

## ULTRASOUND DIAGNOSTIC OF STURGEONS AND EXPLOITATION OF REPRODUCTIVE FEMALES AT STURGEON HATCHERY OF AZERBAIJAN

CHINGIZ MAMEDOV<sup>1\*</sup>, RAUF HAJIYEV<sup>1</sup>, MEKHMAN AKHUNDOV<sup>2</sup>

<sup>1</sup>*Baku State University, Az1141, 23, Z.Xalilov str., Baku, Azerbaijan*

<sup>2</sup>*Azerbaijan Fishery Research Institute, Az1008, 16, Demirchizade str., Baku, Azerbaijan*

*\*Corresponding author: e-mail: m\_chingiz@yahoo.com*

### ULTRAZVUČNA DIJAGNOSTIKA JESETARSKIH VRSTA I KORIŠĆENJE REPRODUKTIVNIH ŽENKI NA MRESTILIŠTU ZA JESETARKE VRSTE U AZERBEJDŽANU

#### *Apstrakt*

Formiranje matičnog stada jesetarskih vrsta za obnavljanje populacija započeto je još 2004. godine u Azerbejdžanu, gde je ostvaren uzgoj riba od stadijuma larvi do adultnih jedinki. Reproaktivne ženke perijske (Kura) jesetre (*Acipenser persicus* Borodin) koje su uzgajane "od ikre" u uzgojnim sistemima za ribe u Azerbejdžanu su 2013. godine po prvi put korišćene za veštački mrest u uslovima mrestilišta.

Cilj ovih studija je bila komparativna evaulacija kvaliteta reproduktivnih ženki koje su uzgajane u uslovima mrestilišta za jesetarske vrste riba i dobijanje njihovog potomstva. Istraživanja su takođe trebala da razviju naučno potkrepljenu dokumentaciju za eksploataciju matičnog stada jesetarskih vrsta, koja bi se bazirala na mrestilišima za jesetarske vrste u Azerbejdžanu.

Studija formiranja matičnog stada jesetarskih vrsta "od ikre" u uslovima Khyly ribnjaka za jesetarske vrste u Azerbejdžanu je bila eksperimentalne prirode. Rad na proizvodnji potomstva/mlađi od reproduktivno zrelih ženki je obavljen 2013. godine. Da bi se stimuliralo sazrevanje dve selektivno odabrane ženke persijske (Kura) jesetre iz generacije 2004. uzgajane u mrestilištu, data im je intramuskularno injekcija sa suspenzijom aceton-vodenog ekstrakta hipofize jesetri u odnosu 2.5 mg na 1 kg težine tela. Da bi se izbegao inbriding putem inseminacije ikre, uzeta su dva mužjaka persijske (Kura) jesetre iz generacije 2005. uzgojene u mrestilištu. Ultrazvučna dijagnostika pola i stepena polne zrelosti, hormonalna stimulacija matičnih riba, fertilizacija i sprečavanje adhezija ikre (odlepljivanje), kao i inkubacija u "Osetr" inkubacionom sistemu je sprovedena u saglasnosti sa postojećim metodama (Dettlaff et al., 1981; Chebanov and Galich, 2010; Mamedov et al., 2009).

Analiza uzgoja riba i bioloških indikatora juvenilnih riba koje su čuvane u mrestilištu je pokazala da je razvoj produkovane mlađi korišćenjem reproduktivno zrelih ženki iz matičnog stada bio u saglasnosti sa normama. Jedan deo proizvedene mlađi je zadržan u mrestilištu sa ciljem daljeg istraživanja njihovih morfoloških, bioloških i fizioloških karakteristika.

Time su u praksi uzgoja riba u Azerbejdžanu po prvi put korišćene za veštački mrest ženke persijske (Kura) jesetre iz matičnog stada koje je uzgojeno "od ikre" u mrestilištu. Ovi podaci ukazuju na mogućnost dostizanja reproduktivnog stanja različitih jesetarskih vrsta u uslovima mrestilišta. Stvaranje reproduktivnog matičnog stada jesetarskih vrsta u mrestilištima će omogućiti očuvanje genetskog pula jesetarskih vrsta i proširiti mogućnosti za njihov veštački mrest u cilju poribljavanja otvorenih prirodnih vodenih ekosistema.

*Ključne reči: matično stado, ultrazvuk, reproduktivne ženke, persijska (Kura) jesetra, „od ikre”, mrestilište za jesetarske vrste*

*Keywords: Broodstock, ultrasound, reproductive females, Persian (Kura) sturgeon, „from eggs”, sturgeon hatchery.*

## INTRODUCTION

Formation of recovery-maternal stock of sturgeon fish «from roe» assumes long growing of fish industrially. Meanwhile it is known that quality of a breeding material in many respects depends on conditions of their maintenance (Mamedov and Salmanov, 2009; Mamedov, 2011, 2012). Considering that the morphological analysis of blood is one of delicate and objective methods of an estimation of a physiological condition of an organism (Ivanova, 1983), we have previously studied the fish breeding and biological and hematological parameters of individuals of different ages Persian (Kura) sturgeon in the course of their long-term breeding in captivity.

Definition of the sex and maturity stage at sturgeon grown in aquaculture conditions at early ages and in the reproductive runs of various age groups by using the non-invasive diagnostic methods (ultrasound) and using them depending on development stages for fish growing purposes are one of the urgent matters.

The aim of the present study is a comparative evaluation of the quality of reproductive females holding under the conditions of sturgeon hatcheries and obtaining their off spring. This research is also needed to develop scientifically substantiated documents on exploitation of sturgeon broodstocks on the basis of sturgeon hatcheries of Azerbaijan.

## MATERIALS AND METHODS

The study was conducted in April 2012 at Khyly Sturgeon Fish Farm of Azerbaijan within the framework of the regional training project "The formation of sturgeon-getter, using them and ultrasound definition of their sex" of the UN Food and Agriculture Organization (FAO). Ultrasound examination was carried out in repair-getter Persian (Kura) sturgeon (*Acipenser persicus*) formed since 2004, in the Russian sturgeons (*A. gueldenstaedtii*) formed in 2005, in the Russian and Siberian sturgeon hybrids (*A. gueldenstaedtii* x *A. baerii*), sterlet (*A. ruthenus*) and beluga (*Huso huso*) fishes, as well as on part of stellate sturgeon (*A. stella-*

*tus*) formed in 2006, and their sex and maturity stages were determined. The total number of repair-getter sturgeon fishes examined was 70 pcs. The „Mindray DP-6600” device was used at ultrasound examination. Methods of research works and required recommendations have been described in details in the scientific literature (Chebanov and Galich, 2010).

Works on the production of off spring/fry from reproductive females carried out in 2013. To stimulate maturation of selected two elite females of the Persian (Kura) sturgeon of the generation 2004 from hatchery – reared broodstock intramuscularly was injected by the suspension of acetone-water extract of sturgeon pituitaries based on the rate of 2.5 mg per 1 kg of body weight. To avoid inbreeding with the view of insemination eggs two males of Persian (Kura) sturgeon of the generation 2005 from hatchery were used. Ultrasound diagnosis of sex and maturity stages, hormonal stimulation of breeders, fertilization and de-adhesion of eggs (unsticking), as well as its incubation in the “Osetr” incubation system were conducted according to the existing method (Dettlaff et al., 1981; Mamedov et al., 2009).

The entire biotechnical process starting from the maturation of females until the release of juveniles into a natural water area has been traced. Research on the reproductive females included a comparative study of the size-weight characteristics, gamete-somatic index, and fecundity in both absolute and relative values as well as a response to hormonal simulation/treatment. The quality of offspring was evaluated by fish cultural and biological indicators of eggs, larvae and fries. The Persian (Kura) sturgeon natural population’s off spring was served as control.

## RESULTS AND DISCUSSION

The global practice of fresh-water aquaculture proves a possibility of the maintenance and growing of sturgeon fishes in artificial conditions (Smolyanov, 1987; Popova et al., 2007; Chebanov and Galich, 2010; Cotenev et al., 2001). The principle of a bundling of maternal stock of sturgeon fish from spawn up to spawn is based on selection of elite posterity with the subsequent growing within 8-10 years up to mature sires. On the basis of posterity of artificial generation the recovery-maternal stock of sturgeon fish is formed on Khyilly Sturgeon Hatchery since 2004 (Mamedov and Salmanov, 2009; Mamedov et al., 2009). At the moment ten-year old (400 pieces), nine-year old (250 pieces), eight-year old (200 pieces), seven-year old- (1200 pieces), six-year old- (500 pieces), five-year old – (500 pieces) Persian sturgeons and different-years old Persian sturgeon stellate sturgeon and Kura-river ship (*A.nudiventris*) (more than 1000 pieces) of industrial manufacture are grown as an experiment in conditions of the Khyilly Sturgeon Hatchery. Additionally, some ten of specimen of Siberian sturgeon (*A.baerii*), sterlets beluga besters (*H.huso x A.ruthenus*) and Russian sturgeons are grown as an experiment in the Khyilly Sturgeon Hatchery. The general quantity of recovery–maternal stock of different kinds of sturgeon fish on Khyilly Sturgeon Hatchery constitutes about 5000 specimens.

It is necessary to note that ecological conditions at growing of young fish and adult forms of sturgeon fish in pools radically differ from conditions in which their various stages of ontogenesis passed during many millions years of evolution. For this reason the long maintenance of young fishes and adult forms of sturgeon in the closed space, that is in ecologically impoverished environment, can lead to simplification of their behavioral reactions, to decrease in impellent activity, formation of the certain stereotype of food behavior, so, to serious changes in functioning of various physiological systems.

In 2012 based on these studies on the definition of sex and stage of maturity of the sturgeon broodstock by ultrasonic diagnosis we were able to identify several females of Persian (Kura) sturgeon from the broodstock of the generation 2004, the state of maturity of the gametes were at III-IV and at IV unfinished stages of development (Table 1). These females were replaced in a separate tank for the purpose of formation of reproductive females at the hatchery.

So, creation of sex in sturgeon having no signs of external dimorphism at early ages and the reproductive run by using the non-invasive diagnostic methods of the maturity stage and implementation of these works according to required rules may be used in sturgeon fish farms of our republic and for commodity fish products.

In the 2013 year during the hatchery season previewing reproductive females by diagnostic ultrasound was done after intramuscular injections of aqueous suspension acetone-dried sturgeon pituitary. The duration of maturation of reproductive females after hormonal injection was 30-32 hours.

The average fecundity of two females was 131,150 of eggs. The number of eggs in 1 g sample amounted to 52 and 53 pieces, respectively. The incubation of sturgeon eggs took place in the individual "Osetr" incubation unit at a water temperature of 18,0-18,5°C. The incubation period was 4,5 days. The percentage of eggs development determined at the stage of the small yolk tube (17<sup>th</sup> stage) was 90.4 and 88.5%, respectively.

The duration of the early ontogenesis of the experimental and control larvae was similar and was made up 8 days at the water temperature from 18.5°C to 19.5°C. The onset of transition of larvae to active exogenous feeding was determined by their behavior, the release of melanin and the flow rate of the yolk mass, which by this time accounted for both versions 91.8%.

**Table 1.** Results of ultrasound diagnosis of sex and maturity stages of gonads of broodstock sturgeon in Khyilly Sturgeon Hatchery of Azerbaijan (2012)

Species of sturgeons	Years of hatching	Sex	Stage of maturity						The total number of sturgeon fishes	
			I	II	II-III	III	III-IV	IV		
Acipenser persicus	2004	♂		16	3	5	3	1	28	47
		♀		10	4	3	1	1	19	
Acipenser gueldenstaedtii	2005	♂					1		1	3
		♀					1	1	2	
Acipenser gueldenstaedtii x A. baerii	2005	♂				1	2		3	7
		♀				2	2		4	
Acipenser stellatus	2006	♂		1	1	2	2		6	10
		♀		1	1	2			4	
Acipenser ruthenus	2005	♂		1	1				2	2
		♀								
Huso huso	2005	♂		1					1	1
		♀								

During the transition to active feeding the larvae length increased up to 19.5 mm (experiment) and 20.0 mm (control), the weight of larvae increased respectively to 38.0 mg and 42 mg. The average daily growth rate on length of larvae in the experimental and control conditions amounted to 6.7 and 7.0% by weight 9.6% and 12.0%, respectively. The survival rate of larvae switched to active feeding averaged 86.5% (experiment) and 92.5% (control) with the standard index of 90%. The obtained offspring subsequently reared in plastic basins and after reaching the standard sample of 1.0 g were released into the Kura River's estuary.

The results of growing out of juveniles were assessed by using the method of total enumeration. The survival rate of stocking larvae to fingerlings was in average 67.4%. On separate basins this index varied from 64.5% to 73.6%. For the offspring obtained from fish of the natural complex, it is equal to 72.4% (standard - 70% - the temporary biotechnology standards for hatcheries of Azerbaijan 2007).

The analysis of fish breeding and biological indicators of hatchery-reared juveniles showed that the development of produced fry using reproductive females from broodstock was in compliance with the norm. Some of hatchery-reared fingerlings were kept at the hatchery with the view of further studying their morphological and biological and physiological features.

Thus, in fish cultural practices of Azerbaijan for the first time hatchery-reared broodstock females of the Persian (Kura) of sturgeon raised «from eggs» at the hatchery were used for artificial reproduction. These data indicate the prospects of the formation of the reproductive stage of various species of sturgeon at hatcheries. The creation of reproductive broodstock at sturgeon hatcheries will allow conservation of the sturgeon gene pool and expanding opportunities of their artificial reproduction to be released into natural water bodies.

## REFERENCES

- Cotenev, B.N., Bursev, I.A., Nikolayev, A.I. and Dergalieva, J.T. (2001). Strategy of preservation of sturgeon fish. *Pisciculture and fishery*, 1: 10-13. (in Russian).
- Chebanov, M.S. and Galich, E.V. (2010): Ultrasound diagnostics of sturgeons. "Proshvshchenie –Yug", Krasnodar, 134 pp. (in Russian).
- Dettlaff, T.A., Ginsburg, A.S. and Schmalhausen, O.I. (1981): Development of sturgeon fishes. "Nauka", Moscow, 223 pp. (in Russian).
- Ivanova, N.T. (1983): The atlas of blood cells of fish. "Leqkaya i pishevaya Promishlennost", Moscow, 184 pp. (in Russian).
- Mamedov, Ch.A., Hajiyev, R.V. and Akhundov, M.M. (2009): New technologies for sturgeon-breeding in Azerbaijan. "Science", Baku, 260 pp. (in Russian).
- Mamedov, Ch.A. and Salmanov, Z.S. (2009): Aquaculture in Azerbaijan: Pool method rearing of sturgeon fishes and their repair-maternal stock in Khyilly Sturgeon Hatchery. The Proceedings of the VI International Symposium on Sturgeon. Wuhan. 208-210.
- Mamedov, Ch.A. (2011): Morfophysiological and hematological features of recovery-maternal stock of sturgeon fish rearing in sturgeon fish hatchery of Azerbaijan. The Proceedings of the V International Conference. Belgrade. 258-260.
- Mamedov, Ch.A. (2012): The Results of experimental-industrial works on Formation of reproduction Stock of Sturgeons in the conditions of the Aquaculture of Azerbaijan.

Proceedings Azerbaijan National Academia Sciences: biological and medical sciences, 67. 43-47. (in Russian)

Popova A.A., Krupi V.A., Chernova P.V. and Daudova G.P. (2007): The state of reproductive potential of the Volga-Caspian sturgeons under present conditions. The International Scientific and Practical Conference, Astrakhan. 258-260.

Smolyanov, I.I. (1987): Technology of formation and operation maternal stock of the Siberian sturgeon in warm-water facilities, "VNIIPRX", Moscow, 33 pp.