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The chemical composition of golden grey mullet *Liza aurata* in southern Caspian Sea during sexual rest and sexual ripeness

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Abstract. This study was conducted to examine the chemical composition (fat, protein, moisture, and ash) of *Liza aurata*'s muscle during sexual rest and sexual ripeness. The samples were collected from 100 adult *L. aurata* in 10 stations in the south coasts of the Caspian Sea (from Astara on the border with Azerbaijan to Gomishan on the border with Turkmenistan) in November and April 2014. The samples were taken to the laboratory in ice box. The weight and length of the fish were measured in each phase. According to the bioassay results, the weight and length of all the fish during the sexual ripeness were significantly higher than those during the sexual rest. The level of chemical substances during the sexual rest and sexual ripeness 3.94% and 2.22% of fat, 22.85% and 21.81% of protein, 77.39% and 78.13% of moisture, and 1.48% and 1.35% of ash, which showed a decrease in the fat and protein content and an increase in the moisture of muscles during the sexual ripeness. The decrease in fat and protein content of the muscles might be due to the growth of gonads and other processes related to spawning in the fall, and restoration of the fat content in the spring might be due to the termination of the spawning season and appropriate feeding.
Key Words: chemical composition, *Liza aurata,* sexual periods, body composition.

Introduction. Golden grey mullet *Liza aurata* (Risso, 1810) is a typical marine schooling fish. It is found in the southern part of the Caspian Sea throughout the year. It is a euryhaline fish, sensitive to a decrease in water temperature. Age of maturity is 3-4 years old. Spawning lasts from 10-20 August till the end of September, at water temperature of 22-23°C. It feeds on detritus, silt, and occasional mollusks. *L. aurata* is a valuable commercial fish (Velikova et al 2012).

The biochemical composition of fish muscle varies greatly depending on species, sexual cycle, age, food, stage of maturity, environment, season, organs and muscle location (Noël et al 2011; Roy et al 2006). The lipid and protein balance is important in assessing the flesh quality and as an indicator of seasonal cycles of reproduction and feeding (Clay 1988). During starvation periods, the fish uses the energy depots in the form of lipids and also may utilize protein, thus depletion of these reserves results in a general diminution of biological condition (Huss 1995). It also seems likely that principal constituents of developing gonads arise, at least in part, from other tissues of the body (Takama et al 1985).

To ensure the nutritional value as well as eating quality of fish, the seasonal biochemical evaluation is necessary. In Iran, Caspian Sea coast is considered a main seafood resource; very little information is available on the biochemical constituents in relation to reproductive cycle of commercial Iranian fishes. *L. aurata* constitutes one of the largest portion of catches and the highly consumed fish species in south Caspian Sea coast (Abdoli & Naderi 2009). In order to use this species as good as possible, the need for further studies aiming to elucidate many factors such as the amount and the variation of its composition over the year are significant. Studies on the biochemical composition of wild fish populations are rarely undertaken due to the difficulties in sampling, preservation of the samples and availability of a wide range of specimens from different ages, sizes, sexes and seasons from unpredictable catches of professional fisheries. The knowledge on biochemical composition of fishery species has fundamental importance in the application of different technological processes (Zaboukas et al 2006; Connell 1975). Moreover, biochemical composition is important as an aspect of quality of raw material, sensory attributes and storage stability (Osibona et al 2009) and is often dependent on sex (De Metrio et al 1989) and stage of sexual maturity (Connell 1975; Huss 1988, 1995). Yet, no detailed chemical study of biochemical composition of this fish is available. Hence, the objective of this study was to examine the seasonal changes over the year in the chemical compositions (including protein, lipid, ash and moisture) of *L. aurata* from the South Caspian Sea in the ripeness and sexual rest stages.

Material and Method. The samples were randomly collected from 100 adult *L. aurata* in 10 stations in the south coasts of the Caspian Sea, including stations 1, 2, 3 and 4 in Gilan Province; stations 5, 6, 6, 7 and 8 in Mazandaran Province; stations 9 and 10 in Golestan Province during sexual rest (April 2014) and sexual ripeness (November 2014) (Figure 1).

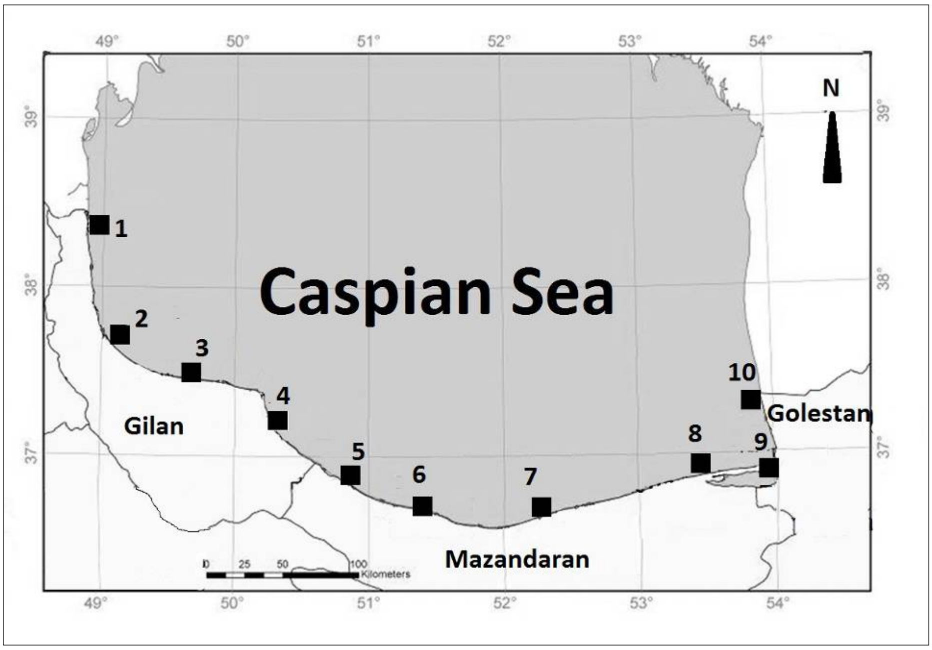

Figure 1. Map of sampling stations of fishes in the south coasts of the Caspian Sea.

Ten (10) adult fish were supplied from each station (5 fish during the sexual rest and 5 other fish during the sexual ripeness). The samples were taken to the Fisheries Research Laboratory Azad University Tonekabon Branch in ice box. Once the fish were washed, the bioassay indexes, including the weight (W) and total length (TL), were measured respectively using a scale of ±2 g precision (a digital scale, SE-62 DY-888, Iran) and a graded board of ±1 mm precision.

To measure the body composition, 200 g fillet from each individual without skin (1 cm thick) was prepared and refrigerated until the beginning of the tests (Ali et al 2005). The body composition was measured (3 times) through standard methods (AOAC 2005). To measure the moisture of muscles, 2 g from every single individual was placed on an aluminum plate (weighed beforehand). Then, the samples were placed in an oven at 105°C (Shimazco, Iran) for 24 hours until the weight of plates did not change (AOAC 2005). The protein content was measured using Kjeldtherm (James 1995) (a Kjeldahl system, VELP Scientifica Heating Digester, Europe) (AOAC 2005). The ash content was determined through placing the raw sample in a furnace (Barnstead, France) at 600 °C (AOAC 2005). The fat content was measured using Kinsella et al (1997)'s method and chloroform and methanol solvent (1:2) and was shown in g per 100 g of the muscle.

Data are expressed as Mean±S.D. Significant differences between groups were determined by a One-way analysis of variance (ANOVA) followed by Duncan test. Statistical analysis was performed using the SPSS for Windows software, version 18 (SPSS Inc., Chicago IL, USA). Mean values are considered significantly different at $p < 0.05$.

Results. Table 1 provides the mean bioassay results of the different stations for the sexual rest period and the sexual ripeness period. During the sexual rest, the maximum and minimum weights were 1071.66 g and 544.66 g, respectively, and maximum and minimum lengths were 53.66 cm and 42.53 cm, respectively ($p < 0.05$). During the sexual ripeness, the maximum and minimum weights were 1352.33 g and 427.33 g, respectively, and maximum and minimum lengths were 56.63 cm and 46.70 cm, respectively ($p < 0.05$). Based on the results, the fish at sexual ripeness had higher weight and length than the fish at sexual rest ($p < 0.05$) (Figure 2).

Table 1

Mean bioassay results (± standard deviation) in *L. aurata* for both sexual periods

	TL (cm)	*W (g)*
Sexual rest	48.45±3.78	821.10±197.02
Sexual ripeness	50.11±5.68	934.56±331.33
Sig.	0.018	0.030

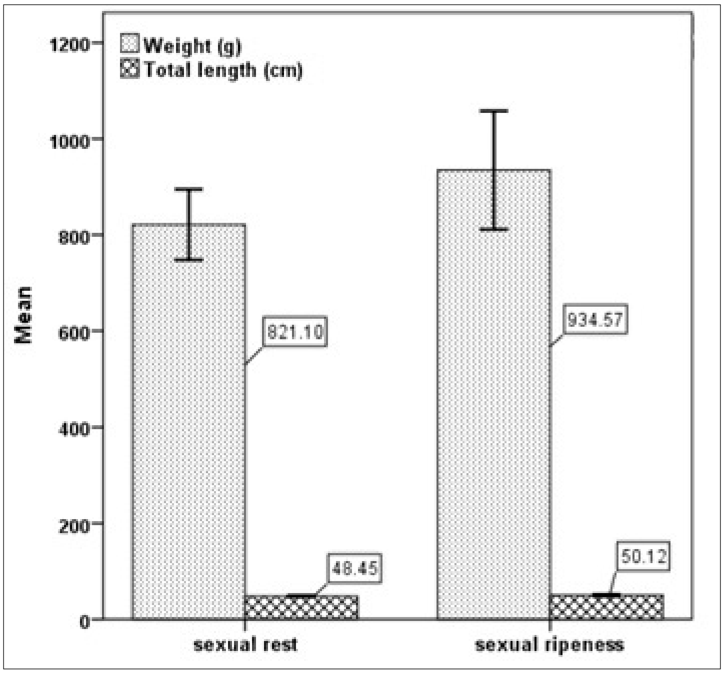

Figure 2. The biometric composition of *L. aurata* during both sexual periods.

The results on the chemical composition of the fish muscle during the sexual rest were as follows: maximum and minimum moisture, ash, protein and lipid contents were respectively 79.54% and 75.65% ($p > 0.05$); 2.07% and 0.90% ($p > 0.05$); 24.75% and 21.52% ($p > 0.05$); 4.83% and 3.7% ($p > 0.05$). The results on the chemical composition of the fish muscle during the sexual ripeness were as follows: maximum and minimum moisture, ash, protein and lipid contents were respectively 81.57% and 75.32% ($p > 0.05$); 1.72% and 0.96% ($p > 0.05$); 23.96% and 19.24% ($p < 0.05$); 2.73% and 0.78% ($p < 0.05$).

According to Table 2 and Figure 3, the results on the chemical composition of the fish muscle during both sexual periods showed that the mean fat content of the fish at sexual rest (3.94%) was significantly higher than that of the fish at sexual ripeness

(2.22%). Mean protein content of the fish at sexual rest (22.85%) was also significantly higher than that of the fish at sexual maturity (22.81%). However, the mean moisture content of the fish at sexual ripeness (78.13%) was significantly higher than that of the fish at sexual rest (77.39%). The mean ash content of the fish at sexual ripeness (1.35%) was insignificantly higher than that of the fish at sexual rest (1.48%). Table 3 shows Pearson Correlation and Significant between biometric parameter and biochemical composition.

Table2

Mean nutrient content (± standard deviation) in *L. aurata* for both sexual periods

	Fat %	*Protein %*	*Moisture %*	*Ash %*
Sexual rest	3.94±0.975	22.85±1.660	77.39±1.655	1.48±0.504
Sexual ripeness	2.22±0.707	21.81±2.296	78.13±2.916	1.35±0.433
Sig.	0.043	0.030	0.018	0.840

Table 3

Pearson Correlation (PCC) and Significant (2-tailed) between biometric parameter and biochemical composition

		W	*TL*	*Fat*	*Protein*	*Moisture*	*Ash*
				Sexual rest			
W (g)	PCC	1	0.933**	0.193	-0.193	-0.326	0.240
	Sig.		0.00	0.308	0.306	0.079	0.201
TL (cm)	PCC	0.0933**	1	0.132	-0.146	-0.221	0.301
	Sig.	0.00		0.486	0.441	0.240	0.106
Fat (%)	PCC	0.193	0.132	1	-0.131	-0.310	-0.013
	Sig.	0.308	0.486		0.491	0.096	0.947
Protein (%)	PCC	-0.193	-0.146	-0.131	1	-0.096	-0.105
	Sig.	0.306	0.441	0.491		0.614	0.581
Moisture (%)	PCC	-0.326	-0.221	-0.310	-0.096	1	0.314
	Sig.	0.079	0.240	0.096	0.614		0.091
Ash (%)	PCC	0.240	0.301	-0.013	-0.105	0.314	1
	Sig.	0.201	0.106	0.947	0.581	0.091	
				Sexual ripeness			
W (g)	PCC	1	0.919**	0.0505**	-0.046	-0.120	-0.068
	Sig.		0.000	0.004	0.811	0.529	0.720
TL (cm)	PCC	0.0919**	1	0.398*	-0.224	0.046	-0.195
	Sig.	0.000		0.029	0.234	0.811	0.301
Fat (%)	PCC	0.0505**	0.398*	1	0.187	-0.432*	0.148
	Sig.	0.004	0.029		0.324	0.017	0.434
Protein (%)	PCC	-0.046	-0.224	0.187	1	-0.297	0.215
	Sig.	0.811	0.234	0.324		0.111	0.254
Moisture (%)	PCC	-0.120	0.046	-0.432*	-0.297	1	-0.400*
	Sig.	0.529	0.811	0.017	0.111		0.028
Ash (%)	PCC	-0.068	-0.195	0.148	0.215	-0.400*	1
	Sig.	0.720	0.301	0.434	0.254	0.028	

** Correlation is significant at the 0.01 level (2-tailed); * Correlation is significant at the 0.05 level (2-tailed).

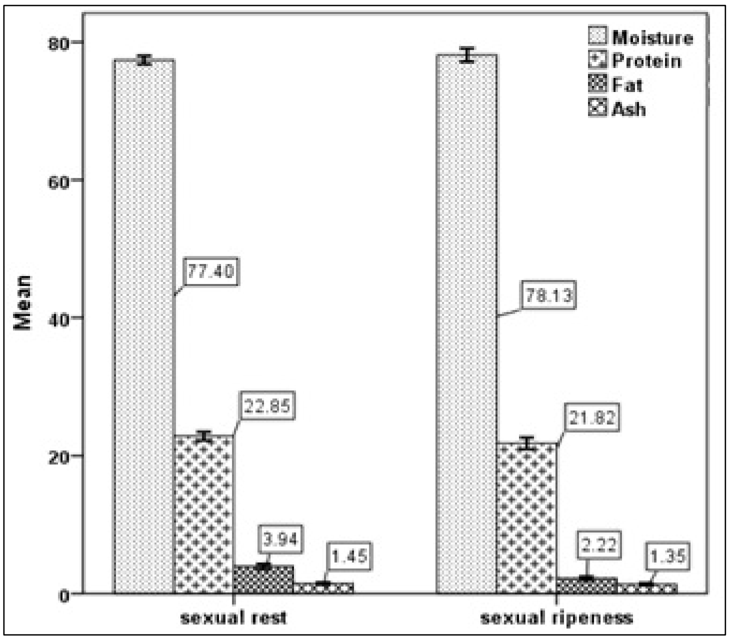

Figure 3. The chemical composition of the fish muscle during both sexual periods.

Discussion. The comparison of sexual rest and sexual ripeness periods in terms of the biomarkers showed that both weight and length were higher in the fall than in the spring. The increased weight of fish during sexual ripeness might be due to the increased weight of gonads. According to study of Abdoli & Naderi (2009), *L. aurata* spends its sexual rest in the spring and its sexual ripeness in the fall. The fish ovaries would be filled up with sex cells (oocytes and spermatozoa) in the fall. Therefore, mean weight of the fish is higher in the fall than in the spring. Gonads release the sex cells completely, which justifies the decrease in the weight in early spring.

The growth of fish is accompanied with various changes, including the biochemical composition of the body, which is a suitable marker for studying the physiological conditions of the fish. Approximate body composition includes moisture, protein, fat, and ash; and the remaining substances are carbohydrates and non-protein substances (Aberoumad & Pourshafi 2010). The biochemical substances largely vary from one fish to another and much depend on the age, sex, environment and season; the level of fish protein, fat, ash, and moisture in the body usually ranges from 16-21%, 0.1-25%, 0.4-1.5%, and 60-81% of the body weight, respectively (Muraleedharan et al 1996).

Given that the chemical composition of fish varies to a large extent depending on the species of the fish, age, and environmental conditions, such as the temperature and season (FAO 2004), the role and direct effects of the sea on biochemical substances of *L. aurata* would be clear.

Our results showed that the mean fat and protein contents in the fish tissue during the sexual rest were higher than those during the sexual ripeness. However, the moisture content during the sexual rest was lower than that during the sexual ripeness. Regarding that the sexual ripeness of this type of fish occurs in the fall, and body reservoirs (fat and protein) are used for producing sexual products, the decrease in fat and protein is compensated with water. Furthermore, the fish uses less food due to its physiological condition in that time. Fats comprise a part of the chemical composition of the fish muscle and differ in amount from one fish to another (Rehbein & Oehlenschlager 2009). The fat content depends on the composition of the aquatics' food. Fat is stored in the fish when the gonad is inactive because fats are energetic substances necessary for division of spermatogonia or when oocytes begin to grow. Moreover, an amount of the cholesterol (structural fat) is used as a precursor of sexual maturation hormones during the sexual ripeness (androgens, estrogens, and progesterone), and thus, a downward trend is observed in the total fat content (Khalko & Khalko 2002). The reproductive cycle and lack of nutrition are important causes of the changes in the biochemical composition of the body because the body uses the fat reservoir to supply energy (Begum & Minar 2012). Moreover, Merayo (1996) explained that the seasonal changes in moisture, fat, and protein content of the muscle and liver have been recognized in several species of the family Gobiidae, and the growth of gonads and other processes related to spawning have been introduced as the causes of those changes. Merayo (1996) also reported that the maximum moisture content and minimum protein content were observed during the sexual ripeness, and the fat content of the liver was retrieved in the non-spawning season probably due to the termination of the spawning season and appropriate feeding.

In a study similar to the present study, Khitouni et al (2014) examined the seasonal changes in the chemical composition of the muscle of grey mullet living in Gabs Bay in Tunisia and found a negative correlation between fat content and water content of the muscle. Khitouni et al (2014) observed the maximum fat content in September and maximum protein content in the spring and summer. In a similar study performed on gray mullets by Das (1978), maximum and minimum fat content was respectively observed in the adult fish and spawning fish (Bhuyan et al 2003), as the decrease in fat content of the muscle during the spawning period was referred to the decreased feeding (Pilla et al 2014). Other previous studies have also reported the changes in biochemical substances of the fish influenced by different factors. Karsli et al (2014) examined the biochemical composition of *Barbus* species in Turkey in different seasons and attributed the difference between the seasons to the reproduction period and sex. Karaton & İnanli (2011) attributed the seasonal changes in the body composition to the species of the fish, sex, season, and region. Pawar & Sonawane (2013) also introduced the food, migration, sexual changes at spawning as causes of the variation in the chemical composition of the fish body.

According to Table 5 the fat to biometric parameter relationship was strong in the sexual ripeness that showed decries of lipid and protein content percent and increasing moisture percent of muscle in the ripeness stage. Probably, decreasing of fat and protein content percent of muscle is spent on energy needs and complete maturation of genital products in autumn and increasing of muscle lipid and protein content in spring is probably connected with feeding.

According to Stansby (1962), the fish with fat less than 5% and protein of 15%-20% are called high-protein low-fat fish, and its seems that *L. aurata* belongs to this group of fish. Moreover, fish are often classified into three groups based on their fat content: thin fish (with fat content lower than 5%), medium-fat fish (with fat content of 5%-10%), and fat or fatty fish (with fat content higher than 10%) (Rahman et al 1995; Jabeen & Chaudhry 2011). Based on the results of this study, *L. aurata* belongs to the thin fish group.

The protein content of fish varies between 15% and 25% that may highly decrease and reach 15% when the fish cannot access food for a long time and during ripeness and spawning (Pilla et al 2014; Rehbein & Oehlenschlager 2009). A similar study conducted by Pirestani et al (2009) also showed that *L. aurata, Cyprinus carpio*, and *Sander lucioperca* were considered as low-fat fish based on the fat content lower than 5%, while, *Clupeonella cultriventris caspia* and *Rutilus frisii kutum* were classified as fatty fish. Therefore, the protein content in the present study was higher than that of other fish, such as *Clarias gariepinus*, *C. carpio*, *Cyprinion macrostomum*, *Capoeta damascina*, *Barbus* sp., *Squalius cephalus*, *Tilapia zillii* and the fat content was in the fat range *C. carpio*, Barbus sp., *T. zillii*, though lower than, of other fish, such as *C. gariepinus*, *C. damascina*, *S. cephalus* (Table 4).

Table 4

Mean values of the content (% wet mass) in lipid and crude protein of fish species muscle, caught in different areas

Author	*Area*	*Species*	*Lipid*	*Protein*
Khitouni et al (2014)	Tunisian coast	*L. aurata*	15.7±0.12	17.41-22.75
Kumaran et al (2012)	Southeast Coast of India	*Mugil cephalus*	2.42±0.21	17.56±0.22
Pirestani et al (2009)	South Caspian Sea	*L. aurata*	4.9±0.0	21.3±0.2
		Rutilus frisii kutum	6.7±0.0	21.4± 0.6
		Cyprinus carpio	3.6±0.0	19.4± 0.9
		Sander lucioperca	2.0±0.0	20.4± 0.4
		Clupeonella cultriventris caspia	10.2±0.1	18.4± 0.4
Yedukondala et al (2014)	East coast of India	*Apogon quadrifasciatus*	2.82±0.0	16.65±0.43
		Photopectoralis bindus	13.87±0.0	15.16±0.58
		Parachaeturichthys polynema	4.24±0.0	19.68±0.99
		Uranoscopus archionema	3.08±0.0	19.80±0.41
Hosseini-Shekarabi, et al (2013)	Oman Sea	*Atrobucca nibe*	1.01±0.0	18.19±0.145
	South Caspian Sea	*C. carpio*	2.71±2.01	15.99±0.29
		Ctenopharyngodon idella	2.71±1.56	15.18±0.61
Present study	South Caspian Sea	*L. aurata*	3.94±0.97-	22.85±1.66-

Conclusions. The results showed that the biochemical composition of *L. aurata* changed under the influence of the reproductive cycle in the fall, as the fat and protein content of the fish decreased, and the moisture content increased; and the fat and protein content significantly increased, and the moisture content decreased during the sexual rest in the spring. The knowledge of the chemical composition of the fish contributes to clarifying the effect of the environment on the physiology and nutritional condition of the fish, better fishing, prevention of fishing in the spawning season, and protection of the fish diversity.

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