Research Article

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Estimation of the Fatty Acid Concentration in Muscle Tissue of Carp Fish in Dukan Lake and A Local Pond in Sulaimani, Iraq.

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Abstract

This research aimed to measure the concentration of various fatty acids, in the dorsal muscle tissue of *Cyprinus carpio* fish from Dukan Lake and Qaikand pond using gas-liquid chromatography (GLC) VARIAN gas chromatograph (GC) (CP3800 Walnut Creek model). The presence of highly digestible proteins and fatty acids in fish is critical for a healthy lifestyle since they provide nutritional value. Fatty acids play significant roles in metabolism and function as hormones and signalling molecules. this study was conducted in November and December 2020, 30 fish samples (length: 28 - 38 cm and weight: 965-1640 grams) were collected from Dukan Lake and Qaikand Pond. The average levels of the saturated, monounsaturated, and polyunsaturated fatty acids in the fish from the Dukan were 22.51%, 51.47%, and 21.94%, respectively, and in fish from the Qaikand, they were 25.80%, 44.79%, and 18.93%, respectively. The present study's findings suggest carp could be an excellent component of low-fat, high-polyunsaturated fatty acid diets.

Keywards: Carp Fish (*Cyprinus Carpio*), Dukan Lake, Fatty Acids.

تقدير تركيز الأحماض الدهنية في الأنسجة العضلية لأسماك الكارب في بحيرة دكان وبركة محلية في السليمانية ، العراق

الخلاصة

هدف هذا البحث إلى قياس تركيز الأحماض الدهنية المختلفة ، والمتعددة غير المشبعة ، في الأنسجة العضلية الظهرية لأسماك Cyprinus carpio المأخوذة من بحيرة دوكان وبحيرة قايكاند باستخدام الكروماتوغرافيا الغازية والسائلة (GLC). يعد وجود البروتينات والأحماض الدهنية ذات القابلية العالية للهضم في الأسماك، أمرًا بالغ الأهمية لنمط حياة صحي ،نظرًا لأنها توفر قيمة غذائية. تلعب الأحماض الدهنية دورًا مهمًا في عملية التمثيل الغذائي، وتعمل كهرمونات وجزيئات مرسالية. تعد البروتينات التركيبية لبنات البناء الأساسية للحياة. في هذه الدراسة ، خلال شهري نوفمبر وديسمبر لسنة 2020 ، تم جمع 30 عينة) الطول: 28 - 38 سم والوزن: 965 - 1640 جرام (من الأسماك من بحيرة دوكان وبحيرة كيكاند. كان متوسط مستويات الأحماض الدهنية المشبعة ، والأحادية غير المشبعة ، والمتعددة غير المشبعة في الأسماك المأخوذة من البحيرة: 22.51٪ ، 51.47٪ و 21.94٪ على التوالي ، وفي الأسماك المأخوذة من الأحواض كان: 25.80٪ ، 44.79٪ و 18.93٪ ، على التوالي. تشير نتائج الدراسة الحالية إلى أن الكارب يمكن أن يكون مكونًا ممتازًا للأنظمة الغذائية قليلة الدسم وعالية التشبع بالأحماض الدهنية.

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Introduction

Freshwaters and marine fish are good sources of polyunsaturated fatty acids, which are important for human nutrition. Polyunsaturated fatty acids have been shown to lower cholesterol levels in the blood. (1). Aquaculture appears to have become the fastest-growing animal food-producing area, via an annual growth rate of approximately 8.3 % since 1970, and it produces nearly half of the world food fish supply (52.5 million tonnes in 2008). (68.3 million tonnes involving aquatic plants) (2). Fish is a rich supply of protein, vitamin D, and minerals such as taurine, omega-3 fatty acids, iodine & selenium. According to the structure of such hydro-carbonated chains (PUFA), fatty acids could be either saturated or unsaturated, which could then be either monounsaturated or polyunsaturated (3). The essential fatty acids (EFAs) of the omega-6 series, particularly linoleic acid (LA), C18:2n-6, as well as arachidonic acid (AA), C20:4n-6, as well as the omega-3 series, particularly linolenic acid (LNA), C18:3n-3, eicosapentaenoic acid (EPA), C20:5n-3, & docosahexaenoic acid (DHA), C22:6n-3 is required for maintenance as well as development, so it plays an impact role in the elimination also the treatment of a wide range of illness like hypertension, coronary heart illness, diabetes, cancer, arthritis, as well as other inflammatory as well as autoimmune disorders (4,5,6). As mentioned by (4,5,6), the fatty acid distribution can lower total cholesterol and low-density lipoprotein levels in the blood, but not high-density lipoprotein levels. Overall, lipid and fatty acid composition data are vital for nutritionists, including food investigators in dietary compositions, nutritional labeling, processing, product creation, pharmaceutics, aquaculturists, feed production, fish type raising, and so on. The scientific literature describes the fatty acid composition of various fish types from various geographical areas (7, 8, 9, 10, 11,12). This investigation aims to analyze the fatty acid amount of *C. carpio*, a common fish species found in Dukan Lake and Qaikan Pond that is economically important and is captured and consumed in huge amounts by regional people.

Materials and Methods

Fish sampling

In each Dukan lake and Qaikand pond (which locates in Tasluja, about 22 km far away from west of Sulaimani), 15 live fish samples (from 09:00 AM to 4:00 PM) were collected (a total of 30 carp fish) from local fishermen and directly transported to the laboratory of the University of Sulamani for analysis, and oxygen was provided for the fish during transportation. The length of the fish was between 28 - 38 cm (about 3 years old), and their weight was between 965-1640 grams during November and December.

Water Condition and Quality Changes

Monthly variation of water quality parameters for some parameters was measured by using a professional multi-parameter water testing meter (WQM-243/PHTK-243), the device was calibrated before use, such as the air and water temperature, in both November and December throughout the experiment, ranging between 16 °C to 28 °C and 11 °C to 22.4 °C (09:00 AM and 4:00 PM) respectively. The pH values were 7.11–7.94 for the lake and 8.03 – 8.79 for the pond. Saturated dissolved oxygen levels also ranged from 6.2 – 7.2 in the pond and 7.1 – 8.9 for the Dukan lake and Qaikand pond.

Determination of fatty acid composition by Chromatograph

One gram of white dorsal muscle tissue was added to a container containing five milliliters of methanol to extract fish fat. For one minute, the container was shaken quickly. After adding 10 ml of chloroform, the mixture was violently stirred for 1 minute. The samples were then left in a dark, quiet area for 24hrs. After that, 5 ml of distilled water was added. Also, the mixture is transferred to a decanter and left in a dark area for 2 to 3 hours to separate into 3 phases. The fat that had been dissolved in the solvent phase and was at the bottom of the decanter was poured into a spotless glass tube. Glass containers containing the solvent and the fat were put in a water bath while nitrogen gas was injected into the tubes to separate the solvent from the fat. After a short time, the solvent evaporated in this method, and the fat was eventually collected. The recovered oils were mixed with 5 ml of 2% methanolic NaOH (2 g of NaOH in 100 ml of methanol) to esterify the fat. The tube lid was sealed, given a vigorous shake, and submerged for ten minutes in boiling water. The chemicals above were combined with 2.2 ml of BF3 solution (triborofluoride) and heated for 2–3 minutes. 1 mL of n-hexane was added to the resultant mixture, followed by 1 mL of saturated salt solution (300 g NaCl in 1 liter of distilled water). The resultant solution was forcefully shaken before settling in a stationary location. The top phase was meticulously separated after developing two distinct phases (13).

The fatty acids in the sample were investigated and identified utilizing a VARIAN gas chromatograph (GC) (CP3800 Walnut Creek model). The detector and injection site temperatures were set at 280 & 240 °C, respectively. A 1 l syringe injected 0.2 l of the esterified material into the gas chromatography system. The column's initial temperature is set at 160 ° C; after five minutes, the temperature reaches 180 ° C. (at a rate of 20 degrees per minute). A temperature stayed at this level for 10 minutes before rising to 200 degrees at a pace of 1 degree Celsius per minute. After one minute, the temperature climbed at a pace of 30 ° C. per minute, reaching 230 degrees. Finally, the column was kept at 230 ° C for 5 minutes to remove all chemicals. In this process, helium gas (purity 99.999%) was utilized as a carrier gas, hydrogen gas as a fuel, and nitrogen (purity 99.999%) as an auxiliary gas. Also, dry air was supplied. The retention time of the unidentified specimen's chromatograms was compared to the iav chromatograms achieved in the standard methyl ester fatty acids solution. The results were described as a percentage of the sub-peak region of

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the total. This allowed determine the fatty acids found in fish oil (5,14).

Statistical Analysis

Data obtained were presented as Mean±SD. The differences for all variables studied were tested by unpaired t-test. All statistical tests were two-tailed and a ($P \leq 0.05$) was considered statistically significant it was determined that there was a statistically significant difference among the groups (* P < 0.05, ** P < 0.01, *** P < 0.001). All the graph, calculation, and statistical analyses were performed using GraphPad Prism 9.1.1 (GraphPad Software, San Diego, California, USA).

Results and Discussion

The outcomes of the mean and standard error of the mean of fatty acid in fish specimens of the lake and pond were recorded in table 1, there is only a statically significant difference between the level of the saturated fatty acids also monosaturated fatty acid of lake and pond, and there were no significant differences among other fatty acids (P < 0.05).

This investigation revealed a significant difference in the amounts of saturated as well as monounsaturated fatty acids (MUFA) in the fish samples of Dukan lake and Qaikand pond, with a mean of 22.51 AND 21.95 polyunsaturated fatty acids, respectively. Polyunsaturated fatty acids (PUFA), omega-3 polyunsaturated fatty acid (n3-PUFA), and omega-6 polyunsaturated fatty acids (n6-PUFA) were not significantly different among the lake and pond compared to other fatty acid classes. The data are presented in table 1 as the mean ±SD.

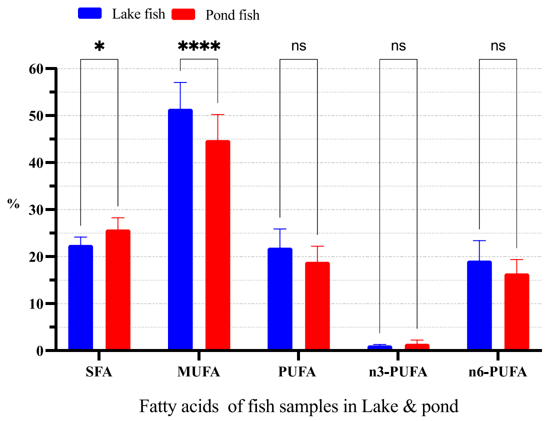

Figure 1: The level of different Fatty acids in different fish from Dukan lake and Qaikand pond.

(ns = nonsignificant, * = P < 0.05, ** = P < 0.01, *** = P < 0.001).

Except for the spring, the fish's essential activity began before the increase in fodder supply, the fat concentration in fish muscle remained practically constant for the whole year. The hepatopancreas, the just section of the carp body that is not usually eaten, has the highest fat content. Saturated fatty acids, such as palmitic acid & stearic acid, do not vary significantly over the year (15). Several factors, including species, geographical region, seasons, feeding patterns, food, and so on, are known to influence the number of lipids, protein & fatty acids in fish. However, nutritional changes are caused not only by inter-species variances but also by intra-species differences due to seasonality as well as maturity level. (16, 17, 18, 19).

Table 1: Mean concentration of different types of fatty acids (mg) in both Dukan lake and Qaikand pond fish samples.

Fatty acids (N=15)	Lake fish	Pond fish	P value
	Mean ± SD	Mean ± SD	
SFA	25.42± 0.41	25.81± 0.62	0.01 **
MUFA - monounsaturate fatty acid -double bond	51.48± 0.48	44.80± 0.38	0.0001 ***
PUFA - polyunsaturated acid	21.95± 0.99	18.93± 0.93	0.9936 ns
n3-PUFA	1.171± 0.038	1.51± 0.18	0.8875 ns
n6-PUFA	19.20± 1.05	16.47± 0.73	0.3630 ns

(ns = nonsignificant, ** = P < 0.01, *** = P < 0.001).

Abu mullet *(Liza abu)* was one of the freshwater fish types examined in the Tigris River, Turkey, for their fatty acid compositions (20). The researchers discovered that this type SFA, MUFA, & PUFA levels were 48.94%, 41.34%, as well as 9.75%, respectively (9). The fatty acid profile and the proximate component of Pacific mullet (Mugil soiuy) captured in the Black Sea were examined. The authors discovered that the total Saturated fatty acids (SFA), Monounsaturated fatty acids (MUFA), and polyunsaturated fatty acids (PUFA) levels in muscle specimens were 29.59, 29.26, and 18.06%, accordingly. The biological activity of fish can be disturbed by increases in heavy metals, as can nucleic acid and ribosome activity. The latter rise can result in declines in fatty acid production, such as the fish's muscle content. (21) discovered that heavy metals have an inhibitory impact on protein and fatty acid biosynthesis with their impact on RNA and ribosomal functionality. The availability of fodder and fish activity affects the compotation of specific fatty acids in various carp tissues throughout the year (22). Table 2 shows the range of saturated and unsaturated fatty acids in Dukan lake and Qaikand pond fish.

Table 2. Range of composition of fatty acids contained in *C. carpio* oil in Dukan lake and Qaikand pond.

Types of Fatty Acids	Name of Fatty Acids	Range (%) Lake fish	Pond fish
Saturated fatty acids	Capric acid methyl ester (C10:0)	0.09 - 0.03	0.07 – 0.02
	Lauric acid methyl ester (C12:0)	0.77 - 0.019	0.68 – 0.015
	Myristic acid methyl ester (C14:0)	1.05 - 0.66	0.94 – 0.65
	Palmitic acid methyl ester (C16:0)	18.94 - 13.54	18.48 – 12.57
	Stearic acid methyl ester (C18:0)	5.59 - 1.65	5.62 – 1.46
	Arachidic acid methyl ester (C20:0)	2.64 - 1.03	2.76 – 1.06
Unsaturated fatty acids	Palmitoleic acid methyl ester (C16:1)	6.55 - 3.42	5.31 – 2.78
	Oleic acid methyl ester (C18:1n9c)	54.26 - 40.13	51.72 – 38.73
	Linoleic acid methyl ester (C18:2n6c)	26.11 - 13 0.5 - 0.02	25.71 – 13.67 0.63 – 0.01
	y-Linolenic acid methyl ester (C18:3n6)	1.9 - 1.02	1.3 – 0.9
	alpha-Linolenic acid methyl ester (C18:3n3)	0.79 - 0.01	0.58 – 0.16
	cis-8,11,14 Eicosatrienoic acid, methyl ester (C20:3n6)	0.59 – 0.04	0.44 – 0.03
	Arachidonic acid methyl ester (C20:4n6)		

Fat tissue contains a surprising amount of oleic acid, eicosapentaenoic acid, and docosahexaenoic acid, and their quantities remain rather constant throughout the year (20). Even though the use of carp fatty tissue for human utilization is uncertain, a component of carp viscera that is only frequently utilized as human food could serve as researcher very beneficial resource for these fatty acids. (23,24). (6) found that around 60% of total SFA was analyzed for three age groups, similar to our findings. (12) discovered that the SFA for *V. vimba tenelle* & *C. capoeta*, which live in Lake Sugla, was 23.45% and 21.36%, respectively. (15) investigated the fatty acid amounts of various carp body parts. The authors reported significant seasonal differences in polyunsaturated fatty acid quantities and seasonal differences among respective parts (25,26). Unexpectedly, polyunsaturated fatty acid concentrations were discovered to change. A variability in saturated fatty acids was significantly lower, as well as various research on the impacts of freshwater fish place, diet, age, size, & ambient temperature on these materials were also evaluated. Nevertheless, no data on seasonal variation in the fat content of individual tissues were obtainable. In edible fat, the ratio of unsaturated to saturated fatty acids is essential, and a value greater than 0.35 is usually thought to be beneficial. (27,28,29).

Conclusions

Fish has traditionally been utilized to produce omega-3-enriched oil. Our research indicates that carp can be a great addition to diets high in polyunsaturated fatty acids and low in fat.

Conflict of Interests

The authors declare that the publication of this paper does not involve any conflicts of interest.

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