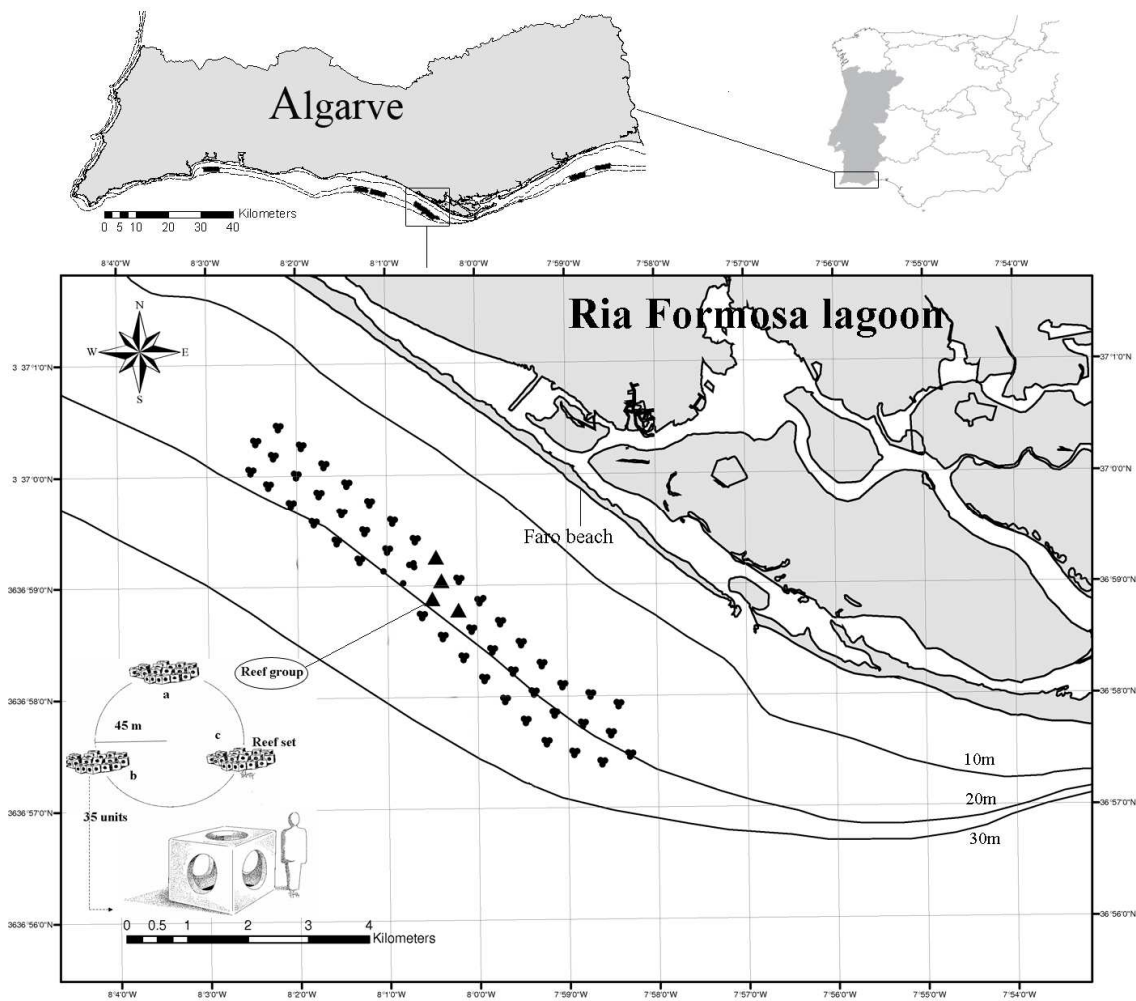


Figure 1. Algarve artificial reef complex, study site i.e Faro/Ancão AR system (large triangles are AR groups where data were collected), the reef modules and their spatial and structural organization.

During each trial the diver sampled all three reef sets of each reef group. In each reef set, three independent random counts were carried out. As daily variation of reef fish fauna is found in these ARs (Santos *et al.* 2002), the visual censuses were always carried out between 9:00 and 12:00AM and under similar tide conditions.

Sea bass specimens were collected by spear fishing every month from October 2003 to September 2004, except for February due to rough sea conditions. This method increases the probability of prey identification and guarantees that fish were collected in the AR. In order to associate predators with prey (diet), sea bass were caught immediately after the census by a second diver and once onboard placed in ice in order to stop digestion. At the laboratory total length of each specimen was recorded (cm). After removal, the stomachs were fixed in a 4% formaldehyde solution during 48 hours. After washing with running water, stomach contents were sorted under a stereoscopic lens. All prey items were separated by *taxon*, counted and weighed to the nearest 0.01g. Depending on the state of digestion, prey items were identified to the lowest possible taxonomic level.

Statistical analysis

Species with high site fidelity, such as cryptic and sedentary species, which generally take a stationary position in the AR or on the bottom around it, were defined as residents (e.g. Blenniidae) and species that do not shelter in AR units and that show a swarming response to the reef, (e.g. *Boops boops*) upon the approach of piscivores or divers were designated as reef associated species. Reef associated species are those that occurred within a distance of approximately 1.5m to the reef as recorded during the visual censuses.

Stomach content analysis

The frequency of occurrence (FO), calculated as a percentage of the number of stomachs with prey divided by the number of stomachs with items, and the percentage in number (%N) and weight (%W) (Hyslop 1980) were used to assess diet composition

of the sea bass. The most important food items were determined using the feeding coefficient ($Q = \%N \times \%W$), which characterizes the relative importance of the different preys in a diet (Hureau 1970). Using Q , prey were separated into three categories (principal prey, $Q > 200$; secondary prey, $20 < Q < 200$; Occasional prey, $Q < 20$). The index of relative importance (IRI) (Pinkas *et al.* 1971) was also estimated, $IRI = (\%N + \%W) \times FO$. The measurement of the stomach fullness (Full stomach $\geq 75\%$; Quite full 50 – 74.9%; Half-full 25 – 49.9%; Almost empty 5 - 24.9%; Empty 0 – 4.9%) and prey digestion state was carried out according to Sigurdsson and Astthorsson (1991). Predation (%) was calculated based on monthly data as the number of times sea bass fed on AR fish prey species, when both sea bass and prey species were simultaneously observed in the AR group reef set. Predation validation was made in the laboratory after checking if predators had the prey species observed in the AR in their stomachs.

Predator - prey relationships

Several hypotheses concerning prey-predator relationships were tested based on the visual census and stomach content data, and the assumption that there are no resident AR piscivores:

- 1) The first approach examined the relationships between prey and predator abundances (Hypothesis I):

Ho: there is no relationship between sea bass and prey abundances

H_A: there is a relationship between sea bass and prey abundances

The relationships between predator and prey abundances were analysed through Spearman correlations (Zar 1993). If predation is the predominant process regulating the number of reef-associated prey, then there should be a negative correlation between the

mean number of predators and the mean number of prey per reef averaged over all censuses. Mean density of both *D. labrax* and the reef prey species found in the stomach contents of the sea bass were calculated by averaging monthly reef set data counts. Given that the data were based on visual census methods, a probability level $\alpha = 0.1$ was used in all analyses in order not to reject the null hypothesis when this is true (Type II error).

2) The second approach was to quantify the number of prey consumed (diet) by the predator when AR prey numbers varied (Hypothesis II):

Ho: There is no relationship between the numbers consumed and the number of prey available

H_A: There is a relationship between the numbers consumed and the number of prey available

Visual census data and stomach data were used in order to evaluate if predation is influenced by the abundance of the AR available reef fish prey. Regression analysis was carried out between mean reef set prey abundance per month (independent variable) and the mean number of preys per stomach.

3) If bass predation influences the number of reef-associated prey-fish, then there should be a positive relationship between the mean number of predators and the mortality of prey (Hypothesis III):

Ho: There is no relationship between prey mortality and the number of predators.

H_A: There is a relationship between prey mortality and the number of predators

Prey mortality, M(%), was estimated as follows:

$$M (\%) = (CN_i / TN_i) * 100 \quad \text{eqn. 1}$$

where, CN_i is the number of prey i consumed, calculated as the product of the mean number of prey i per bass stomach (a_i) and the total number of bass (b) observed on the reef ($CN_i = a_i * b$), and TN_i is the estimated total number of prey species i on the reef, that is calculated as the sum of the estimated total number of prey i consumed on the reef and the number of species i on the reef estimated by visual census. Given the condition of the prey species in the bass stomachs, it is assumed that predation is recent and took place at the reef where the prey and predator species were quantified. Regressions were carried out between the monthly prey mortality (%) and predator mean monthly abundance per reef set.

4) Finally, the question of predator preferences was examined (Hypothesis IV):

Ho: The bass has no preference

H_A: The bass is a selective feeder

Ivlev's index (Ivlev 1961) was used as a measure of electivity (E) for the reef fish species in the fish diet: $E = (d_i - p_i)/(d_i + p_i)$ where d_i is the % of food item in the diet, and p_i is the % of food item in the environment. The mean Ivlev's electivity index, and standard deviation, was calculating using monthly data. Values of Ivlev's electivity index range from -1.00 (complete avoidance) to $+1.00$ (exclusive selection). The p_i contribution of the different fish prey items in the AR was estimated. This was possible because visual census observations were carried out monthly for the whole reef fish assemblage. Nevertheless, considering the aim of the study, only information regarding fish species was used in electivity index estimation.

RESULTS

Predator Diet

A total of 74 sea bass specimens were caught, comprising 46 males, 26 females and 2 of undetermined sex. Their total lengths ranged from 33 to 74.2cm, with a mean value of 47.1 ± 10 cm.

A total of 17 stomachs (23%) were empty. A total of 142 prey items were found and identified in the remaining stomachs (57). The mean number of items found per stomach was 2.5. Full stomachs and quite full stomachs represented 5% and 36%, respectively. The remaining stomachs sampled were half full and almost empty, representing 33% and 26%, respectively. The majority of preys found in the stomach contents were very easy to identify (69%), having been recently ingested or showing an early phase of digestion. In fact, of the above proportion 31% of the items were assigned as non-digested (partially intact items) and 38% as slightly digested.

Sea bass diet was composed of a low variety of food items, including crustaceans, gastropods and finfish (Table 1). Crustaceans and fishes were the items contributing most to the sea bass diet, as far as the number (N%) and weight (W%) of prey is concerned (Table 1).

The most important taxonomic groups (Q and IRI) contributing to the sea bass diet were the crab *Polybius henslowi* Leach (principal prey) and several reef associated demersal fish species. The latter include *Boops boops* Linnaeus (principal prey), *Pagellus acarne* Risso and *Scomber japonicus* Houthuym (secondary preys), and *Trachurus trachurus* Linnaeus (occasional preys). The bass diet also includes resident species belonging to the Blenniidae family (occasional preys). Moreover, stomach content of sea bass also revealed hard reef associated species such as *Necora puber* Linnaeus (Table 1). However, the most frequently observed items were *B. boops*, *S.*

japonicus and *P. acarne*. The most important prey in numbers (N%) were *B. boops* and *S. japonicus* (Table 1), with the latter having a greater mean length (Table 2) and thus contributing more in weight (W%) to predator diet than the other fish species (Table 1).

Table 1. Diet composition of the predator (*Dicentrarchus labrax*). %N – numeric percentage; %W- weight percentage; FO - frequency of occurrence; Q - feeding coefficient; IRI - index of relative importance.

<i>Taxa</i>	N%	W%	FO	Q	IRI
Crustacea					
Amphipoda	1.43	0.00	2.33	0.00	3.32
<i>Necora puber</i>	2.86	0.87	4.65	2.48	17.33
<i>Polybius henslowi</i>	44.29	48.25	18.60	2136.66	1721.54
Total	48.57	49.12	25.58	2385.61	2498.97
Gastropoda					
Unidentified	2.86	2.73	4.65	7.81	26.00
Osteichthyes					
<i>Boops boops</i>	24.29	19.33	37.21	469.34	1622.76
Blenniidae	5.71	0.25	2.33	1.42	13.87
<i>Pagellus acarne</i>	4.29	7.10	6.98	30.44	79.46
<i>Scomber japonicus</i>	10.00	19.30	6.25	192.98	183.11
<i>Trachurus trachurus</i>	2.86	2.17	6.98	6.19	35.05
Total	47.14	48.14	67.44	2269.51	6426.13
Algae					
Unidentified	1.43	0.01	2.33	1.42	3.35

Prey-predator behaviour

In situ observations found that sea bass swims both around and in the inner part of the AR (top, middle and bottom layers of the AR). In relation to reef fidelity the sea bass was categorized as a transient species. Although the sea bass was observed entering the ARs to avoid divers, they usually leave after 5 to 10 minutes, probably due to the stress induced by the presence of the divers.

All the associated reef fish species found in the sea bass stomachs, except the blennies (resident species) were demersal (mid-water) species. These prey species showed a relatively wide home range, moving throughout the external reef area, in the middle and top layers. When disturbed by divers or by the presence of the sea bass, these species never take refuge within the ARs.

Predator - prey assemblage structure and variations in abundance

The frequency of occurrence of sea bass was high (86%) with a mean number of 55 individuals per reef set and a mean total length class of 40cm that was slightly above the size at first maturity (Table 2). The most frequently occurring prey species found in the ARs were *B. boops* and blennies, with both *taxa* being observed in all counts (Table 2). The frequency of occurrence of *P. acarne* was also high, 54% (Table 2), while *T. trachurus* and *S. japonicus* showed a lower frequency of occurrence.

In terms of abundance, the most important species was *B. boops*, followed by the blennies and *P. acarne* (Table 2). Both the mean and maximum length of all demersal reef fish species found in sea bass stomach were below their size at first maturity (Table 2).

Table 2. General information regarding the predator (*Dicentrarchus labrax*) and reef fish preys. FO - Frequency of occurrence; Mean (\pm standard deviation) and maximum (Max) species abundance (n.º individuals per reef). ML – mean total length; L-1st Mat - size at first maturity (reference between brackets); M – range of prey percentage mortality; Juvenile -percentage of juveniles. Predation - percentage of times *D. labrax* forage upon available preys; E - Ivlev electivity index (standard deviation).

	FO	Abundance ^c		ML ^c (cm)	L-1st-Mat	M (%)	Juvenile ^c (%)	Predation (%)	E
		Mean	Max						
<i>Dicentrarchus labrax</i>	86	55 \pm 24	264	40 \pm 3	38.5 (Farrugio and Le Corre 1986)		56		
<i>Boops boops</i>	100	499 \pm 3 67	1266	6 \pm 3	15.7 (Monteiro <i>et al.</i> 2006)	4 - 35 ^b	99.7	73	+ 0.38(0.20)
<i>Pagellus acarne</i>	54	74 \pm 16	388	10 \pm 2	21 (Santos <i>et al.</i> 1995b)	7 – 48 ^a	100	43	+ 0.29(0.36)
<i>Scomber japonicus</i>	25	46 \pm 92	269	21 \pm 3	31 (Anon. unpublished)	13 – 40 ^a	100	50	+ 0.09(0.10)
<i>Trachurus trachurus</i>	29	54 \pm 101	318	12 \pm 2	22 (Borges and Gordo unpublished)	25 – 36 ^a	100	75	- 0.07(0.03)
Blenniidae	100	76 \pm 21	678	6 \pm 2	-	-	-	9	- 0.18
Total prey	-	749 \pm 493	2242		-	7 - 25 ^b	-	90	

^a Based on the range of monthly mean mortality estimations (just for demersal reef associated species)

^b Based on regression analysis (just for demersal reef associated species)

^c Based on visual census.

Predator - prey abundance variation

The monthly variations in abundance of sea bass and reef fish prey found in bass stomachs are shown in Figure 2. High abundances of *D. labrax* per reef set occur between December and February. High densities of *B. boops* were found in May and August, with low values in February and March. High densities of *P. acarne* were observed in April and May. *T. trachurus* was only observed between February and June, with high abundances recorded in May and June. *S. japonicus* was recorded in April and later again between July and September when abundance was high. High overall prey abundances were recorded between April and May (2004) and the lowest between January and March (2004). The mean number of total prey available was 749 individuals per reef set (Table 2).

Predator - prey relationships

The highest abundances of *D. labrax* were recorded when the total abundance of preys was below their mean abundance value (Figure 2; Table 2). In contrast, highest total prey values were recorded from April to July when the numbers of sea bass were below the mean. Relationships between prey-predator abundance showed negative correlations between *D. labrax* abundance and both *B. boops* ($r = -0.6$; $P=0.03$; $df= 13$) and *P. acarne* ($r = -0.5$; $P = 0.09$; $df= 6$). Negative correlations between *D. labrax* abundances and both the latter prey species abundance suggests that predation affects prey abundance (Hypothesis I). However, correlations between sea bass and both *S. scomber* ($r = -0.3$; $P = 0.33$; $df= 3$) and *T. trachurus* ($r = 0.1$; $P = 0.77$; $df= 4$) were not statistically significant. Nevertheless, a strong negative correlation was observed ($r = -0.6$; $P = 0.03$; $df= 13$) between sea bass and total reef prey species abundance,

suggesting that sea bass predation significantly affects the abundance of AR demersal prey.

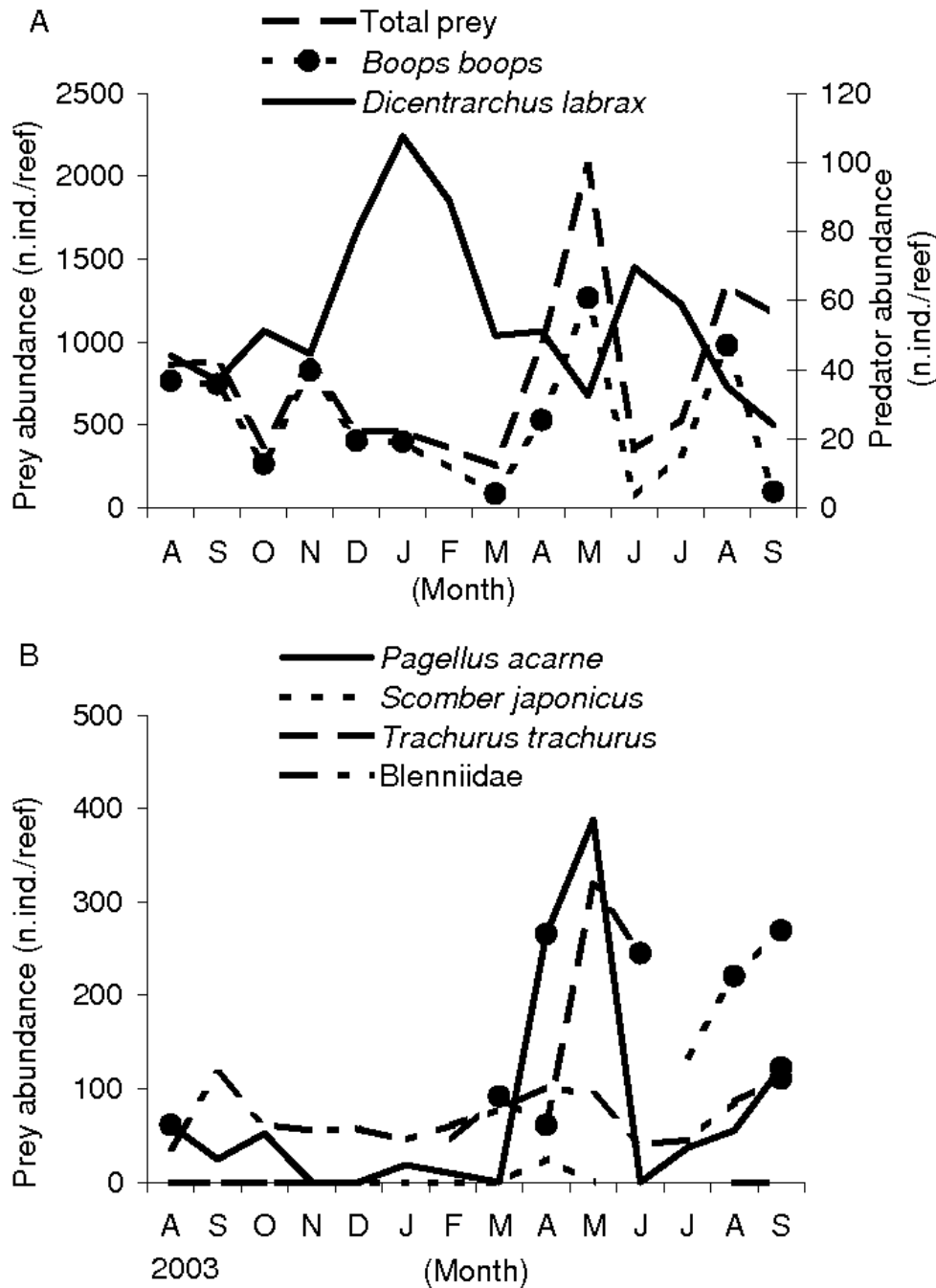


Figure 2. A - Monthly density variation of *D. labrax* (predator), *B. boops* (principal prey) and overall reef fish preys. B - Mean abundance of the other prey species (*P. acarne*, *S. japonicus*, *T. trachurus* and Blenniidae). Filled circle (●) represent the presence of a given prey in sea bass stomachs in a given month.

The relationships between the number of each prey species per sea bass stomach and prey abundance (Hypothesis II) is shown in Figure 3. There was a significant relationship between consumption and abundance (or availability) of *B. boops* ($P = 0.04$; $df = 10$) and *T. trachurus* ($R^2_{T. trachurus} = 0.56$, $P_{T. trachurus} = 0.02$; $df = 3$). For *S. japonicus* ($R^2_{S. japonicus} = 0.42$, $P_{S. japonicus} = 0.15$; $df = 3$) and *P. acarne* ($R^2_{P. acarne} = 0.05$, $P_{P. acarne} = 0.9$; $df = 6$) the relationships were not significant. A significant positive relationship between consumption and overall prey abundance was found ($P = 0.01$; $df = 10$; Figure 3B). These results suggest that for Hypothesis II, the null hypothesis can be rejected and we can conclude that there is a relationship between prey abundance and predation by sea bass on the artificial reefs.

Independently of the monthly availability of prey abundance (Figure 2), predation on reef fish species occurred every month, with the exception of July when all reef fish prey species were absent from the bass diet. However, in the latter month the overall abundance of fish reef species was very low (Figure 2). Nonetheless, overall predation occurred 90% of the times that both sea bass and prey species were simultaneously observed in the AR (Table 2; Figure 2). Predation was high for *T. trachurus* (75%) and *B. boops* (73%), but lower for *S. japonicus* (50%) and *P. acarne* (43%) (Table 2). It is worth noting that although several AR fish species were available to *D. labrax*, it preferentially fed on *B. boops*, that was the only prey species that was always available on the ARs (Table 2). Indeed, despite other prey species being available in the AR, they were not always eaten by the sea bass (e.g. *P. acarne* in January, May and June). In November and December, although *S. japonicus* was not observed in the AR, it was found in the bass stomachs.

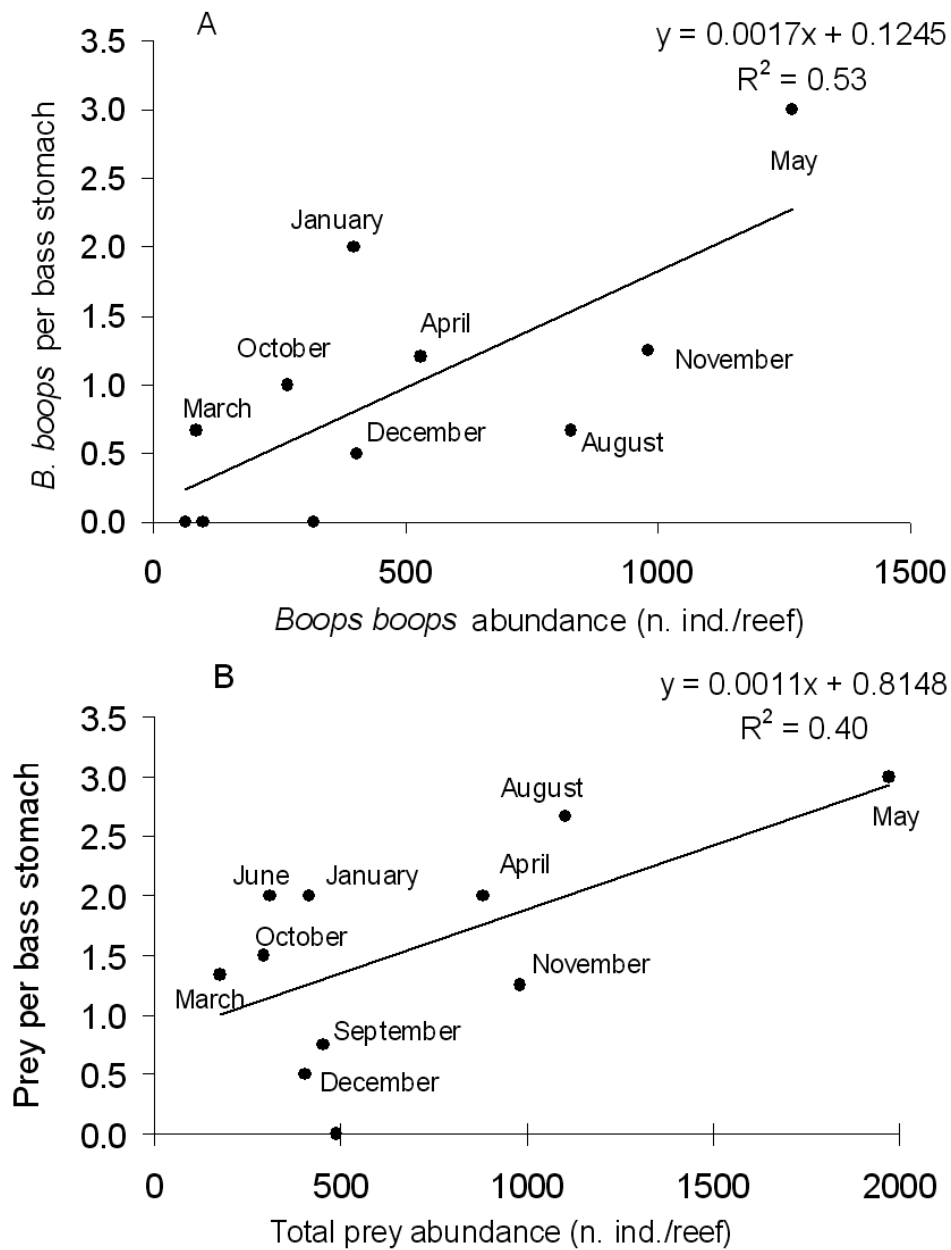


Figure 3. Predator-prey relationship between the number of individuals (A for *B. boops* and B for total demersal reef fish associated prey species) found per sea bass stomach as a function of prey abundance on the reef.

A positive relationship (Hypothesis III) was found between *B. boops* mortality and bass abundance ($P = 0.07$; $df = 10$, Figure 4A). For the remaining species, relationships were not significant. Nevertheless, a strong significant positive relationship ($P = 0.03$; $df = 10$) was also found between predator abundance and overall

prey mortality (Figure 4B). Therefore, we reject the null hypothesis and accept that predation abundance affects prey mortality. Prey mortality depends on bass abundance. For instance, the highest number of preys available was recorded in May but mortality was low as bass abundance was low. In April bass abundance was high and consequently mortality increased (Figure 3 and 4). Overall demersal prey mortality varied between 7 and 25 %. However, the maximum mortality value was high ($\geq 35\%$) for all prey species (Table 2). In June mortality was exclusively of *T. trachurus* (36%) that was the most abundant species. In October mortality was exclusively of *P. acarne* (48%).

Ivlev's electivity index denotes high positive electivity values for *B. boops*, indicating bass prey selective preference (exclusive selection) on the latter species (Table 2). The Ivlev electivity index was also positive for *P. acarne*. However, deviance values for *P. acarne* and *S. scomber* allow the predator to be categorized as being both selective and with no preference for these species (Table 2). The electivity values and associated deviance to *T. trachurus* (Line 481) suggested little or no bass preference for this species. The electivity value was negative in the case of the blennies, meaning that bass do not forage on this resident reef fish group. Considering Ivlev's electivity index, and respective associated deviances, it is possible to state that the feeding preference of sea bass is affected by the available abundance of demersal reef species, such as the *B. boops* and *P. acarne* that were the reef fish species that contributed most to the *D. labrax* diet. Considering Ivlev's index (Table 2) and both Q and IRI (Table 1) for hypothesis VI, the tentative conclusion is that the bass is selective. However, this preference was strongly affected by the abundance of the available prey (see Hypothesis II).

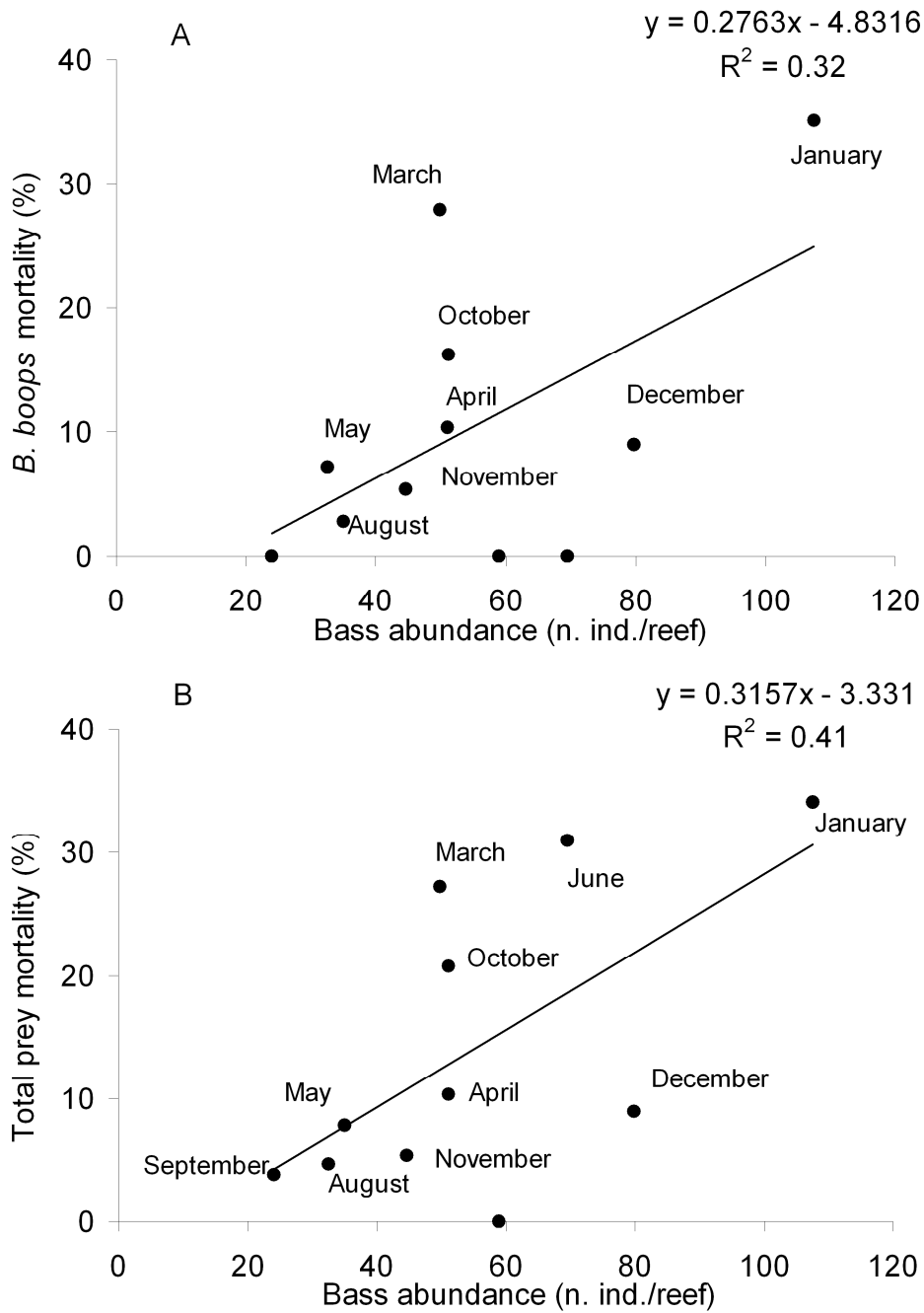


Figure 4. Prey percent mortality (M%), for *B. boops* (A) and for all demersal (B) reef fish associated prey species, as a function of the number of predators.

DISCUSSION

The effect of predation by piscivores in AR fish assemblages is poorly known. Nonetheless, in coral reef areas (Hixon and Bettes 1993; Hixon and Jones 2005), rocky intertidal (Sih *et al.* 1985; Connell 2002; Johnson 2006) and freshwater systems (Zaret

1980) it has been shown that predation can have a significant effect on the distribution and abundance of prey fish species within a community. Our study showed that *D. labrax*, a transient predator, feeds on juvenile demersal species, such as *B. boops*, *T. trachurus*, *P. acarne* and *S. japonicus*, which are attracted to the ARs. These species are among the most abundant in ARs (Santos 1997; Santos *et al.* 2005). Moreover, they occasionally also feed on some resident fish species (belonging to the Blenniidae family) and hard reef macrobenthic species such as *N. puber*, a common species in Algarve ARs (Leitão *et al.* 2007). Other studies also found several finfish species (*D. labrax* (cannibalism), *Pomatoschistus spp.*, *Sprattus sprattus*, *Atherina boyeri*) and a variety of macrozoobenthos species in the sea bass diet (<http://www.fishbase.org>). In the present study, demersal reef fish prey species were found in the stomach of sea bass in almost all months and in addition, predation on total reef fish prey items occurred 90% of the time. It seems that the sea bass is a very opportunistic species that takes advantage of the overwhelming concentration/aggregation of small fish attracted to ARs. The number of empty stomachs was low, with the prey digestion level indicating that feeding activity was recent, probably during the early morning hours.

The idea that fish predation is strongly influenced by prey abundance is highlighted in many studies (Hixon and Carr 1997; Stewart and Jones 2001; Connell 2002). Connell (2000) discovered that larger schools of fish suffer greater rates of mortality, a direct challenge to the idea of safety-in-numbers. A similar phenomenon may also occur in the Algarve ARs with sea bass predation on juvenile (0^+ and 1^+ age classes) demersal reef species, especially on those that were recorded in high abundances and frequency of occurrence, such as *B. boops* and *P. acarne*. Carr and Hixon (1995) observed that the low survivorship (40-80%) of new recruits on reefs where resident predators had been removed may be due to transient predators (e.g.

jacks, Carangidae). The influence of predation on early juveniles of commercially valuable species, such as sea bass, was observed to cause density-dependent mortality (Laffaille *et al.* 2000). In fact, in our study a negative correlation was found between total demersal AR prey abundance and the sea bass numbers, suggesting that predation can influence prey abundance (Hypothesis I). Moreover, the mean numbers of prey per sea bass stomach increased with the demersal reef fish prey abundance (Hypothesis II) and higher predator density caused higher mortality of demersal reef fish associated species (Hypothesis III).

Understanding natural causes of density dependence is essential for identifying possible sources of population regulation (Hixon 1991). Hixon and Beets (1993) found that reef associated piscivores do not always control the number of co-occurring fish prey, but rather set the upper limit to the number of fish prey that occupy a reef. On some reefs, where predator numbers were high, the latter authors showed that average abundance of prey decreases over time as predators increase. Moreover, in the same study the authors reported that there was a clear negative correlation between predator numbers and prey abundance. Nevertheless, they considered these relationships causal and explained the results by the rapid consumption of recruited cohorts on reefs by resident piscivores, with predation occasionally directly observed. Herrera *et al.* (2002) observed that some piscivore species were chiefly responsible for controlling AR fish productivity. The authors report that the seasonal arrival of dense schools of small pelagic species seems to attract piscivores from the sandy bottom biotope with 1-2 months time lag.

The hypothetical estimates of mortality bridge the conceptual link between density and loss of prey at varying predator density. The issue of whether predator aggregation is strong enough to cause proportionally greater predator-driven declines as

prey abundance increase is fundamental to the concept of artificial habitats fish assemblage regulation. Results showed that prey consumption was dependent on prey abundance and that mortality increase with bass abundance. Predators often account for a large fraction of mortality (e.g. Carr and Hixon 1995; Connell 1997; Hixon and Carr 1997; Forrester and Steele 2000) and predator impacts can be reduced by habitat manipulations that increase the availability of shelter for prey or reduce prey encounter rates with predators (Anderson 2001, and references therein; Forrester and Steele 2004; Overholtzer-McLeod 2006).

The deployment of ARs in the Algarve may contribute to an increase in the natural mortality of juvenile, or young of the year, demersal reef prey associated species by facilitating predator-prey interactions. In this study prey mortality varied between 4 and 48% and may be particularly high ($\geq 35\%$) for all prey species. When properly designed, located and constructed, with an adequate quantity of stable and durable substrate, man-made reefs can, in theory, be equally as productive as naturally occurring hard-bottom habitats, limited only by the life-span of the materials utilized. Given the material used in the construction of the Algarve ARs, the structures in place could favour prey-predator encounters for the next several hundred years. Given the large number of ARs in Algarve waters (588 AR sets, 20580 units), constituting the largest complex of this type in European waters, the predation of bass on aggregations of juveniles could have a cumulative effect on local prey populations, especially those with high frequency of occurrence, abundance and that suffer higher mortalities on AR (e.g. *B. boops*). Thomas (1974) showed that fishes decrease the linear distance travelled after successfully discovering food and increased it after rejecting a food item. This behaviour facilitates avoiding the unproductive foraging areas, increasing the changes of discovering productive areas, and remaining in the proximity of discovered food.

Optimal foraging theory (reviewed by Krebs 1978; Hart 1986) could also be applied to movements of bass between reefs. Predators are predicted to distribute themselves and to move between reefs so as to maximize net energy gain. Therefore fishes should distribute between reefs according to reef profitability. Theories of “marginal value” (Hart 1986) and “giving up time” (Krebs 1978) predict that predators should leave a reef when the energy yield from food resources is reduced to a certain level. Foragers should spend more time at reefs with abundant food resources than at reefs with low food availability.

Results showed that predation was the primary cause of density-related mortality. Nevertheless, prey mortalities range considerably according to bass density. Rather than density-dependence, 'density-vague regulation' (Sale and Tolimieri 2000; Strong 1986) may be more useful for explaining bass predation/mortality on ARs. The “density-vague” approach to population regulation recognizes that the influence of density is weak if present over a broad range of densities and that variation within this range is caused by other factors (Strong 1986).

Some AR species were not always present in the bass diet. This may be related to predator feeding preference. Indeed, although prey selection was strongly affected by the abundance of some prey (*B. boops*) our results (Hypothesis IV) suggest that the bass is selective. Nevertheless, for less frequent and abundant prey the variability in Ivlev's index allows bass to be categorized as either selective or with no preference (*P. acarne*; *S. japonicus*) or exclusive avoidance and no preference (*T. trachurus*). This means that in the presence of several fish prey species, bass feed preferentially on the more abundant prey species. The concept of accessibility (Ivlev 1961) or vulnerability (Lewis and Helms 1964) of prey has important implications for understanding resource utilization by predators. Lewis and Helms (1964) proposed that the vulnerability of the

prey was more important than morphological and behavioural characteristics of the predator, while Diggins *et al.* (1979), suggest that both prey density and vulnerability are of importance in predator selection. Ivlev's electivity index denoted positive electivity for *B. boops* and *P. acarne*, no preference for *S. japonicus* and *T. trachurus* and almost a complete avoidance for blennies. The Algarve AR sets are composed of open cubic concrete reef modules of low complexity/heterogeneity that provide relatively few refuges for blennies. Nevertheless, they are not very vulnerable to bass as they take refuge in the concavities (authors' personal observation) provided by the AR macrobenthic community (e.g. dead oyster shells, bryozoans, etc.).

Hixon and Beets (1993) established refuge from predation as an important function of reef fish structure. Bayle-Sempere *et al.* (2001) showed that the small number of recruits in large volume ARs might be associated to predation by larger fishes, since predators have a greater ability to capture prey in low structural complexity artificial habitats. Indeed, juveniles of the demersal species such as *B. boops* and *P. acarne*, that are highly vulnerable to sea bass predation, do not use ARs to avoid predation. However, it is worth noting that for hypothesis II and III significant results were observed only for the total prey community and/or *B. boops*. This suggests that while bass respond to overall prey density, they select (Hypothesis IV) the most available and vulnerable species.

The Faro/Ancão AR system covers an area of 12.21km², at a depth range of 17 to 24m, in a 36km² zone where natural reefs are scarce (only 2.7km²). This AR system located off Faro and the Ria Formosa lagoon was designed to provide suitable hard structure habitat for juveniles that seasonally migrate from the lagoon to coastal waters as reported by Monteiro *et al.* (1990). Thus, in addition to the high capacity of man made structures to attract/aggregate fish, the Faro/Ancão reef fish assemblages may be

strongly influenced by the input of large numbers of fish migrating from the Ria Formosa to the AR, favouring the local increase of prey. This may contribute to creating optimal foraging areas for bass. High levels of bass predation on juveniles at these sites may affect the transfer of stock from the lagoon to the adjacent coastal waters. Several authors report that predation is affected by the abundance of available prey (Hixon and Carr 1997; Stewart and Jones 2001; Connell 2002). Therefore, predation rate and consequently effects on reef species may be higher on ARs that are associated with annual recruitment of juveniles from nearby nurseries. However, this is not the case of the juvenile fish species found in the sea bass stomachs, which are very common in Algarve coastal waters but not particularly abundant in the Ria Formosa lagoon.

At present there is not enough data to prove that sea bass predation rates are higher in ARs located near nursery grounds or whether predation is similar throughout the Algarve AR complex, which occupies an area of 43km² within a coastline of approximately 110km. However, as both predator and prey species are very common on the southern coast of the Algarve, it is expected that this *D. labrax*-prey interaction occurs along all the local ARs. Density-dependent predation can occur on patchy habitats because predators disproportionately forage in patches of high relative to low prey density (Hixon and Carr 1997; Stewart and Jones 2001). Spatially density-dependent predation may be common in some reef settings, particularly on isolated patches (such as those off the Algarve), and less common or important on continuous reefs (Sandim and Pacala 2005). Overholtzer-McLeod (2006) showed that mortality was density dependent on coral reef patches that were spatially isolated (separated by 50m) and density independent on reef patches that were aggregated (separated by 5m). The sea bass, a transient and opportunistic open-water species, can easily pursue their prey

throughout the AR structures. The Algarve AR organization was designed to act as a “chain net” so that fish can move from one reef set to the other (~70m) and/or between reef groups (~300m). The presence of *D. labrax* in the ARs has been particularly noted in the last three years, a period which coincided with the enlargement of the Faro/Ancão AR system during 2002 (Leitão *et al.* 2008). Because of the durability of AR material (concrete), we can predict a continuous long-term predation effect of sea bass on reef fish assemblages. Hueckel and Buckley (1987) found that as an AR increases in age, food resources and predator populations associated with the reef also increase. Moreover, as greater numbers of ARs are deployed, the probability of prey–predator encounters increases and consequently, fish prey become more vulnerable to predation.

D. labrax predation has contributed to the decrease of the abundance of prey demersal species on the ARs. Whether this decrease results in lower catches or recruitment of prey species to the fishery, either locally or in an adjacent region, depends on stock dynamics. If the prey species are migratory, which is not the case here, then heavy predation mortality in one region will probably result in lower levels of recruitment/abundance (exploitable biomass) in adjacent areas. By attracting and aggregating juvenile fish, ARs create feeding areas for predators. Therefore, heavy AR mortality due to predation occurring along Algarve ARs, could reduce recruitment of prey species to the local small-scale fisheries. Caley (1993) showed that predators may affect community structure of older age classes through time-lagged effects on the survivorship of younger age classes and that a greater number of species of recruit and resident fishes were more abundant on reefs from which predators had been removed. On the other hand, while the juvenile prey species are too small to be caught by hook or net gear, they provide food for the sea bass, an economically very important species for inshore artisanal and recreational fisheries. Increases in catches of predators, such as the

sea bass, in the vicinity of ARs is a consequence of their attraction to ARs due to the feeding opportunities provided by higher prey concentrations. However, due to their economic value, such a “bait” effect, may lead to an increase in vulnerability of predators to local fisheries, as suggested by Polovina (1991).

The present study suggests that it is necessary to consider ecological effects such as predator-prey interactions, prior to the development of artificial habitats as a tool for habitat rehabilitation. The effects of predator- prey interactions, particularly in the vicinity of artificial bottom habitats, on fish resources are poorly understood, complex and require in-depth study. Both short and long-term effects of sea bass predation on the Algarve AR reef fish assemblage are difficult to monitor due to the constant evolution of these ecosystems. If fish attraction from surrounding areas is rapid, then long-term, cumulative and indirect effects due to fishing and predation (or both), become difficult to infer.

Spatial associations (or lack thereof) between predator and prey density will, however, be species and context specific. In some situations, the density of predatory fish does covary with prey density (Stewart and Jones 2001), and an aggregative response is the putative cause of density-dependent mortality in other reef fishes (Hixon and Carr 1997; Anderson 2001). Our ability to assess the nature and effects of density-dependent interactions on population dynamics should thus be improved by work that identifies the underlying biological interactions causing density-dependent mortality, and characterizes the spatial and temporal domains at which those interactions operate (Forrester and Steele 2004; Overholtzer-McLeod 2006).

The results obtained in this study highlight the importance of defining a suitable strategy to manage these artificial habitats. Therefore, understanding inter-specific interactions (e.g. predator-prey) is important for conservation and management and for

evaluating the long-term effects of reef deployment, especially in areas where nearby lagoons supply juvenile fish to the adjacent coastal waters.

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CHAPTER 5

***Diplodus* spp. assemblages on artificial reefs: importance for near shore fisheries**

Leitão F, Santos MN, Erzini K, Monteiro CC. *Diplodus* spp. assemblages on artificial reefs: importance for near shore fisheries. Fisheries Management and Ecology 16: 88-99.

ABSTRACT

Artificial reefs (AR) have been deployed along the southern coast of Portugal (Algarve) since 1990 to enhance local artisanal fisheries. The objectives of the study were to: i) describe the colonisation process; ii) assess the role of the AR in terms of juvenile recruitment and growth and as mating/spawning areas and iii) evaluate the AR potential as near shore artisanal fishing grounds for three economically important fish species, *Diplodus bellottii* (Steindachner, 1882), *D. sargus* (Linnaeus 1758) and *D. vulgaris* (Geoffroy Saint-Hilaire, 1817). The fish assemblages were monitored monthly by visual census for two years after the deployment of a large AR (Faro/Ancão) in 2002. Colonisation rates for the three species were fast. The Faro/Ancão artificial habitats play a multiple role for *Diplodus* spp., acting as recruitment, growth and nursery areas for juveniles, and spawning/mating areas for adults, and can thus be considered essential fish habitat. Three months after deployment of the ARs, exploitable biomass was 16, 29 and 8 kg per reef group, respectively, for *D. bellotti*, *D. sargus* and *D. vulgaris*. These results indicate that ARs quickly become good fishing grounds, where suitable financial yields may be obtained by local artisanal fisherman. Moreover, the ARs became new and alternative fishing grounds, allowing reduction of fishing effort over traditional rocky areas that are scarce along the Algarve coast. Management measures for these artificial habitats, in terms of fishing strategies, are discussed.

INTRODUCTION

The deployment of artificial reefs in European waters has increased considerably over the last 30 years (Jensen 2002 and references therein). Fisheries enhancement, increasing diversity and protection/mitigation of marine habitats are among the different arguments used to justify the creation of these artificial habitats (Jensen *et al.* 2000). In the southern coast of Portugal (Algarve), multi-purpose ARs have been deployed since 1990. Their deployment aims to enhance the amount of hard bottom habitat, which is scarce on the sandy southern grounds, to provide suitable habitat for fish and to increase fishing yields (Monteiro and Santos 2000). The idea behind the deployment of AR for fishery enhancement relies on the assumption that habitat is a limiting factor and that ARs will provide critical habitat to promote the production of new biomass through increased growth and survival of juveniles (Bohnsack 1989). Nevertheless, if ARs act like attraction devices, increasing fish density, then higher density may increase catchability, and the greater accessibility increases fishing effort, potentially resulting in greater fishing mortality. An increase in fishing mortality will decrease exploitable biomass in the area Polovina (1991). Whether this decrease results in lower catches, either locally or in an adjacent region depends on stock dynamics and exploitation levels. Therefore, understanding ARs functioning is essential for the management of these areas. Presently, the Algarve (South Portugal) artificial reefs cover an area of 43.5 km², representing the largest area of artificial habitat of its kind in European waters.

Knowledge of the size structure of AR fish assemblages is essential as it may provide a useful tool for determining management measures in AR areas, namely for commercial species. Moreover, determining AR long-term fishing potential requires the knowledge of reef fish species size structure. *In situ* visual methods have been widely used to characterize the structure of AR fish communities (Bortone and Kimmel 1991),

as they provide accurate information regarding the AR fish assemblages. These include species abundances, size-structure, recruitment patterns and reproductive behaviour. Size information allows the evaluation of recruitment patterns and indirectly, through weight-length relationships, the estimation of exploitable available biomass. On the other hand, it is essential to know how fast these man-made structures become profitable fishing grounds and to understand their biological/ecological role for fish assemblages inhabiting the ARs, from early on after deployment, aiming towards fisheries management. In this study the size structures of three economically valuable reef fish species, *Diplodus bellottii* (Steindachner 1882), *D. sargus* (Linnaeus 1758) and *D. vulgaris* (Geoffroy Saint-Hilaire 1817), were monitored on the Faro/Ancão ARs during the first two years after deployment in 2002. Both *D. sargus* and *D. vulgaris* contribute greatly to the total fish abundance (in number) in southern European rocky infralittoral zones, representing up to 56% in the NW Mediterranean (Garcia-Rubies 1997). In the Faro/Ancão ARs, these three species account for 40 and 63% of overall resident reef fish assemblages and 20% and 20% of the overall reef fish assemblage, in terms of abundance and biomass, respectively (Leitão *et al.* 2008). The role of the artificial reefs as areas for recruitment, growth and spawning and/or mating was evaluated, and the biomass of each species potentially available to the local artisanal fisheries estimated. Finally, the data were used to propose appropriate fishing strategies and management plans for the AR.

METHODS

Study site

The Faro/Ancão AR system consists of 52 reef groups (RG) deployed off Faro beach on a sandy ground at depths between 20 and 22 m, covering an area of 12.2 km² (Figure 1).

Each RG is composed of three reef sets (RS) occupying a volume of approximately 529 m³ and consists of 35 cubic modules organised in a two layer structure. The reef units were 1.25 m³ of concrete with the cubic block having an external volume of 2.7 m³ (1.4 m length x 1.4 m width x 1.4 m height). The distance between RSs is 70 m and between RGs is 400 m. The ARs run parallel to the coast line in a NW and SE direction along the 20 m isobath and were set in 2002 on either side of 12 year-old ARs (Figure 1). The new reefs are identical to the old reefs in terms of organisation and module type.

Data collection

The study was carried out between October 2002 and September 2004. The same three randomly selected (within the 52) artificial reef groups (RGs), were monitored monthly, except for February 2004 (month 17) because of rough sea conditions, using scuba diving. Fish distribution was determined by visual census using the stationary point method (for details see Santos *et al.* 2005). At each reef set fish counts (numbers and sizes) were made at three randomly selected points (6-minute interval) in a radius of 3 to 6m, depending on water turbidity during the study period. To reduce the bias caused by daily fish density variation, all counts were carried out from 10.00 to 12.00 am, at similar tidal conditions. *D. bellotti* and *D. vulgaris* were generally assigned to 3 cm size classes (or 5 cm whenever > 20 cm), while *D. sargus* was assigned to 5 cm size classes. Size class calibration/correction was done by spearing specimens of each species and comparing actual and estimated total lengths. Density was calculated as the number of fish per reef set (volume of 529 m³ of water). The calculation of biomass was made using: (1) the estimated midpoint of fish total length class size, (2) length-weight relationships (Santos *et al.* 2002) and (3) the estimated density of fish.

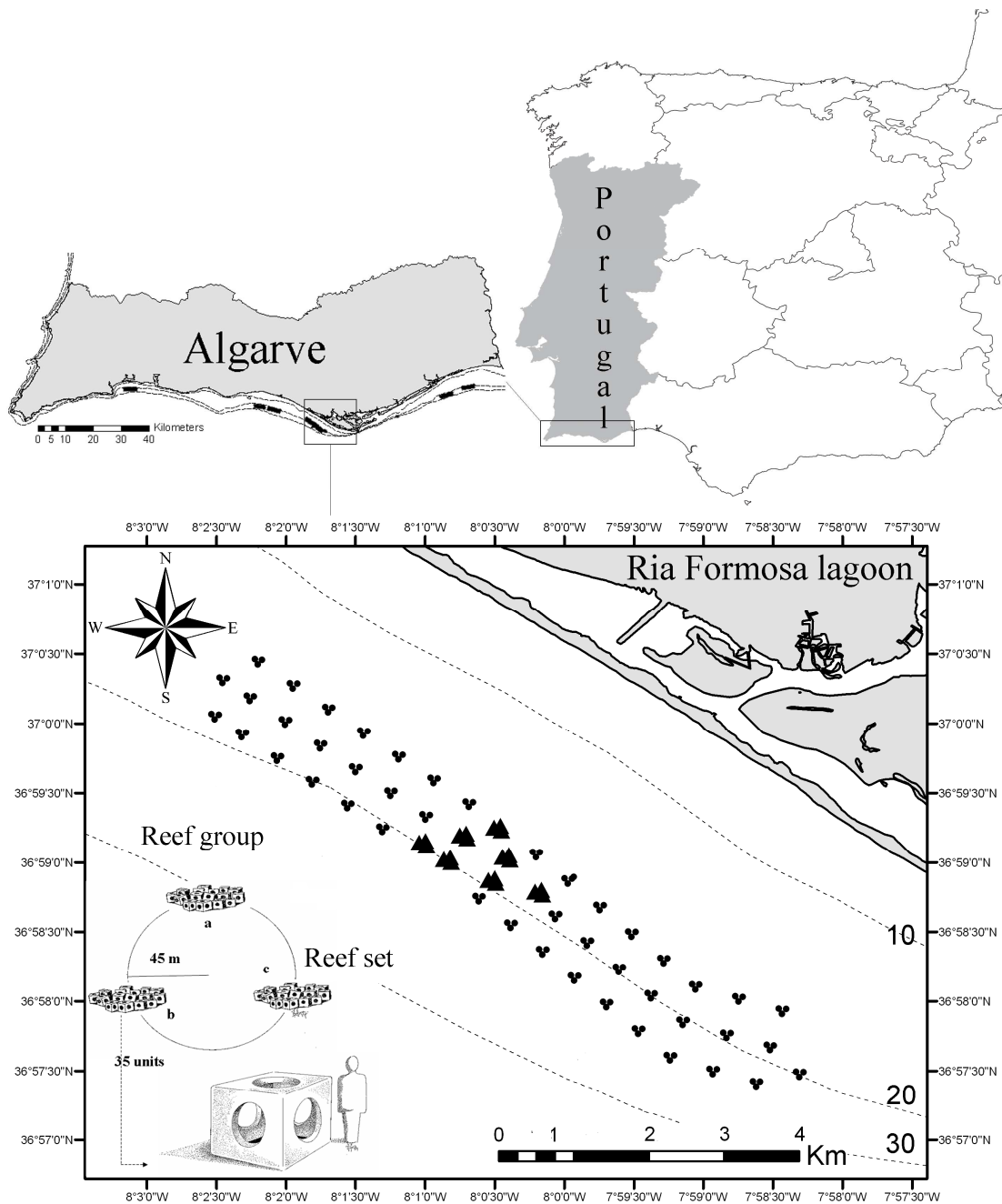


Figure 1. Map of the study area showing reef organization and the location of the AR in relation to the Ria Formosa lagoon and the 10, 20 and 30m isobaths. Triangles for old AR sets and circles for new AR sets.

The role of ARs in terms of mating and/or spawning areas for *D. sargus* was also studied. After finishing the underwater counts and measurements, a number of specimens were caught at the AR by spear fishing between October 2003 and

September 2004. Speared fish were measured to the nearest millimetre below at the laboratory. Each *D. sargus* individual was sexed and maturity stages were determined according to the Buxton and Garratt (1990) macroscopic scale. The percentage of mature fish was calculated considering mature fish those in pre-spawning, spawning and post-spawning stages.

Data analysis

Species colonisation, evolution and stabilisation were studied using mean monthly RS density and biomass. For each species, annual and seasonal (intra-annual) variations were analysed separately for juveniles, adults and the total of all individuals. Temporal shifts (Time effect) in species density, biomass and length were analysed, based on quarterly data (Winter: January-March; Spring: April-June; Summer: July-September; Autumn: October-December), with seasonal samples as the time unit. Seasonal comparisons were only performed after one species was observed on the AR for at least two consecutive months.

The mean numbers of juveniles and adults were calculated taking into consideration the size at first maturity of the species (Santos *et al.* 1998; Gonçalves and Erzini 2000; Erzini *et al.* 2002). *Diplodus bellotti*, *D. sargus* and *D. vulgaris* were considered juveniles when they belonged to length classes equal to or below 12, 15 and 18-cm, respectively. Mean monthly densities and associated standard deviations (\pm SD) were calculated using the RS means. Seasonal values were calculated using monthly RSs density/biomass values. Mean lengths and associated standard deviations (\pm SD) were calculated from the observed numbers per size class.

As monthly sampling was always conducted in the same 3 RGs (sampled sites), data may be RG correlated over time (Zar 1996). Therefore, repeated measures two-

way-ANOVA (site and time) was used, based on RG monthly means ($\alpha= 0.05$) (Zar 1996). Mean RG data were obtained by averaging RS data. Whenever normality (Kolmogorov-Smirnov test) and/or heterogeneity (Bartlett test) assumptions failed, data were transformed by $\ln(n+1)$ to produce normality and homogeneity of variance (Zar 1996). In situations where the null hypothesis was rejected, the Tukey multiple comparison test was performed. Analysis of the results of repeated measures ANOVA did not show a statistically significant difference ($P>0.05$) to RGs and RGs x year/season/time interaction for density biomass and length. Therefore, results of the statistical analysis were only given for season, year and time variables.

The importance of ARs for recruitment and growth was studied by following the progression of monthly length structure data using multivariate hierarchical cluster analyses. Cluster analysis applied the unweighted pair group average algorithm to the Bray-Curtis similarity matrix based on monthly reef group data, after standardisation and square root transformation. Cluster and non-Metric Multidimensional Scaling (MDS) analysis was also used to investigate seasonal size structure. The size classes that most contributed to dissimilarity between seasons were super-imposed in the MDS. The multivariate analysis was carried out using the Primer 5.1 software package (Clarke and Warwick 2001).

The evaluation of AR potential in terms of suitable exploitation by artisanal near shore fisheries was assessed by estimating the monthly available adult (exploitable) biomass per reef set.

RESULTS

Diplodus bellottii

Diplodus bellottii was recorded immediately after AR deployment, but was absent during the next three months (Figure 2). At the 5th month a surprisingly strong recruitment (160 reef set⁻¹) was observed (Figure 2). The minimum number of individuals was recorded in the 1st month (22 reef set⁻¹) and the maximum (243 reef set⁻¹) 16 months after AR deployment. The mean number of *D. bellottii* was 104±69 reef set⁻¹ (Figure 2). The mean number of adults (> 12 cm = 78%), was greater than the mean number of juveniles. No differences for adults (Year: P=0.10, Season^{1st year}: P=0.41, Season^{2nd year}: P=0.46, Time: P=0.54), nor for the overall densities (Year: P=0.54, Season^{1st year}: P=0.62, Season^{2nd year}: P=0.68, Time: P=0.76) were observed during the study period (Figure 3).

The length-frequency analysis indicated that the youngest individuals were mainly sub-adults belonging to the 12-cm length class (Figure 2). A lack of juveniles but the constant presence of the 15-cm length class was noted throughout the study (Figure 2 and 3). Adults were always present but showed no clear trend in terms of density throughout the different seasons (Figure 3). Clusters 1 and 2 grouped the months when the species assemblage was mainly composed of adults (Figure. 4). The latter clusters included mostly spring and late summer months. In spring, the fish assemblages were almost exclusively represented by the 15-cm length class (Figure 2). Cluster 3 and 4 included the months corresponding to the recruitment of sub-adults, mainly between late autumn/winter and summer 2004 (Figure 2). The recruitment of sub-adults was also meaningful in winter (Figure 2). Cluster analyses revealed a high intra-annual seasonal similarity (Figure 4). Moreover, the seasonal size structure was very similar in all seasons, with the exception of spring due to the lack of sub-adults.

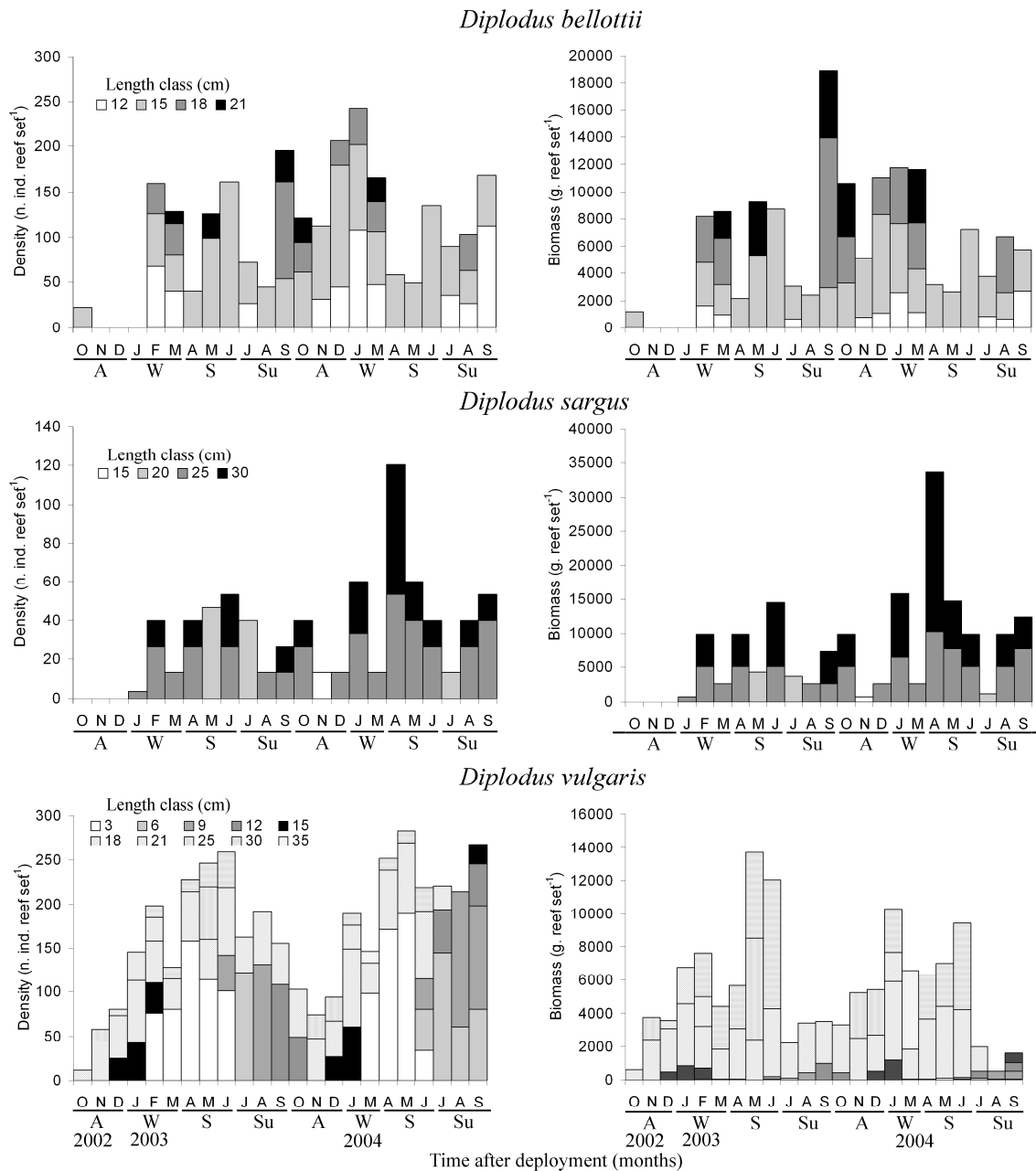


Figure 2. Monthly length-class density and biomass assemblage variation (per reef set) for *D. bellottii*, *D. sargus* and *D. vulgaris*. A– Autumn; W-Winter; S-Spring; Su-Summer.

The mean total biomass was 5926 ± 4474 g reef set⁻¹, of which the majority (90%) was adults, mostly belonging to the 15-cm length class. The mean adult biomass values ranged between 1207 and 18839 g, with a mean value of 5333 ± 4026 g reef set⁻¹ (Figure

2). Mean biomass values for adults (Year: $P=0.45$, Season^{1st year}: $P=0.89$, Season^{2nd year}: $P=0.08$, Time: $P=0.30$) and for the overall *D. bellottii* assemblage did not differ significantly (Year: $P=0.28$, Season^{1st year}: $P=0.90$, Season^{2nd year}: $P=0.10$, Time: $P=0.20$) during the study period (Figure 3). The mean length of the species (15 ± 1.1 cm) also did not vary significantly (Year: $P=0.34$, Season^{1st year}: $P=0.50$, Season^{2nd year}: $P=0.58$, Time: $P=0.81$) during the study period (Figure 5).

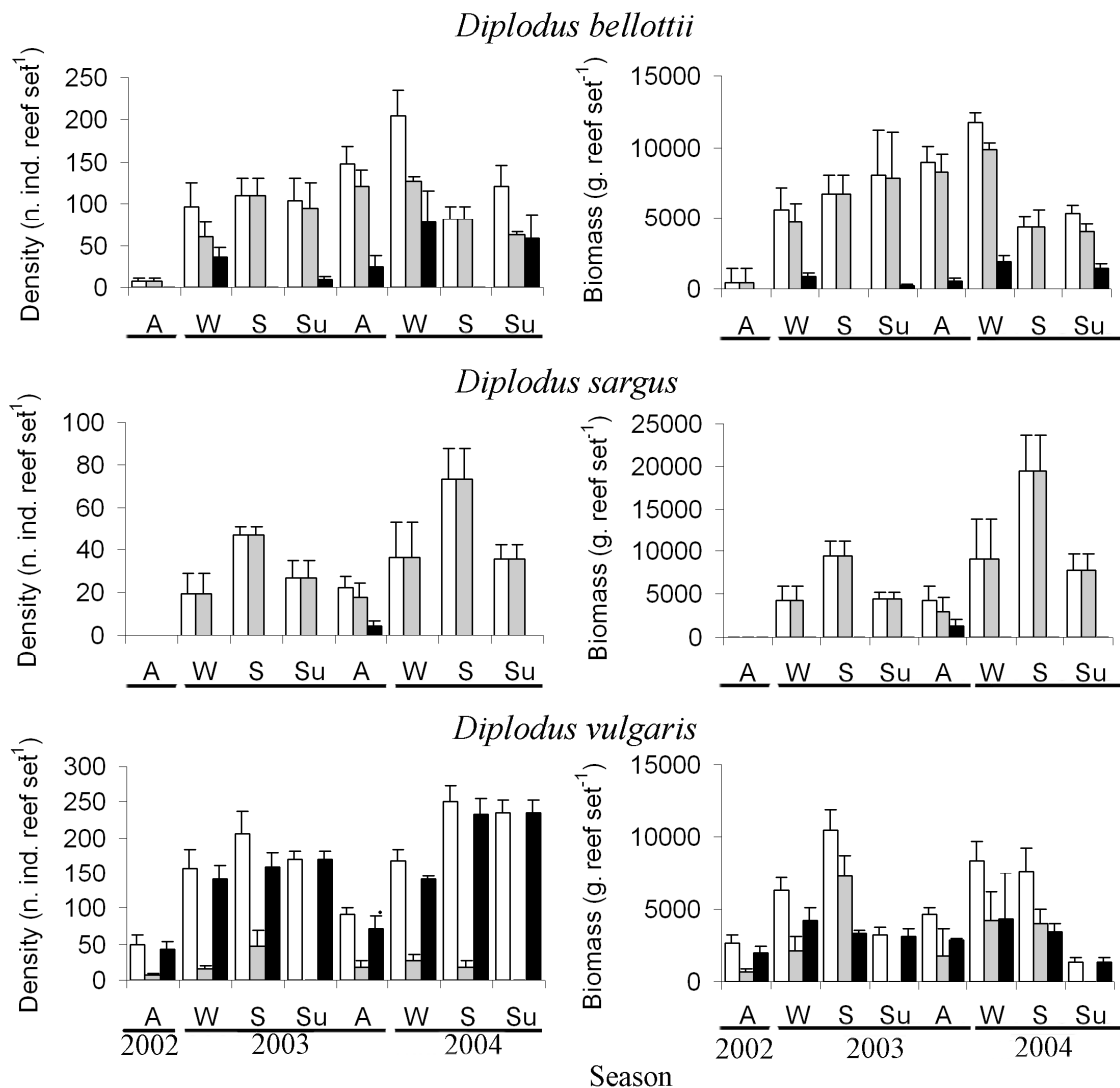


Figure 3. Seasonal juvenile, adult and total density ($n.^{\circ}$ ind. reef set⁻¹) and biomass (g reef set⁻¹) of *Diplodus bellottii*, *D. sargus* and *D. vulgaris*. White bars for total, grey bars for adults and black bars for juveniles mean density and biomass. Lines refer to standard deviation. A– Autumn; W–Winter; S–Spring; Su–Summer

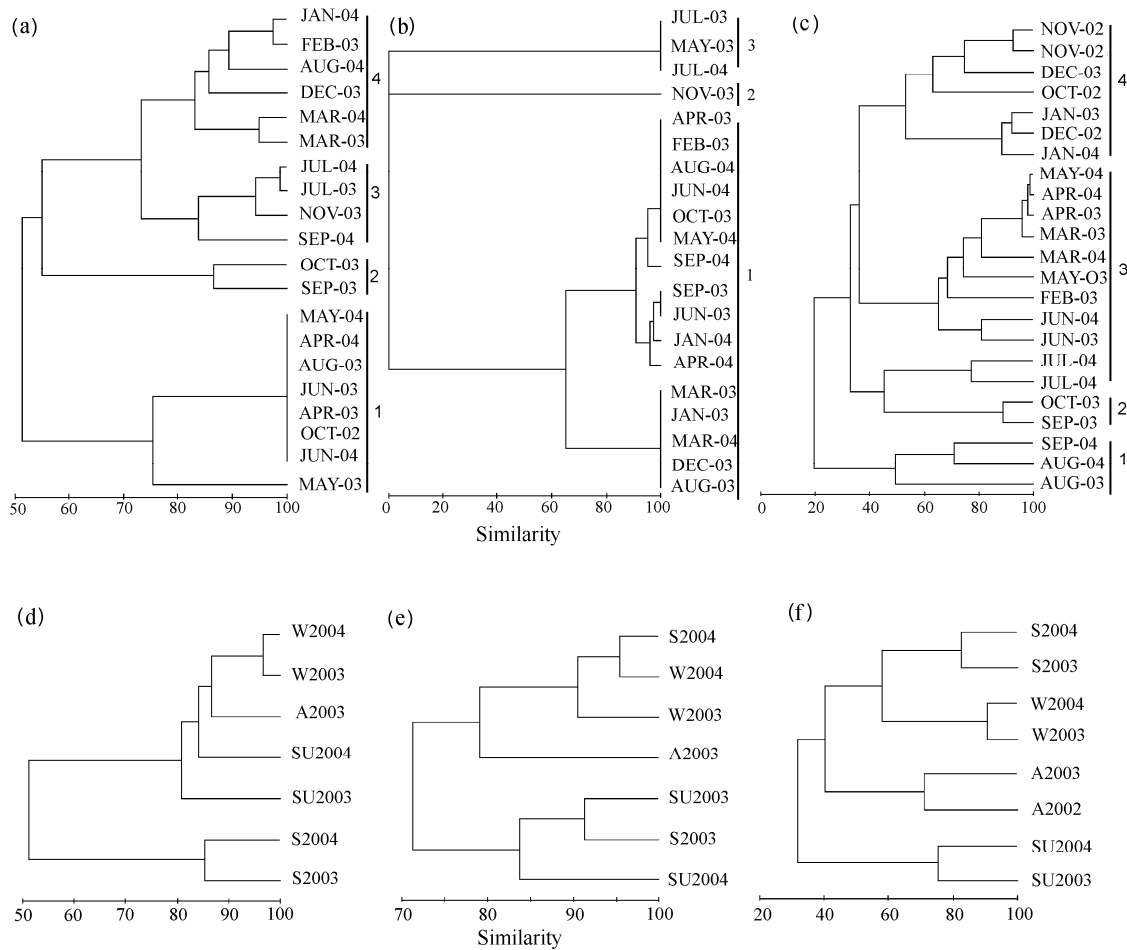


Figure 4. Length-class monthly and seasonal evolution (Cluster analysis) for *Diplodus bellottii* (A, D), *D. sargus* (B, E) and *D. vulgaris* (C, F). A– Autumn; W-Winter; S-Spring; Su-Summer.

Diplodus sargus

Recruitment of the species occurred in the 4th month but density was very low (Figure 2), increasing markedly thereafter. The highest density, 120 reef set⁻¹ was observed in April 2004. *Diplodus sargus* recorded on the reefs were mostly adults (98%) (Figure 2 and 3).

The mean total number of *D. sargus* was 38 ± 29 per reef set. No statistically significant differences in *D. sargus* density (Year: $P=0.13$, Season^{1st year}: $P=0.15$, Season^{2nd year}: $P=0.28$, Time: $P=0.38$) were observed during the study period (Figure 3).

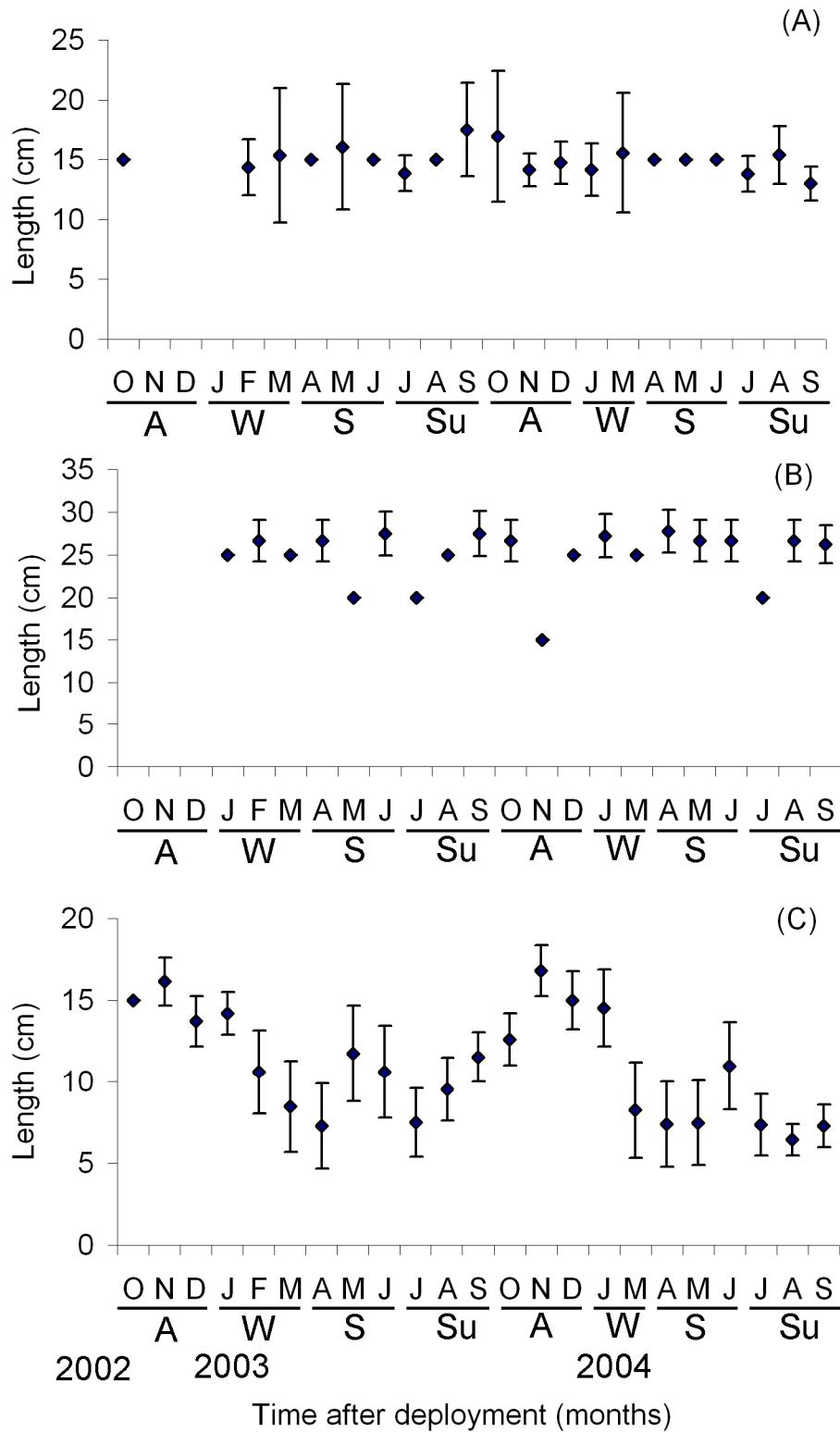


Figure 5. Monthly mean lengths (\pm standard deviation) for *Diplodus bellotti* (A), *D. sargus* (B) and *D. vulgaris* (C). A– Autumn; W-Winter; S-Spring; Su-Summer.

Cluster analysis showed that the *D. sargus* size structure was similar along almost the entire period of the study (Figure 4). The exception was November 2003 (autumn, cluster 2), when juveniles belonging to the 15-cm length classes were observed (Figure 2). Cluster 3 grouped months when solely sub adults (20 cm) were registered.

The mean total monthly biomass (Figure 2) was 9796 ± 10236 g per reef set⁻¹. Adult biomass, ranged between 724 and 33625 g with a mean value of 9723 ± 7074 g reef set⁻¹. The mean biomass (Year: $P=0.08$, Season^{1st year}: $P=0.29$, Season^{2nd year}: $P=0.45$, Time: $P=0.06$) and mean length (24 ± 5.1 cm) of the fish did not vary significantly (Year: $P=0.56$, Season^{1st year}: $P=0.68$, Season^{2nd year}: $P=2.13$, Time: $P=0.82$) during the study period (Figure 3 and 5).

For the analysis of gonads, a total of 104 individuals comprising 50 females and 54 males were caught. The mean lengths of males (26.10 ± 7.52 cm) and females (26.05 ± 6.74 cm) were similar, with an overall mean of 26.45 ± 5.65 cm. The species exhibited a prolonged spawning season, extending from winter to spring (Figure 6). Over 80% of the specimens caught in this period were spawning or presented visible evidence of either pre- or post-spawning. During summer, the reproductive activity decreased (11% for males and 21% for females) and there were no signs of mature gonads in autumn (Figure 6).

Diplodus vulgaris

The mean number of individuals was low in the first three months, but increased markedly between the months 4 and 5 after AR deployment. After this initial period, mean density was always greater than 74 individuals per reef set. The maximum density for the species (283 reef set⁻¹) occurred in month 20 (Figure 2). These values were mainly due to a high density of juveniles (270 reef set⁻¹), namely young of the year

(YOY, 3 cm length class) and sub-adults (18 cm length class) (Figure 2). The mean number of *D. vulgaris* in AR was 166 ± 71 reef set⁻¹.

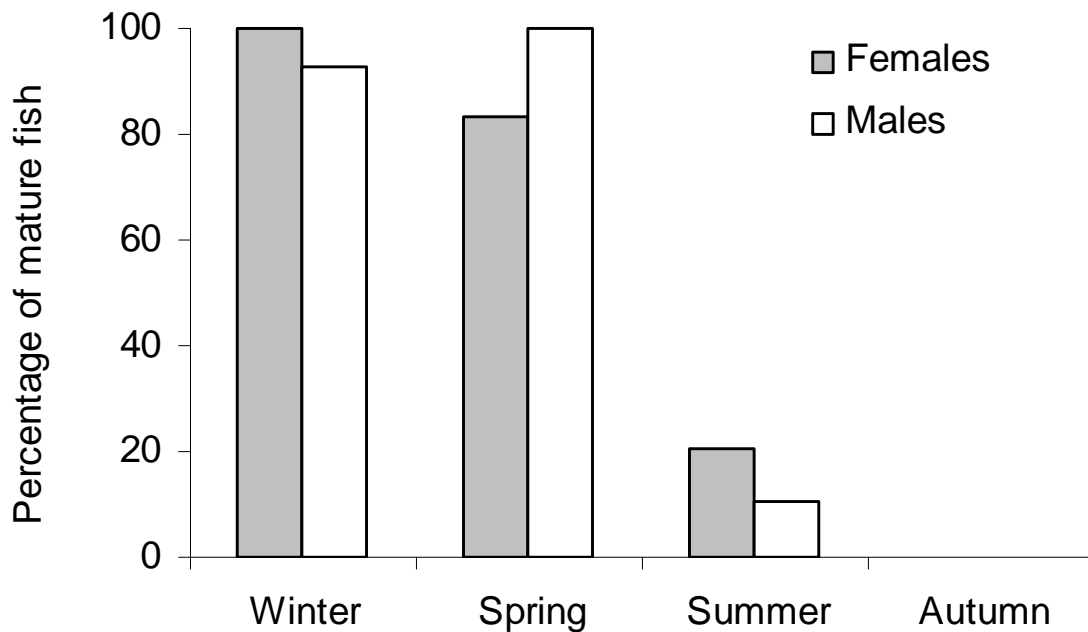


Figure 6. Percentages of mature *Diplodus sargus* females and males caught in the artificial reefs, by season.

Juveniles (≤ 18 cm) accounted for 90% of the *D. vulgaris* observed during the course of the study (Figure 2 and 3). *Diplodus vulgaris* monthly size structure grouped into four clusters (Figure 4). Cluster 1 and 2, grouped months when the size structure was dominated by the 6 to 12-cm length classes (summer months) (Figure 4). Cluster 3, grouped months when the size structure was mostly YOY, in winter and spring. Cluster 4, mainly grouped autumn/early winter months when size structure was mainly composed of juveniles (15 to 18-cm). The *D. vulgaris* size frequency distributions showed a clear seasonal pattern (Figure 4). Seasonal cluster analyses reveal a high intra-annual similarity between seasons. In addition, the low similarities observed between seasons indicate a clear seasonal variation in *D. vulgaris* population size structure. The

seasons when similarity was highest were winter and spring (February to June), coinciding with the recruitment of young of the year (YOY) in both periods. In winter, juvenile recruitment was mainly related to the increased density of YOY after January (Figure 2). In spring, juvenile recruitment was not exclusively associated with 3-cm length class YOY high densities, but also with YOY belonging to the 6 and 9-cm size classes. The most abundant length classes observed in spring, i.e. 3-cm, were replaced by the 6, 9 and 12-cm length classes during summer (Figure 7).

A seasonal variation in species density was verified. Total (Season^{1st year}: $P < 0.01$, Season^{2nd year}: $P = 0.18$) and juvenile (Season^{1st year}: $P = 0.37$, Season^{2nd year}: $P = 0.19$) densities during autumn and winter were lower than in spring and summer. Low numbers of adult were recorded in winter and spring (Figure 3 and 7). Juvenile density was significantly higher in spring 2004 than in spring 2003. Moreover, juvenile ($P < 0.01$) and total ($P < 0.01$) density varied over time, showing low values in autumn.

The highest biomass was found in spring, eight months after AR deployment, with $13735 \text{ g reef set}^{-1}$, mainly due to the presence of adults belonging to the 21 and 25-cm length class (Figure 2). Juvenile biomass ranged between 603 and $5902 \text{ g reef set}^{-1}$ (mean $3102 \pm 11564 \text{ g reef set}^{-1}$). Adult biomass ranged between 1362 and $11322 \text{ g reef set}^{-1}$ (mean $2808 \pm 2565 \text{ g reef set}^{-1}$). In the first year, total biomass ($P = 0.02$) was higher in spring, due to the presence of adults belonging to the 21 and 25-cm length class (Figure 2). In the second year total biomass was lower in summer ($P = 0.01$). Temporal evolution of biomass was associated to species seasonal recruitment episodes being higher in winter and spring compared to summer and autumn ($P < 0.01$).

The observed mean length for *D. vulgaris* was $11 \pm 3.7 \text{ cm}$. The mean size decreased from November to April, increasing after June (Figure 5). The highest lengths were recorded during autumn followed by a decrease between winter and spring due to

recruitment of YOY specimens. However, due to the large standard deviations, no significant differences were recorded in mean lengths during the study period (Year: $P=0.31$, Season^{1st year}: $P=0.37$, Season^{2nd year}: $P=0.09$, Time: $P=0.06$).

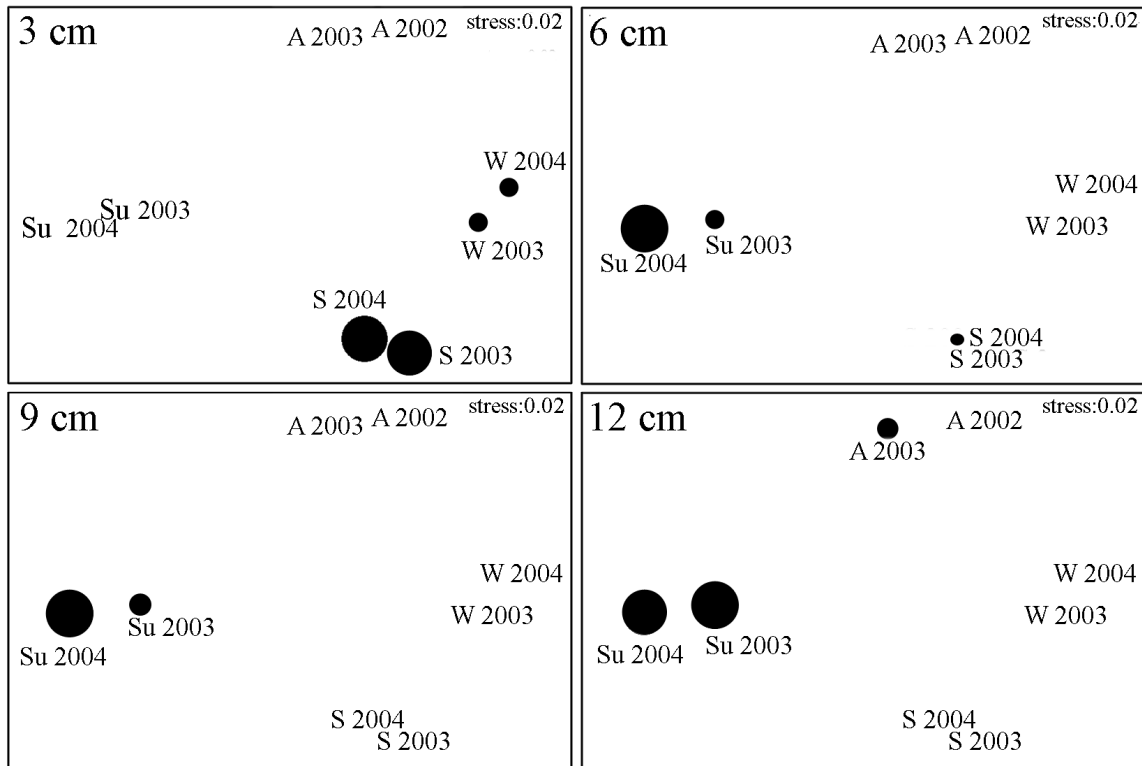


Figure 7. MDS showing seasonal evolution of young-of-the-year length-classes of *Diplodus vulgaris* (3, 6, 9 and 12-cm). A– Autumn; W-Winter; S-Spring; Su-Summer.

DISCUSSION

Diplodus vulgaris was the first of the three species to colonize the AR. A similar pattern of early colonisation by the latter species was also reported by Ramos-Esplá *et al.* (2000) for the western Mediterranean Sea. In the present study the fast colonization rate by this species is related to the vicinity of older ARs. The probability that an individual fish locates a reef is expected to increase with habitat connectivity (Fernández *et al.* 2008 and references therein), and with the increase of AR reef sets, which affects the

rate of fish relocation among habitat patches. However, the hypothesis that older ARs may contribute with fish to the new reefs does not seem to be true for *D. bellotti* and *D. sargus* as these species were not observed in ARs during the first three months (autumn 2002). Moreover, rapid colonisation could also be possibly linked with the migration of juveniles of *D. vulgaris* and sub-adults of *D. bellottii* in late summer and early autumn from the Ria Formosa, an important nursery habitat, to the adjacent coastal waters (Monteiro *et al.* 1990).

Habitat limitation is the primary factor determining the specific composition of AR fish assemblages through availability of food or shelter (Bohnsack 1989). Shelter is generally considered more important (Sale 1980), especially because food availability does not directly influence settlement or survivorship of early larvae (Shulman 1984). A study on the colonisation of these ARs by macrobenthic organisms (Boaventura *et al.* 2006) showed a marked increase in cover (~50%) by month 3 (mostly species belonging to Cirripedia, Serpulidae, Bryozoa and Ascidiacea groups), with the AR surface totally colonised by month 6. *Diplodus sargus* and *D. bellottii*, colonisation matches with the time when macrofauna colonisation increased most rapidly. In fact, the macrofauna *taxa* found in ARs substrate shortly after deployment were reported in the diet of *Diplodus* spp. in AR areas (Pepe *et al.* 1998; Badalamenti 2000; Leitão *et al.* 2007).

Diplodus bellottii and *D. sargus* observed in the reefs were mainly adult individuals and monthly length frequency distributions were similar over the course of the two years of study. Therefore, these AR-associated species present a high potential in terms of adult stock to local fisheries. The results presented on the maturity of *D. sargus*, together with those reported by Santos *et al.* (1998) for *D. bellottii*, suggest that ARs play an important role as mating and/or spawning areas for these species. Thus, the Faro/Ancão ARs extending for 12.2 km² are particularly important in a 36 km² zone

where natural reefs are particularly scarce (2.7 km²). In contrast to both *D. bellottii* and *D. sargus*, *D. vulgaris* schools were mainly of juveniles, which highlight the importance of the ARs as recruitment/nursery areas for the species. In the present study, after month 4, few changes in species density were observed for any of the three species. However, a clear seasonal change in length class structure was noted in the case of *D. vulgaris*. Based on age and growth data (Gonçalves *et al.* 2003), *D. vulgaris* 3- to 9-cm and 12 and 15-cm length classes correspond to juvenile fishes of age classes 0 and 1, respectively. Seasonal recruitment of schools of 3-cm and 6-cm YOY (0) was observed in winter and spring. As the fish grow in the summer, the latter classes were replaced by the 6 and 9 (0) and 12- and 15- and 18-cm length classes. The seasonal development of *D. vulgaris* cohorts highlights the importance of ARs for species growth during their juvenile stage.

Results discussed above show that artificial reefs play a dual role as far as the fishery *Diplodus* spp. is concerned. For *D. bellottii* and *D. sargus* the ARs act as fishery recruitment areas by constituting new fishing grounds. In addition, ARs may be considered an extension of natural mating/spawning grounds. However, for *D. vulgaris*, the ARs represent an inshore extension of the coastal lagoon protection effect, because of local scarcity of natural rocky habitats in shallow waters. In fact, in coastal waters of the Algarve ARs offer a large portion of the available hard habitat substrate between 16 to 24 m depth. Moreover, ARs have a high importance for *Diplodus* spp. as feeding areas (Leitão *et al.* 2007; Leitão *et al.* 2008). Therefore, ARs can be considered essential fish habitat [defined as *substrate necessary for fish for spawning, feeding or growth to maturity* (Benaka 1999)]. Additionally, ARs areas can play an important role in fishery enhancement, as recruitment and fish growth influences the sustainable catch that can be taken from a stock (King 1995). Bohnsack (1989) suggested that biomass production

and catches will increase as some function of the amount of AR material deployed. In the latter context, the availability of a large AR area and the life span of the ARs (concrete blocks) suggests that due to their biological and ecological role, these man-made structures can contribute in the long-run to reef fish production and consequently fisheries enhancement. In addition, the Faro/Ancão ARs is a new fishing ground allowing the decrease of the traditional fishing effort over scarce natural reefs.

After the initial stage (3 to 4 months) of reef colonisation, no differences were observed in terms of adult biomass for the three species. This shows that ARs have a high capacity to aggregate fish, becoming viable fishing grounds within a short period of time. Moreover, this denotes a certain temporal stability in *Diplodus* spp. biomass schools and in practice means that AR can provide year-round exploitable biomass. During the study period the ARs area was fished. However, the mean exploitable biomass (adults) available for *D. bellotti*, *D. sargus* and *D. vulgaris* per reef group (three reef sets) during these initial two years after reef deployment was 16, 29 and 8 kg, respectively. Management of the fishing effort is crucial, as the vulnerability of fish aggregated in small patchy AR areas increases fishing catchability and may lead to greater fishing mortality (Polovina 1991). For reef associated species, ARs increase the chances of gear-resident fish encounters compared with open water fisheries. Moreover, the AR organisation was designed to act as a “chain net” so that fish can move from one reef set to the other (~70m) and/or between reef groups (400m). The patchy distribution of ARs sets contributes to fragmentation of the artificial area. Therefore, due to AR organisation fishing gears deployed between reefs are likely to act like barriers to fish, contributing to the increase in probability of gear-fish encounters during fish movements from reef to reef.

Over the years, because of the technical developments in the fishing gears, artisanal fishermen have abandoned gears that require more man power (e.g. bottom longlines) in favour of the gears that are easier to operate and provide higher yields (e.g. gill nets). However, in the Algarve AR areas, a return to longline fishing may be an alternative to nets, as significant exploitable biomass of economically important species can be found. Since the enlargement of the Faro/Ancão AR (from 0.6 to 12.2 km²), the local fishing communities have re-started using longlines. Within AR assemblages, *D. sargus* is one of the most important target species of longlines and fish traps because of the available exploitable biomass and its high economic value (landing price ~10€ per kg). Experimental longline fishing trials in the Algarve showed that the dominant species in terms of weight (29%) was *D. sargus* (Erzini *et al.* 1996). Thus, cumulative fishing effort as well as gear size selectivity may result in decreased recruitment.

To avoid AR overexploitation, additional management measures are required. Management may be particularly important for resident commercial species as these are more vulnerable/available to the fishery. Fishery management must necessarily be based on the knowledge of reef fish assemblages and take into consideration the AR biological potential as recruitment, growth and mating/spawning areas. In Algarve ARs areas intensive near shore multi-species fisheries make use of a wide variety of fishing gears targeting on sparids, such as gill and trammel nets, long-lines and traps. Based on previously studies on gill/trammel net selectivity (Santos *et al.* 1998; Erzini *et al.* 2003; Erzini *et al.* 2006) a 70mm mesh size would be the best alternative for the exploitation of *Diplodus* spp in the ARs. This represents an increase of 10 mm in the minimum mesh size for gill nets. In the case of longlines, hooks ranging in size between 13 and 11 would avoid the capture of juveniles of all three species (Erzini *et al.* 1996; Erzini, *et al.* 1998). Both the mesh and hook sizes would minimize catches of juveniles of other

commercial species, e.g. *Mullus surmuletus* (L); *Spondylionoma cantharus* (L); *Spicara smaris* (L); *Pagellus* spp.; other *Diplodus* spp.; *Serranus cabrilla* (L) that are also common in the ARs. Moreover, for seasonally recruiting species such as *D. vulgaris*, non-selective gears such as purse seine nets should not be allowed to fish in ARs during periods of recruitment of YOY. As suggested by Santos (1997) the AR exploitation strategies must be goal-oriented and adapted to optimise fishing yields, but avoiding disruptions in the natural evolution of AR and associated fish assemblages. Thus, the exploitation strategies must include the use of different gears, thereby diversifying the catches.

ARs have been developed as part of management plans for coastal fisheries of many countries, but in practise several have failed to contribute to fisheries enhancement, as usually no guidelines for AR sustainable exploitation are enforced. Potential uses of ARs have been demonstrated worldwide and include AR as a tool to halt a global decline in many artisanal fisheries. In this context, marine protected areas (MPAs) and ARs are increasingly regarded as interesting management measures, in that they contribute to ecosystem conservation, improve fish stocks and fisheries sustainability, as they can be helpful in zoning coastal areas in order to reduce conflicts between users (Roberts *et al.* 2001; Claudet and Pelletier 2004). Moreover, the identification of ARs as artificial essential fish habitats can be an important step towards the creation of MPAs based on ARs deployment, despite this study was only a first step that did not take into consideration much of the complexity of coastal ecosystems. The role of ARs as tools for the management of fish stocks is particularly important in the case of artisanal fisheries that in shallow, near shore waters are highly dependent on local coastal fishing resources. Nevertheless, the success or failure of ARs will directly depend on AR management and the adoption of appropriate fishing strategies.

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CHAPTER 6

Influence of benthic community structure on artificial reef fish assemblages

Santos MN, Leitão F, Moura A, Cerqueira M, Lino P, Monteiro CC. Influence of benthic community structure on artificial reef fish assemblages. *Marine Biology* (submitted).

ABSTRACT

The role of suitable habitat structure in determining local-scale patterns in fish species structure is a key issue in ecology. To test whether the established benthic communities influence the density of three artificial reefs (ARs) associated species (*Diplodus bellotti*, *D. sargus* and *D. vulgaris*), we compared the benthic composition and biomass availability with the fish assemblages structure among three similar ARs deployed in 1990, 1998 and 2002 off the Algarve (Southern Portugal). We kept the analyses of *Diplodus* spp assemblage structure (size, abundance and biomass) and benthic communities across age separated to avoid effects of any *a priori* assumptions about relationships between fish and biota. Then, we tested if the benthic communities from the different ARs affected the fish densities. *D. sargus* densities and biomass differed across the 3 ARs, being higher in older ARs. For *D. bellotti* and *D. vulgaris* higher densities and biomass were observed only between the oldest and youngest AR. The total available benthic biomass did not differ across habitat age, but the composition of macrofauna differed between older (1990 and 1998) and most recently deployed AR (2002). Higher biomass values were observed in older reefs (1990 and 1998) for turf algae, Crustacea and Gastropoda and for some sessile benthic *taxa* (Sipunculida and Bryozoa). Moreover, density and biomass of *Diplodus* spp. varied between habitats deployed within the same proxy area but did not vary between ARs separated from kilometres. Overall, results indicated that benthic community' structural differences among reefs, rather than ARs age, affected the *Diplodus* spp. fish assemblages at both local and large spatial scales. These results reinforced the our believe that ARs can play an important role on the management of degraded coastal fishing grounds, as a greater amount of effort on conserving entire ecosystems will hopefully be developed in

coming years rather than focus exclusively on maintaining sustainability of fish resources.

INTRODUCTION

The abundances of fish species are influenced by both biogeographic features variation in local-scale processes (Chabanet 1997; Floeter *et al.* 2007) and by processes acting at very large spatial scales, such as larval supply (Caley 1995). Some important environmental factors known to influence the local distribution and abundance of reef fishes are exposure (Godoy *et al.* 2002; Munday 2002), depth (Friedlander and Parrish 1998) and habitat selection (Gaston and Lawton 1990; Packer and Hoff 1999; García-Charton and Pérez-Ruzafa 2001; Hinz *et al.* 2005). These factors may occur synergistically on reef or rocky systems, mediating the availability of specific factors such as shelter, which is thought to influence the impact of predation and competition (Connel 1997; Hixon and Jones 2005), and food resources that may effect exploitative competition for food (Edgar and Shaw 1995; Horta *et al.* 2004).

Habitat availability is usually assumed to play a major role in determining the composition of fish assemblage's abundance (Ross 1986; Chabanet 1997; Holbrook *et al.* 2000; Almada and Faria 2004), which has been found to explain approximately half of the variation in the abundances of coral-dwelling fishes among locations separated by thousands of kilometres (Munday 2002). According to Arias-González *et al.* (2006), habitat is rarely defined explicitly and often mixes geomorphology (e.g. crest, back reef, or spur and groove zone) and biotic cover (e.g. coral percentage cover, turf algae) and substrate irregularity (e.g. heterogeneity, complexity, rugosity). This result means little uniformity in the scales or use of habitat classification, which has led to confusion in

interpretation (Mumby and Harborne 1999), not allowing easy direct comparisons between studies.

A question of fundamental importance for ecologists is whether communities are limited by a lack of food resources, as would inevitably occur if other processes do not intervene (Malthus 1966) or whether recruitment failure, predation, competition for non-food resources or environmental-induced catastrophes occur with such frequency that communities rarely reach their food resource limits. The intrinsic importance of fluxes of energy and of matter means that community processes are particularly strongly linked with the abiotic environment (Begon *et al.* 2006). The study of the relationship between fish and food resources in a community is a fundamental aspect of population ecology. Most investigations on fish habitat in the coastal zone have focused on differences in abundance, biomass or size distribution of fish, with the aim to evaluate the importance of different habitats on the dynamics of the fish populations under study (Packer and Hoff 1999; Lloret and Planes 2003), but only a few studies have attempted to link the food availability of habitats to the distribution of the fish assemblages (Hinz *et al.* 2005; Stål *et al.* 2007). The food availability in a specific habitat is one indication of habitat quality, reflecting habitat requirements (Sala and Ballesteros 1997; Benaka 1999; Packer and Hoff 1999; Stål *et al.* 2007) and will, in combination with the overall distribution pattern for the selected fish species, provide valuable information for the management of sustainable ecosystems and fisheries.

The Algarve artificial habitats have been deployed in sandy bottoms of coastal waters since the 1990's in order to mimic rocky areas that are scarce and aiming at fisheries enhancement. In these artificial reefs, as well as in NW Mediterranean waters, sparid species belonging to the Genus *Diplodus*, are within the most represented fish assemblages in shallow rocky infralittoral habitats. This is the case of *D. sargus*

(Linnaeus 1758), *D. vulgaris* (E.G. Saint-Hilaire 1817) and *D. bellottii* (Steindachner 1882), three commercial and local important species that account for 40% of AR fish assemblage in terms of density (Leitão *et al.* 2008b). These species are thought to be highly generalist omnivorous, feeding upon preys produced over natural and artificial habitats (Pepe *et al.* 1996, 1998; Sala and Ballesteros 1997; Gonçalves and Erzni 1998; Relini *et al.* 2002; Horta *et al.* 2004; Leitão *et al.* 2007). It has been demonstrated that the benthic composition and production, diversity, abundance and/or habitat structural complexity influence rocky fish assemblages (Brown 1984; Packer and Hoff 1999; Edgar and Shaw 1995; Arias-González *et al.* 2006; Stål *et al.* 2007). Thus, food resources available on the man made structures can be the putative cause for determining assemblage's fish abundance. Comparisons of data collected in the same way at replicate sites at different age reefs, and locations, can provide insights regarding how benthic communities can influence patterns of distribution and abundance of fish. Thus, similar habitats with different ages present a good opportunity to study the influence of benthic communities on the fish assemblages. Herein we studied the benthic community and *Diplodus* spp. assemblages across three artificial habitats with different ages. The specific objectives of this study were to: i) assess the reef fish assemblages, in terms of size structure, abundance and biomass; ii) assess the benthic community composition and *taxa* (biomass) availability; iii) evaluate the relationship between the benthic community and the *Diplodus* spp. assemblages.

MATERIAL AND METHODS

Study area and reef organisation

In the Algarve, southern coast of Portugal, a project of AR deployment started in 1990 with two pilot reefs (one of which off Faro). Later, in 1998 and 2002 new ARs were

deployed, corresponding to the Vilamoura and Faro/Ancão ARs (the latter corresponding to an enlargement of the former Faro AR), respectively (Figure 1).

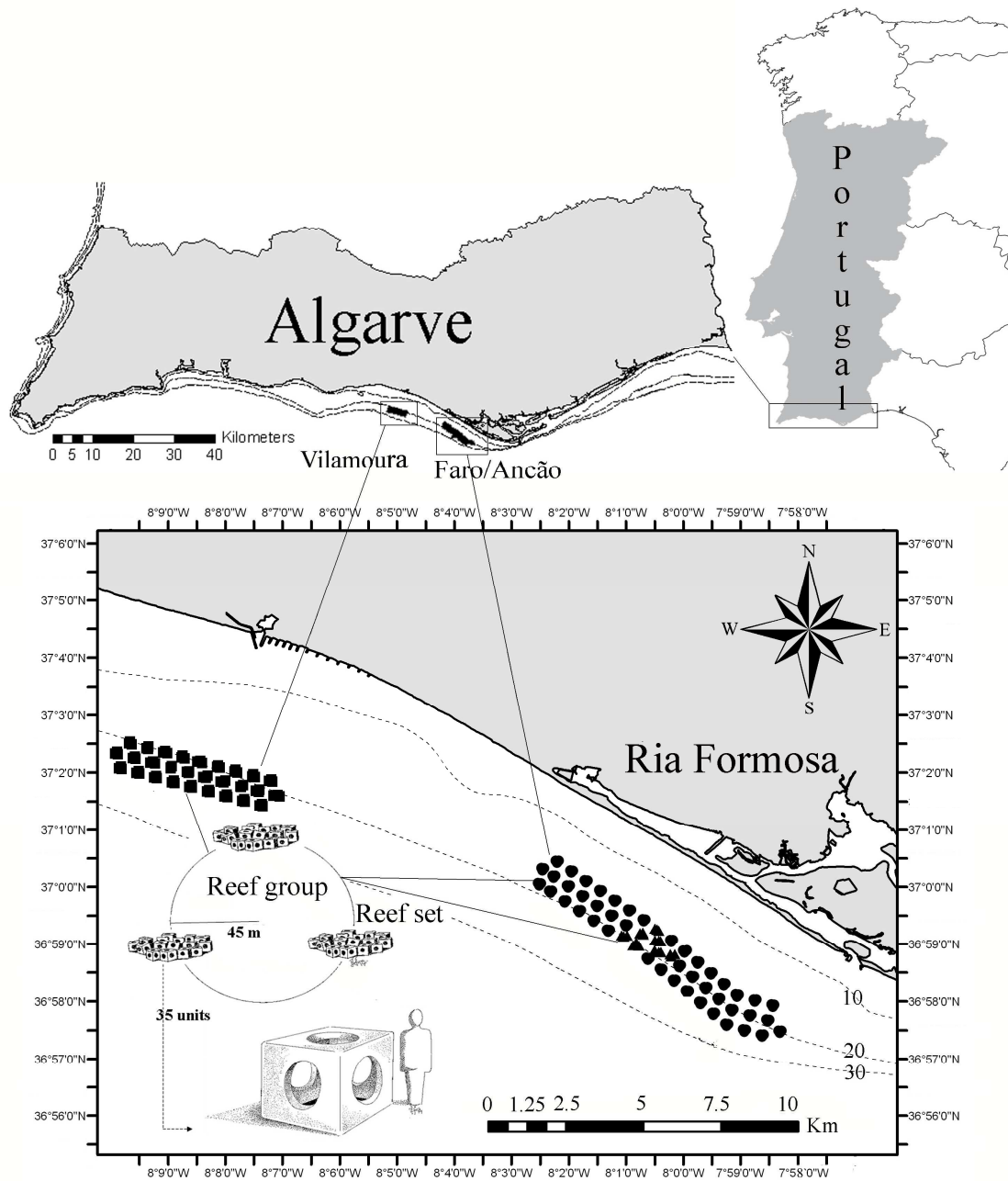


Figure 1. Map of the study area showing the Vilamoura and Faro/Ancão reef organization in relation to the Ria Formosa lagoon and the 10, 20 and 30m isobaths. Triangles, circles and squares for reefs deployed in 1990, 1998 and 2002, respectively.

All ARs made use of concrete blocks and have a similar arrangement, consisting of a number of reef groups (RGs), each consisting of 3 reef sets (RSs). Each RS occupies a volume of approximately 529 m³, consisting of 35 concrete cubic modules organized in a chaotic two layer structure. The reef units used consisted of 1.25m³ of concrete with an external volume of 2.7m³. The distance between RSs is 70m and between RGs is of 300-400 m. The Faro Pilot AR has 7 RGs occupying a total area of 39 ha. The Vilamoura AR with 28 RGs extends for 375 ha, while the Faro/Ancão AR has 52 RGs reef groups extending for 876 ha. The ARs run parallel to the coast line along the 20m isobath. All reefs were deployed during summer, on sandy grounds and isolated from the nearby natural rocky substrate that is scarce in the area. The study area is subjected to similar hydrodynamic conditions with strong events arriving from the SE direction.

Experimental design and Methods

Between June and July 2006 the 3 different age ARs (deployed 16, 8 and 4 years before) were surveyed. For that purpose, for each AR four RGs were randomly selected. Each RG accounts for three replicates, each corresponding to each of the respective RSs. Therefore, overall for each AR a total of twelve (4 RG x 3 RS) fish surveys were carried out. Additionally, three random counts were made at each RS. In order to reduce the bias due to daily fish abundance variation, all counts were carried out from 10 to 12 AM, at similar tide conditions.

Fish distribution was determined by visual census using the stationary point method as described in Santos *et al.* (2005). The observer, always the same diver with years of training, recorded the number and sizes of fishes in a radius of 3 to 6 m (depending on the water turbidity), in a 6-minute interval - the minimum time required to count (for details see Santos *et al.* 2005). The diver recorded the different *Diplodus*

assemblages present within the AR, along with their relative abundance and midpoint size-class value for each species shoal. The number of individuals by species was tallied in 3 cm size classes in the case of *D. bellotti* and *D. vulgaris* (or 5 cm when total length > 20 cm) and 5 cm size classes for *D. sargus*. The determination of fish size near the limits of size classes implied a certain degree of subjectivity, especially when fish were several meters away from the observers. Size class calibration/correction was done by spearing specimens of each species and comparing actual and estimated total lengths.

Benthic samples were collected in June 2006. At each studied AR, three RS were randomly selected among those censused for fish, for benthic macrofauna samples collection. At each RS, three macrofauna benthic samples, each consisting of two pooled quadrates (15 x 15 cm each), were hazardly collected from the surface of the modules. Thus, a total of nine benthic macrofauna samples were collected per reef age. Quadrates samples were obtained directly by scuba-divers who scraped benthic organisms off the reef structure. An airlift pump was used to collect macrofauna, which was retained in a 1mm mesh bag attached over the end of the air lift tube. Once in the laboratory benthic samples were fixed and preserved in 4% buffered formalin. Samples were later washed in running water, sorted and *taxa* were identified under a stereoscopic lens. The wet weight of each *taxa* was obtained after a 5-min drying period on blotting paper with a digital balance with a precision of 0.001 g.

Data analysis

Fish assemblages

In this study the year of AR deployment (1990 for Faro AR, 1998 for Vilamoura AR and 2002 for Faro/Ancão AR) was used throughout the manuscript to discriminate the different habitat ages. The density was calculated as the number of fish per RS (529 m³

of water). The calculation of biomass index was made using: (1) the estimated midpoint of fish total length size class, (2) the length-weight relationship (Santos *et al.* 2002) and (3) the estimated density of fish. Species assemblage biomasses were obtained by multiplying the weight for each size class for each species by the total number of individuals observed in that size class.

The mean numbers of juveniles and adults were calculated taking into consideration the size at first maturity of the species. *Diplodus bellotti*, *D. sargus* and *D. vulgaris* were considered juveniles (Santos *et al.* 2006) whenever belonging to length classes equal to or below 12, 15 and 18cm. Mean density (n.º ind/RS) and biomass (g/RS) for total, juveniles and size classes and standard deviation (\pm SD), according with age, were calculated using mean RS observations, after pooling the data regarding the three counts made at each RS.

Several hypotheses were stated in order to evaluate differences in fish assemblages size structure, density and biomass (dependent variables) across reef age (independent variable), under the assumption that the three species are site fidelity resident species in the ARs (Leitão *et al.* 2008b).

The first approach was to test whether the *Diplodus* spp. assemblage varied between the different habitats ages, in terms of size structure (Hypothesis I). Independently of age, it is expected that assemblage size within each AR varied among RG counts. This RG variability may affect fish assemblage results across habitat age. Therefore Hypothesis I was analysed by two-way (age and RG within age effects) multivariate analysis of similarity (ANOSIM) nesting RG within reef age (Clarke and Warwick 2001). Hypothesis I was stated according to the following hypotheses:

H₀₁: there are no differences in length size distribution among habitats within each age;

H₀₂: there are no differences in length size distribution among habitats with different age;

where, the approach to H₀₂ depends on the outcome of testing H₀₁ (Clarke and Warwick 2001). Size structure data was square root transformed. The SIMPER analysis was used in order to highlight those size classes that most contributed to dissimilarity among reef age. Secondly, we tested if artificial habitats with different age have different *Diplodus* spp. density (Hypothesis II) and biomass (Hypothesis III):

H₀: *Diplodus* spp total density/biomass do not differ across habitat age;

H_A: *Diplodus* spp total densities//biomass differ across habitat age;

The two-way Nested ANOVA was used to compare both density and biomass differences between reefs with different ages. In order to comply with the protocols of the experimental design (random selection of RGs), we nested RGs within age and considered RGs as a random effect (Zar 1996). Both Hypothesis II and III are better stated as described above for Hypothesis I, where the outcome of H₁ influenced the H₂ (Zar 1996). Whenever normality (Kolmogorov-Smirnov test) and/or heterogeneity (Bartlett test) assumptions failed, the data was $\ln(n+1)$ transformed. The ANOVA was carried out for juvenile, adults and total. Nevertheless, given that the final goal was to understand how habitat structure affects reef fish assemblage, the final conclusion regarding both hypothesis II and III were based on the analysis of total assemblage data for each fish species. The non-parametric SNK test was used in order to enable significantly different averages (size class, density and biomass) to be identified (Zar 1996). For census data, statistical significance was attributed to univariate and multivariate tests at the 10% level. While this *a priori* decision allows for greater

probability of falsely concluding that a significant effect exists (i.e. making a Type I error) than the traditional level of 5%, it is important that due regard also be paid to Type II error, i.e. falsely concluding that no differences exists when there really is one (Zar 1996).

Benthic community

Increasingly, studies are separating benthic and geomorphic attributes in the development of habitat classifications (Mumby and Harborne 1999; Áries-González *et al.* 2006). Generally, most studies focusing on benthic habitat communities use the percentage cover method (see introduction section). However, the percentage cover is not suited for the mobile or less conspicuous fauna (Moura *et al.* 2008) that may have an important role in explaining benthic fauna production and fish assemblage's relationships. One common technique is to use biomass, allowing solitary and colonial organisms to be compared on the same scale (Relini and Relini 1997; Moura *et al.* 2008), although it is a laborious method. This was the approach adopted in the present study. Habitats across ARs age were characterised according to the ecological biotic biomass attribute. We used benthic habitat structure term to describe both the composition (presence/absence) of benthic macrofauna and flora and also their availability (biomass) according to each *taxa*. Mean benthic biomass (per m²) and standard deviation (\pm SD) per reef age were calculated using RS samples. Fish assemblage density might be related to fish habitat selection and thus with both food limitations (Malthus 1966; Edgar and Shaw 1995) and fish food selectivity preferences (Pepe *et al.* 1996, 1998; Horta *et al.* 2004; Leitão *et al.* 2007 and references therein). Therefore, first we evaluated if benthic habitat structure differed across age (Hypothesis IV) by means of MDS. The MDS analysis was based on the Bray-Curtis similarity

biomass matrix after square root transformation of the data. Differences in benthic habitat structure across reef age were assessed by multivariate two-way (habitat age and RS samples within age) analysis of similarity (ANOSIM, $\alpha=0.05$), nesting RSs samples within reef age (Clarke and Warwick 2001). SIMPER was used to determine benthic macrofauna *taxa* that most contributed to the differences verified between habitat age (Clarke and Warwick 2001). Secondly, we tested if total and mean *taxa* biomass differed across habitat age (Hypothesis V). Two way nested ANOVA ($\alpha=0.05$) were used to assess statistical differences regarding *taxa* biomass due to habitat age. As benthic samples across reef age were taken randomly, we treated variability among benthic samples as a random factor nested in age (fixed factor) (Zar 1996). Whenever normality (Kolmogorov-Smirnov test) and/or heterogeneity (Bartlett test) assumptions failed, the data were $\log(x+1)$ transformed to achieve parametric assumptions. Whenever the null hypothesis (equality of means) was rejected, the SNK non-parametric test was used in order to enable significantly different averages to be identified.

Fish assemblages and benthos relationships

One of the main problems in planning habitat deployment is predicting equilibrium steady states in benthic and nektonic communities along the reef life cycle. Nevertheless, fish assemblage and benthic communities are expected to be at different levels of colonisation and succession. Herein we assumed that fish assemblages in the artificial marine environment are strongly influenced by population relationships to biotic gradients, such as benthic macrofauna and algae composition and availability. If fish abundance is influenced by hard substrata benthic community composition and

availability across habitat age, then there should be a positive relationship between the total density of fish and benthic habitat structure. Thus, it was tested (Hypothesis VI):

H₀: There is no relationship between fish density and habitat structure across age.

H_A: There is a relationship between fish density and habitat structure across age;

The assumption due to specialised relation of fish with habitat is important as fish mobility may invalidate relationships interpretations. So, our hypothesis were tested under the assumption that species have high site fidelity (Santos *et al.* 2005; Leitão *et al.* 2008b), are highly generalist omnivores (Pepe *et al.* 1996, 1998; Horta *et al.* 2004; Leitão *et al.* 2007) and that they partition the same space and food resources (Sala and Ballesteros 1997; Horta *et al.* 2004). In addition, the choice of the RGs took into consideration the distance between reefs with different ages that were selected far enough to inhibit movement of fish between RGs with different age and thereby serve as independent replicates.

Diplodus spp. density (response variable) and benthos biomass (explanatory variables) relationships were analyzed through Redundancy Analysis (RDA). This ordination method is considered to be a powerful multivariate technique that is useful to extract synthetic environmental gradients from ecological data (Zuur *et al.* 2007). The result of the RDA was plotted on a correlation triplot. In addition, the correlation matrix was also used for the response variables. RS were used as nominal covariables. Some benthic *taxa* were not used in the final model (Echiura; Foraminifera), as previous data analysis showed that these *taxa* reduce the model inertia - the amount of variation that can be explained by the explanatory variables of the model (Zuur *et al.* 2007).

RESULTS

Diplodus bellottii

D. bellottii ranged in size between the 9 and 21-cm size classes, being mainly composed of juveniles that accounted for 67, 64 and 76% of the total density for the reefs deployed in 1990, 1998 and 2002, respectively. The 12cm size class recorded the highest number of fish, independently of habitat age (Figure 2).

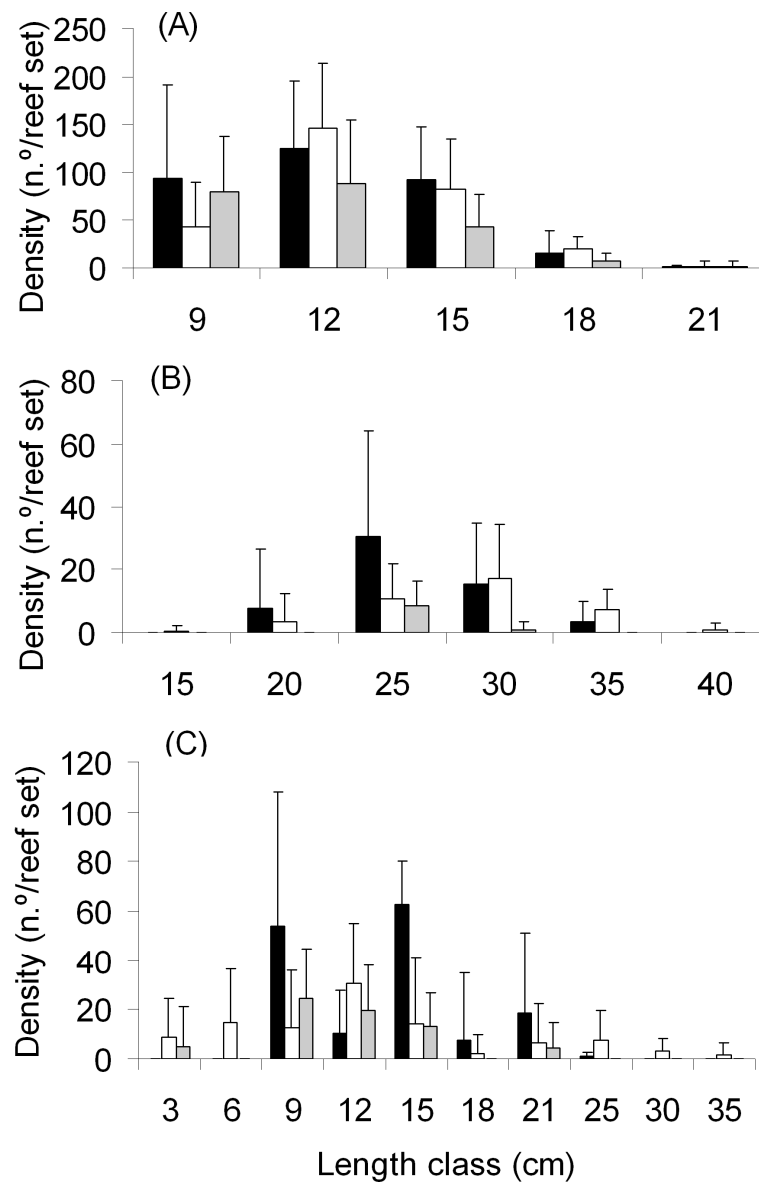


Figure 2. Mean density distribution (\pm standard deviation) per length size classes for *Diplodus bellottii* (A), *D. sargus* (B) and *D. vulgaris* (C) assemblages. Black, white and grey bars refer to the year of reef deployment (1990, 1998 and 2002, respectively).

The size composition of the *D. bellottii* assemblages (Hypothesis I) varied both among RGs counts ($R=0.370$; $P=0.002$) and among habitats ($R=0.338$; $P=0.002$). These differences were significant between habitats deployed in 1990 and 2002 ($R=0.344$; $P=0.066$) and 1998 and 2002 ($R=0.496$; $P=0.086$). The 9 (28<dissimilarity %<38) and 12-cm (29<dissimilarity %<41) juvenile size classes were those that most contributed to the density differences (>67%) verified in fish size distribution frequencies among habitat age (Table 1), followed by the 15-cm size class (22<dissimilarity %<25).

Table 1. Size classes that most contributed (%D) to dissimilarity (SIMPER) among artificial habitat age (year of deployment: 1990, 1998 and 2002). Results of the SNK pair-wise test among size classes reef age NS: non-significant; * $p < 0.1$; ** $p < 0.05$; *** $p < 0.01$.

	<i>D. bellottii</i>			<i>D. sargus</i>			<i>D. vulgaris</i>		
	1990 vs 2002	%D		1990 vs 2002	%D		1990 vs 2002	%D	
Length class (cm)	9	NS	35	25	*	47	9	*	32
	12	NS	32	30	**	36	12	*	15
	15	**	25	35	***	8	15	*	33
		1998 vs 2002			1998 vs 2002			1998 vs 2002	
	9	*	28	25	NS	27	25	*	27
	12	NS	41	30	**	38	30	*	38
	15	NS	22	35	***	20	35	*	20
		1990 vs 1998			1990 vs 1998			1990 vs 1998	
	9	*	38	20	NS	13	9	*	24
	12	NS	29	25	*	42	12	*	15
	15	NS	22	30	NS	30	15	*	28

Graphical analysis (Figure 3) showed that mean *D. bellottii* density increased with reef age (329 ± 118 , 295 ± 114 and 220 ± 57 /RS on reefs deployed in 1990, 1998 and 2002, respectively). However, differences in density were only found for the total (Hypothesis II) and adults assemblages, among the oldest and youngest habitats (Table 2). The mean total biomass (11138 ± 4797 , 10989 ± 4422 and 6642 ± 2043 g/RS for reefs deployed in

1990, 1998 and 2002, respectively) increased with habitat age too (Figure 3). Total *D. bellottii* biomass (Hypothesis III) was significantly higher in the habitats settled in 1990 and 1998 than in 2002 reef (Table 2).

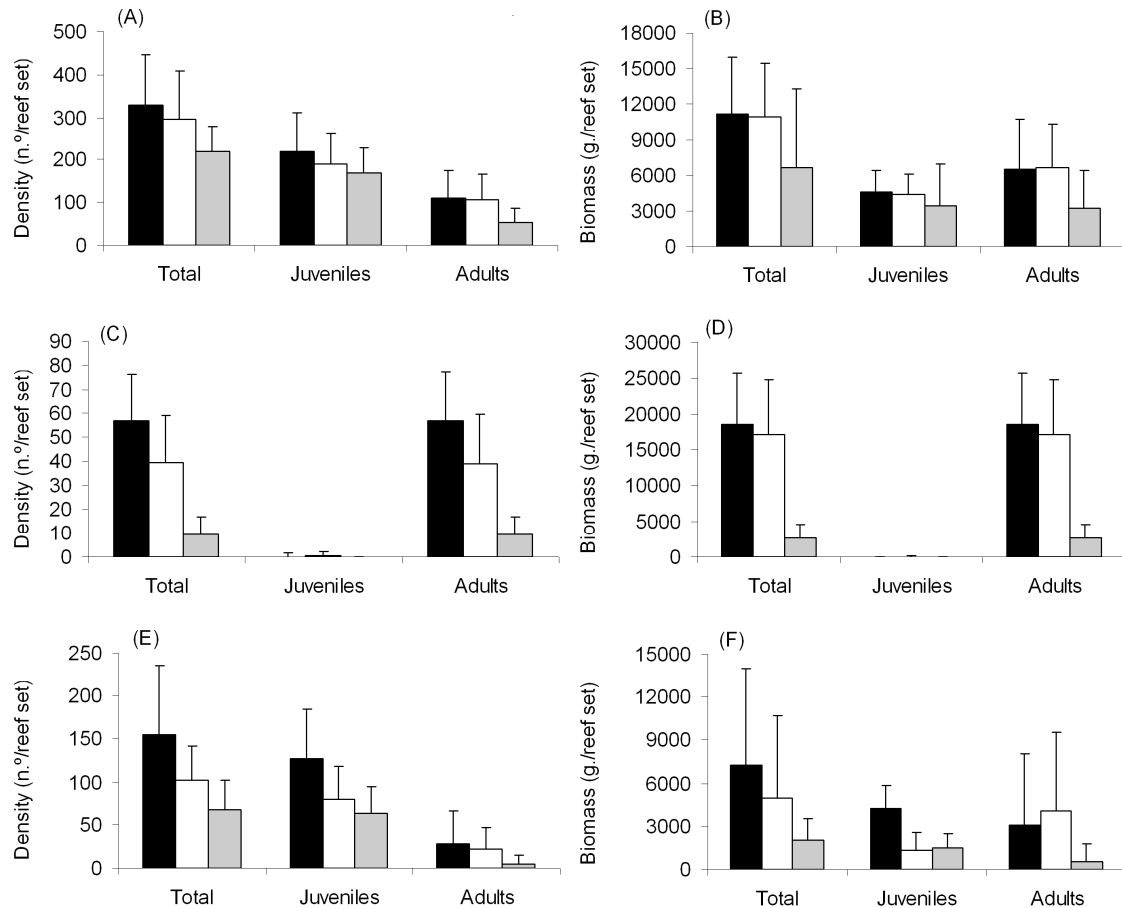


Figure 3. Density and biomass per reef set of *Diplodus bellottii* (A and B), *D. sargus* (C and D) and *D. vulgaris* (E and F) by category. Black, white and grey bars refer to the year of reef deployment (1990, 1998 and 2002, respectively).

Diplodus sargus

D. sargus ranged in size between the 15 and 40-cm size classes, being almost exclusively composed of adults (Figure 2). The size composition of the *D. sargus* assemblages (Hypothesis I) differed within RGs counts ($R=0.504$; $P=0.001$) and among

habitat ages ($R=0.360$; $P=0.022$). Significant differences were found both between 1990 and 2002 habitats ($R=0.347$; $P=0.057$) and 1998 and 2002 ($R=0.667$; $P=0.029$) habitats.

Table 2. Results of two way nested ANOVA (factors: RG within age; Age) to total, juvenile and adults density and biomass (per reef set) of *Diplodus bellottii*, *D. sargus* and *D. vulgaris* assemblage among artificial habitat age (year of deployment: 1990, 1998 and 2002). NS: non-significant; * $p < 0.1$; ** $p < 0.05$, *** $p < 0.01$. SNK – Student-Newman-Keuls pair wise comparison test.

	Density			Biomass		
	RG	Age	SNK	RG	Age	SNK
<i>Diplodus bellottii</i>						
Total	NS	**	1990>2002	NS	**	1990/1998>2002
Juveniles	NS	*		NS	NS	
Adults	NS	*	1990>2002	NS	**	1990>2002
<i>Diplodus sargus</i>						
Total	**	***	1990>1998>2002	**	***	1990/1998>2002
Juveniles						
Adults	**	***	1990>1998>2002	**	***	1990/1998>2002
<i>Diplodus vulgaris</i>						
Total	NS	***	1990>2002	NS	**	1990/1998>2002
Juveniles	NS	**	1990>2002	NS	**	1990>2002
Adults	*	***	1990/1998>2002	*	***	1990/1998>2002

The size classes that most contributed to dissimilarity across reef age were the 25, 30 and 35-cm size classes (Table 1). The SNK test showed that the differences in fish abundance by size classes across reef age are associated to higher fish density of the larger size classes of the older habitats (Table 1; Figure 2). Total *D. sargus* density was 57 ± 32 , 40 ± 19 and 9 ± 7 /reef set, for 1990, 1998 and 2002, respectively (Figure 3). A significant increase of *D. sargus* density (Hypothesis II) with reef age was observed (Table 2). Density variability among RGs counts, within age, was significant. The observed *D. sargus* biomass was 18496 ± 7354 , 17051 ± 7864 and 2611 ± 1976 g/RS for

the habitats deployed in 1990, 1998 and 2002, respectively (Table 2; Figure 3). Total *D. sargus* biomass (Hypothesis III) was significantly higher in the habitats settled in 1990 and 1998 than in 2002 reef (Table 2).

Diplodus vulgaris

D. vulgaris ranged in size between the 3 and 35-cm size classes, being mainly composed of juveniles that accounted for 82, 80 and 93% of the species assemblages on the reefs deployed in the 1990, 1998 and 2002, respectively (Figure 2). There was notable density variation in *D. vulgaris* size class distribution across habitat age. The size composition of the *D. vulgaris* assemblages (Hypothesis I) varied within RGs counts ($R=0.536$; $P=0.001$) and among habitat ages ($R=0.427$; $P=0.002$). Dissimilarity in size frequency distributions among habitat ages was mainly due to the 15, 9 and 12-cm (juvenile) size classes (Table 1). The comparison between both 1990 and 2002 and 1990 and 1998 habitats showed differences that were mainly due to the higher density of juvenile fish belonging to the 9 and 15-cm size classes in older habitats. These size classes contributed with 65 and 52 % to the dissimilarities among latter pair-wise habitats (Table 1; Figure 2). The density of *D. vulgaris* for 25 and 30 and 35-cm size classes were significant higher in 1998 than in 2002 habitat. Graphical analysis showed an increase of *D. vulgaris* mean density with reef age (Figure 3). Mean *D. vulgaris* density found was 154 ± 82 , 102 ± 82 and 67 ± 36 /RS for reefs deployed in 1990, 1998 and 2002, respectively. Statistical differences in total (Hypothesis II), juvenile and adult density were verified between the oldest and youngest habitats (Table 2). Mean *D. vulgaris* biomass found was 7244 ± 6695 , 5730 ± 1284 and 1491 ± 917 g/RS for reefs deployed in 1990, 1998 and 2002, respectively (Figure 3). Total (Hypothesis III) and adult assemblage biomass was significantly higher in both 1990 and 1998 than in 2002

habitat. The juvenile biomass was significantly higher in 1990 than in 2002 habitat (Table 2).

Benthic community

A total of 13 *taxa* were identified, all being present on every habitat independently of age. Overall, the *taxa* that most contributed for the overall biomass were the Cirripedia (barnacles), the turf algae, Gastropoda, Polychaeta, Bryozoa and Crustacea (Table 3).

Table 3. Mean biomass ($\text{g/m}^{-2} \pm \text{SD}$) value per *taxa* for the different artificial habitat age (year of deployment: 1990, 1998 and 2002). Results of two way nested ANOVA (factors: RS within age; Age) among benthic biomass across reef age: NS: non-significant; * $p < 0.05$; ** $p < 0.01$, *** $p < 0.001$. SNK - Student-Newman-Keuls pairwise test.

	1990	1998	2002	RS	Age	SNK
Anthozoa	3.47 ± 2.88	6.66 ± 11.53	0.25 ± 0.52	NS	NS	
Bivalvia	3.91 ± 2.82	3.26 ± 1.77	38.09 ± 75.82	NS	NS	
Bryozoa	25.33 ± 21.64	7.59 ± 5.51	46.04 ± 40.1	NS	*	2002>1990/1998
Cirripedia	1020.81 ± 135.83	863.55 ± 76.24	713.85 ± 454.6	NS	NS	
Crustacea	20.99 ± 19.95	7.46 ± 4.08	7.98 ± 4.5	NS	*	1990>1998/2002
Echinodermata	0.43 ± 0.35	1.47 ± 1.48	0.72 ± 0.85	NS	NS	
Echiura	11.01 ± 12.21	1.13 ± 2.55	4.4 ± 3.52	NS	NS	
Foraminifera	0.09 ± 0.04	0.13 ± 0.11	0.09 ± 0.08	NS	NS	
Gastropoda	54.63 ± 53.08	37.81 ± 18.67	28.99 ± 20.1	NS	*	1990/1998>2002
Hydrozoa	2.04 ± 3.25	0.35 ± 0.72	0.54 ± 0.43	NS	NS	
Algae	376.62 ± 194.83	315.78 ± 170.71	152.64 ± 64	NS	*	1990>1998/2002
Polychaeta	16.90 ± 20.65	18.47 ± 11.21	27.36 ± 18.9	NS	NS	
Sipunculida	17.80 ± 10.97	2.59 ± 5.85	3.14 ± 3.01	*	*	1990>1998/2002
Total biomass	1554 ± 965	1266 ± 568	1024 ± 686	NS	NS	

The two older reefs were closely related in MDS space than the younger reef, suggesting that macrofauna and algae habitat composition differ with AR's age (Figure 4). However, the above analyses did not take into account the Cirripedia (barnacles), since their higher biomass contribution mask the role of the other benthic *taxa*

composition across reef age. Statistical analysis corroborated that benthic habitat composition (Hypothesis IV) varied among habitat age (ANOSIM: $R=0.191$; $P<0.01$) but not among RS with the same age (ANOSIM: $R=0.136$; $P=0.168$). Statistical differences in benthic biomass structure were found between the most recently deployed reefs and both the two other habitats ($AR_{s1998vs.2002}$: $R=0.231$; $P<0.022$; $AR_{s1990vs.2002}$: $R=0.283$; $P=0.011$). The *taxa* that most contributed (SIMPER) to the differences observed between reefs (Table 4) were the turf algae, Sipunculida, Bryozoa, Bivalvia, Echiura, Gastropoda and Polychaeta.

Table 4. SIMPER results highlighting biomass *taxa* that most contributed to dissimilarity (%D) across artificial habitat age (year of deployment: 1990, 1998 and 2002).

1990 <i>vs</i> 1998		1990 <i>vs</i> 2002		1998 <i>vs</i> 2002	
<i>taxa</i>	%D	<i>taxa</i>	%D	<i>taxa</i>	%D
Algae	21.93	Algae	25.09	Algae	24.38
Sipunculida	13.37	Bryozoa	11.04	Bryozoa	16.28
Bryozoa	10.53	Sipunculida	10.3	Bivalvia	10.89
Echiura	9.85	Bivalvia	10.1	Polychaeta	9.54
Gastropoda	9.72	Polychaeta	9.69	Gastropoda	8.06
Polychaeta	9.21	Gastropoda	9.55		

Nevertheless, not all *taxa* groups highlighted by the SIMPER analysis differed across reef age. Statistically significant biomass differences across reef ages (Hypothesis V) were solely verified for Gastropoda, Sipunculida and Crustacea, with higher mean biomasses on reefs deployed in 1990 than on the other two reefs (Table 3). Mean biomass values for turf algae were also statistically higher in older habitats (1990 and 1998). Bryozoa biomass was statistically higher in 2002 than in both 1990 and 1998 reefs. An increase in total biomass was observed with reef age (1554 ± 965 , 1266 ± 568

and 1024 ± 686 g/m² for the reefs deployed in 1990, 1998 and 2002, respectively). However, the total benthic biomass did not differ significantly across habitat age (Table 3).

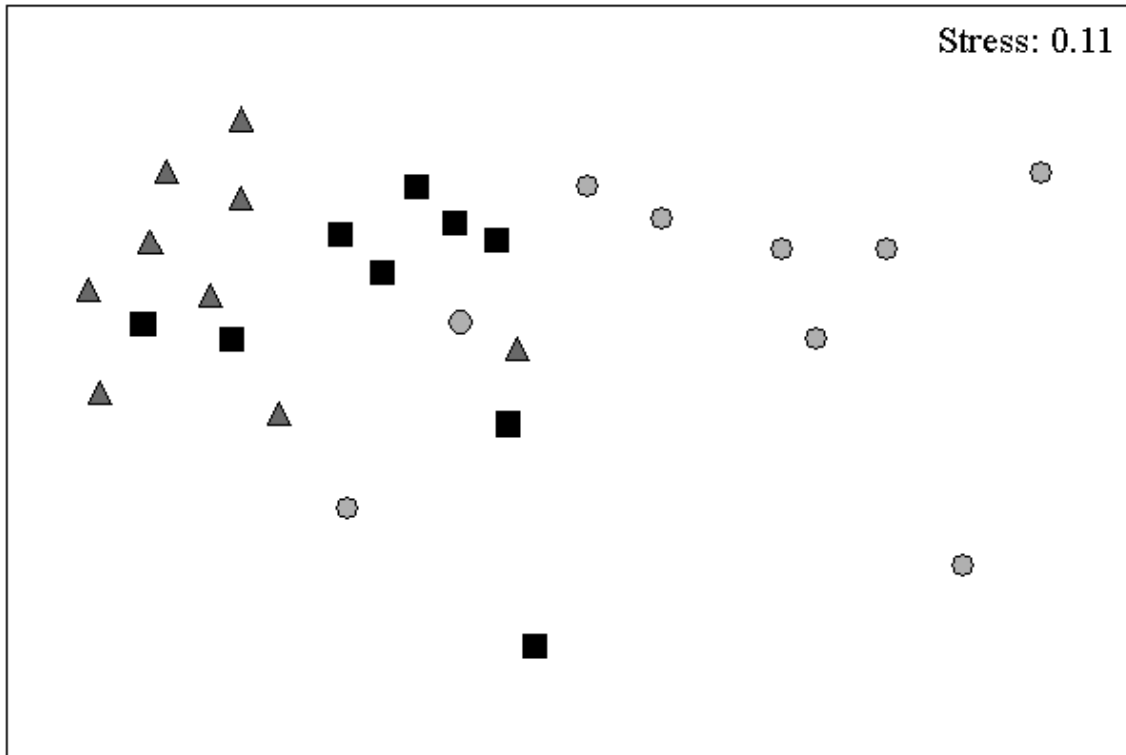


Figure 4. Benthic samples MDS ordination showing the biomass composition of different age habitats. Symbols refer to year of reef deployment (■ - 1990; ▲ - 1998; ● - 2002, respectively).

Fish assemblages and benthic community relationship

The numerical output of the RDA for the habitat structure across habitat age showed that the explanatory variables explain 75% of the variation in the *Diplodus* spp assemblages. For this 75%, the first two axes explain 96%. The triplot of the habitat structure data showed that the density of the *Diplodus* spp. was positively correlated with reef age (Figure 5). Age, Algae, Sipunculida, Gastropoda and Cirripedia were

within the explanatory variables those that most contributed to the RDA model explanation. *Diplodus* spp density was also positive correlated with Anthozoa, Gastropoda, Sipunculida, Crustacea and Hydrozoa biomass.

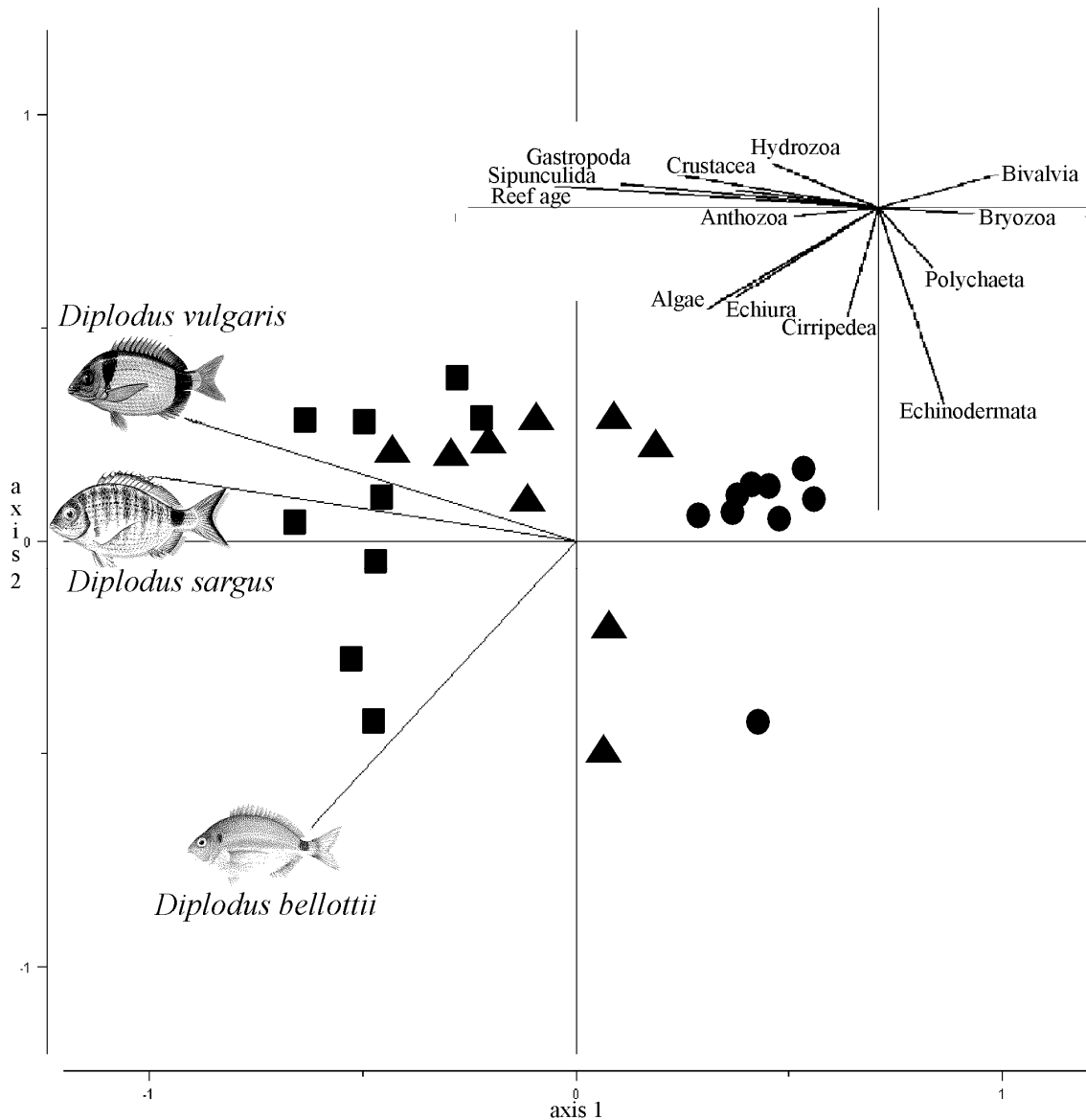


Figure 5. Redundancy analysis (triplet) showing relationship between benthic macrofauna biomass (explanatory variables), *Diplodus* spp. abundances (responses) and samples. Symbols refers to the year of reef deployment (■ - 1990; ▲ - 1998; ● - 2002, respectively).

A relationship between both *D. sargus* and *D. bellottii* and algae were observed, being particular meaningful in the case of the latter species. The relationship was not so strong between algae and *D. vulgaris*, but it was in the case with both Crustacea and Sipunculida. *D. sargus* density was also strongly related with Crustacea. Both *D. sargus* and *D. vulgaris* were strongly associated with Gastropoda. A positive relationship was verified between both *D. sargus* and *D. bellottii* and Cirripedia, being markedly strong in the case of the latter species.

Overall, there was a positive relationship between reef age and benthic *taxa* (explanatory variables) that most contribute to the RDA model (Anthozoa, Gastropoda, Sipunculida, Hydrozoa, Crustacea and turf algae). In addition, the latter explanatory variables were also generally related with the oldest AR benthic samples, but not correlated and/or negatively correlated with AR deployed in 1998 and 2002. This means that algae biomass increase with reef age. Polychaeta, Bryozoa and Bivalvia *taxa* were positively related with recently deployed habitat samples (i.e higher biomass) and negatively correlated with fish density. *Diplodus* spp. densities were negatively correlated and/or not correlated with *taxa* samples recorded on the two newest ARs. Overall, for hypothesis VI, but considering also the hypothesis IV and V results, the tentative conclusion was that habitat structure, namely due to specific *taxa* availability, strongly influence *Diplodus* spp. density variability across habitat age.

DISCUSSION

We analysed the fish assemblage data first, and then used habitat structure as a variable to test for statistical difference in *Diplodus* spp. density across habitat with different ages. This strategy kept the analyses separate and avoided effects of any *a priori* assumptions about relationships between fish and biota (Godoy *et al.* 2002). The size

structure differences among habitat age were generally due to high densities of juvenile of *D. bellottii* and *D. vulgaris* on the older reefs (Hypothesis I). In the case of *D. sargus* the density was generally higher on the older habitat, independently of size class. Size structure variability among RG counts, within each reef age, was also considerable. Nevertheless, when abundance and biomass differences across habitat age were analysed (all size classes pooled) differences among RG counts (within age) were not so evident, except for *D. sargus*. Willis *et al.* (2006) showed that counts separated by months return quite different assemblages whereas counts separated by days or weeks (as in the present study) did not. The *Diplodus* spp. mobility in the area of study is still poorly known but a three month acoustic telemetry surveys conducted in Faro/Ancão area showed that both *D. vulgaris* and *D. sargus* marked fish had not moved outside the study area (Lino *et al.* in press). Mobility makes the analysis performed conservative, because mobility would tend to break up any spatial pattern as it tends to break up any abundance or length structures appearing in a fish assemblage (García-Charton and Pérez Ruzafa 2001). Therefore, it is not possible to state that the specimens' counted on the different aged habitats, are permanent inhabitants of the respective habitats. Instead, we must consider that fish have habitat preferences because in some habitats they have better conditions for growth, feeding and spawning/reproduction (Lloret and Planes 2003; Leitão *et al.* 2007).

Ecologically it is assumed that fish abundance exists close to the carrying capacity of the environment with reef fish abundance considered limited by habitat or space partly because artificial bottom reefs are patchy habitats, limited in geographic coverage and separated from other reefs. Habitat limitation is the primary factor in determining the specific composition of AR fish assemblages, due to the availability of food or shelter (Bohnsack 1989). Both space and shelter requirement in our study were

conservative across reef age, as well as reef unit type (e.g. substrata type, complexity, space availability) and structural organisation (e.g. number of modules, chaotic organisation). In addition, other factors known to affect reef fish assemblages (e.g. depths, bottom type and lack of proxy rocky areas), were also kept across habitat age. Artificial habitats were deployed to mimic coastal rocky areas where larval supply is usually not a limiting factor (see Almada and Faria 2004 for a review), suggesting that post-recruitment deterministic mechanisms in our study are exerting a strong influence on assemblage organisation.

A pattern in *Diplodus* species density (Hypothesis II) and biomass (Hypothesis III) variation among habitat age was found. One of the most significant factors explaining the fish density pattern found in infra-littoral fish assemblage, (e.g. species richness, biomass, number of individuals), despite the fact that they deal with distinct faunas in many different parts of the world, is habitat benthic composition (Ardizzone *et al.* 1997; Friedlander and Parrish 1998; Godoy *et al.* 2002; Munday 2002; Almada and Faria 2004; Floeter *et al.* 2007). Fishes move to exploit resources, principally food and shelter and as a general rule they select foraging areas to maximize food intake where proper habitat is available. The degree of habitat selectivity exhibited by a species has scale-related implications for patterns of abundance (Fox and Morrow 1981; Brown 1984; Gaston and Lawton 1990; Munday 2002). *Diplodus* spp. are closely associated with rocky habitat environments and respective benthic *taxa* (preys), as commonly described in diet studies, which characterise these as high generalist species (Pepe *et al.* 1996; Sala and Ballesteros 1997; Gonçalves and Erzini 1998; Horta *et al.* 2004; Leitão *et al.* 2007). Therefore, under such conservative habitat experimental design, the benthic community became the most likely explanation for reef fish assemblage density differences between habitats with different ages. For instance, differences in *Diplodus*

spp. density (Hypothesis II) were verified between the ARs deployed in 1990 and 2002, which have different benthic communities too (MDS analysis, Hypothesis V). No differences in fish density were observed between the ARs deployed in 1990 and 1998, nor in their respective benthic community. In addition, redundancy analysis (RA) showed that *Diplodus* spp. density increased with AR age, being associated to specific benthic *taxa* and flora availability (Hypothesis VI).

However, *D. vulgaris* and *D. bellottii* total assemblage density (Hypothesis II) were similar in both 1998 and 2002 habitats, while benthic structure differed among the 1998 and 2002 habitats. The MDS ordination analysis, (Hypothesis IV) grouped the 1990 and 1998 habitats and cross correlation procedures of redundancy analysis (Hypothesis VI) separated more clearly macrofauna samples according to both habitat age and *taxa*. Therefore, redundancy analysis was stronger in showing an increase in fish abundance with reef age and highlighting that higher density of fish in older reefs was associated with specific benthic *taxa* biomass availability. It also showed strong relationships between *Diplodus* spp. density and both specific benthic *taxa* (Crustacea, Gastropoda, Sipunculida and Bryozoa) and turf algae on older habitats. Moreover, those *taxa* were important as preys for *Diplodus* spp as previously reported by several authors (Pepe *et al.* 1996, 1998; Horta *et al.* 2004; Leitão *et al.* 2007).

Different analyses have showed different types of information and results, thus discussion may be open to interpretation. We should mention here that our non-manipulative approach may appear less powerful than a field experiment and may be criticised because mobility and fish assemblages' variability would introduce substantial variability. Nevertheless, *Diplodus* spp density differences were still found between habitats with different ages, and being related with habitat structure. Therefore, despite such weakness, we favour the general interpretation of our results, including natural

observations (graphical analysis and numerical analysis) and redundancy analysis (model inertia was high for ecological data set, Zuur *et al.* 2007), because we believe they provide opportunities to understand local and large scale fish dynamics due to habitat composition and fish habitat preferences, even if they do not satisfy the accepted protocols of experimental design, such as: i) the lack of stomachs contents to link information (Stål *et al.* 2007); and ii) the differences in the size of habitat across age (attraction hypothesis). However, *Diplodus* spp showed a strong substrate preference for rocky (artificial) habitats and large amounts of literature supports this linkage. It might be also argued that large size (area) reefs attract more fish (attraction hypothesis, Bohnsack 1989). However, even under such argument, marked differences in *Diplodus* spp density were observed among the two different habitats off Faro/Ancão area. In the smallest Faro AR, higher fish density was found than in the nearby large sized Faro/Ancão AR. As both latter reefs differed in terms of benthic assemblages, these findings favour the hypothesis that benthic habitat structure and fish habitat selectivity are the putative cause of fish density differences among habitat age. In addition, it is also noteworthy that the result achieved with regards to the Faro/Ancão nearby habitats, with different benthic habitat structure, have implication in terms of the spatial organisation of *Diplodus* spp. assemblages. The fast rate of colonisation of the youngest AR was attributed to the migration of fish from the existing older proxy AR deployed in 1990 (Leitão *et al.* 2008b). Nevertheless, these two habitats which are side by side, showed differences in terms of *Diplodus* spp. density/biomass, whereas ARs separate from kilometres (Faro/Ancão and Vilamoura) did not. Thus, the habitat structure has contributed to the separation of local fish assemblages off the Faro/Ancão area. Other studies also referred that the causes of fish abundance variability over spatial scales

have been showed to be related to local-scale processes and associated to benthic habitat selection (Packer and Hoff 1999; Morris 1992; Munday 2002).

Overall results suggested habitat selective preferences by *Diplodus* spp. Ross (1986) showed that in the case of fish assemblages, food segregation has proven to be the most important resource partitioned by species and seems to play a more important role than habitat, spatial or temporal separations. Ardizzone *et al.* (1997) showed that the gradual increase of site fidelity AR fish abundance with reef age is related to changes in benthic community. Spanier (2000) demonstrated in an experiment based on the enrichment of the substrate, that the abundance of the mostly represented fish species (*Epinephelus alexandrinus* and *E. guaza*) decreased after enrichment effect was reduced. Edgar and Shaw (1995) investigated the importance of food resources by quantifying the production of food and relating this to the consumption and production of the local fish community. The latter authors showed that preferred prey availability may limit the production of fishes. In fact, the debate regarding the structure of a fish population can be thought of in terms of the trophic energy available/produced within an ecosystem, the main question being related to its carrying capacity. At one extreme, once fish have colonised reef structures, assemblages are assumed to be at a maximum carrying capacity allowed by the habitat environment. The fish assemblage structure will result from competition between and within species. This view is based on the assumption that fish communities are ordered assemblages composed of populations, each in numerical equilibrium with their resource supply (Smith and Tyler 1972; Sale 1978). It is important to note that in the present studied sites the fish assemblages were assumed to have already reach stability (Santos *et al.* 2005; Leitão *et al.* 2008b). However, the variation in benthic habitat composition across habitat age means that benthic succession, defined as a change in species dominance through time initially by

the opening of space as species extinct (Odum 1970), has influenced the associated *Diplodus* AR assemblages. Therefore, a dynamic equilibrium between fish and food resources is expected to be reached continuously along the several levels of habitat colonisation as postulated by Smith and Tyler (1972) and Sale (1978).

The present study stresses the importance of within-reef benthic habitat variability as determinant to the composition of local reef fish assemblages. Moreover, results suggested that fish assemblage density varied within-site spatial variation according to benthic habitat characteristics. Artificial habitats deployed in coastal waters of the Algarve offer a large portion of the available hard habitat substrate between 16 to 24 m depth and have a high importance for *Diplodus* spp as nursery, feeding (Leitão *et al.* 2007; Leitão *et al.* 2008a), growth and mating/reproductive areas (chapter 5). Thus, these habitats were considered as “Essential Fish Habitats” [EFHs - defined as “those waters and substrate necessary for fish for spawning, feeding or growth to maturity” (Benaka 1999)]. One of the primary purposes of artificial habitats is to attract and concentrate fish by providing them with more favourable conditions than are present in the original environment. Coastal zones are known for maintaining a high diversity of fishes compared to offshore areas, offering organisms multiple habitats and food resources. Thus, the man made structures acting as artificial EFHs should have an important role to play in sustainable management of exploited sparids fisheries, as conservation and ecosystem issues will dominate fisheries management in coming years. In fact, management will hopefully focus a greater amount of effort on conserving entire ecosystems, including the variety of habitats, rather than target exclusively on maintaining the sustainability of fish resources. Thus, the artificial reefs can have an important role in future management plans for coastal fishing ground.

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CHAPTER 7

FINAL DISCUSSION AND CONCLUSIONS

FISH ASSEMBLAGES AND COLONISATION

There are many studies on AR colonisation and fish assemblage's variation over time after reef deployment. However, few studies have carried out monthly sampling over a prolonged period such as our two year study (for a review see Brickhill *et al.* 2005). A rapid increase in fish colonisation occurred in the first months after the deployment of the large-scale multi-reefs near to the small pilot AR (chapter 2). The probability that an individual fish locates a reef is expected to increase with habitat connectivity (Fernández *et al.* 2008 and references therein), and with the increase of reef set numbers (fish instinctive response to ARs), which affects the rate of fish relocation among habitat patches. Therefore, it follows that the degree of connectivity, or isolation, among reefs and other suitable habitats (Ria Formosa- new AR; old AR-new AR; coastal areas-AR) favour fish movement (e.g. increasing numbers of *Dicentrarchus labrax* have been observed in ARs just after the enlargement of the AR off Faro/Ancão) and post-settlement relocation of individuals, contributing to a fast colonisation in an area characterised by a largely sandy bottom. Rapid reef fish colonisation was also suggested to be related with trophic attraction and seasonal (late summer/Autumn) migration of sub-adults fish from Ria Formosa (chapter 2 and 5). However, the Ria Formosa effect was not the same for the studied sparids species (*Diplodus bellottii*, *D. sargus* and *D. vulgaris*, chapter 5). In addition, fish assemblage colonisation was also associated with reef trophic attraction.

ARs were proposed to serve as shelter areas for juvenile fish (Monteiro and Santos 2000). Shelter from predation is important as the greater the fish survivorship, the greater the probability of increased reef production (Bohnsack 1989). However, the role of ARs as shelter habitats and the enhancing of fish post settlement survivorship, is dependent on species specific behaviour and response to sunken structures, rather than

by the availability of space and shelter itself. For instance, young of the year and juvenile demersal species (*Boops boops*, *Trachurus trachurus* and *Pagellus* spp.) that contribute up to 40% of reef density, do not shelter in AR, schooling in the water column where they become more vulnerable to *D. labrax* predation.

Fish assemblages inhabiting rocky areas are described as high resilience despite the fact that they deal with distinct faunas in many different parts of the world (Almada and Faria 2004). ARs are deployed to mimic rocky areas. Nevertheless, excluding seasonal demersal fish episodes and inter-annual peaks in Blenniidae recruitment, ARs fish assemblage (e.g density, species richness) do not undergo marked fluctuations (chapter 2 and 5). This shows that ARs have the capacity to aggregate and stabilize fish assemblages, as previously suggested by Santos (1997).

The deployment of large-scale multi-reefs near to a small pilot AR contributed to an increase in the mean values of ecological indices (higher species richness and diversity), as well as in fish density and biomass abundances, compared to what was previously reported for fish assemblages in the old smaller pilot AR deployed in 1990 (chapter 2). Economically important species accounted for a mean of 180 g/m³ (87%) of total reef fish biomass (95 kg/reef set). *D. Labrax*, along with sparids of the genus *Diplodus* accounted for most of the reef exploitable (adult) biomass, 159 g/m³ (84 kg/reef set). The Algarve and namely Faro/Ancão reef area was enlarged significantly in 2002. An increase of 7.5 times the previous AR area (from 7 to 52 AR groups), generated an increase from 1.49 to 2.8 ind/m³ and from 81.0 to 207 g/m³, corresponding to 2 and 3 times higher density and biomass of fish, respectively. Therefore, with the enlargement of the Faro/Ancão system *new* fishing grounds were created, allowing a reduction on the fishing effort over traditional scarce natural reef grounds. This suggests

that ARs, throughout their fish assemblages, can have a beneficial effect in terms of enhancing local fisheries.

CONTRIBUTION OF AR TO FISH PRODUCTION/FISHERY ENHANCEMENT (FISH-AR RELATIONSHIP)

All biological entities require matter for their growth and energy for their activities. This is true not only for individual organisms, but also for the populations and communities that they form in nature. One of the questions regarding Algarve AR is whether food growth and/or fish attraction to the AR contributed to the production of new fish biomass. This is an important issue as ARs that produced significant biomass may be useful for increase/recovery of fish resources. The underlying rationale of ARs deployment is the production hypothesis, whereby AR provide additional critical habitat that increases the environment's carrying capacity and eventually the abundance/biomass of reef fishes. Mechanisms for increase in fish production include providing additional food. Documenting food web relationships could demonstrate the potential of AR to increase production. In this regard, two commercial species, *D. sargus* and the *D. labrax*, were studied (chapter 3 and 4).

As stomach content surveys have demonstrated, AR's sessile invertebrates and algae provide a food source for *D. sargus* (chapter 3), which is a species with a strong site fidelity to reef structures. Moreover, results showed a strong relationship between AR's macrobenthic communities and *D. sargus* diet, suggesting a strong dependence on these artificial habitats (chapter 3). However, in the case of *D. sargus*, this contribution to the trophic ecology of the species is exclusive to the adult phase of the species life cycle. This is, for *D. sargus* increase biomass production by fish-AR trophic transference is predicted to depend on individual ages. This highlights the role of

ARs as an extension of the scarce local rocky habitat feeding grounds in the area for adults (spawning biomass) of *D. sargus*.

The *D. labrax* do not benefit directly from AR production but rather indirectly by feeding on juvenile reef demersal species attracted to these structures. Sale (1969) predicted that fish accumulate in preferred habitats through a process of appetitive exploration, where fishes in adequate environment will spend little time exploring new environments, compared to fish in less adequate environments. Diet studies and census data showed that *D. labrax* frequency of predation was high, taking place whenever preys were available. On the other hand, it was showed that predator consumption (prey mortality) increases with prey density. These results highlight the importance of ARs as feeding areas. Therefore, predation may assume a significant importance regarding ARs, as the structure of the food web can be controlled by the biodiversity within the system and/or by higher predators (top-down control). Whether or not *D. labrax* predation in AR is similar to that in natural areas, the addition of man made substrate favours an increase in prey-predator encounters and therefore prey consumption and higher prey mortality. Thus, *D. labrax* predation on demersal prey has implications for the understanding of fish assemblage's evolution and thus coastal resources. That is, fish prey-predator interactions are important in terms of conservation and management, as well as for the evaluation of the long-term effects of ARs deployment.

Deployed substrate undoubtedly provides and/or attracted additional food but it remains to be shown how much new fish biomass is consequently produced and whether the added biomass is a significant contribution to stock size. Estimates of reef fish production (in relation to rocky areas) has proven to be exceedingly difficult. An initial estimate of resources provided by an AR can be made under the assumption that all the fish on an AR (i.e the standing stock) are produced entirely on the reef (Ambrose

and Swarbrick 1989). However, this is likely to considerably overestimate true fish production (Bohnsack and Sutherland 1985; Ambrose and Swarbrick 1989). Nevertheless, this procedure is a starting point for evaluating reef size and production in relation to natural areas. Santos *et al.* (1995) showed that fish biomass in ARs (44.4 kg/reef set) was higher than in proxy rocky area (38.2 kg/reef set). This means that, if the biomass on the nearby natural reef is currently of the same order of magnitude and the proportion between the different species remains, to produce the same amount of biomass as a natural area, a much smaller AR area is needed (approximately 3 times less). Moreover, this suggests that the Faro/Ancão AR system (12.2 km²) would compensate for the loss (due to fishing) of biomass available in the natural reef. Nevertheless, a time lag between fish production (through reproduction and growth) and fishing catches increase is expected.

Inferences regarding future catches are difficult to make. While for some species AR can promote biomass increases due to AR production (e.g. *D. sargus*) to others might not (demersal fish preys), this is fish production is species-specific. Increases in catches due to reef production will depend on species-specific biological life cycles, fish adaptation to the reef environment and to fishing/exploitation strategies. Consequently, the increase of fish catches also necessarily needs to be addressed in a completely different way, rather than through quantification of the catches, such as evaluating the biological and ecological value of ARs for fish assemblages. This requires the identification of each species habitat use, fish-AR relationships (e.g. spawning, feeding, merely attraction), to predict long-term possibilities of fishery enhancement. As far as the fishery is concerned, fish exploitable biomass increase depends on recruitment and fish growth that influences the sustainable catch that can be taken from a stock (King 1995). Hence, AR provides the necessarily habitats for the biological cycle of marine

fish populations. For instance, size-length frequency analyses and biological studies revealed that ARs play a multiple role for *Diplodus* spp as: nursery, spawning/mating, growth and recruitment areas (chapter 5 and 6). In addition, AR can favour fish production through the creation of optimal areas for fish feeding (AR-fish trophic transference) Therefore, artificial habitats can be qualify as essential fish habitats (EFH) defined by Benaka (1999) as “those waters and substrate necessary for fish for spawning, feeding or growth to maturity”. That is, for those species that make extensive use of artificial habitats (e.g sparids and *D. labrax*) ARs can provide adequate conditions for their biological cycle. Owing to their extent, durability and benthic production and/or food attraction, we can postulate that long term AR habitats can contribute to increase fish overall exploitable biomass by providing fish with optimal areas for feeding, reproduction and growth.

MANAGEMENT

In the case of AR, independently of whether or not fish are produced our merely attracted to these man-made structures, the long term sustainability of these areas for fishing depends on management. One of the problems that might limit the increase of fish biomass and catch, is reef overexploitation due to increased catchability in ARs areas (Polovina 1991). For instance, an artisanal gill net used by local fishermen can extend to approximately 3000 m, over six AR groups. This means that in an area where fishing effort is high, a small number of boats can easily cover all the Faro/Ancão AR area. Moreover, commercial fishing tends, through the use of size-selective gears (such as gill nets, the most widely fishing gear used by the Algarve artisanal fleet), to selectively remove the largest individuals in the local fish assemblage. As suggested by Berkeley *et al.* (2004), this can consequently alter species assemblage structure, with

results including truncated size and age distributions and reduced fish abundance. In the latter context, length frequencies analysis and demographic composition (juvenile, adult) are highly important as indicators of the fishing pressure in Algarve AR areas and for proposing management measures.

The management plan for AR areas must be based on a monitoring program, which should be used to assess whether the management goals are being met. On the other hand, scientific research should be conducted to improve management, and to increase the AR production and ecosystem conservation potential. For instance, for sparids a 70 mm gill net mesh size would be the most appropriate for AR exploitation (chapter 5). The enforcement of the latter mesh size means an increase by 10 mm on the current minimum legal mesh size. Moreover, fishing yields also can increase if there is an adaptation of the fishing techniques to the fish assemblage structure. For example, on the ARs adult *D. sargus* ranged in size between 20 and over 40 cm, sizes that have a low probability of retention by gill nets with mesh size between 60 and 80 mm. Thus, due to their constant exploitable biomass availability an alternative to gill nets is to use longlines (chapter 5). Moreover, seasonal recruitment events of large numbers of occasional category demersal young of the year (YOY) and resident fish (e.g. *D. vulgaris*) schools also necessarily need to be taken in consideration, thus avoiding the use of fishing gears (such as purse seines) that could catch these juveniles during their recruitment periods (Chapter 2). That is, it is necessarily for managers to adapt the fishing exploitation regimes to the bio-ecology of the reef fish assemblages.

Until 1998 the artificial areas were probably too small to justify their specific management. However, management is particularly important in the case of artisanal fisheries, such as those off the Algarve, that are highly dependent on the availability of fishing resources. In addition, AR management is particularly important for resident

economically valuable species, as these are more vulnerable/available to the fishery. Moreover, as diversification of the catches and fish density/biomass increase due the augmentation of material deployed, greater responsibility for management is required.

Artificial habitats deployed in coastal waters of the Algarve account for large portion of the available hard habitat substrate within the depth range of 16 to 24 m. A key element in moving toward sustainable fisheries is the identification of EFH, i.e. high quality habitats for fishery species. Thus, although this study is only a first step and ignores much of the complexity of coastal ecosystems, the information gather, showed that due to ecological (e.g. feeding areas) and biological (e.g. spawning areas) value of these man made habitats can be qualify as EFHs (chapter 3, 4 and 5). This is a step towards the argument to justify the creation of marine protected areas (MPA), which are increasingly regarded as interesting management measures. ARs used in conjunction with a MPA can be an alternative option for the coming years, as together they can contribute to ecosystem conservation, that in the future will dominate fisheries management/sustainability. But they can be helpful in zoning coastal areas in order to reduce conflicts between users too. In addition, the MPA must be dynamic in the understanding that each AR system is independent and thus should be managed separately as “Marine life conservation districts” (chapter 2).

Studies on artificial reefs have focused predominantly on fish assemblages and have largely disregarded the development of sessile biota and their structural and functional relationships (Svane and Petersen 2001). One important argument that highlights the necessity of conservation of high quality habitats is the recognition of the value of different habitats for determining the structure of exploited fish assemblages. Despite the evidences of important ecological linkages between habitat(s) and fishery production, the management of most commercial resources worldwide has historically

concentrated on assessing stock size and controlling fishing mortality. However, the relationship between benthic fauna and flora and fish assemblages (chapter 6) showed that different benthic communities affect fish assemblage's composition. Therefore, a factor predicted to be important for ARs success is benthic habitat availability and composition. To maintain a sustainable fisheries industry, it is important for managers to regulate not only the total allowable catch of the fisheries, for example, but also to have in mind the role of shallow coastal habitats on the production of fish to the fisheries, i.e., the ecosystem services provided by the habitats and thus also regulate society's exploitation of the coastal environment (Costanza *et al.* 1997; Stål *et al.* 2007). It is most probable that in the future fisheries management will focus a great effort directly on the conservation of coastal zones, such as near shore ARs areas, that are known for supporting a high diversity of fishes compared to offshore areas, offering organisms multiple habitats and food resources. Consequently management necessarily needs to preserve the entire ecosystems, aiming the long term sustainability of the fishery, instead of focus particularly on fish exploitations (catches).

FUTURE STUDIES

It has been found that different sized reefs have different fish assemblage characteristics (Grove and Sonu 1985; DeMartini *et al.* 1989; Stephens *et al.* 1994; Bombace *et al.* 1995; Pickering 1996; Jensen *et al.* 2000) and that reef size significantly influences the biomass and the total number of species and individuals, with the efficiency of artificial reefs as attractors being far greater when formed into a structure than disaggregated into pieces (Moffitt *et al.* 1989; Pickering 1996; Bohnsack *et al.* 1994;). The study of ARs organization and complexity is beyond the scope of this dissertation. However, it is useful to mention that complexity may promote the enhancement of the diversity and

abundance (Carr and Hixon 1997; Sherman *et al.* 2002 and references therein) of species that are typical of coastal areas at reef depths. This might be the case of some crustaceans recorded in low abundances in AR, such as *Panulirus elephants*, *Necora puber* and *Scyllarus arctus*. These species are probably high vulnerable to predation after post settlement in AR due to the lack of shelters (Spanier 1991, 1994; Lozano-Alvarez *et al.* 1994; Barshaw and Spanier 1994; Frazer *et al.* 1994). It would also be interesting to evaluate the possibility of increasing complexity in the ARs areas, as different species along their life cycle require different refuge sizes and types. With the above mentioned crustacean species for example, it does not require a large number of animals to be caught to sustain a small fishery. It is possible that AR adaptation to lobster biology could optimise the catch and that quite a few new/alternative fishing jobs could be supported. Offshore crustacean farming units using large-scale multi ARs started in Canada and USA and are being deployed in Scotland's (Loch Linnhe) coastline (<http://www.earthdive.com/site/news/newsdetail.asp?id=1402>; Anon 2003). A survey of artificial reefs established in the recent decade has revealed that lobsters were reported in only a small proportion of the projects. It is suggested that the majority of these reefs has not been designed and constructed to fit the behavioral-ecological preferences of the target lobster species for shelter (Spanier 1994). Moreover, ARs are considered of particular interest with regards re-colonisation (restocking) using reared aquaculture fish, such as *Epinephelus marginatus*, a species that in the past was common in Algarve waters. However, this prized species and other serranidae of economic importance found in Algarve AR (e.g. *Serranus cabrilla*) have strong shelter requirements and they are territorial sedentary species (Beets and Hixon 1994). In the case of the grouper *E. striatus*, Beets and Hixon (1994) found that ARs of appropriate design (providing species sized holes) provide higher abundances than the natural reefs.

However, it is necessary to consider that this long-lived species is highly voracious and territorial, with strong habitat area requirements. So, the success for some species restocking also depends on species adaptation and fixation in AR environments. Nevertheless, the role of re-colonisation by species such as *E. marginatus*, must be also studied as the species preys on crustaceans, that could negatively impact production of crustaceans in ARs. Probably reefs with different area sizes and complexity/heterogeneity could be adapted to different species.

The lack of knowledge regarding Algarve reef design studies raises several questions. Would another type of reef organization (e.g. number of modules per reef set) produce/attract/aggregate more or different fish species? Is the minimum area (n. ° of reefs) enough to provide suitable area for such territorially sedentary species as *E. marginatus*. It is increasingly recognised that this is one of the major areas where further work is needed (Frazer *et al.* 1994; Pickering and Whitmarsh 1997). It would be worthwhile to investigate if small levels of Algarve ARs manipulation, by the deployment of man made material, would create better conditions for fishery diversification, such as crustaceans and juveniles of serranids of commercial species. Horizontal shelters with opaque cover and multiple den openings were preferred by most crustacean species mentioned above, as these characteristics are considered anti-predator adaptations (Spanier 1994). That is, the effectiveness of ARs in increasing productivity depends in particular on whether it meets the specific habitat requirements of individual target species and age groups and also on the design and size of a reef structure.

Spatial information is often underutilised owing to the lack of capability to explore spatial and temporal relationships between species distributions and environmental gradients across large spatial scales (Battista and Monaco 2004). ARs

should be managed to conserve and enhance fisheries production. In this context, the use of geographical information systems (GIS), and ecosounder surveys (fast/low cost sampling survey method), might be important tools for better understanding the temporal and spatial dynamic of reef assemblage's population and for collecting information regarding fish resources. The integration of mapping (GIS) and monitoring of ARs ecosystems and reef fish habitat utilization patterns can help managers make informed decisions about AR/MPA design and effectiveness, as well as help to define essential fish habitat and understand ecosystem function. Research involving the use of artificial reefs and GIS tools tied to zoning strategies is a positive approach to assessing the best means to protect coastal marine habitats, to enhance spawning stocks, and to manage reef fisheries. Moreover, for the development of regional zoning strategies and community-based participation ("Marine life conservation districts") it will be necessary to implement such (GIS) marine ecosystem management.

Finally, in association with the data collected by the monitoring component of AR Algarve project, results achieved in this study are important for modelling the pelagic ecosystem, to identify the functional groups, how energy and matter flow through these groups and how they are affected by physical and biological changes as well as by human activities (fisheries).

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