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TÜBİTAK

Research Article

External morphology of European seabass (*Dicentrarchus labrax*) related to sexual dimorphism

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Abstract: The relationships among 15 morphometric measurements in 219 aliquots of European seabass (*Dicentrarchus labrax*) were examined with respect to sexual dimorphism. The ratio of females to males was determined as 1:5.1 and gonadosomatic indices (%) were found for males (2.13 ± 0.86) and females (3.63 ± 1.18). There were 4 morphometric characteristics that differed between genders: ventral margin of cleithrum (VMC), postanal fin length (post-AFL), postdorsal fin length (post-DFL), and preanal fin length (pre-AFL). Although there were significant differences between morphometric characteristics (post-AFL, post-DFL, pre-AFL) and total length, no significant differences were found in the VMC. These average ratios were determined as pre-AFL to TL 0.61 ♀, 0.71 ♂; post-AFL to TL 0.72 ♀, 0.84 ♂; post-DFL to TL 0.68 ♀, 0.81 ♂, respectively. The aim of this study was to investigate if sexual differences could affect external morphology in cultured seabass and, additionally, to determine sexual dimorphism related to morphological differences during the maturation and spawning seasons in this species.

Key words: European seabass, *Dicentrarchus labrax*, external morphology, sexual dimorphism

Eşeye bağlı olarak levrek (*Dicentrarchus labrax*) balıklarında gözlenen morfolojik farklılıklar

Özet: Bu çalışmada, 219 adet levrek balığında (*Dicentrarchus labrax*) 15 farklı morofometrik ölçüm arasında eşeye bağlı herhangi bir farklılık olup olmadığı incelenmiştir. Dişi ve erkek oranı 1:5,1 olarak tespit edilmiş ve gonadosomatik indeks (%) erkekler (2,13 ± 0,86) ve dişiler (3,63 ± 1,18) için hesaplanmıştır. Yapılan incelemeler sonucunda 4 morfometrik karakterin (kleithrumun ventral kenarı, postanal yüzgeç uzunluğu, postdorsal yüzgeç uzunluğu, preanal yüzgeç uzunluğu) dişi ve erkek bireyler arasında farklılık gösterdiği tespit edilmiştir. Bununla birlikte, bu 4 morfomedrik karakter total boya'ya (TB) oranlanması sonucunda elde edilen değerlerin 3 tanesi istatistikî olarak önemlilik göstermiştir. Bunlar pre-AFL/TB 0.61 ♀, 0.71 ♂; post-AFL/TB 0.72 ♀, 0.84 ♂; post-DFL/TB 0.68 ♀, 0.81 ♂, olarak tespit edilmiştir. Dişi ve erkek levrek balıklarının üreme dönemine bakılmaksızın eşeye göre ayrımı bu çalışma ile elde edilen sonuçlar kullanılarak mümkün olabilmektedir.

Anahtar sözcükler: Levrek, *Dicentrarchus labrax*¸ morfoloji, eşey farklılaşması

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Introduction

It is well known that sex-determination processes in fish reveal complex patterns of variables and mechanisms utilized among different fish species. Over the last 30 years, numerous studies have been carried with regard to gonadal types, hermaphroditism, genetic sex determination, and the influence of sex steroids on sex differentiation. Studies about *D. labrax* have been generally focused on effects of abiotical factors on sexual determination and differentiation (Pavlidis et al., 2000; Koumoundouros et al., 2002; Begtashi et al., 2004). The present study constitutes the first attempt at identifying sex determination by using external morphology in cultured *D. labrax*.

The European seabass is a gonochoristic species with the gonad remaining sexually undifferentiated until the end of the first year of life (7-12 months of life) (Roblin and Bruslé, 1983; Blázquez et al., 1995). The first primordial germ cells of the gonads appear at 10.6 mm standard length (SL), but gonads become differentiated only after 90-120 mm SL (Roblin and Bruslé, 1983; Blàzquez et al., 1999). However, in fish maintained under intensive culture conditions, the sex ratio is consistently skewed in favor of males (reaching in some instances values over 90%) and a significant proportion of precocious males (males that mature soon after gonadal differentiation) is always present (Blázquez et al., 1995, 1999; Carrillo et al., 1995). This is undesirable for fish farmers, because male fish exhibit reduced somatic growth, resulting in an 18%-40% smaller body weight at 2 years of age (Carrillo et al., 1995). Therefore, during the last 10 years, for both scientific and economic reasons, little research has focused on the genetic and endocrine basis of sexual differentiation in European seabass (Chatain et al., 1997; Koumoundouros et al., 2002).

Morphometric data can be used in the biological evaluation of a species, especially if viewed in terms of its ontogenetic interactions between rearing conditions. For example, growth patterns of morphometric characters have been used to identify developmental thresholds in the ontogeny of several fish species (Kovac et al., 1999). Significant advances in morphometric analysis have occurred in the last 2 decades, offering more efficient and powerful techniques, such as image analysis (Cadrin and Friedland, 1999) and geometric landmark methods (Rohlf and Marcus, 1993) for detecting shape variation within groups. However, traditional methods naturally include several advantages for measurements and analysis (Adriaens et al., 2005).

There have been several papers dealing with European seabass morphometry (Whitehead and Wheeler, 1966; Pickett and Pawson, 1994), providing mainly descriptive morphology with implications for the systematic position of the species and/or its interpopulation variability. Some of these studies specifically focused on sexual growth dimorphism (Saillant et al., 2001); however, to date, no specific study has been carried out regarding the differentiation of morphometric characters on the sex assumption in this species. The objective of the present study was to analyze morphometric characters in European seabass (*Dicentrarchus labrax*) and determine whether they could be used for sex discrimination.

Materials and methods

A total of 219 European seabass specimens (length ranging between 29.7 and 37 cm) were sampled from commercial on-growing cage farms (19 February 2008). The left side of each European seabass sample was photographed using a digital camera (Nikon Coolpix 5000) and then sex was determined by visual examination of the gonads in vivo. The samples consisted of 38 females (29.6-36.4 cm) and 181 males (30.5-36.9 cm). These were killed with an overdose of benzocaine, and weighed (±0.01 g), and measurements were taken for 15 morphometric characters (Figure 1 and Table 1). The morphometric characters were measured using TpsDig software (ver. 1.37; Rohlf, 2004; http://life.bio.sunysb.edu/morph) with 0.01 mm on the photographs. Gonads were dissected and weighed (±0.01 g), and the gonadosomatic index (GSI = 100 × gonad weight/body weight) was calculated.

To examine sexual differences in morphometric measurements, each variable was plotted against TL and the regression slope was visualized in graphics. The differences in the regression slope between males and females were tested by ANCOVA (intercepts correlation coefficient was not analyzed due to

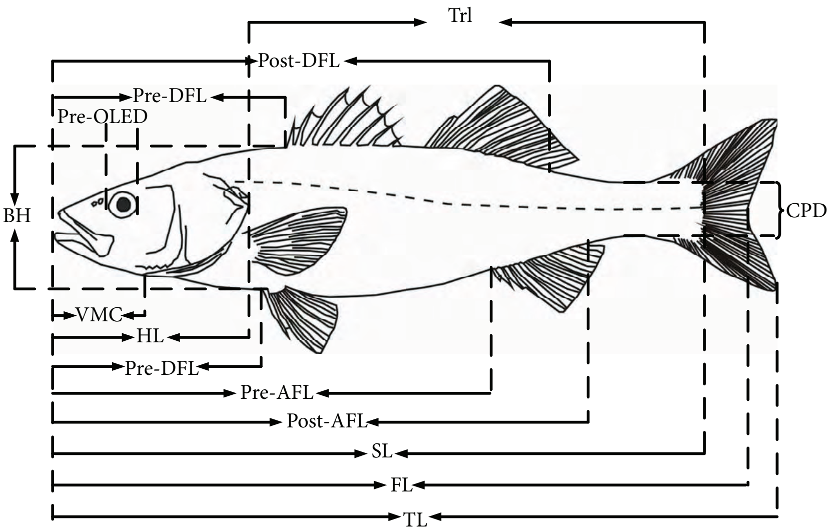

Figure 1. Morphometric measurements taken on *Dicentrarchus labrax* individuals. Morphometric abbreviations are listed in Table 1.

Table 1. Description of measured morphometric character of *Dicentrarchus labrax*.

Abbreviations	Character	Description on Fish
TL	Total Length	From tip of snout to the end of the tail
FL	Fork Length	From tip of snout to the fork of the tail
SL	Standard Length	From tip of snout to base of caudal fin rays
PreAFL	Pre-Anal Fin Length	From tip of snout to anterior margin of the anal fin base
PostAFL	Post-Anal Fin Length	From tip of snout to posterior margin of the anal fin base
PostDFL	Post-Dorsal Fin Length	From tip of snout to posterior margin of the dorsal fin base
PrePFL	Pre-Pelvic Fin Length	From tip of snout to the base of the pelvic fin
PreDFL	Pre-Dorsal Fin Length	From tip of snout to anterior margin of the dorsal fin base
CPD	Caudal Peduncle Depth	Minimum depth of the caudal peduncle
ED	Eye Diameter	Paralel to the longitudinal axis of the body
VMC	Ventral Margin of Cleithrum	From tip of snout to ventral margin of the cleithrum
PreOL	Pre-Orbital Length	From tip of snout to anterior margin of the eye
HL	Head Length	Posterior to gill cover
BH	Body Height	Maximum depth of body
TrL	Trunk Length	From gill cover to anterior margin of the caudal fin

insignificance), and variance analysis of differences between indices of the measured morphometric characteristics and TL between males and females were tested by Student's t-test. Statistical analysis and graphics were generated by SPSS 15.0 software.

Results

Metric characters for females and males in *D. labrax* are given in Table 2. Although whole samplings were collected at the end of February, all gonads from the specimens examined had differentiated into males and females, and only the females' gonads were functional. The sex ratio of female and male was determined as 1:5.1. Gonadosomatic indices (%) of European seabass were estimated for males (2.13 ± 0.86) and females (3.63 ± 1.18).

Regression slope analysis revealed that 4 morphometric characters (VMC, post-AFL, post-DFL, and pre-AFL) were significantly different between males and females (ANCOVA, $P < 0.05$)

Table 2. Meristic character for females and males of *Dicentrarchus labrax*. Morphometric abbreviations are listed in Table 1 (Female = 38; Male = 181).

Character	Sex	Mean (mm)	Min. (mm)	Max. (mm)	Std. Dev.
TL	F	32.89	29.66	36.40	1.341
	M	34.27	30.49	36.99	1.505
FL	F	31.53	28.32	34.53	1.290
	M	32.81	29.28	35.73	1.508
SL	F	28.95	26.66	31.73	1.143
	M	29.84	26.44	32.58	1.465
HL	F	7.93	6.70	9.40	0.648
	M	7.98	6.39	9.33	0.522
TrL	F	20.98	19.27	24.63	1.044
	M	21.89	19.00	27.50	1.302
Pre-AFL	F	20.07	18.36	22.27	0.966
	M	20.93	18.33	25.89	1.120
Post-AFL	F	23.66	21.14	26.00	0.982
	M	24.91	20.82	27.59	1.276
Pre-PFL	F	9.41	7.97	10.63	0.522
	M	9.40	7.76	10.61	0.565
VMC	F	4.56	3.22	5.84	0.652
	M	4.69	3.06	6.00	0.472
ED	F	1.07	0.64	1.39	0.195
	M	1.38	0.94	1.91	0.178
Pre-OL	F	2.26	1.37	3.16	0.388
	M	2.06	1.30	2.88	0.281
Pre-DFL	F	10.86	9.94	12.28	0.654
	M	10.85	8.80	12.46	0.658
Post-DFL	F	22.49	20.18	24.50	0.795
	M	23.78	21.02	26.52	1.172
BH	F	7.40	6.58	8.41	0.463
	M	7.54	6.23	8.73	0.469
CPD	F	2.90	2.48	3.46	0.206
	M	2.97	2.52	3.55	0.168

(Figure 2). However, pre-AFL/TL, post-AFL/TL, and post-DFL/TL were found to be significantly different between sexes, although VMC/TL (Table 3) was not significant. The average pre-AFL to TL ratio was calculated as 0.61 (range: 0.57-0.64; SD: 0.01) for females and 0.71 (range: 0.64-0.87; SD: 0.17) for males, respectively ($P < 0.05$). Similarly, post-AFL to TL ratios of the females and males were on average 0.72 (range: 0.68-0.76; SD: 0.01) and 0.84 (range: 0.73-0.89; SD: 0.16), respectively ($P < 0.001$). However, the ratio of post-DFL to TL was calculated as 0.68 (range: 0.64-0.72; SD: 0.02) for females and 0.81 (range: 0.73-0.94; SD: 0.16) for males, respectively ($P < 0.001$).

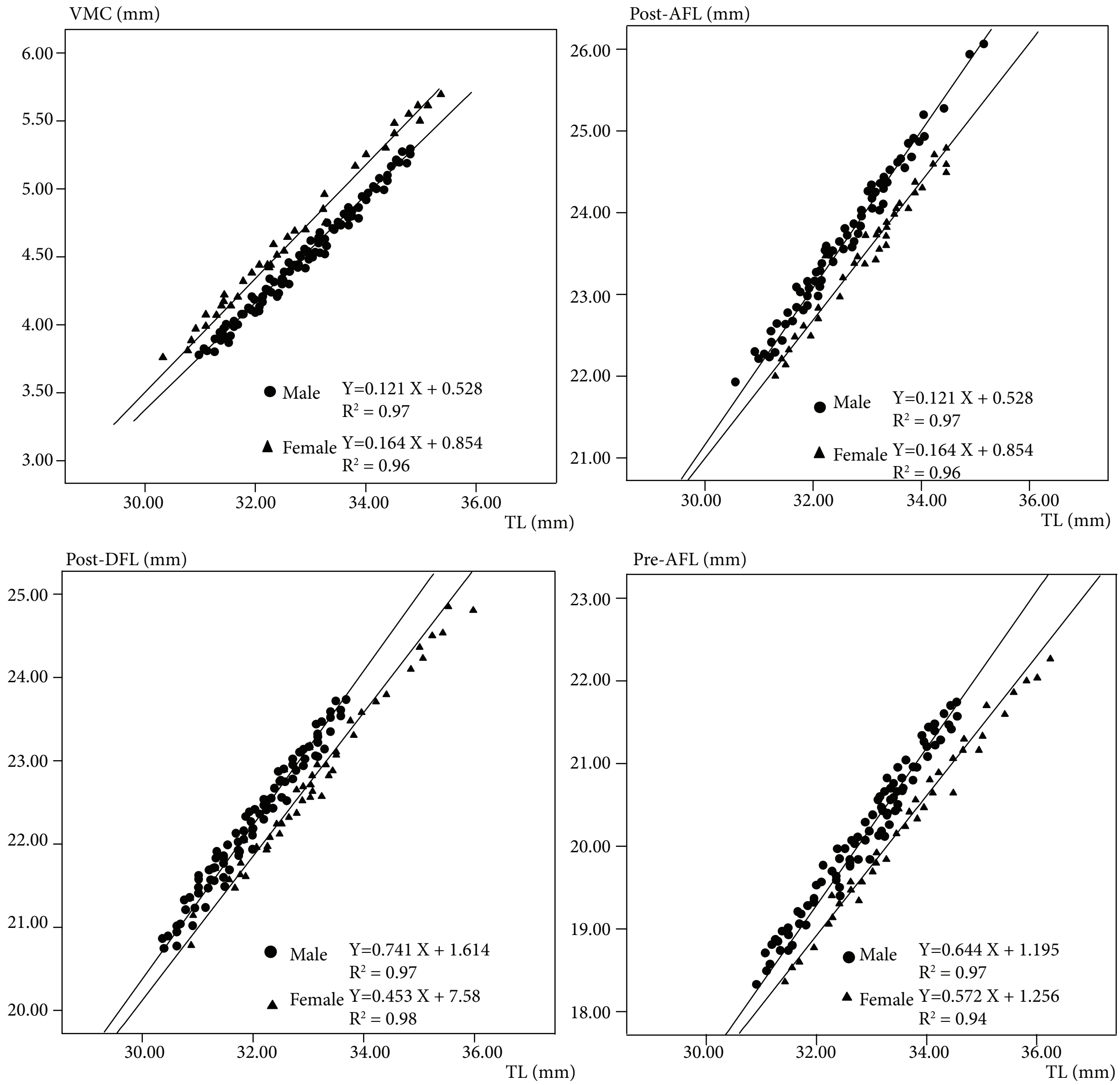

Figure 2. Regression slope analysis of males (●) and females (▲) against total length in *Dicentrarchus labrax*.

Table 3. Proportion of the measured morphometric characters against TL. Morphometric abbreviations are listed in Table 1.

Character	Sex	N	Mean	Min.	Max.	Std. Dev.	*t*-test
FL/TL	F	38	0.9588	0.94	0.97	0.01184	
	M	181	0.9574	0.92	0.98	0.00852	
SL/TL	F	38	0.8802	0.85	0.93	0.01862	
	M	181	0.8689	0.24	0.96	0.04821	
HL/TL	F	38	0.6380	0.59	0.70	0.02393	
	M	181	0.6409	0.60	0.78	0.01853	
TrL/TL	F	38	0.2411	0.20	0.28	0.01764	
	M	181	0.2342	0.20	0.67	0.03473	
Pre-AFL/TL	F	38	0.6103	0.57	0.64	0.01785	*
	M	181	0.7111	0.64	0.87	0.17657	
Post-AFL/TL	F	38	0.7194	0.68	0.76	0.01695	**
	M	181	0.8448	0.73	0.89	0.16831	
Pre-PFL/TL	F	38	0.2865	0.25	0.32	0.01495	
	M	181	0.2767	0.24	0.74	0.03618	
VMC/TL	F	38	0.1385	0.10	0.17	0.01839	
	M	181	0.1366	0.09	0.16	0.01246	
ED/TL	F	38	0.0327	0.02	0.04	0.00648	
	M	181	0.0400	0.03	0.05	0.00487	
Pre-OL/TL	F	38	0.0688	0.04	0.09	0.01095	
	M	181	0.0600	0.04	0.08	0.00715	
Pre-DFL/TL	F	38	0.3304	0.30	0.37	0.01713	
	M	181	0.3156	0.28	0.36	0.01332	
Post-DFL/TL	F	38	0.6844	0.64	0.72	0.01831	**
	M	181	0.8073	0.73	0.94	0.15988	
BH/TL	F	38	0.2250	0.20	0.25	0.01149	
	M	181	0.2189	0.19	0.25	0.01119	
CPD/TL	F	38	0.0865	0.08	0.10	0.00336	
	M	181	0.0881	0.08	0.10	0.00419	

*0.01 < P < 0.05; **0.01 < P < 0.001

Discussion

Sexual growth dimorphism is widely encountered in teleost fish. It is generally recorded that, at adult size, males are found to be larger than females. For instance, this has been observed in many tilapias (Toguyeni et al., 1997); in salmonids such as *Oncorhynchus mykiss*, *Salmo trutta* (Bonnet et al., 1999), and *O. kisutch* (Fleming and Gross, 1994); and in silurids such as the American and European catfishes *Ictalurus punctatus* and *Silurus glanis* (Haffray et al., 1998). Conversely, several papers demonstrated that females are larger than males in many flatfish such as dab, *Limanda limanda* (Lozan, 1992),and turbot, *Scophtalmus maximus* (Imsland et al., 1997); in the perch, *Perca fluviatilis* (Fontaine et al., 1997); and in some cyprinids, including *Cyprinus carpio* (Hollebecq and Haffray, 1994). *D. labrax* is already known to exhibit sexual growth dimorphism at commercial size (300-400 g). The females are larger than males at this stage of development with a relative advantage estimated at 20%-40% (Carrillo et al., 1995; Chatain et al., 1997). However, our results, obtained from the identical age group, demonstrated that there were no significant differences in terms of total length and weight between genders. Saillant et al. (2003) reported that high densities and size grading applied to intensive seabass are not responsible for the sexual dimorphism in farmed populations. However, the feeding rates and growth conditions of fish may account for a part of the sex ratio variation. In the very early life stage of the European seabass, before sexual maturation, females start growing faster than males (Sailant et al., 2003). According to Roblin and Brusle (1983) and Blazquez et al. (1998), morphological sexual differentiation should not have begun at the stage of the first and second size grading (5.1 and 8.9 cm, respectively). It is thought that this situation could have originated by size grading in the hatcheries and on-growing cages, in order to prevent cannibalism under culture conditions.

Studies focusing on sex determination in fish are important both for ecologists and aquaculturists, as morphometric characters have been used as a tool to identify developmental thresholds and sexual dimorphism in ontogeny of several species of fish in both culture and wild stocks, such as *Rutilus rutilus* (Copp and Kovac, 1996), *Barbatula barbatula* (Kovac et al., 1999), *Perca fluviatilis* (Sediva et al., 2000), *Lepomis gibbosus* (Sumer et al., 2005), and *Pagrus pagrus* (Minos et al., 2008). Moreover, several studies related to sexual dimorphism have focused on sex allometric growth (Minos et al., 2008) and/or sexual size dimorphism (Ji et al., 2006), and have also been designed in order to determine physiological changes occurring in spawning season in experimental fish. In this study, although pre-AFL, post-AFL and post-DFL indices changed with intraspecific population, it could be used for determination of sexual dimorphism in this species. Additionally, Minos et al. (2008) reported that 6 morphometric characters (fork length, head height, trunk length, postorbital distance, pre-orbital distance, and standard length) were significantly different between females and males in the red porgy *P. pagrus*. However, head height and preorbital distance had a higher growth rate in males, and also there was a higher postorbital distance increase in females. Siryova (2004) reported that 22 of 43 measured morphometric characters were found to be different between genders in *Alburnoides bipunctatus*. A similar result has also been observed in *Lepomis gibbosus*, where predorsal fin distance was different for both females and male individuals (Sumer et al., 2005).

Results obtained from this study show that there are morphological differences between males and females in *D. labrax* in pre-AFL, post-AFL and post-DFL indices, which could be used for determination of sexual dimorphism. The morphometric analysis supported that indicated changes in some morphologic characters could be observed during the formation of sex characters. As mentioned in previous studies, determination of these differences could be used for selection of female fish in the population both for ecologists and aquaculturists. In respect to aquaculture, modifications in rearing strategies to improve the profitability of seabass cultures could be undertaken by: (1) the selection of strains carrying genetic traits such as a) high survival, b) accelerated growth rate, c) disease resistance, d) age at first maturity, or e) skewed female sex ratio; (2) the induction of sterility, production of all-female culture, or the combination of both, and (3) the prevention or delay of male first maturation (puberty). It could be concluded that scientific approaches about sexual dimorphism could possibly be used in order to select a broodstock in the second year of life, for selection in cage culture due to rapid growth of females, and to prevent cannibalism depending on size differentiation. Further work should be undertaken in the future for the determination of sexual dimorphism during the relatively early life stages of fish.

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