Comparative study on the liver fatty acid profiles of the red toothed trigger fish (*Odonus niger*) from

southwest coast of India

Immanuel G.* and Palavesam A.

Received: August 2008

Accepted: June 2009

Abstract

In order to find an alternative source for the highly unsaturated fatty acids oil with lower cost, a marine trash fish Odonus niger has been analysed and characterised. The body weight and the corresponding liver weight of the fish were studied and the results showed that for one gram increase in the size of the animal, the liver weight can increase to 0.04 g. The oil yielding capacity of the liver was assessed by four different methods namely Soxhlet, Bligh and Dyer, Direct steaming and Solar extraction. The percentage of oil yield was high in Soxhlet method (67.7%), but it was 54.3% in Bligh and Dyer method, 42.5% in direct steaming method and minimum of 32.0% in solar extraction method. The solidification point of all the extracted oils remained at $29 \pm 0.5^{\circ}$ C. The specific gravity (0.95 to 0.96) and refractive index (1.42µ to 1.48µ) of the oil extracted by the four different methods were not varied significantly. The cholesterol contents of the oil extracted by Bligh and Dyer and direct steaming extraction methods were at the highest level (1991.00 and 2059.00mg 100ml⁻¹) but it was 50% less in other methods. The percentages of PUFA in the total fatty acid of the oils were 13.78, 20.46, 19.07 and 22.54% (by weight) in solar extraction, direct steaming, Soxhlet and Bligh and Dyer methods, respectively. Thus the physico-chemical properties of liver oil of O. niger were found to be influenced by the extraction adopted methods. Also it is clear that Bligh & Dyer method is the suitable method for the extraction of liver oil from marine fishes without much loss of nutrients.

Keywords: Odonus niger, PUFA, FFA, Soxhlet, Bligh and Dyer, Direct steaming, Solar extraction

Marine Biotechnology Laboratory, Centre for Marine Science and Technology, Manonmaniam Sundaranar University, Rajakkamangalam – 629 502, K.K. District, Tamilnadu, India

^{*}Corresponding author's email: gimmas@gmail.com

Introduction

Among the fishes of the world, the family Balistidae constitutes a substantial fishing in certain areas during particular months in a year (Immanuel *et al.*, 2000). Among them *Odonus niger* (Ruppel), is a demersal fish, which occurs in enormous shoals and form regular fishery in the south west coast of India (Immanuel *et al.*, 2000, 2009) and obtained in large quantities in trawling during monsoon season as well as in other months. This species is commonly treated as trash fish and used for poultry feed along with huge quantity of livers (Immanuel *et al.*, 2000).

In fishery industries, the value added fishery products such as fats and oils are extracted from whole body or liver of several fish species. Various authors have studied the extraction methods of oil from body and livers of fishes and analysed the physical and chemical properties, e.g., shark liver oil (Shenoy & Dey, 1984; Davidson & Cliff, 2002) and dogfish liver oil (Sunarya et al., 1992); rays liver oils (Garcia et al., 2004) and teleost fish species liver oil (Zheng et al., 2004). The fish oils and marine animal oils are characterized by a large group of fatty acids. These fatty acids can be classified as saturated, monounsaturated and polyunsaturated groups. Moreover these oils are having rich source of energy and vitamin A and D (Nair & Gopakumar, 1985; Rossell, 1986; Adrianus, 1993; Aidos et al., 2002; Adebiyi and Bawa, 2006; Immanuel et al., 2009).

Liver oil of different sources will have various physical and chemical properties.

Sunarya *et al.* (1992) reported that very small amount of cholesterol, which is the un-saponifiable fraction of oil present in the liver oil of dogfish. Iwasaki and Harada (1984) reported that the blue shark liver tissues contain large quantity of cholesterol.

Adopted methods for isolation of fats have significant influence on its composition. Sunarya et al. (1992) reported signifycant changes in fatty acid profile as well as cholesterol percentage of dogfish liver oil extracted by different methods (i.e., direct steaming, Soxhlet, Bligh and Dyer methods). Literature pertained to the above is still wanted as for O. niger is concern. Therefore the present study was under taken to find out the liver index, suitable methods for liver oil extraction and physicochemical characterization of oil.

Materials and methods

A total of 500 specimens (*O. niger*) were randomly collected from the landing centres of Kanyakumari District, Southwest coast of India in different fishing seasons (From June to November, 2004). The length (cm, total length) and weight (g, fresh weight) of each specimen was measured. The lengthweight pairs were plotted in order to find their growth. The parameters and related statistics of the log transformed LWR (logW = loga + blogL;

Where W = weight of fish in grams, L= total length of fish in centimeters, a=constant of proportionality, b=allometry coefficient) were obtained via ordinary least square regression. Then the abdomen of each specimen was dissected and the liver was carefully removed and weighed separately using an electrical monopan balance (accuracy \pm 0.1 g). The weight of the fish and the liver were related using the hepatosomatic index (HSI) formula.

HSI (%) =
$$\frac{\text{Weight of liver (g)}}{\text{Weight of fish (g)}}$$
 x 100

Liver of *O. niger* were weighed and chilled in ice and packed in air tight containers prior to arrival in the laboratory. They were in good condition and did not show any visual trace of spoilage. In the laboratory, the ice cubes were discarded and all the unwanted materials such as blood vessels and visceral materials were removed, cleaned well and were stored at -20° C in a deep freezer until use.

Four different methods were adopted for the extraction of oil from the liver.

- i) Solvent Extraction (Soxhlet) method (Folch *et al.*, 1957)
- ii) Bligh and Dyer method (Bligh & Dyer, 1959)
- iii) Direct steaming method (Sunarya *et al.*, 1992)
- iv) Solar extraction method (crude method)

Solar extraction method was standardized in our laboratory after being thawed the livers at ambient temperature. The livers were weighed and placed in a glass bowl, exposed under hot sunlight $(35\pm2^{\circ}C)$. Within 15 min. the oil started to melt and at every 30 min intervals the melted oil has been collected in a well-cleaned glass beaker and within 4 to 5 hours, the complete oil was separated. The percentage of oil yield was calculated as:

 $\begin{array}{c} \text{Weight of the extracted oil (g)} \\ \text{Yield of oil=} & \hline \\ \hline \\ \text{Weight of the liver (g)} \end{array} x 100 \end{array}$

Similarly all the oil extracted by the three methods was quantified.

After extraction, the oils were kept in separate containers in freezer $(-20^{\circ}C)$ until further use. The physical properties such as solidification point, specific gravity and refractive index of the different extracted oils were analysed by the method of Narasimhan *et al.* (1965) and AOCS (1998). The cholesterol content was estimated by the method of AOCS (1998).

Fatty acid methyl esters (FAMEs) of oil samples were prepared according to the AOCS (1998) official method Ce 1b-89 and analyzed with regard to the amount of individual fatty acids. The different FAMEs were separated from each other with gas chromatography (GC) (Miller & Berger, 1985).

The results obtained were analysed through one-way ANOVA and linear regression tests followed by Zar (1974).

Results

The harvested fish samples were measured 13.0cm length and the largest one was 22.5cm length. The corresponding weight was also varied from 45.0 to 120.0g (Table 1). The length-weight data obtained for the 500 fish were plotted and obtained a linear trend relationship (Fig. 1). Linear regression statistics computed (logW = 0.0296 + 2.966 logL) was correlated (r² = 0.9727) and significant (P<0.001). The liver weight of

the smallest fish was 4.0g and increased to 7.40g in the largest fish sampled. The relation between the body weight and the liver weight was highly significant (P<0.001), indicating that increasing by gram caused the animal size and the liver weight increase (Table 1).

The percentage of oil yielded from the fish liver in different extraction methods were given in Figure 2. Among the tested methods, the difference between the percentage of oil yielded ($67.7\pm0.68\%$) in Soxhlet extraction method and the former was significantly higher (P<0.001) than that in other methods. In Bligh and Dyer and direct steaming methods, the percentage of oil yield recorded were 54.3 ± 0.66 and $42.5\pm0.40\%$, respectively. In solar extraction method the lowest yield of $32.0\pm0.91\%$ oil was registered.

The physico-chemical properties of the oils extracted by the different methods were

given in Table 2. Irrespective of the different methods followed for the extraction of oil, the solidification point remained at 29.0±0.5°C. The specific gravity of the oils extracted by four different methods were 0.9581, 0.9519, 0.9523 and 0.9525 in Soxhlet, Bligh and Dyer, direct steaming and solar extraction methods, respectively. The refractive index at 29°C was also almost similar in all the four methods; they were 1.426, 1.466, 1.483 and 1.477µ in Soxhlet, Bligh and Dyer, direct steaming and solar extraction methods, respectively. The cholesterol content of the oil extracted by Bligh and Dyer and direct steaming extraction methods was at higher level (1991mg and 2059mg $\times 100$ ml⁻¹). In the other two methods, the content of cholesterol was reduced.

Length of fish (cm)	No. of fish (n = 500)	Body weight range (g)	Mean body weight (g)	Liver weight range (g)	Mean liver weight (g)	Liver index (%)
12.5 - 13.5	26	45 - 47	45.75±0.82	4.0 - 4.5	4.12±0.21	9.15
13.5 - 14.5	48	46 - 50	48.60±1.85	4.0 - 4.5	4.30±0.24	8.84
14.5 - 15.5	73	50 - 57	53.93±2.20	4.5 - 5.0	4.76±0.35	8.45
15.5 - 16.5	52	57 - 65	61.00±2.79	5.0 - 5.5	5.15 ± 0.22	8.44
16.5 - 17.5	89	70 - 77	74.11±1.85	5.0 - 5.5	5.41±0.18	7.29
17.5 - 18.5	104	80 - 87	83.57±2.27	5.5 - 6.0	5.88±0.26	7.03
18.5 - 19.5	57	87 - 93	90.00±1.63	6.0 - 6.5	6.16±0.23	6.85
19.5 - 20.5	23	98 - 104	100.00±2.44	6.5 - 7.0	6.70±0.26	6.70
20.5 - 21.5	16	108 -114	110.66±2.49	7.0 - 7.5	7.30±0.24	6.59
21.5 - 22.5	12	118 -120	119.00±1.00	7.0 - 7.5	7.40±0.10	6.21

Table 1. Longth and weight relationship	n of trach fich A	niger and its live	n index (n - 500)
Table 1: Length and weight relationshi	p of it asil fish 0.	. <i>mger</i> and its inv	muex (m - 300)

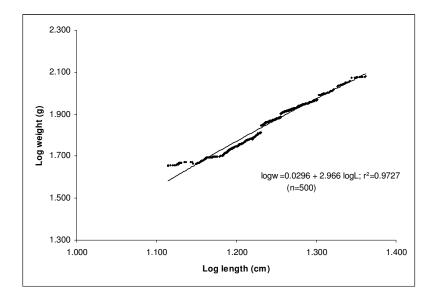


Figure 1: The relationship between the total length (cm) and body weight (g) of trash fish (*O. niger*) in the present study

Parameters	Soxhlet	Bligh &Dyer	Steaming	Solar
	Extraction	Extraction	Extraction	Extraction
Solidification point (°C)	29±0.5	29±0.5	29±0.5	29±0.5
Specific Gravity	0.9581	0.9519	0.9523	0.9525
Refractive Index (μ) (at 29°C)	1.426	1.466	1.483	1.477
Cholesterol (mg 100ml ⁻¹)	1159±7.34	1991±3.55	2059±6.53	1091±5.71

Table 2: Physical and chemical composition of extracted liver oil in *O. niger* with different methods

Each value is a mean (± SD) of three replicate samples

The details of the fatty acid profile of the trash fish liver oil extracted by different methods were given in Table 3. The most abundant fatty acid in the *O. niger* liver oil was C16: 0 (Palmitic acid), i.e., 21.70% in Soxlet method, 18.50% in Bligh and Dyer method, 19.26 % in direct steaming method, and 24.60% in solar extraction method.

The fatty acid level showed that the saturated fatty acid Methyl esters (Fame) in the different methods of extracted oils were, 52.73, 47.05, 49.05 and 59.54 % in Soxhlet, Bligh and Dyer, direct steaming and solar extraction methods, respectively. The mono unsaturated fatty acid percentage in the oil extracted by solar extraction method was lower (26.68%), more or less same percentage in the oils of direct steaming and Bligh and Dyer methods (30.49 and 30.41%). In Soxhlet method, it was 28.20%.

The detected polyunsaturated fatty acids (PUFA) in the fame were minimum of 13.78% in solar extraction method and maximum of 22.54 % in Bligh and Dyer method. Soxhlet and direct steaming extraction methods have more or less same percentage of PUFA (19.07% and 20.46 %).

Carbon No.	Soxlet extraction	Blig & Dyer extraction	Steaming extraction	Solar extraction	Summary of One-way ANOVA
C9:0	nd *	0.39	0.004	nd	S (F = 1460.036)
C 9 . 0 C10 : 0	0.86	1.43	1.21	nd	S(F = 1400.030) S(F = 132.213)
C10:0 C11:0	2.43	1.43	1.21	2.62	S(F = 132.213) S(F = 14626.063)
					· · · · · · · · · · · · · · · · · · ·
C12:0	0.83	nd	0.65	0.30	S (F = 726.333)
C13:0	1.00	1.16	0.96	1.21	S (F = 65.296)
C14:0	4.86	2.05	4.39	5.92	S (F = 2179.184)
C15:0	2.82	2.06	2.65	2.60	S (F = 160.082)
C16:0	21.70	18.50	19.26	24.60	S (F = 757.556)
C17:0	1.74	1.03	1.53	4.07	S (F = 730.676)
C18:0	13.96	14.70	14.23	15.88	S (F = 288.090)
C19:0	nd	nd	nd	nd	_
C20:0	1.78	2.42	1.76	1.45	S (F = 265.533)
C21:0	nd	0.12	nd	0.14	NS (F = 0.500)
C22:0	0.55	0.78	0.69	nd	S(F = 33.580)
C23:0	0.20	nd	0.41	0.63	S(F = 462.330)
C24:0	nd	0.27	nd	0.12	S(F = 112.500)
C14:1	0.29	0.43	0.38	0.13	S(F = 173.580)
C16:1	5.17	4.32	5.32	6.46	S (F = 1935.063)
C18:1	18.29	17.84	18.05	14.43	S (F = 1331.210)
C20:1	2.32	4.02	4.14	3.29	S (F = 1121.747)
C22:1	2.13	3.80	2.60	2.37	S (F = 449.332)
C18:2	11.64	12.87	11.62	8.89	S (F = 1131.053)
C18:3	nd	0.21	nd	0.01	S (F = 800.000)
C20:4	2.48	2.66	2.74	1.54	S (F = 491.253)
C20:4 C20:5	2.05	2.86	2.80	1.86	S (F = 290.916)
C20:5 C22:5	1.40	1.64	1.70	1.00	S(F = 161.440)
C22:5 C22:6	1.40	2.30	1.60	0.48	S(F = 101.440) S(F = 12549.000)
C22:0	1.30	2.30	1.00	0.40	$S(\Gamma = 12349.000)$
εSAFA	52.73	47.05	49.05	59.54	S (F = 120.740)
εMUFA	28.20	30.41	30.49	26.68	S(F = 13.625)
εPUFA	19.07	22.54	20.46	13.78	S(F = 55.881)

Table 3: Fatty acid composition (% - by weight) in liver oil extracted by different methods in	the
present study	

* Not detected

SAFA= Saturated fatty acids; MUFA= Mono unsaturated fatty acids;

PUFA= Poly unsaturated fatty acids

Fatty acid content is expressed as area percent FAMEs and % (by dry weight). Each value is a mean of three replicate samples. S = Statistically significant (P<0.05); NS = Statistically non significant (P>0.05)

S = Statistically significant (P < 0.05); NS = Statistically non significant (P > 0.05) Values in parenthesis are the Statistical One way- ANOVA - F-value.

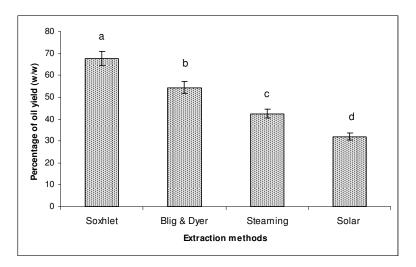


Figure: 2 Percentage oil yield (w/w) from different methods of extraction of *O. niger* liver Each value is a mean of three replicate samples; Figure with different alphabets is statistically significant (P < 0.05)

Discussion

According to Le Cren (1951) length of the fish is often more rapidly and accurately measured than weight, for calculating weight from length, it may also give indication of taxonomic difference and events in the life history of the studied fish. The general expectation is that the weight of fishes would vary on the cube of length (Lagler, 1952; Beverton & Holt, 1957; Brown, 1957; Rounsefell & Everhart, 1959; Brody, 1965). This aspect has been taken into consideration in the present study of length-weight relationship of trasfish O. niger of Southwest coast of India and the correlation coefficient obtained for the relation between them was statistically significant.

Shenoy & Dey (1984) reported that the liver of shark usually weighs about one fourth (25%) of the total weight of the fish. In Balistid fish *Sufflamen capistratus*, the percentage was 5.6% (George *et al.*, 1976). Whereas in *O. niger*, the percentage of liver to the body weight was 9.15% in the smallest weight group (45-47g) and 6.21 % in the largest weight group (118-120g). The result obtained in the present study elucidates the impact of method of extraction on oil yield as well as its properties. The oil extracted from the liver of O. niger was higher in Soxhlet extraction and Bligh and Dyer methods. The reason for the lowest yield of oil in the steaming and the solar extraction methods may be due to rupture of the liver cells during processing (Sunarya et al., 1992; Immanuel et al., 2009). Bailey (1942) found after decanting the first portion of free oil from the steamed and centrifuged dogfish liver, the residue gave oil yield of 2 to 10 % when it was solvent extracted. The Soxhlet and Bligh and Dyer extractions gave results in agreement with the data for the oil content of dogfish liver caught from many areas of Pacific region and Japan, which have been cited as 50-70% (Brody, 1965; Kayama et

al., 1969; Kizevetter, 1973; Murrey & Burt, 1983).

Fats have definite melting point. They are usually low and vary in each case. The melting point of a fat is always higher than the temperature at which it solidifies. The melting points of natural fats are not sharp because they are mixture of several fats. Presence of saturated fatty acids in fact increases the melting point and vice versa. This property causes the fat to be liquid or oil state at room temperature. Solidification temperature of beef fat is $36^{\circ}C$ (Shanmugam, 1990). Similarly in the present study the solidification point for the oils remained at $29\pm0.5^{\circ}C$ in all four methods.

Shanmugam (1990) pointed out the specific gravity of solid fat (0.86) to be less than the specific gravity of liquid fat (0.91 to 0.95). The specific gravity of *O. niger* liver oil extracted by the four methods have the same value of 0.95 (at 29° C) which describes that the oil extracted from the fish is liquid fat (0.91 to 0.95).

Fats have definite angles of refraction and any deviation from the normal value indicate mixture of fats. According to Pillai (1963), the refractive index of *O. niger* liver oil extracted by steaming method has 1.45μ at 28° C, whereas in the present study the refractive index was slightly varied in between 1.42 and 1.48 μ at 29° C in the four methods.

All fats and oils contain 0.1 to 2% complex alcohol known as sterols (Jacobs, 1965; Young, 1986) and this level was varied in the oil extracted by various methods. In the present study in Bligh and

Dyer and steaming extraction methods, the level of cholesterol was almost 50 % higher than the other two methods. But Sunarya *et al.* (1992) found that the cholesterol content was in similar level (1.1%) in the three different methods of extraction of oils from dogfish liver (Bligh & Dyer, Soxhlet and steaming methods). In dogfish liver oil, Kayama *et al.* (1969) reported 0.93% cholesterol. Iwasaki and Harada (1984) reported 1.11% cholesterol in blue shark liver oil.

The oils of fish and other marine animals are characterized by a large group of saturated and unsaturated fatty acids, which are commonly associated with mixed triglycerides. In addition to these, the oils contain small quantity of fatty acids in phospholipids. Compared to body fat, liver oils and oils from particular organs of fish and other marine animals can often contain large amount of fatty acids associated with phospholipids, glycerol, ethers and wax esters (Lovern, 1962; Young, 1986; Davidson & Cliff, 2002; Garcia *et al.*, 2004; Immanuel *et al.*, 2009).

In the present study, the total saturated fatty acid content of the fish *O. niger* liver oil extracted by different methods were much varied (47.05-59.54%). The saturated fatty acids of marine animals are ranged between 17.3 and 61.6%. For instance, in cod liver oil, the saturated fatty acid percentage was 17.3 and in Atlantic herring oil and Menhaden oil, it was 21.5 and 33.3% respectively (New, 1987). In Madura anchovy and gizzard shade, it was 48.9 and 61.6% (Nair & Gopakumar, 1977) etc. Nair

& Gopakumar (1982) studied the saturated fatty acid percentage of Tilapia mossambica both brackish water and fresh water separately and they found that T. mossambica brackish water species has 50.67 to 53.45% of saturated fatty acids, whereas fresh water species has 47.80 to 48.56% of saturated fatty acids. Ackman et al. (1963) pointed out the oil from Newfoundland capelin (Mallotus villosus) associated with 22.60 to 23.50% of saturated fatty acids and also Indian oil sardine has 40.32 % of saturated fatty acids (Raj, 1993). Similar to the above, in the present study also a remarkable variation was found on saturated fatty acid level, it is because of the differences between oil extraction methods. For instance, the saturated fatty acid level of oil samples extracted by Soxlet, Blig & Dyer, Steaming and Solar extraction methods was 52.73. 47.05, 49.05 and 59.54% respectively. In accordance with this, Immanuel et al (2009) reported that the saturated fatty acid level of liver oil samples of S. capistratus was ranged between 36.32 and 63.84% in the above extraction methods.

The monounsaturated fatty acid content of oils of marine fishes varied from 16.9 to 77.1%. Madura anchovy fish oil has the maximum level of 16.9% monounsaturated fatty acids (Nair & Gopakumar, 1977). New (1987) reported the highest level of 77.1% monounsaturated fatty acids in cod liver oil. The oil separated from Atlantic herring contains 73.9% of monounsaturated fatty acids, which was separated from the whole body (New, 1987). Meanwhile very low percentage of monounsaturated fatty acids was separated from the whole body oil of Madura anchovy. Liver oil of fish normally contains high level of monounsaturated fatty acids (New, 1987; Davidson & Cliff, 2002), but in the present study, the monounsaturated fatty acid content of different oil samples extracted by various methods was ranged from 26.68 to 30.49%. Similarly, Immanuel *et al.* (2009) reported the monounsaturated fatty acid level of *S. capistratus* liver oil samples extracted by different methods was ranged from 23.76 to 40.90%.

In O. niger liver oil, oleic acid (C18: 1) and palmitoleic acid (C16: 1) were found to be the major monoenic acids of the oil extracted by various methods. These two acids were found to be more or less same percentage reported for sardine oil by Gopakumar & Nair (1966) and less in dog fish liver oil reported by Sunarya et al. (1992). C14: 1 was absent in several fish oils (e.g. dog fish liver oil: Sunarya et al., 1992). But in O. niger liver oil, the amount of C14: 1 is too low (0.13 to 0.43%), whereas C20: 1 and C22: 1 were in higher percentage in dogfish liver oil than the liver oil of O. niger. In the present study, the polyenic acids (PUFA) constitute 13.78 to 22.54% of the total fatty acids. Among these cislinoleic acid (C18: 2) was the major constituent (8.89 to 12.87%). In evidence to this, Immanuel et al (2009) also reported that cislinoleic acid was the dominant fatty acid among the PUFA and the level of this particular acid recorded was ranged between 5.05 and 11.85%. But in certain fish oils the cislinoleic acid reported was very low or absent. For eg., in New found land capelin fish oil, it was absent (Ackman *et al.*, 1963), whereas it is less in Atlantic Herring oil (1.7%), Menhaden oil (2.0%), Cod liver oil (1.8%) and Pollack liver oil (5.0%) (New, 1987; Akiyama & Dominy, 1989).

According to Ackman (1982) in marine oils, the dominant PUFA are those of the series of C20: 5n-3 and C22: 5n-3, but in the present study the percentage of these two acids occupied second position next to C18: 2 in the PUFA series. C18: 3 were present in trace in solar and Bligh and Dyer methods, but in the other two methods it was not detected. In Atlantic herring and cod liver oils also this acid was very low (New, 1987). Among C20 PUFA series, C20: 4 and C20: 5 were the major acids, i.e., 1.54 to 2.74% and 1.86 to 2.86% respectively in the present study. Whereas in cod liver oil C20: 4n-6 is in trace amount, but presence of 20: 5n-3 was relatively higher amount (8.9%) (New, 1987). In dogfish liver oil, C20: 4n-3 was absent and C20: 5n-3 was representing 4.4 to 4.8% (Sunarya et al., 1992). In pollack liver oil, C20: 4n-3 was absent, but C20: 5n-3 was present in fairly good amount of 12% (Ackman et al., 1963).

In the C22 PUFA series, only C22: 2, C22: 3 and C22: 4 were not detected in the present study. In dogfish liver oil also these series of PUFA were absent except C22: 5 and C22: 6 (Sunarya *et al.*, 1992). The percentage of C22: 5n-3 and C22: 6n-3 in dogfish liver oils was 2.3 to 2.6% and 9.9 to 11.7%, respectively. But in the present

study the amounts were 1.0 to 1.7% (C22: 5n-3) and 0.48 to 2.30% (C22: 6n-3) respectively. In Atlantic herring oil 1.0 and 3.8%, Menhaden oil 1.7 and 7.9%, cod liver oil 1.6 and 9.3% of C22:5 and C22: 6 (New, 1987).

Considering the quantity of oil yielded and their biochemical constituents, Soxhlet and Bligh and Dyer methods were found to be suitable for *O. niger* liver oil extraction.

Acknowledgement

The first author wishes to thank Department of Science and Technology (DST), Ministry of Science and Technology, Govt. of India, for its financial support, in the form of a research grant (Young Scientist Fellowship – SR/FT/L-147/2004).

References

- Ackman, G., 1982. Fatty acid composition of fish oils. *In*: Nutritional evaluation of long chain fatty acids in fish oils. (S.M. Barlow and M.E. Stansby eds.), Academic press, London, UK. pp.25-88.
- Ackman, G., Jangaard, P.M., Burgher, R.D., Huges, M.L. and Macallum, W.A., 1963.
 An account of fatty acid components of Newfoundland capelin (*Mallotus villosus*).
 Journal of American Oil Chemical Society, 40:564-567.
- Adebiyi, O.D. and Bawa, A.A., 2006. Ackeral (Scomber scrombrus) oil extraction and evaluation as raw materials for industrial utilization. Leonardo Journal of Science, 8:33–42.

- Adrianus, J.K., 1993. Phospholipids of Marine origin- The squid (*Loligo Vulgaris*). Journal of Scientific Food and Agriculture, 61:129-132.
- Aidos, I., Van der Padt, A., Luten, J.B. and Boom, R.M., 2002. Seasonal changes in crude and lipid composition of herring fillets, by products and respective produced oils. Journal of Agriculture Food Chemistry, 50:4589- 4599.
- Akiyama, D.M. and Dominy, W.G., 1989. Penaeid shrimp nutrition for the commercial feed industry. Proceedings of the people republic of China, Aquaculture and feed work shop, Ed. D.M. Akiyama, pp.189-236.
- AOCS, 1998. Official methods and recommended practices of the American oil chemists' Society. 5th edn., AOCS: Champaign, Illinois, USA. 848P.
- Bailey, B.E., 1942. Vitamin A investigation III. The preparation of Vitamin A oil from dogfish livers. Progress Report on Pacific Coast Station. 50:10-12.
- Beverton, R.J.H. and Holt, S.J, 1957. On the dynamics of the exploited fish populations.
 Fisheries Investigation, Ministry of Agriculture, Fisheries & Food, Great Brittan, Series, 2:19-533.
- Bligh, E.G. and Dyer, W.J., 1959. A rapid method of total lipid extraction and purification. Canadian Journal of Biochemistry and Biophysics, 37:911-917.

- Brody, L., 1965. Fishery byproducts Technology, The AVI Publishing Company Inc. Westport (Connecticut), pp.47-95.
- Brown, M.E., 1957. Experimental Studies on growth. *In*: Physiology of fishes, Ed. M.E. Brown, Academic Press, New York, USA. pp.361–400.
- Davidson, B. and Cliff, G., 2002. The liver lipid fatty acid profile of seven Indian Ocean shark species. Fish Physiology and Biochemistry, 26:171-175.
- Folch, J., Lees, M. and Sloane Stanely, H., 1957. A simple method for the isolation and purification of total lipids from animal tissues. Journal of Biological Chemistry, 226:497-509.
- Garcia, G.N., Aguilar, R.P., Alvarado, L.B. and Garcia, J.O., 2004. Characterisation of the lipid composition and natural anti oxidants in the liver oil of *Dasyatis brevis* and *Gymnura marmorata* rays. Food Chemistry, 87:89-96.
- George, A.I., Nazir Ahamed, M.S. and Sivadason Asari, T.N., 1976. Observations on an unusually heavy landing of *Sufflamen capistratus* (Shaw) (Balistidae) along South Kerala coast during 1971-1972. Bulletin of the department of fisheries, Kerala, India. 1(1):56-60.
- Gopakumar, K. and Nair, M., 1966. Studies on fish lipids –II. Fatty acid composition of lipids of oil sardine (*Sardinella longiceps*). Fisheries Technology, 3(1):21-25.

- Immanuel, G., Peter Marian, M. and Palavesam, A., 2000. A note on the trashfish resources of Southwest coast of India during the years 1991- '93. Ecology and Ethology of Aquatic Biota. Ed. K. Arvind Kumar, Daya Publication. I(24):332-336.
- Immanuel, G., Sathasivan, S., Selva Shankar, V., Punitha Peter, M.J. and Palavesam, A., 2009. Processing and characterization of low cost Balistid fish *Sufflamen capistratus* liver oil for edible purpose. Food Chemistry, 115:430-435.
- Iwasaki, M. and Harada, R., 1984. Cholesterol content of fish gonads and livers. Bulletin of Japanese Society of Science and Fisheries, 50(9):1623P.
- **Jacobs, M.B., 1965.** The Chemical analysis of food and food products. 3rd edn., D. Van Nostrand Company, Inc. New Jercy, London, UK. 662P.
- Kayama, M.; Tsuchiya, Y. and Nevenzel, J., 1969. The hydrocarbons of shark liver oils. Bulletin of the Japanese Society of Science and Fisheries, 35(7):653-664.
- Kizevetter, K., 1973. Chemistry and technology of Pacific fish. Israel Programme for Scientific Translation (IPST) Catalogue No. 600818, pp.7-18.
- Lagler, K.F., 1952. Fresh water fishery biology. Dubuque, Iowa, WmC. Brown company, 360P.
- Le Cren, E.D., 1951. The length, weight relationship and seasonal cycle in gonad weight and condition in the perch (*Perca*

fluviatilis). Journal of Animal Ecology, 20(2):201–219.

- Lovern, J.A., 1962. Fish Nutrition, Eds. E. Heen and R. Kreuzer. *In*: Fishing News Ltd., London, UK. 86P.
- Miller, L. and Berger, T., 1985. Bacteria identification by Gas Chromatography of whole cell fatty acids. Gas chromatography application note 228-41. Hewlettpackard, 3000 Hanover, St. Palo Alto, CA 94304 -1181, pp.1–8.
- Murrey, J. and Burt, J.R., 1983. The composition of fish. Torry advisory note, No. 38, Torry Research Station, Ministry of Agriculture, Fisheries & Food, Aberdeen, Scotland, 11P.
- Nair, R.K.G. and Gopakumar, K., 1977. Fatty acid composition of marine fish body fat. Journal of Food Science and Technology, 14(6):268-270.
- Nair, R.K.G. and Gopakumar, K., 1982. Effect of age on the fatty acid composition of Tilapia (*Tilapia mossambica*). Journal of Food Science and Technology, 19:250-254.
- Nair, R.K.G. and Gopakumar, K., 1985.
 Influence of habitat on the fatty acid composition of Tilapia (*Tilapia mossambica*).
 In: Proceedings of the symposium on Harvest and Post-harvest Technology of Fish, Eds.
 K. Ravindran *et al.* Society of Fisheries Technologists, Cochin, India, pp.467-469.
- Narasimhan, A.G., Ramamurti, R. and Raman, R., 1965. A text book of practical physics, 2nd edn.. Paul & Co. Publ., Madras, India, 158P.

- New, M.B., 1987. Feed and feeding of fish and shrimp - A manual on the preparation and presentation of compound feeds for shrimp and fish. Aquaculture Development and Coordination Programe, FAO, Rome, Italy. ADCP/REP/87/26.
- Pillai, S.K., 1963. The fatty acid constituents of the liver of *Odonus niger* (Ruppel) Ph.D. Thesis of the University of Kerala, Trivandrum, India. 352P.
- Raj, P.R., 1993. Aquaculture feed. A handbook on Aquafarming MPEDA Publication, Cochin, India. pp.1-64.
- Rossell, J.B., 1986. Classical analysis of oils and fats. In : Analysis of oils and fats, Eds. R.J. Hamilton and J.B. Rossell. Elsevier Applied Science publisher, London, U.K. pp.1-90.
- Rounsefell, G.A. and Everhart, H., 1959. Fishery Science and its Methods and Applications, John Wiely and Son Inc. New York, USA. 444P.
- Shanmugam, A., 1990. Fundamentals of biochemistry for Medical students. Published by the author, Madras, India. pp.1-147.
- Shenoy, A. S. and Dey, V.K., 1984. Shark and its utility. Seafood Export Journal, 1:5-12.

- Sun, T., Pigott, G.M. and Herwig, R.P., 2002. Lipase assisted concentration of n-3 polyunsaturated fatty acids from vicera of formed Atlantic Salmon (*Salmo salar* L). Journal of Food Science, 67:130-136.
- Sunarya, S., Hole, M. and Anthony Taylor, K.D., 1992. Extraction and composition of Dog fish liver oil, FAO fisheries report, No. 470 supplement. pp.267-275.
- Young, F.V.K., 1986. The chemical and physical properties of crude fish oils for refiners and hydrogenators. International association of fish meal manufacturers. Fish Oil Bulletin. 18:1-19.
- Zar, J.H., 1974. Biostatistical analysis. Prentice–Hall Inc. New Jersey, USA. 620P.
- Zheng, X., Seiliez, I., Hastings, N., Tocher, D.R., Panserat, S., Dickson, C.A., Bergot, P. and Teale, A.J., 2004. Characterisation and comparision of fatty acyl Δ 6 desaturax C DNAs from fresh water and marine teleost fish species. Comparative Biochemistry and Physiology, Part B. 139:269-279.

چکیدہ

كلمات كليدي: FFA ،PUFA ،Odonus niger، سوكسله، بلاى وداير، بخار مستقيم، استخراج خورشيدى

آزمایشگاه بیوتکنولوژي دریایي، مرکز علوم و فنون دریایي، دانشگاه مانومانیام ساندارانار، راجاکا مانگالام، ۶۲۹ ۵۰۲، منطقة ک.ک.، تامیلنادو، هند

ساندارانار، رابخانا مانخانم، ۲۰۱ ۲۰۱۰، منطقة ک.ک.، نامید ' پست الکترونیکی نویسندة مسئول: gimmas@gmail.com