

Induction of spawning in *Capoeta aculeata*, (Valenciennes in Cuv. & Val., 1844) (Teleostei, Cyprinidae), using carp pituitary extract

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Abstract. *Capoeta aculeata* is a benthopelagic fresh-water fish that lives in subtropical climates and has a widely distribution in Iranian river basins. Hatchery production of fish for stocking is an important means of temporarily supplementing insufficient natural reproduction, thus help to maintain or restore eroded fish population. This study was done in May and June 2007 on captured brood fish from Zayandeh-Rood River, Iran in order to reach a suitable technique of artificial reproduction of *C. aculeata*. Caught brood fish were injected with carp pituitary extract at doses of 0.75 and 1.5 milligram per kilogram body weight of fish. Brood fish were tested for ovulation each 12 hours from final injection for 3 days, but no sign of readiness for spawning was observed. To gain confidence of experiment, the study was repeated after one month from the first try with new fish. The result, however, was the same and didn't attain to any success. Inadequate information about reproduction biology of this species is one of the most important reasons of the negative result. Furthermore, unsuitable hormone and dosages as well as inappropriate injection times can be probable causes of this failure that should be evaluated in later studies.

Keywords: *Capoeta aculeata*, Spawning Induction, pituitary extract, Zayandeh-Rood River.

چکیده (In Persian)

سیاه ماهی، *capoeta aculeata* یک ماهی آب شیرین بنتوپلاژیک است که در مناطق نیمه گرمسیری زندگی می کند و پراکنش وسیعی در حوزه رودخانه های ایرانی دارد. تکثیر مصنوعی ماهی شیوه ای مناسب جهت افزایش ذخائر این موجودات در شرایطی که قابلیت تکثیر طبیعی آن ها فراهم نیست بوده و لذا در حفظ و بازسازی جمعیت آن ها کارساز خواهد بود. این مطالعه در ماه می و ژوئن 2007 بر روی سیاه ماهی های صید شده از رودخانه زاینده رود و به منظور یافتن تکنیکی جهت تکثیر مصنوعی آن در جهت کمک به بازسازی ذخائر آن صورت گرفت. مولدین صید شده توسط عصاره هیپوفیز کپور معمولی با دوز های 0.75 و 1.5 میلی گرم بر کیلوگرم وزن بدن، مورد تزریق قرار گرفتند. مولدین هر 12 ساعت پس از تزریق نهایی و به مدت سه روز جهت تخم کشی مورد ارزیابی قرار گرفتند. ولی هیچ گونه موفقیتی در زمینه القای تخم ریزی حاصل نگردید. جهت حصول اطمینان از صحت آزمایش، تمامی مراحل یک ماه پس از اولین مرحله با مولدین جدید تکرار گردید ولی نتایج بدست آمده مشابه مرحله اول بود و هیچ موفقیتی در این امر حاصل نشد. عدم وجود شناخت کافی از زیست شناسی تولید مثل این ماهی را می توان از جمله دلایل عمده عدم موفقیت این مطالعه نام برد. ضمن اینکه دلایلی مانند نامناسب بودن عصاره هیپوفیز کپور معمولی برای این ماهی، نامناسب بودن دوز های بکار رفته و حتی زمان تزریق می تواند از جمله دلایلی باشد که امید است با تغییر آن ها بتوان به موفقیت احتمالی دست یافت. واژه های کلیدی: *Capoeta aculeata*، القای تخم ریزی، عصاره هیپوفیز، زاینده رود.

Introduction. The genus *Capoeta* has a wide distribution in Southwest Asia and contains about 20 species of which seven occur in Iran (Bianco & Bănărescu 1982; Eschmeyer 1999; fishbase.org). *Capoeta aculeata* (Fig. 1) is a benthopelagic fresh-water fish that lives in subtropical climates (Mc Alister 1990) and has a widely distribution in Iranian river basins (Coad 2005) but has not been reported in other countries yet. Spawning presumably takes place in late spring and summer (Coad 2005).

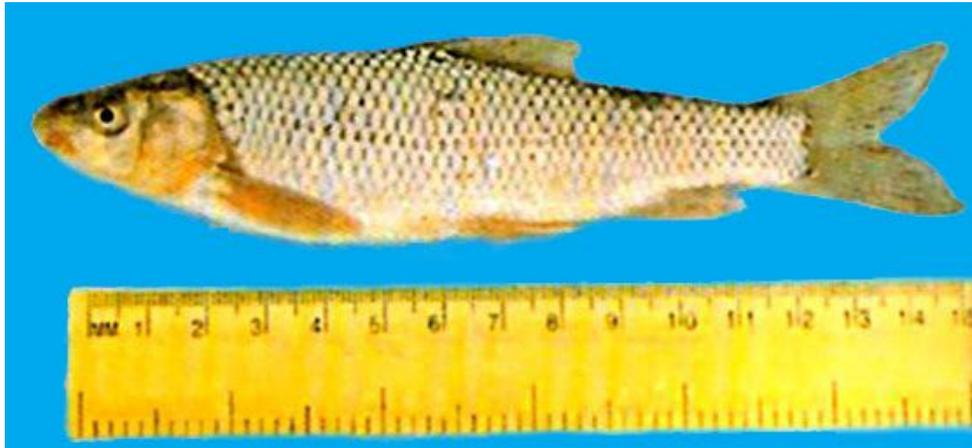


Fig 1. *Capoeta aculeata*

Changes in the natural character of Iranian streams as a result of human activities have caused the decrease of their plant and animal populations. Pollutants discharge, spawning areas damaging, over fishing, fragmentation of streams by the construction and operation of dams are major destructive factors to populations of these organisms. Recent droughts also have had an extreme effect on these ecosystems. Due to its limited distribution, *C. aculeata* can be in danger from these changes.

Techniques of induced spawning would help fish breeders to increase artificial reproduction. Hatchery production of fish for stocking is an important means of temporarily supplementing insufficient natural reproduction, thus helping to maintain or restore eroded fish population.

It is well-known that reproductive processes in fish are controlled by endogenous biological rhythms as well as by environmental cues (Munro 1990). Endogenous control is mediated through actions of various hormones along the hypothalamus-pituitary-gonad axis. Under natural conditions, environmental stimuli are detected and relayed to the brain, resulting in a release of hormones and neurotransmitters that regulate ovulation (Yaron 1995; Peter & Yu 1997).

There is limited study on biological and ecological characteristics, and no published data on artificial breeding of *C. aculeata*, hence the present study is the first reference on induced breeding for this species.

The objective of this study was to assay the effectiveness of Carp Pituitary Extract (CPE) in inducing breeding in this fish.

Materials and Methods. The present study was done in Fisheries Laboratory of Natural Resources Faculty in Isfahan University of Technology, Iran. Brood fish were captured by gill nets (20-30 mm mesh sizes) from Zayandeh-Rood River, located in centre of Iran (32°25'N, 51°33'E) (Fig. 2), in May and June 2007. Water temperature was 18°C. Totally 78 specimens were caught in two steps and carried to a cage located in a pond with a temperature of 18°C. After a few days the specimens were transported in fiberglass tanks to Fisheries Laboratory, and the fish were allowed to acclimatize for 2 days.



Fig. 2. Situation of sampling zone in Zayandeh-Rood River, Esfahan province, Iran.

Spermiation in the males was verified by applying gentle pressure on the abdomen and expressing a small volume of milt. In females, the stage of oocyte maturation was determined by dissecting. That showed female fish are in pre-spawning condition.

Fish were randomly divided in two experimental groups and transferred into two separate tanks. Each tank contained 10 females and 8 and 9 males for groups one and two, respectively. Table 1 shows different groups of males and females and the doses of pituitary extract used, for various groups of each step.

Anesthetized fish were tagged and injected with dry carp pituitary dissolved in 0.7% NaCl at a dose of 0.75 and 1.5 mg.kg⁻¹ body weight for groups one and two, respectively. Injection was done in two steps for females and one step for males. In the first injection only 10% of dose was utilized for both sexes. The second stage of injection for female was done at an interval of 12 hours from the first stage.

Table 1

Different groups of males and females and used doses of pituitary extracts in two steps

	<i>dose of pituitary extract (mg.kg⁻¹ body weight)</i>	<i>males</i>	<i>females</i>	<i>total</i>
Step 1				
Group 1	0.75	8	10	18
Group 2	1.5	9	10	19
Step 2				
Group 1	0.75	11	9	20
Group 2	1.5	12	9	21

Results and Discussion. Brood fish were tested for ovulation 12 hours after the final injection. At this stage, no sign of readiness for spawning was observed. These examinations were continued at each 12 hour for 3 days, but we couldn't induce females to spawn during this period.

To gain confidence of experiment, this study was repeated after one month from the first try with 41 new fish (18 female and 23 male fish). All stages in the second experiment were done as the first time, the result, however, was the same as in the former experiment and didn't attain to any success.

Inadequate information about reproduction biology of this species is one of the most important reasons of the negative result. More information about biological, ecological and environmental requirements of *C. aculeata*, such as suitable water temperature, substrate, photo periods, and exact time of natural reproduction, as well as efficient agents for artificial spawning are vital for induction of ovulation in captured brood fish. Unfortunately, no data are available on the role of various reproductive hormones and induced spawning in this species. Therefore, more investigations and studies appear to be essential. Some other important reasons of negative result include, first, suitability of carp pituitary extract to induce breeding in *C. aculeata*. Perhaps utilization of carp pituitary extract is not appropriate for this fish and using of specific hormones such as GnRH, HCG and so on would lead to better results. We should test the other hormones and agents to find a good one. Besides, as we didn't have any references about the suitable hormone dosage for *C. aculeata*, as well as we wanted to use common carp pituitary extract, we preferred to assess these doses according to suitable dose for common carp in this weight range (56-138g). Therefore, changing the dose of the hormone would cause induction of ovulation in this fish.

Conclusion. Doses of 0.75 and 1.5 mg per kilogram body weight of common carp pituitary extract, are not suitable to induce breeding of *C. aculeata*. Since the present study is the first try in artificial reproduction of this species and even in other species of this genus, we are not sure about the reasons of the negative responses. Nevertheless, we assume that further studies with dissimilar doses or using other usual hormones are necessary. More studies concerning the reproductive biology of this fish will be necessary to reach accurate information about biological and environmental requirements and subsequently, to attain successful results in induction of ovulation. Hence, we think biological and ecological studies should be in priority of later studies.

Another direction of research would be using pituitary extract of *C. aculeata* or other species of genus *Capoeta*, to induce artificial ovulation.

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