academicJournals

Vol. 8(1), pp. 19-24, January, 2014
DOI: 10.5897/AJBR2013.0732
ISSN 1996-0778 ©2014 AcademicJournals
http://www.academicjournals.org/AJBR

African Journal of Biochemistry Research

*Full Length Research Paper*

# The composition of fatty acids stored in liver, muscle and fat tissues of the African lungfish *Protopterus annectens* (Owen, 1839)

**H. A. Elagba Mohamed[1]* and Jamal N. Al-Sabahi[2]**

[1]Natural History Museum, Faculty of Science, University of Khartoum, P.O. Box 321, Khartoum, Sudan.
[2]College of Agricultural and Marine Sciences, Sultan Qaboos University, P.O. Box 34, Al-Khoud, 123, Muscat, Sultanate of Oman, Sudan.

Accepted 4 December, 2013

**The composition and profile (%) of fatty acids stored in liver, muscle, and fat tissues of the fresh-water African lungfish *Protopterus annectens* were determined by gas chromatography. High content of total fatty acids (TFA) was found in fat (62.06 ± 3.4 g/100 g) followed by liver (50.68 ± 4.72 g/100 g) and muscle tissues (10.9 ± 0.81 g/100 g). Saturated fatty acids (SFA) formed 72.9% TFA in the liver and muscles and 53.3% in fat. Fat was rich in polyunsaturated fatty acids (PUFA), containing 31.1%, while muscle and liver contributed 12.9 and 10.6 % respectively. Fat and liver also contained, respectively, 9.59 ± 1.1 and 8.38 ± 1.9 g/100 g of monounsaturated acids (MUFA) compared to 1.56 ± 0.3 g/100 g in muscles. The major SFA were C16:0, C18:0 and C14:0. The MUFA were characterized by the Oleic (C18:1n9c) and Palmitoleic (C16:1) acids. The major PUFA in all three types of tissues was the Linoleic acid (C18:2n6c), while C20:3n6 was detected in fat only, and C18:3n3 was detected in muscles only. The n-3/n-6 ratio in the muscles was 0.5. The results of this study show that fatty acid composition in the African lungfish *P. annectens* can considerably vary, depending on the storage organ. According to the present results, *P. annectens* was considered a semi-fatty fish, storing its lipids in muscles, liver and fat tissues. These organs are potential source of essential fatty acids and may play an important role in human health. More focus on lipid extraction from *P. annectens* is recommended.**

**Key words:** Fat, fatty acids, freshwater, liver, lungfish, muscle, *P. annectens*.

## INTRODUCTION

Fat is stored in form of fatty globules in various parts of the fish such as the muscle and the liver, and as layers under the skin and in the body cavity (Sheridan, 1988; Ben Smida et al., 2009; Khoddami, et al., 2009). Various fats and oils from both seawater and freshwater fish were found to be rich sources of omega-3 long chain poly-unsaturated fatty acids (PUFAs), particularly eicosapen-tacenoic acid (EPA, 20:5n-3) and docasahexaenoic acid (DHA, 22:6n-3), as well as its precursor, alpha linolenic acid (C18:3n3) (Bays and Lansing, 1994; Shahidi and Wanasundara, 1998; Saba and Muhammad 2000; Ackman, 2002; Su et al., 2004; Bergé and Barnathan, 2005). An increasing interest in the health benefits of fish oil consumption has emerged, because these fatty acids play an important role in the prevention and treatment of cardio-vascular diseases (Nestel, 2000), improving of learning ability and lowering the levels of lipid in blood plasma (Uauy et al., 2001; Lim and Suzuki, 2002).

Fish oils extracted from the livers of fishes are commonly a rich source of beneficial fatty acids, including

*Corresponding author. Email: elagba2000@yahoo.com.

long chain polyunsaturated fatty acids like omega-3s, and fat-soluble vitamins A and D (Covadonga et al., 2004; Mnari et al., 2007; Guil-Guerrero et al., 2010). Fish oils are also extracted from non-consumable parts of the fish, such as head, skin, central bones, viscera (Shahidi et al., 1991; Sathivel et al., 2002; Stocknes et al., 2004; Nuraini et al., 2008; Khoddami et al., 2009). These parts may contribute to the total level of fatty acids, thus increasing the nutritional value of the fish. However, little is known about the fatty acids composition and profiles (%) of the Nile fishes (Elagba and Al-Sabahi, 2011) compared to other freshwater fishes (Ahlgren et al., 1994; Suriah et al., 1995; Zenebe et al., 1998; Bieniarz et al., 2000; Ackman et al., 2002; Fawole et al., 2007; Sharma et al., 2009; Ugoala et al., 2009). The present study is a first approach to determine the fatty acid composition and profiles of the African lungfish *Protopterus annectens*. It is important to analyze the fatty acid composition of the lungfish because is commonly eaten in western states of the Sudan and it is a popular marketed species among consumers in these rural areas. Therefore, the objective of this study was to determine the fatty acid composition and profile (%) in the muscle, liver and fat tissues of the freshwater lungfish *P. annectens*, to encourage the consumer to use these storage organs of the of the fish.

## MATERIALS AND METHODS

### Collection of samples

Fresh specimens of the African lungfish *P. annectens* (Owen, 1839) collected from the Nile water was obtained from the fish market in Khartoum. Muscle from dorsal and ventral parts of the fish, liver and fat tissues were carefully removed and minced. Three 1 g samples from each of the three tissues of each specimen were taken. The total lipids (TL) were extracted from the tissue samples with chloroform-ethanol mixture (2:1, v/v) according to the extraction method of Folch et al. (1957).

### Fatty acids analysis with gas chromatography

Fatty acids were analyzed as their methyl esters with a gas chromatography-mass spectrometry (GC-MS; Hewlett- Packard 5890 GC), according to the procedure of Ahlgren et al. (1994). The different fatty acids in the lungfish 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 concentration of individual fatty acid was calculated using heneicosanoic acid (C21:0) as internal standard. The results (means ± standard deviation, SD) were calculated both as concentration (g/100 g of wet tissue) and weight percentage (fatty acid profile). The results represent the mean values of a series of repetitions (n = 9).

## RESULTS AND DISCUSSION

The fatty acid contents in liver, muscle and fat tissues of the African lungfish *P. annectens* are shown in Table 1. Comparatively, the fat tissues express the highest content (62.06 ± 3.4 g/100 g) of TFA followed by the liver (50.68 ± 4.72 g/100 g) and the muscle tissues (10.9 ± 0.81 g/100 g). The levels of fatty acids stored in different tissues are considered as important criterion of classification for fish species. According to Ackman (1994), fish with a medium lipid storage rate contains 4-8 g/ 100 g of fatty acids in the muscular tissue. Therefore, according to the level of fatty acid found in liver (50.68 ± 4.72 g/100 g) and muscles (10.9 ± 0.81 g/100 g) of the lungfish, *P. annectens* can be considered a semi-fatty fish, storing its lipids in the muscles, the liver and fat tissues. The fat and liver tissues also contain higher levels of MUFA (9.59 ± 1.1 and 8.38 ± 1.9 g/100 g) and PUFA (19.52 ± 4 and 5.37 ± 2.7 g/100 g), respectively, compared to 1.56 ± 0.3 and 1.42 ± 0.3 g/100 g, respectively, for the muscle tissues. The fat tissues contributed 46.7% of TFA as unsaturated *fatty acid* (USFA), of which 31.1% were PUFA, while each of the liver and muscle tissues contributed 27.1% of TFA as USFA. This composition was similarly found in other fish, such as salmon, rainbow trout and some Nile fishes (Ahlgren et al., 1994; Zenebe et al., 1998; Bieniarz et al., 2000; Ackman et al., 2002; Fawole et al., 2007; Mnari et al., 2007; Khoddami et al., 2009; Elagba and Al-Sabahi, 2011).

The total fatty acids (UFA and SFA) in the liver, muscle and fat tissues of the lungfish are expressed in (g/100 g) fresh weight (Figure 1). There was a high level of SFA in the liver and muscle of lungfish (72.9 and 72.9%), respectively, compared to 53.3% in the fat tissues. The fat and liver tissues also contain higher levels of MUFA and PUFA (Figure 2) compared to the muscle tissues. A clear variation between the different organs can be observed. Comparatively, the results which expressed the different proportion of MUFAs and PUFAs with other fish lipids may be due to the environmental effect of tropical fish species (Suriah et al., 1995; Zenebe et al., 1998; Lim and Suzuki, 2002; Kwetegyeka et al., 2008). An analysis of the UFA/SFA ratio (Figure 3) gives a better idea of these differences in distribution and confirms the important variations between the organs. The PUFA are very high in the fat tissues, forming 31.1% of TFA. The PUFA/MUFA ratios (Figure 4) were, respectively, 0.6, 0.9 and 2.0 for the liver, muscle and fat tissues. There was a significant concentration of MUFA (9.59±1.1 and 8.38±1.9 g/100 g) in the fat and liver tissues (Figure 5).

The fatty acid compositions of total lipid (TL) from liver, muscle and fat tissues of the lungfish are presented in Table 1. A total of 17 fatty acids were identified in the analyzed tissues of the lungfish. As shown in the table, the profile of each fatty acid group and the distribution of SFA and unsaturated fatty acid UFA (MUFA + PUFA) vary also in different organs. The predominant SFAs in all samples were tricosanoic, palmitic, stearic and myristic acids. The highest levels of C16:0 (21.7%) were determined in the muscle lipid, and the highest levels of C18:0 (38.2 %) were determined in liver lipid. It was reported that palmitic acid was the predominant in SFA group in freshwater channel catfish, *Ictalurus punctatus*

**Table 1.** The contents of fatty acids (Mean ± SD, % of TFA) in the muscle, liver and fat tissues of the lungfish *Protopterus annectens*.

| Fatty acid | Liver | | Muscle | | Fat | |
|---|---|---|---|---|---|---|
| | % | g/100 g | % | g/100 g | % | g/100 g |
| C14:0 | 13.4 | 6.79 ± 1.52 | 11.1 | 1.22 ± 0.56 | 8.9 | 5.44 ±1.39 |
| C15:0 | 4.4 | 2.23 ± 0.34 | * | * | 4.6 | 2.82 ± 0.1 |
| C16:0 | 5.5 | 2.76 ± 15 | 21.7 | 2.3 ± 0.05 | 12.4 | 7.65 ± 2.13 |
| C17:0 | 7.8 | 3.95 ± 0.73 | 2.8 | 0.31 ± 0.06 | 12.4 | 7.61 ± 0.31 |
| C18:0 | 38.2 | 19.36 ± 3.26 | 14 | 1.53 ± 0.36 | 4.4 | 2.71 ± 0.68 |
| C22:0 | * | * | * | * | 2.8 | 1.72 ± 0.03 |
| C23:0 | 3.6 | 1.84 ± 2.9 | 23.3 | 2.56 ± 0.56 | 7.8 | 4.58 ± 0.62 |
| C14:1 | * | * | * | * | 1.4 | 0.88 ± 0.1 |
| C15:1 | * | * | 2.6 | 0.29 ± 0.05 | 2.1 | 1.28 ± 0.1 |
| C16:1 | 8.3 | 4.2 ± 1.09 | 4.1 | 0.45 ± 0.35 | 1.5 | 0.9 ± 0.1 |
| C17:1 | * | * | * | * | 3 | 1.84 ± 0.04 |
| C18:1n9c | 8.2 | 4.18 ± 1.8 | 7.5 | 0.82 ± 0.29 | 1.5 | 0.94 ± 0.07 |
| C20:1 | * | * | * | * | 6.1 | 3.75 ± 0.51 |
| C18:2n6c | 10.6 | 5.37 ± 0.99 | 6.5 | 0.72 ± 0.13 | 15 | 9.6 ± 0.52 |
| C18:3n3 | * | * | 3.3 | 0.36 ± 0.1 | * | * |
| C20:2 | * | * | 3.1 | 0.34 ± 0.08 | * | * |
| C20:3n6 | * | * | * | * | 16.1 | 9.92 ± 3.3 |
| ΣTFA | 50.68 ± 4.72 | | 10.9 ± 0.81 | | 62.06 ± 3.4 | |
| ΣSFA | 36.93 ± 6.7 | | 7.92 ± 1.1 | | 32.53 ± 2.4 | |
| ΣUFA | 13.75 ± 2.24 | | 2.98 ± 0.31 | | 29.53 ± 3.76 | |
| ΣMUFA | 8.38 ± 1.9 | | 1.56 ± 0.3 | | 9.59 ± 1.1 | |
| ΣPUFA | 5.37 ± 2.7 | | 1.42 ± 0.3 | | 19.52 ± 4 | |
| SFA / TFA | 72.9 | | 72.9 | | 53.3 | |
| MUFA / TFA | 16.5 | | 14.2 | | 15.6 | |
| PUFA / TFA | 10.6 | | 12.9 | | 31.1 | |
| UFA / SFA | 0.37 | | 0.38 | | 0.89 | |
| ΣN6 | 5.37 ± 0.99 | 10.6 | 0.72 ± 0.13 | 6.5 | 19.52 ± 0.27 | 31.1 |
| ΣN3 | * | * | 0.36±0.1 | 3.3 | * | * |

SD, Standard deviation; *, Not detected; TFA,total fatty acids; SFA, saturated fatty acids, MUFA, monounsaturated fatty acids; PUFA, polyunsaturated fatty acids; N, Omega 6; N3, Omega 3.

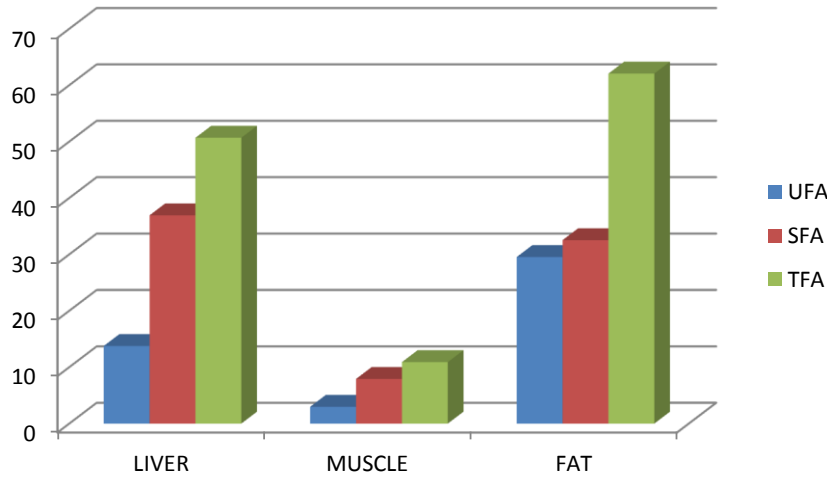


**Figure 1.** Contents of UFA, SFA and TFA (g/100 g) in different tissues of the lungfish.

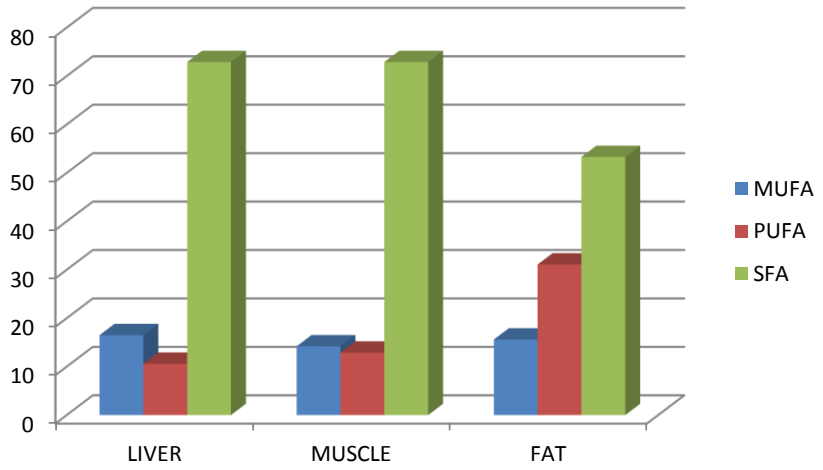


**Figure 2.** MUFA, PUFA and SFA in different tissues of the lungfish (as % of the TFA).

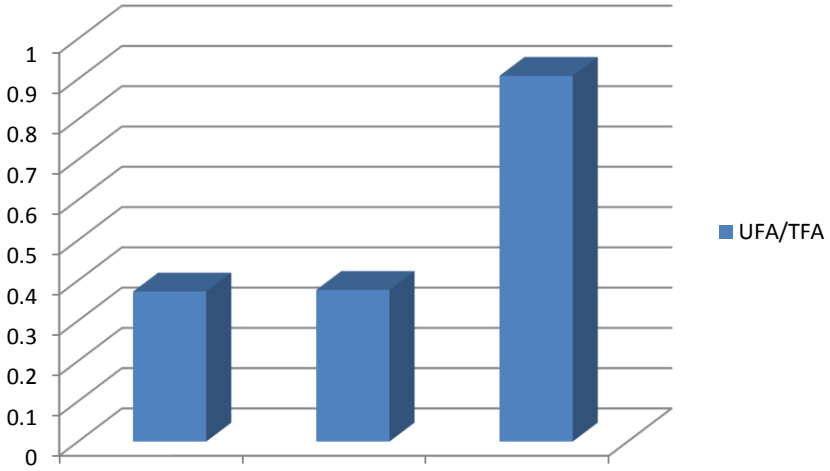


**Figure 3.** UFA / SFAin different tissues of the lungfish.

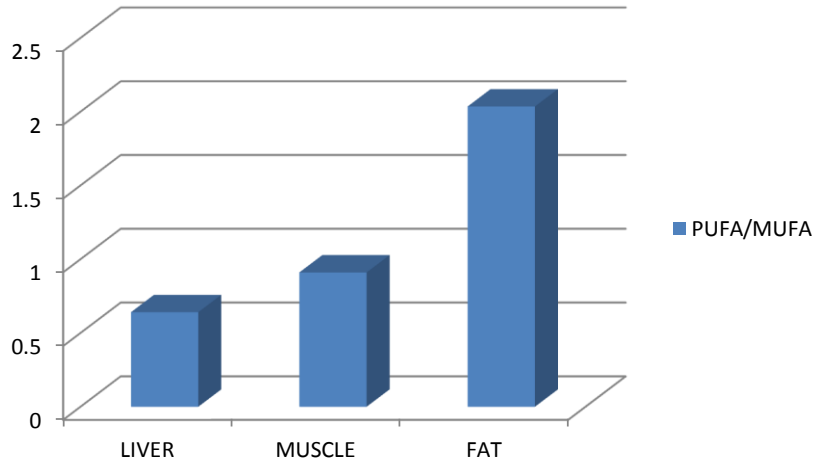


**Figure 4.** PUFA/MUFA in different tissues of the lungfish.

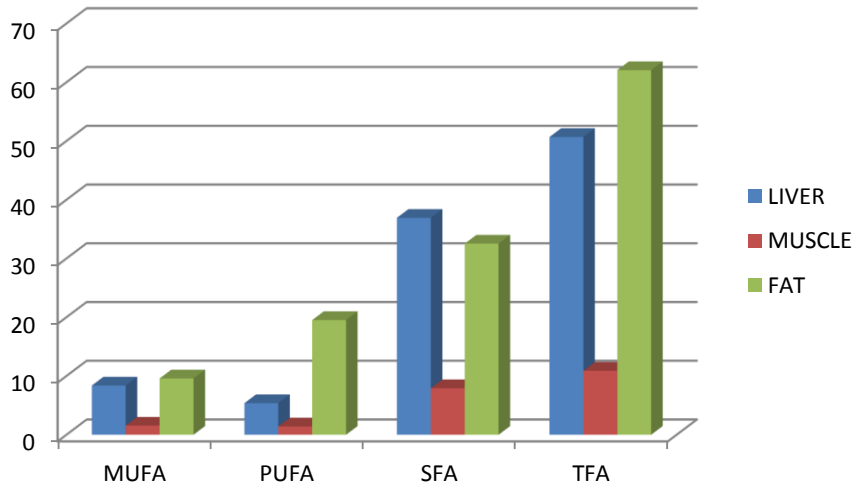


**Figure 5.** Contents of MUFA, PUFA, SFA and TFA (g/100 g) in different tissues of the lungfish

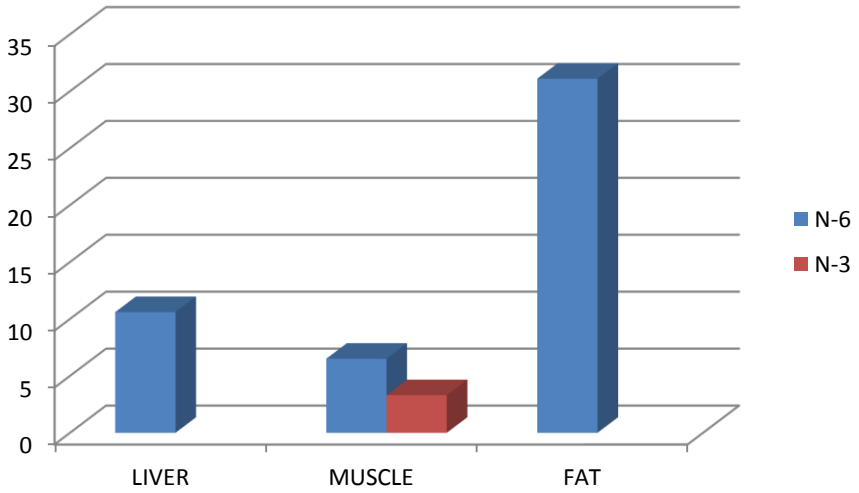


**Figure 6.** Content of omega-6 (N-6) and omega-3 (N-3) in different tissues of the lungfish (as % of the TFA).

(Sathivel et al., 2002), in freshwater rainbow trout, *Oncorhynchus mykiss* (Haliloglu et al., 2004) and in the catfish *Pangasius hypophthalmus* (Ho and Paul, 2009). The percentage of PUFA in fat, muscle and liver of the lungfish, was 32.1, 12.9 and 10.6%, respectively. The major PUFA was the lenoleic acid (C18:2n6). Eicosatrienoic (C20:3n6) and lenolenic (C18:3n3) acids formed 16.1 and 3.3%, respectively, in the fat. The percentage of monounsaturated fatty acid (MUFA) of liver, fat and muscle lipids was 16.5, 15.6 and 14.2%, respectively. Oleic (C18:1n-9) and palmitoleic (C16:1) acids were the major MUFA. Oleic acid constituted 8.2 and 7.5%, and palmitoleic acid constituted 8.3 and 4.1%, in liver and muscle, respectively. The high level of oleic acid found in the lungfish was consistent with its level in other species of freshwater fish. Steiner-Asiedu et al. (1991) found that freshwater tilapia (*Tilapia* sp.) had significantly higher level of oleic acid than the flat sardine (*Sardinella* sp.) and sea bream (*Dentex* sp.). The level of oleic acid in the American freshwater channel catfish flesh was also high compared to sardine and sea mullet (Ackman, 1994). Although level of oleic acid was lower than the level found in some freshwater fish (Khoddami et al., 2009), it was almost similar to the level reported in other freshwater fish (Zenebe et al., 1998; Bieniarz et al., 2000; Saba et al., 2000; Ackman et al., 2002; Ben Smida et al., 2009; Sharma et al., 2009; Ugoala et al., 2009; Elagba and Al-Sabahi, 2011). The variation of n-3 PUFA detected in the different studied organs of *P. annectens* (Figure 6) was to some extent within the range found in other fish species (Suriah et al., 1995). The difference may be due simply to differences in the fatty acid content

in the diet or it may be related to environmental conditions, sex and age of the fish (Steffens, 1997).

## Conclusions

This study indicates that the lipids of the African lungfish *P. annectens* are rich in linoleic and linolenic acids which are essential for human healthy diet. Although, the level of SFA was high in the liver and fat, PUFA accounted for the highest proportion of FA in the fat. The liver and fat (which are considered as waste) may provide a rich unexploited source of polyunsaturated fatty acids. Therefore, more focus on lipid extraction from the lungfish is needed and more detailed studies are necessary.

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