

**Short communication****Correlation between body weight and nutritional value of *Alosa kessleri*****Mohammad Reza Ghomi<sup>1\*</sup>, Elena Tricarico<sup>2</sup>, Mehdi Nikoo<sup>3</sup>, Ahmad Ghenaatparast<sup>4</sup>**<sup>1</sup>*Department of Fisheries, Tonekabon Branch, Islamic Azad University, 46817 Tonekabon, Iran*<sup>2</sup>*Università degli Studi di Firenze, Department of Biology, Florence, Italy*<sup>3</sup>*Department of Fisheries, Faculty of Natural Resources, Urmia University, 5756151818, Iran*<sup>4</sup>*Shahid Ansari Hatchery Center, Rasht, Iran*<sup>\*</sup>*Corresponding author: mghomi@tonekabon.iau.ac.ir***Abstract**

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In this study, chemical composition and fatty acid content in *Alosa kessleri* regarding to body weight was determined. Thirty fish (average weight =  $47.43 \pm 33.47$  g) in the weight range of 10.3 to 123.0 g were caught by gill net from Caspian Sea (Chalous, Iran) and transported to laboratory for further analysis. A significant correlation between fish weight and protein ( $r = 0.458$ ,  $P = 0.011$ ) and fat ( $r = -0.622$ ,  $P = 0.001$ ) content was observed. There was a close range of the three groups of fatty acids to each other, saturated fatty acids (SFA = 31.41%), polyunsaturated fatty acids (PUFA = 30.69%), and monounsaturated fatty acids (MUFA = 29.18%). Palmitic acid (C16:0) (20.85% of total fatty acids), and oleic acid (C18:1) (24.95% of total fatty acids), were the most abundant SFAs and MUFA, respectively. A higher content of n-3 fatty acids was observed than n-6 fatty acids, resulting in n-3/n-6 ratio of 5.25. A significant correlation was found for DHA/EPA ratio ( $r = 0.449$ ,  $P = 0.013$ ) with fish weight. The results of the study indicated that protein and lipid content and DHA/EPA ratio were influenced by body weight.

**Keywords:** *Alosa kessleri*; chemical composition; fatty acid; body weight

**Introduction**

Seafood is major source of high-quality protein with well-balanced essential amino acids and nutritionally valuable lipids and fatty acids (Chaijan et al., 2006). It is rich in n-3 polyunsaturated fatty acids (PUFAs) and contains low cholesterol level (Barrento et al., 2010). The main bioactive omega-3 fatty acids are eicosapentaenoic acid (EPA) (20:5n-3) and docosahexaenoic acid (DHA) (22:6n-3) (Jacobsen et al., 2008; Barrento et al., 2010; Orban et al., 2011). N-3 is not synthesized in the human body (Simopoulos, 2002), and seafood is reported to be the only main source of n-3 PUFA in the human diet (Ghomi et al., 2012). Due to the strong clinical support for the role of EPA and DHA in maintaining

health, the intake of omega-3 fatty acids, specifically EPA and DHA is recommended (Simopoulos, 2002; Barrow et al., 2007). With the growing emphasis on the importance of n-3 PUFA on human health and nutrition, it is important to determine the n-3 content of fish and fish products (Ghomi et al., 2013).

Fatty acid composition of fish flesh is influenced by several biological factors such as diet, season, fillet portions, species, habitat, water salinity and age of the fish (Badiani et al., 1997; Katikou et al., 2001; Shirai et al., 2002; Celik et al., 2005; Gonzalez et al., 2006; Palmeri et al., 2007; Özogul et al., 2007; Huynh and Kitts, 2009; Xu et al., 2010; Usydus et al., 2011; Ghomi et al., 2012; Tao et al., 2012; Ghomi et al., 2013). However, information on the effect of weight of

fish on fatty acid composition is limited. Ghomi et al. (2013) indicated that in farmed beluga (*Huso huso*), protein content and essential n-3 PUFA (content of n-3, n-6, EPA, DHA and n-3/n-6 ratio) increased significantly with increasing body weight of beluga. Palmeri et al. (2007) investigated the effect of Murray cod (*Maccullochella peelii peelii*) size on the proximate composition and found a lower fat content in smaller fish, more likely due to the utilization of fat at a faster rate during early growth stages. A study on three different fish species including brackish-water kutum (*Rutilus frisii kutum*), warm-water silver carp (*Hypophthalmichthys molitrix*) and cold-water rainbow trout (*Oncorhynchus mykiss*) indicated the correlation between fish weight and protein content and fatty acid composition (Ghomi et al., 2012).

*Alosa kessleri* Grimm 1887 is a type of bony species in the family of *Clupeidae* living in the Caspian Sea. *Clupeidae* includes several species like *Alosine* which has six other species mostly outspread in Mexico gulf and North Atlantic (Munroe and Nizinski, 2002). *A. kessleri* lives along shores of central and northern parts of Caspian Sea but in south and especially southeast in winter. *A. kessleri* is assumed to be the tastiest clupeid because of its high fat content (Coad, 2013). Due to the presence of highly nutritious omega-3 fatty acid in flesh, this species is becoming more popular and the consumption has been increasing with market demand.

However, little information is yet available on the nutritional quality of *A. kessleri* based on the fatty acid composition of flesh. As so far, there is no record of work done in assessing the nutritional quality of this species based on fatty acid composition, particularly in relation to weight, to provide nutritional information for better utilization of this species, the chemical compositions and nutritional values are required. Therefore, this study aimed to analyze the proximate and fatty acid composition of *A. kessleri* muscle and to explore the nutritional values in relation to the body weight of the fish as a correlation study model for *Clupeidae*.

## Materials and Methods

### Fish samples

Fresh *A. kessleri* (average weight =  $47.43 \pm 33.47$  g,  $n = 30$ ) in the weight range of 10.3 to 123.0 g were caught by gill net from Caspian Sea (Chalous, Iran) and transported to laboratory in boxes containing ice. The ratio of fish to ice was 1:2. Upon arrival, the fish were decapitated, gutted, skinned, and washed with tap water to remove blood and slime. Slices from the mid sections of the fish were collected to analyze the proximate composition and fatty acid content. The weight of fishes was recorded to the nearest of 0.01 g.

### Proximate composition

Moisture was determined by drying the samples in an oven (D-63450, Heraeus, Hanau, Germany) at 105°C to a constant weight (AOAC 2005). Ash was determined by incineration in a muffle furnace (Isuzu, Tokyo, Japan) at 600°C for 3 h (AOAC 2005). Crude protein was determined by the Kjeldahl method ( $N \times 6.25$ ) using an automatic Kjeldahl system (230-Hjeltec Analyzer, Foss Tecator, Höganäs, Sweden) (AOAC, 2005).

### Lipid extraction

For lipid extraction, the method of Bligh and Dyer (1959) was followed. Fifty grams of fish chopped muscle was homogenized using Ultra Turax homogenizer (IKA® T25 digital Ultra Turax®, Staufen, Germany) at 6000 rpm for 2 min in a solution contained 50 ml chloroform and 100 ml methanol. Then 50 ml of chloroform was added and the mixture was further homogenized for 30 s. Finally, 50 ml of distilled water was added to the mixture and homogenized for 30 s as above. The homogenate was centrifuged (Avanti J-E, Beckman Coulter, Inc., Fullerton, CA, USA) at 4.000 rpm for 15 min at 4°C. The supernatant was then transferred into a separating flask, and the lower phase (chloroform phase) was drained off into a 250-ml Erlenmeyer flask containing 4 g anhydrous sodium sulfate and shaken vigorously to remove any water residues which may remain in the solution. The solution was then filtered through a Whatman No. 4 filter paper into a round-bottom flask. A rotary evaporator (Rotavapor R-114, Buchi, Switzerland) was used for solvent evaporation at 50°C.

### Fatty acid analysis

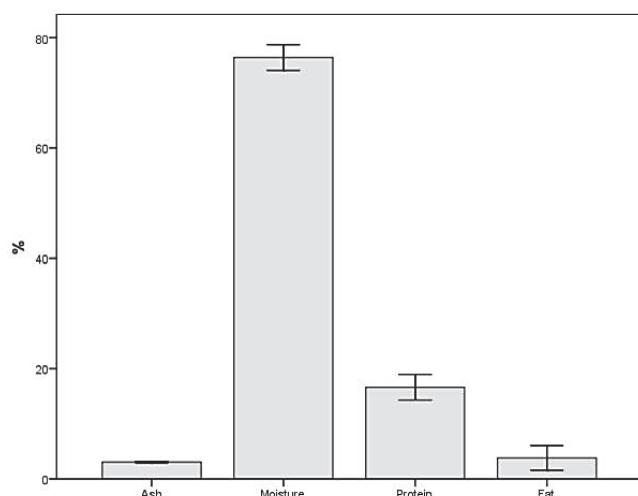
Fatty acid composition was determined on the oils extracted by the method of Bligh and Dyer (1959). Fatty acid methyl ester was prepared as follows: lipid samples (1 g) were diluted with 2 ml of 2 M potassium hydroxide in methanol followed by the addition of 7 ml n-hexane in a sealed tube. The mixture was then shaken using a vortex for 1 min and left for about 20 min in a water bath (temperature 50°C to 55°C) until it was separated into two phases. From the top layer, fatty acid methyl ester was then taken for analysis using Trace GC (Thermo Finnigan, Rodano, Italy). The GC conditions were as follows: capillary column (Bpx-70, 60 m, 0.32 mm, i.d. 0.25 µm), split ratio of 90:1, injection port temperature of 250°C, and flame ionization detector temperature of 270°C. The oven temperature was set at 195°C. The flow rate of carrier gas (helium) was 1 ml/min, and the makeup gas was N<sub>2</sub> (30 ml/min). The sample size injected for each analysis was 1 µl. The data are expressed as grams/100 grams of total fatty acids.

### Statistical analysis

Pearson's correlation coefficient with a significance level of  $P < 0.05$  was used for establishing linear correlations. Data are expressed as mean  $\pm$  standard deviation (SD).

## Results and Discussion

The proximate content of *A. kessleri* is shown in Fig. 1 and varied as follows: protein 9.19-22.4% (mean = 16.39%), fat 0.22-7.12% (mean = 3.69%), moisture 73.0-80.0% (mean = 76.48%), and ash 2.91-3.61% (mean = 3.05%). A similar protein content of kutum (18.3%), silver carp (15.5%) and rainbow trout (17.64%) (Ghomí et al., 2012), common carp (15.64%), silver carp (18.01%), bighead carp (18.03%), grass carp (14.73%), Wels catfish (17.34%), and zander (19.27%) were recorded (Ljubojevic et al., 2013). According to fat content, fish are usually classified into lean (fat content < 2%), low-fat (fat content 2% to 4%), medium fat (fat content 4% to 8%), and high-fat (fat content > 8%) (Haard, 1992). The present study indicated that the *A. kessleri* with a fat content 3.69% could be placed amongst those fish with low fat content (fat content 2-4%).



**Fig. 1. Proximate composition of *A. kessleri*, error bars shows standard deviation**

A significant correlation between fish weight and protein ( $r = 0.458$ ,  $P = 0.011$ ) and fat ( $r = -0.622$ ,  $P = 0.001$ ) content was observed (Table 1). Palmeri et al. (2007) analyzed the proximate composition of Murray cod of different sizes and reported a lower fat content in smaller fish, more likely due to the utilization of fat at a faster rate during early growth stages. In beluga, a significant correlation between

fish weight and protein content was observed ( $r = 0.504$ ,  $P < 0.05$ ) (Ghomí et al., 2013). Similarly, in silver carp, body protein was significantly correlated with weight ( $r = 0.574$ ) (Ghomí et al., 2012).

**Table 1. Correlation between *A. kessleri* weight and proximate composition**

	Ash	Moisture	Protein	Fat
r	0.375*	0.241	0.458*	-0.622*
P-value	0.041	0.209	0.011	0.001
n	30	29	30	27

\*Significant at  $P < 0.05$

The fatty acid composition of *A. kessleri* is shown in Tables 2 and 3. Saturated fatty acids (SFAs) were the most predominant fatty acids (31.41% of total fatty acids) followed by polyunsaturated fatty acids (PUFAs; 30.69% of total fatty acids) and monounsaturated fatty acids (MUFA; 29.18% of total fatty acids) (Fig. 2).

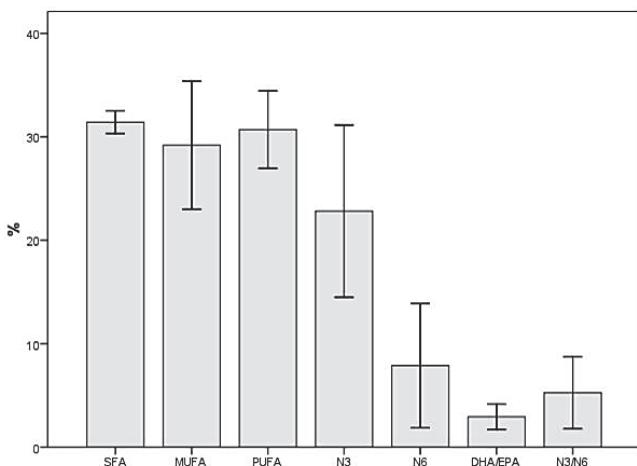
**Table 2. Fatty acid composition (% of total fatty acids) of *A. kessleri***

Fatty acids	n	Min.	Max.	Mean	$\pm$ SD
C14:0	30	1.51	6.90	3.599	$\pm$ 1.50
C16:0	30	18.00	24.99	20.846	$\pm$ 2.05
C18:0	30	1.80	6.99	4.361	$\pm$ 1.29
C20:0	30	.90	3.90	2.607	$\pm$ 0.97
C16:1	30	1.35	7.90	4.229	$\pm$ 1.76
C18:1	30	10.10	36.20	24.955	$\pm$ 6.67
C18:2-N6	30	1.10	18.05	6.509	$\pm$ 6.25
C18:3-N3	30	1.34	2.80	2.051	$\pm$ 0.40
C20:4-N6	30	.70	2.80	1.369	$\pm$ 0.41
C20:5-N3(EPA)	30	1.50	11.01	5.634	$\pm$ 2.69
C22:6-N3(DHA)	30	3.39	29.12	15.126	$\pm$ 6.85

Palmitic acid (C16:0) (20.85% of total fatty acids), and oleic acid (C18:1) (24.95% of total fatty acids), were the most abundant SFAs and MUFA, respectively (Table 2). Docosahexaenoic acid (C22:6) was the main n-3 fatty acid (15.126% of total fatty acids), while linoleic acid (C18:2) was the major n-6 fatty acid (6.51% of total fatty acids). Higher content of DHA (15.126%) was observed than EPA (5.634%). Essential polyunsaturated fatty acids such as eicosapentaenoic acid (EPA) (20:5n-3) and docosahexaenoic acid (DHA) (22:6n-3) are not synthesized in the human body; therefore, their inclusion in diets is essential (Ghomí et al., 2012). A higher DHA/EPA ratio is more beneficial to health as DHA is more efficient than

**Table 3. Correlation between *A. kessleri* body weight and fatty acid composition (% of total fatty acids)**

	C14:0	C16:0	C16:1	C18:0	C18:1	C18:2	C18:3	C20:0	C20:5	C22:6	C20:4
r	-.026	.079	-.190	-.140	.222	.121	.176	-.036	-.328	-.129	-.331
P-value	.890	.677	.313	.462	.239	.524	.351	.851	.077	.498	.074
n	30	30	30	30	30	30	30	30	30	30	30

**Fig. 2. Major class of fatty acids (% of total fatty acids) of *A. kessleri*, error bars shows standard deviation**

EPA in reducing the risk of coronary heart disease (Simopoulos, 2002). Higher content of DHA than EPA (equals with a higher DHA/EPA ratio) were also observed for Japanese sardine (*Sardinops melanostictus*) (Shirai et al., 2002), sardine (*Sardina pilchardus*), anchovy (*Engraulis encrasicholus*) and picarel (*Spicara smaris*) (Zlatanos & Laskaridis, 2007), sardine (*Sardinella gibbosa*) (Chaijan et al., 2006), Sardine (*Sardina pilchardus walb*) (Garcia-Arias et al., 2003), Brazilian sardines (*Sardinella brasiliensis*) (Saldanha et al., 2008), wild perch (*Perca fluviatilis* L.) (Jankowska et al., 2010), sea bass (Özogul et al., 2007), cultured sea bass, gilthead sea bream, and rainbow trout (Testi et al., 2006); carp, walleye Pollock, cod, and Baltic salmon (Usydus et al., 2011). On the other hand, a significant positive correlation was found for DHA/EPA ratio ( $r = 0.449$ ,  $P = 0.013$ ) with fish weight for *A. kessleri* in present study (Table 4). Thus, another positive point for nutritional quality of this species could be raised by

increasing body weight and expecting the DHA/EPA ratio to be boosted as well.

The ratio of n-3/n-6 is an important nutritional index of lipid quality (Simopoulos, 2002). The n-3 PUFA is useful for suppressing increases in plasma cholesterol, preventing cardiovascular diseases and visual function (Shirai et al., 2002). Seafood is the only significant source of n-3 PUFA, predominantly EPA and DHA (Orban et al., 2011). The n-3/n-6 ratio in *A. kessleri* was 5.25 (ranging from 0.40 to 11.16) (Fig. 2). The optimal ratio of n-3/n-6 fatty acids is 0.2 (Sargent, 1997). The n-3/n-6 ratios observed for other species were 0.6 in farmed beluga (*Huso huso*) (Ghomi et al., 2013), 0.99-1.36 in Asian hard clam (*Meretrix lusoria*) (Karnjanapratum et al., 2013) and 2.13-3.73 in small sized Murray cod (*Maccullochella peelii peelii*) (Palmeri et al., 2007). In general, the n-3/n-6 ratio in seafood is higher than the recommended value (1:5), and from a nutritional viewpoint, this is highly beneficial and desirable for the daily human diet (Usydus et al., 2011). Therefore *A. kessleri* with a high ratio of n-3/n-6 (5.25), is recommended for human nutrition. There was no correlation between n-3, n-6 and their ratio (n-3/n-6) with the body weight in *A. kessleri* ( $P > 0.05$ ) (Table 4). Ghomi et al. (2013) reported that larger farmed beluga contained significantly higher n-3/n-6 ratio ( $r = 0.828$ ,  $P = 0.000$ ) when compared to smaller fish which could indicate the beneficial health effect of larger farmed beluga fillets.

## Conclusions

The nutritional value of *A. kessleri* based on proximate and fatty acid composition was established. However, none of individual fatty acids exhibited a correlation with body weight, a significant correlation solely for DHA/EPA ratio, together with protein and fat was observed. *A.*

**Table 4. Correlation between *A. kessleri* body weight and major fatty acids (% of total fatty acids)**

	n3	n6	DHA/EPA	SFA	MUFA	PUFA	N3/N6	TFA
r	-.204	.103	.449*	-.083	.185	-.287	-.175	-.009
P-value	.280	.588	.013	.661	.329	.124	.355	.962
n	30	30	30	30	30	30	30	30

\*Significant at  $P < 0.05$

*kessleri* is a good source of essential omega-3 fatty acids (EPA and DHA) and a high ratio of n-3/n-6 fatty acids (5.25) which indicating this species as a high nutritive value.

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