

1  
2  
3  
4  
5  
6  
7  
8  
9  
10  
11  
12  
13  
14  
15  
16  
17  
18  
19  
20  
21  
22  
23  
24  
25  
26

**DISTRIBUTION OF LIPIDS AND TRACE MINERALS  
IN DIFFERENT MUSCLE SITES OF FARMED AND  
WILD TURBOT (*Psetta maxima*)**

**Santiago P. Aubourg<sup>1,\*</sup>, Vanesa Losada<sup>1</sup> and Ricardo Prego<sup>2</sup>**

<sup>1</sup> Department of Seafood Chemistry, Instituto de Investigaciones Marinas (CSIC), Eduardo Cabello, 6, 36208-Vigo (Spain)

<sup>2</sup> Department of Oceanography, Instituto de Investigaciones Marinas (CSIC), Eduardo Cabello, 6, 36208-Vigo (Spain)

\* Correspondent: Fax: +34 986 292762; e-mail: [saubourg@iim.csic.es](mailto:saubourg@iim.csic.es)

## SUMMARY

Lipid and trace mineral composition were studied in different sites of the edible flesh of farmed and wild turbot (*Psetta maxima*). Lipid matter (total content, sterols, tocopherols) showed to accumulate in the edge zone, except for phospholipids (PL) that provided a distribution that was found to be independent from the kind of turbot and the zone considered. Fatty acid composition of total lipids showed a non-homogeneous distribution, since the edge zone exhibited a different fatty acid group composition (higher monounsaturated and lower polyunsaturated and  $\omega 3/\omega 6$  ratio values) than the other zones considered; less differences were observed by considering the PL fatty acid composition. Most minerals (Ca, Cu, Fe, Mn and Se) studied showed to be homogeneously distributed along the muscle sites of the wild fish, while more differences were obtained when considering the farmed one. For both kinds of turbot, the most important difference was obtained in the case of Zn, since a largely higher content in the edge zone than in the other zones was detected. A close relationship between Zn and total lipid contents ( $r^2=0.90$  and  $r^2=0.76$  for farmed and wild turbot, respectively) was observed.

Keywords: Turbot, muscle sites, farmed, wild, lipids, minerals

Running title: Distribution of lipids and minerals in turbot

## INTRODUCTION

Turbot (*Psetta maxima*, also known as *Scophthalmus maximus*) is a flat fish species of high commercial value found in North Europe waters (North Sea, specially) widely appreciated for its firm, white, and flavourful flesh (FAO, 2004a), whose consumption undergoes a marked decrease because of its low availability and high cost. In recent years, the increasing production of this species as an aquaculture product (FAO, 2004b) has made it more available to consumers. Previous work has been carried out on the effects of diet on turbot growth (Danielssen & Hjertnes, 1993; Regost *et al.*, 2001), the development of tools for the identification of turbot with respect to other fish species (Prost *et al.*, 1998; Etienne *et al.*, 2000), comparison between wild and farmed composition (Sheehan *et al.*, 1994; Sérot *et al.*, 1998) and quality loss during processing (Chevalier *et al.*, 2001; Ruff *et al.*, 2002; Aubourg *et al.*, 2005).

Seafood products are known to provide high contents of important constituents for the human diet (Piclet, 1987; Simopoulos, 1997). The lipid fraction is now the subject of a great deal of attention due to its high content on  $\omega$ 3 polyunsaturated fatty acids, which have shown potential benefits to human health, particularly in the prevention of cardiovascular diseases (Illingworth & Ullmann, 1990; Ackman & Ratnayake, 1990). Marine organisms have shown to accumulate minerals from the diet and deposit them in their skeletal tissues and organs (Lal, 1989), so as to be considered a good source of essential minerals (Piclet, 1987; Alasalvar *et al.*, 2002).

Fish constituents have been shown to be inhomogeneously distributed along the body of a fish (Pearson *et al.*, 1977; Aubourg *et al.*, 1989). Previous research has shown wide differences in lipid content and composition according to the body zone considered in fatty (Gallardo *et al.*, 1989; Body & Vlieg, 1989) and lean (Ingemansson *et al.*, 1991; Aubourg *et al.*, 1999) fish. Further, several studies have indicated that the concentration of trace minerals in fish is influenced not only by seasonal factors (food source, environment) but also by biological aspects (anatomical, physiological) (Bodsha & Sainsbyry, 1978; Farmer *et al.*, 1979; Lal, 1995; Alasalvar *et al.*, 2002).

The present work was focused on the lipid and main trace mineral compositions. Its objective was to identify elements of differentiation that characterise wild and farmed turbot. Such differentiation is considered important if cultured turbot is to be used as a replacement for the wild one in the seafood European marketing system. Different edible sites of the turbot body were considered for the present study, according to a non-homogeneous distribution of constituents in the fish body.

## MATERIALS AND METHODS

### Fish material and sampling

Wild turbot (*Psetta maxima*) specimens (n=6) were caught (June, 2003) near the Galician Atlantic coast and obtained in a local market 10 h after being caught. From catching till arrival to laboratory the fish were kept in ice.

1 Two-year old farmed turbot specimens (n=6) were obtained from Stolt Sea Farm, S.  
2 A. (Carnota, Spain). The feed employed contained 54 % protein, 20 % fat, 8 %  
3 carbohydrate, 9 % ash and 9 % moisture. Fish specimens were sacrificed in a water-ice  
4 mixture and then kept in ice for 10 h until they arrived at our laboratory.

5 For both kinds of fish specimens, the length was in the 40-45 cm range, while the  
6 width was in the 30-34 cm range; the weight was 1.6-1.8 kg. The white muscle  
7 corresponding to the turbot eyed side was considered for the present study and divided into  
8 the three following sites (Figure 1): i) the muscle zone placed in the body margin (edge  
9 zone, EZ); ii) the muscle zone placed between the head and the end of viscera (ahead zone,  
10 AZ); and iii) the muscle zone placed between the end of viscera and the tail (back zone,  
11 BZ). Chemical analyses were carried out separately on each of the selected zones. Each  
12 individual fish was studied independently (n=6) in order to carry out the statistical analysis.

#### 13 14 Water and lipid contents

15 Water content was determined by weight difference of the homogenised muscle (1-2  
16 g) before and after 24 h at 105 °C; the results were calculated as g water kg<sup>-1</sup> muscle. The  
17 lipid fraction was extracted by the Bligh & Dyer (1959) method. Quantification results  
18 were calculated as g lipid kg<sup>-1</sup> wet muscle.

#### 19 20 Lipid analyses

21 The phospholipid (PL) fraction was purified from the total lipid extract according to  
22 the Aubourg *et al.* (1996) procedure. Total lipid extract and PL fraction were converted into  
23 fatty acid methyl esters (FAME) according to the Lepage & Roy (1986) method. FAME  
24 were analysed by GC (Perkin-Elmer 8700 chromatograph) employing a fused silica  
25 capillary column SP-2330 (0.25 mm i.d. x 30 m, Supelco, Inc., Bellefonte, Pa, USA)  
26 according to Aubourg *et al.* (1996). Content of each fatty acid is expressed as g/ 100g total  
27 FAME.

28 Total PL were quantified by measuring the organic phosphorus on total lipid  
29 extracts according to the Raheja *et al.* (1973) method based on a complex formation with  
30 ammonium molybdate. Results are expressed as g kg<sup>-1</sup> wet fish tissue.

31 Total sterols were determined on total lipid extracts by the method of Huang *et al.*  
32 (1961) based on the Liebermann-Buchardt reaction. Results are expressed as g kg<sup>-1</sup> wet fish  
33 tissue.

34 Tocopherol isomers were analysed according to the Cabrini *et al.* (1992) method.  
35 The presence of the different tocopherol isomers was checked. Only the  $\alpha$ -tocopherol  
36 isomer was detected, and its content was expressed as mg kg<sup>-1</sup> wet fish muscle.

#### 37 38 Trace mineral analysis

39 Seven essential minerals (Ca, Cu, Fe, Mg, Mn, Se and Zn) were chosen for the  
40 present study (Sheehan *et al.*, 1994; Alasalvar *et al.*, 2002). Edible flesh samples were dried  
41 in a stove at 50° C until constant weight and later ground in a mortar. Then, a fraction of  
42 300-400 mg of each sample was weighed in a Teflon vessel of 40 ml and a mixture of 4 ml  
43 of 65 % HNO<sub>3</sub> and 1 ml of H<sub>2</sub>O<sub>2</sub> was added to carry out the microwave digestion. To be  
44 digested, the different muscle mixtures were introduced in a Milestone 1200 Mega  
45 microwave grouped in series of seven samples plus one blank and one certified material  
46 reference (DORM-2, National Research Council Canada) to verify the correct sample  
47 dissolution (Table 1).

1 Mineral contents of the digested samples were determined by atomic absorption. Ca,  
2 Fe, Mg and Zn were analysed by means of flame atomic absorption spectrometry (FAAS)  
3 using a Varian 220 FS apparatus. Cu, Mn and Se by means of electrothermal atomic  
4 absorption spectrometry (ETAAS) using a Varian 220 apparatus equipped with Zeeman  
5 background correction. Analyses were carried out in triplicate. Quantification results are  
6 expressed as mg kg<sup>-1</sup> wet flesh muscle, except for Ca and Mg (g kg<sup>-1</sup> wet flesh muscle).

#### 7 8 Statistical analysis

9 Data from the different chemical measurements were subjected to the ANOVA one-  
10 way method ( $p < 0.05$ ) to explore differences among the different edible flesh sites  
11 considered and between wild and farmed turbot (Statsoft, 1994); comparison of means was  
12 performed using a least-squares difference (LSD) method. Correlation values between lipid  
13 and mineral contents were also analysed.

## 14 15 16 **RESULTS AND DISCUSSION**

### 17 18 Water and lipid contents

19 Moisture results are given in Figure 2. Water content did not provide significant  
20 ( $p > 0.05$ ) differences among the three sites considered under study for the wild turbot.  
21 However, in the case of the farmed fish a lower ( $p < 0.05$ ) content was obtained for the edge  
22 zone when compared to the two others, while no differences ( $p > 0.05$ ) were obtained  
23 between the ahead and back zones. Comparison between both kinds of turbot led to a  
24 lower ( $p < 0.05$ ) water content for each zone of farmed fish when compared to its  
25 counterpart site in wild fish.

26 Lipid contents are shown in Figure 3. Comparison among the three sites under study  
27 provided a higher ( $p < 0.05$ ) lipid content in the edge zone than in the two others, while no  
28 differences ( $p > 0.05$ ) were observed between the ahead and back zones; these conclusions  
29 were found valid for farmed and wild fish. Comparison between both kinds of turbot led to  
30 a higher ( $p < 0.05$ ) lipid content for the farmed one in the edge zone, when compared to its

1 counterpart in the wild fish; no differences ( $p>0.05$ ) could be assessed in the two other sites  
2 between both turbot.

3 An inhomogeneous fat distribution has been found for turbot, in the sense that the  
4 edge zone would play the role of acting as a fat depot zone (Pearson *et al.*, 1977) according  
5 to a similar behaviour observed in other kind of fish species (Ke *et al.*, 1977; Aubourg *et*  
6 *al.*, 1989). In the present experiment, this accumulation was specially great in the case of  
7 the farmed fish.

8 In previous research (Sheehan *et al.*, 1994; Sérot *et al.*, 1998; Grigorakis *et al.*,  
9 2002; Orban *et al.*, 2003), where a differential zone study had not been carried out, a higher  
10 lipid content in farmed than in wild fish was observed; in such cases, the diet was identified  
11 as the main reason for this difference (Bell *et al.*, 1985; Robin *et al.*, 2003) and the higher  
12 lipid content was followed by a lower water content, according to a known inverse ratio  
13 between water and lipid constituents (Piclet, 1987).

14

#### 15 Fatty acid analysis

16 Fatty acid composition of total lipids and PL was studied in the different chosen  
17 sites of farmed and wild turbot; results are included in Tables 2-3. In all cases, the most  
18 abundant fatty acids were 22:6 $\omega$ 3, 16:0 and 20:5 $\omega$ 3. Discussion concerning distribution  
19 differences among the different sites and between farmed and wild fish is now focused to  
20 fatty acid group contents and fatty acid ratio values.

21 The fatty acid study on total lipids (Table 2) showed a higher content in  
22 monounsaturated fatty acids (MUFA) and a lower content in polyunsaturated fatty acids  
23 (PUFA) for the edge zone when compared to the two other zones, being both conclusions  
24 valid for the farmed and wild fish. Comparison between both kinds of turbot led to lower

1 contents in saturated fatty acids (SFA) (except for the edge zone) and MUFA and higher in  
2 PUFA for the different sites of wild fish when compared to their counterpart sites in the  
3 farmed one; results on MUFA and PUFA agree to previous research carried out on different  
4 fish species where no zonal differences had been carried out (Sérot *et al.*, 1998; Grigorakis  
5 *et al.*, 2002; Alasalvar *et al.*, 2002).

6         When considering the fatty acid composition of the total PL (Table 3), a more  
7 homogeneous composition among sites was obtained than in the case of total lipids, so that  
8 less differences were obtained. Thus, a higher SFA content and a lower PUFA value for the  
9 edge zone than in the back zone is to be mentioned for the farmed fish; in the case of the  
10 wild fish, only a higher SFA content was detected for the ahead zone when compared to the  
11 two other sites. PL is a lipid group known to develop a structural role in living bodies, so  
12 that its composition is known to reflect less variations as a result of internal (anatomic,  
13 physiological and other aspects) and external (diet, water temperature) factors (Pearson *et*  
14 *al.*, 1977; Sérot *et al.*, 1998).

15         A majority of the Western population does not consume adequate levels of  $\omega$ 3 fatty  
16 acids through natural dietary sources, such as fish. In this sense, a great interest has been  
17 accorded recently to the  $\omega$ 3/ $\omega$ 6 ratio of foods included in human diet. Its value has shown a  
18 great effect on the development of certain health problems (Illingworth & Ullmann, 1990;  
19 Weber, 1992), being the recommended ratio for the whole diet near to 1:6 ( $\omega$ 3: $\omega$ 6)  
20 (Simopoulos, 1994). In the present work, the site comparison showed a lower  $\omega$ 3/ $\omega$ 6 ratio  
21 in the edge zone when compared to the two others for the total lipid in farmed turbot, while  
22 no differences were obtained for the wild fish. Comparison between both kinds of turbots  
23 led to a higher ratio for the wild fish in each zone when compared to each counterpart zone  
24 in farmed turbot, according to previous research where no site distribution had been

1 considered (Sheehan *et al.*, 1994; Sérot *et al.*, 1998; Alasalvar *et al.*, 2002; Grigorakis *et*  
2 *al.*, 2002). The PL study did not provide differences ( $p>0.05$ ) for the  $\omega 3/\omega 6$  ratio as a result  
3 of the site distribution or kind of turbot. It is concluded that both cultivated and wild turbot  
4 provide a profitable  $\omega 3/\omega 6$  ratio in order to maintain the recommended value for the whole  
5 human diet. However, a better value ( $p<0.05$ ) is obtained for the wild fish.

6         According to the interest on the  $\omega 3$  series fatty acids, the 22:6 $\omega 3$  / 20:5 $\omega 3$  ratio  
7 distribution in the different sites and kinds of turbot was studied. The content ratio showed  
8 a lower value in the edge zone than in the two other zones for the farmed and wild total  
9 fatty acids, while no site differences were obtained when considering the PL fraction.  
10 Comparison between both kinds of turbot led to lower values for farmed fish in total and  
11 PL fractions, according to previous research where no site distribution study had been  
12 attained (Sheehan *et al.*, 1994; Alasalvar *et al.*, 2002).

13

#### 14 Lipid group analysis

15         Total PL contents obtained (Figure 4) are included in the range 1.7-2.2 g kg<sup>-1</sup> wet  
16 flesh muscle. Results did not provide differences ( $p>0.05$ ) among zones or by comparing  
17 both kinds of turbot. PL are known to be an important constituent of cell membranes and,  
18 although its content and composition has shown to be altered by the diet (Igene, 1976), the  
19 changes are relatively small as compared to those of depot fats (Pearson *et al.*, 1977). This  
20 homogeneous distribution of PL content in different sites of fish flesh muscle agrees to  
21 previous research carried out on fatty fish (Aubourg *et al.*, 1989) and to the above  
22 mentioned results on fatty acid composition.

23         Figure 5 shows the results obtained for total sterols. Values in the range 0.2-1.1 g  
24 kg<sup>-1</sup> wet flesh muscle were obtained, that agree to common values for lean fish species



1 (Piclet, 1987). The site distribution study showed a higher ( $p < 0.05$ ) content in the edge  
2 zone when compared to the two other zones for both kinds of turbot. This result agrees to  
3 previous research on albacore fish where a higher total sterol content was obtained in the  
4 lipid depot site (Aubourg *et al.*, 1989). Comparison between both farmed and wild fish did  
5 not provide differences ( $p > 0.05$ ) for the ahead and back zones, but a higher ( $p < 0.05$ )  
6 content in the edge zone of the farmed fish was obtained when compared to its counterpart  
7 site in the wild fish, according to the results obtained for the total lipid contents (Figure 3).  
8 Orban *et al.* (2003) obtained no differences for cholesterol content when comparing wild  
9 and farmed sea bass and gilthead sea bream; however, a site distribution study was not  
10 encountered in such experiment. Present results on total sterols show an inhomogeneous  
11 distribution of this lipid group, so that its accumulation is carried out in the edge zone, and  
12 this behaviour is particularly strong in the farmed fish.

13         Alpha-tocopherol contents obtained are shown in Figure 6. The values were within  
14 range reported for different fish species (Piclet, 1987; Ruff *et al.*, 2002; Aubourg *et al.*,  
15 2004). Farmed fish showed an inhomogeneous distribution for this endogenous antioxidant.  
16 Thus, a higher ( $p < 0.05$ ) content in the edge zone was obtained when compared to the two  
17 other zones under study; no differences ( $p > 0.05$ ) were obtained between the ahead and  
18 back zones. Wild fish provided a higher mean value for the edge zone; however,  
19 differences were not significant ( $p > 0.05$ ) among the three sites under study. When both  
20 kinds of turbot are compared, no differences ( $p > 0.05$ ) in the edge zone were obtained,  
21 while the ahead and back zones in wild turbot showed higher ( $p < 0.05$ ) levels than their  
22 counterparts in farmed turbot. Alpha-tocopherol is a known lipid-soluble chain- breaking  
23 antioxidant, whose main role is protecting the polyunsaturated fatty acids from oxidation  
24 (Kamal-Eldin & Appelqvist, 1996; Kulås & Ackman, 2001). Its higher presence in the edge

1 zone agrees to the fact that this zone provides the highest lipid content. If the alpha-  
2 tocopherol/total lipid ratio is considered, a higher ( $p < 0.05$ ) value is observed in each zone  
3 of the wild turbot when compared to its corresponding site in the farmed one. This  
4 difference, in addition to the one found for alpha-tocopherol content in ahead and back sites  
5 could be explained as a result of a different diet intake between both kinds of turbot.

6

### 7 Mineral analysis

8 Trace mineral contents obtained are expressed in Table 4. Levels obtained for Ca  
9 and Mg were higher than in the five remaining minerals according to common distribution  
10 in marine species flesh (Piclet, 1987). Expected values were obtained for most minerals  
11 (Ca, Mg, Mn, Se and Zn) and low for Fe and Cu, when compared to previous research  
12 (Piclet, 1987; Engman & Jorhem, 1998). The relative low value of Fe could be explained as  
13 a result of discarding the dark muscle in the present study (Alasalvar *et al.*, 2002).

14 The Ca content revealed an homogeneous distribution among the three sites studied.  
15 However, comparison between both kinds of turbot showed a higher content in the farmed  
16 one for the back zone.

17 The Cu distribution did not show differences as a result of the flesh site considered.  
18 However, comparison between wild and farmed turbot provided a higher value in the three  
19 zones for the farmed one when compared to their counterpart in wild fish.

20 Large variations from fish to fish were obtained when considering the Fe presence.  
21 The farmed fish showed a higher content in the edge zone than in the back zone for this  
22 mineral; no site differences could be observed for the wild turbot. Comparison between  
23 both kinds of fish did not provide differences at any of the sites studied.

1 The Mg content showed to be higher in the ahead zone than in the edge zone in both  
2 turbot. Comparison between both kinds of fish led to higher values for the farmed one in  
3 the ahead and back zones.

4 A higher Mn content was observed in the edge zone than in the ahead zone for the  
5 farmed turbot; no site differences were obtained for the wild fish. Comparison between  
6 both turbot showed a higher content in the farmed one in the three sites studied when  
7 compared to their counterparts in wild fish.

8 The Se content provided an homogeneous distribution among the different sites.  
9 Also, no differences could be outlined by comparing both kinds of turbot.

10 A higher Zn content was obtained in the edge zone than in the two other zones for  
11 both kinds of turbot. Farmed turbot showed a higher Zn content than the wild one when  
12 comparing each site to its counterpart in the wild fish.

13 Mineral elements corresponding to the transition and electronegative elements from  
14 the Periodic Table have been reported to be strongly bound to other constituents (Piclet,  
15 1987; Gordon, 1988). Accordingly, the relationship between mineral presence and lipid  
16 content was investigated in the present work. For it, correlation values between fat and  
17 mineral contents were studied. Farmed fish showed a good correlation value for Zn ( $r^2 =$   
18  $0.90$ ), and fair in the cases of Fe, Mg and Mn ( $r^2 = 0.71$ ,  $r^2 = -0.76$  and  $r^2 = 0.78$ ,  
19 respectively); poor correlation values were obtained in the case of considering the PL  
20 content instead of the total lipid content. For wild fish, fair correlation values were obtained  
21 for total lipid content and presence of Cu and Zn ( $r^2=0.77$  and  $r^2=0.76$ , respectively); again,  
22 unfair correlations were obtained in the case of considering the PL content.

23 Content relationship between water and mineral elements was also checked. Farmed  
24 fish showed a good correlation for Zn ( $r^2 = -0.85$ ), and fair in the cases of Fe, Mg and Mn

1 ( $r^2 = -0.67$ ,  $r^2 = 0.76$  and  $r^2 = -0.69$ , respectively). These results agree to the above  
2 mentioned comparison to lipid content, and can be explained as a result of the already  
3 mentioned inverse ratio between water and lipid contents (Piclet, 1987). For wild turbot,  
4 poor correlation values were obtained between water and mineral elements.

5

6

7

### **CONCLUDING REMARKS**

8

9 A non-homogeneous distribution of lipid composition among the different sites  
10 considered was observed for turbot. Thus, the edge zone showed a higher lipid content, a  
11 different fatty acid group contents and fatty acid ratios and a higher total sterol and  $\alpha$ -  
12 tocopherol contents than the two other sites considered. These differences were more  
13 marked in the farmed fish than in the wild one, showing an important effect of diet  
14 provided on the development of an heterogeneous lipid distribution. The PL fraction  
15 provided far less differences as a result of the zone or the kind of turbot considered,  
16 according to its structural role in living bodies and being less influenced by the diet.

17 Most trace minerals studied showed a more homogeneous distribution among sites  
18 than in the case of the lipid matter. According to lipid fraction results, less differences were  
19 obtained for the wild fish than for the farmed one. The most interesting difference was  
20 found for the Zn content that led to a higher value in the edge zone for both kinds of  
21 turbot. Further, fair correlation values were obtained between Zn and total lipid contents.

22

1 ACKNOWLEDGMENTS

2           This work was supported by the Secretaría Xeral de I + D from the Xunta de Galicia  
3 (*Project PGIDT01MAR40202PR*). The authors thank Mrs. Ana Labandeira, Mr. Marcos  
4 Trigo and Mr. José Antonio for their excellent technical assistance.

## **FIGURE LEGENDS**

1

2

3

4 **Figure 1**: Position of the three white muscle zones considered in the present study: ahead  
5 (AZ), back (BZ) and edge (EZ) in the turbot body.

6

7 **Figure 2**: Water content ( $\text{g kg}^{-1}$  wet flesh muscle) obtained in the edge (EZ), ahead (AZ)  
8 and back (BZ) zones of farmed and wild turbot.

9

10 **Figure 3**: Lipid content ( $\text{g kg}^{-1}$  wet flesh muscle) obtained in the edge (EZ), ahead (AZ)  
11 and back (BZ) zones of farmed and wild turbot.

12

13 **Figure 4**: Total phospholipid content ( $\text{g kg}^{-1}$  wet flesh muscle) obtained in the edge (EZ),  
14 ahead (AZ) and back (BZ) zones of farmed and wild turbot.

15

16 **Figure 5**: Total sterol content ( $\text{g kg}^{-1}$  wet flesh muscle) obtained in the edge (EZ), ahead  
17 (AZ) and back (BZ) zones of farmed and wild turbot.

18

19 **Figure 6**: Alpha-tocopherol content ( $\text{mg kg}^{-1}$  wet flesh muscle) obtained in the edge (EZ),  
20 ahead (AZ) and back (BZ) zones of farmed and wild turbot.

## REFERENCES

- 1
- 2
- 3 Ackman, R. & Ratnayake, W. (1990). Chemical and analytical aspects of assuring an  
4 effective supply of omega-3 fatty acids to the consumer. In: *Omega-3 fatty acids in*  
5 *health and disease* (edited by R. Lees & M. Karel). Pp. 215-233. New York (USA)  
6 and Basel (Switzerland): Marcel Dekker, Inc.
- 7 Alasalvar, C., Taylor, K., Zubcov, E., Shahidi, F. & Alexis, M. (2002). Differentiation of  
8 cultured and wild sea bass (*Dicentrarchus labrax*): total lipid content, fatty acid and  
9 trace mineral composition. *Food Chemistry*, **79**, 145-150.
- 10 Aubourg, S., Medina, I. & Pérez-Martín, R. (1996). Polyunsaturated fatty acids in tuna  
11 phospholipids: Distribution in the sn-2 location and changes during cooking.  
12 *Journal of Agricultural and Food Chemistry*, **44**, 585-589.
- 13 Aubourg, S., Pérez-Martín, R. & Gallardo, J. (1989). Stability of lipids of frozen albacore  
14 (*Thunnus alalunga*) during steam cooking. *International Journal of Food Science*  
15 *and Technology*, **24**, 341-345.
- 16 Aubourg, S., Piñeiro, C. & González, M<sup>a</sup> J. (2004). Quality loss related to rancidity  
17 development during frozen storage of horse mackerel (*Trachurus trachurus*).  
18 *Journal of the American Oil Chemists' Society*, **81**, 671-678.
- 19 Aubourg, S., Piñeiro, C., Gallardo, J. & Barros-Velázquez, J. (2005). Biochemical changes  
20 and quality loss during chilled storage of farmed turbot (*Psetta maxima*). *Food*  
21 *Chemistry*, **90**, 445-452.
- 22 Aubourg, S., Rey-Mansilla, M. & Sotelo, C. (1999). Differential lipid damage in various  
23 muscle zones of frozen hake (*Merluccius merluccius*). *Zeitschrift für Lebensmittel*  
24 *Untersuchung und Forschung*, **208**, 189-193.

- 1 Bell, M., Henderson, R. & Sargent, J. (1985). Changes in the fatty acid composition of  
2 phospholipids from turbot (*Scophthalmus maximus*) in relation to dietary  
3 polyunsaturated fatty acid deficiencies. *Comparative Biochemistry and Physiology*,  
4 **81B**, 193-198.
- 5 Bligh, E. & Dyer, W. (1959). A rapid method of total extraction and purification. *Canadian*  
6 *Journal of Biochemistry and Physiology*, **37**, 911-917.
- 7 Bodsha, K. & Sainsbyry, M. (1978). Aspects of the biology and heavy metal accumulation  
8 of *Ciliata mustela*. *Journal of Fish Biology*, **12**, 213-220.
- 9 Body, D. & Vlieg, P. (1989). Distribution of the lipid classes and eicosapentaenoic (20:5)  
10 and docosahexaenoic (22:6) acids in different sites in blue mackerel (*Scomber*  
11 *australasicus*) fillets. *Journal of Food Science*, **54**, 569-572.
- 12 Cabrini, L., Landi, L., Stefanelli, C., Barzanti, V. & Sechi, A. (1992). Extraction of lipid  
13 and lipophilic antioxidants from fish tissues: A comparison among different  
14 methods. *Comparative Biochemistry and Physiology. Biochemistry and Molecular*  
15 *Biology*, **101**, 383-386.
- 16 Chevalier, D., Sequeira-Muñoz, A., Le Bail, A., Simpson, B. & Ghoul, M. (2001). Effect  
17 of freezing conditions and storage on ice crystal and drip volume in turbot  
18 (*Scophthalmus maximus*). Evaluation of pressure shift freezing vs. air-blast  
19 freezing. *Innovative Food Science and Emerging Technologies*, **1**, 193-201.
- 20 Danielssen, D. & Hjertnes, T. (1993). Effect of dietary protein levels in diets for turbot  
21 (*Scophthalmus maximus*) to market size. In: *Fish Nutrition in Practice* (edited by S.  
22 Karushik & P. Luquet). Pp. 89-96. Biarritz (France): INRA Editions.



1 Engman, J. & Jorhem, L. (1998). Toxic and essential elements in fish from Nordic waters,  
2 with the results seen from the perspective of analytical quality assurance. *Food*  
3 *Additives and Contaminants*, **15**, 884-892.

4 Etienne, M., Jérôme, M., Fleurence, J., Rehbein, H., Kündiger, R., Mendes, R., Costa, H.,  
5 Pérez-Martín, R., Piñeiro, C., Craig, A., Mackie, I., Malmheden Yman, I., Ferm, M.  
6 Martínez, I., Jessen, F., Smelt, A. & Luten, J. (2000). *Journal of Agricultural and*  
7 *Food Chemistry*, **48**, 2653-2658.

8 FAO (2004a). Fishery statistics. Capture production. In: *Food and Agriculture*  
9 *Organization of the United Nations*. Vol. 94/1. Pp. 121-122. Rome (Italy):  
10 Yearbook 2002.

11 FAO (2004b). Fishery statistics. Aquaculture production. In: *Food and Agriculture*  
12 *Organization of the United Nations*. Vol. 94/2. P. 75. Rome (Italy): Yearbook 2002.

13 Farmer, G., Ashfield, D. & Samant, H. (1979). Effects of zinc on juvenile Atlantic salmon  
14 (*Salmo salar*): acute toxicity, food intake, growth and bioaccumulation.  
15 *Environmental Pollution*, **19**, 103-117.

16 Gallardo, J., Aubourg, S. & Pérez-Martín, R. (1989). Lipid classes and their fatty acids at  
17 different loci of albacore (*Thunnus alalunga*): Effects of precooking. *Journal of*  
18 *Agricultural and Food Chemistry*, **37**, 1060-1064.

19 Gordon, D. (1988). Minerals in seafoods: their bioavailability and interactions. *Food*  
20 *Technology*, **42**, 156-160.

21 Grigorakis, K., Alexis, M., Anthony Taylor, K. & Hole, M. (2002). Comparison of wild  
22 and cultured gilthead sea bream (*Spaurus aurata*); composition, appearance and  
23 seasonal variations. *International Journal of Food Science and Technology*, **37**,  
24 477-484.

- 1 Huang, T., Chen, C., Wefler, V. & Raftery, A. (1961). A stable reagent for the  
2 Liebermann-Buchardt reaction. *Analytical Chemistry*, **33**, 1405-1407.
- 3 Igene, J., Pearson, A., Merkel, R. & Coleman, T. (1979). Effect of frozen storage time,  
4 cooking and holding temperature upon extractable lipids and TBA value of beef and  
5 chicken. *Journal of Animal of Science*, **49**, 701-707.
- 6 Illingworth, D. & Ullmann, D. (1990). Effects of omega-3 fatty acids on risk factors for  
7 cardiovascular disease. In: *Omega-3 fatty acids in health and disease* (edited by R.  
8 Lees & M. Karel). Pp. 39-69. New York (USA) and Basel (Switzerland): Marcel  
9 Dekker, Inc.
- 10 Ingemansson, T., Olsson, N., Herslöf, B. & Ekstrand, B. (1991). Lipids in light and dark  
11 muscle of farmed rainbow trout (*Oncorhynchus mykiss*). *Journal of the Science of*  
12 *Food and Agriculture*, **57**, 443-447.
- 13 Kamal-Eldin, A. & Appelqvist, L. (1996). The chemistry and antioxidant properties of  
14 tocopherols and tocotrienols. *Lipids*, **31**, 671-701.
- 15 Ke, P., Ackman, R., Linke, B. & Nash, D. (1977). Differential lipid oxidation in various  
16 parts of frozen mackerel. *Journal of Food Technology*, **12**, 37-47.
- 17 Kulå, E. & Ackman, R. (2001). Properties of  $\alpha$ -,  $\gamma$ -, and  $\delta$ -tocopherol in purified fish oil  
18 triacylglycerols. *Journal of the American Oil Chemists' Society*, **78**, 361-367.
- 19 Lal, S. (1989). Minerals. In: *Fish nutrition* (edited by J. Halver). Pp. 220-257. San Diego  
20 (CA, USA): Academic Press.
- 21 Lal, S. (1995). Macro and trace elements in fish and shellfish. In: *Fish and fishery*  
22 *products: composition, nutritive properties and stability* (edited by A. Ruiter). Pp.  
23 187-214. Wallingford: CAB International.

- 1 Lepage, G. & Roy, C. (1986). Direct transesterification of all classes of lipids in a one step  
2 reaction. *Journal of Lipid Research*, **27**, 114-120.
- 3 Orban, E., Névigato, T., Di Lena, G., Casini, I. & Marzetti, A. (2003). Differentiation in  
4 the lipid quality of wild and farmed seabass (*Dicentrarchus labrax*) and gilthead  
5 sea bream (*Sparus aurata*). *Journal of Food Science*, **68**, 128-132.
- 6 Pearson, A., Love, J. & Shorland, F. (1977). “Warmed-over” flavor in meat, poultry and  
7 fish. *Advances in Food Research*, **23**, 2-61.
- 8 Piclet, G. (1987). Le poisson aliment. Composition – intérêt nutritionnel. *Cahiers de*  
9 *Nutrition et Diététique*, **XXII**, 317-335.
- 10 Prost, C., Serot, T. & Demaimay, M. (1998). Identification of the most potent odorants in  
11 wild and farmed cooked turbot (*Scophthalmus maximus*). *Journal of Agricultural*  
12 *and Food Chemistry*, **46**, 3214-3219.
- 13 Raheja, R., Kaur, C., Singh, A. & Bhatia, A. (1973). New colorimetric method for the  
14 quantitative determination of phospholipids without acid digestion. *Journal of Lipid*  
15 *Research*, **14**, 695-697.
- 16 Regost, C., Arzel, J., Cardinal, M., Robin, J., Laroche, M. & Kaushik, S. (2001) Dietary  
17 lipid level, hepatic lipogenesis and flesh quality in turbot (*Psetta maxima*).  
18 *Aquaculture*, **193**, 291-309.
- 19 Robin, J., Regost, C., Arzel, J. & Kaushik, S. (2003). Fatty acid profile of fish following a  
20 change in dietary fatty acid source: model fatty acid composition with a dilution  
21 hypothesis. *Aquaculture*, **225**, 283-293.
- 22 Ruff, N., Fitzgerald, R., Cross, T. & Kerry, J. (2002). Comparative composition and shelf-  
23 life of fillets of wild and cultured turbot (*Scophthalmus maximus*) and Atlantic  
24 halibut (*Hippoglossus hippoglossus*). *Aquaculture International*, **10**, 241-256.

- 1 Sérot, L., Gandemer, G. & Demaimay, M. (1998). Lipid and fatty acid compositions of  
2 muscle from farmed and wild adult turbot. *Aquaculture International*, **6**, 331-343.
- 3 Sheehan, E., Sheehy, P., Morrissey, P. & Fitzgerald, R. (1994). Compositional analysis on  
4 wild and farmed turbot and fish feeds in Ireland. In: *Turbot culture: Problems and*  
5 *prospects* (edited by P. Lavens & R. Remmerswaal). Gent (Belgium): European  
6 Aquaculture Society, Special Publication No. 22.
- 7 Simopoulos, A. (1994). Fatty acids. In: *Functional foods, designer foods, pharmafoods,*  
8 *nutraceuticals* (edited by I. Goldberg). Pp. 355-392. New York (USA): Chapman  
9 and Hall.
- 10 Simopoulos, A. (1997). Nutritional aspects of fish. In: *Seafood from producer to consumer,*  
11 *Integrated approach to quality* (edited by J. Luten, T. Børrensen & J.  
12 Oehlenschläger). Pp. 589-607. London (UK): Elsevier Science.
- 13 Statsoft. (1994). *Statistica for Macintosh*. Statsoft and its licensors. Tulsa, Ok (USA).
- 14 Weber, P. (1992). Dietary fatty acids and eicosanoid Biochemistry. In: *Advances in Seafood*  
15 *Biochemistry* (edited by G. Flick & R. Martin). Pp. 181-184. Lancaster (PA, USA):  
16 Technomic Publishing.
- 17

**TABLE 1**

Analysis results of certified reference material (CRM)\*

<b>Concentration</b>	<b>Ca</b>	<b>Cu</b>	<b>Fe</b>	<b>Mg</b>	<b>Mn</b>	<b>Se</b>	<b>Zn</b>
Found	5.14±0.53	2.00±0.14	148± 3	1.01±0.04	3.53±0.22	1.30±0.13	27.6±0.5
Certified	NC	2.34±0.16	142±10	NC	3.66±0.34	1.40±0.09	25.6±2.3

\* CRM consisted on fish muscle material (DORM-2) from the National Research Council of Canada. Results are expressed in  $\mu\text{g g}^{-1}$  flesh muscle. "NC" indicates a value not certified.

**TABLE 4**

Trace mineral contents in different muscle zones\* of farmed and wild turbot\*\*

Trace mineral	Farmed Turbot			Wild Turbot		
	EZ	AZ	BZ	EZ	AZ	BZ
Ca	0.93 (0.20)	<sup>y</sup> 1.05 (0.10)	0.99 (0.12)	0.90 (0.26)	<sup>z</sup> 0.75 (0.17)	0.78 (0.10)
Cu	<sup>y</sup> 0.33 (0.05)	<sup>y</sup> 0.24 (0.09)	<sup>y</sup> 0.23 (0.06)	<sup>z</sup> 0.16 (0.06)	<sup>z</sup> 0.09 (0.01)	<sup>z</sup> 0.11 (0.03)
Fe	1.91 b (0.80)	0.80 ab (0.49)	0.73 a (0.25)	1.78 (0.69)	1.32 (0.82)	1.65 (0.68)
Mg	0.22 a (0.02)	<sup>y</sup> 0.27 b (0.01)	<sup>y</sup> 0.26 ab (0.02)	0.19 a (0.02)	<sup>z</sup> 0.23 b (0.02)	<sup>z</sup> 0.22 ab (0.02)
Mn	<sup>y</sup> 0.41 b (0.12)	<sup>y</sup> 0.22 a (0.05)	<sup>y</sup> 0.29 ab (0.09)	<sup>z</sup> 0.14 (0.06)	<sup>z</sup> 0.12 (0.04)	<sup>z</sup> 0.12 (0.03)
Se	0.27 (0.09)	0.35 (0.10)	0.37 (0.09)	0.29 (0.10)	0.32 (0.06)	0.31 (0.03)
Zn	<sup>y</sup> 12.11 b (2.46)	<sup>y</sup> 6.32 a (0.74)	<sup>y</sup> 6.33 a (0.66)	<sup>z</sup> 7.81 b (1.17)	<sup>z</sup> 3.77 a (0.28)	<sup>z</sup> 4.28 a (0.37)

\* Muscle zone abbreviations: Edge (EZ), ahead (AZ) and back (BZ) zones.

\*\* Mean values (mg kg<sup>-1</sup> wet flesh fish, except for Ca and Mg expressed as g kg<sup>-1</sup> wet flesh fish) of six independent determinations are expressed. Standard deviations are indicated in brackets. For each mineral and for each kind of turbot, mean values followed by different letters (a-b) indicate significant differences (p<0.05) among muscle zones. For each mineral, means preceded by different superscripts indicate significant differences (p<0.05) between both kinds of turbot in the muscle zone indicated.