A STUDY OF SOME OF HISTO-CHEMICAL FEATURES FOR RED MUSCLES SKELETAL IN SOME LOCAL IRAQI FISHES ; BUNNI FISH, *Mesopotamichthyes sharpeyi* (GUNTHER, 1874) AND HIMRI FISH, *Carabarbus luteus* (HECKEL, (1843)

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Abstract

The current study was conducted to determine differences in some of the chemohistological features of red skeletal muscles in two different regions in the body of two types of teleosts. These species were Bunni fish, *Mesopotamichthyes sharpeyi* (Gunther, 1874) and Himri fish, *Carabarbus luteus* (Heckel, 1843) collected from Qurna market , north of Basrah province, Iraq , between November - December 2018. The results revealed differences in the proportions and diameters of the red muscle fibers in R1 and R2 regions which ranged between 3.68 - 4.42% in *M. sharpeyi* , while ranged 2.44 - 2.96 % in *C.luteus* , but the statistical results showed no significant differences (p > 0.05) . The diameters averages of red muscle fibers ranged from 26.45 to 23.45 micron and 17.13 to 14.42 microns in R1and R2 regions in *M.sharpeyi* and *C.luteus* , respectively . But the statistical results did not indicate significant differences (p > 0.05) in the two regions (R1 and R2) in the studied species. Results indicated a difference in the studied fishes in protein, lipid, moisture and ash contents in R1 and R2 regions. *M.sharpeyi* fish had values ranging from 19.08 - 18.64% protein, 5.0 - 5.52 % lipid, 74.15 - 73.82 % moisture and 1.64 - 1.54 % ash while the values ranged between 18.16 - 17.8 % protein, 2.35 - 2.88 % lipid, 77.86 - 77.58 % moisture and 1.31 - 1.28 % ash content in the *C*. *luteus* fish. The results pointed that the *M.sharpeyi* fish had higher energy values compared to *C.luteus* fish, which recorded 122.58 kcal/g and 95.454 kcal/g respectively, so the statistical results showed significant differences (P < 0.05) between studied species in lipid content in R1 and R2. So the *M.sharpeyi* fish placed in a medium-lipid fishes whereas the *C.luteus* fish were in a low-lipid fishes.

Key words: *Mesopotamichthyes sharpeyi*, *Carabarbus luteus*, Red muscle, Chemical content.

Introduction

Local Iraqi fish are a consumption source in daily human life because they contain high nutritional value as well as their health benefits which reflect the amount of the unsaturated fatty acids in their muscles (Mansour, 2005). The main role for fish muscles are movement, also they give important support to the skeleton. The difference between striated skeletal fish muscles and other vertebrates is firstly the separation of muscle fibers types into discrete layers in fish where the constituting 90-95% of the mass of muscle tissue in most fish species. Secondly is the continued growth of muscle fibers in fish during life stages which leads to the addition of more muscle fibers and increased fibers size (Kiessling *et al.* 2006).

In majority of fish, there are three types of muscle fibers are found; red fibers form a thin lateral superficial layer under the skin. White fibers make up the larger mass of

the myotome (Mansour, 2005; Rabah, 2005) whereas the intermediary or pink muscle are positioned between the red and white muscle (Martinez et al . 2000). Red muscle fibers are the smallest in diameter, while white muscle fibers are dominant in number and largest in diameter beside to the capable of bearing a heavier load (Spierts, 2000). Intermediary (or pink) fibers are larger in diameter than red fiber but shorter than the white fibers (Martinez et al . 2000). Principal chemical composition of fish is (16-21% protein, 0.2 - 25% fat, 66 - 81% moisture (water), 1.2 -1.5 % mineral and 0 - 0.5% carbohydrate (Love, 1970). Generally, In most fish the chemical composition for muscles varies not only between species, but also between individuals depending on age, feed, size, sexual cycle, stage of maturity, environment, season and muscle location in fish body (Huss, 1995; Kozlova, 1997; Khitouni et al. 2010; Mansour, 2018). The current fish are considered from the economic fish in Iraq, namely bunni fish, Mesopotamichthyes sharpeyi (Gunther, 1874) and Himri fish, Carabarbus luteus (Heckel, 1843), both species are belong to the Cyprinidea, so the present study aims at clarifying the differences in muscle fiber ratios and its diameters, as well as differences in the values of the chemical composition of the muscles between the studied body regions in the same species and between the differenced fish species, thus can be determination the nutritional value of the studied fish .

Materials and Methods

Sampling

Forty local important fish were chosen, also they weight ranged between 150-250g for both studied species and collected from Qurna market, Northern of Basrah city,Iraq. Samples were collected between November and December 2018. They were transported to the laboratory to perform the tests associated with account of the proportions and diameters the red skeletal muscle fibers as well as the chemical composition of the studied fish muscles .

Account the red muscle proportions

To accounting the proportion of red muscle, the cross sections were taken from the body of the studied fishes; R1 represents the trunk region while R2 represents the caudal peduncle, then determined red muscle with drawing on transparent paper, then weighed with a sensitive balance and calculated the proportion of red muscles according to Broughton *et al*., (1981).

Measurement the diameters of red muscle fibers

To measure the diameters of the red muscle fibers in R1, R2 in the studied fish. Cross-sections of the red fibers were taken with 8 micron thickness. Fifty muscle fibers were measured randomly using the Ocular micrometer and the Calibration according to Broughton *et al*., (1981).

Estimation of chemical composition

The contents of the chemical composition of the muscles were assessed in the studied fish, which included protein, fat, moisture and ash according to AOAC (Association Of Analytical Chemists Procedures (2005). The energetic (Calorific) value of the protein and lipid contents to the red muscles were calculated by Jabeen and Chaudhry (2011).

Statistical Analysis

Statistical analyses were conducted using the IBM SPSS Statistical 25 software. The data of proportions, diameters and chemical composition obtained from the red muscle fibers of studied fish were analyzed using one-way analysis of variance (ANOVA) followed by Tukay's multiple comparison test. Differences were considered to be significant when P value ≤ 0.05 .

Results

Tables (1 and 2) showed differences in the averages of proportions of the red muscle fibers in the body regions (R1and R2) in the studied species, which ranged between 1.8 - 5.6 % and 2.4 - 6.8 % in *M.sharpeyi* while were 1.4 - 3.7 % and 1.8 - 4.4 % in *C.lateus*. Despite these variations in current values, the statistical results showed no significant differences (P > 0.05) between the studied fish (Table 6). Also, The results revealed an increase in the total average for proportions red muscle fibers in R2 compared with R1 where were 4.42 % in R2 while 3.68 % in R1 in *M.sharpeyi* fish (Table 1) whereas recorded 2.96 % , 2.44 % in R2 and R1 respectively in *C.lateuc* fish (Table 2).

The current results pointed an approximation the averages of diameters of the red muscle fibers in (R1and R2) for both studied species which ranging from 17.13 to 37.70 micron and ranged 15.76 - 34.27 micron in *M.sharpeyi* (Table 1) while ranged between 10.28 - 23.99 micron in R1 and 8.22 to 20.56 micron in R2 in *C.lateus* (Table 2) . As the results showed that the *M.sharpeyi* fish had a total averages for diameters of red muscle fibers compared to the *C.lateus* fish in (R1and R2), which was between 26.45 - 23.22 microns in *M.sharpeyi* (Table 1), while was17.13 and 14.42 micron in *C.lateus* (Table 2). However , the statistical results did not record

significant differences (P > 0.05) between the studied fish (Table 6). Also , the results demonstrated a decrease in the averages of diameters the red muscle fibers in the posterior region of the body (R2) in both studied species.

	R1	R2	R1	R2
Weight group				
	Red fiber	Red fiber	Diameter red	Diameter red
(g)	proportion %	proportion %	fiber (µ)	fiber (µ)
	1.8 ± 0.06	2.4 ± 0.08	17.13 ± 1.65	15.76 ± 1.87
	2.6 ± 0.10	2.9 ± 0.12	19.18 ± 1.46	17.98 ± 1.72
150 – 225	3.6 ± 0.12	4.6 ± 0.22	27.42 ± 1.72	23.99 ± 1.65
	4.8 ± 0.18	5.4 ± 0.16	30.84 ± 1.80	27.12 ± 2.42
	5.8 ± 0.12	6.8 ± 0.18	37.70 ± 2.15	34.27 ± 1.45
Mean	3.68	4.42	26.45	23.82

Table (1): Proportions and diameters red muscle fibers in (R1and R2) regions in *M.sharpeyi* . $\pm \dots SE$.

Table (2) : Proportions and diameters red muscle fibers in (R 1 and R 2) in C. luteus . $\pm \dots SE$.

	R1	R2	R1	R2
Weight group				
(g)	Red fiber	Red fiber	Diameter red fiber	Diameter red fiber
	proportion %	proportion %	(μ)	(μ)
	1.4 ± 0.05	2.4 ± 0.06	10.28 ± 1.78	8.22 ± 2.15
	1.8 ± 0.08	2.9 ± 0.08	13.71 ± 1.65	11.24 ± 1.85
150 - 225	2.2 ± 0.14	4.6 ± 0.06	17.13 ± 2.42	14.56 ± 1.65
	3.0 ± 0.16	5.4 ± 0.12	20.56 ± 1.84	18.50 ± 1.87
	3.7 ± 0.08	6.8 ± 0.16	23.99 ± 2.14	20.56 ± 1.82
Mean	2.44	2.96	17.13	14.42

The results founded that there were significant differences (P < 0.05) between the fish studied for the values of protein content in the R1 region, while they were not significant (P>0.05) in R2 region (Table 6). The observations showed that the protein content was the second most valuable chemical component after the moisture content for chemical analysis of muscles, where ranged from 18.2 to19.8% and 17.4 -18.8% in R1 region for *M.sharpyei* and *C.luteus*, respectively(Tables 3 and 4), while the values in R2 region ranged from 17.8 to 19.2 %. for *M.sharpyi* fish(Table 3) whereas ranged from 17.0 - 18.6 % in *C.luteus* fish (Table 4). The values of the lipid content in R1 region ranged from 4.10 to 6.20 % whereas the values in R2 region ranged from 4.20 to 3.4 % in R1 and R2 regions in *C.luteus* , respectively (Table 4). As a result of these variations in lipid content values in the studied fish body regions, the statistical results indicated significant differences (P < 0.05)between the studied species (Table 6). As the current results pointed that the total lipid content in *M.sharpeyi* was 5.26% and 2.615% in the *C.luteus* (Table 5).

	Chemical composition in R1			Chemical composition in R2				
Weight group (g)	Protein %	Lipid %	Moisture %	Ash %	Protein %	Lipid %	Moisture %	Ash %
	18.20	4.10	76.20	1.30	17.8	4.50	76.0	1.22
	±	±	±	±	±	±	±	±
	0.10	0.08	0.22	0.04	0.18	0.05	0.14	0.04
	18.6	4.50	75.14	1.54	18.2	4.90	74.8	1.42
150	±	±	±	±	±	<u>+</u>	<u>+</u>	±
-	0.16	0.06	0.26	0.08	0.14	0.08	0.18	0.05
225	19.2	4.80	74.20	1.68	18.6	5.60	73.9	1.56
	<u>±</u>	<u>+</u>	<u>±</u>	±	±	±	<u>±</u>	±
	0.24	0.08	0.18	0.06	0.16	0.06	0.16	0.04
	19.6	5.40	73.12	1.82	18.9	5.80	72.8	1.72
	±	±	±	±	±	±	±	±
	0.15	0.08	0.22	0.08	0.15	0.06	0.12	0.02
	19.8	6.20	72.10	1.84	19.2	6.80	71.6	1.76
	±	±	±	±	±	±	±	±
	0.12	0.06	0.14	0.06	0.12	0.08	0.15	0.04
Mean	19.08	5.00	74.15	1.64	18.54	5.52	73.82	1.54

Table (3) : chemical composition of muscles (R1 and R2) in *M*. sharpeyi $\pm \dots$ SE

Table (4): chemical composition of muscles (R1 and R2) in C. $luteus \pm \dots$ SE

	Chemical composition in R1			Chemical composition in R2				
Weight group (g)	Protein %	Lipid %	Moisture %	Ash %	Protein %	Lipid %	Moisture %	Ash %
	17.40	1.68	79.50	1.02	17.00	2.40	79.20	1.00
	<u>+</u>	<u>+</u>	±	<u>±</u>	<u>±</u>	<u>+</u>	±	<u>±</u>
	0.12	0.04	0.24	0.06	0.12	0.04	0.18	0.04
	17.80	2.10	78.40	1.24	17.40	2.60	78.10	1.20
150	±	\pm	±	±	±	±	±	±
-	0.08	0.06	0.18	0.08	0.10	0.06	0.14	0.06
225	18.25	2.40	77.80	1.30	17.80	2.80	77.60	1.28
	±	±	±	<u>±</u>	<u>+</u>	<u>+</u>	±	<u>±</u>
	0.12	0.06	0.14	0.08	0.14	0.06	0.18	0.04
	18.60	2.60	77.20	1.40	$18.20 \pm$	3.20	76.80	1.36
	±	±	±	<u>±</u>	0.10	<u>+</u>	±	<u>±</u>
	0.10	0.08	0.18	0.06		0.08	0.22	0.06
	18.80	2.80	76.40	1.60	18.60	3.40	76.20	1.55
	±	±	±	±	±	±	±	±
	0.10	0.06	0.22	0.05	0.12	0.08	0.15	0.04
Mean	18.16	2.35	77.86	1.31	17.80	2.88	77.58	1.28

The results showed that the moisture content (water) values were the highest compared to the other components in the muscles of the studied regions in the studied species, which ranged from 76.2 - 72.10% and 79.5 - 76.4%. In R1 region for *M.sharpeyi* fish and *C. luteus* fish Respectively (Table 3 and 4), while the values in R 2 region ranged from 76.0 - 71.6% and 79.20 - 76.20% in *M.sharpeyi* and *C.luteus* respectively (Table 3 and 4). The results showed that the total average of moisture content in the *M.sharpeyi* fish was 73.985%, while 77.72% was recorded in *C.luteus* fish (Table 5). Due to differences in values of moisture content in R1 and R2 regions. The results of the statistical analysis showed no significant differences (P>0.05) in R1while the differences were significant (P<0.05) in R2 between the studied fish (Table 6).

Tables (3 and 4) showed a variation in ash content values between the studied species, which ranged between 1.30 - 1.84% and 1.02 - 1.60% in R1 region for *M.sharpeyi* fish and *C.luteus* fish respectively, which revealed significant differences (P<0.05)between the studied fish (Table 6). But the statistical results did not indicate significant differences (P>0.05) between the studied species, which reflect the values of the moisture content in R2 region, which ranged between 1.22 - 1.76% and 1.0-1.55% in the *M.sharpeyi* fish and *C.luteus* fish, respectively (Tables 3 and 4).

Table (5) indicates that the highest values of calories (energy) were found in the *M.sharpeyi* fish, which recorded 122.58 kcal/g while was noted 95.455 kcal/g in the *C.luteus* fish, these data reflecting the energy values for protein and lipid contents for studied fish which were 47.34 kcal/g in the *M.sharpeyi* fish while it was 71.92 and 23.55 kcal/g in the *C.luteus* fish (Table 5).

Fish species	Protein %	Energy value (Kcal/g)	Lipid %	Energy value (Kcal/g)	Total energy value (Kcal / g)
M.sharpeyi	18.81	75.24	5.26	47.34	122.58
C.luteus	17.98	71.92	2.615	23.535	95.455

Table(5): Total averages of protein, lipid contents and caloric (energy) values of *M. sharpeyi* and *C.luteus*.

Table (6) : Statistical analysis of proportions , diameters and chemical composition ofmuscles of R1 and R2 in *M. sharpeyi and C. luteus* .

R1 in M.sharpeyi and C.luteus				R2 in M.sharpeyi and C.luteus			
Parameter	F– values	Significant level	Differences	F – values	significant level	Differences	
Proportion of	2.355	0.163	Non-	2.439	0.157	Non-	
red fiber			significant			significant	
Diameter of	4.300	0.072	Non-	5.268	0.051	Non-	
red fiber			significant			significant	
Protein	5.426	0.048	significant	3.867	0.085	Non-	
content						significant	
Lipid content	42.885	0.000	significant	36.338	0.000	Significant	
Moisture	17.168	0.003	Non-	16.539	0.004	Significant	
content			significant				
Ash content	5.513	0.048	significant	3.678	0.091	Non-	
						significant	

Discussion

The musculature tissues in the teleosts fish are arranged into pieces of fillet (myomers) (Ayala *et al.*, 2005; *kiessling et al.*, 2006). The main muscle fibers are classified into three separated muscle layers(red,pink and white). The superficial red muscle is a thin layer of slow muscle fibers which are used for slow and sustained swimming (Love,1980;Mansour,1998;Coughlin,1999;Mansour,2005,2018;Al-

Muhanna,2018). Red muscle fibers have abundant mitochondria, high lipid and glycogen content (Ayala *et al.*, 2005, Mansour,2005). Fish are generally different in muscle proportions, depending on species and muscle location in the same species (Martinez et al. 2017; AL- Muhnna, 2018). This variation is related to the mode of life (Love, 1980), which reflects the level of fish activity (Pauly, 1989; Mansour, 2005). Pauly (1989) noted that the fish activity depends on three factors: the gill respiratory area, red muscle proportions and the content of alimentary tract. Therefore, the current fish showed a variation in red muscle proportions in the studied regions. Also, the results revealed that red muscle proportions are increasing in the posterior region of fish, which reflects the importance of this region in the fish activity. Al-Badri (1985) suggested when studying the muscles of the Carpet shark, *Chiloscyllium*

arabicum, that the posterior region (peduncle caudal) with the caudal fin formed the main locomotion organ in fish.

Several previous studies indicated that the red muscles in fish have small and homoloegenous diameters compared to white muscles that have larger and heterogeneous diameters. Greer-walker (1970) explained that the red muscle fibers ranging from 30 to 40 micron in fish with lengths of 250-350 mm while white muscle fibers ranging from 80 to120 micron. Also the current study indicate the decreasing the averages of the red muscle fibers diameters in posterior region in studied fish body, where previous studies pointed that the decreasing of the muscle fibers were due increases in the number and size of small muscle fibers to (Stickland, 1983; Graham et al., 1983; Rowlerson and Vegetti, 2001; Mansour; 2005; Karahmet et al., 2014). Rowlerson and Vegetti (2001) explained that fish continue to grow which reflecting an increase in muscle mass (increased number and diameter of muscle fibers). These processes are called hyperplasia and hypertrophy, both of which produce muscle growth.

The growth fish is accompanied by many changes, one of them the chemical composition of the fish muscles, which varies from species to another depending on internal and external factors including sex, age, size, muscle location, physiological condition, environment and season (Bosch, 2012; Pawar and Sonawane, 2013; Martines *et al.*, 2017). So the current results showed a difference in the values of the components of the chemical composition in the studied species.

In the study of the chemical composition for muscles of some freshwater fish in Iraqi waters, Hantoush et al., (2015) found that the values ranged 14.74 - 19.15% protein, 3-16-6.03% lipid, 71.23-78.15% moisture and 1.28 - 3.25% ash. These values were associated with factors such as geographical area, age, size, season and sexual activity. Therefore, the current results showed a convergence in the values of chemical composition components in the current fish muscles with the results of the previous studies. Pawar and Sonawane (2013) pointed to variation the values of protein content in five bonny fish in India. They found the protein content in Rasbora daniconius were 32.79% and they results were related to season, size, sex and reproductive cycle. Therefore, the current results of protein content have been linked to factors of sex, size and red muscles location in the body. Ackman (1989) classified fish into four groups according to lipid content values: high lipid fish > 8%, medium lipid fish 4-8%, low lipid fish 2-4% and lean lipid fish < 2. According to the values of the lipid content in the current fish, the *M.sharpeyi* fish can be placed in medium lipid fish (5.26%) while the C.luteus fish in low lipid fish (2.615). The differences in the values of moisture and ash

contents in the studied fish muscles may be due to differences in species, size, and muscle location. Oliveira et al. (2003) observed differences in the values of chemical composition in fish might due to factors such as water quality, feeding, species and sexual maturity . Younis *et al.*, (2015) noted the season had an affecting on the values of chemical composition components in Nile tilapia, *Oreochromic niloticus*. The results revealed that the *M.sharpeyi* fish has higher calories (energy) 122 .58 kcal /g compared with 95.45 kcal/g in the *C.luteus* fish. These values are a reflection of the contents of the protein and lipid, which reflects the amount and proportions of the muscles (Bosch, 2012 ; Martines *et al.*, 2017 ; Mansour, 2018).

Conclusions

1. The variation in the proportions and diameters of the red muscle in the current fish is due to the species, size and muscles location in the body.

2. The importance of the posterior region (caudal peduncle) in the fish movement.

3. According to the values of the lipid content, the *M.sharpeyi* fish can be placed in medium lipid fish while the *C.luteus* fish is within low lipid fish.

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