

Determination of Sex Hormones Levels in the Seminal Plasma of Persian Sturgeon, *Acipenser Persicus*

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Abstract

In the present study, we investigate the steroid levels in the seminal plasma of Persian sturgeon in order to obtain the basic information about steroid changes in surrounding liquid of ejaculated spermatozoa. In this regard some steroids were measured by Radio Immune Assay (RIA) including: Testosterone (T), Progesterone (P), 17α , hydroxyprogesterone (OHP) and 17β -estradiol (E_2). According to results, the steroid values had high variations between seminal plasma samples. The values of assay steroids were significantly lower than their concentrations in serum. T had highest concentration while E_2 showed lowest concentration in the seminal plasma. As well as, there was significant correlation between T, P and OHP in the seminal plasma. It seems that androgens comprise the main steroids of seminal plasma of Persian sturgeon males. Probably, the concentration of steroids in seminal plasma is depending on their concentrations in serum.

Key words: Ejaculation, steroid hormones, seminal plasma

INTRODUCTION

The sex steroids are the main factor for inducing of gonad maturation in fish [3, 5]. During final maturation of spermatozoa, the spermatozoa acquire fertilization capacity in the spermatid duct under control of sex steroids [7, 8]. After spermiation, the spermatozoa are plunged in seminal plasma, a liquid media composed of organic and inorganic components which preserve sperms till spawning or stripping [2]. However many studies investigated the steroid levels of serum during maturation, but information about sex steroids in seminal plasma and its role on sperm physiology is unknown. Until now, a few studies have reported the various levels of steroids in fish seminal plasma. The Persian sturgeon is a critically endangered anadromous species that has been considered for a biological conservation program in the southern part of the Caspian Sea [6]. However, there is a little information about steroid levels in sturgeon seminal plasma especially Persian sturgeon. Therefore, the study aimed to measure the values of steroids in seminal. Steroids including: Testosterone (T), Cortisol (C), Progesterone (P), 17α , hydroxyprogesterone (OHP) and 17β -estradiol (E_2).

MATERIAL AND METHODS

The Persian sturgeon males (n=9) were captured in the southern part of the Caspian Sea during March to June 2008 and transported to Shahid Beheshti Artificial Sturgeon Propagation and Rearing Center (SAPRC), Iran, Rasht. After delivery to SAPRC, the males were kept in pond (1m×4m×8m) with flow of river water (about 1.5 m³/min). During the experiment, the water temperature was 16–18 °C, dissolved oxygen was 8–8.3 mgL⁻¹ and pH 7.3–7.5. After spermiation, from each spermiating males, the milt samples were collected by hand. After milt collection, the milt samples were centrifuged (Heraeus, Sepatech, Labofuge 200, Germany, 5000 rpm for 10min) to separate the SF. The SF samples were stored at -20 °C until hormonal analysis.

Steroid assays

Radio Immuno Assay (RIA) for Testosterone, Progesterone, 17α , hydroxyprogesterone and 17β -estradiol measurements:

We used Direct Radio Immuno Assay method (DRIA) which was a simplified method of Canario and Scott (1989)

method according to (Stanczyk, Røed & Fjalestad 2007). This method, do not incorporate a purification step (extraction) before quantification of the analyte. Assay kits of C, T, P, E₂ and OHP were obtained from IMMUNOTECH Company, France. Each commercial assay kit contained two necessary reagents [i.e. Antibody and Tracer (labeled antigen)]. The Kit properties were described in details by Hajirezaee et al. 2011 [4]

The SPSS software was used for data analysis. The correlations between the steroid concentrations of the seminal plasma were tested using the bivariate correlation coefficients of Pearson. Then, the linear and non-linear regression models were investigated using regression fits.

RESULTS AND DISCUSSION

The minimum, maximum and mean of sex steroids of seminal plasma have been presented in Fig 1. The steroid values had high variations between seminal plasma samples. The values of assay steroids were significantly lower then their concentrations in serum. T had highest concentration while E₂ showed lowest concentration in the seminal plasma. Also, there was significant correlation between T, P and OHP in the seminal plasma (Fig 2 and Fig 3, P<0.01). Fish seminal plasma has a unique composition regarding the presence of organic and inorganic compounds which support the spermatozoa till

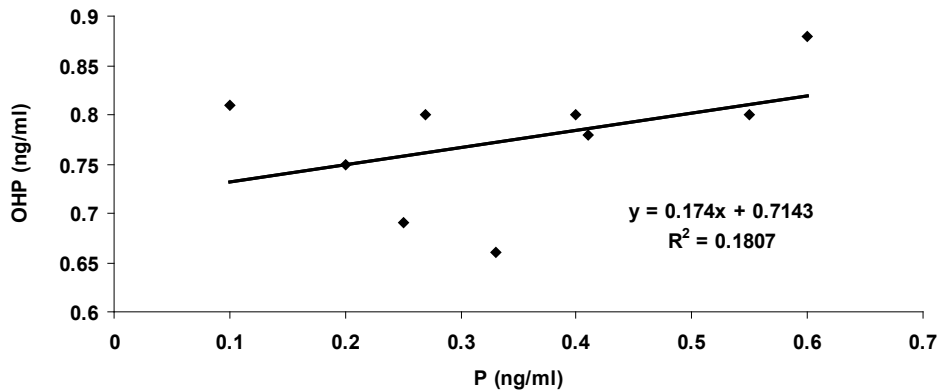


Fig 1. the steroid levels in the seminal.plasma of Persian sturgeon (mean ± sd).

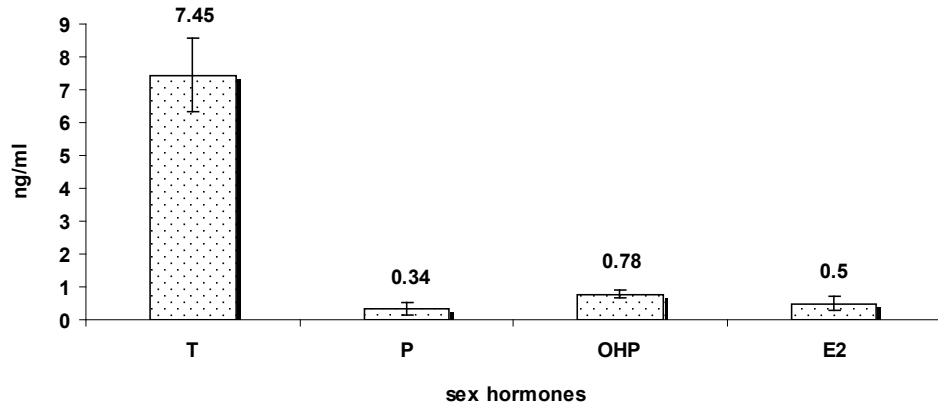


Fig 2. Relationship between P and OHP concentrations of seminal fluid.

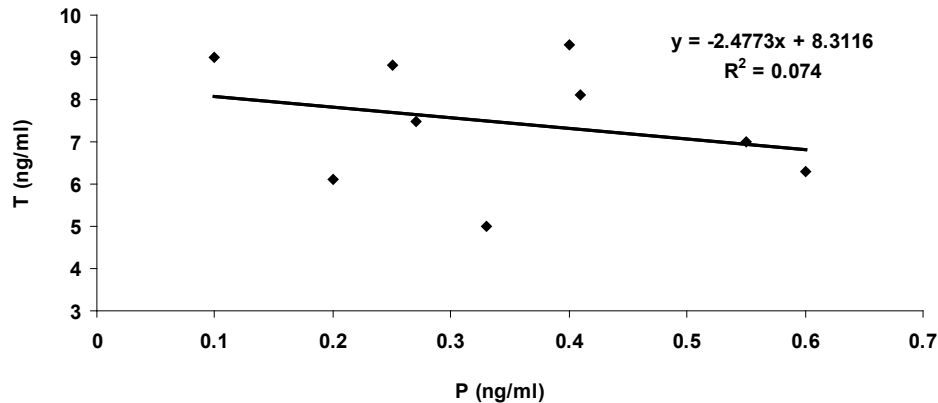


Fig 3. Relationship between P and T concentrations of seminal fluid.

spawning or stripping. Sex steroids are organic components which play an important role in hormonal control of the reproduction in fish including sturgeons [1, 5]. In this study, various levels of steroids were found in seminal plasma of Persian sturgeon. These steroids may be in relation to their levels in blood. As well as, several studies showed that urine could be one of potential sources of steroids in seminal fluid, although we did not carry out any analysis of urine in this study. Scott et al. (1991) [9] demonstrated that contamination of milt with urine during the hand stripping procedure is one of sources of steroids in the seminal fluid of plaice *Pleuronectes platessa* (L.,1758), dab *Limanda limanda* (L.,1758), flounder *Platichthys flesus* (L.,1758), goldfish *Carassius auratus* (L.,1758) and Rainbow trout. It is obvious that contamination of milt with urine complicates the precise analysis of steroids and probably other materials in seminal fluid. As a suggestion, simultaneous analysis of seminal fluid and urine and also to be sure of urinary bladder emptiness could be good strategies for precise analysis of chemical parameters and their changes in seminal fluid during reproduction. In the present study, the significant correlations were found between the concentrations of P, OHP and T in the seminal plasma. According to steroidogenesis pathway in fish, steroids were converted to each other. In fact, some steroids are precursor for next steroid as the significant correlations were found between the concentrations of P, OHP and T.

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