# Opeatogenys gracilis (Pisces: Gobiesocidae): an overlooked species or another 'Mediterranean endemism' found in Atlantic waters?

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The occurrence of *Opeatogenys gracilis* outside the Mediterranean Sea is confirmed for the first time. This is probably a more common species than previously thought, but its apparent complete dependence on seagrass beds suggests the possibility of including it in the IUCN threatened species list. Some sex differences are described and a complete meristic and morphometric description of the species is presented. The occurrence of the species in the north-east Atlantic indicates that it might be a recent dispersal from the Mediterranean Sea, or an overlooked part of the autochthonous fauna.

Key words: north-east Atlantic; Opeatogenys gracilis; seagrass beds.

# INTRODUCTION

Opeatogenvs gracilis (Canestrini) is a poorly studied gobiesocid that lives in seagrass beds (Hofrichter, 1995). It is a very small fish with a total length  $(L_T)$ of only c. 3.0 cm (Briggs, 1986). This species was first described by Canestrini (1864) as Mirbelia gracilis, based on one specimen collected in Nice (southern France). Moreau (1881) did not agree with Canestrini's description of the new genus and placed this species in the genus Lepadogaster, as L. gracilis (Griffini, 1903). Briggs (1955) examined a specimen noted by Carus (1893) and described the genus *Opeatogenys*, at the time a monospecific genus. Later, Briggs (1957) described a second species, Opeatogenvs cadenati Briggs, also occurring in seagrass beds in western Africa (Ghana and Senegal; Briggs, 1957). Opeatogenys cadenati is now also known from Morocco (Brownell, 1978) and the Canary Islands (Brito, 1991). Briggs (1986) does not provide any common name for O. gracilis, probably because its very small size and cryptic habits means that it is infrequently encountered by non-scientists who might give it a vernacular name. Bini (1968), however, cites the name 'pygmy clingfish', and Debelius (1997) uses the name 'seagrass clingfish'.

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No type specimens of O. gracilis are known (Eschmeyer, 2004) and, until recently, only a few specimens had been collected in the Mediterranean Sea. Briggs (1986) described the distribution of this species as extending from Nice to the Adriatic. One specimen was described by Carus (1893) from Messina (Sicily); another specimen was reported by Kolombatovic (1900) associated with Posidonia oceanica (L.) Delile in the Adriatic. Dieuzeide et al. (1955) refer to the occurrence of some specimens of O. gracilis in P. oceanica meadows in Algeria. More recently, Reina-Hervás & Nuñez Vergara (1985) described two specimens found in Cymodocea nodosa (Ucria) Ascherton beds in Málaga (Spain). Cardona & Guerao (1992) collected five specimens from C. nodosa and Caulerpa prolifera (Forsskaal) Lamouroux meadows in Cataluña (Spain). Hofrichter (1995) confirmed the presence of the species at previous sites (Adriatic and Sicily) and also described its occurrence in Cyprus, France (Banyuls-sur-Mer) and Ibiza (capturing, for the first time, a large number of specimens). Hofrichter & Patzner (2000), studying clingfishes associated with P. oceanica and C. nodosa, cited a maximum abundance of eight individuals  $m^{-2}$ , and noted the occurrence of O. gracilis at depths of up to 20 m. Finally, Vizzini & Scilipoti (1999) and Vizzini et al. (2001) captured 275 specimens from P. oceanica meadows in western and northern Sicily, and suggested that this must be a more common species than previously thought. Gonçalves (1941), reviewing an old collection by King D. Carlos I of Portugal, found a specimen classified as Mirbelia gracilis collected in the Sado River (western Portugal) in 1897. This author has not confirmed the identity of the specimen and refers to it as Lepadogaster sp.

In this paper the occurrence of *O. gracilis* outside the Mediterranean Sea was investigated, some old records were revised, and a complete meristic and morphometric description of the species was undertaken to determine the main differences between the sexes. Hypotheses explaining the Atlantic occurrence of this species, otherwise known only from the Mediterranean Sea are also discussed.

# MATERIALS AND METHODS

Specimens were collected at two locations on the Portuguese coast, the Ria Formosa (n = 26, collected on 23 February 2001), southern Portugal  $(36^{\circ}59 \text{ N}; 7^{\circ}51' \text{ W})$  and the Sado estuary (n = 4, three collected on 6 June 2001 and one on 10 July 2003), western coast (38°27 N; 8°43 W). Ria Formosa is a coastal lagoon in permanent connection with the sea, formed by a succession of sand barrier islands along the south coast of Portugal. Inside the lagoon extensive seagrass beds (C. nodosa and Zostera marina L.) occur at shallow depths (<6 - 7 m) (Alberto et al., 2001; Billingham et al., 2003). The Sado estuary has high salinity levels throughout the year (Cabral, 2000). Some sparse beds of Z. marina and Zostera noltii Hornemann are present at depths of <5-6 m. At both sites, fish were collected using SCUBA by dragging a fine mesh (<1 mm) hand net along seagrass leaves. Specimens were killed with an overdose of the anaesthetic MS222, fixed in formalin (10% diluted in sea water) and preserved in 70% alcohol. Biometric measurements were carried out using callipers ( $L_T$  and standard length,  $L_S$ ) and a dissecting microscope with an ocular micrometer to the nearest 0.01 mm. The following measurements were taken, after Briggs (1955):  $L_T$ ,  $L_S$ , head length (HL), head width (HW), head height  $(L_{\rm H})$ , snout length (SnL), eye diameter (Eye), interorbital distance (IOD), dorsal fin length (DFL), anal fin length (AFL), caudal peduncle length (CPL), caudal peduncle height (CPH), caudal fin length (CFL), pelvic disc length (DiL), pelvic disc width (DiW),

distance from the anus to the anal fin base (AnAFB), distance from the anus to the pelvic disc (AnDi), pre-pelvic disc distance (PreDi), pre-dorsal fin length (PreDFL), pre-anal fin length (PreAFL), length of the urogenital papilla ( $L_{\rm UP}$ ), width of the urogenital papilla (UPW), depth at anus (dA), dorsal-caudal distance (DC), postdorsal distance (PDD) and postdorsal-caudal distance (PDC). Specimens were sexed by external examination of the urogenital papilla. The number of rows of flattened dermal papillae in the pelvic disc regions A, B and C, follow Briggs (1955, 1986) descriptions. All specimens were catalogued in the collection of the Arrábida Nature Park Oceanographic Museum (Portinho da Arrábida, Portugal).

The specimen from the King D. Carlos I collection deposited at the Vasco da Gama Aquarium in Lisbon (catalogue number 293) was carefully analysed to confirm its taxonomic status. An additional Mediterranean specimen from the Natural History Museum (London) (catalogue number BMNH 1995.3.15.1), captured at Banyulssur-Mer, France, was examined for comparative purposes. The only other specimen found is no longer suitable for meristic and morphometric measurements. This is the specimen noted by Carus (1893) and examined by Briggs (1955), which belongs to the California Academy of Sciences collection (catalogue number 147898), and has been cleared and stained.

Size differences  $(L_{\rm S})$  between sexes were tested using a t-test for independent samples. All other variables, which could be influenced by allometry, were divided by  $L_{\rm S}$ , arcsine  $(\sqrt{p})$  transformed (Sokal & Rohlf, 1981) (where p is the value of the proportion) and tested for sex-differences using the t-test for independent samples. In one case  $(L_{\rm UP}L_{\rm S}^{-1})$ , the homogeneity of variances assumption could not be met (Levene's test) and a Mann-Whitney U test was used. To test for sex differences in the body proportions (as in Briggs, 1955), variables were  $\log_{10}$  transformed (Sokal & Rohlf, 1981) and tested using the t-test for independent samples. Results were considered significantly different if P < 0.05 (two-tailed). All tests were performed using the STATISTICA computer package.

### **RESULTS**

Morphometric characteristics of the specimens collected in this study are presented in Table I. No difference in size  $(L_{\rm S})$  between sexes was found (P=0.46). Males and females, however, differ significantly in the following measurements (as proportion of  $L_{\rm S}$ ) (Table II): anal fin length (AFL; P<0.05), urogenital papilla length  $(L_{\rm UP};$  Mann-Whitney U test, P<0.001) and width (UPW; P<0.001), and dorsal-caudal distance (DC; P<0.05). In the body proportions analysed (as in Briggs, 1955), only the eye diameter in interorbital space was also significantly different between sexes (P<0.05) (Table III).

For meristic characters the values are (number of individuals in parentheses): dorsal fin rays 3(21) or 4(9); anal fin rays 3(11) or 4(19); pectoral fin rays 15(10) or 16(20); caudal fin rays 8(16), 9(6) or 10(8); rows of flattened papillae in pelvic disc region A 4(26) or 5(4), pelvic disc region B 4(20) or 5(10) and pelvic disc region C 3(20), 4(8) or 5(2).

Briggs (1957) stated that *O. cadenati* can be distinguished from *O. gracilis* based on the following characteristics. *Opeatogenys cadenati*: body depth 6.0 (5.7 -6.2) in  $L_s$ ; pectoral fin rays 21 (20–22); upper attachment of gill membrane opposite 4th (4–5) pectoral ray; upper attachment of axial, dermal flap opposite 10th (9–11) pectoral ray; seven short, blunt rakers present on the third gill arch. *Opeatogenys gracilis*: body depth 7.1 in  $L_s$ ; pectoral fin rays 16; upper attachment of gill membrane opposite 3rd pectoral ray; upper attachment of axial,

TABLE I Mornhometric measurements (mm) for 30 specimens of O oracilis (16 females and 14 males). For Lyand CFL n=15 females and

I ABLE I.	Table 1. Proprometric measurements (min) for 50 specimens of $C$ , $gratins$ (10 females and 14 mates). For LT and CT L, $n = 15$ females and $n = 13$ males, for PDD, $n = 13$ females and $n = 13$ males, due to damaged fins; and for dA $n = 15$ females	n = 13 males;	ales;	for F	DD,	n = n	13 fe	en spe emale	s and	n > 0	7. gr 13 ma	iles, d	due to	dama dama	aged	i+ ma fins; e	ucs). I'c und for	$dA = \frac{1}{4}$	for PDD, $n = 13$ females and $n = 13$ males, due to damaged fins; and for dA $n = 15$ females	n — 1. ales	7 15111	ales a	nii
	$L_{ m T}$ $L_{ m S}$	$L_{\mathrm{T}}$ $L_{\mathrm{S}}$ HL HW $L_{\mathrm{H}}$	/ L <sub>H</sub>	SnL	Eye	IOD	DFL	AFL	CPL	СРН	CFL D	iL D	iW An	AFB /	AnDi F	reDi	PreDFL	PreAFL	SnL Eye IOD DFL AFL CPL CPH CFL DiL DiW AnAFB AnDi PreDi PreDFL PreAFL Lup UPW dA DC PDD PDC	V dA	DC	PDD P	DC
Females																							
Mean	23.5 19.6	23.5 19.6 6.47 4.48 2.77	3 2.7	7 1.67	<u>;</u>	1.62	1.09	1.67  1.44  1.64  1.09  1.20  3.10  1.68	3.10	1.68	3.85 3.95 2.94	95 2.		2.96	4.21 4.15	4.15	15.97	15.52	0.22 0.19	2.65	4.18	2.65 4.18 7.15 3.25	3.25
S.D.	2.7 2.1	2.7 2.1 0.77 0.54 0.36	4 0·3t	6 0.22	0.15	0.15 0.32		0.19 0.15	0.49	0.49 0.37	0.59 0.38		0.44 0	0.43	09.0	0.53	1.72	1.51	90.0 90.0	0.35 0.48	3.48	0.80 0.40	.40
Maximum		28.3 23.8 7.60 5.43 3.60	3 3.60		2.06 1.65 2.30	2.30	1.36 1.44	<u>+</u>	4.14	4.14 2.69	4.49 4.49	49 3.	3.78 4	4.29	5.05	5.15	18.65	18.50	0.30 0.30		3.38 5.05	8.40 4.06	90:
Minimum	20.0 16.6	20.0 16.6 5.40 3.45 2.35	5 2.3;		1.21	1.20	99.0	1.35 1.21 1.20 0.66 0.99	2.13	96.0	2.13 0.96 2.47 3.18	18 2.	2.27 2	2.32	3.28	3.43	13.40	13.50	0.12 0.07		3.60	2.20 3.60 6.10 2.58	.58
Males																							
Mean	24.4 20.2	24.4 20.2 6.66 4.67 2.79	7 2.75		1.41	1.78	1.70 1.41 1.78 1.26 1.37	1.37	3.17 1.73	1.73	3.97 4.17	17 3.	3.17 3	3.23	4.08	4.12	16.34	15.80	0.68 0.29		4.10	2.70 4.10 7.29 3.22	3.22
S.D.	2.7 2.4	0.94 0.86 0.47	5 0.4		0.28 0.11 0.29	0.29		0.31 0.24		0.57 0.24	0.39 0.57 0.45	57 0.		0.41	0.87	0.59	2.10	1.91	0.19 0.10	0.45	0.59	0.45 0.59 1.28 0.47	.47
Maximum	29.8 25.3	8.00 6.50 3.90	3.90		1.61	2.30	1.97	2.20 1.61 2.30 1.97 1.76	4.90	4.90 2.06	4.70 5.50	50 3.	3.94 4	4.16	6.50	5.20	19.50	19.40	1.00 0.48		3.80 5.60	9.90 4.10	·10
Minimum	20.5 17.1	20.5 17.1 5.35 3.35 2.20	5 2.20	0 1.20	1.20 1.26 1.15	1.15	0.78	0.78 0.92 2.43 1.16	2.43	1.16	3.38 3.53	53 2.	2.12 2	2.62	3.00	3.38	13.30	12.70	0.41 0.17	2.10 3.05		5.70 2.58	.58
Total																							
Mean	23.9 19.9	23.9 19.9 6.56 4.57 2.78	7 2.7		1.42	1.71	$1.68  1.42 \ 1.71  1.17  1.28$	1.28	3.13	3.13 1.70	3.91 4.05 3.05	05 3.		3.09	4.15	4.14	16.15	15.65	0.44 0.24		4.15	2.68 4.15 7.22 3.24	.24
S.D.	2.7 2.3	0.84 0.70 0.41	0.4		0.25 0.13 0.31	0.31		0.26 0.21	0.52	0.52 0.31	0.50 0.48	48 0.	0.45 0	0.44	0.73	0.55	1.88	1.68	0.27 0.09		0.53	$0.40 \ 0.53 \ 1.05 \ 0.43$	.43
Maximum	29.8 25.3	8.00 6.50 3.90	3.90		1.65	2.30	1.97	2.20 1.65 2.30 1.97 1.76	4.90	4.90 2.69	4.70 5.50	50 3.	3.94 4	4.29	6.50	5.20	19.50	19.40	1.00 0.48		3.80 5.60	9.90 4.10	·10
Minimum	20.0 16.6	20.0 16.6 5.35 3.35 2.20	5 2.2(		1.21	1.15	99.0	0.92	2.13	96.0	$1 \cdot 20  1 \cdot 21  1 \cdot 15  0 \cdot 66  0 \cdot 92  2 \cdot 13  0 \cdot 96  2 \cdot 47  3 \cdot 18$	18 2.	2.12 2	2.32	3.00	3.38	13.30	12.70	0.12 0.07	2.10 3.05		5.70 2.58	.58

Lr. total length; Ls. standard length; HL, head length; HW, head width, LH, head height; SnL, snout length; Eye, eye diameter; IOD, inter orbital distance; DFL, dorsal fin length; AFL, anal fin length; CPL, caudal peduncle length; CPH, caudal peduncle height; CFL, caudal fin length; DiL, pelvic disc length; DiW, pelvic disc width; AnAFB, distance from the anus to the anal fin base; AnDi, distance from the anus to the pelvic disc; PreDi, pre-pelvic disc distance; PreDFL, pre-dorsal fin length; PreAFL, pre-anal fin length; Lup, length of urogenital papilla; UPW, width of the urogenital papilla; dA, depth at anus; DC, dorso-caudal distance; PDD, post dorsal distance; PDC, post dorsal distance.

Table II. Proportions (as percentage of  $L_{\rm S}$  in mm) for 30 specimens of O. gracilis (16 females and 14 males) which revealed significant differences between sexes (see Table I for abbreviations)

	AFL	$L_{ m UP}$	UPW	DC
Females				
Mean	6.12	1.14	0.98	21.34
S.D.	0.74	0.26	0.28	1.24
Maximum	8.23	1.52	1.52	23.08
Minimum	5.29	0.66	0.38	18.93
Males				
Mean	6.77	3.33	1.43	20.28
S.D.	0.93	0.70	0.33	1.59
Maximum	8.39	4.42	2.11	22.33
Minimum	5.24	2.36	0.97	17.04

dermal flap opposite 7th pectoral ray; five short, blunt rakers present on the third gill arch.

Hofrichter (1995), however, examined the differences between these two species and reported that the only character that presents no overlap between the

Table III. Body proportions as in Briggs (1955) for the 30 specimens of O. gracilis (16 females and 14 males), and for the BMNH specimen.  $L_{\rm H}$  in  $L_{\rm S}$  is not presented for this specimen since the head is slightly damaged (see Table I for abbreviations)

	$L_{ m H}$ in $L_{ m S}$	CPH in CPL	$_{ m in}^{ m HL}$	$\frac{\mathrm{HW}}{\mathrm{in}\ L_{\mathrm{S}}}$	SnL in HL	Eye in IOD	Eye in HL	${ m DiL}$ in $L_{ m S}$
Females								
Mean	7.11	1.91	3.04	4.40	3.90	1.14	4.50	4.97
S.D.	0.55	0.43	0.14	0.32	0.26	0.19	0.28	0.32
Maximum	8.13	3.03	3.25	5.00	4.37	1.43	4.96	5.74
Minimum	6.01	1.18	2.77	3.99	3.57	0.79	3.81	4.39
Males								
Mean	7.31	1.85	3.05	4.39	3.96	1.26	4.73	4.87
S.D.	0.52	0.30	0.19	0.41	0.38	0.14	0.45	0.25
Maximum	8.24	2.53	3.32	5.25	4.83	1.53	5.38	5.32
Minimum	6.49	1.51	2.78	3.89	3.55	0.91	3.93	4.52
Total								
Mean	7.20	1.88	3.05	4.39	3.94	1.21	4.62	4.91
S.D.	0.55	0.38	0.16	0.36	0.32	0.17	0.38	0.29
Maximum	8.24	3.03	3.32	5.25	4.83	1.53	5.38	5.74
Minimum	6.01	1.18	2.77	3.89	3.55	0.79	3.81	4.39
BMNH specimen								
1		2.66	2.89	4.37	3.28	1.00	4.21	4.27

BMNH, British Museum of Natural History.

species is the number of pectoral fin rays. In order to establish that the specimens belonged to *O. gracilis* and not *O. cadenati* or some undescribed species a search was also made for available material from museum collections. Although only one specimen was available (BMNH 1995.3.15.1; see Material and Methods), data from this specimen and from the literature show that there are no morphological differences between the present specimens and Mediterranean Sea ones (Table III). Also, the numbers of pectoral fin rays in the present specimens (15 or 16) are clearly different from the numbers reported for O. cadenati by Briggs (1957) (20 to 22 pectoral fin rays).

Thus, the description by Briggs (1955) of *O. gracilis*, based on a single specimen, can now be updated based on material examined in this study (Table III): body depth  $7\cdot2$  ( $6\cdot0-8\cdot2$ ) in  $L_{\rm S}$ ; caudal peduncle height  $1\cdot9$  ( $1\cdot2-3\cdot0$ ) in its length; head length  $3\cdot1$  ( $2\cdot8-3\cdot3$ ) in  $L_{\rm S}$ ; head width  $4\cdot4$  ( $3\cdot9-5\cdot3$ ) in  $L_{\rm S}$ ; snout length  $3\cdot9$  ( $3\cdot6-4\cdot8$ ) in head length; eye diameter  $1\cdot2$  ( $0\cdot8-1\cdot5$ ) in bony interorbital space and  $4\cdot6$  ( $3\cdot8-5\cdot4$ ) in head length; length of disc  $4\cdot9$  ( $4\cdot4-5\cdot7$ ) in  $L_{\rm S}$ . Taking into consideration all specimens described in the literature (Briggs, 1955; Reina-Hervás & Nuñez Vergara, 1985; Cardona & Guerao, 1992; Hofrichter, 1995; Vizzini & Scilipoti, 1999; Vizzini *et al.*, 2001), the meristic counts for the species are: dorsal fin rays 2-4; anal fin rays 2-4; pelvic fin rays 4; pectoral fin rays 14-18; caudal fin rays 8-14; rows of flattened papillae in disc region A 4-5, disc region B 4-5 and disc region C 3-5. The maximum known size is  $27\cdot0$  mm  $L_{\rm S}$  ( $33\cdot0$  mm  $L_{\rm T}$ ) (Cardona & Guerao, 1992).

Body colouration of the specimens analysed varies from light green to light brown and dark red sometimes with small blue iridescent spots along the flanks (Reina-Hervás & Nuñez Vergara, 1985; Hofrichter & Patzner, 2000).

The specimen of the King D. Carlos I collection, in spite of being in very bad condition, could clearly be identified as O. gracilis ( $L_T = 25.0 \text{ mm}$ ;  $L_S = 20.0 \text{ mm}$ ; fin rays: dorsal 3; anal 3; pectoral 15/16).

#### DISCUSSION

A marked sexual dimorphism was found in the size and shape of the urogenital papilla in *O. gracilis* [also described for *O. cadenati* by Briggs (1957)], which can clearly be used to separate both sexes. This dimorphism was predicted by Briggs (1955) who described a long, slender genital papilla in the specimen he examined (a male). Males and females also differ significantly in anal fin length, dorsal-caudal distance and eye diameter in interorbital space. These characters are thus useful for distinguishing the two sexes in this species, since they present quite similar colours and morphology.

The occurrence of *O. gracilis* outside the Mediterranean Sea is described here for the first time. The presence of this species on the Portuguese coast, however, can be traced back to the late 19th century (given the existence of the specimen collected during the King D. Carlos I oceanographic expeditions). The occurrence of Mediterranean Sea coastal fish species on nearby Atlantic shores has been documented in recent years (Nieto & Alberto, 1992; Almada *et al.*, 1993; Gonçalves *et al.*, 1993; Henriques *et al.*, 1999; Bañón *et al.*, 2002). It is not clear, however, if these recent findings result from an increased research effort in the Portuguese and Spanish Atlantic coasts (Almada *et al.*, 2001) or if the

distributions of these species are changing as a result of global warming. In fact, water temperature has been increasing steadily on the Portuguese coast since the 1940s (Lemos & Pires, 2004), but the nearshore ichthyofauna was largely unknown until recently. The case of *O. gracilis* is further complicated by the fact that this is a very cryptic species that occurs exclusively in seagrasses and that can only be sampled using small mesh size nets. Although there is no direct evidence to support the hypothesis that the Portuguese distribution is associated with global warming, the fact that a specimen of this species was collected in the late 19th century in Portugal could also support either argument; that this is an abundant but overlooked species of small size which occurs exclusively in seagrasses, or that the late 19th century was a period of increase in seawater temperature in the north-eastern Atlantic (Southward *et al.*, 1995; Levitus *et al.*, 2000), resulting in the presence of this species in this region at that time.

Although this might be an abundant species, its apparent strict dependence upon seagrass beds and the overall decline of these beds in the Atlantic due to anthropogenic action and seaweed infestations (den Hartog & Polderman, 1975; Bortone, 2000) suggest that this is a possible candidate for the IUCN Red List of Threatened Species. The species might be listed in the data deficient (DD) category (IUCN, 2004) because more information is required for this species and there is the possibility that future research will show a threatened classification is appropriate. More detailed studies focused on seagrasses in the northeastern Atlantic and the Mediterranean Sea using adequate techniques for collecting this very small and cryptic species, however, are needed to clarify the distribution, range, and ecology of *O. gracilis*.

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