Survey of some Chemical Compositions and Fatty Acids in Cultured Common Carp (*Cyprinus carpio*) and Grass Carp (*Ctenopharyngodon idella*), Noshahr, Iran

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Abstract: Ecological studies have found a negative correlation between the risk of developing heart diseases and fish consumption because of their long chain omega-3 fatty acids. In this study to determine of some chemical compounds (proteins, lipids, moisture and ash) and fatty acids were determined for two cultured species, common Carp (*Cyprinus carpio*) and Grass carp (*Ctenopharyngodon idella*). Results of this study showed amount of saturated fatty acids (SFA) in common carp and Grass carp were 35.21 ± 2.19% and 27.18 ± 2.63% respectively, SFA in common carp was higher compared to grass carp (P<0.05). Levels of polyunsaturated fatty acids (PUFA) in the common carp and grass carp were 23.5±2.59 and 31.55±1.38 %, respectively and there was significant difference between the two species (P<0.05). Mono unsaturated fatty acids (MUFA) in the common carp and grass carp were 31.41±2.06 and 35.12±1.78 % respectively. There was no significant difference in MUFA between two species (P>0.05). This study showed in grass carp PUFA was higher than SFA, while in common Carp SFA was higher than PUFA (P<0.05). There were significant differences in protein, lipid and moisture in both studied species (P<0.05) but there was no significant difference in the amount of ash in the two fish species (P>0.05).

Key words: Chemical Composition, Fatty Acid, Common Carp (*Cyprinus Carpio*), Grass Carp (*Ctenopharyngodon Idella*)

INTRODUCTION

Fish and other sea food consumption increased in recent years and demand for sea products is growing with the increasing of population, income and also priority of sea products toward other food. Fishes have essential unsaturated fatty acids, protein with high biological value, minerals and vitamins that make them distinguished from other creatures [1]. Fish is a major source of food for mankind, providing a significant amount of the animal protein diet in many countries. As compared to red meat, fish flesh is easily digestible because it contains long muscle fibers. Moreover, the consumption of fish has been linked to health benefits, such as reduced risk of coronary heart diseases, which are largely attributable to the polyunsaturated fatty acids (PUFA) in fish oils [2].

An increase in seafood consumption, especially fish, has been suggested as an alternative to elevate the T3 PUFA level in the western diet. Another alternative that is being well accepted by the population is the use of food supplements based on sea oils, with a high T3 PUFA concentration [3]. Data about many clinical and observational studies concerning the role of fatty acids in human health have revealed that saturated and trans-fatty acids increase cardiovascular (CVD) risk, while long chain n-3 PUFA (EPA and DHA), which appear to play amultifactorial role in CVD risk, the mechanisms of which have yet to be fully elucidated, have been associated with a reduced risk [4]. Fish body like other animals consists of water, protein, non-protein nitrogen compounds, fat, minerals, vitamins and low amount of hydrocarbon. Amount of these ingredients and their changes in fish

body can be used as an indicator for physiological conditions. Chemical composition in fish muscle is different and is depend on the (type) species, ration and diet composition, farming operation and environmental conditions [3], size, age, reproductive cycle, salinity, temperature, geographical location and fishing season [5] and genetic factors in muscle [5, 6].

Due to fishing restriction in water resources, aquaculture is the only way to sea food supply [7]. Cyprinus carpio and Ctenopharyngodon idella are two species of fishes that could be seen in most environments and due to rapid growth, easiness of breeding and high food efficiency; they are cultured in almost all places in the world [8]. These two species are counted as the important farming species in Iran, as now are included about 50% of fish in warm freshwater fish ponds. Accordingly, considering abundance of producing these two species, surveying their body composition is very important and this study has been done for investigating the level of fatty acids and some chemicals composition (protein, lipid, moisture and ash) of Cyprinus carpio and Ctenopharyngodon idella muscle tissues.

MATERIALS AND METHODS

Fish Samples: Fishes samples including *Cyprinus carpio* and *Ctenopharyngodon idella* were from the fish ponds in Noshahr on July 2009. They were kept in iced boxes and transported to the laboratory where they were washed with cold water, weighed and measured.

Analyses of Protein: Total protein was determined by using Kjeldahl method [9]. A conversion factor of 6.25 was used to convert total nitrogen to crude protein for all varieties of fish..

Moisture and Ash Analyses: The muscle was homogenized by food processor (Braun Combimax 600) and moisture content of 5 g of homogenized sample was determined by drying the sample in oven at 105° C until a constant mass was obtained [3]. Ash was determined by using the basic AOAC method [10] by heating the samples in the furnace at 550°C for 8-12 h. Each sample analyzed three times.

Lipid Analyses: Total lipids of each one of the above mentioned fish species were extracted (separately) according to the Bligh and Dyer method [11]. After phase equilibration, the lower Chloroform layer (TL) was

removed and dried in a rotary vacuum evaporator at 32°C. The extracted lipids were weighed in order to determine the TL and then redissolved in chloroform/methanol (9:1, v/v) and finally stored at 0°C until used. Additionally, in order for that to be confirmed, aliquots were evaporated in preweighed vials to constant weight to determine the lipid content. To prevent oxidation t-butyl-hydroquinone was added to all samples during preparation.

Fatty Acid Methyl Esters and Gas Chromatographic Analyses: Separation of the methyl esters was by gas chromatography, using a VARIAN Mod. 3300 gas chromatograph equipped with a flame ionization detector and a fused silica DB-WAX capillary column (30 m-0.25 mm i.d.) (J and W Scientific, California, USA). The operation parameters were as follows: detector temperature, 280°C; injection port temperature, 250°C; column temperature, 170°C for 16 min, programmed to increase at 2°C/min up to 210°C with a final holding time of 25 min; carrier gas, hydrogen at 0.8 ml/min, linear velocity of 38 cm/s, with an oxygen filter coupled to the line; nitrogen was used as the makeup gas at 30 ml/min, hydrogen and synthetic air at 30 ml/min and 300 ml/min for the detector; split injection at 1:100 ratio. All the stages, from the transesterification to the final injection, were accomplished under nitrogen. Retention times and peak area percentages were automatically computed by a Varian 4290 integrator. Fatty acid methyl esters used as GC standard were: lauric acid M-E, L7272, cis-5,8,11,14,17eicosapentaenoic acid M-E, E2012 and cis-4,7,10,13,16,19docosahexaenoic acid M-E, D2659 (purity =98%) and they were purchased from Sigma Chemical Co; Matreya Bacterial Acid Methyl Esters CPTM Mix, Catalog No: 1114; SupelcoTM 37 Component FAME Mix, Catalog No: 47885-U. All solvents used for sample preparation were of analytical grade and the solvents used for GC analysis were of HPLC grade from Merck (Darmstadt, Germany). All water used in this work was re-distilled. All reagents used were of analytical grade from Mallinckrodt Chemical Works (St. Louis, MO) and from Sigma Chemical Co (Sigma-Aldrich Company St. Louis, MO).

Statistical Analysis: At first we checked normality distribution of our data by using Kolmogrov-Smirnoff Test. Significant differences between chemicals characteristic between two fishes was determined by using T test about 5% probabilities. All statistical analysis was done by SPSS (ver. 19.5) software.

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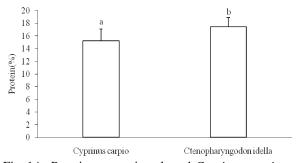
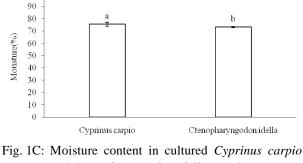


Fig. 1A: Protein content in cultured Cyprinus carpio and Ctenopharyngodon idella muscles.



and Ctenopharyngodon idella muscles

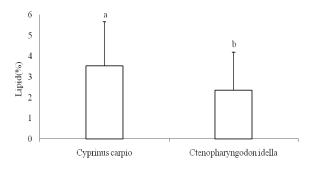


Fig. 1B: Lipid content in cultured Cyprinus carpio and Ctenopharyngodon idella muscles

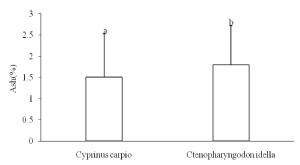


Fig. 1D: Ash content in cultured Cyprinus carpio and Ctenopharyngodon idella muscles

Table 1: comparison of amount of fatty acids in cultured Cyprinus carpio and Ctenopharyngodon. idella muscles (N=10)

spices	PUFA/SFA	T-3/T-6	EPA+DHA	PUFA	MUFA	SFA
Common carp	0/71±0/04a	0/82±0/18 ^a	7/18±0/19 ^a	23/5±0/2a	37/41±1/2 ^a	35/21±3/32a
Grass carp	$1/04\pm06^{b}$	0/52±0/35a	$6/68\pm2/14^a$	$31/55\pm3/12^{b}$	35/12±0/9a	$27/18\pm0/54^{b}$

^{*}Our data is expressed based on average ± standard deviation. Different letters in each column show the significant difference (P<0.05).

RESULTS

Fish biometry showed the average length and weight were 37±1cm and 860±72 g in Cyprinus carpio and 50±1 cm and 900±50 g in Ctenopharyngodon idella respectively. Results of chemical compounds (protein, lipid, ash and moisture) show in muscle tissue of both Cyprinus carpio and Ctenopharyngodon idella in figure 1. Amount of protein in Ctenopharyngodon idella $(17.41 \pm 1.47 \%)$ was more than Cyprinus carpio (15.2 ± 1.9) %) (P<0.05). Lipid in Ctenopharyngodon idella was 2.35± 1.83 % and in *Cyprinus carpio* was 3.53 ± 2.12 percent % (P<0.05). Amount of ash in grass carp was 1.8 ± 0.92 % and in common carp 1.5±1.02 %, there was no significant difference in muscle tissue of both fishes. Also results showed that there was a significant difference in muscle tissue of both fishes and in the level of moisture, as the amount of moisture in Ctenopharyngodon idella (73.1±10.38 %) was less than *Cyprinus carpio* (75.48±1.58 %) (P<0.05) (Fig.1).

Amount of saturated fatty acids (SFA) and Mono unsaturated fatty acids (MUFA) in Cyprinus carpio muscles was orderly 35.21±2.19% and 31.24±2.06%. Also the amount of poly unsaturated fatty acids (PUFA) in Cyprinus carpio was 23.5±2.59 %. In Ctenopharyngodon idella the level of saturated fatty acids (SFA) and Mono unsaturated fatty acids (MUFA) was orderly 27.18±2.63 % and 35.12±1.78%. And about poly unsaturated fatty acids (PUFA) in Ctenopharyngodon idella was 31.55±1.38 %. Saturated fatty acids in *Cyprinus carpio* muscles is more than Ctenopharyngodon idella muscles (P<0.05) (Fig. 1). The results of statistical analysis showed the total amount of Mono unsaturated fatty acids (MUFA) in Cyprinus carpio does not have any significant difference with its amount in Ctenopharyngodon idella (P<0.05). Also the amount of poly unsaturated fatty acids (PUFA) in Cyprinus carpio was less than Ctenopharyngodon idella (P<0.05). In Ctenopharyngodon idella poly unsaturated fatty acids was more than saturated fatty acids while in Ctenopharyngodon idella polysaturated fatty acids was more than poly unsaturated fatty acids (P<0.05). Amount of T-3/T-6 in *Ctenopharyngodon idella* was 0.52±0.03 and in *Cyprinus carpio* was 0.82±0.13, there was no significant difference between two species (p<0.05). Also PUFA/SFA ratio in *Ctenopharyngodon idella* (1.04±0.10) was more than *Cyprinus carpio* (0.71±0.03) (P<0.05) (Table, 1).

DISCUSSION

Investigating chemicals characteristic of freshwater fishes is very important, because useful information for experts related to food resources having low fat, high protein and being easily accessible. Results of this study shown level of moisture in Cyprinus carpio was 5.48±1.57 % and in Ctenopharyngodon idella was 73.11±0.38 % that was according to the findings of others in this background [12, 13]. Fish is considered to be not only food with good source of quality protein but also food with healthy components. Fish lipid Demand continues to increase in the world. Fish lipid that contributes to the nutritional needs is currently being extracted from muscle or liver of cod, mackerel, herring and sardine [14]. Fish can be grouped into four categories according to their fat content: lean fish ($\langle 2\% \rangle$), low fat (2-4%), medium fat (4-8%) and high fat (N8%; [15]). In this study, amount of fat in Cyprinus carpio and Ctenopharyngodon idella was orderly 3.53 ± 2.12 and 2.53 ± 1.83 % that was according other research results [12, 13]. The investigated fishes in this study, was of lean fishes group, which was different by Jabben and Chaudhry's findings [16] This different May be due sampling location, sampling season, nutrition condition of fish, environmental condition, size and age and etc.

The protein content is important when considering quality and texture of the fish muscle. Fish muscles that contain small amounts of protein tend to lose much water upon cooking, which ruins the texture of the meat. The amount of protein in *Ctenopharyngodon idella* was 17.41 ±1.47% and in *Cyprinus carpio* was 15.2±1.9 % which was according to Khorramgah *et al.* [17] findings. Also the amount of ash in this survey was more than Khorramgah *et al.* [17].

So many surveys showed that chemical compounds in fish muscles in different species or even in one species has difference depends on gender, age, environmental condition and season. According to results of this study, chemical compounds of two cultured fishes, *Ctenopharyngodon idella* and *Cyprinus carpio* considering being in the same environmental condition

and having differences with each other and with other done study in this background.

Cultured *Cyprinus carpio* have more saturated fatty acids (SFA) than cultured *Ctenopharyngodon idella*, while the amount of poly unsaturated fatty acids in *Cyprinus carpio* is less than its amount in *Ctenopharyngodon idella*. This difference could be related to the type of feeding and culture system. *Cyprinus carpio* that cultured as a semi-extensive in the earth ponds mainly depends on natural foods and benthic communities in the ponds, while *Ctenopharyngodon idella* has herbivore diet and in addition to feeding from reed around the earth pond, fed manually by forage and hay [17].

Transferring of PUFA and particularly EPA and DHA in fish food chain shows which normally plankton feeders has the highest PUFA and benthic carnivores feeds from invertebrates, has the lowest amount of PUFA[18]. Nutrition recommendations for daily intakes of n-3 PUFAs (DHA + EPA), ranging from 1.6 to 0.5 g for healthy adults, infants, pregnant and lactating women have been published by several international scientific authorities [British Nutrition Foundation [19]; International Society for the Study of Fatty Acids and Lipids [20]. Considering type of food diet in Cyprinus carpio toward herbivore Ctenopharyngodon idella and that the benthic invertebrates form much of the foods of Cyprinus carpio, in this survey also the amount of PUFA in Cyprinus carpio and Ctenopharyngodon idella is less than amount of MUFA. The lowest amount suggested for PUFA/SFA ratio, is 0.45 [21] which in this study for Cyprinus carpio calculated 0.71 and for Ctenopharyngodon idella calculated 1.04.

N-3/n-6 ratio is the appropriate indicator for relative comparison of nutritional value of fish fat [8]. Generally amount of n-6 among freshwater fishes is more than n-3 [22]. The n-3/n-6 ratio is an important index of the fatty acid role in human health. As the appropriate balance for n-3/n-6 ratio, as recommended by Simopoulos [23], varies from 1.1 to 1.4 depending on the disease under consideration. The n-3/n-6 ratio differences may be explained by the large variability of the oil level in the fish muscle, which depends on the species, period of the year, age, size, reproduction period, the specific species as well as the fatty acid composition of the diet [24].

Also in present study like cultured *Cyprinus carpio* or wild one Khorramgah *et al.* [16] and cultured channel catfish Tokur *et al.* [25] and higher amount of n6 than n-3 was observed in cultured *Cyprinus carpio* and *Ctenopharyngodon idella.* Lots of surveys showed that

the amount of saturated fatty acids in freshwater fishes is more than unsaturated fatty acids. In this survey the amount of saturated fatty acids in Cyprinus carpio was unsaturated fatty more than acids but Ctenopharyngodon idella unsaturated fatty acids was more than saturated fatty acids that it likely because of the type of feeding of Ctenopharyngodon idella that feeds from plants. It should however be noted that the net content of n-3 and n-6 is low because of the low lipid content of these fish species and only regular consumption of such fish species could contribute to increase the amount of n-3 and n-6 fatty acids in the diet.

Traditionally, fish with high fat content, like rainbow trout, has been considered to be nutritional important species since they have a relatively high content of n-3 FAs. However, it has been demonstrated that there is an inverse relationship between amount of n-3 FAs and total fat content [16]. This implies that it is important from the nutritional point of view to place attention upon getting species with high proportions of n-3 FAs instead of only focusing on the fat content. Previous studies have also demonstrated a beneficial role of lean fish, as well as fatty fish consumption, in the prevention of cardiovascular diseases [25].

Data derived from various long-term epidemiological studies, which have investigated fish intake in relation to a reduced mortality from cardiovascular disease, suggest a combined DHA/EPA intake of 500 mg/day for normal healthy adults. (International Society for the Study of Fatty Acids and Lipids,) [20] This intake is both safe and achievable by diet alone, even for pregnant and lactating Women [20]. Data from the present study indicate that the consumption of 500 g of cultured *Cyprinus carpio* muscle tissue covers the amount mentioned above, while as *Ctenopharyngodon idella* well as respectively.

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