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Evaluation of the seminal plasma of captive and wild scale carp *Cyprinus carpio* var. *communis* in Kashmir, India

Ishrat Mohd, Farooz Ahmad Bhat* M. H. Balkhi, Tasaduq Hussain Shah, Bilal Ahmad Bhat, Feroz Ahmad Shah and Asifa Wali

Faculty of Fisheries, Sher-e-Kashmir University of Agricultural Sciences and Technology of Kashmir (SKUAST-K), Rangil, Ganderbal - 190 006, Jammu and Kashmir, India

Abstract

Evaluating gamete quality in fish is vital for aquaculture success. This study explores how the habitat, whether wild or farmed, affects the physical and biochemical composition of scale carp seminal fluid. Understanding these factors is essential for assessing the reproductive capabilities of fish species. The seminal plasma contained 70.64±10.75 mg dl^{-1} glucose, 1.99±1.15 g dl^{-1} total protein, 12.99±5.6 mg dl^{-1} triglyceride, 5.98±0.629 mg dl^{-1} cholesterol and 25.22±4.047 mg dl^{-1} urea in wild conditions, whereas in farmed conditions, the seminal plasma contained 85.06±9.29 mg dl^{-1} glucose, 0.917±0.62 g dl^{-1} total protein, 12.3±5.22 mg dl^{-1} triglyceride, 5.53±1.161 mg dl^{-1} cholesterol and 28.4±5.75 mg dl^{-1} urea. The physical parameters like mean sperm volume, mean sperm motility, mean movement duration, mean sperm density and mean pH recorded in the in the wild fishes were 2.393±1.64 ml, 75.038±10.162%, 50.367±13.92, 3.534±.272 109 ml and 8.29±.494, while the parameters in farmed fishes were 1.486±0.88 ml, 68.9±12.46%, 44.66±13.48 s, 3.84±0.181109 ml and 8.5±0.311 respectively. In correlation matrix, the sperm volume (R2=0.247, p<0.01), pH (R2=0.059, p<0.01) and total protein (R2=0.1882, p<0.01) showed significant positive correlations with sperm motility. Wild brooders outperformed cultured counterparts in several aspects, *viz*., milt volume, sperm motility percentage, duration, total protein, triglyceride and cholesterol levels. However, cultured males exhibited elevated glucose and urea concentrations, possibly indicating stress from captivity, handling, or confinement and increased urea due to ammonia in ponds. These findings underscore the importance of considering these parameters in standardising artificial fertilisation or cryopreservation techniques for common carp spermatozoa.

*Correspondence e-mail:

fabhat_fb@yahoo.com; ishratkq786@gmail.com
fabhat_fb@yahoo.com

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Introduction

The common carp *Cyprinus carpio* Linneus, 1758, is a widely distributed freshwater cyprinid fish (Economids, 1991). Because of its high growth rate and ability to resist harsh climatic conditions, the fish has been successfully introduced into freshwaters all over the world (Welcomme, 1988; Seegers *et al*., 2003). Ensuring consistent production of the species in aquaculture requires effective reproduction management, which in turn demands a thorough understanding of sperm characteristics, including both physical and biochemical characteristics. Use of high-quality spermatozoa for breeding under captive conditions has great importance in ensuring quality offspring production in aquaculture (Hajirezaee *et al*., 2010a). Effective gamete management in fish farms, can be accomplished by high fertilisation success (Billard *et al*., 1995). Study of sperm physiology of fish can offer fundamental knowledge for effectively managing fertilisation (Alavi and Cosson, 2005).

Both intrinsic gamete quality and quantity can influence fertilisation success for cryopreservation or short-term storage (Billard and Cosson, 1992; Dreanno *et al*., 1998). The constituents of a gamete (sperm), including seminal plasma and spermatozoa, can serve as indicators for assessing the quality of the gamete (sperm).

Basic research on seminal plasma contents such as metabolites, carbohydrates, vitamins, amino acids, fatty acids and other inorganic compounds can reveal a lot about sperm health (Rurangwa *et al*., 2004; Cabrita *et al*., 2008). Spermatocrit, sperm density, osmolarity and pH of seminal plasma, chemical composition of seminal plasma, motility, morphology and ultrastructure, fertilising capability, enzymatic activity, adenosine triphosphate (ATP) concentration, and several other biomarkers of sperm quality have all been examined (Billard and Cosson, 1992; Ciereszko and Dabrowski, 1993; Lahnsteiner *et al*., 1998). The most typical metrics used to measure spermatozoa quality in fish are sperm volume, concentration and motility, as well as the composition of seminal plasma (Linhart *et al*., 2000; Alavi and Cosson, 2006). The motility of spermatozoa is an important component in determining the fertilisation potential (Alavi *et al*., 2004). Several parameters influence spermatozoa motility, including pH (Alavi and Cosson, 2005), cations or osmolality (Alavia, 2007), and dilution ratio (Alavi *et al*., 2004) in both aqueous and diluent environments (Alavi and Cosson, 2006). The ionic and organic elements of the seminal fluid can indicate fertilisation potential. The reproductive potential of cultured fish for controlled production in aquaculture systems relies heavily on physical and biochemical features of the milt (Ciereszko *et al.,* 2000). For proper evaluation of a fish species' reproductive capabilities, knowledge of physical parameters and chemical composition of seminal plasma is required. In terms of mineral and organic contents of seminal plasma, however, some species-specific properties are important for artificial insemination or sperm preservation. In this backdrop the present study was undertaken to evaluate the seminal plasma of scale carp collected from captive and wild stocks.

Materials and methods

Source of broodstock and milt collection

For the experiment, two groups (Group A and B) of scale carp (male and female) were chosen and thirty brooders from each group were used for semen collection. Group A, comprised male brooders (Weight, W = 293±270.84 g; Total length, TL = 254.1±64.2 mm). sourced from captive stock maintained in the hatchery/farm of Faculty of Fisheries, Sher-e-Kashmir University of Agricultural Sciences and Technology of Kashmir (SKUAST-K) and in the Pandach fish farm (J&K state Govt. owned farm). Group B comprised wild male brooders (W=462.5±393.06 g; TL=311.1±77.12 mm) collected from Dal Lake during spawning season. Each male brood fish was anaesthetised and stripped once. Clean 15 ml graduated centrifuge tubes were used to collect the sperm, with utmost care taken to avoid contact with water, urine, blood, and faeces. The tubes were sealed and sent to the laboratory in ice (4°C) for analysis.

Evaluation of semen volume and pH

Milt volume was calculated using a digital pH meter (Model pH ep® Hanna instruments, Italy). Each pH measurement was carried out three times.

Sperm motility (%) and duration

Within the first hour after semen collection, the motility (percent) and duration (s) of spermatozoa in each sample were assessed at room temperature (25°C). To activate the sperm, 10 µl of milt was placed on a glass slide and 100 µl activation solution (0.3% NaCl) was added (Mims, 1991). Motility was observed using an Olympus CX31 microscope (×400, Olympus CX31). Only forward movements of spermatozoa were considered motile. The counting process was repeated three times and motility was expressed as percentage. The duration of spermatozoa motility was measured using a stopwatch from the start of forward spermatozoa movement after 3-5 s of activation to the end of the last spermatozoa movement in that field (× 400, Olympus CX31).

Sperm density

The sperm was diluted 1000 times in 0.3% NaCl. The density of spermatozoa was measured using a haemocytometer counting chamber and expressed as the number of cells per mililitre. A droplet of diluted milt was placed on a haemocytometer slide (ROHEM INDIA, B.S.748, I.S.10269) with a cover slip (Marienfeld, Laudakonigshofen, Germany) and counted under light microscopy (400, Olympus CK2, Tokyo, Japan). The number of spermatozoa was counted in at least 5 large squares (area 0.04 mm^2) of total 25 squares (one large square has 16 individual cells) after 3-5 min (to allow sperm sedimentation) and then the counted cells were computed to estimate the sperm density using the formula:

No. of cells per ml = Average No. of cells counted in the large squares $\times 10^4$ x Dilution factor

Evaluation of seminal plasma composition

After centrifuging the semen at 4000 rpm for 10 min at room temperature (20°C), the seminal plasma was collected and stored in Eppendorf vials at -20°C until analysis. To avoid contamination with spermatozoa, seminal plasma was centrifuged twice. A spectrophotometer (Systronic UV–VIS Spectrophotometer 117) was used to measure the levels of the metabolites (glucose, total protein, cholesterol, triglyceride and urea). The glucose oxidase/peroxidase (GOD/POD) method was used to measure the total glucose content in seminal plasma. Total protein concentration was determined by Biuret method; cholesterol by cholesterol oxidase/phenol + aminophenazone (CHOD/PAP) method; triglycerides by glycerophosphate oxidase-peroxidase (GPO-PAP) method and urea by modified Berthelot method. The methods (GOD/POD; CHOD/PAP; GPO-PAP and the modified Berthelot method) used for the estimation of semen metabolites provides high specificity and sensitivity particularly in the biological fluids like seminal plasma.

Statistical analyses

Statistical analyses were performed using the statistical software package SPSS16 for Windows. Significant difference between the wild and captive brooders was analysed by t test. Results are presented as means±SEM. The significance level was set at $p<0.05$. Correlations between spermatological parameters and plasma composition were estimated using Pearson's correlation test. The spermatozoa motility was used as dependent and other observed parameters as independent variables for linear regression.

Results and discussion

The physical parameters of the milt of *C. carpio* var. *communis* from wild and captive reared brood fishes are presented in Table 1. In the case of wild brooders, sperm volume, motility rate, duration, sperm density and pH were 2.393±1.64 ml, 75.038±10.162% 50.367±13.92 s, 3.53±0.272 $\times 10^9$ ml and 8.29±0.49 respectively whereas In case of captive brooders, the parameters recorded were 1.486±0.88 ml, 68.9±12.46%, 44.66±13.48 s, 3.84±0.181$\times 10^9$ ml and 8.5±0.311 respectively. Statistical analysis revealed non-significant difference in motility duration and pH between the two groups. The sperm volume and sperm motility (%) showed significant ($p<0.05$) difference between the two groups. The wild fish was having higher sperm volume and motility percentage than that of captive reared fish. In addition, the sperm density in the captive reared brooders was significantly higher ($p<0.01$) than that in the wild brooders (Table 2). The relationships of spermatozoa motility with sperm volume, pH, sperm density and total protein content are depicted in Fig. 1-4. Spermatozoa motility showed significant positive correlation with the sperm volume, while it presented significant negative correlation with sperm density ($R^2 = -0.48$; $p<0.05$) (Fig. 3).

Seminal plasma composition

The mean values, standard errors and ranges of the biochemical parameters are summarised in Table 1. The glucose (mg dl^{-1}) and total protein (g dl^{-1}) ranged from 35-81, 0.14-4.23 in wild and 71-99, 0.12-1.9 in captive brooders. The range of triglyceride (mg dl^{-1}) and cholesterol (mg dl^{-1}) was 4.3-21, 4.89-6.99 in wild and 4-20, 3.1-9 in captive brooders. Urea (mg dl^{-1}) content was in the range of 15-38.33 in wild and 21.89-45 in captive fishes. Biochemical parameters of milt from the wild and farmed scale carp are given in Table 2. In the case of wild brooders, the mean values (mg dl^{-1}) of glucose, protein, triglyceride, cholesterol and urea were 70.64±10.75; 1.99±1.15; 12.99±5.6; 5.98±.629 and 25.22±4.047 respectively and the mean values for captive brooders were 85.06± 9.29;

Table 1. Descriptive statistics of the physico-biochemical characters of the sperm of scale carp from wild and captive conditions

Parameter	Habitat	Range	Mean	Standard error
Sperm volume (ml)	Wild	0.3-5.6	2.39	0.29
	Farmed	0.1-3	1.48	0.17
Motility (%)	Wild	61.2-94	75.03	1.85
	Farmed	44-89	68.9	2.27
Motility duration (s)	Wild	20-85	50.36	2.54
	Farmed	20-69	44.66	2.46
Sperm density ($\times 10^9$ ml^{-1})	Wild	3.16-3.98	3.53	0.04
	Farmed	3.27-4.21	3.84	0.03
pH	Wild	7.2-8.9	8.29	0.09
	Farmed	7.9-8.9	8.5	0.05
Glucose (mg dl^{-1})	Wild	36-81	70.64	1.96
	Farmed	71-99	85.06	1.69
Protein (mg dl^{-1})	Wild	0.14-4.23	1.99	0.21
	Farmed	0.12-1.9	0.917	0.11
Triglyceride (mg dl^{-1})	Wild	4.3-21	12.99	1.023
	Farmed	4-20	12.307	0.95
Cholesterol (mg dl^{-1})	Wild	4.89-6.99	5.98	0.114
	Farmed	3.1-9	5.53	0.29
Urea (mg dl^{-1})	Wild	15-38.33	25.22	0.73
	Farmed	21.89-45	28.49	1.05

Table 2. Comparative analysis of physico-biochemical parameters of the milt of scale carp from wild and captive conditions

Parameters	Mean±SD		t value	P value
	Wild	Farmed		
Sperm volume (ml)	2.393±1.64	1.486±0.88	2.625	<0.05
Motility (%)	75.038±10.162	68.9±12.46	2.0899	<0.05
Duration (s)	50.367±13.92	44.66±13.48	1.61	>0.05
Sperm density ($\times 10^9$ ml^{-1})	3.534±.272	3.84±0.181	-5.148	<0.01
pH	8.29±.494	8.5±0.311	-1.905	>0.05
Glucose (mg dl^{-1})	70.64±10.75	85.06±9.29	-5.55	<0.01
Protein (mg dl^{-1})	1.99±1.15	0.917±0.62	4.48	<0.01
Triglyceride (mg dl^{-1})	12.99±5.6	12.3±5.22	0.494	>0.05
Cholesterol (mg dl^{-1})	5.98±0.629	5.53±1.61	1.4207	>0.05
Urea (mg dl^{-1})	25.22±4.047	28.4±5.75	-2.549	<0.05

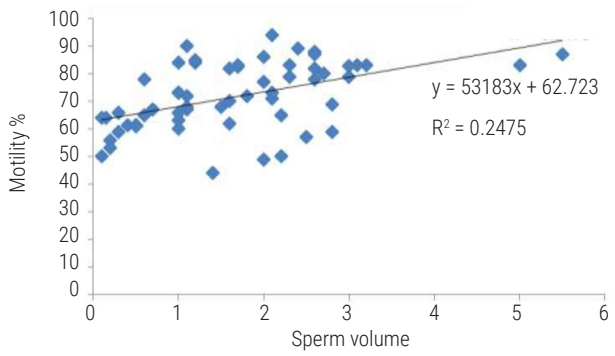

Fig. 1. Relationships between spermatozoa motility and sperm volume

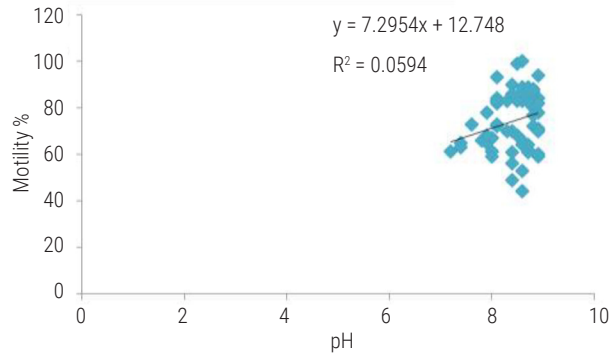

Fig. 2. Relationships between the spermatozoa motility and pH

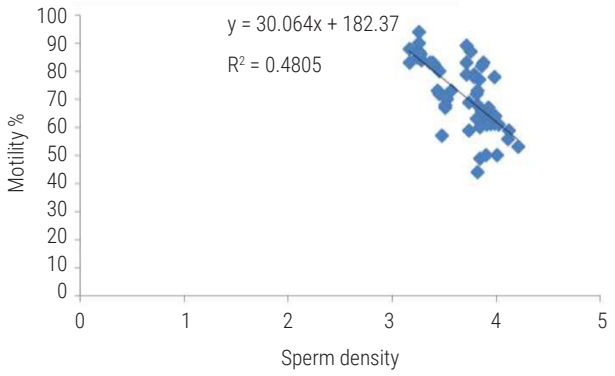

Fig. 3. Relationships between the spermatozoa motility and sperm density

0.917±0.62; 12.3±5.22; 5.53±1.61 and 28.4±5.75 respectively. The levels of glucose and urea observed in farmed scale carp were significantly higher ($p<0.05$) than those in the wild carp. In contrast, the concentration of total protein in cultured individuals was significantly lower than that in their wild counterparts ($p<0.05$), Cholesterol and triglyceride though higher in wild condition displayed insignificant variation between the two groups. The high level of glucose and urea in farmed males may be related to stress conditions (confinement, holding or handling). Captive rearing can negatively impact sperm traits in aquatic animals, including an increased percentage of abnormal spermatozoa, reduced number of sperm in spermatophores, reduced percentage of viable sperm (Leung-Trujillo and Lawrence, 1987) and the degeneration of the male reproductive tract (Talbot *et al*., 1989). High stocking densities, or crowding of fish in confined spaces, can lead to increased stress levels. Fish in overcrowded conditions may exhibit behaviours indicative of stress, such as aggressive interactions, reduced feeding and impaired growth. Elevated stocking densities can result in physiological changes, including increased glucose level and compromised immune function. No significant correlation was found between spermatozoa motility and other parameters of seminal plasma except total protein. Significant correlations were observed between sperm motility and total protein ($R^2 = 0.188$, $p<0.05$) (Fig. 4). The difference in seminal quality characteristics between farmed and wild scale carp was investigated in this study to determine gamete quality in the two groups. Male age and weight, sampling period, sampling method (Suquet *et al*., 1994), rearing conditions, nutrition, breeding seasonality, method of spawning induction, spawning behaviour (Rurangwa *et al*., 2004), feeding conditions and regime, environmental factors, or spawning time have all been linked to differences in sperm production (Bozkurt *et al*., 2006). The mean sperm volume in wild brooders (2.393±1.64 ml) was greater than in farmed individuals (1.486±0.88 ml) in this study. There was a significant difference in sperm volume between wild and farmed scale carp ($p<0.05$). The average sperm volume in farmed and wild fish was found to be similar to that reported by Belova (1981), who observed sperm volumes in the range of 1-9 ml in scale carp. Milt volume in scale carp was also reported to be 2.75 ml by Bozkurt (2006). The volume of milt in *Cyprinus carpio* ranged from 1.83 to 1.98 ml, according to Thamizhselvi and Thirumathal (2016).

Sperm density is an important factor in the determination of sperm quality (Suquet *et al*., 1992). Its role in fertilisation of spermatozoa is well documented and reported (Aas *et al*., 1991; Pool and Dillane, 1998). In the present study, the average sperm density of $3.534±0.27x10^9$ ml^{-1} in the wild and $3.8415×10^9±0.18$ ml^{-1} in captive brooders was recorded, which are in conformity with the results of Bozkurt *et al*. (2009a) for grass carp ($2.87-33.914 x10^9$ ml^{-1}). Chutia *et al*. (1998) have reported sperm density of $6.6x10^9$ sperm cells ml^{-1} in *C. carpio*. Thamizhselvi and Thirumathal (2016) recorded average sperm density of $2.25x10^9$ cells ml^{-1} in *C. carpio*. Lahnsteiner *et al*. (2000) observed sperm density of *C. carpio* as 0.5 to $1.0×10^{11}$ cells per ml of milt. Results of the present study revealed that sperm density of farmed fish was higher than that of wild fish and similar results have been reported by Hajirezaee *et al*. (2011).

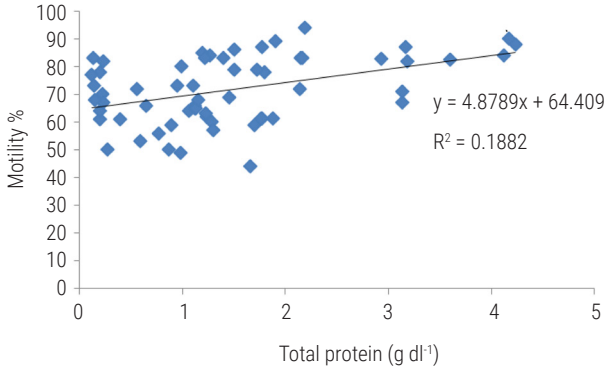

Fig. 4. Relationships between the spermatozoa motility and total protein

The motility and longevity of spermatozoa have a significant impact on fertilisation success. Spermatozoa motility, on the other hand, varies in intensity and duration not just across males, but also within individual males, depending on maturity (Tekin *et al*., 2003). Motility is a criterion used to assess the spermatozoa's quality (Terner, 1986). Fertilisation success is largely determined by sperm motility (Billard *et al*., 1995). In several teleost fish, sperm motility performance (% of motile sperm and time of motility) changes during the spawning season (Koldras *et al.,* 1996). In this study, the mean motility percentage and duration in wild fish were 75.03% and 50.36 s, respectively, while the values in farmed fish were 68.9% and 44.6 s, respectively. Although there was a significant difference in motility percent across the groups, the difference in motility duration was not significant. Scale carp had a mean sperm motility of 63.18% and a mean motility duration of 56.81 s, as per Bozkurt *et al*. (2009b). According to Bozkurt *et al.* (2008), grass carp motility percentages range from 70 to 95%, with durations ranging from 35 to 117 s. These findings are consistent with current findings for both farmed and wild fish. According to the results of this study, wild brooders had higher mean sperm motility and longer mean motility duration than farmed individuals and Hajirezaee *et al*. (2011) have reported similar findings. According to Skjaeraasen *et al.* (2009), wild males of cod had higher sperm velocity, motile cell percentages and progressive cell percentages than farmed counterparts. Many workers, however, claimed that there was no difference in sperm motility between wild and cultured brooders (Aydin *et al*., 2011). Striped and white sea bass that were collected from wild during the spawning season and brought to captivity produced milt with non-motile sperms (Berlinsky *et al*., 1997).

The sperm volume and motility had a significant positive correlation ($p<0.05$, $R^2 = 0.247$) (Fig. 1). Also sperm motility had a negative correlation with sperm density ($p<0.05$, $R^2 = 0.4805$) (Fig. 2). For *Cyprinus carpio,* the correlation between the two parameters (volume and motility rate) corresponds with Bozkurt *et al*. (2009b) and Nahiduzzman *et al.* (2014). The number of components involved in sperm motility is anticipated to decrease when seminal fluid production by the spermatic duct epithelium diminishes. This appears to be one of the reasons why wild males with greater motility rates were found to have higher spermatic duct epithelial secretion than farmed males in the current investigation.

In aquatic animals, a change in the pH of the external medium is one of the sperm activation factors (Stoss, 1983). The pH of the milt was found to be alkaline in both fish groups. There was no significant difference observed in pH between the wild and farmed fish. The pH of wild fish was 8.2±0.49 in this study, while the pH of farmed fish was 8.56±0.3. The pH of the milt measured in this study agrees with Bozkurt *et al.* (2009b), who found that the pH of scale carp milt ranged from 6.9 to 9.2. Verma *et al*. (2009) recorded pH of 7.8±0.07 in catla, 7.3±0.06 in rohu, 7.9±.05 in mrigal, 8.1±0.09 in kalbasu, 7.8±0.03 in silver carp and 7.9±0.06 in grass carp. There was a substantial positive relation between semen pH and motility ($p<0.05$, $R^2 = 0.059$) (Fig. 1). According to the relationship, pH is one of the most essential seminal plasma features controlling sperm activation. Morisawa and Morisawa (1986) and Billard *et al.* (1986) found comparable results (1995). According to them, an increase in external pH is responsible for the acquisition of motility in some salmonid fish during the passage of spermatozoa from the testis to the spermatic duct and the seminal fluid. pH also affects the final maturation of spermatozoa (Lahnesteiner *et al.,* 1998). pH polarises the cell membrane and promotes the movement of fish spermatozoa, according to Morisawa *et al.* (1999).

Seminal proteins defend spermatozoa from oxidative damage and microbial attack (White and Macleod, 1963; Dietrich *et al.*, 2010). Seminal plasma proteins, according to Lahnsteiner *et al*. (2004), extend the viability of rainbow trout spermatozoa as determined by sperm motility. During the present study, concentrations of total proteins were found as 1.99±1.15 g dl^{-1} in wild condition and 0.917±0.62 g dl^{-1} in farmed condition and are in conformity with the findings of Bozkurt *et al.* (2009b) for scale carp and Faramarzi (2012) for silver carp. Total protein concentration in the wild brooders was found to be significantly higher than that recorded for captive brooders. High protein concentration has been considered as positive characteristic of fish semen. The high protein content of fish sperm has long been seen as a favourable trait. Butts *et al*. (2011), who studied seminal plasma proteins in cod species found that they play an important role in spermatozoa viability, which agrees with the present findings. When Percin and Konyalioglu (2008) compared the serum biochemical profiles of captive and wild northern blue fin tuna (*Thunnus thunnus* L. 1758) in the Eastern Mediterranean, they discovered that the total protein content of wild tuna was higher than that of farmed fish. Total protein levels in wild trout were likewise higher than in farmed trout, according to Phillips *et al.* (1957). According to Phillips *et al*. (1957) for trout species, the cause for this variation is natural food and higher activity by wild fish. In both conditions, there is a significant association between the percentage of motility and total protein in seminal plasma of common carp, which could be related to the important role of motility in sperm cells (Lahnestiner *et al*., 2004).

The major sugar in seminal plasma is glucose. The presence of glucose in seminal plasma has been linked to the testes' increased energy demand during spermatogenesis or spermatozoa lipid synthesis (Soengas *et al*., 1993). Mean glucose level reorded from wild brooders was 70.643±10.75 mg dl^{-1} while that in captive brooders was 85±9.29 mg dl^{-1} during the current investigation. According to Kruger *et al*. (1984), glucose levels in carp species range from 9 to 100 mg dl^{-1}. The glucose content in wild common carp was 19.7 mg dl^{-1}, according to Bastami *et al.* (2012). A wide range of environmental stresses, such as captivity, hypoxic environment, and malnutrition, are thought to generate higher glucose concentrations in farmed scale carp compared to wild counterparts (Hardy and Audet, 1990; Cech *et al.,* 1996; Santos and Pacheco, 1996). High glucose levels may be linked to stressful situations (*i.e.,* confinement). The findings of this study are in agreement with the findings of Coz-Rakovac *et al*. (2005), who found that farmed *Dicentrarchus labrax* had much higher glucose levels than wild *D. labrax*. Similar findings have been reported in Dojo loach by Zhou *et al*. (2009).

Seminal plasma lipids are linked to spermatozoa metabolism and serve as energy sources for sperm motility in fish (Stoss, 1983; Piironen, 1994). The triglyceride concentration was found to be 12.99±5.605 mg dl^{-1} in the wild and 12.308±5.22 mg dl^{-1} in the farmed fish during the current study. Secer *et al*. (2004) found that the mean level of triacylglyceride in *C. carpio* seminal plasma was greater than that of *O. mykiss* (8 mg dl^{-1}), but it was near to that of *C. idella* (14.58 mg dl^{-1}) (Bozkurt *et al*. 2008b). Despite the fact that triglyceride levels in wild fishes were greater than in farmed ones, the difference in triglyceride levels between the two settings

was shown to be statistically insignificant. While researching the metabolic properties and hatching performance of cultured and wild Atlantic salmon (*Salmo salar*) eggs. Srivastava and Brown (1991) discovered that eggs collected from the wild had a higher lipid content than farmed eggs.

The cholesterol level was found to be 5.928±0.629 mg dl^{-1} in wild and 5.5±1.610 mg dl^{-1} in farmed fish. There is limited information about the role of cholesterol in seminal plasma (Billard et al., 1995). Cholesterol protects against environmental changes, particularly temperature fluctuations that occur when fish semen is expelled (Bozkurt *et al*., 2008). The cholesterol content measured in both groups during the present study was similar to that found in scale carp by Bozkurt *et al.* (2009b). The cholesterol level observed in scale carp during this investigation was lower than that reported by Bozkurt *et al*. (2008) and Bastami *et al.* (2012) for *Ctenopharyngodon idella* (12.02 mg dl^{-1}) but higher than that seen by Secer *et al*. (2004) for *Oncorhynchus mykiss* (2.55 mg dl^{-1}). According to Billard *et al.* (1995), the differences in the values could be attributed to differences in age, season, environment, and physiological circumstances of fish.

Urea in semen may cause reduced sperm motility and fertilising ability (Dreanno *et al.,* 1998) affecting the variability of other semen parameters (Glogowski *et al.,* 2000). During the present study, notable concentrations of urea were determined in seminal plasma with 25.221±4.047 mg dl^{-1} recorded in wild brooders and 28.497±5.75 mg dl^{-1} in captive brooders . The urea content in scale carp was reported to be 24.457.96 mg dl^{-1} by Bozkurt *et al.* (2009b), which agrees with the current results in both groups. Secer *et al.* (2004) reported a urea content of 3.16 mg dl^{-1} in *O. mykis*. The urea concentration of mirror carp was found to be between 38 and 97 mg dl^{-1} (Bozkurt *et al.,* 2009a). The mean serum urea levels in cultured individuals were higher than those in wild individuals, according to our findings. The amount of ammonia-N in intensively cultured ponds tended to be higher than in the wild environment, since high ammonia-N levels combined with hypoxic conditions generate an alternative osmotic pressure, which appears to be responsible for the higher urea levels in farmed scale carp (Shen *et al*., 1991).

The study concludes that physical and biochemical characteristics of seminal plasma are critical factors in determining sperm quality in scale carp. The sperm motility rate, motility duration, and sperm volume of scale carp from the wild were all higher compared to captive brooders . In addition, total protein and other metabolites were also found to be higher in wild fish. The sperm quality of wild scale carp was shown to be higher than that of farmed scale carp. Better spermatozoa motility contributes considerably to achieving a higher fertilisation rate. This information can greatly aid in the enhancement of captive reproductive management, particularly the efficacy of fertilisation techniques and semen storage, including cryopreservation.

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References

Aas, G. H., Refstei, T. and Gjerde, B. 1991. Evaluation of milt quality of Atlantic salmon. *Aquaculture,* 95: 125-132. https://doi.org/10.1016/0044-8486(91)90079-M.

Alavi, S. M. H. and Cosson, J. 2005. Sperm motility and fertilizing ability in the Persian sturgeon (*Acipenser persicus*). *Aquac. Res*., 36: 841-850. https://doi.org/10.1111/j.1365-2109.2005.01292.x.

Alavi, S. M. H. and Cosson, J. 2006. Sperm motility in fishes. (II) Effects of ions and osmolality: A review. *Cel. Biol. Int.,* 30: 1-14. https://doi.org/10.1016/j.cellbi.2005.06.004

Alavia, S. M. H., Psenicka, M., Rodina, M., Policar, T. and Linhart, O. 2007. Changes of sperm morphology, volume, density and motility and seminal plasma composition in *Barbus barbus* (Teleostei: Cyprinidae) during the reproductive season. *Aquat. Liv. Resour.,* 21: 75-80.

Aydin, I., Sahin, T., Polat, H. and Kucuk, E. 2011. A study on the spermatological characteristics of hatchery-reared flounder (*Platichthys flesus luscus* Pallas, 1814). *J. Fish. Sci. Com,* 5(4): 270-278.

Bastami, K., Rezaei, M., Haghparast, S., Eghtesadi Araghi, P., Najafi, H. E., Khansari, A., Jam, A. and Faraz-Ghasemi, A. 2012. A comparative study on the biochemical parameters of seminal and blood plasma and their correlation in the wild common carp (*Cyprinus carpio*). *Clin. Pathol*., 21: 5781-784.

Belova, N. V. 1981. The ecological and physiological peculiarities of sperm in pond cyprinids. Communication I. Production and ecological and physiological peculiarities of the sperm of cyprinids. *J. Ichthyo.,* 21: 90-102.

Berlinsky, D. L., William, K., Hodson, R. G. and Sullivan, C. V. 1997. Hormone induced spawning of summer flounder *Paralichthys dentatus. J. World Aquac. Soc.,* 28: 79-86.

Billard, R. 1986. Spermatogenesis and spermatology of some teleost fish species. *Reprod. Nut. Devel.,* 26: 877-920.

Billard, R. and Cosson, M. P. 1992. Some problems related to the assessment of sperm motility in freshwater fish. *J. Exp. Zool.,* 261: 122-131. https://doi.org/10.1002/jez.1402610203.

Billard, R., Cosson, J., Crim, L. W. and Suquet, M. 1995. Biology of sperm and artificial reproduction in carp. *Aquaculture,* 124: 95-112.

Bozkurt, Y. 2006. Relationship between body condition and spermatological properties in scaly carp (*Cyprinus carpio*) semen. *J. An. Veter. Adv.,* 5(5): 412-414.

Bozkurt, Y., Ogretmen, F. and Secer, F. S. 2009a. Effect of different extenders and storage periods on motility and fertilisation success of grass carp (*Ctenopharyngodon idella*) sperm during spawning season. *J. Agric. Sci.,* 15 (3): 277-284. https://doi.org/10.1501/Tarimbil_0000001101.

Bozkurt, Y., Secer, S., Bukan, N., Akcay, E. and Tekin, N. 2006. Relationship between body condition, physiological and biochemical parameters in brown trout (*Salmo trutta fario*) sperm. *Paki. J. of Bio. Sci.,* 9(5): 940-944.

Bozkurt, Y., Ogretmen, F., Ercin, U. and Yildiz, U. 2008. Seminal plasma composition and its relationship with physical spermatological parameters of grass carp (*Ctenopharyngodon idella*) semen: With emphasis on sperm motility. *Aquac. Res.,* 39:1666-1672. https://doi.org/10.1111/j.1365-2109.2008.02041.x.

Bozkurt, Y., Ogretmen, F., Secer, F. S. and Ercin, U. 2009b. Relationship between seminal plasma composition and spermatological parameters in scaly carp (*Cyprinus carpio*). *J. Anim. Vet. Adv.,* 8: 2745-2749.

Butts, I. A. E., Trippel, E. A., Ciereseszko, A., Soler, C., Alavi, S. M. H., Litvak, M. K. and Babaik, I. 2011. Seminal plasma biochemistry and spermatozoa

characteristics of Atlantic Cod (*Gadus morhua*) of wild and cultivated origin. *Comp. Biochem. Physiol.,* 159: 16-24. https://doi.org/10.1016/j.cbpa.2011.01.014.

Cabrita, E., Robles, V. and Herraez M. P. 2008. Sperm quality assessment. In: Cabrita, E., Robles, V. and Herráez, M. P. (Eds.), *Methods in reproductive aquaculture: Marine and freshwater species. Biology Series*. CRC Press, Taylor and Francis, London, UK, pp. 9: 31-47. https://doi.org/10.1201/9780849380549.

Cech, J. J., Bartholow, S. D., Young, P. S. and Hopkins, T. E. 1996. Striped bass exercise and handling stress in freshwater: Physiological responses to recovery environment. *Tran. Am. Fish.. Soc*., 125: 308-320.

Chutia, I. P., Krishna, G. and Chaudhary, A. 1998. In: Ponniah, A. G., Das, P. and Verma, S. R. (Eds.), *Fish genetics and biodiversity conservation,* Nature conservators, Muzaffarnager, Uttar Pradesh, India, pp. 205-213.

Ciereszko, A., Glogowski, J. and Dabrowski, K. 2000. Biochemical characteristics of seminal plasma and spermatozoa of freshwater fishes. In: Tiersch, T. R. and Mazik, P. M. (Eds.), *Cryopreservation in aquatic species*. World Aquaculture Society, Baton Rouge, Louisiana, USA, pp. 20-48.

Coz-Rakovac, R. Strunjak-perovic, I., Hacmanjek, M., Topic, P. N., Lipez, Z. and Sostaric, B. 2005. Blood chemistry and histological properties of wild cultured seabass (*Dicentrarchus labrax*) in the North Adriatic Sea. *Vete. Res. Commun*., 29: 677-687. https://doi.org/10.1007/s11259-005-3684-z.

Dietrich, G. J., Dietrich, M., Kowalski, R. K., Dobosz, S., Karol, H., Demianowicz, W. and Glogowski, J. 2010. Exposure of rainbow trout milt to mercury and cadmium alters sperm motility parameters and reproductive success. *Aquat. Toxicol.,* 97(4): 277-284 https://doi.org/10.1016/j.aquatox.2009.12.010.

Dreanno, C., Suquet, M., Desbruyeres, E., Cosson, J., Delliou, H. and Billard, R. 1998. Effect of urine on semen quality in turbot (*Psetta maxima*). *Aquaculture,* 169: 247-262.

Economids, P. S. 1991. *Check list of freshwater fishes of Greece (recent status of threat and protection)*. Hellenic Society for the Protection of Nature, Special publication 48 p. https://doi.org/10.1080/14634980601027855.

Faramarzi, M. 2012. Assessment of reproductive parameters in silver carp (*Hypophthalmichthys molitrix*) *World J. Fish Mari. Sci.,* 4(3): 244-248.

Glogowski, J. Kwasnik, M., Piros, B., Dabrowski, K., Goryczko, K., Dobosz, S., Kuzminski, H. and Ciereszko, A.2000. Characterization of rainbow trout milt collected with a catheter: Semen parameters and cryopreservation success. *Aquac. Res*., 31: 289-296. https://doi.org/10.1046/j.1365-2109.2000.00400.x

Hajirezaee, S. and Rafiee, G. R. 2010a. Stress responses of Persian sturgeon, *Acipenser persicus* to repetition of a management stressor (hand stripping of milt). *J. Appl. Biol.. Sci.,* 4: 9-12.

Hajirezaee, S., Rafiee, G. R. and Hushangi, R. 2010b. Comparative analysis of milt quality and steroid levels in blood and seminal fluid of Persian sturgeon males, *Acipenser persicus* during final maturation induced by hormonal treatment. *Biology,* 66(1): 160-169. https://doi.org/10.2478/s11756-010-0140-5.

Hajirezaee, S., Rafiee, G. R. and Hushangi, R. 2011. Comparative analysis of milt quality and steroid levels in blood and seminal fluid of Persian sturgeon males, *Acipenser persicus* during final maturation induced by hormonal treatment. *Biologia,* 66(1): 160-169.

Hardy, D. and Audet, C. 1990. Evaluation of plasma glucose as an indicator of mild chronic stress in brook charr (*Salvelinus fontinalis*). *Bull. Aquac. Assoc. Can.,* 90: 54-56.

Koldras, M., Loir, M., Maise, G. and Le-Gac, F. 1996. Study of the composition of seminal fluid and of sperm motility along the genital tract, during the spawning season, in rainbow trout (*Oncorhynchus mykiss*). *Aquat. Liv. Resour.,* 9: 337-345.

Kruger, J. C., Smit, G. L., Vuren, J. H. J. and Ferreira, J. T. 1984. Some chemical and physical characteristics of the semen of *Cyprinus carpio* L. and *Oreochromis mossambicus. J. Fish. Biol.,* 24: 263-272. https://doi.org/10.1111/J.1095-8649.1984.TB04797.X

Lahnsteiner, F., Berger, B., Weismann, T. and Patzner, R. A. 1998. Determination of semen quality of the rainbow trout, *Oncorhynchus mykiss*, by sperm motility, seminal plasma parameters and spermatozoa metabolism. *Aquaculture,* 16: 163-181. https://doi.org/10.1016/S0044-8486(98)00243-9.

Lahnsteiner, F., Berger, B., Horvath, A., Urbanyi, B. and Weismann, T. 2000. Cryoperservation of spermatozoa in cyprinid fishes. *Therioge,* 54: 1477-1498. https://doi.org/10.1016/s0093-691x(00)00469-6

Lahnsteiner, F., Mansour, N. and Berger, B. 2004. Seminal plasma proteins prolong the viability of rainbow trout (*Onchorhynchus mykiss*) spermatozoa. *Therioge,* 62: 801-808. https://doi.org/10.1016/j.theriogenology.2003.12.001.

Leung-Trujillo, J. R. and Lawrence, A. L. 1987. Observations on the decline in sperm quality of *Penaeus setiferus* under laboratory conditions. *Aquaculture,* 65: 363-370. https://doi.org/10.1016/0044-8486(87)90249-3.

Linhart, O., Rodina, M. and Cosson, J. 2000. Cryopreservation of sperm in common carp *Cyprinus carpio* Sperm motility and hatching success of embryos. *Cryobiology,* 41: 241-250. https://doi.org/10.1006/cryo.2000.2284.

Mims, S. D. 1991. Evaluation of activator solutions, motility duration and short term storage of paddle fish spermatozoa. *J. World Aquac. Soc.,* **22:** 224-229. https://doi.org/10.1111/j.1749-7345.1991.tb00738.x.

Morisawa, M., Hirano, T. and Suzuki, K. 1979. Changes in blood and seminal plasma composition of the mature salmon (*Oncorhynchus keta*) during adaptation to freshwater. *Comp. Biochem. Physiol.,* 64: 325-329.

Morisawa, M., Oda, S., Yoshida, M. and Takai, H. 1999. The Male Gamete: From Basic Knowledge to Clinical Applications (Ed. Gagnon, C.), Cache River Press, Vienna, USA 1999, pp. 149-160.

Morisawa, S. and Morisawa, M. 1986. Acquisition of potential for motility in Rainbow trout and Chum salmon. *J. Exp. Biol.,* 126: 89-96. https://doi.org/10.1242/jeb.126.1.89.

Nahiduzzaman, M., Akter, S., Hassan, M. M., Azad Shah, A. K. M. and Hossain, M. A. 2014. Sperm biology of artificially induced common carp, *Cyprinus carpio* (Linnaeus, 1758) *Int. J. Fish. Aquat. Stud.,* 1(6): 27-31.

Percin, F. and Konyalioglu, S. 2008. Serum biochemical profiles of captive and wild northern bluefin tuna (*Thunnus thynnus* L. 1758) in the Eastern Mediterranean. *J. Fish Biol.,* 56: 1-20. https://doi.org/10.1111/j.1365-2109.2008.01954.x.

Phillips, A. M., Brockway, D. R., Lovelace, F. E. and Podollak, H. A. 1957. Chemical comparison of hatchery and wild brook trout. *The Progressive Fish Culturist*, 19(1): 19-25.

Piironen, J. 1994. Composition and cryopreservation of sperm from some Finnish freshwater teleost fish. *Fin. Fish. Res.,* 15: 27-48.

Poole, W. R. and Dillane, M. G. 1998. Estimation of sperm concentration of wild and reconditioned trout. *Salmo trutta* L. *Aquac.Res.,* 29: 439-445. https://doi.org/10.1046/j.1365-2109.1998.00223.x.

Santos, M. A. and Pacheco, M. 1996. Stress biomarkers recovery in clean water and secondary treated pulp mill effluent. *Ecol. Environ.,* 35:96-100.

Secer, S., Tekin, N., Bozkurt, Y., Bukan, N. and Akçay, E. 2004. Correlation between biochemical and spermatological parameters in rainbow trout (*Oncorhynchus mykiss*) semen. *Isr. J. Aquac. Bamidgeh,* 56: 274-280. https://doi.org/10.46989/001c.20390.

Seegers, L. D., Vos, L. and Okayo, D. O. 2003. Annotated checklist of the fresh water fishes of Kenya from Lake Victoria. *J. East Afr. Nat. Hist.,* 92: 11-47. https://doi.org/10.2982/0012-8317(2003)92[11:ACOTFF]2.0.CO;2.

Shen, X. M., Zhang, H. Y. and Hua, R. 1991. Effect of environmental factors on haematological characters of blunt-snout bream **(***Megalobrama amblycephala) Ac. Ecol.. Sini.,* 11: 92-94.

Skjaeraasen, J. E., Mayer, I., Meager, J. J., Rudolfsen, G., Karlsen, O., Haugland, T. and Kleven, O. 2009. Sperm characteristics and competitive ability in farmed and wild cod. *Mar. Ecol Prog. Ser.,* 375: 219-228. https://doi.org/10.3354/meps07774.

Soengas, J. L., Sumatran, B., Barciella, P., Aldegunde, M. and Rozas, J. 1993. Changes in carbohydrates metabolism in domesticate rainbow trout, *Onchorhynchus mykiss* related to spermatogenesis. *Comp. Biochem. Physiol.*, 105: 665-671.

Srivastava, R. K. and Brown, J. A. 1991. The biochemical characteristics and hatching performance of cultured and wild Atlantic salmon (*Salmo salar*) eggs. *Can. J. Zool.,* 69: 2436-2441. https://doi.org/10.1139/z91-342

Stoss, J. 1983. Fish gamete preservation and spermatozoan physiology. In: Hoar, W. S., Randall, D. J. and Donaldson, E. M. (Eds.), *Fish physiology,* Academic Press, NewYork, USA, pp. 305-350. https://doi.org/10.1016/S1546-5098(08)60306-4.

Suquet, M. J., Omnes, Y., Normant and Fauvel, C. 1992. Assessment of sperm concentration and motility in turbot *Scophthalmus maximus. Aquaculture,* 101: 177-183. https://doi.org/10.1016/0044-8486(92)90241-C.

Suquet, M., Billard, R., Cosson, J., Dorange, G., Chauvaud, L., Mugnier, C. and Fauvel, C. 1994. Sperm features in turbot (*Scophthalmus maximus*): a comparison with other freshwater and marine fish species. *Aquati. Livi. Resour.,* 7: 283-294.

Talbot, P., Howard, D., Leung-Trujillo, J., Lee, T. W., Li, W.Y., Ro, H. and Lawrence, A. L. 1989. Characterization of male reproductive tract degenerative syndrome in captive penaeid shrimp (*Penaeus setiferus*). *Aquaculture*, 78: 365-377. https://doi.org/10.1016/0044-8486(89)90112-9.

Tekin, N., Secer, S., Akcay, E., Bozkurt, Y. and Kayam, S., 2003. The effect of age on spermatological properties in rainbow trout (*Oncorhynchus mykiss* W., 1792). *Turk. J. Vet. Anim. Sci.,* 27: 37-44.

Terner, C. 1986. Evaluation of salmonid sperm motility for cryopreservation. *Progressive Fish. Culturist,* 48: 230-232.

Thamizhselvi, N. and Thirumathal, K. 2016. A study of interrelationship between physico-chemical characteristics of water and spermatological qualities of *Cyprinus Carpio. Int. J. Fish. Aqat. Stud.,* 4(3): 621-625.

Verma, D. K., Routray, P., Dash, C., Dasgupta, S. and Jena, J. K. 2009. Physical and biochemical characteristics of semen and ultrastructure of spermatozoa in six carp species. *Turk. J. Fish. Aquat. Sci.,* 9: 67-76.

Welcomme, R. L. 1988. International introductions of inland aquatic species. *FAO Fisheries Technical Paper No.* 285. Food and Agriculture Organisation of the United Nations, Rome, Italy, pp. 237-278.

White, I. and Macleod, J. 1963. Composition and physiology of semen. In: Hartman, C. G. (Ed.), *Mechanisms concerned with conception***.** Pergamon Press, London, UK, pp. 135-172.

Zhou, X., Li, M., Abbas, K. and Wang, K. 2009. Comparison of haematology and serum biochemistry of cultured and wild Dojo loach *Misgurnus anguillicaudatus. Fis. Physiol. Biochem.,* 35: 435-441. https://doi.org/10.1007/s10695-008-9268-4.