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Determination of cutting point of oviduct in minimally invasive surgical technique in Persian sturgeon (*Acipenser persicus*) and Starry sturgeon (*Acipenser stellatus*)

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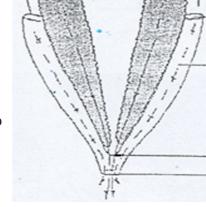
Abstract. Similar to other Chondrostean fish, Persian sturgeon (*Acipenser persicus* Borodin, 1897) and Starry sturgeon (*Acipenser stellatus* Pallas 1771) have got Gymnovarian ovaries, and structure form of their reproduction system is such that eggs cannot readily leave it. In minimally invasive surgical technique, a small incision is made in the wall of oviduct so that the eggs can easily leave it with the minimal manipulation and also the broodstock can survive. The aim of this work is to determine the distance between gonopore and incision area according to size and species which results in a more accurate and flawless incision. The results of this study showed that the distance between gonopore and conjuncture area of the two oviducts depends on species and size of fish; this distance in Persian sturgeon with 115-191 centimeters fork length and Starry sturgeon with 122-143 centimeters fork length were 4.2-6.3 and 3.8-4.5 centimeters, respectively. This distance is more than the incision depth that is generally made on the wall of one of the oviducts.

Key Words: oviduct vessel, minimally invasive surgical technique, Persian sturgeon, Starry Sturgeon.

چکیده:
قره برون و ازون برون مانند دیگر ماهیان غضروفی- استخوانی دار ای تخمدان جیمنوو ارین می باشند و ساختل شکلی دستگاه تولید مثلی این ماهیان به گونه ای است که تخم ها نمی تو انند به سهولت از آن خارج شوند. در روش ریز برش یک برش کوچک در دیواره مجرای تخمک بر زده می شود تا تخمک ها بدون کمترین دستکاری خارج گردند و مولد نیز زنده بماند. هدف از اجرای این پروژه تعیین فاصله بین منفذ تناسلی با محل برش با توجه به سایز گونه ماهی است که منجر به برش دقیق و بی نقص تری می شود تا تخیی است که منجر به برش دقیق و بی نقص تری می شود تایج این تحقیق نشان داد که فاصله بین منفذ تناسلی و محل اتصال دو مجرای تخمک بر به گونه و اندازه ماهی بستگی دارد و در قره برون با طول چنگالی 115- 143 سانتی متر بین 3/8 تا 4/5 سانتی متر بود. این فاصله بیشتر از عمق برشی است که عموما بر روی جداره یکی از مجاری تخمک بر داده می شود.

Introduction. Sturgeons are of oviparous fish which release their eggs and sperms into water and have external zygosis. In Persian and Starry sturgeons (*Acipenser persicus* Borodin, 1897 and *Acipenser stellatus* Pallas, 1771) similar to other chondrostean fish, the eggs are released to ventral cavity (Billard 2000, Conte et al 1988) and goes to one of two ovaries through short Mullerian duct (ciliated funnel) which is connected to Mesonephric duct; this configuration of ovary is called Gymnovarian (Hoar 1969). In sturgeons, the bottom of ovary capsule is not set next to the oviduct and the eggs that released to ventral cavity, entered to rather long tube of oviduct in order to leave body (Fig. 1). Therefore, unlike Cytovarian fish (such as salmon family) forming structure of female sturgeons breeding system is such that eggs cannot readily release out of body (Burtsev 1969). Thus, in female sturgeons, only a part of ripe eggs which have entered oviduct can be gained by pressure. These ratios of eggs are small part of whole eggs in female fish.

- 1. Ovary
- 2. Spout (funnel) vessel of the eggs conductor
- 3. Vessel of the eggs conductor
- 4. Gonopore
- Place of incision (conjuncture area of two oviducts)



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Figure 1. Form of ovarian in sturgeons (orig.).

Nowadays, numerous methods are being used in order to collect eggs from female sturgeons which most popular one is cutting abdominal region and achieving eggs after determinate ripening stage and by use of probe (Graham et al 1986, Conte et al 1988). Another method to collect eggs is called periodic collection in which the amount of eggs present in oviducts is collected every time. Another method is spawning of injected fish in ponds which has not been succeeded. In another method called caesarian, eggs are collected through an 8-10 cm incision in upper part of gonopore in abdominal region (Burtsev 1969, Conte et al 1988).

Because of problems of above-mentioned methods (cited by Mokhayyer 1993, Mims et al 2002, Parandavar et al 2006), a new method called minimally invasive surgical technique (MIST) has been proposed which was made fast collection of eggs possible along with the minimal manipulation of brood stocks of sturgeons (Mims et al 2004, Stech et al 1999).

Considering a mild bleeding in incision area of oviduct after the surgery, the incision is healed and repaired fast. If the incision is not made properly, the fish may die due to bleeding. One of the most important points is to make an accurate incision is to know the distance between gonopore and incision area, which depends on size of the fish. The aim of this work is to determine the distance between gonopore and incision area in two sturgeon species according to the size of the fish which results in a more faultless incision.

Material and Method. In order to reach the aim of this work, 8 mature-sized Persian sturgeon and 3 mature-sized Starry sturgeon brood stocks were caught from Sefidrood River in April and May of 2008 using gillnet and then were transported to Shahid-Beheshti hatchery, Guilan, Iran. The fish were placed in circular Kourenski ponds filled with the water from Sefidrood River with 50 L min⁻¹ module current rate. Choos the most appropriate brood stocks and calculation the time of hormone injection were done on the basis of germinal vesicle (GV), sexual maturity index (Detlaf et al 1993) and estiminated to the method of Van Eenennam et al (2001).

The female brood stocks were intramuscularly injected with the hormone acquired from pituitary of sturgeons (50-70 mg kg $^{-1}$ (BW) 3). The female brood stocks were injected two times with 12 hours interval (10% and 90% of total dose, respectively). The temperature of water was 14.2-15.1 $^{\circ}$ C through the experiment. 22-26 hours after the second injection, ovulation occurs. When female brood stocks become ready to spawn, they were anesthetized using 300 mg L $^{-1}$ clove oil after 5 minutes and then, they were transported to reproduction room. Biometry was done after sacrificing the fish and eggs were removed through an incision in abdominal region according to the traditional method (Conte et al 1988). The incision was made near the gonopore. After cutting the upper part of oviduct to urogenital pore and removing, this part was fixed in formalin.

Afterward, the samples were taken to laboratory and then, diameter of gonopore and also the distance between gonopore and conjuncture areas of two oviducts were measured using a caliper (see Figs 2-3).



Figure 2. Measuring the diameter of gonopore by a caliper.



Figure 3. The area that eggs pass from oviduct to gonopore (head of pen).

Results and Discussion. The mean standard length and total weight of Persian sturgeon and Starry sturgeon before eggs collections were 133.5 \pm 23.19 cm and 13.65 \pm 9.03 kg and 124.33 \pm 6.50 cm and 9.26 \pm 0.75 kg, respectively (Table 1 and Table 2).

Module parameters in Persian sturgeon (n=8)

Table 1

Total	Standard	Fork Diameter of		Distance between gonopore to	
weight	length	length	gonopore	conjuncture areas of two oviducts	
(kg)	(cm)	(cm)	(cm)	(cm)	
14	108	115	2.75	5.5	
13.3	112	123	2.5	5.5	
33	152	167	1.8	5	
35.5	177	191	2	6.3	
34	157	171	2.2	5	
33	150	167	2.5	6	
29	132	151	1.6	4.2	
32	140	162	2.75	5.8	

Total weight (kg)	Standard length (cm)	Fork length (cm)	Diameter of gonopore (cm)	
10	118	122	1	4.5
9.3	124	131	1	4.5
8.5	131	143	1.5	3.8

The results show that there is a direct correlation between standard length of the brood stocks and the distance between gonopore and conjuncture area of two oviducts (incision area in MIST method) (in Persian and Starry sturgeons, it is $r^2=0.014$ and $r^2=0.75$, respectively); this distance is longer in heavier fish. The comparison between the above mentioned two species show that these characteristics exist in both species of sturgeons.

Poorasadi et al (2009) compared MIST techniques and traditional method of eggs collection from Persian sturgeon brood stocks. The mean weight and fork length of their female brood stocks were 28.3 ± 2.3 kg and 154.2 ± 4.6 cm, respectively. Depth of stripping via scalpel into gonopore was 2-3 cm; the distance between gonopore and conjuncture area of two oviducts (depth of stripping via scalpel) was shorter than the distance which used in this study for the brood stocks with similar mean weight and length.

Also, Bani & Banan (2010) compared MIST and traditional methods to collect eggs from Starry sturgeon brood stocks. The mean weight and fork length of 8 female broodstocks were 9.75 \pm 1.7 kg and 116 \pm 5.7 cm, respectively. The depth of influx of their scalpel into gonopore was 1-5 cm according to the size of the fish; the distance between gonopore and conjuncture area of two oviducts and the depth of stripping via scalpel was consistent with our findings.

On the basis of results of this work, female Persian and Starry sturgeon brood stocks have no vent in their oviducts. Nevertheless, Conte et al (1988) stated that oviduct of White sturgeon (*Acipenser transmontanus* Richardson, 1836) has got vent.

Compared to other mentioned methods, MIST method has some advantages such as fast collection of eggs (Mims et al 2004, Bani & Banan 2010, Poorasadi et al 2009), absence of inflammation or infection after some post period (Stech et al 1999, Mims et al 2004, Poorasadi et al 2009, Bani & Banan 2010), lower stress to the brood stocks and absence of problem in devouring food again (Parandavar et al 2006), possibility of returning the brood stock back to ponds in case eggs are not ovulated (Conte et al 1988, Padushka 1999), and also could extracted more than 90% (Bani & Banan 2010) to 30-90% (Parandavar et al 2006) of brood stocks eggs. Other experiments results have shown that there is not a significant correlation between position of GV, weight of collects eggs, number of eggs, productivity rate, and percentage of hatching comparing MIST and traditional methods for collecting of brood stocks eggs (Poorasadi et al 2009, Bani & Banan 2010).

As mentioned above, on the basis of our results, in MIST method, the incision is done in conjuncture area of walls of two oviducts and accuracy in measurement of the distance between the incisions to anus results in lesser damage to brood stocks. Unlike our findings, Mims et al (2002) showed that incision in this method in done in urogenital vessel.

Conclusions. The results of this study showed that the distance between gonopore and conjuncture area in two oviducts of Persian sturgeon with 115-191 cm fork length and 13.3-35.5 kg weight and Starry sturgeon with 122-143 cm fork length and 8.5-10 kg weight were 4.2-6.3 and 3.8-4.5, respectively. This distance is more than the depth of stripping into the wall of one oviduct (Mims & Shelton 1999, Mims et al 2002).

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