

## NOTES ON THE BIOLOGY OF *Centrolabrus caeruleus* AZEVEDO, 1999 (TELEOSTEI: LABRIDAE)

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*Centrolabrus caeruleus* is a recently described littoral fish whose biology is poorly known, despite the fact that it is very abundant in the Azores. This paper presents general information about the age and growth, food and reproduction of this species. Males grow to larger sizes than females, and there are sexual differences in colour and behaviour. Preliminary von Bertalanffy growth curves are given for both sexes, based on modal analysis of the monthly length-frequency distributions. The length-weight relationship is also given. Food is dominated by small phytal invertebrates. Reproduction occurs mainly between March and June. Young of the year are seen from July on. Territorial males build algal nests where females spawn. Details of nest building, and of the courtship and spawning behaviours are given.

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*Centrolabrus caeruleus* é um peixe litoral recentemente descrito e cuja biologia é pouco conhecida, apesar de ser muito abundante nos Açores. Este artigo apresenta informação geral acerca da idade e crescimento, alimentação e reprodução desta espécie. Os machos atingem tamanhos superiores aos das fêmeas e existem diferenças de cor e comportamento entre os dois sexos. Apresentam-se as equações de crescimento de von Bertalanffy preliminares para cada sexo, calculadas a partir da análise modal das distribuições de frequências de comprimentos mensais. A relação comprimento-peso é também apresentada. A alimentação baseia-se sobretudo em pequenos invertebrados fitais. A reprodução decorre entre Março e Junho, podendo os juvenis ser observados a partir de Julho. Nesta espécie, machos territoriais constroem ninhos de algas onde as fêmeas colocam os ovos. São apresentados detalhes da construção do ninho e dos comportamentos de corte e de desova.

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### INTRODUCTION

*Centrolabrus caeruleus* is a very common shallow water wrasse that inhabits the rocky coasts of the Azores islands. Only very recently was this species distinguished from *C. trutta*, a similar fish that inhabits the coasts of Madeira and presumably also the Canary and Cape Verde islands (AZEVEDO 1999). *Centrolabrus exoletus*

is the only other species of this genus. It can be found in the coasts of northern Europe, from Portugal to Norway, and in Greenland (QUIGNARD & PRAS 1986).

The only published information on aspects of the biology and ecology of *C. caeruleus* is the brief account of PORTEIRO et al. (1996), as *C. trutta*. This species is reported to live around boulders and rocks covered with algae, from 0 to 30 m depth, being most abundant between 2 and

15 m. Those authors also stated that this species is quite frequent, being the third most common wrasse between 0 and 20 m depth. The main diet items reported were small benthic invertebrates. Reproduction was said to occur in late spring and summer, and the species was described as having benthic eggs with some degree of parental care by the male, which build nests with algae. Ontogenetic differences in colouration were described, with the young said to show a greenish or brownish irregular striped pattern, while larger individuals become progressively turquoise blue. This latter colour pattern was reported by AZEVEDO (1999) to occur only in territorial males. The present paper investigates and attempts to clarify different aspects of the reproductive biology, age and growth of this species.

## MATERIAL AND METHODS

Work was carried out in the rocky reef outside and to the east of the Ponta Delgada marina, São Miguel Island, in the eastern Azores. Rotenone sampling was conducted monthly from December 1993 to November 1995. In each sample, an area of approximately 100 m<sup>2</sup> was encircled with a 1 cm mesh net. The net was placed in water 2-4 m deep, and extended from the bottom to the surface, enclosing a cylindrical area. Inside this area a diver would spread 1,5 l of commercial rotenone emulsion. Fish were collected by hand or with dipnets after the rotenone cloud dispersed, usually within 5-10 minutes. Fish affected by the rotenone outside the net were also collected, but kept separate. For purposes of the present study, all collected specimens (a total of 479 fishes) were considered.

Collected fish were placed on buckets with salt water and transported to the laboratory, where they were measured (total length, TL, to the nearest mm) and weighed (total weight, TW, with 0,01 g accuracy). They were then fixed in 15% buffered formalin for two weeks and preserved in 70% alcohol. Later, each specimen was re-weighed, dissected, and the stomach and gonads removed and kept separately in 70% alcohol. The gonads were weighed (gonad weight, GW, 0,01 g accuracy) and sex determined, when possible, by

their macroscopic appearance. Stomach contents from the first year (n=212) were spread in water in a Petri dish, so that the items could be identified to the lowest possible taxonomic rank using a binocular microscope and counted. A vacuity coefficient was calculated as the percentage of the number of stomachs without recognisable items over the total number of stomachs examined. Results for each item are expressed as percentage occurrence in the stomachs (number of stomachs with item over the total number of stomachs analysed) and percent abundance of the item (number of the given item over total number of items).

From January to June 1997 a second sampling, was carried out in the same area, specifically designed to obtain more data on the reproduction. A total of 95 specimens were captured by divers using a small speargun. After capture the fish were placed in saltwater and transported to the laboratory where each was measured and weighed as above, and photographed using an adaptation of the method described by EMERY & WINTERBOTTOM (1980). Each animal was dissected fresh and the gonads removed and weighed (GW) with 0,001g accuracy. The gonad was then processed histologically: fixed in AFA (ethanol 50% - 600 ml, formalin - 300 ml, acetic acid glacial - 120 ml) for 24 hours, dehydrated and embedded in paraffin. Longitudinal sections 5-7µm thick were stained with haematoxylin and eosin. Underwater observations of fish behaviour were conducted while scuba diving. Observations were recorded on polyester paper attached to an acrylic clipboard. A total of 23 dives were made, giving approximately 20 hours of observation. One to three nests were visited in each dive.

Modal analysis of the length frequency distributions, and the fitting of the resultant age-length distribution to a von Bertalanffy curve were done following the methods in SPARRE & VENEMA (1992) and using program FiSAT (GAYANILO et al. 1994). January 1st was used as the birth date.

A gonadosomatic index (GSI) was calculated as  $GSI = TW / (TW - GW) * 100$ . Due to the small usable sample size, data from 1993-95 and 1997 were pooled. It was assumed that weight differences between fresh and preserved material

would affect TW and GW in an approximately equal way, and therefore have negligible impact on their ratio, the GSI.

## RESULTS

### AGE AND GROWTH

Monthly length frequency distributions are shown on Fig. 1. Young of the year (2-8 cm TL) were first caught in July, in both 1994 and 1995. They were abundant enough for the mean length of the respective cohorts to be followed until November of each year (Table 1). In that four-month period, their mean length increased by approximately 4 cm. The 0+ cohort was the only one, which could be positively identified on the length frequency distributions.

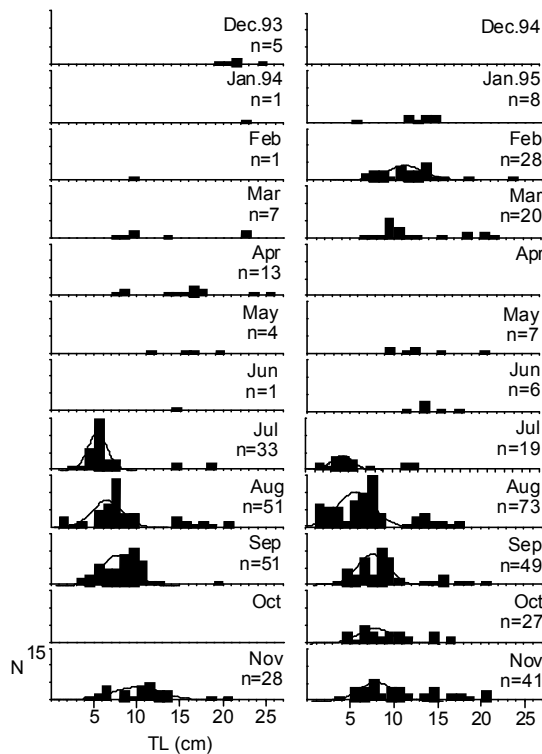


Fig. 1. Monthly length frequency distributions, with superimposed curves resulting from the modal analysis. Data from these curves in Table 1.

Males grow larger than females: the largest male measured 27 cm TL, whereas the largest female was only 23 cm TL. A preliminary estimation of the von Bertalanffy growth curve for this species was obtained forcing an  $L_{\infty}$  of 23 (females) and 27 (males) on the data of Table 1. The resulting curves are given in Fig. 2, and the respective parameters are:

- females:  $L_{\infty} = 23$ ;  $K = 0.823$ ;  $t_0 = 0.164$ ;
- males:  $L_{\infty} = 27$ ;  $K = 0.646$ ;  $t_0 = 0.141$ .

The length-weight relationship of the specimens collected is given by:

$$TW = 0.0106 * TL^{3.149} \quad (n = 474; r = 0.996)$$

Table 1.  
Age (in years) and mean total length (TL, in cm) of 0+ fish, cohorts of 1994 and 1995.

	Age	1994		1995	
		TL	n	TL	n
Jul.	0.53	6.4	29	5.1	15
Aug.	0.62	7.6	40	6.4	58
Sep.	0.7	9	50	8.5	41
Oct.	0.79			8.9	22
Nov.	0.87	10.8	26	8.9	25

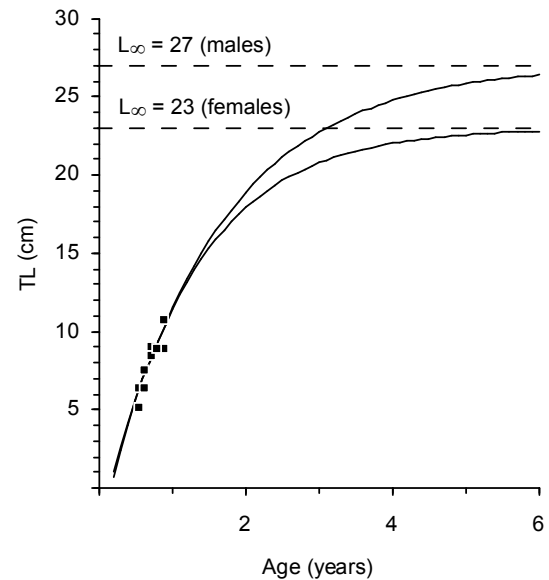


Fig. 2. Growth curves for both sexes. Black squares represent the monthly mean lengths of the 0+ fish used for the calculations. See text for curve parameters.

## FOOD

Feeding is carried out with the animal swimming slowly near the bottom, amongst the algal fronds. The eyes are very active, indicating an important role of visual cues to prey location. Short and fast forward movements are associated with prey ingestion. Consistent with these observations, stomach contents were found to be dominated by small (2-5 mm) benthic invertebrates (Table 2), mostly crustacea. Amphipods were the more frequent and abundant items. Small pieces of algal fronds were found in most stomachs, but they reached the terminal portion of the intestine undigested.

Table 2.

Main items in the stomach contents: percent occurrence in the stomachs (Occur.) and percent abundance of each item (Abun.).

	Occur. (%)	Abun. (%)
Algae	98	-
Polychaeta	4	0
Gastropoda	13	4
Bivalvia	11	2
Amphipoda	96	83
Isopoda	29	4
Brachyura	17	1
Caridea	9	1
Ophiuroidea	8	3
N=212, coefficient of vacuity= 8%		

## REPRODUCTION

### *Gonad morphology and histology*

The gonads of both sexes have the typical elongated shape and position above the intestines and below the gas bladder found in most teleosts. Outside the reproduction season, gonads are small and thread-like, ranging in colour from white to beige and rose. Macroscopic sex determination is difficult in most cases. Mature ovaries are big, brownish yellow or beige. Oocytes can be seen through the transparent ovary wall. Mature testes are white, sometimes with grey or blue tones. Blood vessels can be seen on the outside wall of the gonads of both sexes.

Histological sex determination was always possible, except in some 0+ fish. No histological indication of hermaphroditism, according to the criteria delineated by SADOVY & SHAPIRO (1987), was found.

### *Size at maturity*

The relationship between GSI and size for each sex indicated that the smallest mature specimens were a 14 cm female and a 12 cm male (Fig. 3). Small males had higher GSI values than large ones, comparable with the highest female GSI values. These small males with large testis (GSI 8-9 %) did not have the blue colouration associated with the territorial, nesting males (see below), which, in turn, had GSI values not exceeding 3 %. There are thus sexually mature males that do not build nests, and these are the smallest ones.

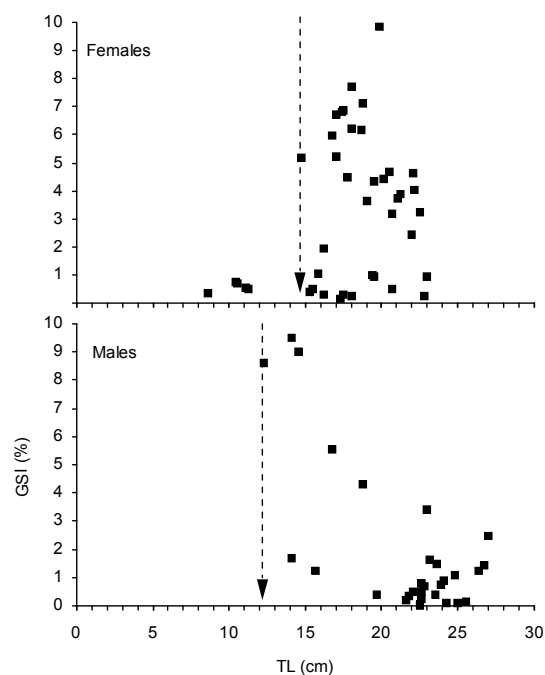


Fig. 3. Individual GSI values as a function of size, for each sex.

### *Reproductive season*

Highest values of GSI were recorded between March and June, the time of the year when the sea water temperature is lowest (Fig. 4). Underwater

observation of reproductive activities also showed more nest building, courtship and spawning behaviours during this period. This observation is consistent with the capture of the first juveniles in July. Differences in colouration were related to

the sex and size of the specimens. Mature females and small males presented a similar colouration, whereas large males were very distinct. Initial and terminal phase males (*sensu* WARNER & ROBERTSON 1978) therefore exist in this species.

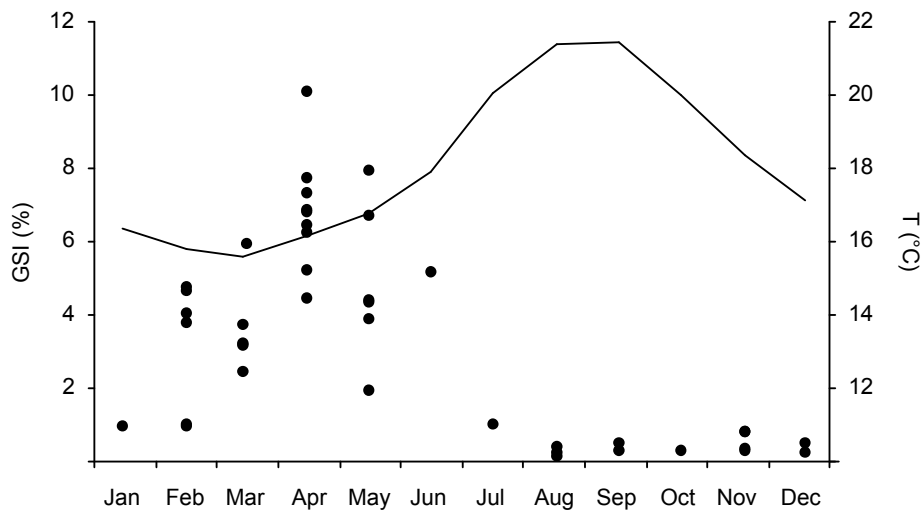


Fig. 4. Monthly values of female GSI (dots) and of sea water surface temperature (line, daily means for Ponta Delgada, São Miguel Island, 1993 to 1995. Source: INMG).

#### *Sexual dichromatism*

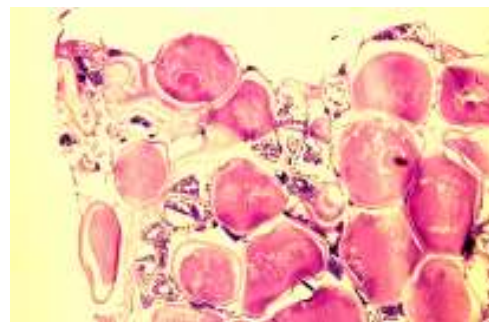
The colour pattern of the females is described based on a ripe female as confirmed histologically (Fig 5a, b). General appearance was brown. The eye had a blue iris surrounded by an orange circle. The lips were white. The head was dark brown dorsally and pale rose with small dark spots ventrally. A dark brown line originated near the mouth and curved upwards around the sides of the snout, ending close to the eye. In some other specimens, additional grey-blue lines were also present on the snout. Body scales were pale brown with a grey-blue spot in the centre. The body had a chequered pattern, although some other specimens appeared rather mottled. The dorsal and middle region of the body was darker, showing three longitudinal bands. Ventrally, the pattern was less well marked. The scales along the ventral aspect of the body were white or pale pink, some of them having a dark brown centre. A conspicuous black spot was present on the

caudal peduncle, below the lateral line and near the caudal fin rays. Genital papilla prominent, with a blue base and a white tip. The colour of the genital papilla ranged from light to very dark blue in other specimens.

A typical terminal phase male whose sex was confirmed histologically, had a blue general appearance (Fig. 5c, d). The eye had a blue iris surrounded by a white circle. The mouth was blue, but in most other terminal males it was contrastingly white. The head was blue, darker dorsally. The area around the eyes was greyish. Body scales were blue, with a light brown centre. The body had a chequered dark grey pattern, similar to that of the females, but without spots on the ventral region. The spot on the caudal peduncle was also less visible. It had no developed genital papilla. All fins were blue, the dorsal and anal presenting large dark-grey spots aligned with the body pattern. This colour pattern was only found on males larger than 21 cm.



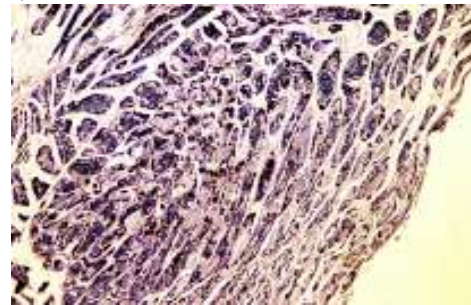
a)



b)



c)



d)

Fig. 5. Colour patterns of *Centrolabrus caeruleus*: mature female, specimen D68, May 97, 19.5 cm TL (a), with ripe gonads (b); territorial, terminal phase male, specimen D64, May 97, 27.0 cm TL (c) with ripe gonads (d).

### Nesting

Only the terminal phase males were observed building algal nests. The algae were either ripped off the substrate or collected from the water column with the mouth. The animals could momentarily release the plants and grab them again, showing a tendency to hold the plants by their base. The algae were collected near the nest, usually not farther than 6 m away. The algae were pushed deep amongst the nest material with the mouth, aided by lateral movements of the body and the propulsion of the caudal fin.

Nests were built on crevices or on small gullies between adjacent boulders. They were elongated in shape (maximum width 20 cm, length between 24 - 55 cm). There was a marked zonation of algal disposition in the nest (see Fig. 6), and some characteristic structural peculiarities. In the more exposed area there was an algal slope where *Taonia* and *Dictyota* predominated. This was called the "yellow" area. This slope levelled off towards the inner part of the nest, which was formed mainly of erect

coralline algae (*Corallina* and *Amphiroa*) that was termed the "pink" area. These two areas differed in texture as well. The yellow area was soft, while the pink area was harder and rough, due to the calcareous, branching nature of the coralline algae. Table 3 gives the species composition of both areas of one particular nest. *Asparagopsis armata*, a red alga with multi-hooked branches, was an important component of the yellow part of this nest. Fish eggs were found attached to the algae, mainly on the yellow area.

Table 3.

Percent algal composition (wet weight, in g) of the "yellow" and "pink" areas of one nest of *Centrolabrus caeruleus*.

	"Yellow" area	"Pink" area	Total
<i>Taonia atomaria</i>	41	6	24
<i>Dictyota dichotoma</i>	30	5	18
<i>Halopteris scoparia</i> , <i>H. filicina</i>	15	65	40
<i>Corallina elongata</i> , <i>Amphiroa</i>	4	24	14
<i>Asparagopsis armata</i>	10	-	5

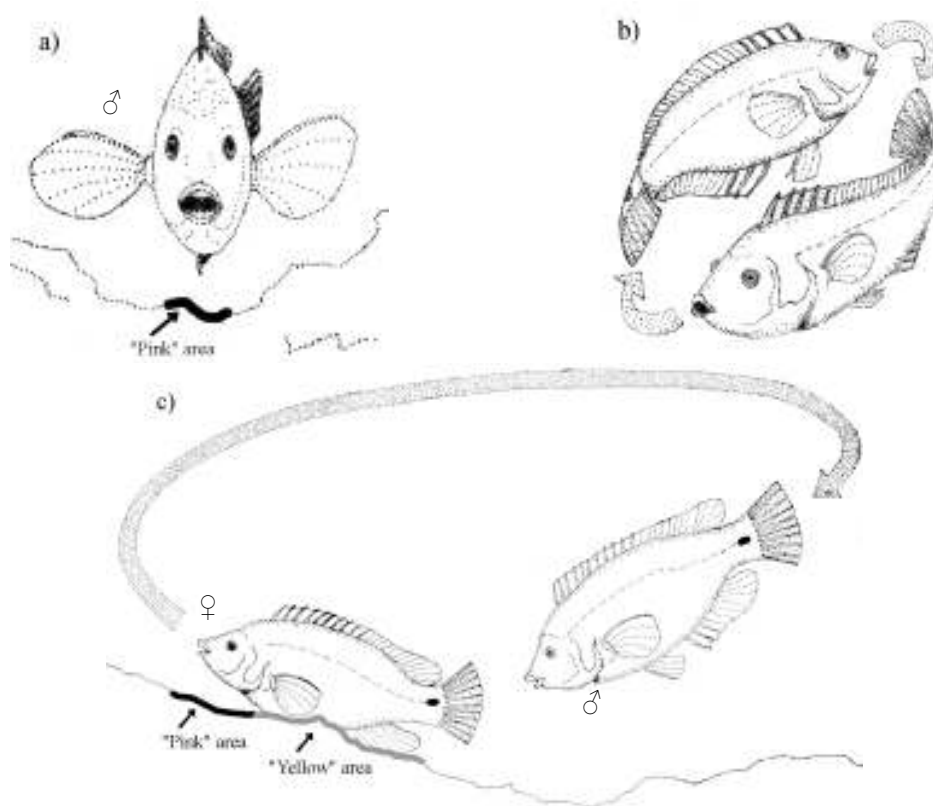


Fig. 6. Typical behaviours of territorial males of *Centrolabrus caeruleus*: (a) aggressive frontal display; (b) tail-to-tail chase of female, in front of the nest; (c) circling in and out of the nest, with female.

The nesting male was always seen at or near the nest, except for brief periods. They showed aggressive behaviour, chasing and biting other fish that came close to the nest. This behaviour was directed towards conspecifics and other species, such as *Coris julis*, *Symphodus mediterraneus*, *Thalassoma pavo* and *Chromis limbata*. On other occasions, the nesting male would face a threat with a frontal display (Fig. 6a). He would place himself over the nest, pectorals spread wide and beating slowly, with the mouth wide open and with exaggerated opercular movements. The white "O" shaped mouth contrasted markedly with the dark blue body, making the male appear larger than it actually was.

#### *Courtship and spawning behaviour*

Females approaching the nest were chased by the nesting male. This behaviour could lead to a tail-

to-tail chase (Fig. 6b). Eventually, the female entered the nest, touching the yellow area with its belly and being followed closely by the male. Presumably, it was then that spawning and fertilisation took place. This behaviour was repeated several times, the two fishes circling in and out of the nest (Fig. 6c), after which the female went away. The male in some cases repeated the courtship and spawning behaviour immediately, with another female. This species is then clearly polygamous.

Simultaneously with these activities, some initial phase *C. caeruleus* hovered around the nest. Some of these would dive into the nest, trying to place themselves near the spawning female. The nesting male would interrupt the mating activities to bite and chase them. These intruders were likely initial phase males, with a streaking strategy (*sensu* WARNER & ROBERTSON 1978). Fishes belonging to other species were

also attracted to the nest during the spawning activity. Once spawning was over, all other fishes, except the terminal male would gradually go away. Only after this would the nesting male resume its normal nest building and repair activities.

## DISCUSSION

Females of *Centrolabrus caeruleus* were seen spawning from late February into July, indicating a prolonged spawning season. It is therefore likely that the range of values of female GSI observed each month is caused by females having several spawning cycles within a season, as documented in related species (e.g. TABORSKY et al., 1987, for *Symphodus ocellatus*). The different modal lengths of the recruits in 1994 and in 1995 also suggest some inter-annual fluctuation of the reproductive period. In any case, the low values of GSI from July onwards indicate that reproduction is centred in spring, when the water temperatures are low, and not in late spring and summer, as stated by PORTEIRO et al. (1996).

The relatively small size of the present data set affected particularly the determination of the von Bertalanffy parameters, and led us to use a non-standard approach. The present results are therefore preliminary, and age-at-length extrapolation for age classes other than 0+ is probably not reliable. In the present context of very little information about the biology of this species, however, even this rough estimate of age-at-length can be useful to generate working hypothesis, as will be seen below.

The observed growth rate of 0+ individuals, and the determined size at maturity, indicate that both sexes can reach maturity in less than one year. At least some individuals can, apparently, reproduce in the season following that in which they were born.

The present results confirm the assertion of AZEVEDO (1999) that only the large, nest-building, males show the blue (terminal) colour pattern. It is not, therefore, a pattern common to all larger specimens, regardless of sex, as assumed by PORTEIRO et al. (1996). The computed von Bertalanffy growth curve suggests that the youngest terminal phase males (TL=21

cm) belong to the 2+ cohort, having therefore spent at least one reproductive season as initial phase males.

Labrids present a wide diversity of reproductive strategies: some species are sequential (protogynous) hermaphrodites while others are gonochoristic, some build and tend nests while others have planktonic eggs, some have sexual dichromatism while others are monochromatic (see WARNER & ROBERTSON 1978, THRESHER 1984, COSTELLO 1991). In this group of fishes, a situation such as that observed in *C. caeruleus*, of initial and terminal phase males, with the latter being chromatically distinct from and larger than the rest of the population, can occur mainly in two situations. If the species is gonochoristic, males will have to have a higher growth rate than the females, changing into the terminal phase colour pattern after reaching a threshold size. In a protogynous species, the larger terminal phase males could also originate from sex-changing females. This would explain the absence of larger females with no need to postulate sexual differences in growth rates.

All species of tropical Labridae whose reproduction has been studied in any detail are pelagic spawners and protogynous hermaphrodites (THRESHER 1984). It is only in temperate areas that gonochoristic species have been found, and (at least in the eastern Atlantic and Mediterranean) these are all demersal spawners with some degree of parental care by the male. Using the temperate genus *Symphodus*, WARNER & LEJEUNE (1985) tentatively concluded that the extensive male mating investment limits the extremes of male reproductive success, and thus reduces selection for protogynous sex change. Thus, the large males of *S. melanocercus* spawn at a much higher rate than small males, in part because there is no nest building or defense. In this species, sex change is apparently common. On the other extreme, in species like *S. ocellatus* and *S. roissali*, in which the larger males construct elaborated nests, ventilate the eggs, and engage in territorial defence (especially against smaller, non-nesting, males), sex change appears to be absent.

In this perspective, it seems likely that *C. caeruleus* is also a gonochoristic species. The



available data (e.g., lack of histological indications of sex change, and intensive parental care of nesting males) support this hypothesis. There is probably a lower size limit for effectively building and defending a nest, and males large enough to do this are also those with the terminal phase colouration. Smaller males are nevertheless sexually mature, and they have several alternative reproductive strategies available. The present observations suggest that at least some of them adopt a streaking behaviour, parasitizing the nest as the female is spawning. It is likely that small males with this behaviour are also those with the relatively large testes, since this may be seen as an adaptation increasing their spawning success. This has been shown in *Symphodus ocellatus* by TABORSKY et al. (1987) and BENTIVEGNA & BENEDETTO (1989).

Further work is necessary in order to fully understand the reproductive biology of *C. caeruleus*. This should involve histological description of the annual gonadal cycle, and its relationship with different colour patterns. Only then can the different sexual stages be positively identified, and field observations of reproductive roles of individual fish be accurately interpreted.

*Centrolabrus caeruleus* is one of a series of medium-sized fishes that swim near the bottom, over the rocky reefs of the Azores, selectively feeding on benthic invertebrates (see revision in AZEVEDO, 1995). Of these, other labrids such as *Symphodus mediterraneus*, *Thalassoma pavo* and *Coris julis*, have similar feeding strategies. Unpublished data by one of the authors (J.B.C.R.) indicates that there is some degree of niche separation in this respect, with *C. caeruleus* and *T. pavo* feeding predominantly on amphipods and *C. julis* concentrating on gastropods. These observations need to be confirmed and placed against the background of the benthic fauna present in this kind of habitat, but this information is not available at present.

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