# Ecology of the whiskered sole in the Sado Estuary, Portugal

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The abundance of the wiskered sole *Monochirus hispidus* in the Sado Estuary showed a marked seasonal pattern, the highest densities occurred in spring and summer. The density of this species was mainly related, non-linearly, to depth, water transparency and percentage of gravel, fine sand and mud in the sediment. *Monochirus hispidus* fed mainly on crustaceans and polychaets. Significant differences were found between the proportions of prey items eaten and total length class; larger fish consumed more decapods and polychaets. The von Bertalanffy growth equation coefficients differed between sexes. The asymptotic length  $L_{\infty}$  obtained for females was higher compared to males, while the growth coefficient (*K*) was higher for males. Individuals with mature gonads were found between March and September, but the main spawning period of *M. hispidus* in the Sado Estuary was from March to June. © 2004 The Fisheries Society of the British Isles

Key words: distribution patterns; feeding; growth; Monochirus hispidus; sexual cycle; Soleidae.

# **INTRODUCTION**

The whiskered sole *Monochirus hispidus* Rafinesque occurs from Ghana to Portugal and in the Mediterranean (Quéro *et al.*, 1986; Desoutter, 1997). It inhabits coastal waters from 10 to 250 m depth in muddy and sandy bottoms and is also found in estuaries (Quéro *et al.*, 1986). The knowledge on *M. hispidus* ecology is extremely limited and includes a study on feeding and growth in the Mediterranean (Cau & Deiana, 1980) and distribution and abundance patterns in the Sado Estuary (Cunha, 1994; Cabral, 2000*a*).

On the Portuguese coast, this species has been reported south to  $39^{\circ}$  N and mainly in estuarine systems, *i.e.* in Lagoa de Albufeira (P. L. Cunha & M. M. Antunes, pers. comm.), in the Sado Estuary (Cunha, 1994; Cabral, 2000*a*) and in the Ria Formosa (Monteiro, 1989). This species is particularly abundant in the Sado Estuary where it is one of the most important fish species, being reported in densities >100 individuals  $1000 \text{ m}^{-2}$  in some areas (Cabral, 2000*a*).

Due to its high densities within the Sado Estuary (Cunha, 1994; Cabral, 2000*a*) and to its absence or extremely low abundance in the adjacent coastal areas (Henriques *et al.*, 1999; Cabral *et al.*, 2000), Cabral (2000*a*) suggested that

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*M. hispidus* could be a resident species in the Sado Estuary. Since eggs and larval stages, however, have not been caught in abundance within the estuary (Duarte, 1994), this is difficult to confirm (Cabral, 2000a).

The Sado Estuary has a high species richness of Pleuronectiformes [18 of the 25 flatfish species that have been reported for the Portuguese coast (Cabral, 2002) occur in the Sado Estuary], including the occurrence of rare species of the Portuguese coast (Cabral, 2000*a*, 2002). Also, along the Sado Estuary latitudinal range, several species from the warm-temperate and cool-temperate Atlantic biogeographical regions have their northern or southern distribution limits, respectively (Cabral, 2000*a*, 2002).

The aim of the present work was to study the main aspects of the ecology of M. *hispidus*, namely distribution and abundance patterns and their relationships with environmental factors, feeding ecology, growth and sexual cycle, in the Sado Estuary, near the northern distribution limit for this species.

# MATERIAL AND METHODS

#### STUDY AREA

The Sado Estuary is located on the west coast of Portugal ( $38^{\circ}28'$  N;  $8^{\circ}50'$  W) and has an area of *c*. 180 km<sup>2</sup>. It is strongly influenced by the sea, and has a large bay in the lower part, and a relatively narrow channel upstream. The estuary opens into a larger bay limited on its northern boundary by rocky shores and on its southern boundary by sandy beaches. In the middle part, the estuary is divided longitudinally by sand banks which influence the estuarine circulation; the water exchange is more intense in the south channel. The Sado River flow varies significantly both seasonally and annually with an annual average flow of  $10 \text{ m}^3 \text{ s}^{-1}$  (unpubl. data).

Although  $\bar{depths} > 25 \text{ m}$  can be found near the mouth of the estuary, the majority of the estuarine area is <10 m deep. The tidal regime is primarily semi-diurnal with a range of 1.5 m in neap tides and 3.5 m in spring tides. Northerly winds which favour upwelling occur in summer (Fiúza *et al.*, 1982) and strong south-west winds are common in winter months. The area is relatively protected from the dominant north-west swell of the Portuguese coast, although it is exposed to seasonal south-west storms.

In the lower part of the estuary, sediment is mainly composed of sand, while more upstream the percentage of mud in the sediment is high. In the narrow channel upstream, sand and gravel are the main components of the sediment.

The Sado Estuary has long been subjected to industrial development, urbanization and port and fishing activities. Important industrial complexes such as chemical, petrochemical, food and smelting are also present. The upper part is bordered by land used intensively for agriculture. The fisheries activity within the estuary is mainly directed to the capture of cuttlefish *Sepia officinalis* and soles *Solea solea* (L.) and *Solea senegalensis* Kaup, and is usually performed using gillnets.

# SAMPLING PROCEDURES AND DATA ANALYSIS

Fish were sampled monthly, from May 2001 to April 2002, in 20 sampling areas throughout the estuary (Fig. 1), using a 4m beam trawl with one tickler chain and 10 mm mesh size. Sampling was only performed in the middle and lower estuary, since fish abundance in the eastern arm of the Sado Estuary is quite low and *M. hispidus* has never been reported in that part of the estuary (only species with freshwater affinities occur). Trawls were towed for 15 min, during daylight, at low water on spring tides. The distance covered by each trawl was calculated using a GPS. Immediately before and after each tow, depth, temperature, salinity and dissolved oxygen concentration were



FIG. 1. Location of the sampling areas (1-20) within the Sado Estuary.

measured with an YSI 600 XLM probe near the surface (0.5 m depth), and water transparency was measured with a Secchi disk. Substratum samples were taken during each tow using a van Veen grab for the granulometric analysis of the sediment. At the laboratory, these samples were washed to remove the fine portion (<0.063 mm), dried at 70° C and then sorted in a series of sieves. For data analysis three sediment components were considered according to grain size: gravel, >2.000 mm diameter; large sand, from 2.000 to 0.500 mm; medium sand, from 0.500 to 0.150 mm; fine sand, from 0.150 to 0.063 mm; mud, <0.063 mm. For the other environmental variables data were averaged per sampling site and month.

To study the diel variation of feeding activity additional samples were taken at *c*. 2 h intervals for 24 h, on two different dates (July 2001 and May 2002), at sampling area 20 (Fig. 1), where the highest abundance values of *M. hispidus* were recorded.

All fish caught were identified, counted, measured (total length,  $L_{\rm T}$ , to 1 mm) and weighed (wet mass to 0.01 g). The digestive tract was removed and its contents preserved in 4% buffered formalin for later identification. Only the contents of the first half of the digestive tract were considered in order to avoid overestimation of prey with exoskeletons or other hard structures. The gonads and eviscerated mass were also weighed. The sagittal otoliths were extracted and dried for later ageing of the fish.

#### DISTRIBUTION AND DENSITY PATTERNS

Relationships between *M. hispidus* densities and environmental factors were analysed by non-parametric multiple regression (locally weighted regression) (Venables & Ripley, 1994; Ryan, 1997). The significance of the additive model terms was tested by nonparametric *F* statistics (Venables & Ripley, 1994). All pairwise correlations of environmental variables were determined *a priori*, and whenever a correlation >0.8 was obtained, one of the variables were excluded from the additive model, in order to avoid multicollinearity (only large sand and medium sand were strongly correlated and, thus, large sand was excluded from the model).

#### FEEDING

The stomach contents of 458 individuals of *M. hispidus* were analysed. Each prey item was identified to the lowest taxonomic level possible, counted and weighed (wet mass to 0.001 g). The relative importance of each prey item in the diet was evaluated by the numerical  $(I_N)$ , occurrence  $(I_O)$  and gravimetric  $(I_G)$  indices (Hyslop, 1980). Feeding activity was evaluated by the vacuity index  $(I_V)$  defined as the per cent of empty stomachs and by the fullness index  $(I_F)$  expressed as the percentage of the mass of the food in the stomachs in relation to the eviscerated mass of the fish (Hyslop, 1980).

In order to study diet variation with fish size, three length classes were considered:  $\leq$ 70, from 70 to 100 and >100 mm  $L_{\rm T}$ . The  $L_{\rm T}$  of the fish captured varied between 39 and 131 mm. Differences in the number of prey per main food item were tested using the *G*-test of independence (Sokal & Rohlf, 1995). A significance level of 0.05 was used.

#### GROWTH

Age was evaluated using otoliths. For each specimen, two counts of the otolith's annuli were made under a dissecting microscope. Whenever the two readings of the same otolith resulted in different age estimates the data were not considered for further analysis.

Estimates of theoretical growth in length were obtained by fitting length-at-age data to the von Bertalanffy growth equation, *i.e.*  $L_T = L_{\infty} (1 - e^{-k(t-t_0)})$  where  $L_T$  is the total length,  $L_{\infty}$  is the asymptotic total length, k is the growth coefficient and  $t_0$  is the theoretical age at zero length. The growth coefficients of this model were estimated iteratively using the least squares method in Statistica software. This analysis was performed separately for males and females.

#### **REPRODUCTIVE CYCLE**

Gonads were observed macroscopically and a maturation stage was given to each individual, according to the scale: I, immature; II, developing; III, spawning; IV, post-spawning (Cabral, 1998). For each month, the percentage of fish in stages II, III and IV was determined.

#### RESULTS

# VARIATION OF ENVIRONMENTAL FACTORS IN THE SAMPLING AREAS

For the majority of the environmental variables considered in the present study, there was a longitudinal pattern in salinity, dissolved oxygen, depth and water transparency, which had higher values at sampling sites near the estuary mouth, while temperature increased from downstream to upstream areas (although the variability near the mouth was extremely high) (Table I). Sediment in contrast was heterogeneous throughout the study area (Table I).

#### DISTRIBUTION AND DENSITY PATTERNS

The distribution and abundance patterns of *M. hispidus* varied according to season. In autumn and winter abundance was low (mean density of 3.7 and 3.3 individuals  $1000 \text{ m}^{-2}$ , respectively), and *M. hispidus* mainly occurred in the

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	1	Vater physic	o-chemical ch	aracteristics				Sediment comp	osition		
Sampling station	Temperature (°C)	Salinity	Dissolved oxygen $(mg l^{-1})$	Depth (m)	Water transparency (m)	Gravel (%)	Large sand (%)	Medium sand (%)	Fine sand (%)	0%) (%)	Organic matter (%)
-	$19.21 \pm 3.37$	$34.55 \pm 0.91$	$5.21 \pm 0.43$	$9.81 \pm 1.06$	$2.07 \pm 0.79$	$2.69 \pm 1.23$	$5.42 \pm 3.22$	$9.09 \pm 3.52$	$31 \cdot 77 \pm 28 \cdot 84$	$51.03 \pm 6.86$	$0.08\pm0.01$
6	$17.3 \pm 4.03$	$34.07 \pm 1.39$	$5.21 \pm 0.32$	$5 \cdot 88 \pm 3 \cdot 18$	$2 \cdot 10 \pm 0 \cdot 73$	$4 \cdot 79 \pm 2 \cdot 27$	$67 \cdot 13 \pm 65 \cdot 65$	$24.04 \pm 24.56$	$0.90\pm0.55$	$3 \cdot 14 \pm 1 \cdot 30$	$0{\cdot}04\pm0{\cdot}01$
б	$18.45 \pm 3.21$	$34.52\pm0.98$	$5.07 \pm 0.48$	$9.93 \pm 1.57$	$1.91 \pm 0.81$	$4.72 \pm 2.46$	$35.24 \pm 35.40$	$29.49 \pm 23.99$	$14 \cdot 1 \pm 5 \cdot 23$	$16.45\pm10.78$	$0.04\pm0.01$
4	$17.53 \pm 4.23$	$33.98 \pm 1.64$	$5 \cdot 19 \pm 0 \cdot 33$	$3.99 \pm 2.10$	$1.79\pm0.77$	$1.91 \pm 1.80$	$57 \cdot 72 \pm 48 \cdot 68$	$32.01 \pm 26.74$	$1.22 \pm 1.12$	$7.13 \pm 0.25$	$0{\cdot}06\pm0{\cdot}02$
5	$18 \cdot 22 \pm 3 \cdot 36$	$34.42 \pm 1.31$	$5 \cdot 17 \pm 0 \cdot 47$	$9.76 \pm 1.10$	$1.71 \pm 0.82$	$4.67 \pm 3.34$	$16.11 \pm 4.97$	$39.69\pm14.4$	$22.24 \pm 5.62$	$17.28 \pm 7.79$	$0{\cdot}01\pm0{\cdot}01$
9	$17.76 \pm 4.25$	$33.8 \pm 1.94$	$5.16 \pm 0.41$	$5.93 \pm 2.34$	$1.88\pm0.52$	$3.93\pm0.05$	$18.54 \pm 15.48$	$14.46 \pm 12.17$	$7.49 \pm 1.87$	$55.58 \pm 54.51$	$0{\cdot}06\pm0{\cdot}02$
7	$17.74 \pm 4.58$	$33.25 \pm 1.8$	$4.98\pm0.36$	$4{\cdot}38\pm1{\cdot}33$	$1.87\pm0.94$	$2.39 \pm 1.78$	$3 \cdot 30 \pm 1 \cdot 83$	$3.72 \pm 1.34$	$12.86 \pm 4.14$	$77.74 \pm 50.75$	$0.06\pm0.01$
8	$18.39 \pm 4.2$	$33 \cdot 30 \pm 1 \cdot 82$	$5.06 \pm 0.38$	$10.63 \pm 1.12$	$1.91 \pm 0.73$	$24 \cdot 24 \pm 1 \cdot 11$	$16.93 \pm 16.35$	$311 \cdot 17 \pm 1 \cdot 48$	$4.77 \pm 3.32$	$42.89\pm40.25$	$0{\cdot}05\pm0{\cdot}02$
6	$18 \cdot 11 \pm 4 \cdot 78$	$33.28 \pm 1.93$	$5.02 \pm 0.39$	$4{\cdot}48\pm0{\cdot}89$	$1.72 \pm 0.76$	$28 \cdot 27 \pm 21 \cdot 77$	$26 \cdot 62 \pm 3 \cdot 84$	$26.61 \pm 3.35$	$8.92 \pm 0.32$	$9.57 \pm 8.77$	$0.01\pm0.01$
10	$18.07 \pm 4.65$	$33.05 \pm 1.82$	$5.01 \pm 0.45$	$7.97 \pm 1.76$	$1.64 \pm 0.62$	$22.67 \pm 11.45$	$3 \cdot 29 \pm 1 \cdot 65$	$3.77 \pm 2.42$	$21 \cdot 90 \pm 1 \cdot 4$	$48.37 \pm 18.44$	$0{\cdot}10\pm0{\cdot}02$
11	$18.33 \pm 5.05$	$32.88 \pm 2.56$	$4.97 \pm 0.41$	$4.79 \pm 1.35$	$1.68\pm0.84$	$0.21\pm0.01$	$0.79 \pm 0.70$	$0.76\pm0.50$	$3.53 \pm 2.14$	$94.72\pm9.78$	$0.13\pm0.06$
12	$18.13 \pm 5.11$	$32.61 \pm 2.80$	$5.12 \pm 0.46$	$4.62 \pm 2.05$	$1.51 \pm 0.68$	$3.58 \pm 3.34$	$26{\cdot}99\pm12{\cdot}35$	$14.52 \pm 5.07$	$8.06\pm6.53$	$46.85\pm41.65$	$0{\cdot}10\pm0{\cdot}01$
13	$18.25 \pm 5.11$	$32.38 \pm 2.17$	$5.03 \pm 0.41$	$3.28\pm0.73$	$1.36\pm0.67$	$1.45 \pm 1.42$	$2.28\pm0.85$	$15.25\pm8.40$	$38 \cdot 18 \pm 35 \cdot 7$	$42.83 \pm 42.62$	$0.07\pm0.02$
14	$18.27 \pm 5.12$	$32.77 \pm 3.14$	$5.00 \pm 0.41$	$4.22 \pm 1.64$	$1.49\pm0.70$	$28 \cdot 19 \pm 12 \cdot 71$	$5.21 \pm 4.69$	$4 \cdot 12 \pm 2 \cdot 47$	$13.03 \pm 2.93$	$49{\cdot}46\pm40{\cdot}98$	$0{\cdot}09\pm0{\cdot}01$
15	$18.14 \pm 4.88$	$33.07 \pm 2.54$	$4.82 \pm 0.42$	$2.97 \pm 1.16$	$1.32 \pm 0.62$	$1{\cdot}69\pm0{\cdot}07$	$8.51 \pm 7.69$	$19.38\pm9.78$	$44.79 \pm 6.22$	$25.64 \pm 7.33$	$0{\cdot}03\pm0{\cdot}01$
16	$18.28\pm5.00$	$33.22 \pm 2.38$	$4.85\pm0.46$	$4.01 \pm 1.31$	$1{\cdot}64\pm0{\cdot}84$	$22 \cdot 76 \pm 2 \cdot 60$	$14.64 \pm 14.06$	$31 \cdot 3 \pm 25 \cdot 27$	$18.35 \pm 10.29$	$12.94 \pm 2.94$	$0.05\pm0.02$
17	$18.11 \pm 5.73$	$32.51 \pm 3.15$	$4.46\pm0.38$	$3 \cdot 13 \pm 1 \cdot 81$	$0.87\pm0.46$	$8 \cdot 12 \pm 2 \cdot 99$	$26.54 \pm 24.76$	$34.03\pm24.02$	$6.82 \pm 4.85$	$24.5\pm6.03$	$0.06\pm0.01$
18	$18.11 \pm 5.66$	$32.54 \pm 2.89$	$4.65 \pm 0.31$	$3.33 \pm 1.17$	$1.05\pm0.65$	$1 \cdot 77 \pm 1 \cdot 19$	$32.53 \pm 13.93$	$38.91 \pm 23.97$	$4.96 \pm 4.19$	$21.84 \pm 2.06$	$0{\cdot}01\pm0{\cdot}01$
19	$18.08\pm5.76$	$33.85 \pm 3.48$	$4 \cdot 18 \pm 0 \cdot 57$	$3.27 \pm 1.52$	$0.63 \pm 0.30$	$0.90 \pm 0.20$	$2.76\pm0.05$	$1.42\pm0.05$	$6.05 \pm 0.97$	$88.88\pm6.45$	$0{\cdot}09\pm0{\cdot}02$
20	$17.96 \pm 5.63$	$31 \cdot 62 \pm 3 \cdot 21$	$4.37 \pm 0.38$	$2.97 \pm 0.97$	$0.70\pm0.48$	$46.8\pm47.5$	$15 \cdot 09 \pm 4 \cdot 67$	$6.08 \pm 2.38$	$4.26 \pm 1.94$	$27.77 \pm 23.15$	$0.07 \pm 0.01$

TABLE I. Mean  $\pm$  s.D. values of the environmental variables in the 20 sampling areas



FiG. 2. Monochirus hispidus mean density values throughout the Sado Estuary per season: (a) spring, (b) summer, (c) autumn and (d) winter. ○, 0; •, 0 to 5; •, 5 to 10; •, 10 to 20; •, >20 individuals 1000 m<sup>-2</sup>.

middle and lower part of the estuary (Fig. 2). In the spring and summer periods an increase in density was noticed (mean values of 9.5 and 11.9 individuals  $1000 \text{ m}^{-2}$ , respectively). In spring, individuals mainly concentrated in the middle and upper estuary, while in summer they occurred in high densities in two main areas: the lower and the uppermost sampling areas (Fig. 2).

From the results obtained from the GLM applied to *M. hispidus* density data, the most important environmental variables explaining density were depth, water transparency and percentage of gravel, fine sand and mud in the sediment (Table II). The relationships between *M. hispidus* density and these variables were not linear: the highest density values were obtained for areas with depths between 2 and 4 m, low water transparency and with a sediment with <10 or >40% of gravel, <10% of fine sand and a percentage of mud from 10 to 50% (Fig. 3).

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Term	Coefficient	d.f.	n.p.d.f.	$F_{n.p.}$	$PF_{n.p.}$
Intercept	-7.27	1			
s (temperature)	-0.33	1	3	0.38	0.77
s (salinity)	0.01	1	3	2.24	0.10
s (dissolved oxygen)	0.13	1	3	0.58	0.63
s (depth)	-0.01	1	3	3.23	0.03*
s (transparency)	0.67	1	3	4.17	0.01*
s (% gravel)	0.45	1	3	8.90	0.00*
s (% medium sand)	0.31	1	3	0.28	0.84
s (% fine sand)	0.01	1	3	4.05	0.01*
s (% mud)	0.10	1	3	5.17	0.00*
s (organic matter of the sediment)	4.01	1	3	1.33	0.28
Residual <i>deviance</i> : $1222 \cdot 71 \pm 39$ d.f.					

TABLE II. Non-parametric regression analysis results for Monochirus hispidus data

\*, significant; s (variable *i*), smooth function for variable *i*; d.f., degrees of freedom; n.p.d.f., nonparametric degrees of freedom;  $F_{n,p,}$ ; non-parametric *F* statistic;  $PF_{n,p,}$ , probability of  $F_{n,p,}$ .

#### FEEDING

The diet of *M. hispidus* was mainly composed of crustaceans and polychaets. Although the relative importance of the main prey goups varied according to the index used, the most important items were cumaceans, amphipods and polychaets (Table III). Significant differences were found between the proportion of the number of the main prey according to  $L_{\rm T}$  class (*G*-test, P < 0.05). The main differences that were noticed were a decrease in the ingestion of cumaceans followed by an increase in the importance of decapods and polychaets in the diet of larger fish (Fig. 4).

The mean value of the  $I_V$  was low ( $I_V = 5\%$ ) and was similar for all the  $L_T$  classes considered ( $I_V = 7\%$  for fish <70 mm, 5% for fish between 71 and 100 and 5% for fish >100 mm).

The diel variation of the  $I_{\rm F}$ , estimated based on the 24 h cycle sampling surveys, showed that the highest values were obtained in the period between 2000 and 0200 hours, and the lowest in the period from 1600 to 1800 hours (Fig. 5).

# AGE AND GROWTH

Among the 186 individuals collected for age determination, the otoliths from 82 females and 93 males were used. The  $L_{\rm T}$  of fish analysed varied from 51 to 128 mm. The oldest fish was age 4 years.

The von Bertalanffy growth equation coefficients differed between sexes (Fig. 6). The asymptotic length  $(L_{\infty})$  obtained for females was higher compared to males (174 and 111 mm, respectively), while the growth coefficient (*K*) estimate of males (K=0.70) was higher than that determined for females (K=0.24). The  $t_0$  estimates were -1.13 and 0.25 for females and males, respectively.



FIG. 3. The density of *Monochirus hispidus* in relation to (a) depth, (b) water transparency, and percentage of (c) gravel, (d) fine sand and (e) mud in the sediment. Note density is  $log_{10}$  transformed and the curves resulted from the locally weighted regression.

#### **REPRODUCTIVE CYCLE**

The variation of the percentage of individuals in each maturation stage per month was similar for both sexes (Fig. 7). The highest percentage of individuals at stage III (spawning) was recorded from March to June. Individuals in the post-spawning stage (stage IV) were found between June and September, for both sexes (Fig. 7).

## DISCUSSION

The distribution and density of M. *hispidus* in the Sado Estuary showed a marked seasonality. Low densities were recorded in autumn and winter and an

Index	$I_{ m N}$	I <sub>O</sub>	I <sub>G</sub>
Polychaeta	17.3	9.8	14.8
Cumacea	57.8	36.0	28.7
Mysidacea	1.0	2.5	3.3
Isopoda	3.8	8.6	13.5
Amphipoda	12.7	29.9	30.7
Decapoda	1.2	4.7	2.5
Others and unidentified	6.2	8.4	6.6

TABLE III. Numerical  $(I_N)$ , occurrence  $(I_O)$  and gravimetric  $(I_G)$  indices of the main prey items found in stomachs of *Monochirus hispidus* in the Sado Estuary

increase in abundance in spring and summer was noticed. Similar abundance variation patterns have been reported for several Soleidae species, namely *S. solea* and *S. senegalensis*, that usually use estuaries as nursery areas (Marchand, 1988; Dorel *et al.*, 1991; Jager *et al.*, 1993; Cabral & Costa, 1999). The density values reported for these species for the winter period (*c.* 0.3 individuals  $1000 \text{ m}^{-2}$ , unpubl. data) were considerably lower compared to those obtained for *M. hispidus* (*c.* 1.1 individuals  $1000 \text{ m}^{-2}$ ), at least in some estuarine areas. Therefore, this may suggest that *M. hispidus* is a resident species in the Sado Estuary. The lower and deeper part of the estuary may act as an overwintering area, since water temperature in the upper part is considerably lower compared to areas near the mouth. In spring and summer periods, the high productive



FIG.4. Relative importance of the main food items ( $\square$ , Polychaeta;  $\bigotimes$ , Cumacea;  $\square$ , Amphipoda;  $\bigotimes$ , Decapoda;  $\square$ , other and not identified) in the diet of *Monochirus hispidus* according to total length class, based on numerical ( $I_N$ ), occurrence ( $I_O$ ) and gravimetric ( $I_G$ ) indices.



FIG. 5. Mean + s.D. fullness index  $(I_F)$  values determined (in two 24 h surveys) for *Monochirus hispidus* according to the time of day.

estuarine areas bordered by saltmarshes and mudflats, mainly located in the upper part of the estuary, are intensively colonized by *M. hispidus*.

The assessment of the influence of environmental factors on fish density is rather difficult, since the relationships may be indirect (Riley *et al.*, 1981; Henderson, 1989). The densities of *M. hispidus* in the Sado Estuary were higher in the northern part, which has shallower water and higher mud content in the sediment and consequently a lower water transparency. As reported in the present study for *M. hispidus*, sediment seems to be of extreme importance in flatfish distribution, since it generally determines epi- and endobenthic prey abundance. Several authors relate Soleidae density to sediment composition. Riley *et al.* (1981) reported *S. solea* preferences for sandy and muddy sediments. For this species, Dorel *et al.* (1991) and Rogers (1992) attributed the patchy distribution within a nursery area to the sedimentary discontinuity. Cabral & Costa (1999) also noticed that *S. solea* and *S. senegalensis* abundance in the Tagus Estuary was related to sediment and prey abundance.

The diet composition described for *M. hispidus* is similar to those reported for other flatfish species, namely S. solea (Quiniou, 1978; Ramos 1981; Lagardère, 1987; Molinero & Flos, 1991; Cabral, 2000b), S. senegalensis (Cabral, 2000b), Buglossidium luteum (Risso) (Nottage & Perkins, 1983; Darnaude et al., 2001), Dicologlossa cuneata (Moreau) (Belghyti et al., 1993) and Pegusa lascaris (Risso) (Marinaro & Bouabid, 1983). Soleidae are basically night feeders consuming less mobile or sedentary prey (de Groot, 1971; Braber & de Groot, 1973). One main difference was noticed in the diet of M. hispidus when compared to these species: an extremely low consumption of bivalves. Cunha (1994), in a previous study conducted in the Sado Estuary, found that M. hispidus fed mainly on amphipods (especially gammarids) and polychaets, and he also emphasized the low importance of bivalves in the diet of this species. Cau & Deiana (1980), in the Mediterranean, identified polychaets, bivalves and echinoderms as the main prey of M. hispidus. These differences in the diet of M. hispidus according to geographical area may be due to the variety of habitats, prey availability, season and range of fish lengths analysed in these studies (Molinero & Flos, 1991; Avsar, 1994; Cabral, 2000b). Nonetheless, bivalves are quite abundant in the Sado Estuary (Rodrigues, 1992) and are eaten by several flatfishes, namely S. senegalensis (V. Amaral, unpubl. data), which contradicts



FIG. 6. von Bertalanffy growth curves fitted to length-at-age data of *Monochirus hispidus* (a) females and (b) males.

the opportunist and generalist nature of most of the flatfish species in coastal habitats (Beyst *et al.*, 1999). Several flatfish species co-occur with *M. hispidus* in the main feeding grounds of the Sado Estuary (Cabral, 2000*a*) and thus the prey selection of *M. hispidus* may be constrained in order to avoid competition with other species, namely *S. solea* and *S. senegalensis* that fed intensively on bivalves (Cabral, 2000*b*).

Diet overlap has been reported within juvenile flatfish assemblages in other geographical areas (Beyst *et al.*, 1999; Cabral, 2000*b*; Darnaud *et al.*, 2001). The existence of trophic niche overlap does not necessarily involve interspecific



FIG. 7. Monthly percentage of individuals at each maturation stage (□, developing; □, spawning; ■, post-spawning) during 2001 and 2002 for (a) females and (b) males.

competition. The main items in the diet of juvenile flatfishes in these coastal assemblages are generally the most abundant prey (Beyst *et al.*, 1999; Cabral 2000*b*; Darnaude *et al.*, 2001), which emphasize a generalist and an opportunistic utilization of these food resources (Lasiak & McLachlan, 1987; Beyst *et al.*, 1999). Furthermore, niche overlap is usually minimized by differences in time and space (Moore & Moore, 1976; Poxton *et al.*, 1983; Burke, 1995).

Stomach vacuity of *M. hispidus* was low. A wide range of mean vacuity values have been reported for other flatfish species. In studies conducted in the northeastern Atlantic and in the Mediterranean, Marinaro & Bouabid (1983), Nottage & Perkins (1983), Belghyti *et al.* (1993) and Cabral *et al.* (2002) obtained low vacuity values for a large number of flatfish species. High vacuity values (mean values between 40 and 85%) for *S. solea* and *S. senegalensis*, however, have been reported (Quiniou, 1978; Ramos, 1981; Molinero *et al.*, 1991; Cabral, 2000*b*). The high vacuity found in some of these species may be related to a high evacuation rate between the stomach and the intestine and the lack of digestion in the stomach (Lagardère, 1987).

The diel variation of the fullness index suggested that *M. hispidus* mainly fed during the night, as has been pointed for other Soleidae (de Groot, 1971; Braber & de Groot, 1973). Some authors found evidence that feeding activity of flatfishes is related to tidal cycle (Wirjoatmodjo & Pitcher, 1984; Burrows *et al.*, 1994; Cabral, 2000*b*). Although this factor was not considered in the sampling

design of the present study, the two 24 h cycle surveys were performed under different tide conditions (in July 2001, high tide occurred *c*. 2000 and 0800 hours; in May 2002, high tide was observed at 1600 and 0400 hours) and showed no differences in the estimates of  $I_{\rm F}$  according to the period of the day, suggesting that feeding intensity is independent of tidal cycle.

The growth pattern of *M. hispidus* was similar to that reported by Cau & Deiana (1980) in the Mediterranean, although length-at-age estimates determined for individuals from the Sado Estuary were higher than those obtained for *M. hispidus* on the Italian coast. A faster growth in males and a larger size for the same age in females were also recorded by Cau & Deiana (1980).

The seasonal variation of gonadal development suggests that the spawning period of M. hispidus in the Sado Estuary is from March to June. Cau & Deiana (1980) reported that the spawning season of this species in the Mediterraean was from July to September. These authors, and also Cunha (1994), indicated that the spawning season of this species may be extremely wide, which may explain the occurrence of abundance peaks of small sized individuals in late autumn and winter (Cunha, 1994; Cabral, 2000*a*) and in late summer (present study). Plankton surveys will be of particular value to find out the main periods of egg and larvae abundance, and to validate the hypothesis of the resident status for this species in the Sado Estuary.

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