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Seasonal fluctuations of steroid hormones in beluga sturgeon *Huso huso* cultured in brackish water

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Abstract

The experiment was carried out in one year on blood sexual hormones in 74 four-years old beluga sturgeon *Huso huso* held in 8 round cement tanks in brackish water with salinity of about 13.5 ppt. Fishes were biopsied at the beginning of the experiment in order to determine the sexuality and gonadic stage. Blood sampling was performed in caudal veins three times during experiment in fall, winter and spring seasons.Testosterone (T), Esteradiol (E2) and Progesterone (P) were measured by Radio Immune Assay (RIA) method. Test of profile was done in duplicate. Statistical results showed a significant elevation ($P > 0.05$) on measured factors in both sexes, especially in males.In males, testosterone (T) depends on gonadic stage had significant elevation; but in females, there was no significant increase ($P > 0.05$) in estradiol (E2) and progesterone (P) which probably resulted from their ages. In males, T level in stages II, II-III, III, IV, were 5.5, 4.4, 6.3, 2.6 ng/ml, respectively.The result indicated that the amount of T increased by development of gonads until III stage, then suddenly decreased in stage IV. Quantities of P and E2 were measured only in females**.**Amounts of P and E2 in stages of II, II-III, were 0.12, 0.13 and 1.455, 0.81 ng/ml, respectively, suggesting the increase of P and decrease of E2 were associated with gonad progress. According to the results, it was demonstrated that sexual hormones were affected by season, environmental conditions and gonadal stages.Therefore, inland underground brackish water as a new medium culture was effective in reduction maturity time in beluga.

Keywords: Steroid hormones, Gonadic stage, Gonad development, Beluga, Brackish water

Introduction

Many parameters such as environmental, nutritional, genetic, and endogenous factors affect fish growth. However, many studies have examined the impact of these factors upon growth rate, the interrelationship between genetic determinism of growth and endocrine status in fish remains poorly understood. From the view point of pisciculture, it is important to establish whether growth rate is related to endogenous levels of growth promoter hormones. They are likely under the same degree of genetic control, and hence could be selected for in-breeding programs.

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The establish relationship between different body tissues is very vital. Among this relationship, hormones have important role. Hormones are regulators which affect cells metamorphism. Hormones are not beginner of reaction and many physiological actions undergo without them. Hormones are divided into three major groups based on their chemical construction (Nagahama 1993): steroid hormones, peptide hormones and amino acid productions. Estrogen and progesterone are produced by ovary and testosterone by testis. In addition, upper kidney gland produces some steroid hormones. Secretion of gonadotropin from pituitary is affected by gonad steroid hormones via a feed back mechanism. Overall, steroid hormones are produced by upper kidney layer, ovary and testis that have similar embryonic origin.

Annual fluctuation of hormones is in relation to reproduction, feeding and growth cycle of fishes (Scott et al; 1983). Increase of 17 β estradiol, progesterone and cortisol during maturity was reported in many teleost fish (Nagahama 1993). Beluga is potentially the commercially most important species in the world for caviar production, but apart from extensive studies on physiological issues (Scott et al. 1983; Qiu-zhi et al. 2005), a little information on sex steroid changes and gonadal development stages of this species has been reported (Yousefian 2005).

Gonads steroid hormones affect the secretion of gonadotropin from pituitary via a feed back mechanism. These hormones that are active before maturity (Yousefian 2005); include: 1) progestins including progestin, 17α Dy Hydroxy progesterone, 20α Dy Hydroxy progesterone and 20β Dy Hydroxy progesterone which is produced by ovary and have an effective role in final oocyt maturity. In this study 17α Dy Hydroxy progesterone was examined. 2) Estrogens including 17-β estradiol and 17estrone that are also produced by ovary and their important role is stimulation of liver to produce vitelogenin (Yousefian 2005). Among these hormones 17β estradiol was examined. 3) Androgens which are hormones responsible for sexual growth and development. The most important androgen is Testosterone that was measured in present study.

Most of studies on steroid hormone carried out in maturity and harmonious with final maturity of gonad hormone fluctuation had examined only in short time. However, with awareness to seasonal fluctuation and effect of environmental condition before maturity, it can be obtained the beneficial information about suitable time of hormonal injection, creation of premature populations and decrease of long time maturity in this valuable fishes. Therefore, in present study, the profile of seasonal fluctuation of steroid hormones was examined before maturation.

Materials and methods

This research was carried out in Inland Salt Water Fishes Research Station, Bafgh, Iran for one year (summer 2005 to spring 2006). Eight round roofed 30 tons cement tanks, which equipped with water distribution system and central aeration, were used. Required water was supplied through a well with constant salinity of about 13.5 ppt. Seventy-two individual four years old great sturgeons with mean weight of about 11.0 ± 1.2 kg from a culturing population in brackish water earth pond were randomly taken and transported to the cement tanks. Fish were fed pelleted food in protein and energy levels of 40% and 4500 kcal/kg, respectively. Fishes were biopsied at the beginning of the experiment in order to sexuality and gonadic stage determination. Histological preparation was stained with hematoxyline and Eosin method (Bahmani and Kazemi 1998) in physiology and biochemistry Dept. of International Research Institute of Sturgeons (Rasht, Iran).

Blood sampling was performed in caudal vein using non-heparinized syringes three times during experiment in seasonal (fall, winter, spring). 4-5 ml blood were collected from each fish. Storage, centrifuging and pipeting of the samples were carried out at 4 ˚C. Plasma for further assays was stored at -20 ˚C. Testosterone (T), Esteradiol (E2) and Progesterone (P) were measured by Radio Immune Assay (RIA) method. The test of profile was done in duplicate. The SPSS software (version 10) was used for statistical analyses. Assignment of data correlation was done by Pearson and Kendal tests. Duncan test was used to compare mean differences. The significant differences were determined at %5 level.

Results

Sexuality determination

For the whole of fish, there were 57% male and 43% female. According to gonadic studies of biopsied fish, 4% of males were in I, 11% in I to II, 60% in II, 12.5% in II to III and 12.5% in III to IV stage. For females, they were 23% in I to II, 60% in II and 17% in II to III stage.

Testosterone (T)

Statistical results showed that amount of T between male and female had strong significant difference (sig = 00, f = 127.3) and was higher in males. In different seasons significant difference was not observed (sig = 0.94, f = 0.062) (Fig. 1). The highest amount of T in males was in fall (5.33 ± 4.4 ng/ml), the lowest was in winter (5.06 ± 2.4 ng/ml) and the average was 5.22 ± 3.2 ng/ml. In females, the most amount of T was also found in fall (0.21 ± 0.21 ng/ml); the lowest was in winter (0.1 ± 0.00 ng/ml) and the average was 0.14 ± 0.13 ng/ml.

In addition, T in different sexes had significant correlation with gonadic index (sig. = 0.01, r = 0.5) (Fig. 2) and would be considered T as index of Gonadic growth in males. Histological results showed that the amount of T of males in stage II, II-III, III and IV was 5.5, 4.4, 6.3 and 2.6ng/ml, respectively and was 0.10 and 0.16 ng/ml in stage II and II-III, respectively, for females.

Progesterone (P)

Amount of P was measured only in females. Statistical results showed that there was no significant difference in different seasons (sig. = 0.064, f = 3.48) (Fig. 3). The highest amount of P was in winter (0.66 ± 0.06 ng/ml); the lowest was observed in spring (0.12 ± 0.04 ng/ml) and the average was 0.37 ± 0.41 ng/ml. There was no significant correlation between gonadic indices. In addition, histological results demonstrated that amount of P was 0.12 and 0.13 ng/ml, in stage II and II-III.

Estradiol (E2)

Amount of E2 also was measured only in females. Statistical results showed significant difference in different seasons (sig. = 0.00, f = 16.95). The highest amount of E2 was in fall (5.51 ± 2.6 ng/ml); the lowest was observed in winter (1.18 ± 0.82 ng/ml) (Fig. 4) and the average was 2.63 ± 2.6 ng/ml. There was no significant correlation between gonadic indices.

Results also showed that the amount of E2 was 1.45 and 0.81 ng/ml, respectively, in stage II and II-III. According to the results, amount of steroid hormones were affected by sexuality. Therefore, quantity of T in males and E2 and P in females was higher. Because of low concentration of P and E2 in males, these hormones were measured only in females. It also was observed that only E2 was affected by season.

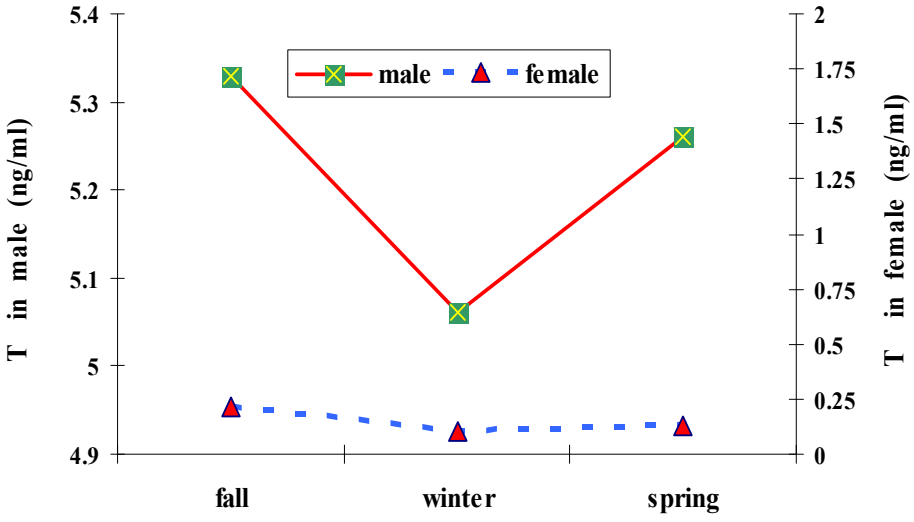

Fig. 1. Serum testosterone in different seasons

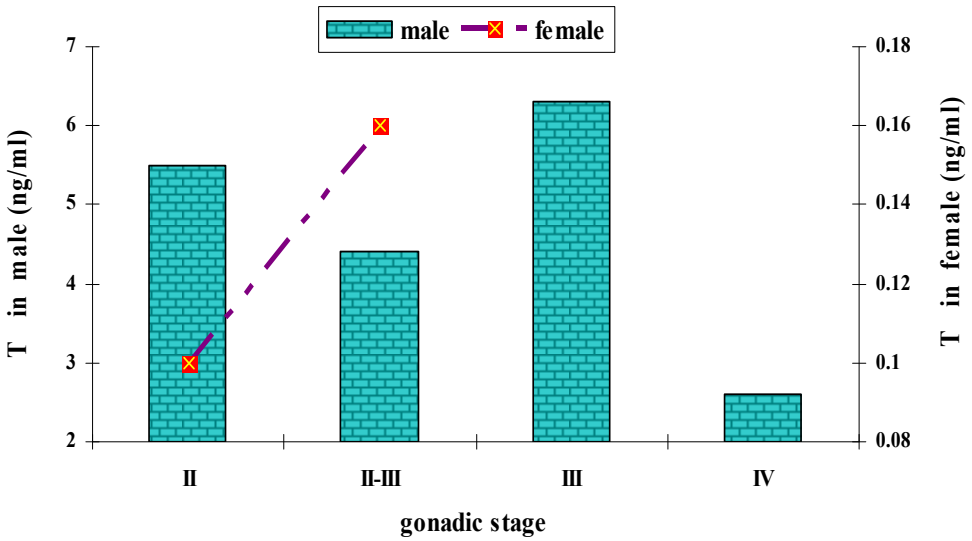

Fig. 2. Serum testosterone in gonadic stage

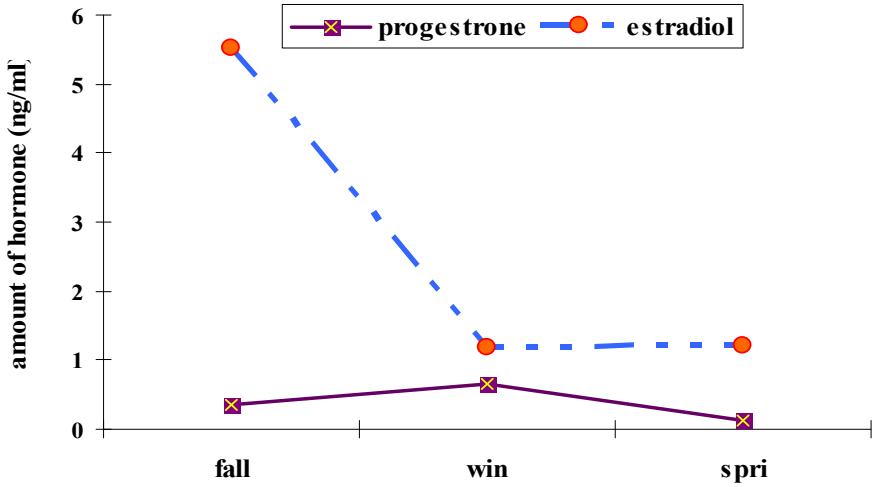

Fig. 3. Serum estradiol and progesterone in different season

Discussion

Hormonal changes are dependent on the alternation of environmental temperature. For suitable temperature for spawning, amount of hormones will be increased (Malollahi 1994). Fluctuation of sexual hormones in annually, monthly and diurnal cycle in teleost fishes was proved (Qiu-zhi et al. 1994).

Results showed that in males by progressing in gonadic stage until stage III, amount of T increased and then intensely decreased in stage IV. In females with progression of gonadic stage, quantity of P increased, but in contrary amount of E2 decreased. This is because of gonad formation that affects amount of hormones.

Direct correlation between secretion of sexual steroid hormones and development of gonad with synthesis and amount of gonadotropine hormones has been reported. In previttelogenic stage and stage II, oocyte is little and also separating animal and vegetable pole is not possible; amount of sexual steroid hormones is low (Nazari 2010).

Therefore, immature fish in this study had the low amount of hormones. Yousefian (2005) examined the fluctuation of E2 and T in young Great sturgeon cultured in freshwater and showed that their concentrations are low until one years old (respectively 0.23 and 0.8 ng/ml in both sexes), and also in two years old. But in age of 2.5 years old, in both sexes, the amount of E2, T and P were 3, 10.1 ng/ml and 7 pg/ml, respectively (Yousefian 2005). When comparing this result with present study, it was demonstrated that in brackish water, the amount of P was higher but amount of T and E2 were lower than freshwater.

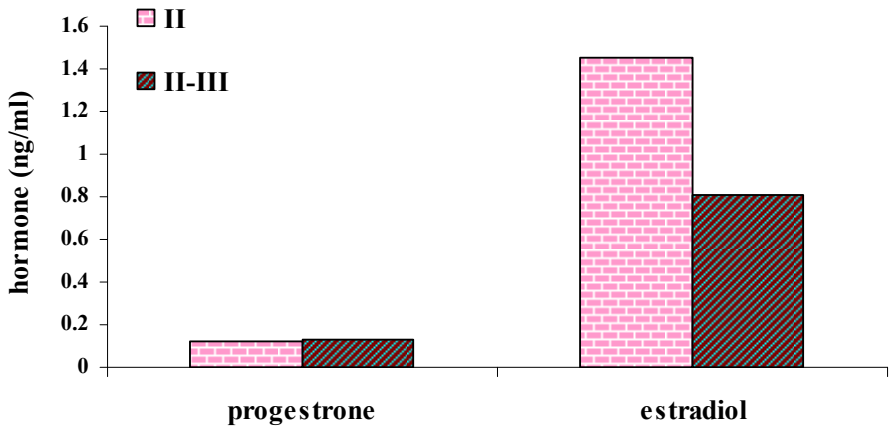

Fig. 4. Serum progesterone and estradiol in different gonadic stages of females

The study on immature white sturgeon clarified that in age 2 years old, amount of T was less than 1ng/ml in both sexes and this amount in males and females was 37.4 and 1.7 ng/ml, respectively (Fitzpatrick et al. 2004). Investigation on mature and immature Acipenser sturio in brackish water proved that the highest amount of steroid hormones was T which in mature males and females was 40 and 20 ng/ml, respectively. Amount of E2 and T in immature fishes was very low and it probably resulted in less gonadic development. This research also showed a seasonal fluctuations of steroid hormone in immature fishes (more in spring) (Cuisset et al. 2005). According to these results, seasonal fluctuations in steroid hormones were not only in matured fish but also could be observed in immature fish like present study.

In other research on Persian sturgeon, quantity of T in stage II was very low (0.25 ng/ml). In stage III, T increased (8.55 ng/ml) and in stage IV had a little decrease but remained at high level (7.44 ng/ml). E2 had a significant correlation with gonadic stage and in stage II was low (0.55 ng/ml) but increased in stage III (harmonizing with ovule growth) (4.53 ng/ml) and then in stage IV decreased (2.65 ng/ml). P also had a significant correlation with gonadic stage. In stage II, III, IV, T was 0.32, 0.52, 0.36 ng/ml, respectively (Nazari 2010). This report confirmed increasing of T and P in present study but decrease of E2 was opposite to other reports and need more investigations about that.

Hormones in stages II, II-III of ovum development are low because in this stage (previtellogenic stage) ovum is small and animal and vegetable poles are not distinct. Results in present study were in agreement with other reports, in which lower hormone in early stage of gonad development was observed (Cuisset et al. 2005). Stage of concentrating vitellin in ovule (is still nucleus in center of ovule) was affected by secretion gonadotropin hormone in pituitary gland. The amount of steroid hormone is increased (Yaron 1995). So, in present study production of steroid hormone in stage II-III also increased.

By comparing the present study with other similar studies, it was ascertained that amount of T in both sexes in brackish water is lower, but amount of E2 is higher than other condition. Great sturgeon cultured in brackish water at stage II had higher amount of sexual hormone than other condition. In addition, both sexes of this species in brackish water have upper gonadic stage than other environments.

According to the results, it seems that sexual hormones fluctuate before maturity and is affected by different seasonal and environmental condition. In order to investigate exact endocrine process and sexual steroid hormone production, these hormones should be measured before and in maturity stage.

Taking in to consideration, hypothesis of being correlation between blood profile with gonadic stage of Great sturgeon cultured in brackish water was confirmed. The lower amount of steroid hormone was observed in early stage. The increase in until stage III and decrease in stage IV were proved. No relationship between P and E2 with gonadic stage indicated that in immature fish in upper stage (near maturity), all of steroid hormones were affected by gonadic stage and had significant correlation in different gonadic stage.

It can be concluded that culture medium of brackish water was more proper than fresh water and had a wonderful effect on gonadic growth and steroid hormones production progress in Great sturgeon. Sustainable management and use of effective factors on gonadic growth can be reduced long time of maturity in sturgeon cultured in brackish water and in order to caviar production in shorter time.

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References

Bahmani M, Kazemi R. 1998. Histological study of gonad in young cultured sturgeon. Fish Sci mag 7: 1-16.

Cuisset BD, Lacomme S, Rouault T, Pelard M, Lepage M, Williot P. 2005. Hormonal profile in adult of critically endangered sturgeon *Acipenser sturio*, adapted to hatchery condition. 5th ISS Symposium, Ramsar, Iran.

Fitzpatrick MS, Feist GW, Eenennaam JV, Doroshov SI, Schreck CB. 2004. Sex identification of white sturgeon at early stage of growth. Oregon State University, USA.

Malollahi A. 1994. Examination of seasonal fluctuation of sexual hormone in *Acanthopagrus latus*. Persian Gulf Fishery Research Center, 47 pp.

Nagahama Y. 1993. Endocrine regulation of gametogenesis in fish. Int J Dev Biol 38: 217-229.

Nazari RM. 2010. Plasma sex steroid hormones of Persian sturgeon *Acipenser persicus* as influenced by gonad development stages and season. Int Aquat Res 2(1): 49-54.

Qiu-zhi QU, Da-jiang S, Bing-qian W, Guo-jan M. 2005. The relationship between gonad development and sex steroids level at different age of *Acipenser Schrenckii*. 5th ISS Symposium, Ramsar. Iran.

Scott AP, Sumpter YP, Hardiman PA. 1983. Hormone changes during ovulation in the Rainbow trout (*Salmo gairdneri*). Gen Com Endocrin 49: 128-134.

Yaron Z. 1995. Endocrine control of gametogenesis and spawning induction in the carp. Aquaculture 129: 49-73.

Yousefian M. 2005. The sex differentiation by gonadogenesis and sex steroid hormones in cultured Grate sturgeon, *Huso huso*. 5th ISS Symposium, Ramsar. Iran.