

## Feeding ecology of the gobies *Pomatoschistus minutus* (Pallas, 1770) and *Pomatoschistus microps* (Krøyer, 1838) in the upper Tagus estuary, Portugal\*

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**SUMMARY:** The sand goby *Pomatoschistus minutus* (Pallas, 1770) and the common goby *Pomatoschistus microps* (Krøyer, 1838) are among the most abundant fish species in estuaries, lagoons and along the Atlantic shores of Europe. In the Tagus estuary these species are particularly important in the food web. Fish samples were taken monthly in the upper estuary, and the gut contents of a total of 431 *P. minutus* and 382 *P. microps* were analysed in order to study preferences and seasonal changes in their diets and to understand intra- and interspecific relations between these two congeneric gobies. The sand goby had no dominant prey, although mysids were particularly important in the diet of this species. Shrimps and polychaetes were also common prey. *P. microps* preferentially ingested polychaetes, with isopods, amphipods, bivalves and copepods as secondary prey items. Significant seasonal variations in the ingestion of the main prey groups of both species were obtained, which reflect variations in prey availability. During the reproductive season there were differences in the food consumed by males and females, mainly for *P. minutus*, reflecting the different activity pattern exhibited by males and females during the breeding season, since parental care is supported by the former. The smallest classes from *P. minutus* increased their niche widths as they grew. Due to the different length reached by each species, the degree of interspecific overlap decreased with increasing body size.

**Key words:** *Pomatoschistus*, feeding ecology, niche overlap, Tagus estuary, Portugal

**RESUMEN:** ECOLOGÍA ALIMENTARIA DE LOS GOBIOS *POMATOSCHISTUS MINUTUS* (PALLAS, 1770) Y *POMATOSCHISTUS MICRIPS* (KRØYER, 1838) EN LA ZONA SUPERIOR DEL ESTUARIO DEL TAJO, PORTUGAL. – El cabuxino *Pomatoschistus minutus* (Pallas, 1770) y el gobio común *Pomatoschistus microps* (Krøyer, 1838) están entre las especies más abundantes de peces de estuarios, lagunas y a lo largo de las orillas atlánticas de Europa. En el estuario del Tajo estas especies son particularmente importantes en la cadena alimentaria. Muestras de los pescados fueron tomadas mensualmente en la parte superior del estuario, y el contenido de la tripa de un total de 431 *P. minutus* y de 382 *P. microps* fue analizado con la finalidad de estudiar las preferencias y cambios estacionales en sus dietas y comprender las relaciones intra e ínter específicas entre estos dos gobios congéneres. El cabuxino no tenía ninguna presa dominante, aunque los misidáceos eran particularmente importantes en la dieta de esta especie. Los camarones y los poliuetos eran también presa común. Los *P. microps* injirieron preferencialmente poliuetos, con isópodos, anfípodos, bivalvos y copépodos como presas secundarias. Fueron obtenidas variaciones estacionales significativas en la ingestión de los grupos principales de la presa de ambas especies, lo que refleja variaciones en la disponibilidad de la presa. Durante la estación reproductiva había diferencias en el alimento consumido por los varones y las hembras, principalmente para el *P. minutus*, reflejando la diferencia en el patrón de la actividad exhibido por los machos y las hembras durante la estación de cría, puesto que el cuidado parental es llevado a cabo por los primeros. Las clases más pequeñas del *P. minutus* aumentaron el tamaño de sus nichos mientras crecían. Debido a la diversidad de longitud alcanzada por cada especie, el grado del solapamiento ínter específico disminuía con el aumento de tamaño del cuerpo.

**Palabras clave:** *Pomatoschistus*, ecología alimentaria, solapamiento de nichos, estuario del Tajo, Portugal

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## INTRODUCTION

The sand goby *Pomatoschistus minutus* (Pallas, 1770) and the common goby *Pomatoschistus microps* (Krøyer, 1838) are among the most abundant fish species in estuaries, lagoons and along the shores of Europe. Their geographic distributions range from the coast of Norway to the Gulf of Lion, in the Mediterranean, for *P. microps* (Bouchereau *et al.*, 1993) and from the coast of Norway to the west coast of the Black Sea for *P. minutus* (Bouchereau *et al.*, 1989). Despite their high abundance and importance in estuarine food webs (Costa, 1988), few studies have focused on the ecology of these species in the South of Europe and they are almost non-existent for *P. microps*. As described for several estuaries and lagoons of Europe, their role in the diet of different organisms of the Tagus estuary has been emphasised by several authors, namely for decapods, *Crangon crangon* Linnaeus and *Carcinus maenas* Linnaeus (Gaudêncio and Guerra, 1991), teleosts, *Alosa fallax* (Lacépède) (Assis *et al.*, 1992), *Anguilla anguilla* Linnaeus (Costa *et al.*, 1992), *Ciliata mustela* Linnaeus, *Gobius niger* Linnaeus, *Platichthys flesus flesus* Linnaeus, *Trigla lucerna* Linnaeus and *Trisopterus luscus* Linnaeus (Costa, 1988) and piscivorous birds such as *Egretta garzetta* Linnaeus (Moreira, 1992), *Recurvirostra avosetta* Linnaeus and *Calidris alpina* Linnaeus (Moreira, pers. comm.).

Several authors have described the feeding ecology of *P. minutus* (Fonds, 1973; Claridge *et al.*, 1985; Zander and Hagemann, 1987; Hamerlynck and Cattrijsse, 1994; Laffaille *et al.*, 1999) and *P. microps* (Magnhagen and Wiederholm, 1982a; Magnhagen, 1986; Pihl, 1985; Gee *et al.*, 1985) but only for the coastal areas of northwest Europe.

The major aim of the present work is to study the feeding ecology of *P. minutus* and *P. microps* in the Tagus estuary and to evaluate niche overlap between these sympatric species that have been described as dominant species in this estuary (Cabral *et al.*, 2001). Several studies have compared the feeding habits of the two gobies, but they were performed in laboratories (Edlund and Magnhagen, 1981; Magnhagen and Wiederholm, 1982b; Wiederholm, 1987) or in a particular time of their reproduction period (Mehner, 1992). Niche segregation may avoid competition between predators of the same trophic level. It could be expected that coexisting fish will differ in their choice of type and size of prey (Zander and Berg, 1984). The diet variation with season, fish

length and sex was also considered. Differences in the diet according to sex are expected during the breeding season, since there are considerable differences in the behaviour pattern between the two sexes (Magnhagen, 1986; Lindström and Wennström, 1994). Parental care is supported by males, which leave their nests only for short feeding bouts and to chase away intruders, so females can have a wider range for food foraging than males (Magnhagen, 1986).

## MATERIALS AND METHODS

The present study was carried out at three sites located in the upper zone of the Tagus estuary (Fig. 1), in southwest Europe.

Fish were sampled monthly during ebb tides, after sunrise during spring tides, using a 4 m beam-trawl with a 10 mm mesh size, from October 1994 to September 1995. Three trawls per site were towed for 20 min at a speed of 1 knot. A GPS (25 m precision) was used to locate sampling stations and to calculate the distance travelled for each trawl. After capture, the gobies were placed on ice to prevent *post mortem* digestion. Water temperature, salinity, turbidity and depth were measured on each sampling occasion using a Hydrolab multiprobe sounder.

In the laboratory, fish total length was measured (to the nearest 1 mm) and the sex was determined.

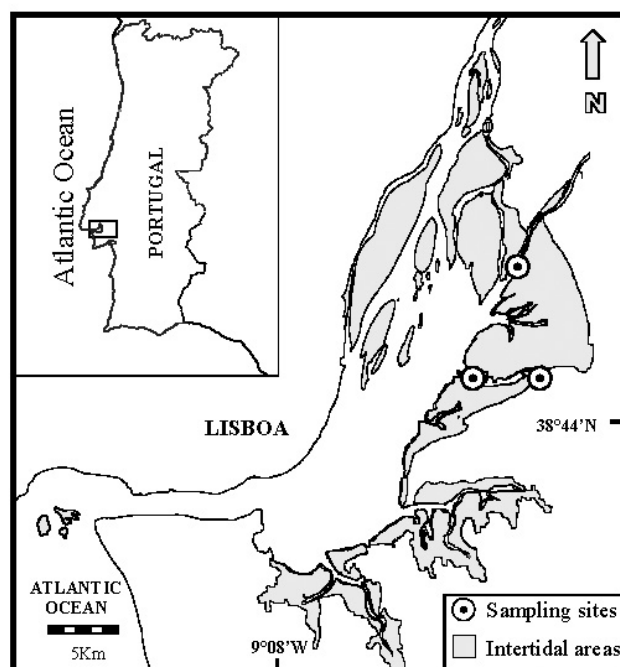


FIG. 1. – Location of the Tagus estuary and of sampling areas.

The food items found in the entire gastro-intestinal tracts (excluding the rectum) were identified to the lowest taxonomic level possible, counted and wet weighed (to the nearest 0.001 g).

The gut contents of 431 *P. minutus* and 382 *P. microps* (with total lengths ranging from 16 to 89 mm and from 22 to 55 mm respectively) were analysed. For quantitative analysis of the diet, numeric ( $N_i$ ), gravimetric ( $W_i$ ) and occurrence ( $O_i$ ) frequencies were used (Hyslop, 1980), as well as an adaptation of a mixed method proposed by Hureau (1970), the dietary coefficient ( $Q_i$ ), defined as  $Q_i = \%N_i \times \%W_i$ . This method, which considers both the weight and the number of ingested prey, classifies them into preferential, secondary and accidental categories. In the present paper another terminology has been used, namely dominant ( $Q \geq 200$ ), secondary ( $200 > Q \geq 20$ ) and unimportant ( $Q < 20$ ).

The fullness index (Fi) was defined as  $Fi = g_i / m_i$  (Hyslop, 1980), where  $g_i$  is the mass of the gastro-intestinal tract content of the individual  $i$  and  $m_i$  is the mass of the individual  $i$ . The functional relationships between fullness index (dependent parameter) and simultaneous water temperature, salinity, turbidity and depth (independent parameters) were analysed by stepwise multiple regressions (Zar, 1984). To study the relations between feeding activity (fullness index) and fish density (ind.  $m^{-2}$ ), a Spearman correlation (Zar, 1984) was used.

In order to determine whether the consumption of the different prey groups was dependent on the seasons, length classes or the sex of the individuals, the G-test of independence (Sokal and Rohlf, 1981; Zar, 1984) with a 0.05 significance level was performed, using the numeric frequencies of the major prey groups. For this purpose, monthly samples were divided into 4 seasons (Autumn = September-November; Winter = December-February; Spring = March-May and Summer = June-August), and species were divided into 4 mm and 8 mm size classes, for *P. microps* (mI = < 32 mm, mII = 33-36 mm, mIII = 37-40 mm, mIV = 41-44 mm and mV = > 45 mm) and *P. minutus* (MI = < 44, MII = 45-52 mm, MIII = 53-60 mm, MIV = 61-68 mm and MV = > 69 mm) respectively. During the reproductive period reported for these species, which in the Tagus estuary according to Moreira (1989) extends from January to May, data was grouped according to sex and comparatively analysed.

Intra- and interspecific food niche overlap was evaluated using Schoener's index (SI), defined as

$$SI_{xy} = 1 - 0.5 \left( \sum_{i=1}^n |P_{xi} - P_{yi}| \right),$$

where  $P_{xi}$  is the relative frequency of prey category  $i$  in the digestive tract of species  $x$  and  $P_{yi}$  the same frequency in species  $y$  (Hurlbert, 1978). Both numerical ( $N_i$ ) and gravimetric ( $W_i$ ) measures of diet were used in the calculation of this index. According to Wallace (1981) and Wallace and Ramsey (1983), overlap values  $> 0.6$  should be considered as biologically significant.

Trophic niche width was calculated for both sexes using Shannon-Weaver's diversity index with the numerical frequencies ( $N_i$ ) of the different prey items. To test the statistical significance of these results, an approximation to the t-test (Hutcheson, 1970) was used. For all test procedures a significance level of 0.05 was considered.

## RESULTS

According to the results of the dietary coefficient ( $Q$ ) (Table 1), *P. minutus* has no dominant prey. Nevertheless, mysids were a particularly important prey with an  $Q_i = 0.40$  and a coefficient of 179.66 (Fig. 2), showing a similar importance

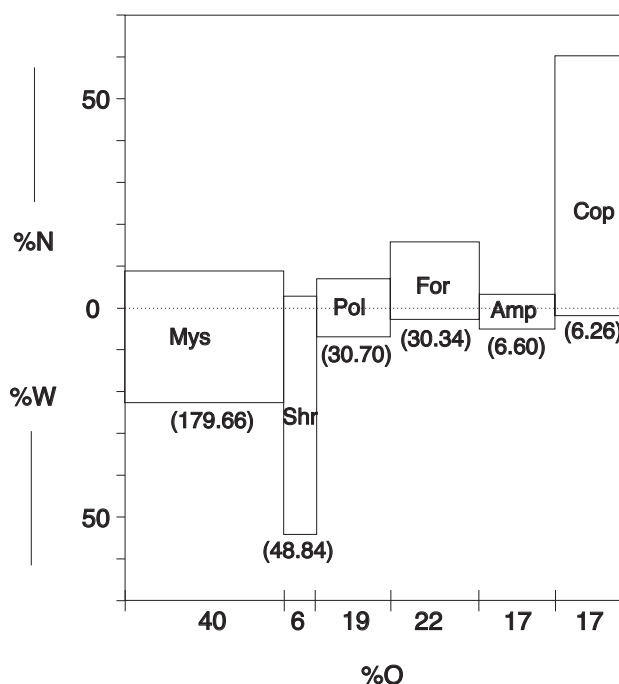


FIG. 2. – Numerical (N), gravimetric (W) and occurrence (O) percentages of the most important prey groups included in the diet of *P. minutus*. Hureau's dietary coefficient ( $Q_i$ ) between brackets. (Amp: amphipods; Cop: copepods; Mys: mysids; Shr: shrimps; For: foraminifers; Pol: polychaetes).

TABLE 1. – Numerical ( $N_i$ ), gravimetric ( $W_i$ ) and occurrence ( $O_i$ ) frequencies and dietary coefficient ( $Q_i$ ) values of the prey found in *P. minutus* and *P. microps* gut contents in the upper Tagus estuary (n.i.: not identified).

Species Prey	<i>P. minutus</i>				<i>P. microps</i>			
	$N_i$	$W_i$	$O_i$	$Q_i$	$N_i$	$W_i$	$O_i$	$Q_i$
<b>Foraminifera (total)</b>	<b>0.15</b>	<b>0.02</b>	<b>0.22</b>	<b>30.34</b>	<b>0.06</b>	<b>&lt;0.01</b>	<b>0.23</b>	<b>0.22</b>
<b>Nematoda (total)</b>	<b>&lt;0.01</b>	<b>&lt;0.01</b>	<b>0.01</b>	<b>&lt;0.01</b>	<b>&lt;0.01</b>	<b>&lt;0.01</b>	<b>0.01</b>	<b>&lt;0.01</b>
<i>Peringia ulvae</i>	<0.01	<0.01	0.01	-	<0.01	<0.01	0.02	-
Opisthobranchia	-	-	-	-	<0.01	<0.01	0.01	<0.01
<b>Gastropoda (total)</b>	<b>&lt;0.01</b>	<b>&lt;0.01</b>	<b>0.01</b>	<b>&lt;0.01</b>	<b>&lt;0.01</b>	<b>&lt;0.01</b>	<b>0.02</b>	<b>&lt;0.01</b>
<i>Scrobicularia plana</i>	<0.01	0.01	0.02	-	0.01	0.03	0.04	-
Siphons	0.01	0.02	0.05	-	0.01	0.19	0.10	-
<b>Bivalvia (total)</b>	<b>0.02</b>	<b>0.02</b>	<b>0.07</b>	<b>3.16</b>	<b>0.02</b>	<b>0.22</b>	<b>0.13</b>	<b>44.22</b>
<i>Nephtys</i> sp.	<0.01	0.01	0.01	-	<0.01	0.16	0.01	-
<i>Hediste diversicolor</i>	<0.01	0.01	0.01	-	<0.01	0.11	0.01	-
<i>Glycera convoluta</i>	<0.01	<0.01	<0.01	-	<0.01	0.01	<0.01	-
<i>Streblospio shrubsolei</i>	0.05	0.01	0.13	-	0.06	0.07	0.17	-
Cirratulidae n.i.	<0.01	<0.01	0.02	-	<0.01	<0.01	0.01	-
Polychaeta n.i.	<0.01	0.01	0.03	-	<0.01	0.02	0.03	-
<b>Polychaeta (total)</b>	<b>0.06</b>	<b>0.05</b>	<b>0.19</b>	<b>30.70</b>	<b>0.07</b>	<b>0.37</b>	<b>0.23</b>	<b>262.53</b>
<b>Oligochaeta (total)</b>	<b>0.01</b>	<b>&lt;0.01</b>	<b>0.03</b>	<b>0.44</b>	<b>0.03</b>	<b>0.03</b>	<b>0.07</b>	<b>8.35</b>
<b>Acaridae (total)</b>	<b>-</b>	<b>-</b>	<b>-</b>	<b>-</b>	<b>&lt;0.01</b>	<b>&lt;0.01</b>	<b>0.01</b>	<b>&lt;0.01</b>
<b>Ostracoda (total)</b>	<b>0.01</b>	<b>&lt;0.01</b>	<b>0.03</b>	<b>&lt;0.01</b>	<b>0.01</b>	<b>&lt;0.01</b>	<b>0.06</b>	<b>0.02</b>
<i>Neomysis integer</i>	0.08	0.21	0.39	-	0.03	0.10	0.17	-
Mysids n.i.	<0.01	<0.01	0.01	-	<0.01	<0.01	0.01	-
<b>Mysidacea (total)</b>	<b>0.09</b>	<b>0.21</b>	<b>0.40</b>	<b>179.66</b>	<b>0.03</b>	<b>0.10</b>	<b>0.17</b>	<b>71.38</b>
Calanoida	0.01	<0.01	0.03	-	0.01	<0.01	0.01	-
Harpacticoida	0.59	<0.01	0.17	-	0.67	<0.01	0.38	-
<b>Copepoda (total)</b>	<b>0.60</b>	<b>&lt;0.01</b>	<b>0.17</b>	<b>6.26</b>	<b>0.68</b>	<b>&lt;0.01</b>	<b>0.38</b>	<b>23.09</b>
<b>Cumacea (total)</b>	<b>&lt;0.01</b>	<b>&lt;0.01</b>	<b>&lt;0.01</b>	<b>&lt;0.01</b>	<b>&lt;0.01</b>	<b>&lt;0.01</b>	<b>0.02</b>	<b>&lt;0.01</b>
<i>Paragnathia formica</i>	0.02	0.01	0.10	-	0.03	0.04	0.15	-
<i>Sphaeroma monodi</i>	<0.01	0.02	0.03	-	0.01	0.05	0.03	-
<i>Eurydice pulchra</i>	<0.01	<0.01	0.01	-	<0.01	<0.01	0.01	-
<i>Porcellio pruinosus</i>	<0.01	<0.01	<0.01	-	-	-	-	-
<i>Idotea chelipes</i>	-	-	-	-	<0.01	<0.01	<0.01	-
Isopoda n.i.	<0.01	<0.01	0.01	-	<0.01	<0.01	0.01	-
<b>Isopoda (total)</b>	<b>0.02</b>	<b>0.03</b>	<b>0.12</b>	<b>5.28</b>	<b>0.04</b>	<b>0.09</b>	<b>0.18</b>	<b>36.35</b>
<i>Amphitoe ferox</i>	-	-	-	-	<0.01	<0.01	<0.01	-
<i>Talorchestia brito</i>	-	-	-	-	<0.01	<0.01	<0.01	-
<i>Corophium volutator</i>	0.01	0.01	0.06	-	0.01	0.07	0.06	-
Gammaridae n.i.	0.02	0.02	0.11	-	0.01	0.06	0.09	-
<b>Amphipoda (total)</b>	<b>0.02</b>	<b>0.03</b>	<b>0.17</b>	<b>6.60</b>	<b>0.02</b>	<b>0.13</b>	<b>0.14</b>	<b>28.21</b>
<i>Crangon crangon</i>	0.01	0.47	0.06	-	<0.01	<0.01	<0.01	-
<i>C. crangon</i> (eggs)	<0.01	0.04	0.01	-	<0.01	<0.01	<0.01	-
<b>Caridea (total)</b>	<b>0.01</b>	<b>0.52</b>	<b>0.06</b>	<b>48.84</b>	<b>&lt;0.01</b>	<b>&lt;0.01</b>	<b>&lt;0.01</b>	<b>&lt;0.01</b>
Homoptera	-	-	-	-	<0.01	<0.01	<0.01	-
Chironomidae	<0.01	0.01	0.01	-	0.01	0.01	0.02	-
Dolichopodidae	<0.01	0.02	0.01	-	0.01	0.02	0.03	-
Coleoptera	<0.01	<0.01	<0.01	-	-	-	-	-
Insecta n.i.	<0.01	<0.01	<0.01	-	<0.01	0.01	0.06	-
<b>Insecta (total)</b>	<b>&lt;0.01</b>	<b>0.02</b>	<b>0.02</b>	<b>0.47</b>	<b>0.02</b>	<b>0.03</b>	<b>0.07</b>	<b>5.52</b>
<b>Crustacea n.i. (total)</b>	<b>0.01</b>	<b>0.02</b>	<b>0.04</b>	<b>6.03</b>	<b>&lt;0.01</b>	<b>&lt;0.01</b>	<b>0.03</b>	<b>0.19</b>
Clupeidae n.i.	<0.01	0.02	<0.01	-	-	-	-	-
<i>Pomatoschistus</i> spp.	<0.01	0.02	0.01	-	<0.01	0.02	0.01	-
Pisces n.i.	<0.01	0.05	0.01	-	<0.01	<0.01	<0.01	-
<b>Pisces (total)</b>	<b>&lt;0.01</b>	<b>0.08</b>	<b>0.02</b>	<b>2.20</b>	<b>&lt;0.01</b>	<b>0.02</b>	<b>0.01</b>	<b>0.21</b>

according to the numeric and gravimetric indices (Table 1). Other secondary preys were shrimps (only *Crangon crangon* (L.)), with high values of gravimetric frequency, polychaetes (mainly Spirogonidae), with identical contributions from the numerical and weight indices, and foraminifera. Despite their high numerical importance ( $N_i = 0.60$ ), copepods were considered unimportant prey.

The remaining prey occurred with frequencies near or below the coefficient value of 0.20.

Polychaetes (mainly *Hediste diversicolor* (Müller), *Nephtys* sp. and *Streblospio shrubsolei* Buchanan) were dominant prey in the diet of *P. microps*, with a coefficient of 262.53 (Fig. 3). This result was mainly due to their high importance in weight (Table 1). Secondary prey, in decreasing

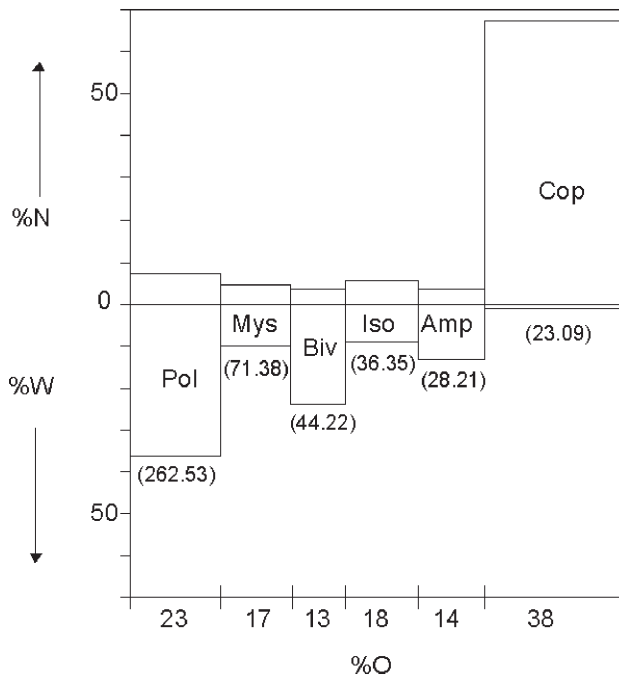


FIG. 3. – Numerical (N), gravimetric (W) and occurrence (O) percentages of the most important prey groups included in the diet of *P. microps*. Hureau's dietary coefficient ( $Q_1$ ) between brackets. (Iso: isopods; Amp: amphipods; Cop: copepods; Mys: mysids; Pol: polychaetes; Biv: bivalves).

order of importance, were mysids, bivalves (exclusively siphons of *Scrobicularia plana* (da Costa)), isopods (mainly *Paragnathia formica* (Hesse)) and amphipods (mainly *Corophium volutator* (Pallas)), with similar contributions from the numerical and

weight components. Copepods were also a secondary prey, mainly due to the high value of the numerical index. This was also the most frequent ingested prey in the diet of *P. microps*, followed by foraminifera and polychaetes.

Significant seasonal variations in the consumption of the main major prey items were observed for both species (Table 2), with Autumn and Summer showing no statistical differences in most of the cases. During these periods, the consumption of amphipods and isopods by *P. minutus* was very high, while there was a reduction in the ingestion of copepods. Mysids were mostly consumed by both gobies during Summer.

The importance of bivalves, mysids and oligochaetes in the diet of *P. microps* (Table 2) was significantly higher in summer. The consumption of copepods was higher in winter, while polychaetes were mainly ingested during spring and summer.

Stepwise multiple regression analysis showed that water temperature ( $x$ ) was, among the measured physical parameters, the only predictor of fullness index for both species ( $y = 0.123025x - 0.644358$  ( $r = 0.56$ ;  $n = 20$ ;  $p < 0.05$ ) for *P. minutus* and  $y = 0.196997x - 1.990352$  ( $r = 0.72$ ;  $n = 20$ ;  $p < 0.05$ ) for *P. microps*), with higher temperatures promoting more intense feeding activity.

The fullness index values determined for *P. minutus* were negatively correlated with fish density ( $r_s = -0.64$ ;  $n = 20$ ;  $p < 0.05$ ), while for *P. microps* no association was obtained ( $r_s = -0.31$ ;  $n = 20$ ;  $p > 0.05$ ).

TABLE 2. – Numerical frequencies according to season of the major food items in the diet of *P. minutus* and *P. microps*. (Iso: isopods; Amp: amphipods; Cop: copepods; Ins: insects; Mys - mysids; Shr: shrimps; For: foraminifers; Pol: polychaetes; Oli: oligochaetes; Fish: fish; Biv: bivalves; G: G-test statistic; Test result: posteriori test; \*  $p < 0.05$ ).

Species	Prey	Autumn	Winter	Spring	Summer	G	Test result
<i>P. minutus</i>	Iso	0.15	0.04	<0.01	0.03	72.9*	A S W Sp
	Amp	0.15	0.02	0.02	0.09	37.4*	A S Sp W
	Cop	0.05	0.61	0.65	0.02	240.6*	W Sp A S
	Ins	-	<0.01	<0.01	-	0.7	A W Sp S
	Mys	0.13	0.05	0.08	0.68	159.3*	S Sp A W
	Shr	0.01	0.02	<0.01	0.02	31.2*	W S A Sp
	For	0.28	0.15	0.16	0.03	20.0*	A Sp W S
	Pol	0.05	0.02	0.08	0.05	45.6*	Sp A S W
	Oli	-	0.03	<0.01	-	42.8*	Sp A S W
	Fish	-	<0.01	<0.01	-	1.9	A W Sp S
	Biv	0.05	0.03	<0.01	-	42.1*	A W S Sp
<i>P. microps</i>	Iso	0.08	0.04	0.02	0.01	24.3*	A W S Sp
	Amp	0.14	0.01	0.02	0.04	64.1*	A S Sp W
	Cop	0.58	0.81	0.47	0.07	506.4*	W A Sp S
	Ins	-	0.02	0.02	0.04	7.7	A W Sp S
	Mys	0.04	<0.01	0.05	0.31	165.3*	S Sp A W
	Shr	-	-	-	0.01	7.4	A W Sp S
	For	0.08	0.04	0.11	0.04	43.3	Sp S A W
	Pol	0.02	0.01	0.22	0.16	424.3*	Sp S A W
	Oli	-	0.02	0.04	0.12	30.2*	S Sp W A
	Fish	-	-	<0.01	-	7.7	A W Sp S
	Biv	-	<0.01	0.05	0.17	106.3*	S Sp W A

TABLE 3. – Numerical frequencies according to sex of the major food items in the diet of *P. minutus* and *P. microps*. (Iso: isopods; Amp: amphipods; Cop: copepods; Ins: insects; Mys: mysids; Shr: shrimps; For: foraminifers; Pol: polychaetes; Oli: oligochaetes; Fish: fish; Biv: bivalves; G: G-test statistic; Test result: posteriori test; \*- p < 0.05).

Species Prey	<i>P. minutus</i>			<i>P. microps</i>		
	Females	Males	G	Females	Males	G
Iso	0.02	0.01	7.4*	0.03	0.04	0.7
Amp	0.02	0.01	5.5*	0.01	0.02	2.1
Cop	0.60	0.76	70.3*	0.69	0.73	2.5
Ins	<0.01	-	3.4	0.02	0.01	2.3
Mys	0.08	0.03	25.1*	0.02	0.06	11.6*
Shr	0.01	<0.01	2.5	<0.01	-	0.1
For	0.17	0.13	7.4*	0.06	0.05	0.5
Pol	0.08	0.03	22.7*	0.09	0.02	22.1*
Oli	0.01	0.02	1.1	0.03	0.01	3.3*
Fish	<0.01	<0.01	0.2	<0.01	-	0.6
Biv	0.01	0.01	0.0	0.03	0.01	3.4

TABLE 4. – Specific richness (n), Shannon-Weaver's (H') index (variance between brackets) and evenness (J) for the diet of females and males of *P. minutus* and *P. microps* during the reproductive period. Result of the t-test, \*- p < 0.05.

Species	n	Fem		J	n	Male		J	t-test
		n	H'			n	H'		
<i>P. minutus</i>	23	0.619	(0.001)	0.455	19	0.420	(0.029)	0.329	3.689*
<i>P. microps</i>	24	0.554	(0.001)	0.402	18	0.486	(0.002)	0.387	1.153

Significant differences in the numerical frequencies of the prey ingested by males and females of *P. minutus* were observed for 6 of the 11 major prey categories (Table 3). Females consumed higher frequencies of isopods, amphipods, mysids, foraminifers and polychaetes, whereas copepods were ingested in larger number by males.

For *P. microps* (Table 3) the differences were not as marked as for the latter species, occurring only in 3 of the 11 prey groups. Males consumed a significantly higher number of mysids, whereas females consumed more polychaetes and oligochaetes than males. Shrimps and fish were only detected in female digestive tracks.

TABLE 5. – Numerical frequencies according to total length classes of the major food items in the diet of *P. minutus* and *P. microps*. (Iso: isopods; Amp: amphipods; Cop: copepods; Ins: insects; Mys: mysids; Shr: shrimps; For: foraminifers; Pol: polychaetes; Oli: oligochaetes; Fish: fish; Biv: bivalves; G: G-test statistic; Test result: posteriori test; \*- p < 0.05).

Species	Prey	MI	MII	MIII	MIV	MV	G	Test result
<i>P. minutus</i>	Iso	<0.01	0.02	0.07	0.07	0.02	55.9*	III IV V II I
	Amp	0.01	0.03	0.05	0.08	0.03	20.1*	III IV V II I
	Cop	0.73	0.62	0.17	0.02	0.03	598.2*	I II III IV V
	Ins	-	<0.01	-	-	0.02	8.9	I II III IV V
	Mys	0.05	0.11	0.18	0.16	0.09	42.3*	III IV V II I
	Shr	-	<0.01	<0.01	0.03	0.23	94.7*	V IV I II III
	For	0.14	0.13	0.37	0.23	0.26	95.9*	II I V IV III
	Pol	0.06	0.05	0.09	0.13	0.02	17.9*	I II III V IV
	Oli	-	<0.01	0.01	0.12	0.12	94.0*	IV V I II III
	Fish	-	<0.01	<0.01	0.01	0.05	64.9*	V IV III II I
	Biv	-	0.02	0.02	0.12	0.03	64.9*	IV V III II I
	<i>P. microps</i>	Iso	0.05	0.06	0.02	0.04	0.07	23.8*
Amp		0.08	0.02	0.01	0.03	0.02	28.4*	I II IV V III
Cop		0.62	0.65	0.76	0.62	0.61	75.3*	III I II IV V
Ins		0.02	0.04	0.02	0.02	0.01	7.7	I II III IV V
Mys		0.09	0.03	0.02	0.02	0.04	21.2*	I II V IV III
Shr		-	-	-	-	<0.01	3.8	I II III IV V
For		0.03	0.06	0.06	0.07	0.07	4.3	V IV III II I
Pol		0.03	0.06	0.05	0.11	0.07	36.3*	IV V III II I
Oli		0.04	0.01	0.03	0.03	0.01	16.1*	V IV I II III
Fish		-	-	<0.01	<0.01	<0.01	1.4	I II III IV V
Biv		0.01	0.02	0.01	0.02	0.05	35.8*	V IV I II III

TABLE 6. – Schoener's index performed with numeric (below x-line) and gravimetric frequencies (above x-line) for intraspecific and inter-specific food overlap between *P. minutus* (MI = < 44, MII = 45-52 mm, MIII = 53-60 mm, MIV = 61-68 mm and MV = > 69 mm) and *P. microps* (mI = < 32 mm, mII = 33-36 mm, mIII = 37-40 mm, mIV = 41-44 mm and mV = > 45 mm) size classes.

Species	classes	<i>P. minutus</i>					<i>P. microps</i>				
		MI	MII	MIII	MIV	MV	mI	mII	mIII	mIV	mV
<i>P. minutus</i>	MI	x	<b>0.600</b>	<b>0.651</b>	0.332	0.025	0.454	0.401	0.446	0.283	0.330
	MII	<b>0.875</b>	x	<b>0.659</b>	0.418	0.203	0.407	0.324	0.408	0.223	0.288
	MIII	0.440	0.545	x	0.419	0.152	0.501	0.487	0.460	0.391	0.438
	MIV	0.288	0.387	<b>0.667</b>	x	0.562	0.354	0.392	0.409	0.344	0.360
	MV	0.254	0.352	0.515	<b>0.628</b>	x	0.025	0.027	0.044	0.062	0.035
<i>P. microps</i>	mI	<b>0.753</b>	<b>0.855</b>	0.459	0.366	0.309	x	<b>0.780</b>	<b>0.806</b>	<b>0.600</b>	<b>0.677</b>
	mII	<b>0.811</b>	<b>0.837</b>	0.451	0.298	0.262	<b>0.839</b>	x	<b>0.776</b>	<b>0.732</b>	<b>0.884</b>
	mIII	<b>0.881</b>	<b>0.813</b>	0.369	0.239	0.225	<b>0.804</b>	<b>0.859</b>	x	<b>0.692</b>	<b>0.757</b>
	mIV	<b>0.789</b>	<b>0.847</b>	0.481	0.365	0.279	<b>0.854</b>	<b>0.894</b>	<b>0.857</b>	x	<b>0.816</b>
	mV	<b>0.799</b>	<b>0.857</b>	0.497	0.364	0.287	<b>0.830</b>	<b>0.922</b>	<b>0.819</b>	<b>0.901</b>	x

A comparison between the diversity index values of the diet for males and females (Table 4) only showed significant differences for *P. minutus* ( $t = 3.689$ ; d.f. = 40;  $p < 0.05$ ), with a higher diversity for females than for males. No statistical differences were detected for *P. microps* ( $t = 1.153$ ; d.f. = 36;  $p > 0.05$ ). In both species the number of taxa preyed (n) and the evenness (j) were higher in the diets of females.

A general increase in the number of prey items ingested in almost all food categories with increasing lengths was observed for *P. minutus* (Table 5). However, statistically significant differences were observed in the higher consumption of copepods by the smaller size classes (MI, MII and MIII), whereas oligochaetes, fish, bivalves and shrimps revealed a significantly greater importance for the higher size classes. Shrimps, ostracods, oligochaetes, fish and bivalves were not ingested by individuals from class I.

No clear trend of diet variation according to fish size was found for *P. microps* (Table 5), although significant differences were observed for 7 prey groups. Nevertheless, it was noticed that fishes were absent from the gut contents of small size classes, while shrimps were only ingested by individuals from the largest size class.

A high intraspecific food overlap, using both gravimetric and numeric frequencies, was obtained between the two smallest classes of *P. minutus* (0.88) (Table 6), and most neighbouring classes showed values close to or above 0.60. Overlap values between *P. microps* size classes for numeric frequencies were always higher than 0.80. Interspecific overlap was high ( $> 0.75$ ) between classes MI and MII of *P. minutus* and all size classes of *P. microps*, while low overlap ( $< 0.5$ ) occurred with the remaining larger classes of *P. minutus*. Low interspecific overlap was observed when gravimetric frequencies were used.

## DISCUSSION

The diets of *P. minutus* and *P. microps* included a high variety of prey. Such versatility is associated with their capacity to use different feeding behaviours such as biting and suction (Hamerlynck and Cattrijsse, 1994).

The diet of the sand goby, based on mysids, shrimps, polychaetes and foraminifera as the most important prey, was similar to those reported in previous studies in the Tagus (Costa, 1988) and the Sado (Cunha, 1994) estuaries in Portugal and for some areas of northern Europe (Fonds, 1973; Claridge *et al.*, 1985; Zander and Hagmann, 1987; Hamerlynck and Cattrijsse, 1994; Hostens and Mees, 1999; Laffaille *et al.*, 1999). However, in all of those previous studies, amphipods showed greater importance than that found in the present study, which could be explained by the low abundance of this prey in the study area (Calvario and Marques, 1983).

The common goby preferentially ingested endobenthic organisms, such as polychaetes. Several authors have reported a similar benthic-feeding habit, but pointed out the isopod *C. volutator* (Magnhagen and Wiederholm, 1982a; Magnhagen, 1986; Pihl, 1985) and bivalve siphons (Gee *et al.*, 1985) as dominant prey. Though they are present in the results of these authors, polychaetes have never revealed the importance they assume in this study. The diet of both gobies in the Tagus estuary shows a high similarity to the results reported for northwest European waters by other authors. The most marked differences are in the lower importance of amphipods in their diets in the Tagus estuary, which results in the opportunistic shift towards other more available prey items, such as mysids for *P. minutus* and polychaetes for *P. microps*.

*P. minutus* and *P. microps* are considered opportunistic carnivores, feeding on organisms that they select on the basis of their availability (Pihl, 1985). Thus, the coincidence in the seasonal variation observed in the ingestion of some prey groups by both species may suggest that diet reflected prey availability. Examples are the ingestion of polychaetes in spring and the higher abundance of copepods observed in both species diets in winter, when densities of this prey type are higher (Sobral, 1987). The latter example also seems to be connected to a decrease in availability of other prey such as polychaetes and isopods (Costa *et al.*, 1992).

During the reproductive season of these gobies, which in the Tagus estuary is longer than in north-west European water, extending from January to May (Moreira, 1989), several differences were identified between the diet of males and females. This may be due to the different activity pattern exhibited by individuals of each sex during the breeding season, since parental care is supported by males (Magnhagen, 1986; Moreira, 1989; Bouchereau *et al.*, 1991). Thus, females show a wider food-niche width than males, particularly for *P. minutus*. Males are probably limited to the food available around the nest, whereas females apparently behave as food maximisers, feeding on a higher number of taxa and ingesting most of the prey types in higher numbers. Although the consumption of some prey, such as polychaetes, isopods and some amphipods, involves more time expended on prey searching and handling, the richer caloric content of these prey compared to copepods is likely to supply more energy for reproduction and growth.

No goby eggs were observed in the gut contents, which according to Hamerlynck and Cattrijsse (1994) may suggest that nest sites were not a limiting factor in this estuary.

Juveniles from both species increased their niche widths as they grew (Thorman, 1983). This was obvious in the comparison between the gut contents of the various *P. minutus* size classes. The smallest classes, which revealed a strong intraspecific food overlap, ingested mainly copepods, as previously reported by Pihl (1985), Mehner (1992) and Hamerlynck and Cattrijsse (1994). For *P. minutus* with a total length greater than 50 mm a dietary shift was observed, with a progressive disappearance of copepods and an increasing importance of larger prey, such as shrimps and fishes. Doornbos and Twisk (1987) identified a gradual shift with the weight proportion of copepods decreasing from nearly 95% in

individuals of 30 mm length to less than 1% in specimens of 65 mm length. Hostens and Mees (1999) described the progressive replacement of copepods in the diet of sand gobies as they grow, but this study reported copepods as an important prey item for specimens larger than 50 mm.

The negative correlation between the *P. minutus* fullness index and fish density suggests the presence of a trophic intraspecific competition in autumn, when densities were high and the fullness index low. Nevertheless, the positive relationship between water temperature and fullness index indicates that the decrease in this goby feeding activity in autumn and winter, the coldest seasons, could be primarily influenced by this abiotic parameter, attenuating the hypothesis of a possible intraspecific competition. The corresponding increase in feeding activity and water temperatures was also observed for *P. microps*, but in this case there was no correlation between fullness index and the fish density. Although some differences were detected in the food ingested by the different size classes considered for *P. microps*, no trend was noticed. A possible explanation for this is that the different size classes were artificial and that there was no feeding segregation between the size classes considered. Doornbos and Twisk (1987) observed a switch in the diet of individuals with 40 mm from a composition dominated by copepods to one including larger epibenthic crustaceans.

The large interspecific food overlap between all size classes of *P. microps* and the two smallest of *P. minutus* using the numeric frequencies suggests potential competition for the same food items in food limited situations. However, the reduced interspecific overlap when gravimetric frequencies are used suggests that although they ingest the same prey items in similar numeric proportions within some size classes, they might select preys with different weights. This could represent an interspecific segregation in the differential consumption of the same prey items due to the different dimensions reached by each species. Vertical spatial segregation was found between *P. minutus* and another goby of the same genus, *Pomatoschistus lozanoi* (de Buen), with the former feeding closer to the bottom (Hamerlynck and Cattrijsse, 1994; Hostens and Mees, 1999; Laffaille *et al.*, 1999). Kislalioglu and Gibson (1977) observed that although they appear to eat the same major food groups, there was low similarity when prey species were compared. Mehner (1992), studying the diet of the two species in the



first weeks after hatching, found that *P. microps* ate a smaller number of prey species than *P. minutus*. Being socially dominated by *P. minutus* (Magnhagen and Wiederholm, 1982b; Wiederholm, 1987; Mehner, 1992), *P. microps* narrowed its trophic niche where both species coexist (Edlund and Magnhagen, 1981). However, in the present study the presence of segregation into different feeding niches is mainly associated with the size of the specimens. Individuals larger than 50 mm apparently ingest more endobenthic preys than specimens smaller than 50 mm. Thorman and Wiederholm (1983) found high diet overlap between the two species, though there was no evidence for the existence of food competition. In these two coexisting species the interspecific competition between individuals of the same length is also avoided or diminished by the time lag in the reproduction of the two species and thus in the colonisation time of the study areas by the young of the year of each species.

With the absence of data about prey availability it is impossible to determine whether the feeding resources of these species are a limiting factor. Further investigations should focus on this matter and also on questions about the reproduction behaviour of both species in the Tagus estuary.

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