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ORIGINAL RESEARCH

The effects of freshwater rearing on the whole body and muscle tissue fatty acid profile of the European sea bass (*Dicentrarchus labrax*)

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Abstract The purpose of this study was to investigate the effect of freshwater rearing on the fatty acid profiles of the whole body and muscle tissue of the European sea bass (*Dicentrarchus labrax*). Half of initial fish were gradually acclimated to freshwater (FW) kept at the same temperature to salt water and grown in same conditions as their counterparts in saltwater (SW). The decrease in salinity caused an increase in the percentages of 18:1$n-9$, 24:1$n-9$, 18:3$n-3$, 18:2$n-6$ and decrease in the percentages of 14:0, 15:0, 20:0, 21:0, 20:5$n-3$ and 22:6$n-3$ both in the whole body and in the muscle tissue fatty acid profiles. The lipids of FW-reared fish contained significantly ($P < 0.01$) higher percentages of 18:2$n-6$ and 18:3$n-6$ than that of SW-reared fish. However, percentages of 20:5$n-3$ and 22:6$n-3$ fatty acids decreased significantly ($P < 0.05$) compared with those of salt water-reared European sea bass. There was a clear trend of decrement in the percentages of $n-3$ PUFA fatty acids due to the decrease in water salinity. However, the percentages of $n-6$ PUFA fatty acids were also increased with the decrease in water salinity. We concluded that the FW acclimation is followed by changes in certain lipid classes of sea bass muscle tissue and whole body samples. $n-3/n-6$ PUFA ratios were characteristic to previously reported ratios for both FW- and SW-reared European sea bass. In addition, EPA/DHA ratios were basically similar for the fish reared in both SW and FW indicating the equal nutritional value of the final products in terms of providing PUFA's for human nutrition.

Keywords Fatty acids · Freshwater rearing · European sea bass · *Dicentrarchus labrax* · Lipid composition

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Introduction

Lipids play an important role in fish nutrition for the provision of energy and essential fatty acids (Sargent et al. 1989). Lipids of marine fish species are generally characterized by low levels of linoleic acid (18:2$n-6$) and alpha-linolenic acid (18:3$n-3$) and high levels of long-chain $n3$ polyunsaturated fatty acid (Steffens 1997). Polyunsaturated fatty acids like eicosapentaenoic acid (EPA, C20:5$n-3$) and docosahexaenoic acid (DHA, C22: 6$n-3$) are of great importance to human beings for prevention of coronary artery diseases (Conner 2000; Mozaffarian et al. 2005) and fish is the most important dietary source supplying these fatty acids to humans. Fish lipids are a good source of EPA and DHA. Among the polyunsaturated fatty acids, EPA and DHA are the dominant $n3$ fatty acids in marine fish species (Ackman 1989). Freshwater fish, however, contain high levels of C18 PUFA and low levels of the $n3$ EPA and DHA compared to marine fish species.

The European sea bass, *Dicentrarchus labrax* is the most popular and valuable euryhaline species for the commercial aquaculture in the Mediterranean. This species can survive in a wide range of environmental salinity levels from open sea to the brackish water river deltas and to the lagoons (Conides and Glamuzina 2006; López-Olmeda et al. 2009). It has previously been shown that the European sea bass could even tolerate direct transfer from 37‰ seawater to brackish water of 3.9‰ salinity, and also to waters of as low as 0.5‰ salinity if salinity is gradually reduced over 24 h (Rubio et al. 2005; López-Olmeda et al. 2009). Due to the lack of sheltered bays suitable for cage culture in some part of the Mediterranean and pollution risk of seawater have been forcing cage farmers to consider estuaries, and even freshwater resources. Therefore, farming this species partly in inland waters could create a chance to better utilization of marine environment (Zanuy and Carrillo 1985).

The fatty acid composition of fish species can be substantially affected by the diets, sex and environmental conditions (Chen et al. 1995; Alasalvar et al. 2002). Earlier it has been reported that the fatty acid compositions of fish reflect that of different environmental conditions (Steffens 1997; Kheriji et al. 2003; Haliloglu et al. 2004; Conides and Glamuzina 2006). Water salinity was demonstrated to have an important effect on fatty acid composition particularly PUFA levels of fish and the $n3/n6$ ratio was much lower in fish living in freshwater than their counterparts in saltwater (Steffens 1997). However, the effects of freshwater acclimation on growth and whole body and fillet proximate compositions of sea bass needs to be fully understood before beginning of commercial rearing in brackish and freshwater environments. Therefore, the aim of this study was to demonstrate the effects of freshwater acclimation of the European sea bass on the whole body and muscle fatty acid profiles and muscle tissue proximate composition.

Materials and methods

Fish and the maintenance

This study was conducted at the Marine Research Station of The Faculty of Fisheries, University of Cukurova, Turkey. *Dicentrarchus labrax* fingerlings (Average mean weight of 0.83 ± 0.22 g. $n = 300$) were obtained from a commercial farm (Akuvatur, Tuzla, Adana, Turkey) and raised in a stock tank until they reach approximately 5 g. Then fish were separated into two groups and designated as seawater (SW) and freshwater acclimated groups (FW). Fish that will be used in freshwater growth trial were acclimated to

freshwater by gradually reducing the water salinity. To acclimate fish to freshwater, water salinity was decreased gradually during 7 days at the rate of 5 ppt in each day by adding freshwater to seawater. During the initial rearing and freshwater acclimation period, fish were both fed by a commercial diet (Çamli/İzmir/Turkey, Granules No: 1.5 and 55% crude protein, 18% crude lipid, 12% moisture and 13% ash).

The experiment was conducted in 6,210 l indoor rectangular tanks (1.5 ml × 0.35 mW × 0.40 mH). 20 fish (average mean weight of 4.80 ± 0.25 g.) were stocked to each tank. Each treatment was replicated three times and natural photoperiod was utilized throughout the experiment. Each tank was continuously supplied with flow through sand filtered freshwater (0.4‰) and seawater (40‰) at a flow rate of 2 l min^{-1}. During the experiment, fish were fed with a commercial European sea bass diet (Çamli/İzmir/Turkey, No. 2 containing 48% crude protein, 18% lipid, 12% moisture and 10% ash) until the 90-day grow-out period completed. Both groups were fed twice a day between 09:00 and 10:00 h and 17:00–18:00 h at 3% BW. The percentage of fatty acid profiles of feed used in the study is given in Table 1.

During the experiment (90 days), rearing water in each tank was permanently saturated with oxygen using air-stones connected to an air-blower. Daily recordings of physical and chemical parameters of tank water (temperature, dissolved oxygen, pH and salinity) were made at 08.00 h. Dissolved oxygen and pH values of tank water were not allowed to fall below 7 mg l^{-1} and 7.5–7.8, respectively, throughout the experiment. Daily recordings of water physical and chemical parameters were made using specifically designed YSI model 30 salinometer (Yellow Springs Instrument, Yellow Springs, OH, USA), an oxygen meter and a pH meter (pH 315i Set, WTW Measurement Systems, Germany). The average temperature was maintained at 26 ± 2°C throughout the experiment as this temperature range was reported optimum for the European sea bass (Barnabe 1990).

Sampling and the chemical analysis

All fish were individually measured for weight before the experiment and end of the experiment. Fish in each tank were also batch weighed for every 2 weeks and feeding rate was adjusted accordingly throughout the experiment.

At the end of the 90-day grow-out period, five fish from each tank were randomly sampled for muscle tissue proximate analysis. Fish were fasted 24 h before slaughter. For the fatty acid profiles of the whole body and muscle tissue, ten more fish were also sampled from each tank. The whole body and muscle tissue samples were homogenized in a blender and stored at −20°C until the chemical analysis. Prior to analysis samples were thawed at +4°C for 24 h. Homogenized and thawed samples from each tank were pooled in equal amounts before proximate and fatty acid analyses were made. Moisture and crude ash contents were according to standard methods (AOAC 1990). Crude lipid content was determined according to Bligh and Dyer (1959). Crude protein content was analyzed using Macro Kjeldahl method ($N \times 6.25$) (AOAC 1990).

Extracted lipids were stored under liquid nitrogen gas at −20°C for the determination of fatty acid profile in the whole body and muscle samples. The fatty acids in the total lipid were saponified into the free form by saponification with 0.5 N methanolic NaOF, followed by esterification with 14% BF_3 (w/v) in methanol (IUPAC 1979). Esterified samples were analyzed using a thermoquest trace gas chromatograph equipped with a Supelco-SP-2330 fused-silica capillary column (30 m × 0.25 mm i.d., 0.20-μm film thickness of polyethylene glycol) (Supelco Inc., Bellefonte, PA, USA) and a flame-ionization detector (FID). Helium (30 ml min^{-1}) was used as a carrier gas. The samples were injected at

Table 1 The percentage fatty acid profile (% of total fatty acids) of feed used in the research

Fatty acids	%
14:0	10.63 ± 0.35
15:0	1.26 ± 0.01
16:0	25.63 ± 0.43
17:0	1.24 ± 0.01
18:0	4.27 ± 0.01
20:0	0.57 ± 0.01
21:0	1.40 ± 0.09
23:0	0.28 ± 0.01
$\sum$ Saturated fatty acids (SFAs)	45.28
14:1	0.30 ± 0.03
16:1	8.65 ± 0.17
17:1	0.73 ± 0.02
18:1$n-9$	17.50 ± 0.03
20:1$n-9$	0.86 ± 0.03
22:1$n-9$	0.86 ± 0.03
24:1$n-9$	0.38 ± 0.06
$\sum$ Monounsaturated fatty acids (MUFAs)	29.28
18:2$n-6$	4.74 ± 0.01
18:3$n-3$	0.94 ± 0.02
18:3$n-6$	0.12 ± 0.01
20:2n11-14c	0.16 ± 0.01
20:4$n-6$	0.85 ± 0.02
20:5$n-3$	6.62 ± 0.38
22:6$n-3$	7.82 ± 0.12
$\sum$ Polyunsaturated fatty acids (PUFAs)	21.25
Unidentified	4.19
$\sum n-3$ PUFA	15.38
$\sum n-6$ PUFA	5.71
$n-3/n-6$	2.69
EPA/DHA	0.85

Values are mean ± SEM and expressed as percentages of total fatty acids (feed samples were analyzed triplicate for fatty acids)

120°C. After 2 min temperature was raised 5°C min^{-1} to 220°C where it was kept for 8 extra minutes. The temperatures of the injector and the detector were set at 240 and 250°C, respectively. Fatty acid methyl esters were identified by comparing their retention times with those of the commercial fatty acid methyl ester standards (FAME mix C4–C24; Supelco LB41302; Code Number: 18919). The relative concentrations of fatty acids were expressed as percentages of their total.

Statistical analysis

Results are reported as mean ± SEM throughout the text. Differences between sea bass reared in SW and FW were analyzed using the Student's *t*-test. The significance was accepted at the probability value of 0.05 or less. The SPSS ver. 9.0 statistical programs were used to evaluate data (SPSS 1993).

Results

Fish seemed healthy during the freshwater acclimation period. Fish started their normal feeding and swimming activities after the 7-day transfer period from SW to FW. The fish grown at SW had significantly higher average growth rate than those at FW in the end of the experiment ($P < 0.05$; Table 2). SW-reared fish had also higher specific growth rate (SGR) than their counterparts in FW. In addition, feed conversion ratio (FCR) was significantly influenced by decreasing salinity. The FW-reared fish had lower FCR and therefore higher SGR than that of SW-reared fish (Table 2).

The fatty acid composition of the sea bass muscle tissue and the whole body samples are presented in Table 3. Dominant fatty acids in both SW- and FW-reared fish were 14:0 (myristic acid), 15:0 (pentadeclic acid), 16:0 (palmitic acid), 18:0 (stearik acid), 20:0 (arachidic acid), 21:0 (henicosonoic acid), 17:1 (heptadeconoic acid), 18:1$n-9$ (oleic acid), 20:1$n-9$ (eicosonoic acid), 18:2$n-6$ (linoleic acid), 18:3$n-6$ (gamma-linolenic acid), 18:3$n-3$ (alpha-linolenic acid), 20:5$n-3$ (EPA), 22:6$n-3$ (DHA; Table 3). SW-reared fish contained significantly higher ($P < 0.05$) proportions of 14:0, 17:1, 20:5$n-3$, 22:6$n-3$ and lower proportions of 18:1$n-9$, 18:2$n-6$, 18:3$n-3$ in muscle tissues than that of FW-reared European sea bass (Table 3). 14:0 and 16:0 were also the major saturated fatty acids (SFA), contributing approximately 80% to the total SFA content of the lipids in both SW- and FW-reared European sea bass muscle tissue samples. Furthermore, the total SFA content of muscle tissue lipids were 35.71 and 35.29% in SW- and FW-reared fish, respectively. However, in the whole body the total SFA content were found to be 38.48 and 31.48% in SW- and FW-reared fish, respectively.

Oleic acid was identified as a primary monounsaturated fatty acid (MUFA) in both muscle tissues and the whole body samples of SW- and FW-reared European sea bass. Moreover, SW-reared fish had significantly ($P < 0.01$) lower amounts of oleic acid than that of FW-reared fish for both muscle tissue and the whole body samples (Table 3). Among $n-6$ series of fatty acids, 18:2$n-6$ was the one of the predominant polyunsaturated fatty (PUFA) in both muscle tissue and the whole body samples of FW-reared fish. FW-reared fish had also significantly ($P < 0.01$) higher amount 18:2$n-6$ than that of SW-reared fish for both muscle tissues and the whole body samples. It appeared that the decrease in water salinity caused an increase in the percentages of 18:1$n-9$, 18:2$n-6$, 18:3$n-3$ and a decrease in the percentages of 20:4$n-6$, 20:5$n-3$, 22:6$n-3$ in both muscle tissues and the whole body samples of the European sea bass (Figs. 1, 2). However,

Table 2 Growth performances of *Dicentrarchus labrax* fingerlings reared in SW and FW for 90 days

	SW	FW
Initial body weight (g/fish)	4.80 ± 0.25	4.80 ± 0.25
Final body weight (g/fish)	39.23 ± 1.84[a]	31.53 ± 1.25[b]
Weight gain (g/fish)	34.43 ± 1.85a	26.73 ± 1.25b
SGR (%/d)	2.32 ± 0.05[a]	2.08 ± 0.04[b]
FCR	1.27 ± 0.85[a]	1.45 ± 1.20[b]

Values are expressed ±SEM of three replicates in each groups

Values in the same row with the different superscript are significantly different ($P < 0.01$)

Weight gain (WG) = final body weight (g)-initial body weight (g); specific growth rate (SGR) = ln final body weight-ln initial body weight (g)/t (time) × 100; feed conversion ratio (FCR) = dry feed fed (g)/weight gain

Table 3 The whole body and muscle tissue fatty acid profile of SW- and FW-reared European sea bass for 90 days

Fatty acids	Muscle tissue			Whole body tissue		
	Seawater	Freshwater	*F*	Seawater	Freshwater	*F*
14:0	6.64 ± 0.21[a]	4.89 ± 0.44[b]	*	8.61 ± 0.60[a]	4.62 ± 0.35[b]	*
15:0	0.84 ± 0.06[a]	0.63 ± 0.02[b]	*	1.01 ± 0.05[a]	0.71 ± 0.09[b]	*
16:0	21.45 ± 084	22.47 ± 0.99	NS	22.42 ± 0.33[a]	20.62 ± 0.27[b]	**
17:0	1.04 ± 0.202	1.12 ± 0.04	NS	0.45 ± 0.04	0.46 ± 0.10	NS
18:0	3.73 ± 0.10[a]	4.55 ± 0.14[b]	**	4.02 ± 0.36[a]	4.36 ± 0.07[b]	*
20:0	0.41 ± 0.03[a]	0.28 ± 0.01[b]	**	0.45 ± 0.02[a]	0.29 ± 0.01[b]	**
21:0	1.25 ± 0.02[a]	1.05 ± 0.01[b]	**	1.22 ± 0.01[a]	0.08 ± 0.01[b]	**
23:0	0.35 ± 0.02	0.30 ± 0.03	NS	0.30 ± 0.01	0.34 ± 0.02	NS
∑SFA	35.71	35.29		38.48	31.48	
14:1	0.24 ± 0.03	0.16 ± 0.01	NS	0.26 ± 0.02[a]	0.14 ± 0.01[b]	**
16:1	6.88 ± 0.25	6.01 ± 0.28	NS	7.71 ± 0.23[a]	6.37 ± 0.16[b]	*
17:1	1.05 ± 0.03[a]	0.52 ± 0.13[b]	*	0.54 ± 0.01	0.58 ± 0.12	NS
18:1*n* − 9	18.93 ± 0.23[a]	24.65 ± 0.31[b]	**	17.99 ± 0.30[a]	26.14 ± 0.67[b]	**
20:1*n* − 9	1.88 ± 0.07[a]	1.62 ± 0.05[b]	*	1.86 ± 0.23[a]	2.92 ± 0.04[b]	**
22:1*n* − 9	1.18 ± 0.05[a]	0.63 ± 0.06[b]	**	1.30 ± 0.06[a]	1.03 ± 0.02[b]	**
24:1*n* − 9	0.50 ± 0.02[a]	1.44 ± 0.08[b]	**	0.38 ± 0.01[a]	1.32 ± 0.06[b]	**
∑MUFA	30.66	35.03		30.04	38.05	
18:2*n* − 6	5.25 ± 0.20[a]	10.06 ± 0.25[b]	**	5.15 ± 0.11[a]	10.24 ± 0.20[b]	**
18:3*n* − 3	0.80 ± 0.10[a]	1.46 ± 0.04[b]	*	0.93 ± 0.02[a]	1.45 ± 0.02[b]	**
18:3*n* − 6	0.18 ± 0.01[a]	0.22 ± 0.01[b]	**	0.25 ± 0.03	0.24 ± 0.01	NS
20:2*n*11-14c	0.35 ± 0.02[a]	0.08 ± 0.01[b]	**	0.32 ± 0.01[a]	0.82 ± 0.02[b]	**
20:4*n* − 6	0.70 ± 0.03	0.60 ± 0.03	NS	0.55 ± 0.03	0.58 ± 0.01	NS
20:5*n* − 3	7.10 ± 0.35[a]	5.65 ± 0.23[b]	*	5.71 ± 0.15	5.43 ± 0.13	NS
22:6*n* − 3	11.37 ± 0.62[a]	9.46 ± 0.15[b]	*	9.08 ± 0.60	8.97 ± 0.29	NS
∑PUFA	25.75	27.53		21.99	28.03	
Undefined	7.88	2.15		9.49	1.99	
∑*n* − 3 PUFA	19.27	16.57		15.72	15.85	
∑*n* − 6 PUFA	6.13	10.88		5.95	11.36	
n − 3/*n* − 6	3.14	1.52		2.64	1.40	
EPA/DHA	0.62	0.60		0.63	0.61	
PUFA/SFA	0.72	0.78		0.57	0.89	

Values are mean ± SEM ($n = 6$) and expressed as percentages of total fatty acids

Means in the same row with the same superscript are not significantly different NS = ($P > 0.05$), ** ($P < 001$), * ($P < 0.05$)

the percentage of EPA and DHA in muscle tissue samples of SW-reared fish was found to be significantly higher ($P < 0.05$) than that of their counterparts in FW. Furthermore, it appeared that decreasing water salinity had a decreasing effect on the proportion of $n - 3$ HUFA (especially EPA and DHA) both in muscle tissues and in the whole body samples of European sea bass (Table 3) Muscle tissue proximate composition values are given in

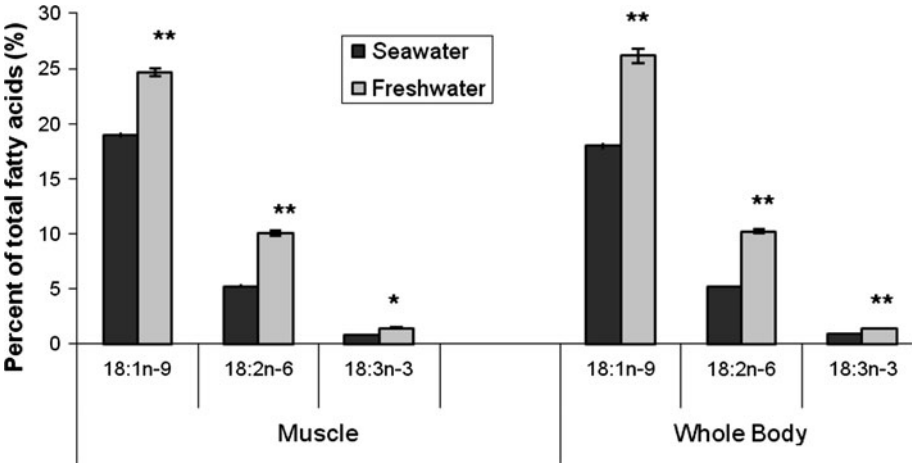

Fig. 1 Percentages (% total fatty acids) of 18:1$n-9$, 18:2$n-6$, 18:3$n-3$ in muscle and whole body of SW and FW living fish. * Indicates significant difference compared with the SW and FW (Student's t-test, $P < 0.05$), ** $P < 0.01$

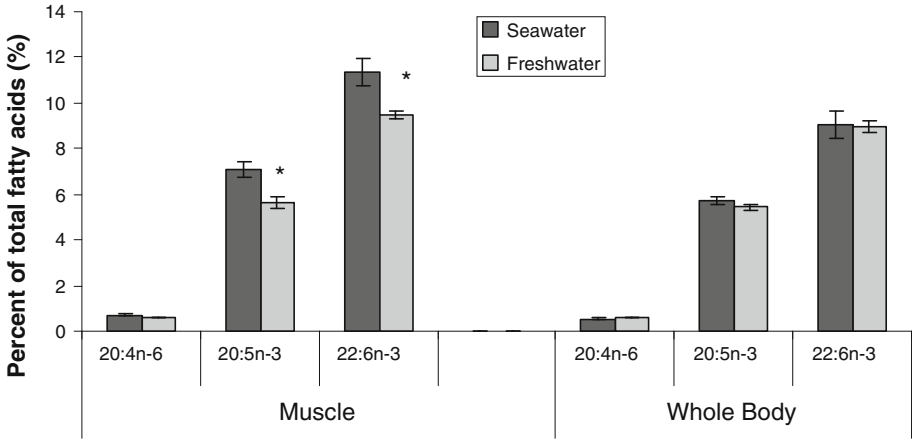

Fig. 2 Percentages (% total fatty acids) of 20:4$n-6$, 20:5$n-3$, 22:6$n-3$ in muscle and whole body of SW and FW living fish. * Indicates significant difference compared with the SW and FW (Student's t-test, $P < 0.05$)

Table 4 Muscle tissue proximate analysis (%) of *Dicentrarchus labrax* in living seawater and freshwater

	SW	FW
Moisture (%)	71.7 ± 0.02[a]	72.5 ± 0.05[a]
Crude protein (%)	19.55 ± 0.01[a]	20.50 ± 0.05[a]
Crude lipid (%)	4.37 ± 0.01[a]	3.55 ± 0.04[b]
Crude ash (%)	2.36 ± 0.03[a]	2.93 ± 0.05[a]

Values are mean ± SEM ($n = 3$). Values in the same row with different superscript are significantly different ($P < 0.05$)

Table 4. The fish reared in SW contained more lipids in their muscle than that of their counterpart in FW ($P < 0.05$). In addition, crude protein, moisture and crude ash levels of the muscle tissues were not significantly affected by freshwater acclimation (Table 4).

Discussion

In this study, European sea bass fingerlings SW-reared fish had better growth rate than those reared in FW (Table 2). During the acclimation period, the fish showed lower interest to feed but it picked up after the 3rd day. When salinity decreases in water media, the fish show lower locomotion in the tanks in order to reduce the food energy consumption and attribute larger portions of it for osmoregulation (Conides and Glamuzina 2006). In addition, a substantial change in environmental salinity may suppress growth rates in fish by simply altering feed intake, the activity of digestive enzymes, which could in turn alter the digestibility and consequently the metabolic availability of feed to fish (Colin et al. 1985; Woo and Kelly 1995; Kelly et al. 1999). Salinity also affects the transportation of some macronutrients resulting in lower bioavailability of these micronutrients to fish hence lower growth rates are occurred (Nordrum et al. 2000). Since feed intake is regulated by several hormones and/or neurotransmitters acting at brain, modifications in any of these factors could also alter the growth rate in fish. In this sense, it has been demonstrated that salinity changes modify cortisol and growth hormone (GH) levels in SW fish species, and prolactin in FW fish species (Nakano et al. 1998). Therefore, differences found in growth rates of European sea bass in SW and FW in this study might primarily be attributed to the alterations in mechanisms involved in both maintaining osmoregulatory balance and feeding physiology and metabolism in FW acclimated fish.

This study showed that crude lipid content of SW-reared European sea bass was significantly higher than that of FW-reared counterparts. Apart from diet, fish size or age, reproductive status, environmental salinity, and season have all been demonstrated to influence fish muscle tissue fat content and biochemical composition as a whole (Ackman 1989; Alasalvar et al. 2002; Eroldogan et al. 2004). At high environmental salinity levels, fish was shown to have higher body lipid content (Kheriji et al. 2003). FW-reared European sea bass muscle tissue had lower lipid content than that of their counterparts in SW in this study and this could be attributed to spending more energy to maintain osmoregulatory balance in freshwater. The main mechanism behind this is to maintain the function of branchial epitheliums, hence the continuity of cell membrane fluidity in freshwater (Tseng and Hwang 2008). The total saturated fatty acid (SFA) content of SW-reared European sea bass was higher than that of FW-reared sea bass, whereas total monounsaturated (MUFA) and polyunsaturated fatty acids (PUFA) was observed to be lower in SW-reared fish than that of its counterparts in FW for both muscle tissue and whole body samples (Table 3). It seemed that fish in FW predominantly used SFAs for energy production than those in SW for osmoregulation as FW species have to excrete higher amount of water from body to the hypo-osmotic environment (Eroldogan et al. 2004; Rubio et al. 2005). Similar results have been reported previously for the European sea bass (Alasalvar et al. 2002; Krajnovic-Ozretic et al. 1994) and in rainbow trout, *Oncorhyncus mykiss* (Haliloglu et al. 2004). It was also shown that the fatty acid composition of fish muscle was significantly affected by rearing trout in salt water. The decrease in salinity was followed by an increase in the percentages of 18:1$n-9$, 18:2$n-6$, 18:3$n-3$ (Fig. 1) and decrease in the percentages of 20:4$n-6$, 20:5$n-3$ and 22:6$n-3$ fatty acids in both muscle tissue and whole body samples in this study (Fig. 2). This could be a result of fatty acid uptake mechanisms by the

intestine working differently in an environment with different salinities. It is known that only a fraction of the fatty acids, mainly long-chain fatty acids, can enter epithelial cells of small intestinal villi via fatty acid transporter protein (FATP; Stahl 2004; Tseng and Hwang 2008). In addition, FATP expression has been reported to be influenced by some hormones and cytokines (Pohl et al. 2004). Recent studies also reported that the profile of gene expressions of known osmoregulation related transporters, enzymes and hormones as well as those related to other cellular events or physiological processes were all affected by environmental salinities in the brain, gills, intestines and kidneys of the European eel (*Anguilla anguilla* L.) and sea bass (*Dicentrarchus labrax*) (Boutet et al. 2006; Kalujnaia et al. 2007a, 2007b).

The increase of total PUFA in FW-reared fish in muscle and whole body may indicate that their synthesis from the essential ones, mainly from 18:2$n-6$ to 18:3$n-3$ is more active in FW than in SW (Henderson and Tocher 1987; El-Kardawy and Salama 1997; Orban et al. 2002). Researchers have shown that FW fish generally contain lower proportion $n3$ PUFA and higher $n6$ PUFA than marine fish. 18:2$n-6$ is a good energy sources for fish, and it is an important fatty acid for keeping cell membrane permeability (Castell et al. 1972; Sargent et al. 1989). Because the mechanism involving the catabolism of 18:3$n-3$ and 18:2$n-6$ fatty acids is more active in FW than in SW (Kheriji et al. 2003; Sargent et al. 1989), the increase level of 18:3$n-3$ and 18:2$n-6$ in muscle tissue and whole body may also indicate that their synthesis from other decreased PUFA. Same conclusion was reached in a study in which *M. cephalus* fry were acclimatized to freshwater (El Cafsi 2000). The increase of PUFA in FW-reared fish could also be related to the decrease in total fatty acids and the catabolism of 18:2$n-6$, 18:3$n-3$ and 14:0 fatty acids (Sargent et al. 1989).

During the freshwater adaptation period, two types of mechanisms can be employed to maintain ionic hemolymph homeostasis and these mechanisms are essentially linked to lipid metabolism. It is well known that phospholipids play an important role in membrane structure, which ultimately effects ion permeability (Luvizotto-Santos et al. 2003). It might be concluded that this difference was due to a change in the proportion of phospholipids to triacylglycerols. Similar results were reported that farmed sea bass changed their fatty acid composition and demonstrated a significant correlation between water salinity and especially the amount of 22:6$n-3$ fatty acid of total phospholipids from gill, liver and muscle tissue samples (Cordier et al. 2002). The findings of this study was also in line with previous findings that the percentages of fatty acids 22:6$n-3$ and 20:5$n-3$ of total fatty acids in muscle tissue samples are a good indication of a specific physiological function for these fatty acids in relation to osmoregulatory challenge in fish species (Daikoku et al. 1982; Jarvis and Ballantyne 2003). The levels of $n-3$ PUFA's were higher in muscle tissue and whole body of SW-reared European sea bass than FW-reared counterparts in this study. Therefore, the changes in lipid composition, in muscle samples could primarily be attributed to the alterations occurring in lipid metabolism due to the salinity challenge.

In conclusion, this research has demonstrated that sea bass could acclimate to freshwater but lower growth rates and $n-3$ fatty acids in their muscle tissues. It also appeared that the lower growth rates in the FW are directly linked to the combined effect of water salinity and the feed intake in this species. Furthermore, this study revealed that the most abundant individual PUFA's were DHA and EPA in muscle tissue and whole body of the European sea bass reared in both SW and FW. However, there were differences in lipid contents and fatty acid composition of SW-reared fish compared with their counterparts in FW. It is known that, EPA and DHA levels are higher in salt water species than in freshwater fish species (Steffens 1997; Czesny et al. 1999). SW-reared fish was shown to

have a rich PUFA content particularly DHA, whereas the level of $n-6$ PUFA was lower in muscle tissue samples indicating the strong relationship between muscle fatty acid composition and water salinity levels. But for human consumption, it was clear that muscle tissue and whole body fatty acid profiles of both SW- and FW-reared fish were in rich of PUFAs which are important beneficial properties for the prevention of human coronary diseases (Pickova and Morkore 2007). A minimum value of PUFA/SFA ratio recommend is 0.45 for humans in their diets (HMSO 1994). Therefore, the PUFA/SFA ratio calculated for fish reared in SW (0.72); and in FW (0.78) were actually higher than the recommended ratio making a FW adapted European sea bass as an equally alternative to SW-reared counterparts in terms of human consumption. Further research targeting specifically the effects of osmoregulation related transporters, enzymes and hormones on the fatty acid metabolism in the European sea bass need to be conducted in order to draw a clear picture about the lipid deposition in an environment with different salinities.

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