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The composition of fatty acids in the tissues of Tunisian swordfish (Xiphias gladius)

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ABSTRACT

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The objective of this study was to compare the composition of fatty acids stored in the various parts of swordfish. Muscle and organ sections of a series of swordfish samples were collected. Their chemical analysis allowed the classification of swordfish as a semi-fatty fish, with its byproducts totalling 35.6% of the total fatty acids (TFA) and its white and red muscles (MR) 7.2%. The highest contents were found in the liver (26%), the gonads (4.7%) and the red muscle (RM) (4.5%). The UFA/SFA ratio as recommended by nutritionists is 3; in the liver, skin and RM samples this ratio was, respectively, 3.5, 2.8 and 2.7. There is a high level of unsaturated fatty acids (UFA) in swordfish. More than 40% of the muscular tissues are made up of polyunsaturated fatty acids (PUFA) of the n-3 series (EPA + DHA). The byproducts contain more than 30% of monounsaturated acids characterised by the fatty acids of the n-9 series and particularly by oleic acid.

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1. Introduction

Fat is stored in various parts of the fish, mainly in the liver, in the muscles and in the perivisceral and subcutaneous adipose tissues ([Sheridan, 1988\)](#page-6-0). So far, it has not been possible to know exactly what determines the fatty acid deposits in the various parts of fish [\(Sheridan, 1994](#page-6-0)). Indeed, the differences both in storage tissues and total fatty acid (TFA) levels represent important parameters for the classification of species [\(Eymard, 2003\)](#page-6-0). In lean fish, for instance, the fatty acid level of the liver can reach as much as 40–70 g per 100 g of tissue, whereas their muscles contain only little fat (5 g per 100 g of tissue). Muscles in fatty fish, however, have a high fat level with more than 10% of the total fat, thus even exceeding the fat part in the liver. Fats occur in the form of extracellular fatty globules, and as layers under the skin and in the abdominal cavity [\(Corraze & Kaushik, 1999\)](#page-6-0). Fish with a medium lipid level are usually flat fish, storing their fats in the liver, but also the muscles and some other tissues, such as perivisceral adipose tissue.

Fish oils ([Richard, 2006; Shahidi & Wanasundara, 1998\)](#page-6-0) are the main source of n-3 fatty acids. They are extracted from non-consumable parts or byproducts of the fish, with these byproducts being defined as unused or recoverable parts subsequent to traditional methods of treatment of the fish (such as head, skin, falling thread, central bones, viscera, liver, and, depending on the fishing season, reproductive elements, such as eggs or milt ([Dumay,](#page-6-0)

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[2006\)](#page-6-0). It is important to analyse the position effect on the composition of the fatty acids in swordfish; in fact, the byproducts may contribute to the total level of fatty acids, thus increasing the commercial value of the fish.

The purpose of this study is (1) to determine the fatty acid composition in juvenile swordfish by exploring its different anatomical areas, and (2) to inform the consumer about the fatty acid level in the different parts of the fish, in order to guarantee an optimal use of the fatty acids, especially $n-3$ and $n-6$ fatty acids.

2. Materials and methods

Fresh swordfish samples were collected at the wholesale market of Tunis between December 2005 and February 2006. They had not yet reached first sexual maturity, which is 140 cm in the Mediterranean Sea [\(De la Serna, Ortiz, & Macias, 1996](#page-6-0)). The lower jaw fork length of our samples $(n = 6)$ varied between 55 and 104 cm, gutted weight between 1.4 and 10.5 kg.

2.1. Sampling

Samples were taken at three different points ([Fig. 1\)](#page-1-0): a frontal section (A) just under the first dorsal fin, a second central section (B) before the first anal fin, and a third section near the second dorsal fin.

Two one-gram samples of white muscle were taken from each of the three sections A, B and C, one in the dorsal area, and the other from the ventral area, i.e., D_1 and V_1 from section A, D_2 and V_2 from section B and D_3 and V_3 from section C. A sample of red

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muscle (RM) was taken from the right side of section C. Only lipids of muscular tissue (white and red muscles) were analysed, disregarding any other type of lipids, such as dermal lipids. On the whole, our analyses are based on seven anatomical samples of muscular tissue, i.e., D_1 , V_1 , D_2 , V_2 , D_3 , V_3 and RM.

Both the eyeballs (O) of the fish were excised after sectioning the optic nerve and removing the surrounding tissue. The eyeballs were crushed in a mixer and were analysed for their fatty acid composition. The brain (C) was excised from the cranial cavity; the fat (E) embedding the brain was drawn with a pipette and stored. Further one-gram samples were taken from the liver (F) and the central part of the gonads (G). Another one-gram sample was taken from the skin of section A (P). So a total of six further samples (O, C, E, F, G, P) were included in our analyses.

Table 1 Fatty acid in the different anatomical areas of swordfish (in% of TFA, mean ± SE) areas

All samples were fixed in boiling water to completely inactivate enzymatic activity, especially phospholipases. Samples along with the fixing liquid were stored in a freezer at -28 \degree C.

2.2. Total lipid extraction

The total lipids were extracted from the tissues according to the [Folch, Lees, and Sloane-Stanley \(1957\)](#page-6-0) method, i.e., chloroform:methanol (2:1, v/v).

2.3. Fatty acid analysis with gas chromatography

For further analysis, the fatty acids were transformed into methyl esters, according to the [Cecchi, Basini, and Castano \(1985\)](#page-6-0) method. A gas chromatograph type HP 6890 with a split/splitless injector with electronic pressure control and a flame ionisation detector was used for the analysis. Separation was performed with a 30 m HP Innowax capillary column with an internal diameter of $250 \mu m$ and a 0.25 μm film thickness, the stationary polar phase of the column being polyethylene glycol.

2.4. Identification and quantification of fatty acids

The different fatty acids in swordfish were obtained by comparing the retention times of the fatty acids under study and those of a mixture of methyl esters (SUPELCO PUFA-3). The quantification of the fatty acids is based on an internal standard not present in our Fig. 1. Sampling areas of section A, B and C. Samples, methyl nonadecanoate or $C_{19:0}$ (Sigma).

SFA: C14:0 + C15:0 + C16:0 + C17:0 + C18: 0; MUFA: C16 :1 + C18:1n-7 + C18:1n-9 + C20:1n-9 + C22:1n-11 + C22:1n-9 C24:1n-9; PUFA: C16:2n-6 + C16: 3n-4 + C16: 4n-3 + C18: 2n-6 + C18: 3n-3 + C18: 4n-3 + C20: 4n-6 + C20: 4n-3 + C20: 5n-3 + C22: 6n-3.

2.5. Statistical analyses

The results represent the mean values of a series of repetitions $(n = 6)$. In order to compare the different anatomical parts with each other, the mean values of the white muscle areas (D_1, D_2, D_3) D_3 and V_1 , V_2 and V_3) were combined to form a single part (M). The result is considered significant if $p < 0.05$. Different mean values were analysed according to the Duncan test. The statistical analyses were carried out with the SAS software program version 6.12.

3. Results

3.1. Lipidic profile of swordfish

Twenty-five fatty acids were identified in the different areas. They are listed in [Table 1.](#page-1-0) Four fatty acids are particularly important in the lipidic composition of the different anatomical areas. The palmitic and stearic acids prevail in the saturated fatty acid (SFA) group with, respectively, $17.08\% - 26.9\%$ and $2.41\% - 9.03\%$. The oleic acid content in the monounsaturated fatty acid (MUFA) group was around 13.3% – 35.5%. As to the polyunsaturated fatty acid (PUFA) group, the contents of docosahexaenoic acid (DHA) varied between 14.5% and 28.8%. These four major fatty acids account for 64.3% – 81.6% of the twenty-five fatty acids making up the different anatomical areas. Among the remaining fatty acids, the presence of palmitoleic acid and eicosapentaenoic acid (EPA) is notable with respectively $0.37% - 5.8%$ and $1.53% - 3.52%$.

It can also be inferred from [Table 1](#page-1-0) that the profile of each fatty acid group varies according to the anatomical area. The muscular zones (M and RM) have a high EPA + DHA content. As to the byproducts, the brain contains a high percentage of palmitic acid, the brain fat (E) is rich in eicosenoic acid, and myristic, palmitoleic, eicosatetraenoic and docosenoic acids characterise the eyeball (O). Oleic, hexadecatrienoic and docosapentaenoic acids are specific to

Fig. 2. Contents of TFA (g/100 g) per anatomical areas. TFA values with different subscripts (a–c) were significantly different at $p < 0.05$.

Fig. 3. Contents of UFA and SFA per anatomical areas. TFA values with different subscripts (a–c) were significantly different at $p < 0.05$.

the liver. Notable amounts of palmitoleic, docosapentaenoic and eicosapentaenoic acids are present in the skin. The gonads contain in particular hexadecadienoic, eicosenoic, stearic, linoleic and alpha linolenic acids.

3.2. Contents of total fatty acids per anatomical area

The total fatty acids (TFA) after extraction was expressed in grams per 100 g fresh weight ([Fig. 2\)](#page-2-0). Significant variation between the different areas is due to the position effect. The highest contents of TFA were found in the liver, the gonads and the red muscles (MR) with respectively 26, 4.7 and 4.5 g.

3.3. Contents of fatty acid groups per anatomical area

As shown in [Table 1,](#page-1-0) each of the SFA and unsaturated fatty acid (UFA) groups has its specific model of distribution. The position effect seems to account for the significant variations of SFA and UFA (MUFA + PUFA). Thus, the brain (C) is rich in SFA (38.61 \pm 1.56%) and poor in UFA (61.39 \pm 1.56%). Inversely, the liver has a low percentage of SFA $(25.41 \pm 3.13%)$ and a high percentage of UFA (74.58 ± 3.12%). [Table 1](#page-1-0) clearly illustrates both the abundance of UFA and the low contents of SFA according to their storage area.. The distribution of UFA and SFA is shown in [Fig. 3.](#page-2-0)

An analysis of the UFA/SFA ratio (Fig. 4) gives a better idea of these differences in distribution and confirms the important varia-

Fig. 4. Changes in the ratio UFA / SFA based on anatomical areas. TFA values with different subscripts (a-b) were significantly different at $p < 0.05$.

Fig. 5. PUFA and MUFA per anatomical area (as% of the TFA).

tions between the areas, such as a ratio of 3.5 for the liver and of 1.6 for the brain, and for the skin and the RM 2.8 and 2.7, respectively.

The UFA comprises the PUFA and the MUFA. The PUFA clearly distinguish ([Fig. 5\)](#page-3-0) the muscular areas M and RM, totalling, respectively, $46.11 \pm 1.01\%$ and $44.2 \pm 3.19\%$. The PUFA/MUFA ratios (Fig. 6) were, respectively, 2.0 and 1.8 for the muscular areas M and RM.

There is a significant presence of MUFA in the byproducts ([Fig. 5\)](#page-3-0), the richest organs being the liver and the eyeballs with, respectively, 42.44 ± 4.33% and 38.08 ± 3.38%.

[Table 1](#page-1-0) and Fig. 7 show that the muscular areas M and RM are high in n-3 PUFA, exceeding 35% of the TFA. The n-6 PUFA vary considerably between the muscles M and the gonads, making up, respectively, 6.06 ± 0.28 % and 6.03 ± 0.39 % of the TFA.

As to the byproducts, there is a high percentage of $n-9$ PUFA in the liver with $37.3 \pm 4\%$ and $n-7$ PUFA in the skin with $7.32 \pm 0.7\%$. EPA + DHA ([Fig. 8\)](#page-5-0) are high in the muscular areas M and RM (respectively, 31.8 ± 0.85% and 31.02 ± 3.04%).

The $n-3/n-6$ ratios range from 4.1 to 8.3, with significant differences from area to area [\(Table 1](#page-1-0)). According to the [Fig. 9,](#page-5-0) the lowest values are the gonads, the skin and the liver, with, respectively, 4.2, 5.2 and 5.7, due to their high contents of n-6 fatty acids.

The most important values are to be found in the eyeball (O) and the RM areas, with high percentages of $n-3$ fatty acids and low percentages of $n-6$ fatty acids.

Fig. 6. PUFA/MUFA ratios per anatomical area.

Fig. 7. $(n-3)$, $(n-6)$, $(n-7)$ and $(n-9)$ fatty acids per anatomical area $(\%$ of the TFA).

Fig. 8. EPA + DHA per anatomical area (% of the TFA).

Fig. 9. $n-3/n-6$ ratios per anatomical areas.

4. Discussion

The areas with the highest TFA contents are the byproducts: eyeball, brain, fat, liver, gonads and skin. They contributed 35.6 g of the TFA (out of 100 g of fresh material) as compared to 7.22 g for the muscular areas (M and RM).

The lipids as stored in the various parts of the species are a criterion of classification for certain categories of fish. The TFA contents (g/100 g fresh material) in the muscular areas of swordfish vary from 1 to 5 g, whereas the byproducts total 0.5 to 26 g. According to the classification of [Corraze and Kaushik \(1999\),](#page-6-0) the swordfish is considered a semi-fatty fish, storing its lipids in the muscles, the liver and other organs, such as the gonads (4.7 g). [Ack](#page-6-0)[man \(1994\)](#page-6-0) confirmed that fish with a medium lipidic storage rate contain 4–8 g out of 100 g of muscular tissue.

The tissues of the swordfish distinguish themselves by a high level of UFA and a relatively low level of SFA ([Table 1](#page-1-0)). The UFA make up between 61% and 74% of the TFA, while SFA make up 23% to 38% of the TFA. [Vlieg, Murray, and Body \(1993\)](#page-6-0) found in swordfish of the Indian Ocean about 24% SFA in the dorsal area, whereas the Mediterranean swordfish under study contains in the same zone 32%.

According to the dietary recommendations of the French Agency for Food Safety ([AFSSA, 2003\)](#page-6-0), the quantity of UFA should be three times the quantity of SFA. The ratios concerning the swordfish, as established in this study, appear perfectly balanced, except for the brain and the gonads. They are 3.5, 2.8, 2.71, 2.65, 2.57 and 2.4 for the liver, the skin, the RM, the eyeball, the fat embedding the brain and the white muscles. As illustrated in [Fig. 5,](#page-3-0) the muscular areas (M and RM) have a particularly high percentage of PUFA (40%), a value confirmed by the studies of Bergé and Barnathan (2005) attributing to fish PUFA values of 25% to 40%. The high contents of PUFA is due to the presence of $n-3$ fatty acids (see [Fig. 7](#page-4-0)), in particular the cumulative value of EPA + DHA ([Fig. 8](#page-5-0)). The highest concentrations of EPA + DHA were found in the muscles (M and RM), totalling 31% of the TFA, whereas Vlieg et al. (1993) report only 14% in the dorsal muscular tissue.

The byproducts are rich in MUFA [\(Fig. 5](#page-3-0)), which is due to the presence of n-9 fatty acids ([Fig. 7](#page-4-0)). Stocknes, Okland, Falch, and Synnes (2004) found, respectively, 48% and 39% of MUFA in the eyeball and the brain of salmon. In the liver of the gilthead sea bream Mnari et al. (2007) found 35.25% of MUFA. As to the swordfish in the present study, MUFA levels of, respectively, 38%, 33% and 42.4% were found in the eyeball, the brain and the liver ([Fig. 6](#page-4-0)).

The highest percentage of $n-3$ fatty acids is 37% in the muscles (M and RM) and the highest percentage of $n-6$ fatty acids is 6% in the area of M and the gonads. The $n-3/n-6$ ratio in dietary recommendations is 0.2 (AFSSA, 2003); in the swordfish of the Indian Ocean this ratio (Vlieg et al., 1993) is 5.8, whereas the findings concerning the Mediterranean swordfish confirm ratios of 6.6 (M) and 8.0 (RM).

The liver is high in $n-9$ fatty acids and the skin is high in $n-7$, fatty acids totalling respectively, $37.38 \pm 4\%$ and $7.32 \pm 0.72\%$ of the TFA. These fatty acids are not considered essential in human nutrition, unlike the linoleic, linolenic, arachidonic and eicosapentaenoic fatty acids which a FAO report declares essential for human dietary needs (WHO/FAO, 1977; Dumay, 2006).

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