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# **Research Article**

# Reproductive biology of *Alburnus mossulensis* (Teleostei: Cyprinidae) in Gamasiab River, western Iran

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**Abstract:** In the present investigation, some reproductive characteristics of *Alburnus mossulensis* were examined. Sampling of 325 specimens were carried out at monthly intervals from Gamasiab River, Tigris River drainage, western Iran, through the year 2013. Age, sex ratio, fecundity, oocytes diameter, gonadosomatic (GSI), modified gonadosomatic, Dobriyal and hepatosomatic indices were measured. Regression analysis was used to estimate the relations between fecundity and standard length, body weight, gonad weight and age. All of the male with 70.0mm and female with 75.0mm in standard length, and all of those older than one year were ripe. The mean egg diameter ranged from 0.11mm (August) to 0.84mm (May). Spawning took place in May, when the water temperature was 18 to  $22^{\circ}$ C. Average GSI values for females at the beginning of the reproduction period (March) and in ripe mature females (May) were 5.05% and 12.03%, respectively. The averages of absolute and relative fecundity (g/body weight) were 1920 eggs (±1025SD) and 99.03 (±94), respectively. The absolute fecundity was significantly related to body length and ovary weight.

**Keywords:** Cyprinid Fish, Spawning Season, Hepatosomatic Index, Gonadosomatic Index, Persian Gulf.

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#### Introduction

Iran harbors approximately 288 freshwater fish species in 107 genera, 28 families, 22 orders and 3 classes distributed throughout different basins (Esmaeili et al. 2017). This ichthyofauna is dominated by the family Cyprinidae. Among the drainage systems, the Caspian Sea basin shows the greatest fish species diversity, followed by the Tigris, Karun, and Persis basins (Jouladeh-Roudbar et al. 2015; Keivany et al. 2016). The cyprinid fishes of genus *Alburnus* in Iran, comprise eight confirmed species including *Alburnus mossulensis* (Mousavi-Sabet et al. 2013, 2015). *Alburnus mossulensis*  is found in the Tigris-Euphrates basin and adjacent basins. In Iran, it is recorded from the Tigris River, Kor River, Persis, Maharlu Lake, and upper reaches of the Hormuz basins (Berg 1949; Abdoli 2000; Mousavi-Sabet et al. 2015; Coad 2017; Esmaeili et al. 2010, 2017; Esmaeili & Teimori 2016) also from the whole middle to upper Karkheh basin including Simareh, Qarasu and Gamasiab rivers (Abdoli 2000). This species is found in streams, rivers, lakes, reservoirs and marshes. *Alburnus mossulensis* feeds on the phytoplankton, organic detritus and insects (Coad 2017).

Reproductive biology investigation of fish species is vital in order to assessing commercial potentialities of its stock, culture practice and actual management of its fishery (Doha & Hye 1970; Soofiani et al. 2006; Dopeikar et al. 2015). It has three key components including: sexual maturity, reproductive period and fecundity, which are important demographic characteristics, essential for understanding a species life history (Cortes 2000). Age data are used in order to assessing the fish population dynamics that include growth, mortality, and recruitment and stock structure; also, this data is an essential component of age-structured population models (Bagenal 1967; Ghanbarzadeh et al. 2014). Estimation of age at first maturity requires a precise determination of gonadal maturation stages, whatever this method used for the estimation of the spawning season (Bagenal 1967; Oso et al. 2013).

Counting of scale or otolith annuli commonly is used in order to conduct age determination and usually requires the measurement of a large number of specimens (Fletcher 1991). Fecundity is the number of ripening eggs in a female prior to the next spawning season. Ripe ovaries are appropriate for fecundity estimation. Gonadosomatic Index (GSI) expresses the weight of the gonad in relation to the body weight and is expressed as percentage (Bagenal 1967; Oso et al. 2013; Mousavi-Sabet et al. 2011; Keivany et al. 2012; Mousavi-Sabet 2012; Mousavi-Sabet et al. 2012a,b). The reproductive potential, i.e. fecundity is an important biological parameter that plays a significant role in evaluating the commercial potentials of fish stocks (Gómez-Márquez 2003; Jamali et al. 2014, 2016).

The main object of this study was to provide data on the reproductive biology of a cyprinid fish, including sex ratio, gonadosomatic index, fecundity, oocyte diameter, and spawning season which are necessary for conducting conservation and management programs of *A*. *mossulensis*, sampled from Gamasiab River.

# Materials and Methods

Fishes were collected from January 2013 to December 2013 by monthly intervals using electrofishing and gillnet from the Gamasiab River (34°22'249"N, 47°54'729"E), the Tigris River drainage (Persian Gulf Basin), in Kermanshah Province, western Iran. All the collected specimens were fixed and preserved in 4% formaldehyde solution and transferred to the ichthyology laboratory.

In the laboratory, for each specimen, total length (TL) and standard length (SL) were measured using digital calipers with a precision of 0.1mm and body weight (W) was taken on a digital balance to the nearest 0.01g (Table 1). Sex determination was done by examination of the gonad tissue either with eye or by microscopic examination of the gonads (by a binocular 25-40×). Three indices were used to investigate the monthly changes in gonads in order to estimating the spawning season of this species: Gonadosomatic Index (GSI), Modified Gonadosomatic Index (MGSI) and Dobrival Index (DI). In order to determine the GSI (GSI =  $[Wg \times W^{-1}] \times 100$  (Biswas 1993), MGSI  $(MGSI = [Wg \times W^{-1}] - Wg \times 100)$  (Nikolsky 1963) and DI (DI=  $\sqrt[3]{Wg}$ ) (Dobriyal et al. 1999), ovaries were weighed (Wg) to the nearest 0.001g. Also, monthly Hepatosomatic Index (HSI) was calculated using the formula:

## $HSI = [W1 \times W^{-1}] \times 100$

Where Wl was the liver weight. The absolute fecundity (Fa) was estimated in 30 ovaries by counting 0.05g of oocytes (Biswas 1993). In

order to determine the number of eggs, pieces of approximately 0.05g were removed from the anterior and median parts of both ovarian lobes. The pieces were weighed and eggs were counted under a binocular microscope. The number of eggs in each female was calculated as the proportion of eggs in the sample to the weight of the whole ovary. In order to calculate absolute fecundity, ovaries in stages IV or V were used. The stage of gonad maturity was determined visually following the Nikolsky's scale (1963). The relative fecundity (Fr) was expressed by dividing the absolute fecundity (Fa) by the fish body weight (Bagenal 1967). To determine the ovum diameter, the ovaries were preserved in 4% formaldehyde solution. The diameters of 30 ova of each female specimen were measured using a binocular microscope model M6C-10 which was fitted with an ocular micrometer.

The age of each specimen was assessed based on the annual growth of scales taken from the left side of the body, between the end of the pectoral fin and beginning of the dorsal fin (Bagenal 1967; Mousavi-Sabet et al. 2012a, b).

In order to compare significant differences in the GSI index among the samples taken in different months and various sizes, the analysis of variance and Tukey's test were applied (P<0.05). The strength and significance of the relationship between the absolute fecundity (Fa) and selected individual features of the females included in the study (standard length, total body weight, the gonad weight and age) were analyzed by determining Pearson's correlation coefficient r (P<0.05) and regression equations (Thulasitha 2013). The obtained data were analyzed by SPSS 16.0 and Microsoft Excel 2010 software packages.

**Table 1.** Standard length (SL), total length (TL) and body weight (W) (mean±SD) in different ages of male and female specimens of *Alburnus mossulensis* from the Gamasiab River.

Age	Ν	SL (mm)			TL (mm)		W (g)	W (g)	
	М	F	М	F	Μ	F	М	F	
1+	14	10	88.50±10.8	80.00 ±9.15	$106.00 \pm 12.64$	97.50±10.60	9·96±4·64	8.80±2.40	
$2^{+}$	53	33	112.36±11.23	$109.04 \pm 13.65$	$131.40 \pm 13.14$	$128.33{\pm}14.08$	22.15±7·20	19.73±7.86	
3+	63	55	$122.57{\pm}11.20$	$117.78 \pm 10.60$	$142.42 \pm 13.13$	136.38±11.08	$28.42 \pm 7.44$	27.02±27.02	
4+	46	35	$133.33 \pm 5.70$	$137.00 \pm 12.04$	$155.00 \pm 5.00$	$158.00 \pm 14.40$	35.40±4·10	49.35±10.11	
5+	7	9	145.47±12.36	$144.43 \pm\! 10.41$	$164.52 \pm 12.10$	164.92±13.31	38.64±17·19	40.23±10.00	

N: number of specimens, SD: standard deviation, M: male specimens, F: female specimens.

### Results

During this study, 325 specimens of *Alburnus mossulensis* were collected from Gamasiab River. The measured standard length (SL) and total weight (TW) were 70.0–155.0mm, and 6.3-54.6g, respectively. The obtained results showed that age of this species ranged from 1<sup>+</sup> to 5<sup>+</sup> years. Most of the fish were 2<sup>+</sup> (27.4%) and 3<sup>+</sup> (35.6%) years old, some were 1<sup>+</sup> (7.4%) and 4<sup>+</sup> (24.6%) and only a few of them were 5<sup>+</sup> (4.9%) years old. Based on the results obtained from the gonad examination, it was observed that all of the males with 70.0mm and females with 75.0mm in standard length and those older than one year (approximately 2 years old) were ripe individuals. From the total number of 325 collected specimens, 142 were females and 183 were males, giving an overall sex ratio of 1F:1.3M. The difference between the number of females and males was significant ( $x^2 = 5.172$ ; P < 0.05). In order to determine the spawning season, the mean GSI (Fig. 1) and HSI (Fig. 2) were calculated monthly. The results revealed that the peaks of GSI and HSI are in the months of May and August, respectively. Significant differences were observed between the female and male GSI and MGSI (P<0.05). There was no significant difference between GSI and MGSI/DI (P>0.05). The female GSI increased gradually in March and reached the maximum value in May (at water temperature of 18-22°C) and decreased gradually from May to August (Fig. 1.) This trend of the gonad indices and ova diameter showed that this species spawns in May (Figs. 1, 3).

Individual values of the absolute fecundity varied in a wide range from 118 to 5720 eggs with an average of 1920 eggs ( $\pm$ 1025.37SD) (Table 2). The relative fecundity was 10.78 to

205.25 eggs with a mean of  $100 (\pm 94)$  per gram of body weight (Table 2). Absolute fecundity was significantly related to female body length and gonad weight, with the regression coefficient values of 0.55 and 0.41 for body length and gonad weight, respectively (Table 3).



**Fig. 1.** Variation of mean gonadosomatic index (GSI) of female (F) and male (M) specimens of *Alburnus mossulensis* from the Gamasiab River in different months.



**Fig. 2.** Variation of mean hepatosomatic index (HSI) of female (F) and male (M) specimens of *Alburnus mossulensis* from the Gamasiab River in different months.



Fig. 3. Variation of mean ova diameter (mm) in female *Alburnus mossulensis* from the Gamasiab River in different months.

Table 2. Absolute (Fa) and relative (Fr) fecundity in particular ranges of body length (SL) and body weight
(W) of female Alburnus mossulensis from the Gamasiab River.

Parameters		Ν	Fa		Fr		
			Range	Mean±SD	Range	Mean±SD	
	7.5-10	9	118-1892	1132.38±544.21	16.62-205.25	87.03±50.11	
SL (mm)	10-12.5	17	330-3600	1809.09±822.36	11.70-158.49	$76.60 \pm 37.29$	
	12.5-15	4	347-5720	2574.48±1148.22	10.78-144.44	67.21±30.78	
	<25	11	118-3249	1415.17±749.08	16.62-205.25	$84.81 \pm 46.00$	
W (g)	25–45	14	347-5720	2294.67±874.76	10.78-144.44	$69.44 \pm 29.87$	
	45<	5	2454-3260	$2832.33 \pm 405.26$	44.95-60.59	52.68±7.83	
	2	15	118-1892	1372.19±544.21	10.78-205.25	73.74±46.26	
Age	3	12	330-3600	2190.82±822.36	29.14-137.65	$75.75 \pm 28.96$	
	4	3	347-5720	3570.50±1148.22	52.51-144.44	85.55±33.79	

n = number of specimens; SD = standard deviation.

**Table 3.** Correlation coefficients r and regression equations for relationships between absolute fecundity (Fa) and body length (SL), body weight (W), weight of ovary (Wg) and age of *A. mossulensis*.

Relationship	N	Linear Regression	r	$r^2$	F	Р
Fa-SL	30	Y=-2209.595X+353.480	0.547	0.300	2.775	0.007
Fa-W	30	Y= 554.504X+50.996	0.547	0.299	1.966	0.056
Fa-Wg	30	Y=1408.53X+290.659	0.409	0.167	7.667	0.001
Fa-Age	30	Y= -654.627X+987.682	0.629	0.396	1.599	0.115

## Discussion

In this study, the most abundant age group in the catches was  $3^+$  (35.6%) with average standard length of 120.18±10.90mm. Jawad (2004) used eye lens diameter for ageing the young (up to age 3) of this species from the marshes north of Basra. Ergene (1993) studied the growth of this species in the Karasu of Turkey and found 4 age groups and mentions 5 age groups for another Turkish study. Ergene

(1993) reported averages fork length of 118.2mm, 131.0mm, 145.2mm and 163.3mm, for 4 age groups, respectively. Türkmen & Akyurt (2000) also worked on this species in the Karasu River and reported age groups of 1 to 6. According to them, age group 3 was the most abundant group. Length and age at first maturity have been reported to be 1.26 years and 9.24cm for males and 1.81 years and 9.65cm for females in Karasu River, Turkey with the oldest age group of 7 years (Yıldırım et al. 2007).

In many species, the HSI is an appropriate index for prediction of gonadosomatic condition and amount of allocated energy for reproduction activity (Yagarina & Marshall 2000; Siami et al. 2017). Typically, in many fish species, at the peak of spawning, the GSI is in highest value and the HSI increases (Galloway & Munkittrick 2006). Based on the obtained results in different months, the lowest value of HSI in A. mossulensis was at the beginning of May, and it was in its highest value in August. Variation in the HSI might be contributed to the vitellogenesis in the liver. present very low Vitellogenin is in concentrations in the plasma of immature or male organisms. During vitellogenesis, the liver stimulated to produce females is of vitellogenin, which in turn, is incorporated into the yolk of developing oocytes. Vitellogenesis induction in general results in enhanced liver metabolism leading to an enlargement of the liver and consequently an increased HSI (Christensen et al. 1999). In order to response to the energy shortage, organism management their reserves energy (Wootton 1999). So, maybe take part of the reproductive cycle of this fish in the cold months (winter), at the beginning of reproductive period fish faced with a cold period. On the other hand, the fishes experienced a poor environmental condition such as food availability. Therefore, in order to increase survival, they save more energy in the summer when environmental condition is favorable. This strategy (saving energy in the summer in order to overwintering) was also

reported in other species (Love 1970; Dopeikar et al. 2015; Siami et al. 2017). The monthly changes in GSI showed that the reproductive period of A. mossulensis began in March and ended in August. According to obtained result from gonadosomatic examining, the GSI values increased in March and got its highest value in May, then decreased gradually from May to August. After that, a secondary increasing was observed in ovaries development from August to September, when some of the studied fish were ready to spawn. The average absolute fecundity of female A. mossulensis from Gamasiab River was 1920 eggs (±1025), and ranged from 120 to 5720 eggs. The linear function was found well enough for expressing the Fa-SL and Fa-Wg relationships for this species in Gamasiab River. Berg (1949) reported mature eggs in a female with 155mm long. Yıldırım et al. (2007) examined this species in Karasu River of Turkey and found a male:female sex ratio of 1:1.08, not significantly different from 1:1, a fecundity range of 3012 to 11427 eggs, significant correlations between fecundity and fork length, total weight, age and gonad weight, and a spawning season from June to August when water temperature attained 15°C. Our results showed that the spawning season occurs within May to August in Gamasiab River which signifies the latitude effect in terms of temperature reproductive activity. on Temperature (which has major effects on gonad maturation and spawning season) is important in the onset of spawning and has a modifying role, particularly in cueing the precise timing of gamete maturation and spawning, providing the capacity for reproductive cycles to be locally tuned (Wootton 1999; Wright & Trippel 2009; Pankhurst & King 2010). Fecundity is affected by different environmental factors, which is different among fish species (Wootton 1999). Several factors, such as the size and age of the females (Thrope et al. 1984), life history strategy (Morita & Takashima 1998), and food supply and water temperature (Fleming & Gross 1990) affect the fecundity. Ayoade

(2011) suggested that variation in fecundity may be due to differential abundance of food. According to Jonsson (1999), fecundity increases with body size because the amount of energy available for egg production and the body cavity accommodating the eggs increases with fish size. The length seems to be a better indicator of the capacity of oocyte production than the weight, because fish dose not decrease significantly in size whereas weight may vary along the year (Bengenal 1967). Usually, the relative fecundity is higher in small females than in large ones (Lobon-Cervia et al. 1997). Bagenal (1967) asserted that fish species exhibit wide fluctuation in fecundity among fish of the same species, size and age.

The present study, on some aspects of reproductive biology of *A. mossulensis* has revealed the followings: the mature males and females were longer than 70.0 and 75.0mm, respectively in standard length (age 1+); the breeding season was from April to June with a peak in May; the mean absolute and relative fecundity were 1920 eggs ( $\pm$ 1025) and 100 ( $\pm$ 94) per gram body weight, respectively, and mean ovum diameter range of 0.11-0.84mm.

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مقاله پژوهشی

# زیستشناسی تولیدمثل شاه کولی جنوبی (Alburnus mossulensis) (ماهیان استخوانی عالی: کپورماهیان) در رودخانه گاماسیاب، غرب ایران

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چکیده: در مطالعه حاضر، برخی جنبههای زیستشناختی تولیدمثل شاه کولی جنوبی Alburnus mossulensis مورد بررسی قرار گرفت. نمونه برداری ها به صورت ماهانه و به مدت یک سال انجام شد و تعداد ۳۲۵ عدد ماهی از رودخانه گاماسیاب، حوضه آبریز تیگره (دجله)، در غرب ایران صید شد. سن، نسبت جنسی، هماوری، قطر تخمک، و شاخصهای گنادی-بدنی، گنادی-بدنی اصلاح شده، دابریال و کبدی-بدنی مورد مطالعه قرار گرفتند. آنالیز رگرسیون برای پی بردن به ارتباط بین هماوری و طول استاندارد، وزن بدن، وزن گناد و سن مورد استفاده قرار گرفت. تمام ماهیان نر و ماده به ترتیب با طول استاندارد ۷۰ و ۲۵ میلی متر و سن بیش از یک سال، بالغ و با رسیدگی جنسی بودند. میانگین ماهانه قطر تخمک از ۱۱/۱ (مرداد) تا ۸۴/۰ میلیمتر (اردیبهشت) اندازه گیری شد. تخمریزی در اردیبهشتماه، زمانی که دمای آب ۱۸ تا ۲۲ درجه سانتی گراد بود، صورت پذیرفت. میانگین شاخص گنادی-بدنی برای ماهیان ماده در شروع فصل تولیدمثل (اسفند ماه) و در رسیدگی کامل (اردیبهشت)، به ترتیب ۵۰/۵ و ۱۲/۱۰ درصد محاسبه شد. ماهیان ماده در شروع فصل تولیدمثل (اسفند ماه) و در رسیدگی کامل (اردیبهشت)، به ترتیب ۵۰/۵ و ۲۰ در ۱۲/۱۰ درصد محاسبه شد. ماهیان ماده در شروع فصل تولیدمثل (اسفند ماه) و در رسیدگی کامل (اردیبهشت)، به ترتیب مازای هر گرم وزن بدن، محاسبه شد. هماوری مطلق به صورت معنیداری با طول بدن و وزن تخمدان رابطه داشت.