Northwest Atlantic Fisheries Organization

Serial No. N4477 **NAFO SCR Doc. 01/89**

SCIENTIFIC COUNCIL MEETING – SEPTEMBER 2001
(Deep-sea Fisheries Symposium – Poster)

New Findings on the Reproductive Biology of the Black Scabbardfish (*Aphanopus carbo* Lowe, 1839) in the NE Atlantic

by

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Abstract

Black scabbardfish is a deep-water species, common in the NE Atlantic at depths between 450 and 1300 m, currently exploited by some European countries. Between May 1998 and April 2000, sampled specimens collected at three different locations in the NE Atlantic - NW of Scotland, Sesimbra (Portugal mainland) and Funchal (Madeira) - were analysed. The evolution of maturity throughout the year of both sexes was studied based on the macroscopic and microscopic analysis of the gonads. Specimens with the largest total length were found in Funchal whereas the smallest size was recorded in the NW of Scotland. Neither spawners nor post-spawners were ever observed in NW of Scotland and Sesimbra. In Sesimbra, only a few number of individuals attained pre-spawning stage and most of early developing females exhibited a high percentage of atresia in their ovaries. In Funchal, all the maturity stages were found, spawners occurred from September to December (females) and from August to December (males). Length of first maturity for females was estimated to be around 1000 mm. Two groups of different size spawners were observed during the spawning period off Madeira.

Introduction

The Black scabbardfish (*Aphanopus carbo* Lowe, 1839) is a deep-water species from the family Trichiuridae. It presents a word-wide distribution, with records in the NW and NE Atlantic, from Iceland to the south of Madeira island (Saemudsson, 1949; Tucker and Palmer, 1949; Templeman and Squires, 1963; Pechenik and Troyanovskii, 1970; Forster, 1971; Bridger, 1978; Anon., 1984; Gordon, 1986; Merrett *et al*., 1991), Gulf of Aden (Norman, 1939), south of Indian Ocean (Piotrovskiy, 1977) and north-western Pacific (Clarke and Wagner, 1976). In what concerns the bathymetric distribution, this species occurs from 200 m depth around British Isles (Tucker, 1956) to 1800 m in the south of Madeira Island (Martins *et al*., 1987). Nevertheless, it is more commonly found from 500 to 800 m in the North of Europe (Ehrich, 1983; Bridger, 1978), between 800 and 1200 m off Portugal mainland (Anon., 2000) and from 700 to 1300 m in Madeira waters (Martins and Ferreira, 1995). It is considered to be bathypelagic (Wheeler, 1969; Le Gall, 1972; Bridger, 1978), however, it was also found in close vicinity to the ocean floor (Ehrich, 1983; Anon., 1999). The water temperature at the depth of capture was recorded around 3.5ºC in the Newfoundland shelf and north of Europe (Templeman and Squires, 1963; Zilanov and Shepel, 1975) and ca. 11.5ºC off the Portuguese continental coast (Le Gall, 1972; Anon., 2000).

Black scabbardfish is the target species of two Portuguese longline fisheries: the long established Madeira's fishery (since the early 19th century) and the more recent fishery off Portugal mainland (since 1983). In northern European areas, the growing landings of this species result from multi-species deep-water trawl fisheries mainly from France and United Kingdom. In fact, during the last decades the European commercial fleets have turned more to the exploitation of continental slope areas.

Despite the increasing interest on this species, little is known about its life cycle. The existing contributions on the reproduction of the black scabbardfish usually allude to a short analysis of maturity and to the size range of captured specimens. In the North of the British Isles, the majority of caught specimens were immature or in a developing maturity stage (Pechenik and Troyanovskii, 1970; Kelly *et al*., 1998); there is only reference of two individuals caught at the Porcupine bank in January with ripe gonads (Ehrich, 1983). Black scabbardfish specimens in spent condition were found in Icelandic waters between January and March (Magnússon and Magnússon, 1995). No spawners were ever observed throughout the year off the Portuguese continental coast (Machado *et al*., 1998; Anon., 2000). Specimens in pre-spawning and ripe condition were observed in Madeira waters between September and February (Carvalho, 1988; Anon., 2000) and in Azorean waters in August (Anon., 2000).

Carvalho (1988) carried out a study on reproduction of the species at Madeiran waters and identified the period comprised between October and December as the spawning season for this species. Later, under the European research project BASBLACK E.U. Study Project CT 97/0084 gonads were collected in order to establish a macroscopic maturity scale, which standardise macroscopic criteria to be adopted to assign maturity stages. This scale was further validated by the microscopic analysis of histological sections from gonads of black scabbardfish caught in three different areas of the NE Atlantic (Gordo *et al*., 2000).

Some authors have suggested that this species undertakes horizontal migrations to spawning and nursery grounds located in south European waters (Bridger, 1978; Geistdoerfer, 1982; Kelly *et al*., 1998; Anon., 2000). Others referred to vertical migrations along the continental slope in north European waters for feeding (Harrison, 1967; Zilanov and Shepel, 1975; Du Buit, 1978; Howe *et al*., 1980; Ehrich, 1983).

The main objective of this paper is to examine recent collected data on maturity from three distinct areas in the NE Atlantic – NW Scotland, Sesimbra (Portuguese continental coast) and Funchal (Madeiran waters) (Fig. 1), focussing the reproductive behaviour of this species and discussing its reproductive strategies.

Material and Methods

Fish samples were obtained from a biological sampling program executed at Sesimbra and Funchal commercial landing ports and from fishing hauls catches of Marine Laboratory - Aberdeen (MARLAB) research surveys (Table 1).

In the laboratory, all individuals were measured to the nearest millimetre below and later grouped in 1 cm Total Length (TL) classes. Individual total weight was also recorded to the nearest 1 g and their gonads removed and weighted to the nearest 0.01 g.

Macroscopic maturity stages were assigned to specimens using the maturity scale defined by Gordo *et al*. (2000). This scale was devised for both sexes comprising five maturity stages: I – Immature/Resting; II – Developing; III – Pre-spawning; IV – Spawning and V – Post-spawning.

Histological sections of ovaries and testes were also made, especially in cases where the assignment of maturity stages was ambiguous. The preparation of sections included the preservation of gonads in Bodian's AFA (Lillie and Fullmer, 1976) that remained in the fixative for a period of time between 1 to 6 days, depending on their thickness. They were later embedded in paraffin wax, sectioned at 4 to 9 µ and stained using Masson trichrome (females) and Heidenhain Azan (males).

Gonadosomatic index (GSI), which expresses gonad weight as a percentage of total weight, was estimated by month, for females and males (only for Funchal and Sesimbra, since no data on gonad weight of specimens from NW Scotland was available). Sex-ratio was calculated as the fraction of the number of females in relation to the total number of individuals captured by month. Based on the fraction of mature specimens (stages III, IV and V) per length class, the maturity ogive for the period where spawners occurred was estimated adjusting the simple logistic model referred in Zar (1996) and expressed by:

$$M = \frac{1}{1 + e^{-(b_0 + b_1 L)}},$$

where M is the proportion of mature specimens at total length class L and $ß_0$, $ß_1$ are model parameters.

Results

A total of 2443 fish were sampled in the three NE Atlantic areas: Funchal (1249), Sesimbra (826) and NW of Scotland (368).

1. Samples length composition

In the NW of Scotland, total length varied from 612 to 1175 mm (Table 2) and its median was about 900 mm in March and September, decreasing to 793 mm in October. Females were dominant in March and September (Fig. 2).In Sesimbra, TL varied from 667 to 1365 mm; but specimens between 1020 and 1110 mm TL were more frequently found. In fact, individuals smaller than 800 mm only appeared in September and specimens larger than 1350 mm were only caught in April. Females dominated samples during all the year (Fig. 2).

In Funchal, TL varied from 712 to 1510 mm (Table 2). The most frequent range was between 1150 and 1190 mm. With the exception of July and December, males were predominant in the remaining months of the year (Fig. 2).

2. Sexual cycle

In the NW of Scotland, the reduced number of samples did not allow the study of the sexual cycle throughout the year. All the individuals caught were either in maturity stages I or II (Table 3). Males were predominantly in maturity stage I. Females were predominantly in stage II in March, in stage I in October and equally represented in these two stages in September.

In Sesimbra, the majority of individuals from both sexes were also in maturity stages I and II (Table 3). From July onwards, most individuals began their gonadal development reaching maturity stage III in August – 2 % of the total number of specimens (Table 3). No stage IV was ever found. Between December and April, the majority of females in stage II showed a high incidence of atresia (Fig. 3) in the early-developed oocytes (Fig. 4).

In Funchal, all the five maturity stages were recorded. Stage II was found through all the year, being more common between March and April (Table 3). Stage III appeared in males mostly in May, while in females appeared later in July. As for Stage IV, it occurred mainly from September to December (females) and from August to December (males).

Regarding the remaining maturity stages, it must be pointed out: a) the very low occurrences (usually below 4%) of stage I for both males and females (Table 3) and b) the long duration period of stage V (from November to June). Spawning should occur preferentially during the last quarter of the year, where the highest GSI values were registered (Fig. 5). Since no sufficient data on maturity was available for males, maturity ogive was only adjusted to female data for the period between September and February, which coincides with the spawning period. Based on the estimates obtained, the length of first maturity is about 1028 mm (Fig. 6).

Due to the frequent macroscopic stage assignment errors (later corrected by microscopic analysis) related with stages I and V, the GSI values of individuals in those stages were analysed by month. In Figure 7, it can be observed that specimens in maturity stage V presented higher values of GSI than specimens in maturity stage I. Actually, the GSI values of stage I and V were significantly different (p-value =0,000089). As a consequence, this index can be useful to differentiate these two stages.

The analysis of histological sections from females in maturity stages III and IV (Fig. 8), suggest that oocyte development is essentially group-synchronous, with only one oocyte recruitment in each spawning season. Indeed, only one group of well-developed and bigger size oocytes was identified prior to the spawning season.

3. Reproductive strategy

To further understand the reproductive strategy of this species, we first analysed the reproductive characteristics of black scabbardfish individuals from Sesimbra and Funchal. In order to make such comparative analysis we have restricted both length and maturity stages to ranges that were common to both areas. In this sense, the GSI values of stage II females from Funchal and Sesimbra with TL between 1100 and 1210 mm were analysed. A distinct behaviour of this variable was observed between those two areas (Fig. 9); specimens from Funchal presented the highest GSI values, particularly from April to July.

Restricting the analysis to stage IV specimens sampled in Funchal and focussing on their TL, it was observed (Fig. 10) that:

a) A first group of individuals with TL smaller than 1250 mm began to spawn early in the spawning season (between September and December);
b) A second group of individuals with TL larger than 1250 mm spawn preferentially in January and February.

Furthermore, this fact is corroborated through the monthly distribution of GSI values of maturity stage V females (Fig. 7). According to this, there is an increasing trend on the GSI values from October to December, which is followed by a decreasing trend that lasts until May. In June the GSI variability raised again. This behaviour was deeper analysed by taken into consideration the length, particularly by analysing the evolution of GSI values for different lengths in the period from March to June (Fig.11), where two groups of post spawners could be observed: one occurring in June with individuals above 1300 mm TL and other from March to May exhibiting lengths predominantly below 1300 mm TL.

Discussion

The range of black scabbardfish total length varied between areas. Largest specimens were sampled off Madeira (above 1400 mm TL) while smallest ones were collected in the NW of Scotland (below 650 mm TL). In all the areas, juveniles (TL below 900 mm) were scarce, which posed some difficulties to the understanding of species life cycle, particularly during pre-recruitment phases. So far, there are only two records of small specimens with 100 mm and 150 mm TL that were found in a stomach of an *Alepisaurus ferox* individual captured off Madeira (Maul, 1954). Females were predominant in the two most northern areas in opposition to the southern area off Madeira.

Black scabbardfish length distribution was different between areas - large size individuals were more frequent at the southern areas than at the northern ones. This difference on length distributions between northern and southern areas (separated by parallel 40º N) was also observed on a recent study based on an enlarged length dataset (Carvalho and Figueiredo, 2001). Both geographic and fishing gear (trawl is used in the north and longline in the south) factors are important although their individual contributions are difficult to disentangle.

The macroscopic assignment of maturity stages was sometimes difficult, because the differentiation between stages was not clear. This might be related to a slow rate of gonadal development, which creates problems in the discreteness of maturity stages at macroscopic level. In fact, within each maturity stage the macroscopic aspect of its initial phase of development is more similar with the previous stage than with the final phase, whose aspect is much more similar with the following stage. Microscopic analysis of gonads proved to be very useful in clarify such misinterpretations, thus reducing the errors on maturity stage assignments. However, the expensive and time-consuming histological technique prevents its routine use to monitor maturity stage assignments. For that reason, GSI values by maturity stage constituted important auxiliary information to be taken into consideration for a correct assignment of maturity stages, being possible to identify distinct and almost non-overlapping ranges of GSI values between different stages. For instance, some problems in assigning maturity stages I and V which were solved by microscopic analysis, presented clear differences on the magnitude of GSI values between the two stages.

Mean GSI values by month were quite different between Funchal and Sesimbra. The largest GSI values obtained in Funchal reflect the continuity of sexual cycle towards spawning in Madeira, while the lower values from Sesimbra indicate its non-evolution. In fact, considering a unique range size between 1100 and 1210 mm of early developing females, the corresponding GSI values from Sesimbra were much lower than those from Madeira.

Our results clearly indicate that black scabbardfish exhibits temporal sexual maturation differences according to areas in the NE Atlantic. Vitellogenesis begins in Funchal and Sesimbra at the same time of the year, however only the specimens from Funchal continue their gonadal development towards maturation and egg release. No spawners were found in Sesimbra and NW Scotland areas. However, while in Portugal mainland a small percentage of specimens reached the pre-spawning stage, in the NW Scotland only initial development stages (I and II) were found.

Nevertheless, at more northern latitudes (Icelandic waters), spent individuals were recorded between January and March by Magnússon and Magnússon (1995). This fact suggests that the species also reproduces in northern European waters at areas not surveyed by the present study. Moreover, considering this hypothesis it is also interesting to notice that the referred period for Icelandic waters occurs slightly after the one observed at the most southern spawning area off Madeira (Dalila, 1988).

The predominance of pre-adult specimens at the NW Scotland, either immature or in an early development stage not evolving to more developed maturity stages, might reflect insufficient levels of energy to proceed.

In Sesimbra, where adults are commonly observed, there is a differentiation of two maturity categories. Immature/resting, which occur along all the year presenting very low levels of atresia and early maturing specimens that are more frequent from July till October after which enter into a intense process of atresia with a maximum in March. This process affects oocytes in different developing stages and reabsortion process occurs throughout the ovary. The length of specimens belonging to state II was above that of first maturity (ca. 1028 mm). This thus reflect that although those specimens are potentially capable of reproducing, do not enter in a spawning process and remain in a resting phase that can extend far beyond the spawning season. Possible reasons for the observed stops in the maturation process could also be related with insufficient levels of accumulated energy or unfeasible prospects of a successful reproduction. This arrest in the maturation development due to low levels of energy accumulated has been also observed for the north European eel (*Anguilla anguilla*) population and related with delays in reproductive migrations (Svedäng and Wickström, 1997).

In Madeiran waters, spawners occurred mainly from September to December. Period that comprises the spawning season referred by Dalila (1988) and Anon (2000) from October to December. Two distinct reproductive strategies seem to be followed by the species in that area, as different size specimens spawn in different time periods. While smaller size spawners occur between September and December, larger individuals preferentially undertake spawning in January and February. These findings were also corroborated by the existence of two female post-spawners groups during the spawning period.

Despite the fact that we only analysed data from three locations, it was clear that black scabbardfish presents different reproductive strategies according to the sampled area in the NE Atlantic. The maturity data collected at those areas are also in favour to the existence of reproductive migrations to spawning areas. However, the movements of Sesimbra and NW Scotland specimens towards spawning grounds are still unknown.

Acknowledgements

This study was supported by DG XIV of the European Commision through Study project 97/0084 entitled ‘Environment and biology of deep-water species *Aphanopus carbo* in NE Atlantic: basis for its management (BASBLACK)’.

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Table 1 – Sampling periods and sources for each study area.

Region (landing port)	Period	Source
Portugal mainland (Sesimbra)	Jun 1998- Apr 2000	Artisanal longline fishery
Madeira (Funchal)	May 1998 – Dec 1999	Artisanal longline fishery
Northwest of Scotland	Sep 1998, Oct 1998 and Mar 1999	MARLAB Research trawl surveys on board R/V "SCOTIA"

Table 2 - Maximum, median and minimum total length estimates and sample size by month in the three studied areas.
■ No sampling.

		Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
NW Scot.	Max			1062						1175	1091		
	Med			911						901	793		
	Min			612						636	687		
	n			80						198	90		
Sesimbra	Max	1215	1229	1207	1365	1232	1174	1342	1202	1247	1299	1317	1238
	Med	1040	1069	1024	1058	1039	1062	1105	1055	1019	1108	1071	1068
	Min	869	920	891	842	934	882	863	925	667	883	868	884
	n	86	94	59	30	67	30	138	32	83	88	62	57
Funchal	Max	1295	1393	1340	1407	1301	1440	1350	1290	1410	1510	1388	1363
	Med	1150	1165	1170	1165	1170	1192	1170	1180	1180	1170	1172	1190
	Min	1000	1050	1059	1047	1040	1030	1022	712	878	1040	1040	1000
	n	89	91	63	81	84	110	114	57	83	236	167	74

Table 3 – Relative frequency in percentage (Female:Male) by month of each maturity stage in the three studied areas.
■ No sampling.

		Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
NW Scot.	I			16:40						32:29	30:50		
	II			38:6						33:6	17:3		
Sesimbra	I	29:17	49:23	37:25	43:23	33:28	43:20	13:15	31:19	37:36	32:27	42:15	19:19
	II	30:23	16:12	19:19	13:20	19:19	23:13	49:23	25:19	19:6	26:14	32:11	32:25
	III	---	---	---	---	---	---	---	3:3	0:1	0:1	---	2:4
Funchal	I	0:0	1:0	0:0	1:0	2:1	0:0	3:1	2:2	2:1	1:0	1:0	1:0
	II	3:13	15:14	21:13	21:35	25:18	23:33	38:4	9:4	7:6	6:5	3:4	8:7
	III	2:2	0:3	0:2	1:2	0:15	0:20	17:33	30:35	24:35	15:21	5:11	5:3
	IV	1:1	3:0	---	---	---	---	---	0:18	11:10	17:32	29:30	16:4
	V	25:52	26:36	22:43	11:28	8:30	18:6	2:3	0:2	0:4	2:3	4:13	36:19

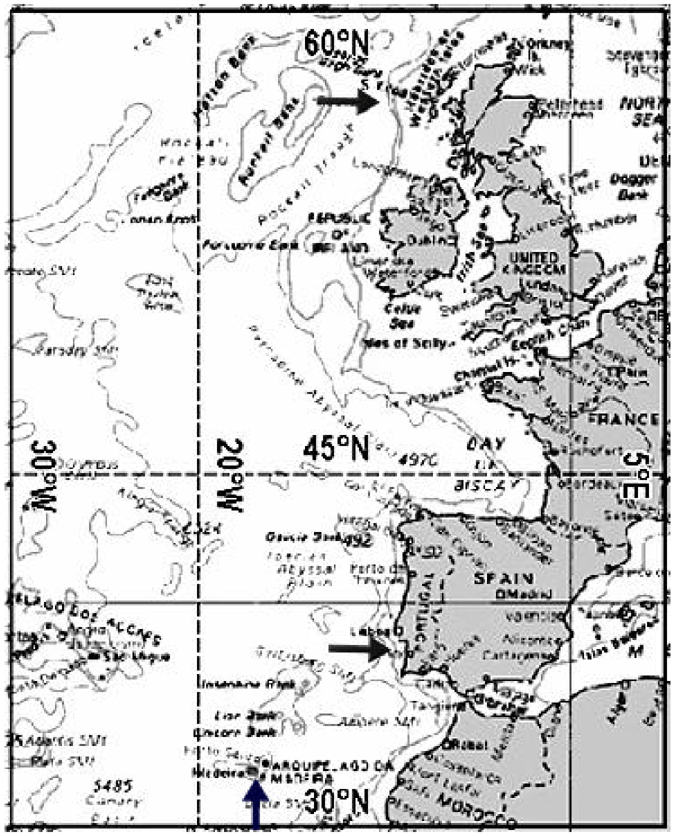


Fig. 1. NE Atlantic areas (black arrows) from where black scabbardfish samples were obtained.

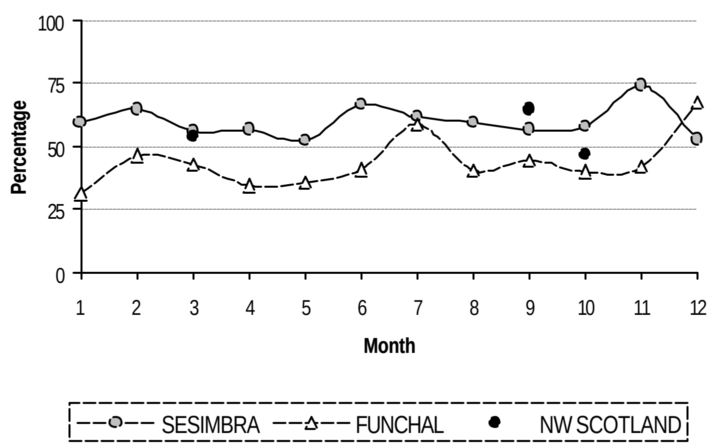


Fig. 2. Sex-ratio (females/total) values of black scabbardfish by month at each of the three sampled areas.

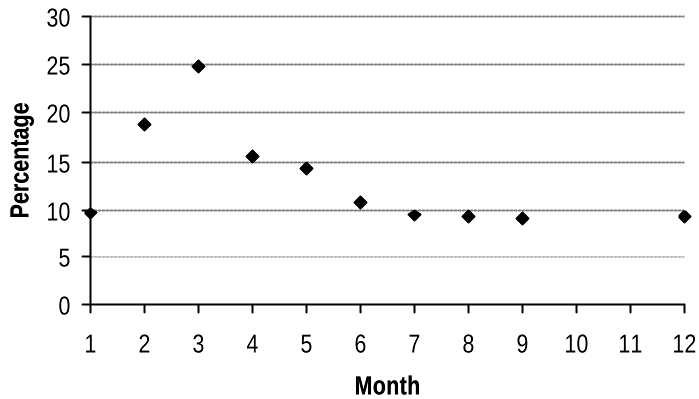
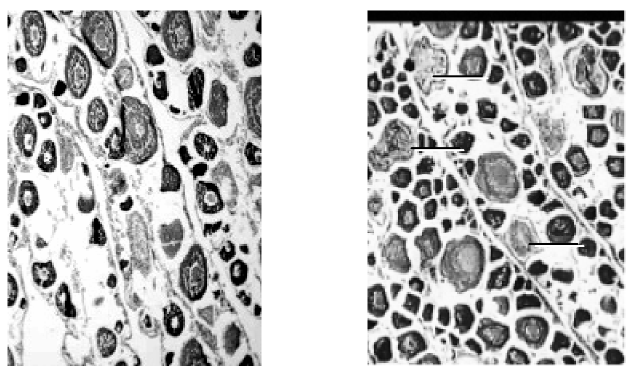


Fig. 3. Monthly relative percentage of atresia in ovaries of black sccabardfish from Sesimbra.



Funchal Sesimbra

Fig. 4. Histological sections of stage II ovaries from a specimen sampled in Funchal and Sesimbra. In the section from Sesimbra some atretic oocytes are indicated by an arrow (10 X Magnif.).

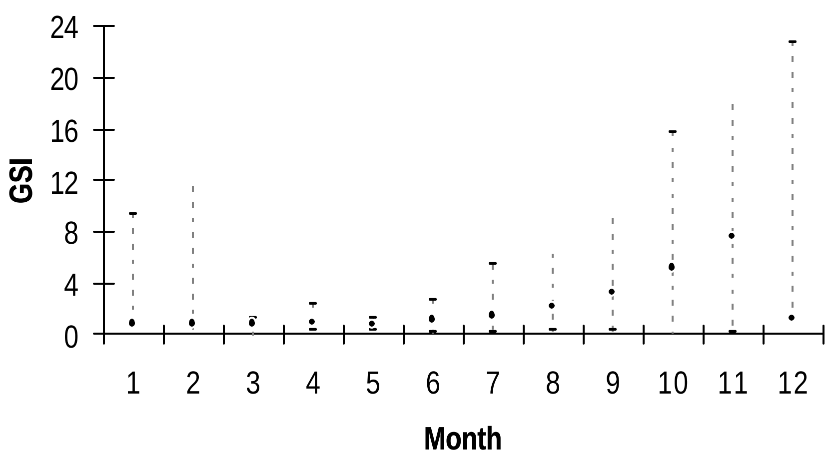


Fig. 5. Maximum (-) median (•) and minimum (-) values of GSI estimated by month for black sccabardfish sampled in Funchal.

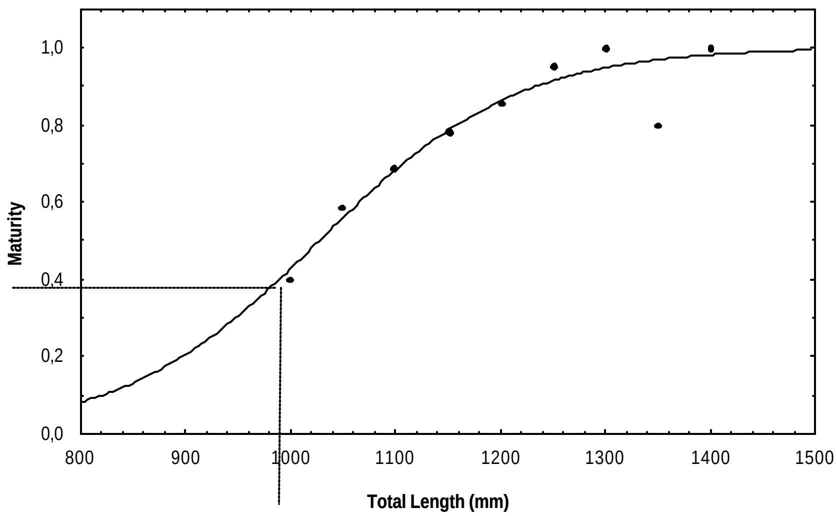


Fig. 6. Maturity ogive of black scabbardfish females sampled in Funchal for the period between September and February.

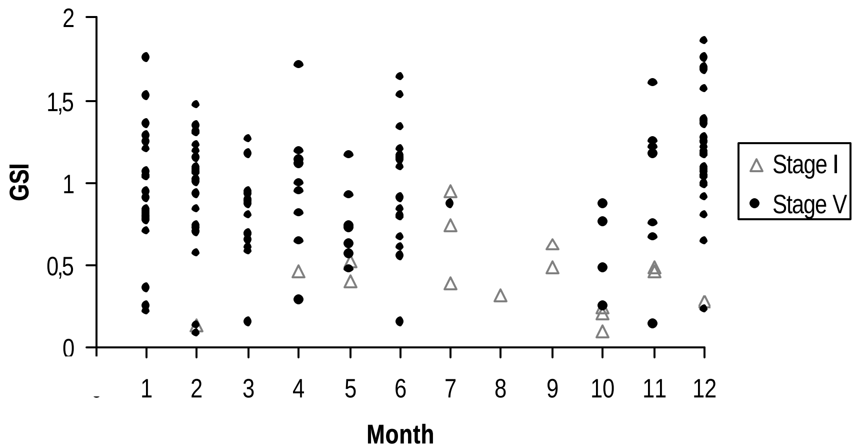
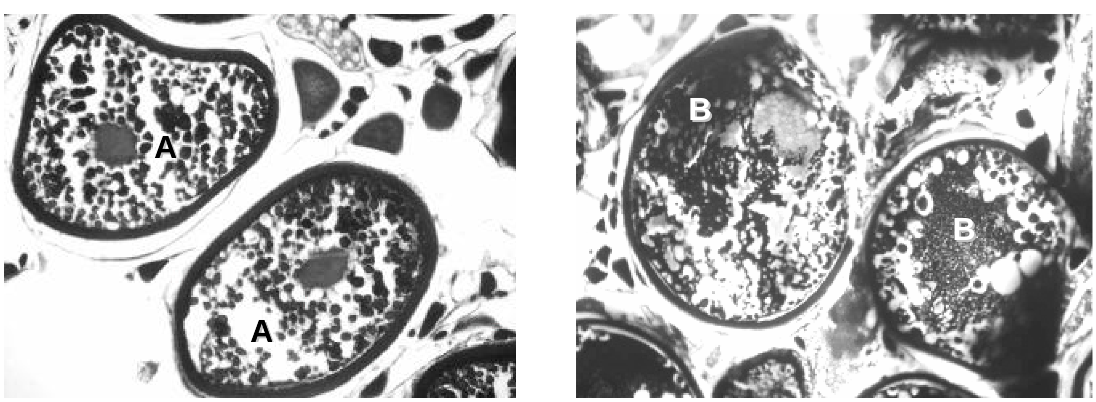


Fig. 7. Distribution of GSI values of maturity stages I and V by month in Madeira.



Stage III

Stage IV

Fig. 8. Successive states of oocyte development present in the ovaries of females in maturity stage III (A) and stage IV (B) sampled in Funchal (10 X Magnif.).

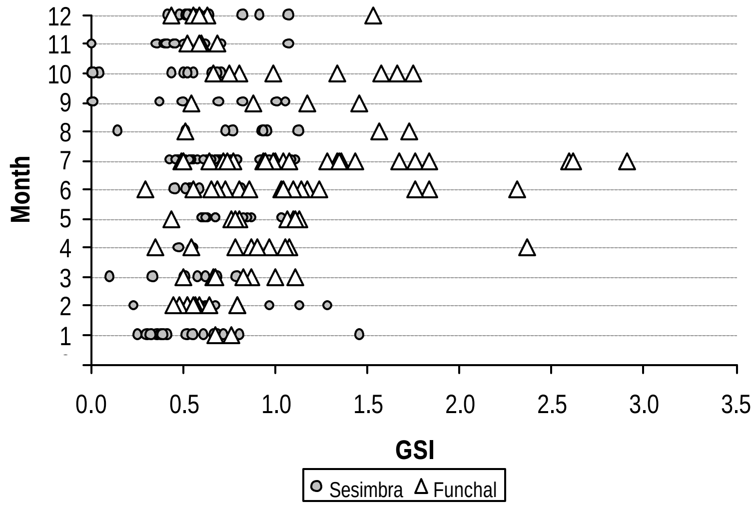


Fig. 9. Monthly distribution of stage II GSI values of females with TL between 1100 and 1210 mm sampled in Sesimbra and Funchal.

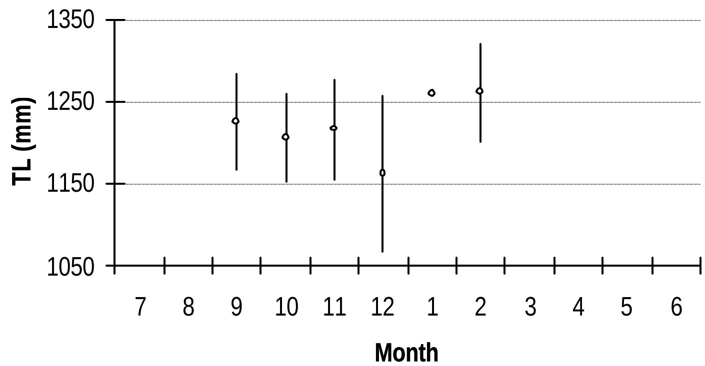


Fig. 10. Monthly variation (Mean+/-Std. deviation) of total length for spawners from Funchal.

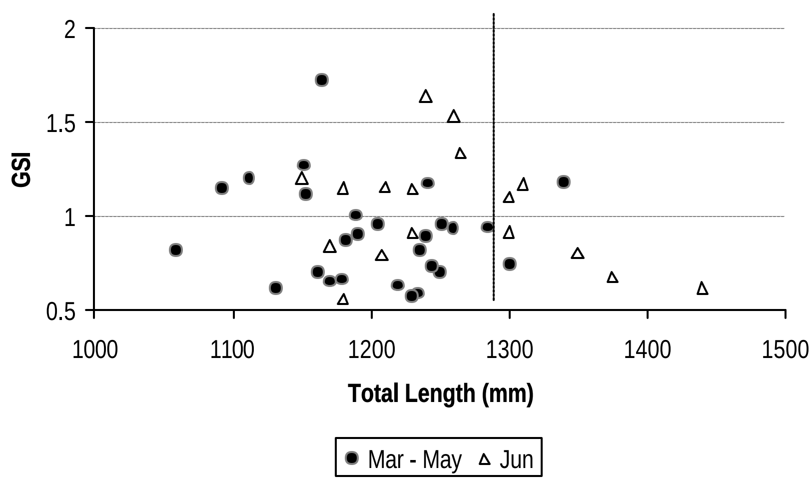


Fig. 11. Variation of GSI values *versus* total length of stage V specimens.