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Effects of water temperature and migration time on some fecundity indices and fertilization rate of female Kutum, *Rutilus frisii kutum*, migratory to Shiroud River in the southwest Caspian Sea

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ABSTRACT

Kutum, *Rutilus frisii kutum*, is an commercially important fish in the Caspian Sea. The fish enters the rivers leading to the Caspian Sea for spawning. One of these rivers is Shiroud River. In the artificial propagation process of Kutum, different factors are involved in quality and quantity of female broods eggs and also in the best temperature and migration time for spawning. The influence of temperature and migration time on some fecundity indices and fertilization rate of female kutum in Shiroud River was studied in the spawning season. In this study, 90 individual females were studied from February to May 2007. Averages of total length, weight and age were 43.26 cm, 832.08 g, and 4.41 respectively. Results showed that maximum egg diameter (1.86 mm), number of eggs per each gram of body weight (309.12), relative fecundity (56.21) and fertilization rate (95.82 %) were obtained from 5 to 20 April (15.95°C). Maximum ovary weight (201.00 g) and absolute fecundity of eggs (49987.18) were obtained from 6 to 20 March (17.74°C). Relationship between temperature and migration time and fecundity indices was linear and weak.

Keywords: *Artificial propagation, migration, Rutilus frisii kutum,Fish.*

INTRODUCTION

Kutum, *Rutilus frisii kutum*, is a unique, endemic species and has a high ecological and commercial food values (Razavi Sayad, 1999). In recent years due to uncontrolled fishing, increased pollution, river bed degradation, migration and etc. its numbers and reproduction has been reduced (Coad, 1980; Ghani Nejhad *et al.*, 2000; Razavi Sayad, 1999; Ralonde & Walczak 1971). Because of the mentioned problems the artificial propagation of Kutum has been on the agenda of the Iranian Fisheries Organization (Razavi Sayad, 1999).

Annually in almost 15 adjacent rivers inflowing to the Caspian Sea, Shiroud River is the main and most important river for Kutum artificial spawning. Shiroud River is formed by the joining of two main streams flowing in two directions rather then parallel to each other. The River runs for almost 30 km and empties on an average about 125.29 million cubic meters of water into the Sea annually. This river is one of the important and primary rivers in the southern Caspian Sea for different fish species which live in or migrate to it and is the main spawning ground for Kutum during their spawning migration (Abu, 1994).

Kutum is an anadromous fish and has a lithophilus spawning behavior. The fish lay their eggs between late winter to mid spring at temperatures between 8-24°C on gravel substrate and bottom of rivers. The optimal temperature for spawning of this fish is 13 to 14 ^{o}C (Abdoli & Naderi 2009). Studying the relationship between an organism and habitat characteristics are most important cues for selecting the

habitat by aquatic organisms such as fish. In this regard temperature and migration time and its duration are very important and noticeable. This study was carried out to study the effect of water temperature and migration time on the artificial spawning efficiency of kutum during the spawning season between March to May 2008.

MATERIALS and METHODS

In this study 90 pieces of Kutum were caught by cast netting in estuaries of Shiroud River. Sampling period of Kutum was carried out according to the fish migration time to Shiroud River which was divided into four periods: 1) 6 to 20 March; 2) 21 March to 4 April; 3) 5 April to 20 April and 4) 21 April to 5 May. Eggs were collected from the caught fish without hormone injections and egg removal took place with slight pressure on the abdominal area. The egg properties such as egg weight and diameter were measured. Then the collected eggs from each female were fertilized by sperm stripped from 2 to 3 males. Several seconds after adding the sperm, water was added to the solution to increase fertilization rate. Eggs were rinsed during incubation to remove adhesion. Loss of adhesion and hydration process was completed inside Seth Green incubators in the River (Yousefian & Mosavi 2008). During this study, diameter of eggs, the number of hydrated eggs per gram of eggs, absolute fecundity, relative fecundity and fertilization rates were calculated for every batch of eggs subjected to artificial reproduction. Also on selected females, fork length was measured using a 1 mm plywood, body and eggs weight was measured by digital scale up to the nearest 0.01 grams. The age of fish was determined using scales from the mid trunk region between rays and chest (Parafkandeh Haghighi, 2000).

To determine fertilization rate, eggs were supplemented with diluted acetic acid solution and distilled water in a petri dish 5 hr after fertilization when fertilized eggs reached stage 8 of cell division. After separating transparent and fertilized eggs from translucent and dead eggs, the number of fertilized eggs was calculated according to the following formula (Razavi Sayad, 1995).

Fertilization rate= (number of fertilized eggs/total eggs number) ×100

Data collected were analyzed using SPSS 10.0.13 (Microsoft Office). one way ANOVA test and also nonparametric tests such as Kruskal - Wallis test, and Mann- Whitney U test were used for comparing data with a confidence rate of 95% ($P<0.05$) and the regression relationships were used to determine the correlation of different factors.

RESULTS

Biometric measurements of migratory female Kutum to Shiroud River are shown in Table 1. The average temperature was 17.14°C in the first sampling period (6 to 20 March), 17.39°C in the second phase (21 March to 4 April), 15.95°C in the third phase (5 to 20 April), and 18.83°C in the fourth stage of sampling (21 April to 5 May).

Table 1. Bioassay of female Kutum migratory to the Shiroud River

parameter	Weight (g) Range	Total length (cm) Range	Fork length (cm) Range	Age Range
mean ± SD	832.08±368.19	43.26±5.82	39.20±5.40	4.41±0.93
	315-2216	32.4-59.6	29.1-54.7	3-7

Effects of migration time on some parameters of egg fertilization have been shown in Table 2 and that of correlation rate of temperature and migration time on fertilization in Table 3. The highest average egg extract (201.00 g), egg diameter after formalin handling (2.05 mm) and absolute fecundity (49987.18 eggs) were obtained in the first period. According to the one-way ANOVA test, significant differences were observed ($P<0.05$) between different times of the mean egg diameter after formalin handling. Duncan test showed correlation between these times and also correlation in terms of egg diameter after formalin hardness. There were also significant differences between these treatments: (1-2, 2-3 and 2-4). According to the Kruskal-Wallis test, the mean weight of eggs obtained from different times and also absolute fecundity were not significantly different ($P>0.05$). Mann-Whitney U test

showed significant differences between the different batches of artificial reproduction (batch 1 and batch 4, and also batch 1 and batch 3) in terms of average weight of eggs. Batch 1 and batch 4 also differed significantly in average absolute fecundity.
In this study, the highest average hydrated egg diameter (2.65 mm) was obtained in the second and the third batches. There were significant differences between different batches in terms of average hydrated egg diameter (One-Way ANOVA test, $P<0.05$). Duncan test showed significant differences in average diameter of hydrated eggs between batches 1 and 2, 1 and 3 and 1 also and 4.

Table 2. Effects of migration time on related parameters of egg fertilization

Parameter	Migration Time			
	6 to 20 March	21 March to 4 April	5 April to 20 April	21 April to 5 May
egg weight (g)	201.00±97.69	151.27±102.87	136.13±57.582	136.94±67.128
Range	66-387	36-416	53-242	52-312
egg diameter (mm)	1.57±1.83	1.83±0.10	1.86±0.09	1.85±0.09
Range	1.30-1.94	1.66-1.99	1.67-2.04	1.65-2.00
hydrated egg diameter	2.19±0.24	2.58±0.19	2.65±0.17	2.65±0.10
(mm)Range	2.56-3.24	2.27-2.85	2.37-3.02	2.39-22.82
Egg diameter after formalin exposure	2.05±0.13	1.86±0.09	1.98±0.14	2.01±0.09
(mm)Range	1.8-2.2	1.7-2.0	1.7-2.4	1.8-22.2
number of dry eggs (g)	261.55±41.81	304.06±39.97	309.12±50.48	288.03±31.58
Range	199-333	233-362	231-446	217-364
number of hydrated	72.00±10.23	85.55±11.48	88.94±19.64	83.7±14.17
eggs (g)Range	61-89	71-105	59-145	39-131
absolute Fecundity	49987.18±20492.92	43036.00±22880.71	41121.70±17772.380	38372.54±`1636 4.42
Range	21959-89144	11903-96824	18550-86346	15935-79363
relative fecundity	46.91±8.45	51.00±11.44	56.21±26.79	49.17±7.45
Range	30-63	38-77	26-148	31-66
fertilization rate	86.55±17.52	11.61	95.82±5.43	93.44±6.97
Range	40-98	60-100	73-100	64-100

In the present study, the highest average of variables such as egg diameter (1.86 mm), number of dry eggs per gram (309.12), number of hydrated eggs per gram (88.94), relative fecundity (56.21) and fertilization rates (95.82) were achieved in the 5-20 April batch. Significant differences were observed between various batches for some factors such as average fertilization rate, number of dry eggs per gram eggs and number of hydrated eggs per gram eggs (Kruskal-Wallis test, $P<0.05$). Significant differences were also observed for the average number of dehydrated eggs per gram (between batches 1 and 2 and between batches 1 and 3) for average number of dehydrated eggs per gram (between batches 1 and 2, batches 1 and 3 and batches 1and 4) and also for the fertilization rate (between batches 1 and 2, batches 1and 3, batches 2 and 4 and batches 3 and 4) (Mann-Whithney U test, $P<0.05$). While the Kruskal-Wallis test showed no significant differences ($P>0.05$) in average diameter of eggs and relative fecundity between different batches time groups, , Mann-Whitney U test showed significant differences ($P<0.05$)between the batches 1 and 4 and batches 1 and 3 in terms of average egg diameter.

Table 3. Correlation between water temperature and migration time with parameters of fertilization.

parameter	water temperature	time of migration
egg weight (g)	0.083	0.236
egg diameter (mm)	0.007	0.264
hydrated egg diameter (mm)	0.095	0.291
after formalin exposed of egg diameter (mm)	0.038	0.07
number of dry eggs (g)	0.01	0.1
number of hydrated eggs (g)	0.04	0.158
absolute Fecundity	0.072	0.191
relative fecundity	0.057	0.014
fertilization rate	0.046	0.178

DISCUSSION

According to the results of this study, the average fertilization rate was 92.3%. Farabi *et al.* (2007) reported that fertilization rate of this species is 89% in Goharbaran River (in Iran). In another study, Golshahi and Morad Nejhad (2008) reported that it is 94.85 %.

In the study of Golshahi and Morad Nejhad (2008), the highest weight of eggs was obtained between 23 March and 2 April. In this study, the highest average weight of eggs was obtained between 6 and 20 March. Differences in results of the present study and these studies might be due to different time of beginning these studies and also different climatic conditions. In the study of Golshahi and Morad Nejhad (2008), the extraction of eggs begun from 12 to 23 April, more than 10 days earlier than the present study (2 to 12 April). In the present study, different results were achieved and there were no significant differences between the 4 periods which may be due to various conditions such as temperature and water flow of the river. The greatest number of migratory fish was observed in the last batch, but the weight of eggs harvested was less than that of the first and second batches. Golshahi and Morad Nejhad (2008) believed that constant meteorological and environmental parameters in the forth batch, compared with those recorded in the other batches, might be due to higher water temperature.

Water temperature showed low correlations with fish fecundity index and fertilization rates. These results show that while temperature is one of the most important causes of Kutum migration, other factors such as environmental and meteorological factors may collaborate with temperature and may have some influence on migration. Similar results were reported by Golshahi and Morad Nejhad (2008) in river Goharbaran and by Yousefian and Mosavi (2008) in Tajan River and Shiroud River on the effects of environmental cues in kutum migration to rivers. In this study, the absolute fecundity was 41370. Farabi *et al.* (2007) and Golshahi and Morad Nejhad (2008) also reported an absolute fecundity of 47600 and 40550 eggs in the third and forth batches of sampling, respectively.

Golshahi and Morad Nejhad (2008) found that although a higher number of eggs was produced between 12 and 23 April, the average weight of eggs was lower than the first and third periods (14 to 23 March and 2 to 12 April), respectively. These results were similar to the results of present study on migratory Kutum entering the Shiroud River. In those study maximum samples were collected during the fourth period, but the weight of eggs was lower than that of the first and second periods. The amount of eggs harvested in the forth period was similar to that of the third period of sampling.

It is evident from the results that during the period between 21 April and 5 May, environmental conditions for spawning were not effective enough, so Kutum could not successfully enter into the adjacent rivers. Golshahi and Morad Nejhad (2008) mentioned that rising in water temperature and meteorological factors could effect the migration of Kutum into these rivers. In this study, the maximum temperature was recorded in the fourth period of study.

It may therefore conclude that environmental cues for spawning of Kutum is related to different parameters, and will affect on artificial reproduction. Average fertilization rate, relative fecundity, eggs diameter before fertilization (dry eggs) and numbers of egg per gram eggs were in the maximum between 5 and 20 April, while average absolute fecundity and weight of extracted eggs were in their maximum range between 6 and 20 March and increased significantly.

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اثر درجه حرارت آب و زمان مهاجرت روی برخی از شاخص های هم آوری و درصد لقاح ماهی سفید (*Rutilus frisii kutum*) ماده مهاجر به رودخانه شیرود در جنوب غربی دریای خزر (ایران)

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چکیده

ماهی سفید (*Rutilus frisii kutum*) یکی از مهمترین ماهی استخوانی دریای خزر می‌باشد که جهت تخم‌ریزی به رودخانه‌های منتهی به دریای خزر از جمله رودخانه شیرود وارد می‌شود. در روند تکثیر مصنوعی ماهی سفید، عوامل متفاوتی دخیل می باشند از جمله کیفیت و کمیت مولدین ماده و تخمک‌های استحصالی از آنها و همچنین بهترین دما و زمان مهاجرت این گونه برای تخم‌ریزی می‌باشد. به همین دلیل تأثیر درجه حرارت آب و زمان مهاجرت بر روی برخی از شاخص های هم آوری و درصد لقاح در جمعیت مولدین مهاجر ماهی سفید به رودخانه شیرود مورد بررسی قرار گرفت. بدین منظور از اسفند ۱۳۸۶ تا اردیبهشت ۱۳۸۷، ۹۰ عدد مولد ماهی سفید ماده مورد بررسی قرارگرفتند. طبق بررسی‌های انجام گرفته میانگین طول کل ۴۳/۲۶ سانتی متر، میانگین وزن ۸۳۲/۰۸ گرم ، میانگین سن ۴/۴۱ بود. نتایج نشان داد که بیشترین میانگین قطر تخمک ۱/۸۶ میلی متر، تعداد در گرم تخم خشک ۳۰۹/۱۲، هم آوری نسبی ۵۶/۲۱ و درصد لقاح ۹۵/۸۲ درصد مربوط به نیمه دوم فروردین ماه (۱۵/۹۵ درجه سانتی گراد) بود. بیشترین میانگین وزن تخمک استحصالی ۲۰۱/۰۰ گرم و هم آوری مطلق ۴۹۹۸۷/۱۸ عدد تخمک مربوط به نیمه دوم اسفند ماه (۱۷/۱۴ درجه سانتی گراد) بود. رابطه ضعیفی بین درجه حرارت و زمان مهاجرت با شاخص های هم آوری و درصد لقاح وجود داشت.