Int Aquat Res (2010) 2: 131-133
ISSN 2008-4935
Available online at www.intelaquares.com

International
Aquatic Research

Short communication

Serum steroid hormones in kutum *Rutilus frisii kutum* during spawning season

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Received:2 June 2010; Accepted:11 July 2010

Abstract

The objective of this study was to measure the concentrations of steroid hormones (testosterone, 17β-estradiol and progesterone) in kutum *Rutilus frisii kutum* during spawning period. Serum testosterone level in gravid fish (14.52 ± 0.03 ng/ml) was higher than that (0.54 ± 0.11 ng/ml) in ripe fish ($P < 0.05$). Levels of 17β-estradiol and progesterone were lower than 1 ng/ml with no significant difference between ripe and gravid fish ($P > 0.05$). The hormonal profile indicates that gravid fish are ready to ovulate.

Keywords: Steroid hormones, Spawning period, *Rutilus frisii kutum*

Introduction

The kutum Rutilus frisii kutum exists in the southern waters of the Caspian Sea and is a commercial fish with high market acceptance. Kutum is an anadromous and litophylous species which spawns on gravel in late winter to mid spring at water temperatures between 8 and 24 ºC with the optimum temperature of 13-14 ºC (Abdoli and Naderi 2009). Natural stock of kutum has been declined in the past decades due to damming of rivers where it used to spawn and overfishing. Capture of wild breeders during upstream reproductive migration is currently practiced by hatcheries for controlled breeding.

Serum steroid hormones such as testosterone, 17β-estradiol and progesterone have been investigated to gain insight into the mechanisms involved in reproductive behavior, gametogenesis and gonadal steroidogenesis (Scott et al. 1983; Rinchard et al. 1997, 2001; Sen et al. 2002; Lee et al. 2002; Suresh et al. 2008). Information on the reproductive physiology of kutum during spawning period could help in better understanding of its reproduction and controlled breeding management.

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Materials and methods

Experimental fish

Fish were caught at Shirood River mouth (northern Iran) by cast net of 2 cm mesh size. Five samplings from 11 March until 1 May 2009 took place and they were classified into two groups as described by Patterson et al. (2004): 1- Ripe fish: which had releasing (ovulating) gametes when caught in the river, and 2- Gravid fish: whose gametes are attached tightly to the body cavity. Twenty five gravid (weight: 1070 ± 56.18 g, length: 44.9 ± 4 cm) and 22 ripe (weight: 1123 ± 45 g, length: 45.8 ± 3.4 cm) fish were used in this study.

Blood collection and steroid assay

Fish were anesthetized with MS_{222} (100 ppm) for one minute. Thereafter blood was collected from the caudal vein in a 1 ml vial and kept in ice until centrifugation at 3000 ×g for 10 minutes. Separated serum was stored at -20 ºC until steroid assay. Serum steroids (testosterone, 17β-estradiol and progesterone) were measured by radioimmunoassay (RIA) according to the method described by Suresh et al. (2008).

Statistical analysis

Differences in the concentrations of steroid hormones between ripe and gravid fish were statistically analyzed by unpaired *t*-test. The differences were considered significant at $P < 0.05$. All data are expressed as mean ± SD using SPSS 16.

Results and discussion

Testosterone concentration in gravid fish was higher than that in ready-to-spawn or ripe fish (Table 1, $P < 0.05$). Higher concentrations of this steroid in gravid fishes were also reported in rainbow trout Oncorhynchus mykiss (Scott et al. 1983), walleye Stizostedion vitreum (Pankhurst 1986) and whitefish Coregonus clupeaformis, (Rinchard et al. 2001). Higher levels of this steroid in gravid fish species are attributed to the shift in the activity of key enzyme involved in steroidogenesis as follicles are prepared for synthesis of progestogens (Nagahama 1987). The same results were also found in goldfish Carassius auratus in which testosterone levels were high when gametes were fully mature and ready to ovulate (Kobayashi et al. 1987).

Table 1. Serum steroid hormones in ripe (n= 22) and gravid (n= 25) kutum during spawning period. Values are expressed as mean ± SD.

Steroid hormones	Gravid	Ripe	Significance
Testosterone	14.52±0.03	0.54±0.11	$P < 0.05$
17β-estradiol	0.75±0.18	0.36±0.07	$P > 0.05$
Progesterone	0.67±0.21	0.52±0.26	$P > 0.05$

17β-estradiol levels were lower than 1 ng/ml with no difference between ripe and gravid fishes ($P > 0.05$) (Table 1). In whitefish Coregonus clupeaformis, 17β-estradiol concentrations in gravid and sexually mature females were low (Rinchard et al. 2001). In Indian major carp Labeo rohita, 17β-estradiol levels showed decreasing trend during spawning (Suresh et al. 2008). Similar result was also reported by Sen et al. (2002) in the same species. In roach Rutilus rutilus, plasma 17β-estradiol levels decreased during the spawning period (Rinchard et al. 1997). It has been reported that decrease in the levels of this steroid may be a consequence of shift in steroidogenic pathway prior to germinal vesicle breakdown (Goetz et al. 1987; Tamaru et al. 1991) which can reduce the negative feedback on the pituatory and allow gonadotropin surge which is needed for maturation (Scott et al. 1983; Lee et al. 2002).

Progesterone levels were lower than 1 ng/ml in both groups of fishes (Table 1). Low levels of progesterone during spawning period may indicate limited role of this steroid in ovarian function and its indirect involvement in final maturation through dihydroxyprogestrone in spawning phase (Suresh et al. 2008).

In conclusion, kutum gravid breeders had high levels of testosterone and low amounts of 17β-estradiol, respectively. According to Rinchard et al. (2001), low levels of 17β-estradiol in gravid fish associated with high

levels of testosterone indicates that vitellogenesis is completed and the fish are ready to ovulate. Thus gravid females may need to remain in the freshwater river conditions so that environmental cues could act their roles in the reproductive axis to effective hormonal secretions.

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