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Life history traits and gonad histology of an endemic cyprinid fish, Mond spotted barb, *Capoeta mandica* from Southern Iran

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ABSTRACT

The life history traits and gonad development of an endemic cyprinid fish, the spotted barb, *Capoeta mandica* (Bianco & Banarescu 1982), from southern Iran was investigated by regular monthly collections from February 2006 through January 2007 and on the basis of microscopic and macroscopic analyses. No information on the spawning characteristics of the fish are available to date. A total of 335 specimens were captured using electrofishing including 253 males and 102 females, resulting in 2.5:1 (male: female) sex ratio, which is significantly different from the expected ratio. The commonly known five standard maturation stages were determined based on the size, shape and weight of the gonads, degree of occupation of the body cavity, presence or absence of ripe oocytes, or milt, diameter of the oocytes in the ovary, and histological observations. These stages were correlated to the reproductive indices. The ovum diameters ranged from 0.04 to 1.31 mm, with the highest mean value in May. The condition factor of male and female specimens was the highest in April. The female GSI increased from March to May, peaking in the middle of spring and decreased significantly in June. Based on the gonad maturation stages, gonadosomatic index (GSI), modified GSI, mean egg diameter, condition factor (K) and Dobriyal index (DI), it was concluded that the spotted barb spawns during May and June. The provided data, contribute baseline data towards management ecology and conservation of this endemic fish species.

Key words: Cyprinidae, Gonad histology, Reproduction indices, Spotted barb.

INTRODUCTION

The order Cypriniformes with 11 families and 4298 species is one of the largest order of the fishes. The family Cyprinidae comprises about 3050 species distributed throughout the world and is the largest family in term of species diversity in the order Cypriniformes (Blanc *et al.* 1971; Eschmeyer & Fong 2016). The cyprinid genus *Capoeta* is distributed from Central Asia to Western Asia including Anatolia, Azerbaijan, Afghanistan, Armenia, Georgia, Iraq, Iran, Israel and Uzbekistan (Banarescu 1991; Levin *et al.* 2012; Alwan *et al.* 2016). They generally occur in lakes and streams with fast and slow-flowing waters (Geldiay & Balik

1996) and include more than 26 species, 12 of which occur in Iran (Eschmeyer & Fricke 2017; Esmaeili *et al.* 2017).

Knowledge of the reproductive cycle and the factors affecting it are important issues in fish and fisheries biology (see Tomkiewicz *et al.* 2003; Chakrabarti & Barun 2017). Reproductive studies of fishes, such as assessment of size at maturity, duration of the spawning season and fecundity, require knowledge of the state of gonad development and a large number of macroscopic maturity scales in individual fish (Carrasson & Bau 2003). Although macroscopic staging can enable detailed recording of the seasonal occurrence of different reproductive

stages, histological analysis of the gonads can provide a more precise determination (Fazeli *et al.* 2015; Mirghiyasi *et al.* 2016). There are numerous studies determining reproductive stages of fishes in terms of both macroscopic and microscopic analyses (e.g. Tomkiewicz *et al.* 2003; Muchlisin *et al.* 2010; Dopeikar *et al.* 2015; Opadokun & Ajani 2015; Mirghiyasi *et al.* 2016) revealing its importance. Without having knowledge on reproduction cycles, it is difficult to effectively manage, and subsequently conserve, endemic fish populations (Al-Saleh *et al.* 2012).

The Mond spotted barb, *Capoeta mandica* is an endemic and poorly known cyprinid fish distributed in exorheic Persis basin in Southern Iran draining to the Persian Gulf. Despite the importance of this endemic species in the Persis basin and the importance of knowledge about reproductive biology of endemic fishes to define strategies for conservation, the reproductive biology of *C. mandica* has not been investigated (see Coad 2016).

This study presents the first detailed descriptions of its reproductive biology identifying criteria to assess the peak of spawning period and provides basic

knowledge in assessing its potential reproductive capability.

MATERIALS AND METHODS

Study site and sampling

The specimens of *Capoeta mandica* were collected monthly from the Rudbal River (near Firouzabad City), Mond River drainage (Persis basin) at 28°42'36"N, 52°38'11"E, Fars Province, Iran (Fig. 1). A total of 355 specimens including 253 males and 102 females were collected from February 2006 through January 2007 using electrofishing device. The specimens were fixed in 10% formalin at the spot, then transferred to the laboratory for further studies. The collected specimens were deposited in the Zoological Museum, Collection of Biology Department, Shiraz University (ZM-CBSU).

Morphometric study

Total length (TL), standard length (SL), fork length (FL), head length (HL), head depth (HD), minimum body depth (MinBD), maximum body depth (MaxBD) of the preserved specimens were measured to the nearest 0.05 mm using Vernier caliper, then weighted to the nearest 0.001g (total weight, W).



Fig. 1. Live specimen of *Capoeta mandica*, from Mond River drainage.

Length-weight relationship

Parameters of the length-weight relationship, $W = aL^b$ were estimated by linear regression of the log-transformed weight and length, where W is the whole body weight (g), L is the total

length, a is the intercept of the regression and b is the regression coefficient (Koutrakis & Tsikliras 2003; Esmaeili *et al.* 2014). Prior to regression analyses, log-log plots of length and

weight values were performed for visual inspection of outliers (Froese 2006).

Reproductive traits

Fish specimens were carefully dissected out ventrally. Sex and stage of maturity were ascertained macroscopically and microscopically followed by taking the weight of fixed gonads (GW) to the nearest 0.001g. Maturation stages were classified as follows: I, immature; II, developing; III, maturation; IV, ripe and V, spawning (running) (Babiker & Ibrahim 1979; Tacon *et al.* 1996). Eggs were measured under microscope using ocular scale. The Sex ratio was determined and a Chi-square (χ^2) test was used to assess deviation from 50:50 sex ratio (Robards *et al.* 1999).

To examine the monthly changes in the gonads and to estimate spawning season, four indices were used: Gonadosomatic index (GSI), modified gonadosomatic index (MGSI), Dobriyal index (DI) and reproductive condition (K) which were calculated as follows:

$GSI = (\text{gonad mass}/\text{fish mass}) \times 100$ (Nikolsky 1963)

$MGSI = (\text{gonad mass}/\text{fish mass} - \text{gonad mass}) \times 100$ (Nikolsky 1963)

$ID = 3 \sqrt{GW}$ (Dobriyal *et al.* 1999)

$K = WG/L$ (Way *et al.* 1998)

Where W is whole fish mass in g, GW is gonad mass in g and L is fish length in mm. Maximum width and length of oocytes were measured to the nearest of 0.01 mm using an ocular micrometer (Ziess model SV 6). The relationship between egg number and body size was calculated by $F = aL^b$ (Bagenal & Braum

1978; Elliott 1995). One-way ANOVA was used to test for significant differences of the reproductive indices at different months.

Histological study

The histological preparations were made according to Bancroft & Stevens (1990): ovary and testis were dehydrated in alcohol, cleared in xylene, embedded in paraffin wax at 56°C melting point, sectioned at 5-7 μm thickness, and then the sections were stained by Hemotoxylin and Eosin (H & E) staining method.

RESULTS

Size range, population structure and Length-weight relationship

Morphometric characters of male and female specimens are given in Table 1. During this investigation, a total number of 355 fish specimens of *C. mandica* ranging 55.90 to 213 mm (SE = 30.04) in total length and 45.80 to 193 mm (SE = 25.45) in standard length were collected, of which 253 were males and 102 were females, giving a ratio of 2.5:1 which was significantly different ($p < 0.05$) from the expected ratio and significantly male biased (Chi square = 64.228, $p < 0.001$). The number of males were more than females in all months (Fig. 2). There was a significant relationship with a high regression coefficient ($r^3 = 0.991$, $P < 0.001$) between the length and weight of the fish (Table 2). There was no significant difference in the parameters b and a between sexes. However, the parameter b for male specimens was comparatively slightly higher than for females.

Table 1. Few measured morphometric character of *Capoeta mandica* specimens collected from the Rudbal River, Mond River drainage, Persis basin, Iran.

Measured character	Number	Min (mm)	Max (mm)	Mean (mm)	Standard deviation
Total length	355	55.9	213	118.38	30.04
Fork length	355	52.4	202	109.42	28.03
Standard length	355	45.8	193	98.30	25.45
Head length	355	13	42	24.89	5.72
Head depth	355	9.3	31.20	18.40	4.06
Max body depth	355	11.4	44.50	24.42	5.91
Min body depth	355	6.25	27.30	11.96	2.92

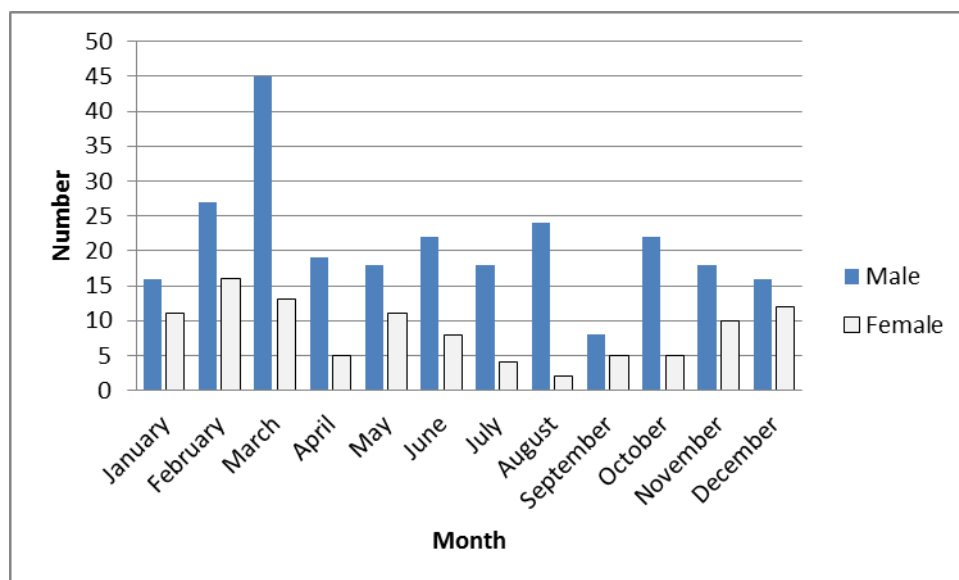


Fig. 2. Number of male and female specimen of *C. mandica* in different months.

Table 2. Length-weight relationship of *C. mandica* in different months collected from the Rudbal River, Mond River drainage, Persis basin. *r*: correlation coefficient, *r*²: coefficient of determination, *a* and *b*: constant of regression equation.

Month	Gender	<i>r</i>	<i>r</i> ²	Log. <i>a</i>	<i>b</i>	<i>F</i>	<i>p</i>
1	Male	0.99	0.99	-4.11	2.72	2205.94	0.000
	Female	0.97	0.94	-3.75	2.51	55.08	0.005
2	Male	0.99	0.98	-4.01	2.65	1572.23	0.000
	Female	0.99	0.99	-4.79	3.06	945.22	0.000
3	Male	0.99	0.98	-3.98	2.64	1755.46	0.000
	Female	0.98	0.96	-3.88	2.59	3612.70	0.000
4	Male	0.96	0.92	-4.43	2.87	210.08	0.000
	Female	0.99	0.98	-4.94	3.13	125.77	0.008
5	Male	0.98	0.97	-4.20	2.76	886.03	0.000
	Female	1.00	1.00	-2.92	2.10	.	.
6	Male	0.97	0.95	-3.67	2.49	114.13	0.000
	Female	0.98	0.96	-6.63	3.99	85.12	0.003
7	Male	0.99	0.98	-3.99	2.64	981.83	0.000
	Female	0.99	0.99	-3.72	2.50	412.73	0.000
8	Male	0.98	0.97	-4.27	2.78	743.81	0.000
	Female	0.98	0.97	-3.90	2.61	313.81	0.000
9	Male	0.99	0.99	-4.17	2.73	2051.83	0.000
	Female	0.99	0.99	-4.27	2.77	1050.95	0.000
10	Male	0.93	0.87	-4.03	2.65	99.43	0.000
	Female	0.99	0.98	-4.35	2.82	447.28	0.000
11	Male	0.98	0.97	-4.35	2.82	1104.50	0.000
	Female	0.99	0.98	-3.84	2.58	802.48	0.000
12	Male	0.98	0.97	-4.09	2.71	1660.98	0.000
	Female	0.99	0.99	-4.10	2.71	1556.15	0.000

Gonadosomatic index, K condition factor, and spawning periods

There was a significant difference in the GSI value of male and female in different months (ANOVA, $p < 0.001$). The female GSI increased from March through May, peaking in the

middle of spring and decreased significantly in June, indicating that the spawning falls after May. In the case of males, the GSI increased from December through April and then decreased in May and June (Fig. 3). MGSI decreased gradually from February through

March, while increased from April through May, exhibiting significant decrease in June and gradually increase after that. MGSI in males gradually increased from December through February and significantly in March and April, while then decreased from May through August in male specimens (Fig. 4). In females and males, the DI increased from February through April, with a clear fall during May and June (Fig. 5). In both sexes, condition factor had a peak in April. From December through February, it showed an increasing

pattern. In the case of males, the condition factor was the lowest in January, while increased up until April (Fig. 6). We observed significant difference between male and female K index in different months. This index affected by some factors such as sex and time of sampling.

In this study, this factor fluctuated in different months. The one-way ANOVA test showed that there was significant difference in K index values between males and females as well as between months ($p < 0.05$).

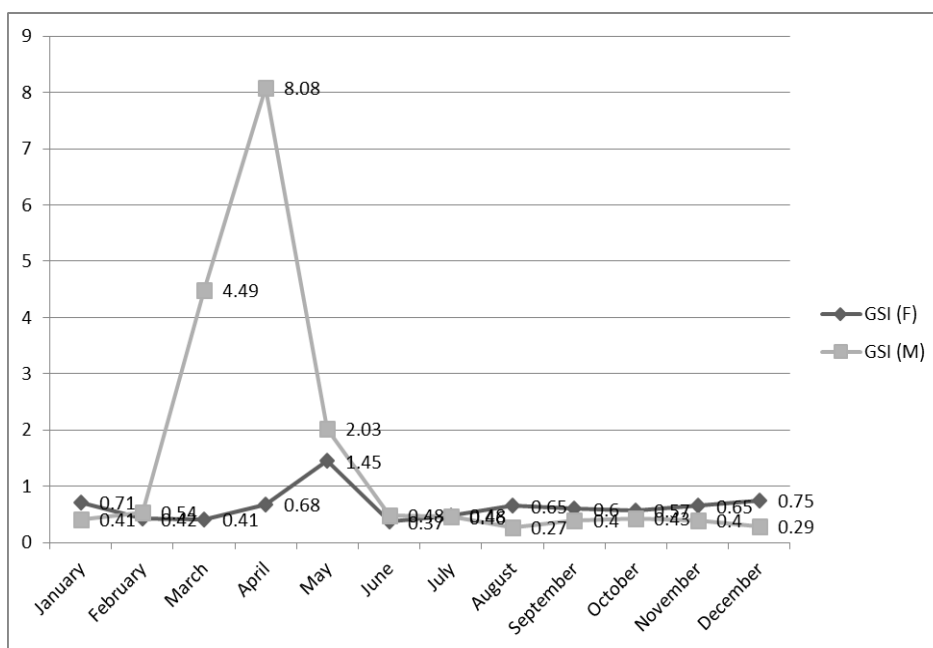


Fig. 3. Monthly variation of mean GSI in male and female specimens of *C. mandica*.

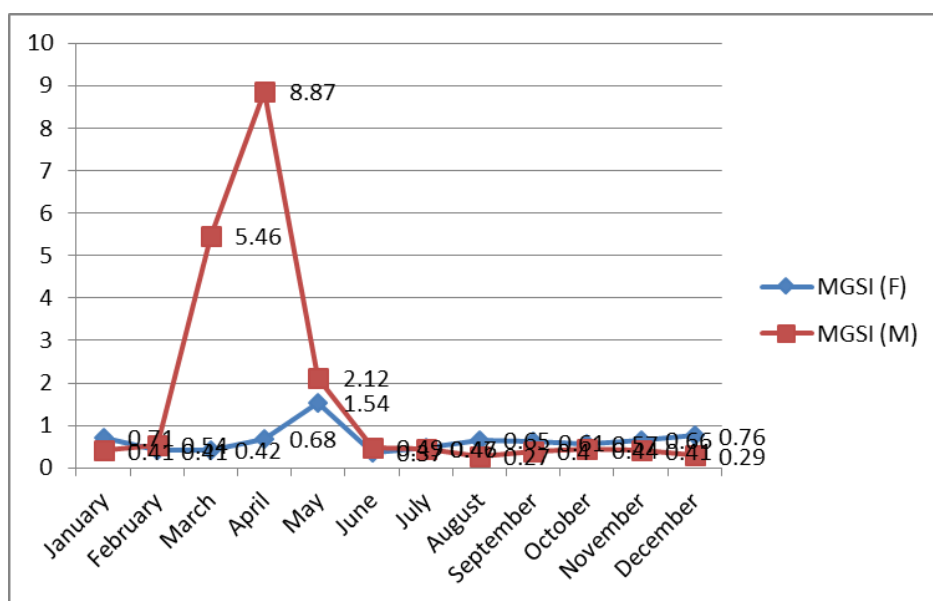


Fig. 4. Monthly variation of mean MGSI in male and female specimens of *C. mandica*.

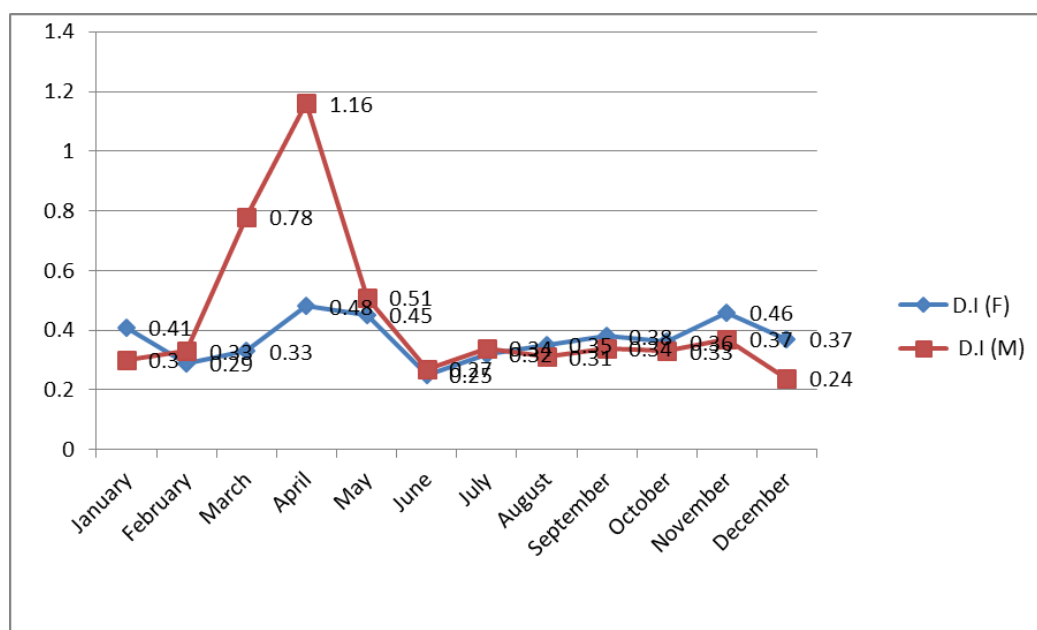


Fig. 5. Monthly variation of mean DI in male and female specimens of *C. mandica*.

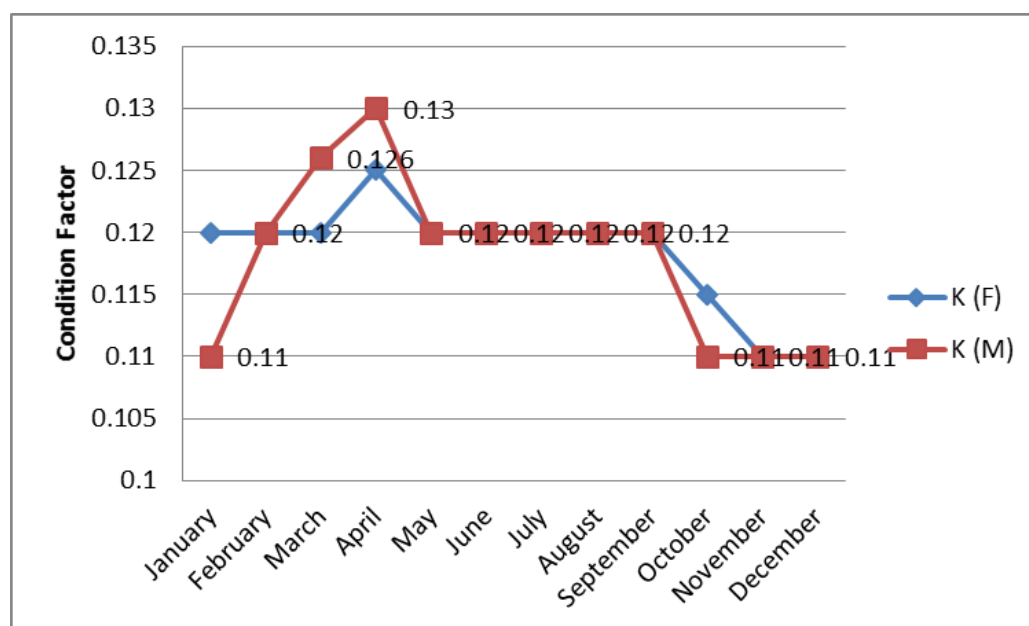


Fig. 6. Monthly variation of mean K in male and female specimens of *C. mandica*.

Gonadal development

The ovum diameters ranged from 0.04 to 1.31 mm, with the highest mean value in May, while the lowest in June. One-way ANOVA test showed significant difference between ova diameter in different months and also different stages ($p < 0.05$; Figs. 3, 7, and 8).

Female maturation stages

Based on the size and weight of the ovary, degree of occupation of the body cavity,

presence or absence of ripe oocytes, diameter of the oocytes, and histological observations, five maturation stages were described as follows (see Fig. 9):

Stage I (immature): Ovaries were small, transparent and thread-like attached to the vertebral column. Oocytes were not visible to the naked eye having mean diameter 0.08 mm. Small, round and transparent oocytes with a central nucleus were observed in histological sections of ovaries having basophilic cytoplasm

and an acidophilic nucleus. The ratio of nucleus to cytoplasm volume was high.

Stage II (developing): Ovaries were elongated and spindle shaped. Small yellow and transparent oocytes with an average diameter of 0.12 mm were not visible to the naked eye. Histological sections showed that the developing oocytes exhibit an acidophilic nucleus and a weak basophilic cytoplasm characterized by small lipid droplets. Very thin follicular layer appeared around the oocyte. Large peripheral nucleoli were distinguished adjoining the nuclear envelope and abundant nucleolus. This stage is called a returning stage, because after spawning, the ovaries return to this stage to start oogenesis.

Stage III (maturation): Ovaries were large, increased in weight and its color changed to yellowish because of the accumulation of yolk materials. Yellow large and densely packed oocytes were obviously visible.

Formation of yolk granules was distinguished. The mean diameter of oocytes was 0.16 mm.

A thin zona radiata surrounded by cubic follicular cells and thecal layer was appeared in this stage.

Stage IV (ripe): Ovaries were yellowish, elongated and spindle shaped having large oocytes with mean diameter of 0.21 mm.

The weight of gonads increased but not achieved their maximum amount. Blood capillaries were observed in the stroma of the ovary, as well as increase in follicular sizes, density of yolk granules and thickness of zona radiata and also decrease in the ratio of nucleus to cytoplasm. The follicular cells shape changed from cuboid to pavement state.

Stage V (spawning): Ovaries were yellow and occupied most of the body cavity.

They had achieved their maximum weight. Large, yellow and dark oocytes, full of yolk with an average diameter of 0.59 mm were distinguishable with loosely connection to each other. Ovaries were characterized by dense yolk and large lipid granules. Oocytes were released by slight pressure.

The zona radiata was completely thin and had separated from the follicular layers; thecal layer

was clearly distinguishable. Blood vessels were clearly observed in the stroma of the ovary.

Male maturation stages

The position of the testes in the males was the same as ovary. The main histological characteristics corresponding to the maturation stages, is the spermatogenesis process. Five spermatogenic cells were diagnosed as a) Spermatogonia: the largest spermatogenic cell, with clear cytoplasm and large nucleus, b) primary spermatocytes: nuclei were densely packed, c) secondary spermatocytes: smaller than primary spermatocytes, d) Spermatids: smaller in size than secondary spermatocytes and possess dense nuclei and finally (e) Spermatozoa: the smallest and densest spermatogenic cells. Based on macroscopic and microscopic observations, five stages of maturation were distinguished for males as follow (see Fig. 10):

Stage I (immature): Testis was thread-like, thin and whitish cream in color. Spermatogonia and primary spermatocytes were the dominant cells in this stage. Spermatogonia had a light cytoplasm and a large nucleus. Some secondary spermatocytes having basophilic cytoplasm were also observed.

Stage II (developing): Testes were opaque and darker than in former stage. Spermatogonia were reduced. Primary and secondary spermatocytes and also some spermatids were observed in the tubules.

Stage III (maturation): The testes were dark cream in color and a little elongated flat. In this stage, the number of large cells was decreased, while the number of small cells (spermatids and sperms) was increased progressively.

Stage IV (ripe): The male gonads were flat. Tubules were characterized by having secondary spermatocytes, large numbers of spermatids and spermatozoa.

Stage V (spawning): Testis was large flat in structure occupying the whole body cavity. Milt was running by employing slight pressure on the abdominal region of the fish. The testes had well-defined lobules with a large number of spermatids and spermatozoa on their inner center.

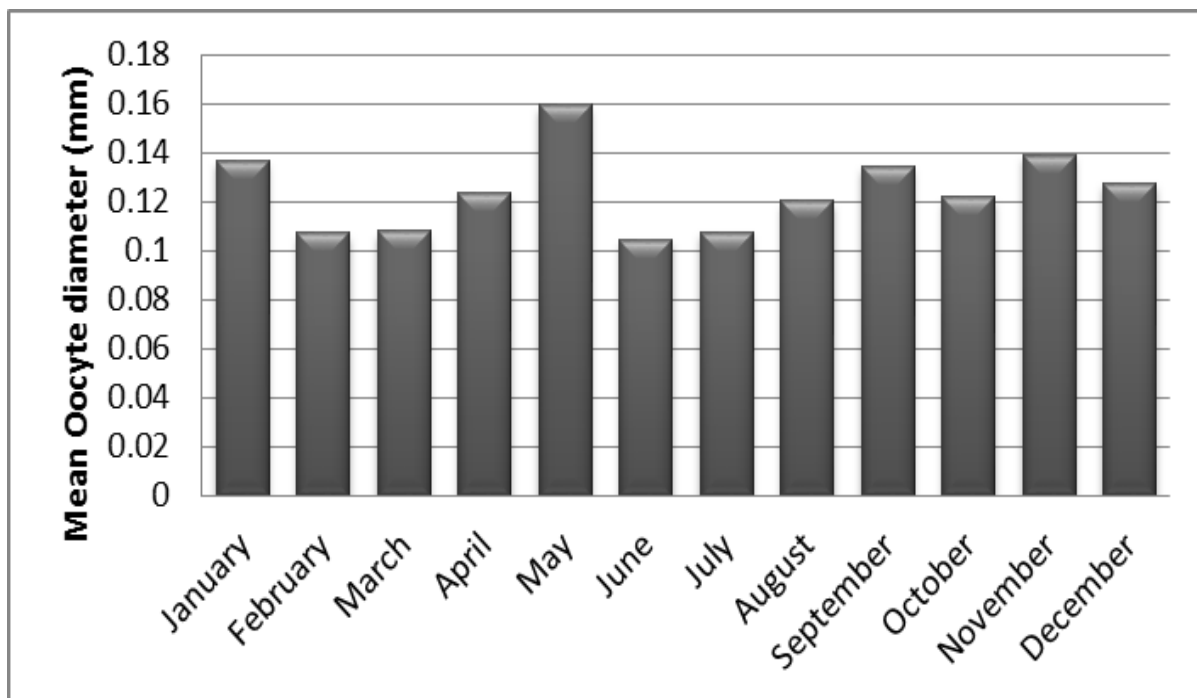


Fig. 7. Mean oocyte diameter (mm) by month for *C. mandica*.

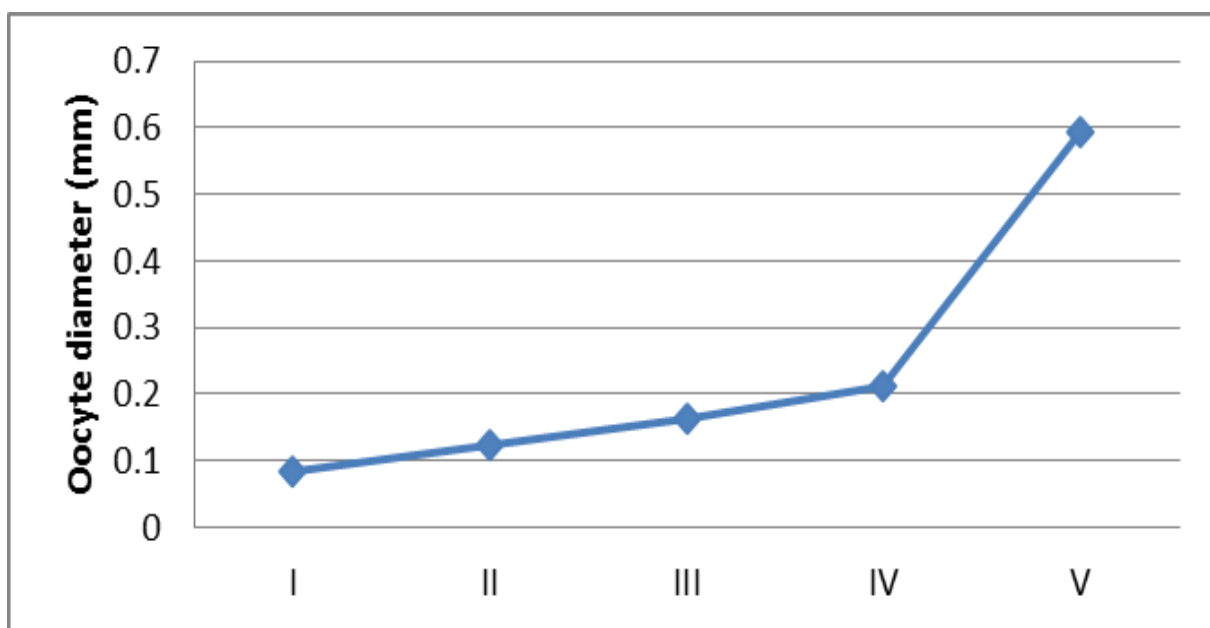


Fig. 8. Mean Ova diameter in different maturity stages of *C. mandica*.

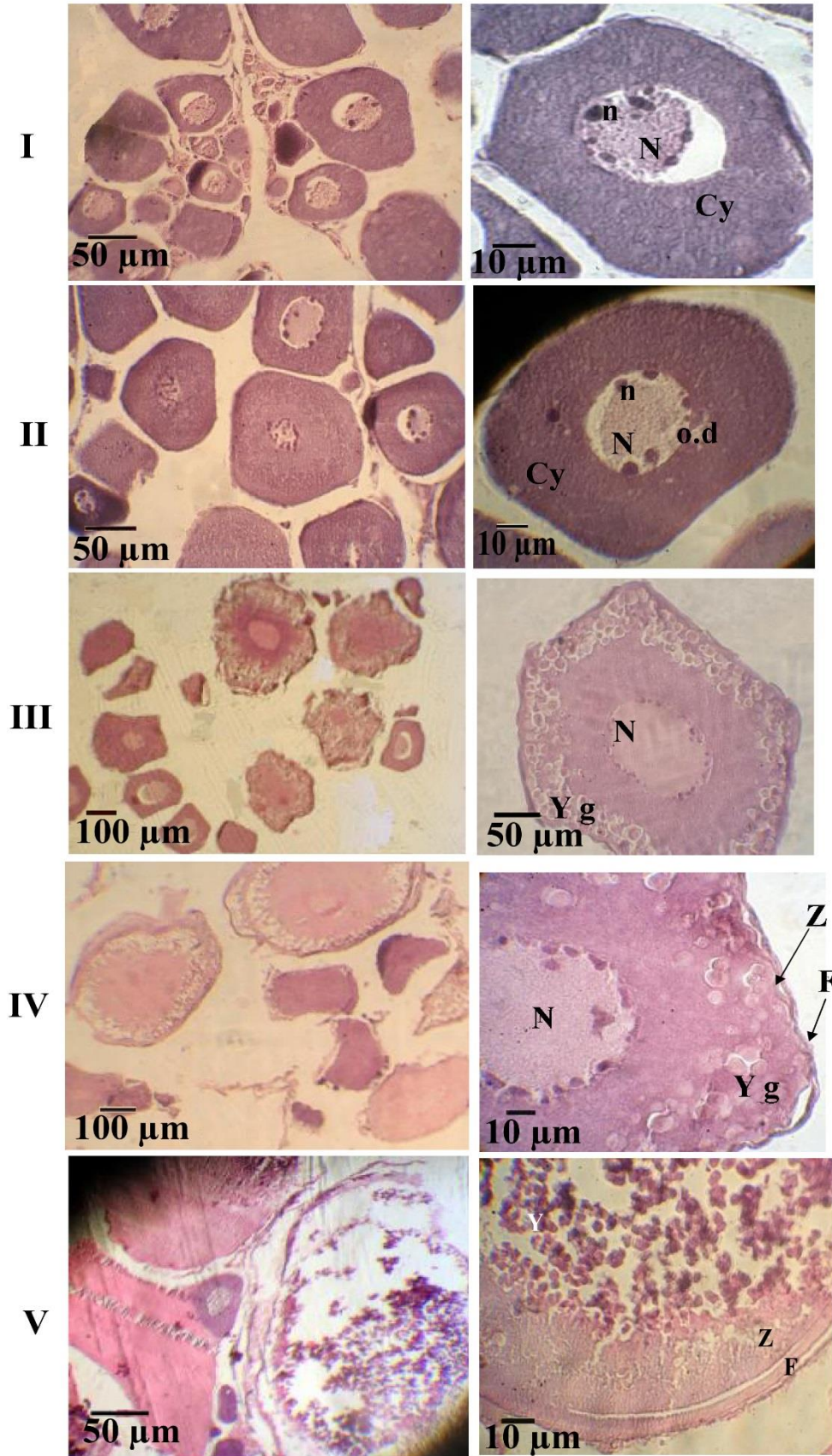


Fig. 9. Histological profiles of the female maturity stages in *C. manduca*, Persis basin, Southwestern Iran. (I) Immature stage, (II) Developing stage, (III) Mature stages, (IV) Ripe stage, (V) Spawning stage. N: Nucleus, n: Nucleolus, Cy: Cytoplasm, Od: Oil droplets, F: Follicular layer, YG: Yolk granules, Y: Yolk.

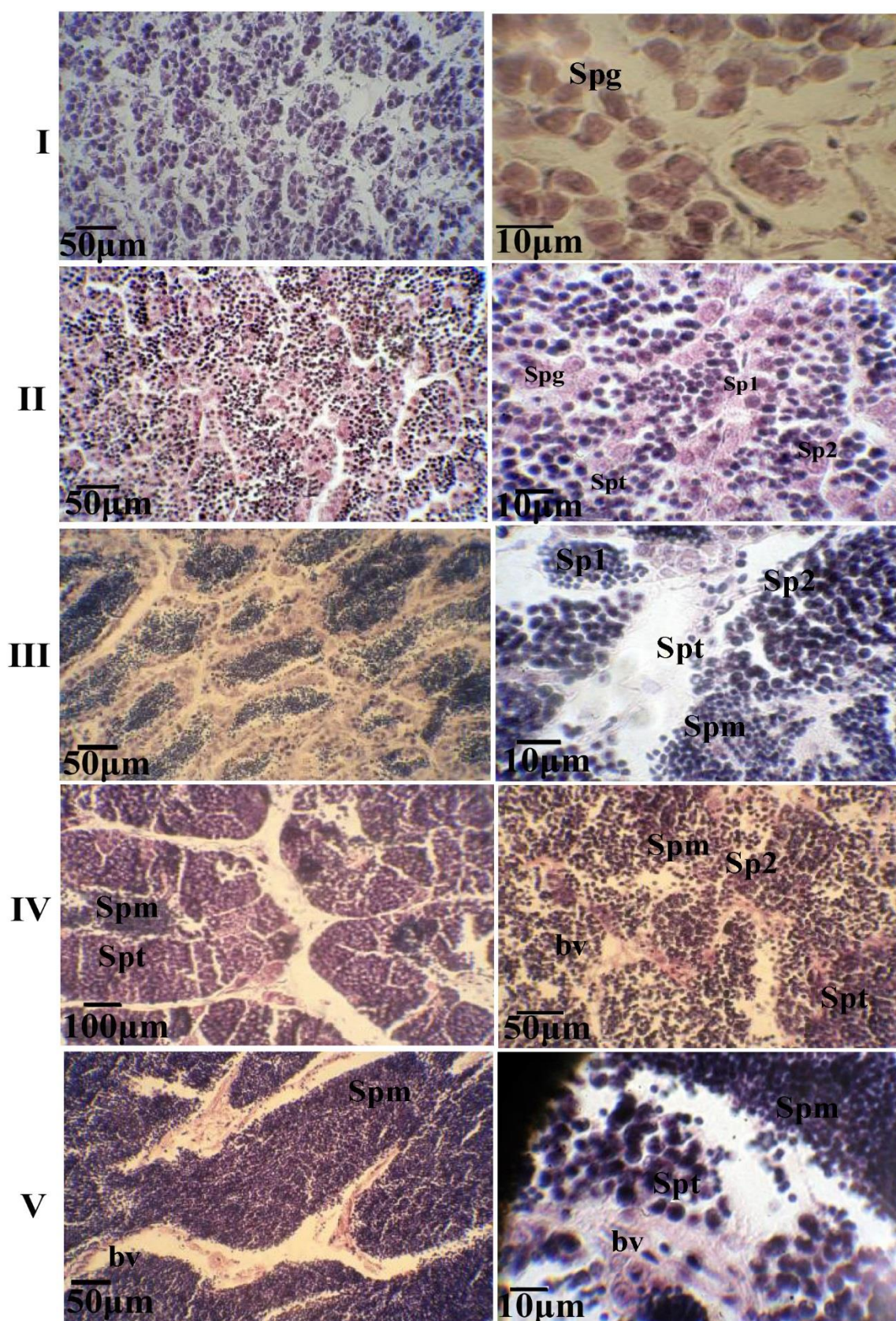


Fig. 10. Histological profiles of the male maturity stages in *C. mandica*, Persis basin, Southwestern Iran. (I) Immature stage, (II) Developing stage, (III) Mature stages, (IV) Ripe stage, (V) Spawning stage. Spg: Spermatogonium, Sp1: Primary Spermatocyte, Sp2: Secondary Spermatocyte, Spt: Sperm, Spm: Sperm, bv: Blood vessel.

Table 3. Ova number and diameter (mm) in different maturity stages of *C. mandica* collected from the Rudbal River, Mond River drainage, Persis basin.

Stage	Number	Min	Max	Mean	SD
1	599	0.04	0.14	0.08	0.01
2	3080	0.05	0.21	0.12	0.02
3	281	0.08	0.26	0.16	0.03
4	40	0.13	0.42	0.21	0.06
5	41	0.08	1.31	0.59	0.37

DISCUSSION

The present study provided the details on life history traits and morpho-histology of gonads in an endemic cyprinid fish species, *Capoeta mandica* in Southern Iran. Our findings indicated that based on GSI, MGSI, mean egg diameter, K, DI and developmental stages of the ovaries, *C. mandica* spawns during May and June. The highest GSI values correspond to when the gonads were at ripe and ripe running stages (IV and V), while the lowest values indicate totally spent stage or starting developing stage (recovering stages) (Mirghiyasi *et al.* 2016). The results of Duman (2004) showed that *Capoeta trutta* spawned between June and July, in consistent with our results for *C. mandica*. It has already been pointed out that several factors including physicochemical, bio-ecological and climatic factors might have direct and indirect effects on fish, promoting sexual maturity of fish (see Nicolsky 1963; Akgul 1986; Ekmekci 1996; Ekmekci & Ozeren 2003) which could also affect reproductive success of *C. mandica*.

In females, mean value of GSI and MGSI were the highest at May supported by histomorphological analyses. Similar increasing was determined for egg diameter value. The ova diameter variation is probably one of the important pieces of evidence used in determination of fish reproductive strategy (Tomasini *et al.* 1996). Dopeikar *et al.* (2015) also observed highest egg diameter in April (2.25 mm) when GSI was the highest in both sexes of a cyprinid fish, *Barbus lacerta*.

The monthly variations in the GSI were highly associated with the seasonality of the maturity stages assigned macroscopically, as found by Morato *et al.* (2003). In our study, a gradually increase in the GSI of females was observed from June through December, which indicated the appearance of pre-spawning individuals. The GSI peaked in spring, corresponding to the first observation of spawning individuals. In the case of males, the GSI increase was from December through April, while decreased in May, showing that male specimens likely start spawning earlier than females. GSI of both sexes of *Capoeta damascina* was reported the least in August and September and then rose gradually from these months to reach maximum levels in June, suggesting spawning of this species occurs between May and July in the Gheshlagh Reservoir in Tigris basin (Bahrami Kamangar *et al.* 2015). Apparently, both species spawn nearly simultaneously. However the time of peak spawning is different. Such a nearly lengthy breeding season is a type of adaptation by some populations, which lives in an unstable habitat, to environmental conditions (Miller 1979). Spawning period during June-July was reported for *Alburnoides* sp. with high value of the GSI in June and its sharp decrease in August (Seifali *et al.* 2012). Abedi *et al.* (2011) also reported that the GSI of female *Garra rufa* increased from November through May, peaking in mid - spring, then decreasing slowly from the late May through November, showing prolonged reproductive activity.

The gonad weight has an obviously better correlation with reproductive capacity than with the body weight. Hence, the DI could also be used for determining the spawning season, sexual maturity, and the spawning frequency of fish. The DI involves only the data related to the sexual organs, which are easy for interpretation and calculation and also provide a narrow range of index if the gonad weight is very low or very high (Esmaili & Shiva 2006). According to our results, in females and males, the DI increased from February through April, with a clear fall during May and June suggesting that Mond spotted barb spawns during May and June consistent with other indices. The condition factor of fishes has been reported to be influenced by a number of factors, of those, gonad size in the spawning period is one of the most important factor (see Murphy & Willis 1996; Alhassan *et al.* 2015).

The condition factor of *C. mandica* was high at the beginning of the spawning season when more ripe eggs were present, while decreased slowly during the long spawning period. This shows the effect of gonad weight on the k value. In fact, releasing the oocytes in females and sperms in males decreased the weight of fish, so they became thin and this decreased the k amount. An increase or decrease in condition factor could be due to availability of food, spawning, stress, changes in temperature, pH or pollution of water, and so on (Turkmen *et al.* 1999; Erdoğan *et al.* 2002).

The length-weight relationship in fishes is affected by a number of factors, including season, habitat, gonad maturity, sex, diet and stomach fullness, health, and preservation techniques (Tesch 1968). Results of the present study indicated that the b value was about 3, concluding that the fish growth is isometric, and that its shape does not change by growth. In general, the value of b lies between 2.5 and 4 and can be varied as a result of changes in fish shape, season, age, and food availability (Ricker 1979).

In the present study, the value of b remained within the expected range. Abedi *et al.* (2011) reported that the growth of *Garra rufa* is

isometric in the Armand rocky stream (one of the branches of Karun River, Iran) and that b value is about 3, in consistent with our study. The length-weight relationships study for *Capoeta damascina* in Gheshlagh reservoir (Tigris basin) exhibited a negative allometric growth (Bahrami Kamangar *et al.* 2015). The overall sex ratio for *C. mandica* displayed that male significantly biased and deviated from the hypothetical distribution of 1:1, consistent with that reported for *Barbus lacerta* (Dopeikar *et al.* 2015) while in contrast to *Capoeta damascina* (Bahrami Kamangar *et al.* 2015). Although, this ratio is close to 1:1 in many species (e.g, *Garra rufa*; Abedi *et al.* 2011) but it may be far from this because of many factors such as growing differences between sexes, differences in sampling equipment, fishing situations, reproductive strategies, predators, environmental conditions, mechanism of population regulation, etc. (Bao-Shan *et al.* 2012; Gupta & Banerjee 2013). Sex ratio biased toward males in *C. mandica*, may be related to its reproductive strategy where eggs become fertilized by several males. In conclusion, this study has provided information on the reproductive cycle of Mond spotted barb, *Capoeta mandica*, an endemic cyprinid fish in the Persis basin which contribute baseline data towards management ecology and conservation of the species. The overall sex ratio for *C. mandica* was significantly male - biased in Persis basin. Based on the different studied indices, it was concluded that the Mond spotted barb spawns during May and June.

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بررسی ویژگی‌های چرخه زندگی و بافت شناسی گناد یک کپورماهی بومزاد، سیاه ماهی خالدار (*Capoeta mandica*)، از جنوب ایران

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چکیده

در این مطالعه زیست شناسی تولید مثل و تکوین گنادهای سیاه ماهی خالدار که یک کپورماهی بومزاد ایران است با نمونه برداری های ماهیانه از بهمن ۱۳۸۵ تا اسفند ۱۳۸۶ در جنوب ایران و بر اساس بررسی های میکروسکوپی و ماکروسکوپی مورد مطالعه قرار گرفت. در کل تعداد ۳۳۵ نمونه ماهی شامل ۲۵۳ نمونه نر و ۱۰۲ نمونه ماده با استفاده از دستگاه شوک دهنده الکتریکی جمع‌آوری گردید. بررسی تعداد ماهیان نر و ماده نشان داد که به ازای هر ماهی ماده ۲/۵ ماهی نر وجود دارد و این نسبت به طور معنی داری با نسبت جنسی مورد انتظار متفاوت است. بر اساس اندازه، شکل و وزن گنادها، میزان اشغال حفره شکمی، حضور و عدم حضور اسپرم یا تخمک رسیده، قطر تخمک‌های درون تخمدان و مطالعات بافت شناسی پنج مرحله بلوغ برای این ماهی تشخیص داده شد که با شاخص های تولید مثلی مرتبط می باشد. قطر تخمک‌ها در سیاه ماهی خالدار بین ۰/۰۴ تا ۱/۳۱ میلی متر متغیر بوده و بیشترین میانگین آن در ماه اردیبهشت اندازه گیری شد. بیشترین میزان ضریب کیفیت در افراد نر و ماده در ماه فروردین مشاهده شد. میزان شاخص GSI در افراد ماده از اسفند تا اردیبهشت ماه افزایش و سپس به طور معنی داری در خرداد ماه کاهش نشان داد. بر اساس مراحل بلوغ گناد، شاخص GSI، شاخص گنادی-بدنی تغییر یافته، میانگین قطر تخمک‌ها، ضریب کیفیت و شاخص دوبریال مشخص شد که سیاه ماهی مند در طول بهار و در ماه های اردیبهشت و خرداد تخم ریزی می کند. این نتایج می تواند اطلاعات مفیدی برای مدیریت و حفاظت از این گونه بومزاد در اختیار قرار دهد.

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