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Tocopherol, Mineral, Heavy Metal, Lipid and Fatty Acid Contents of shark sucker (*Echeneis naucrates*, Linnaeus 1758) Caught by Trawl

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Abstract

The objective of the present study was to investigate some biochemical aspects (tocopherol level, micro and macro minerals, heavy metals, lipid level, and fatty acid compositions) of shark sucker caught by trawl in April in 2011 in the northeastern Mediterranean. Shark sucker has got more than 100 mg kg⁻¹ tocopherol levels. The predominant macro mineral, micro mineral, and heavy metal were P, Fe, and Pb with the values of 1369.45, 4.39, and 0.39 mg/kg, respectively. The levels of the saturated fatty acids (SFA), monounsaturated fatty acids (MUFA), and polyunsaturated fatty acids (PUFA) were calculated to be around 30%, 25%, 40%. Additionally, the average levels of arachidonic acid (ARA, C20:4n6, ranged from 3.24 to 3.51%), eicosapentaenoic acid (EPA, C20:5n3, ranged from 3.28 to 3.93%), and docosahexaenoic acid (DHA, C22:6n3, ranged from 13.86 to 14.07%) in lipid of the shark sucker were found to be very similar that of many bony fish. Based on data, it can be considered that this present study showed that shark sucker showed some similarity to that of the regular consumed fish regarding its fatty acid compositions; however some certain differences were observed.

Keywords: *Echeneis naucrates*, shark sucker, discard fish, fatty acid content, tocopherol, heavy metal

1. Introduction

shark sucker (*Echeneis naucrates*, linnaeus 1758) generally attach itself to large animals, mainly elasmobranchs, including teleost fish, marine turtles, mammals, and some other additional hosts [1] with its oval sucking disk with 21-28 laminae located on top of the head. Shark suckers generally feed mainly on ectoparasites that are scraped from host body by the lower jaw. They also supplement their diet with free-swimming organisms, eggs and larvae and sometimes can be preyed on by dolphins [2].

Shark suckers are not consumed as food item due to its sour and bitter taste and are not a target fish species but a discard one which generally thrown back into the sea. According to fishermen, they are abundant and cause problems to them because they have to clear them from either the target ones or nets which require additional time and extra work for them (Personnel communication, fishermen, 2011).

Having shark suckers these amounts has raised concerns over the biochemical compositions of the fish. If the fish can worth the try in any way to turn it a useful item for any possible usage to get benefit out of something useless for now. There are some studies about discard fish show that some of their biochemical properties could be useful (e.g., liver oil of smoothhound, common guitarfish, common stingray, eagle ray, Lusitanian cownose, and bignose shark from Gulf of Iskenderun (Mediterranean Sea) [5], thornback ray from Mediterranean and Black seas, shark species from the Caribbean and Gulf of California waters [6], *Dasyatis brevis* and *Gymnura marmorata* rays [6].

On the other hand, some scientific studies showed that some whole fish could be useful [3-10] to get benefits and could be used in different part of the industry. If possible to get benefit and answer of this discard's possibility of the usage as raw materials (regarding its fatty acid content) in some industrial branches, e.g., pharmaceutical and cosmetic industries, it is worth for us to investigate the fatty acid content of this discard fish. The aim of this study was to investigate lipid level, fatty acid compositions, total tocopherol content, micro, and macro minerals, heavy metals of shark suckers to get a clue to possibility this discard fish species suitable in any way to fit anywhere.

2. Materials and Methods

Shark suckers were caught from northeastern Mediterranean Sea (Iskenderun Bay, between 36° 02'11"E - 36°35'23"N and 35°46'13"E - 36°23'22"N) Iskenderun Bay/Turkey by trawl in April in 2011. A total of seven samples were chosen out of many for this study. Study area is shown in Fig 1.

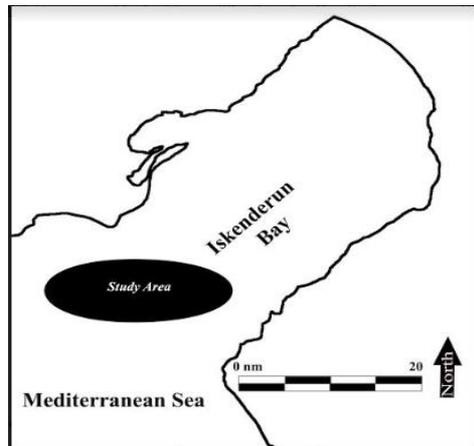


Fig 1: Study area considered in the present study.

Fishes were taken to the laboratory for the analysis. All chemicals used in this presented study were of analytical grade and purchased from Merck (Darmstadt, Germany) and Sigma Aldrich (St. Louis, MO, USA).

2.1. Determination of total tocopherol content in muscle of shark sucker

Determination of total tocopherol content in muscle of shark sucker was carried out according to Turkish Standards 5036 (TS 5036), TSE (Turkish Standards Institute)^[11]. Absorbance was read by using a Hitachi U-1900 model spectrophotometer at 520 nm.

2.2. Extraction and Determination of Macro Mineral, Micro Minerals, and Heavy Metals:

The wet digestion method was performed to digest the muscle of shark sucker. The following steps were performed as described in Ozyilmaz^[12].

The determination and quantification of macro and micro minerals were performed by using an ICP-AES (Inductively Coupled Plasma-Atomic Emission Spectrometry, Varian Model-Liberty series II). Calibration curves for each of the individual elements (macro minerals, micro minerals, and heavy metals) were prepared from ICP Multi element stocks (Merck, Darmstadt, Germany). Wavelengths of the macro and micro minerals determined for this study were depicting in Table 1.

Table 1: Wavelengths of the macro minerals, micro minerals, and heavy metals determined in this study.

Heavy metals, macro and micro minerals	Wavelengths λ (nanometer)
Pb (Lead)	220.353
Cd (Cadmium)	228.802
Fe (Iron)	259.940
Cu (Copper)	324.754
Zn (Zinc)	213.856
P (Phosphor)	213.618
Ca (Calcium)	396.847
Mg (Magnesium)	285.213
Na (Sodium)	588.995

2.3. Crude Lipid Analysis: Determination of crude lipid of shark sucker was carried out by using modified Bligh and Dyer method^[13]. To perform to analyse, a total of 10 g mixed fish muscle was weighed for each analysis. A total of 40 ml chloroform and methanol were used in the experiment. Detailed information is available in Ozyilmaz *et al.*^[4].

2.4. Fatty Acid Methyl Esters (FAMES) Preparation and Chromatographic Conditions:

The conversion and separation of FAMES in lipid of shark sucker were carried out as described in Ozyilmaz and Öksüz^[5]. Fatty acid determination were performed by using a GC-MS (Gas Chromatography-Mass Spectrometry (GC-MS) [a Hewlett Packard GC (model 6890)] coupled with a Hewlett Packard model 5972A HP 6890 system MS detector. The oven programme and the identification of individual fatty acids were performed as described in Ozyilmaz^[3].

2.5. Statistical Analysis: Analysis of descriptive statistics (means and standard deviations) for the results of total tocopherol content, macro minerals (Mg, Na, P, Ca), micro minerals (Fe, Zn, Cu), heavy metals (Cd and Pb), lipid level, and fatty acid profiles of the fish were performed by using SPSS 22.

3. Results and Discussion

3.1. Total tocopherol content of shark sucker

The total tocopherol content of shark sucker is shown in Table 2 and was calculated to be 109.03 ± 5.60 mg kg⁻¹. Ozyilmaz^[3] reported that total tocopherol content of liver oil in male and female thornback ray from both Black sea and Mediterranean Sea were found to be in the range of 42.33 mg kg⁻¹ to 134.45 mg kg⁻¹. Additionally, in another cartilaginous liver oil study, in liver oil's total tocopherol content of smooth-hound, common guitarfish, common stingray, eagle ray, Lusitanian cownose, and bignose shark caught in the Gulf of Iskenderun (Mediterranean Sea) were measured to be in the range of 60.55 mg kg⁻¹ to 114.69 mg kg⁻¹^[5]. The tocopherol levels of the shark sucker in this current study were in accordance in previously reported values of both studies.

Table 2: Total tocopherol content of shark sucker (mg kg⁻¹)

	MM 1	MM 2	MM 3	Mean	SD
Tocopherol content	102.79	113.63	110.67	109.03	5.60

MM: Each mixed muscle was composed of seven individual fish muscle

3.2. Macro Mineral, Micro Minerals, and Heavy Metals of shark sucker

A total of four macro minerals, Mg (Magnesium), Na, (Sodium), Ca (Calcium), and P (Phosphorus) was determined in muscle of shark sucker in this current study and the results are demonstrated in Table 3. Some previously reported scientific studies have shown that muscles of seafood generally have some minerals such as Ca, Mg, Na, P, and K^[15, 16, 17]. While the levels of Mg in muscle of sucker fish this study was found to be very close to that of European hake^[12] it was lower than that of gold band goatfish^[15], striped red mullet^[15], marbled spinefoot^[16], and dusky spinefoot^[16], wild brown trout^[17]. Though Mg toxicity is very unusual, an adequate intake of Mg has been reported to have health beneficial effects on human body system^[18].

Table 3: Macro minerals of shark sucker (mg/kg)

Macro Minerals	MM 1	MM 2	MM 3	Mean	SD
Mg (Magnesium)	207.68	220.24	212.22	213.38	6.36
Na (Sodium)	583.73	586.94	588.87	586.51	2.60
Ca (Calcium)	300.60	321.17	263.78	295.18	29.08
P (Phosphorus)	1295.39	1450.08	1362.87	1369.45	77.55

MM: Each mixed muscle was composed of seven individual fish muscle, SD: Standard Deviation

The levels of Na in muscle of shark sucker fish this study was found to be in the range of some previously reporting [12,15,16,17]. The average amount of Ca in muscle of shark sucker was found to be around 180 mg/kg (Table 3). Parallel to findings in this study many other scientific research have been reported that the amount of Ca in many fish species [15-17]. While the levels of P in muscle of shark sucker was found to be very close to that of European hake [12] it was found to be much lower than that of was found to be lower than that of gold band goatfish [15], striped red mullet [15], marbled spinefoot [16], and dusky spinefoot [16], and wild brown trout [17].

Table 4: Micro minerals and heavy metal of shark sucker (mg/kg)

Micro Minerals	MM 1	MM 2	MM 3	Mean	SD
Zn (Zinc)	3.295	2.913	4.164	3.457	0.641
Cu (Copper)	0.691	0.552	0.619	0.621	0.070
Fe (Iron)	6.000	3.336	3.828	4.388	1.417

MM: Each mixed muscle was composed of seven individual fish muscle

Three micro minerals [Zn (Zinc), Cu (Copper), and Fe (Iron),] was determined in muscle of shark sucker and shown in Table 4. The mean amount of Zn in muscle of shark sucker was found to be around 3.5 mg/kg while that of Cu was 0.6 mg/kg. Both Cu and Zn were found to be lower than some edible fish [12, 15, 17]. Additionally, the level of Fe in muscle of shark sucker was found around 4.4 mg/kg. Fe should be added in the daily diet in a certain amount. A diet includes Fe supplied in it is recommended to be in the range of 10-15 mg/day to meet daily requirement [19] (19 Belitz *et al.*, 2004). According to Camara *et al* [20] Fe helps to prevent some major health problems.

On the other hand, the levels of Cd (Cadmium) and Pb (Lead) in muscle of shark sucker were shown in Table 5. Both Cd and Pb are considered as toxic elements in heavy metals [21]. Higher than certain amount of Cd and Pb intake are known to let many health problems [22] and supposed to be harmful.

Table 5: Heavy metals of shark sucker (mg/kg)

Heavy metals	MM 1	MM 2	MM 3	Mean	SD
Cd (Cadmium)	0.003	0.004	0.013	0.007	0.006
Pb (Lead)	0.125	0.175	0.857	0.386	0.409

MM: Each mixed muscle was composed of seven individual fish muscle

According to FAO and WHO, tolerable daily intake of Cd and Pb by for a 70 kg person is 0.05 and 0.24 mg/day. Findings regarding the mean value of Cd and Pb in muscle of shark sucker in this present study were 0.007 and 0.386 in 100 g.

3.3. The lipid content of shark sucker

The mean lipid content of shark sucker was calculated to be around 13% and results were shown in Table 6. This fish can be categorized as high-fat (>8%) fish regarding Ackman's and Love's reporting [23, 24] about fish lipid level. The average lipid level of shark sucker in this study was found to be higher than many fish in Mediterranean region [12, 14-17].

Table 6: The lipid levels of shark sucker (%)

	MM 1	MM 2	MM 3	Mean	SD
Lipid levels	12.83	13.33	12.86	13.01	0.28

MM: Each mixed muscle was composed of seven individual fish muscle

3.4. Fatty acid profiles (% of total fatty acids) of shark sucker

Fatty acid profiles (% of total fatty acids) of shark sucker used in the present study are given in Table 7. The average total amounts of the saturated fatty acids (SFA) in lipid of the shark sucker were found to be higher than %31. In SFA, palmitic acid (C16:0) was the predominant fatty acid in lipid of shark sucker followed by Stearic acid (C18:0) and acid Myristic (C14:0) with the value of 6.61% and 4.50%, respectively. First three major acid in shark sucker were found to be in given order C16:0> C18:0> C14:0 in this study. Ozogul *et al.*, [25] studied on fatty acid profile of thirty-four fish species caught in the Mediterranean Sea which is the same region in this current study and found out that twenty-seven of them showed the a similar pattern in SFA content having the first three major fatty acids. There are also some scientific studies about relating fatty acid compositions of the fish (caught from the same fishing area (North eastern Mediterranean) In which same situation exist e.g., marbled spinefoot [16] and dusky spinefoot [16], gold band goatfish striped red mullet [15], red mullet (in Autumn, Winter, and Spring) [26], grey mullet [27] and common sole [27], gilthead sea bream and white sea bream (in Spring and Summer) [28], chub mackerel and horse mackerel [29], European hake [12]. The similarity in SFA contents of the some fish previously reporting tells us that the shark sucker in this study have similar SFA contents with many other fish which are often used as food item in the Mediterranean.

The total amounts of the monounsaturated fatty acids (MUFA) in lipid of the shark sucker were found to be lower than those of in lipid of both saturated fatty acids (SFA) and polyunsaturated fatty acids (PUFA). The average level of oleic acid (C18:1n9) in shark sucker was found to be the predominant fatty acid in MUFA with the value of 15.93% and constitute almost more than half of the all MUFA content.

Table 7: Fatty acid profiles (% of total fatty acids) of shark sucker

Fatty Acids	MM 1	MM 2	MM 3	Mean	SD
C12:0	1.51	1.93	1.66	1.70	0.21
C14:0	4.33	4.93	4.24	4.50	0.38
C16:0	18.18	17.71	18.43	18.11	0.37
C17:0	0.46	0.45	0.45	0.45	0.01
C18:0	6.07	7.08	6.68	6.61	0.51
SFA	30.55	32.10	31.46	31.37	1.47
C16:1n9	7.75	6.91	7.42	7.36	0.42
C16:1n7	0.30	0.30	0.38	0.33	0.05
C18:1n9	16.19	15.51	16.08	15.93	0.37
C18:1n7	0.24	0.39	0.32	0.32	0.08
C20:1n9	0.68	0.41	0.58	0.56	0.14
MUFA	25.16	23.52	24.78	24.49	1.05
C16:2n4	0.37	0.37	0.37	0.37	0.00
C16:3n4	0.74	0.71	0.61	0.69	0.07
C22:2	5.48	5.51	4.17	5.05	0.77
C18:2n6	1.39	1.53	1.16	1.36	0.19
C20:3n6	1.09	0.75	0.89	0.91	0.17
C20:4n6	3.25	3.51	3.24	3.33	0.15
C22:4n6	3.56	3.88	3.30	3.58	0.29
C22:5n6	2.61	3.91	2.79	3.10	0.70
n6	11.90	13.58	11.38	12.29	1.51
C18:3n3	0.44	0.82	0.56	0.61	0.19
C20:3n3	0.29	0.28	0.20	0.26	0.05
C20:4n3	0.45	0.59	0.51	0.52	0.07
C20:5n3	3.28	3.93	3.80	3.67	0.34
C22:5n3	2.60	3.06	2.76	2.81	0.23
C22:6n3	13.86	13.92	14.07	13.95	0.11
n3	20.92	22.60	21.90	21.81	1.00
PUFA	39.41	42.77	38.43	40.20	3.34
MUFA+PUFA	64.57	66.29	63.21	64.69	4.38
n6/n3	0.57	0.60	0.52	0.56	1.51
n3/n6	1.76	1.66	1.92	1.77	0.66
DHA/EPA	5.31	3.56	5.04	4.50	0.15

MM: Each mixed muscle of the fish was composed of seven individual fish muscle,

SD: Standard Deviation

Results of studies on fatty acid contents of European hake [12], Spotback herring [30], grey mullet [27], common sole [27] marbled spinefoot [16], dusky spinefoot [16], gold band goatfish [15], striped red mullet [15], *Champsodon nudivittis* [4] red mullet also showed that the level of C18:1n9 is the predominant fatty acid in MUFA.

In PUFA arachidonic acid (C20:4n6, ARA or AA), eicosapentaenoic acid (EPA, C20:5n3) and docosahexaenoic acid (DHA, C22:6n3) were found to be 3.33%, 3.67%, and 13.95% in lipid of shark sucker, respectively. Ozogul et al [31] studied seasonal seven marine fish namely *Sillago sihama*, *Upeneus pori*, *Sparus aurata*, *Saurida undosquamis*, *Epinephelus aeneus*, *Mullus barbatus*, *Solea solea* and found out that the range of C20:4n6, C20:5n3, and C22:6n3 were 2.96–6.52%, 0.21–8.34%, and 14.44–29.99%, respectively. The levels of C20:4n6 and C20:5n3 in this current study were in the range of that previous reporting while the level of C22:6n3 was closer to [31]. There are also some other previous studies about fatty acid of marine fish agrees with both this study [3, 32].

The levels of C22:4n6, C22:5n3, C22:5n6 of shark sucker in this present study were found higher comparing a lot of bonny fish, however cartilaginous fish liver oil have higher levels of these fatty acid. Additionally, the ratio of n6/n3 which is not exceeds 4 that is the maximum level advised by The United Kingdom's health departments [33]. According to previously reported some scientific research, the values higher than 4 (ratio of n6/n3) might negative effect on human health [34].

In this current study, ratio of n6/n3 in shark sucker was calculated to be 0.56 which is much lower than that issued as recommended level. Moreover, there is another recommended value for ratio PUFA/SFA (should be minimum 0.45) [33]. In this present study, ratio of PUFA/SFA in shark sucker was found to be 1.28 which is higher than that.

4. Conclusion

In conclusion, lipid of shark sucker used in this study has got considerable amount of tocopherol. Macro and micro minerals of the fish were found to be in parallel with many fish considered as food item. Some of heavy metals in fish exceed the recommended levels which can be keep in mind, if the fish is going to be used as raw material. Additionally, fish has got substantial quantity of lipid level. Based on data of fatty acid profiles of lipid in this study, shark sucker has got PUFA/SFA and n6/n3 in the range of recommended levels.

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