## Some aspects of the Reproductive Biology of Barbus barbulus Heckel, 1847, from Karoon river, Iran

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#### **Abstract**

The present study was carried out from December 2007, during which to November 2008 more than 210 fish (Barbus barbulus) were caught and measured, including, 57 males, 64 females and 89 immature fishes. Maximum and minimum total lengths were 885 mm and 210 mm respectively. Maximum and minimum total weight was 8460 g and 99 g respectively. The length-weight relationship was Y=0.00002 L<sup>2.90</sup> (n=57, R<sup>2</sup>=0.85) for males, Y=0.000002 L<sup>3.22</sup> (n=64, R<sup>2</sup>=0.87) for females and Y=0.00005  $L^{2.96}$  (n=210,  $R^2$  =0.90) for total fishes, these results showed isometric growth of the studied fish. The mean value of Gonado-Somatic Index (GSI) for the male and female were 1.58  $\pm$  0.41 and  $1.85 \pm 0.22$  respectively. For both sexes, the highest GSI value was in March and the lowest in December. GSI and maturity stages indicated that spawning time was during March to April. The mean size at first sexual maturity (L<sub>m</sub>) was 380-430 mm for males and 470-520 mm for females.

**Key words:** Barbus barbulus, Reproduction, Karoon River, Iran.

#### Introduction

The order Cypriniformes represented by six families, 321 genera and some 3268 species (Nelson, 2006) is one of the most widespread fishes all over the world. Thus cyprinids are major element in the Iranian ichthyofauna, exists in all of its major basins. The genus *Barbus* is found in Europe, Southwest Asia and Africa.

It comprises about 800 species (Coad, 2006). More than 17 species of Barbus had been reported from different basins of Iran. B. barbulus which is locally known as "Berzemlabpan", is one of the most important fishes in the Tigris-Euphrates Basin and Karoon River (Berg, 1949; Marammazi, 1995; Abdoli, 2000). In Iraq it was found in the Tigris River basin, the Gulf basin at the Zohreh River the Shapur Dalaki rivers, the lower Mand River, the Helleh, Dozgah, Dasht-e Palang (and its tributary the Shur). This species was announced as endangered species in Turkey (Fricke et al., 2007). Several studies were conducted on species of this genus like B. grypus (Marammazi. 1995: **Nikpey** et al.. 2000). xanthopterus (Eskandary, 1999), Barbus pectoralis (Ghafari and Jamili, 2010) and Barbus esocinus (Eskandary et al.,2001). Al-Habbib et al. (1986) studied the spawning time of B. barbulus from the Tigris River at Mosul, Iraq.

The objective of this study was to provide information pertaining to reproductive biology of this species in Karoon River and is the first to present the complete reproductive characteristics based on observations and information analysis. These data could be used to improve the management of the fisheries of this valuable species.

#### Material and Methods

Samples of *B. barbulus* were collected monthly from the landing sites and also caught at five stations along the Karoon river: Gotvand, Shoshtar, Molasani, Ahwaz and Darkhoin in. During the period from 2007 to November 2008 (Fig.1, Table 1).

Fish sampling was carried out by using 80m long gill nets, with mesh sizes of 120 and 140 mm (stretched). Nets were anchored at each of the sampling stations at sunset and were removed at sunrise on the following day, remaining 12 hr in water. Total length (T.L., mm) to the nearest millimeter, and total weight (g) to nearest gram, were measured for each fish.

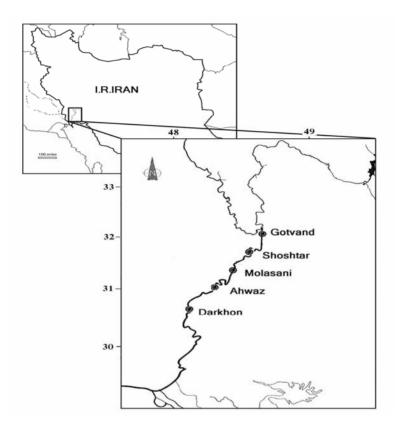


Fig 1: The map of Iran, Karoon river and the studied stations in Khuzestan province (South West of Iran)

Table 1: The coordinates of the five stations in Karoon river

Station	Longitudes E	Latitudes N
Gotvand	48°,50´	32°,12´
Shoshtar	48°,46´	31°,58´
Molasani	48°,52´	31°,29´
Ahvaz	48°,51´	31°,22´
Darkhoin	48°,25´	30°,45´

Parameters of the length weight relationship were obtained by fitting the power equation  $W = a \times L^b$  to length and weight data where: (W) is the total wet weight (g), (a) is a constant determined empirically, (T.L.) is the total length (mm) testing according to Biswas (1993), in order to verify if calculated b was significantly different from 3, the Students t-test was employed (Zar, 1996).

The maturity stage for males and females was determined microscopically using a six-stage maturity key (Kesteven, 1960). These stages included: Virgin (I), maturing virgin (II), developing (III), late developing (IV), spawning (V) and spent (VI) (Table 2).

Gonado-somatic index (GSI), was calculated by expressing the gonad weight as a proportion of the total body weight, and the proportion of females in spawning

condition were plotted against the sample period by month to establish the timing and frequency of spawning (Biswas, 1993). Population sexual structures were examined using Qi square ( $\chi$ 2) *goodness* of fit tests. Independent tests were conducted to determine whether sex ratios differed significantly from unity for whole samples and size categories within samples. The probability level was set at 0.05 (Sokal and Rohlf, 1995), and 1 degree of freedom for each comparison. The equation  $K=W\times10^2$  / L was used to find fish status changes (condition factor) in which W= weight and T= total length (Beckman, 1994).

The mean size at first sexual maturity was estimated for females by fitting the logistic function to the proportion of mature fish in 5-cm (L) size categories

 $Y=1/1+e^{(-a-bX)}$ . In which Y is the proportion of the number of all mature male and females to all immature males and females in the same length class, X in total length in cm and a and b are correlation constants (King, 1995). The mean size at first maturity was taken as that at which 50 % of individuals were mature.

Table 2. Degree of maturation and the morphology of the gonad in different stages of maturity of *Barbus barbulus*.

G.	D 0	D : : 0.1 1
Stage	Degree of	Description of the gonads
	maturation	
		Ovaries very small, thin, thread like pale incolour,
I	Immature	occupying a small part of the body cavity. Testes
	or virgin	are thin, slender translucent and pale in colour.
	and resting	Both the gonad invisible to the naked eye.
	adult	
		Ovaries become slightly larger and increase in
II	Early	weight and volume with minute opaque whitish
	maturing	eggs occupied about half of the body cavity.
		Testes become enlarge, flat, increase in weight and
		volume, and creamy white in colour. Both the
		gonad is readily seen without any aid.
III	D1i	Ovaries distended occupied, about 2/3 of
1111	Developing	abdominal cavity with large pale yellow eggs.
		Testes enlarge, increase in weight and volume,
		light pinkish and larger in size and look more
		vascular. Blood capillaries become conspicuous.
IV	Davalanad	Ovary becomes more enlarged occupying almost the entire body cavity, with large number of big,
1 V	Developed /pre	turgid, spherical, translucent, deep yellow riped
	spawning	
	spawning	ova. Testes become soft, turgid, pinkish red and
		increase in weight and volume. Blood capillaries prominent. Roe to milt run with slight pressure.
		Ovary walls become thin almost transparent.
V	Spawning	Riped eggs are visible through the ovarian wall
ľ	Spawning	and some riped eggs are present in the oviduct.
		Testes become flabby, thin and dull white in
		colour.
		Colour.
		Gonad shrunken having loose walls. Ovaries are
VI	Spent	flaccid, shrinked and sac like, reduced in volume.
	~ F ****	Ovary contains ripped unspawned darkened eggs
		and a large number of small ova. Testes become
		flabby, thin and dull white in colour.

#### Results

During this study *B. barbulus* were measured. From the total number of caught fishes, 57 and 64 were males and females, respectively and 89 specimens were immature.

The mean value of length for the male and female were calculated as  $445.38 \pm 11.5$  mm,  $516.12\pm14.9$  mm respectively and mean value of weight for the male and female were  $991.62 \pm 137.1$  g,  $1676.16 \pm 209.5$  g, respectively and the total length ranged between 885 mm to 210 mm, total weight ranged between 99 g to 8460 g (Table, 3). The length-weight equation were calculated as  $Y=0.00002L^{2.90}$  (n=57,  $R^2=0.85$ ) for males,  $Y=0.00002L^{3.22}$  (n=64,  $R^2=0.87$ ) for females and Y=0.00005  $L^{2.96}$  (n=210,  $R^2=0.90$ ) for all fishes (Fig. 2). The t- test analyses for the constant (b) with typical value (3) verified that there was no significance difference (p>0.05).

Table 3: Average values and standard deviation (±SD) of size corresponding of *Barbus barbulus* from the Karoon river (2007-08)

Month	N	Mean W. ±S.D (g)	Min. – max.	Mean TL. ±S.D (mm)	Min. – max.
January	20	$2029 \pm 159$	702-7000	$555.5 \pm 118$	346-850
February	25	1511±151	218-8460	484.8±16.5	270-885
March	26	1305±718	214-3820	$478 \pm 73.5$	373-660
April	29	545.4±553	99-3035	$355 \pm 92.7$	230-670
May	8	1217±514	353-1905	493±82.3	380-609
June	16	456.5±161	298-7190	140.1±340	293-710
July	13	449±208	167-905	337±63	256-400
August	2	$634 \pm 9.8$	642-627	416.5±9.1	410-423
September	6	1201±134	463-3935	475.5±134	370-665
October	12	919±374	353-2015	483.5±374	362-603
November	45	493.4±243	128-1410	425.4±243	215-430
December	8	957±137	310-580	393.7±92.7	210-400
Average	210	998±109	99-8460	404.1±111	210-885

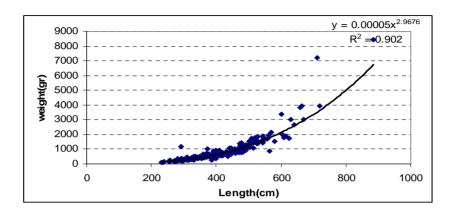


Fig. 2. The length- weight relationship curve for *Barbus barbulus* in Karoon river, Khuzestan province, Iran.

Average length and weight in females were higher than in males. Condition factor for different months is showed in Fig. 3. The K was highest during spring (April) sampling for the total population. The K mean values ( $\pm$ S.D) was calculated for males and females 1.05 ( $\pm$ 0.24), 1.31 ( $\pm$ 0.34) respectively (Fig. 3).

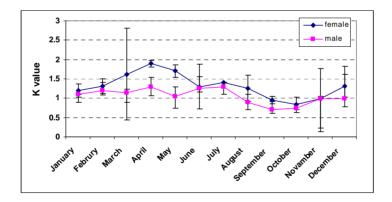


Fig.3. Monthly variation of Condition factor of *Barbus* barbulus (B) in Karoon river, Khuzestan province, Iran. Bars refers to standard deviation.

Fig. (4) Shows the monthly fluctuations of Gonado-Somatic index, the peaks for males and females were well defined and occurred simultaneously in March with spawning time during April. The mean value of GSI for the male and female were calculated as  $1.58\pm~0.41$  and  $1.85\pm~0.22$  respectively. During spawning time the number of fish with the highest mean GSI increased in Shoshtar station.

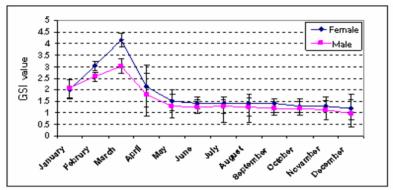


Fig4. Monthly variation of GSI of *Barbus barbulus* (B) in Karoon river, Khuzestan province, Iran. Bars refers to standard deviation.

Pearl organs in mature males were observed, 88.88 % in stage IV of maturation and 50 % in stage V of maturation during the spawning time. The mean size at first sexual maturity ( $L_m$ ) was 38-43 cm for males and 47-52 cm for females. The first maturity stage appeared in 335 mm for males and 423 mm for female. The different sex ratio values were shown in Table (4). The male-to-female sex ratio was 0.83:1, and there was no significant different among study months (p> 0.05,  $\chi$ 2=0.02, df= 1).

Table 4: Sex ratio and significant difference between	male and
female of Barbus barbulus in Karoon river (	2007-08)

Month	Total	Immature	ð	9	Sex	Qi	P-value
				-	ratio	square	
January	20	-	6	14	0.42	1.60	0.05>
February	18	-	7	11	0.63	0.44	0.05>
March	35	5	23	7	3.28	4.27	0.05<
April	39	34	1	4	0.25	0.90	0.05>
May	8	2	ı	6	0	3.00	0.05>
June	10	7	1	2	0.5	0.17	0.05>
July	10	8	1	1	1	0.00	0.05>
August	1	-	-	1	0	0.50	0.05>
September	6	2	1	3	0.33	0.50	0.05>
October	11	4	3	4	0.75	0.07	0.05>
November	44	23	13	8	1.62	0.60	0.05>
December	8	4	1	3	0.33	0.50	0.05>
Average	210	89	57	64	0.89	0.2	0.05>

During this study the water temperature ranged from 14.25 °C (Feb.) to 28.5 °C (Aug.). During the spawning time in March to April the water temperature was 16.75 °C (Fig. 5).

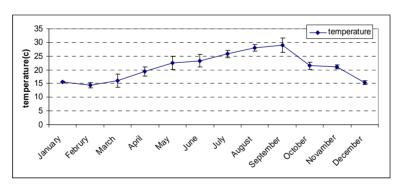


Fig.5. Monthly variation of temperature (°C) in Karoon river. Bars refer to standard deviation.

#### Discussion

Size sexual dimorphism was observed in *B. barbulus* since females dominated in the longer length classes. Pearl organ have been observed in males during spawning in April. Ghafari and Jamili (2010) obtained the Pearl organs of *B. pectoralis* in the spawning time.

The b values in the weight-length model were close to 3 for B. barbulus fishes which mean that the shape of fish is close to the cubic equation of length. The value of b for B. barbulus was b=2.88 at Habbaniya lake, and at Tharthar lake b= 3.02 in Iraq (Szypula et al., 2001). In Khuszestan the b values of Length-weight relationship was 2.98 and 2.97 for female and male of Barbus grupus, while for female and male of Barbus sharpeui was 2.82 and 2.96, respectively (Nikpey, 1996). The variation of b in the different regions could be by seasonal fluctuations in environmental parameters, physiological conditions of the fish at the time of collection, sex, gonad development and nutritive conditions in the environment of fish (Biswas, 1993). The length-weight relationship is a practical index of the condition of fish, and may vary over the year according to several exogenous and endogenous factors such as food availability, feeding rate, health, sex, gonad development, spawning period and preservation techniques (Bagenal, 1978; Tesch, 1968). The highest value of (K) was observed in spring after winter, a feeding season and decreased after spawning time in April which shows the effect of weight. K value decreased after spawning time due to Gonad spending. Condition factor is a well-being value and its increase coincide with fish weight increasing at seasonal growth and can measure growth changes related to fish food or reproduction stage (King, 1995). Sex ratio (males: females) was found to be 0.83: 1. In this study, significant differences from 1:1 sex ratio were observed in March. Sex ratio of Barbus xanthopterus was 1:1 (Eskandary, 1999). Sex ratio of Barbus esocinus differ from 1:1 as the ratio of female: male was 1:4 (Eskandary et al., 2001). These results could be typical of many temperate freshwater and marine fish (Pitcher and Hart, 1990). In general variation in sex ratio is might be a result of immigration, difference in behavior catchability and different mortality rates (Sadovy et al., 1994).

B. barbulus spawning was during March to April according to GSI. Al-Habbib et al. (1986) reported that spawning period during July and August in fish from the Tigris River at Mosul, Iraq. This could be related to the geographical and ecological differences between the stocks. Ghafari and Jamili (2010) found that Barbus pectoralis spawned in Karoon River (Khuzestan province) during March to April. Spawning season of Barbus xanthopterus in April (Eskandary, 1999), Barbus esocinus in May (Eskandary et al., 2001), Barbus grypus and Barbus sharpeyi between March to April (Nikpey, 1996) were reported. The single spawning period during late winter and spring was reported in other Barbus species in Khuzestan provinces (Maramazi, 1995, Eskandary, 1996). Patterns in the monthly GSI fluctuations that shows similar pattern to the reproductive cycles are common among tropical fishes (Sadovy, 1996).

During the ripping stages, temperature was low. It has been observed that cool water induced later maturity stages (Eskandary, 1999; Eskandary *et al.*, 2001). This pattern is similar to that reported by Nikpey (1996); Eskandary (1999) and Eskandary *et al.* (2001).

Mean size at first sexual maturity (Lm) for *B. barbulus* was estimated to be 38-43 cm in males and 47-52 cm in females. Mean size at first sexual maturity have been reported as 30 cm (males) and 49 cm (females) in *Barbus xanthopterus* (Eskandary., 1999), 15 cm (males) and 50 cm (females) in *Barbus esocinus* (Eskandary *et al.*, 2001). Ghafari and Jamili (2010) found length at maturity in *B. pectoralis* ranging from 35-40 cm in males and 50-55 cm in females. The results showed that *B. barbulus* needs a protection during the spawning period from March-April, which could help in the management of its stocks.

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# بعض مظاهر حياتية التكاثر لسمكة Barbus barbulus Heckel, 1847 من نهر كارون/ جمهورية إيران الإسلامية

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#### المستخلص

جمعت حوالي 210 سمكة من اسماك Barbus barbulus من نهر كارون في محافظة خوزستان- جنوب إيران، خلال الفترة من كانون الأول 2007 الى تشرين الثاني 2008، تشتمل على 57 ذكرا و46 أنثى و89 سمكة غير ناضجة. كان أقصى طول كلي حوالي 885 ملم وأدنى طول كلي 210 ملم. وكان أقصى وزن كلي 8460 غم وأدنى وزن 99 غم. كانت علاقة الطول بالوزن كالتالي:

 $(0.85=R^2,\,57=n)~Y=0.00002~L^{2.90}:$ للأناث:  $(0.87=R^2;\,64=n)~Y=0.000002~L^{3.22}$ للإناث:  $(0.90=R^2:\,210=n)~Y=00005~L^{2.96}$ لجميع الاسماك:  $(0.90=R^2:\,210=n)~Y=00005~L^{2.96}$ 

هذه النتائج تظهر نموا متشابها" (isometric) للأسماك المدروسة. كانت قيمة معدل دالة المناسل GSI) Gonado-Somatic الذكور  $(6.41\pm0.21)$  وللإناث  $(6.41\pm0.21)$  وبلغت اعلى قيمة لدالة المناسل لكلا الجنسين في آذار وأدناها في كانون الأول. تبين دالة المناسل ومراحل النضج ان فترة التسرئة (Spawning) كانت خلال آذار الى نيسان. وبلغ طول الجسم الكلي  $(1.60\pm0.21)$  عند النضج الجنسي  $(1.60\pm0.21)$  عند النضج الجنسي  $(1.60\pm0.21)$