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Effect of different salinities on breeding and larval development of common carp, *Cyprinus carpio* (Linnaeus, 1758) in inland saline groundwater



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ABSTRACT

A 90 days experiment was conducted to study the effect of different salinities (0, 5, 10 and 15 ppt) on breeding and larvae development of *Cyprinus carpio* using inland saline groundwater (ISGW). At the end of the experiment, average weight of the matured brooders were 370 ± 20 g, 342 ± 26 g, 275 ± 15 g, 167.5 ± 17.50 g at the salinities of 0, 5, 10 and 15 ppt, respectively. Among different salinities, relatively high gonad weight, fecundity, sperm motility, fertilization rate, embryonic and larval development was observed at the salinities of 0 and 5 ppt. The results showed that the stripping response, latency period, fertilization, and hatching rate were significantly (P < 0.05) reduced at the salinity of 15 ppt. The percentage of larvae survival were 69.40 \pm 1.76, 74.28 \pm 2.22, 49.98 \pm 2.04 and 28.78 \pm 5.34 at the salinities of 0, 5, 10 and 15 ppt, respectively. Long hatching duration (68–70 h) and high levels of larvae deformity (71.21 \pm 5.34%) was also observed at 15 ppt. Thus, it can be concluded that 5 ppt of ISGW can provide the best results for the production of *C. carpio* seed and could be useful in developing a hatchery technology for *C. carpio*.

1. Introduction

India is the second largest fish producer in the world with a production of 12.6 million metric tonnes during 2017-18. The fishery sector of India (both culture and capture) contributes 5.23% to the Gross Domestic Production (GDP) of the agriculture sector (SOFIA, 2018). India has tremendous potential to increase the fish production/ productivity by utilizing the untapped resources for commercial aquaculture. In India, around 8.62 million hectare of agricultural lands have been critically affected by the problem of soil salinity and around 40% of total inland saline soils are located in the states of Haryana, Uttar Pradesh, Punjab, and Rajasthan (Lakra et al., 2014). The inland saline soils are not suitable for agriculture and often considered as barren land. Salinization of inland water occurs due to either primary (natural processes such as rainfall, rock weathering) or secondary (anthropogenic) factors. In arid/semi-arid areas, factors such as excessive usage of irrigation, waterlogging, poor drainage, and indiscriminate use of inorganic fertilizers would lead to increase the salt content of the soil over time and subsequently causes salinization (Beresford et al., 2004). These lands have been converted to ponds for aquaculture practices (Inland saline aquaculture) and different candidate species along with refined aquaculture practices/hatchery technologies are being developed in India. Salt tolerant freshwater fish species such as tilapia

(Oreochromis niloticus), goldfish (Carassius auratus), common carp (Cyprinus carpio), riverine catfish (Pangasianodon hypophthalmus) and Amur carp (Cyprinus carpio haematopterus) were standardized as good candidate species for inland saline aquaculture (Wang et al., 1997; Saoud et al., 2003; Sawant et al., 2001; Kumar et al., 2017; Singh et al., 2019). Among these species; Cyprinus carpio (Linnaeus, 1758) popularly known as common carp, is one of the best candidate species for inland saline aquaculture as it has good domestic market and can tolerate moderate fluctuations in salinity and temperature (Whiterod and Walker, 2006). Nevertheless, C. carpio seed/fingerlings need to be procured from distant places and it often causes mortality of fingerlings. These factors would escalate the production cost and reduce the economic benefit from inland saline aquaculture. Developing a hatchery technology for C. carpio using inland saline ground water could be a viable option to address the above issue. Malik et al. (2018) has investigated the effect of salinity on larval development of C. carpio using artificial seawater and reported a range of 0–10 ppt as salinity tolerance limit for *C. carpio*. However, the ionic composition would be different between inland saline ground water (ISGW) and artificial seawater. Hence, the effect of ISGW on breeding and larval development of fish could be different from artificial seawater. With this background, the present study was carried out with an objective of studying the effect of ISGW salinities on common carp breeding and larval development. The findings of the

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present study are useful in developing a hatchery technology for common carp using ISGW.

2. Material and methods

2.1. Experimental site and design

The experiment was carried out at Rohtak regional centre, ICAR-Central Institute of Fisheries Education, Rohtak, Harvana, India. The experiment used completely randomized design (CRD) with three treatments (5, 10 and 15 ppt) and a control (0 ppt) in duplicates. Inland saline groundwater of 15 ppt (treatment T3) was pumped from a borewell to a non-drainable earthen pond. Later, the water was diluted with freshwater to achieve salinity of 5 and 10 ppt for treatment T1 and treatment T2, respectively. Freshwater with a salinity of 0 ppt was used for control groups (C). Common carp fingerlings (n = 840) with an average size of 32 \pm 2.5 g were reared separately in different earthen ponds ($21 \times 10 \times 1.50$ m), which corresponds to different treatment and control groups. Pelleted feed with a protein content of 32% was given twice a day at a rate of 2% of the fish body weight. After 90 days of culture, healthy brooders (n = 18) with an average weight of $370 \pm 20 \text{ g}$ (0 ppt), $342 \pm 26 \text{ g}$ (5 ppt), $275 \pm 15 \text{ g}$ (10 ppt) and 167.5 ± 17.50 (15 ppt) were selected and transferred to respective breeding tanks (FRP tanks with a capacity of 300 L). Breeding tanks were prepared for each treatment (5, 10 and 15 ppt) and control (0 ppt) in triplicates. The water for breeding tanks was disinfected using bleaching powder at a dosage of 15 mg L^{-1} and aerated for 48 h before releasing the brooders.

2.2. Collection of gametes, artificial fertilization, and incubation of eggs

Cyprinus carpio brooders (n = 9) were maintained in a sex ratio of 2:1 (females: males) in breeding tanks of different salinities (0, 5, 10 and 15 ppt). The carp pituitary extract was prepared following Divaware et al. (2007) method. It was administered at a dose of 6 mg kg^{-1} body weight (BW) to a female fish in two equal doses at an interval of 4 h while the male fish was given a single dose of 2 mg kg^{-1} BW. Eggs and milt were stripped from control (0 ppt) and treatment T1 (5 ppt) after 4 h post injection as they showed oozing of eggs. Whereas, in treatment T2 (10 ppt) and T3 (15 ppt), egg oozing was observed after 10 h post injection. Accordingly, stripping was done in T2 and T3 group fish after 10 h post administration of CPE. The striped eggs were weighed using electronic weighing balance. The adhesiveness of eggs was removed (degumming) by using cream milk (fat: 26-28%) method described by Khan et al. (1986). Around 1 µl of milt was taken for assessing the sperm motility. Later, the eggs were fertilized with the remaining milt by adding freshwater as a hydrating agent. Thereafter, the fertilized eggs were incubated separately in FRP tanks (300 eggs in each tank) at different salinities (0, 5, 10 and 15 ppt) in triplicates. All treatment processes were approved by the ethics and animal care committee of ICAR-Central Institute of Fisheries Education, Mumbai India.

2.3. Evaluation of sperm motility

The sperm motility was observed under a microscope at a magnification range of 10–20X. A drop of fish sperm (1 μ l) was placed on glass slide and 100 μ l of activating solution (0.3% w/v NaCl) was added to estimate the sperm motility. The motility was observed under microscope until around 85% of sperm lost their progressive movement and the time (in seconds) taken for this process was recorded using stopwatch (Ochokwu et al., 2015).

2.4. Water quality parameters of incubation tank

The optimum water quality was maintained by the flow-through system. Water quality parameters were determined regularly according to APHA, (2012). The parameters such as dissolved oxygen (DO), pH, and temperature were measured daily, whereas ammonia, nitrite, total alkalinity, total hardness, calcium, magnesium and potassium content were estimated twice a week. The flow of water in every incubation unit was adjusted via a flow-through system with partial exchange of the water, resulting in a complete turnover of the water in 24 h.

2.5. Latency period, fecundity, stripping response, fertilization rate, rate of embryonic and larval development

The latency period is described as the time interval between injection of the female fish and stripping of eggs. It is varied according to the degree of water salinity. The other parameters were estimated as per the following equations.

Total fecundity =

Number of eggs counted in the sample x weight of both ovary Weight of sample (1 g)

Spawning fecundity =
$$\frac{\text{Total fecundity}}{\text{Weight of sample (1 g)}}$$
 (2)

Stripping response =
$$\frac{\text{Weight of stripped eggs x 100}}{\text{Total gonad weight}}$$
 (3)

Fertilization rate (%) =
$$\frac{\text{Number of fertilized eggs x 100}}{\text{Total number of eggs}}$$
 (4)

Hatching rate (%) =
$$\frac{\text{Total number of hatchlings x100}}{\text{Total number of fertilized eggs}}$$
 (5)

Few samples (fertilized eggs) were observed at regular intervals for the embryonic and larvae developmental stages with a magnification range of 10–40X.

2.6. Mortality and deformation

The unfertilized eggs (dead/moldy eggs) were removed periodically at a regular interval during the entire experiment to prevent the fungal growth. Eggs were considered dead, if they turned opaque and white or if heartbeat had stopped. The percentage of normal and deformed larvae (including dead ones) was calculated as per the given equations.

Normal larvae rate (%) =
$$\frac{\text{Total number of normal larvae x 100}}{\text{Total number of hatchlings}}$$
 (6)

Deformed larvae (%) =
$$\frac{\text{Total number of dead larvae x 100}}{\text{Total number of hatchlings}}$$
 (7)

2.7. Statistical analysis

The data was analyzed by one-way Analysis of Variance (ANOVA) followed by Duncan's multiple range test in order to determine the significant differences between the means with a statistical package SPSS version 22.0 (Kirkpatrick and Feeney, 2014).

3. Results

3.1. Water quality

Water quality parameters observed throughout the experimental period are given in Table 1. Dissolved oxygen (6.40–6.85 mg L⁻¹), pH (7.55–7.85), hardness (420–3250 mg L⁻¹), magnesium (47.0–671.0 mg L⁻¹), potassium (6.25–16.85 mg L⁻¹), and sodium (965–1895 mg L⁻¹), calcium (90–195 mg L⁻¹) alkalinity (92.5–128.80 mg L⁻¹), temperature (26.5 °C), total ammonia nitrogen (0.07–0.27 mg L⁻¹) and nitrite nitrogen (0.02–0.06 mg L⁻¹) were

Table 1

Physico-chemical parameters during incubation of C. carpio eggs at different salinities.

Parameters	Salinities				
	0 ppt (C)	5 ppt (T1)	10 ppt (T2)	15 ppt (T3)	
Dissolved Oxygen (mg L^{-1})	$6.75^{a} \pm 0.01$	$6.85^{a} \pm 0.01$	$6.40^{\rm b}$ \pm .0.22	$6.40^{b} \pm 0.01$	0.01
Temperature (⁰ C)	$26.50^{\rm a} \pm 0.50$	$26.50^{\rm a} \pm 0.50$	$26.0^{a} \pm 0.50$	$26.50^{\rm a} \pm 0.50$	0.47
Free CO_2 (mg L ⁻¹)	Nil	Nil	Nil	Nil	
pH	$7.55^{\rm c} \pm 0.50$	$7.65^{\rm c} \pm 0.50$	$7.85^{\rm b} \pm 0.05$	$8.05^{a} \pm 0.05$	0.01
Ammonia-N (mg L^{-1})	$0.07^{\rm a} \pm 0.03$	$0.05^{a} \pm 0.05$	$0.07^{a} \pm 0.22$	$0.08^{\rm a} \pm 0.11$	0.56
Nitrite-N (mg L^{-1})	$0.02^{\rm a} \pm 0.01$	$0.03^{a} \pm 0.02$	$0.04^{a} \pm 0.01$	$0.06^{a} \pm 0.00$	0.17
Alkalinity (mg L $^{-1}$)	$92.5^{\circ} \pm 2.50$	$124^{ab} \pm 4.00$	$107^{ab} \pm 1.00$	$128^{a} \pm 8.00$	0.01
Hardness (mg L^{-1})	$420^{\rm c} \pm 20.00$	$1975^{\rm b} \pm 125.00$	$2000^{\rm b} \pm 100.00$	$3250^{\rm a} \pm 550.00$	0.01
Ca^{2+} content (mg L ⁻¹)	$90^{\rm a} \pm 2.00$	$130^{\rm a} \pm 10.00$	$150^{\rm a} \pm 10.00$	$195^{\rm a} \pm 69.00$	0.33
Mg^{2+} content (mg L ⁻¹)	$47^{c} \pm 4.00$	$400.5^{\rm b} \pm 36.50$	$364^{\rm b} \pm 12.00$	$671^{a} \pm 92.00$	0.01
Na^+ content (mg L ⁻¹)	$965^{c} \pm 15.00$	$1085^{c} \pm 35.00$	$1335^{\rm b} \pm 85.00$	$1895^{a} \pm 45.00$	0.01
K^+ content (mg L ⁻¹)	$6.25^{\rm a} \pm 2.65$	$8.9^{\rm a} \pm 0.00$	$9.4^{\rm a} \pm 0.20$	$16.85^{a} \pm 7.75$	0.40

Values with different superscripts in the same row differ significantly (P < 0.05) and data is expressed as Mean \pm SE (n = 3).

maintained at optimal levels among the different experimental treatments.

3.2. The effect of salinity on gonad weight, latency period, sperm motility, stripping response, total fecundity and fertilization rate

Fish of treatment T1 (5 ppt) and control (0 ppt) groups showed relatively high average gonad weight of 102.5 g \pm 0.5 and 95.5 g \pm 6.5, respectively. Low values of gonad weight, i.e. $67.0 \text{ g} \pm 12$ and 31.0 ± 7.0 were observed in treatment T2 and T3, respectively. The latency period was less (~2.5 h) in control (0 ppt) and treatment T1 (5 ppt) compared to other treatments (10 h in T2 and T3) (Table 2). The sperm motility was also significantly high (P < 0.05) at 0 (94 s) and 5 ppt (96 s). It was reduced significantly in higher salinity range i.e. in treatment T2 and T3 (Fig. 1). High stripping response was recorded in 5 ppt (74.63 ± 0.85%) followed by 0 ppt (67.73 ± 4.81%). The lowest stripping response was observed in fish reared at 15 ppt $(35.08 \pm 1.75\%)$ (Fig. 2). The total fecundity was relatively high in T1 (139171 ± 2387) followed by control groups (98348 ± 12543) . Compared to T1 and C group, treatment T3 showed a reduction of 13-15% of total fecundity (Fig. 3). The ovulatory response and egg quality are represented in Table 2. The percentage of fertilization rate was high (P < 0.05) in C and T1 treatment groups while T3 group showed significantly low values (Fig. 4).

3.3. The effect of salinity on hatching dynamics and deformation of larvae

The incubation period was significantly less (P < 0.05) in control (0 ppt) (30–32 h) and treatment T1 (5 ppt) (26–28 h) than other treatments (Fig. 5). Long hatching duration (68–70 h) was observed in treatment T3 (15 ppt). The percentage of normal and deformed early life stages (eggs and larvae) in response to different treatments are shown in Table 2. The percentage of larvae survival was significantly

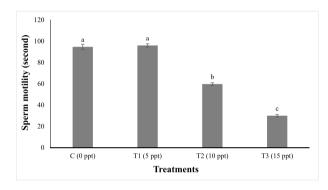


Fig. 1. Effect of different salinities on sperm motility (second) of *C. carpio* Data are expressed as mean \pm S.E., n = 3. Different superscripts signify statistical differences (P < 0.05).

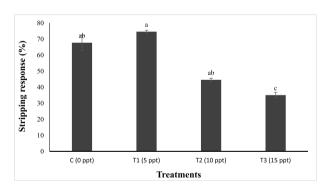


Fig. 2. Effect of different salinities on stripping response (%) of *C. carpio* Data are expressed as mean \pm S.E., n = 3. Different superscripts signify statistical differences (P < 0.05).

Table 2

Ovulatory response and egg quality of Cyprinus carpio under treatment with different salinities in ISGW.

Parameters	Salinities	Salinities				
	0 ppt	5 ppt	10 ppt	15 ppt		
Weight of females (g)	$370.0^{a} \pm 20.0$	$342.0^{ab} \pm 26.0$	$275.0^{\rm b} \pm 15.0$	167.5 ^c ± 17.5	0.00	
Weight of gonad (g)	$95.5^{ab} \pm 6.50$	$102.5^{\rm a} \pm 0.5$	$67.0^{\rm b} \pm 12.0$	$31.0^{\circ} \pm 7.0$	0.00	
Deformed larvae (%)	$30.59^{c} \pm 1.76$	$25.71^{\circ} \pm 2.22$	$50.01^{b} \pm 2.04$	$71.21^{a} \pm 5.34$	0.00	
Normal larvae (%)	$69.40^{a} \pm 1.76$	$74.28^{\rm a} \pm 2.22$	$49.98^{\rm b} \pm 2.04$	$28.78^{\circ} \pm 5.34$	0.00	
Spawning fecundity	$103.25^{\rm b} \pm 7.64$	$159.83^{a} \pm 14.87$	$78.67^{ab} \pm 6.32$	$51.12^{c} \pm 5.36$	0.00	
Latency period (hr)	2.50-3.15	2.0-2.15	0.00 ^a	0.00 ^a		

Values with different superscripts in the same row differ significantly (P < 0.05) and data is expressed as Mean \pm SE. (n = 3). ^a Not naturally spawned.

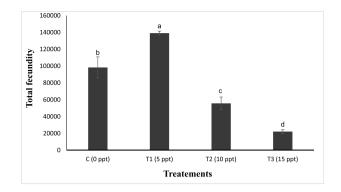


Fig. 3. Effect of different salinities on total fecundity of *C. carpio* Data are expressed as mean \pm S.E., n = 3. Different superscripts signify statistical differences (P < 0.05).

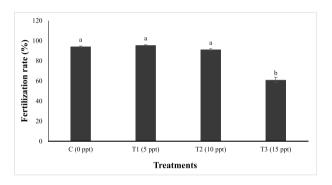


Fig. 4. Effect of different salinities on fertilization rate (%) of *C. carpio* Data are expressed as mean \pm S.E., n = 3. Different superscripts signify statistical differences (P < 0.05).

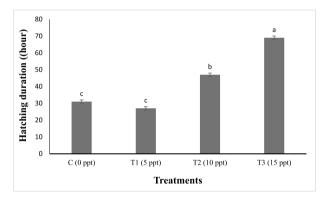


Fig. 5. Effect of different salinities on hatching duration (%) of *C. carpio* Data are expressed as mean \pm S.E., n = 3. Different superscripts signify statistical differences (P < 0.05).

differed (P < 0.05) among treatments. Control (0 ppt) and treatment T1 (5 ppt) showed high percentage of larvae survival than other treatment groups (T2 and T3) (Fig. 6). At salinity of 0 (C) and 5 ppt (T1), egg hatching rate was relatively high and it was gradually decreased as salinity increases (10–15 ppt) (Fig. 7). The percentage of deformed larvae was relatively low in 5 ppt (25.71 \pm 2.22%) compared to 15 ppt (71.21 \pm 5.34%) (Table 2).

3.4. Effect of salinity on embryonic and larval development

The observations showed delayed progression from eggs to juveniles at treatment T2 (10 ppt) and T3 (15 ppt). Normal rate of development was observed in control (0 ppt) and treatment T1 (5 ppt) (Table 3). The important early developmental stages along with time duration to

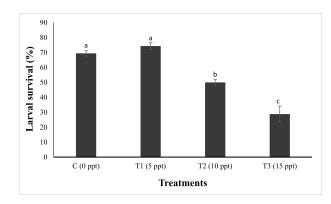


Fig. 6. Effect of different salinities on larval survival (%) of *C. carpio* Data are expressed as mean \pm S.E., n = 3. Different superscripts signify statistical differences (P < 0.05).

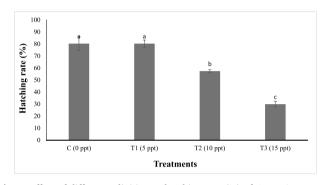


Fig. 7. Effect of different salinities on hatching rate (%) of *C. carpio* Data are expressed as mean \pm S.E., n = 3. Different superscripts signify statistical differences (P < 0.05).

Table 3

Different embryonic stages of the *Cyprinus carpio* at 26–27 $^\circ$ C with different salinities in ISGW.

Embryonic stages	Salinities				
	0 ppt(C)	5 ppt (T1)	10 ppt (T2)	15 ppt (T3)	
Fertilized egg (h: min)	00:15	00:15	00:22	00:25	
Zygote (h: min)	0:26-1:26	0:56-1:06	1:40-2:00	1:50-2:50	
Morula (h: min)	1:26-2:50	1:06-3:45	2.00-3:50	2:50-4:20	
Blastula (h: min)	2:50-3:24	3:45-4:00	3.50-4:50	4:20-5:20	
Gastrula (h: min)	3:24-5:50	4:00-6:00	4:50-7:30	5:20-8:50	
Neurella (h: min)	5:50-7:45	6:00-6:45	7:30-8:45	8:50-10:30	
Appearance of melanophores in the eye (h: min)	7:45–16:25	6:25–13:12	8:45–22:30	10:30-28:03	
Pharyngula (h: min)	16:25-24:25	13:12-21:00	22:30-34:15	28:03-43:03	
Hatching (h: min)	24:25-31:00	21:00-27:00	34:15-47:50	43:03-69:00	
Juvenile stage (days)	24–30	24–28	36–45	42–60	

achieve the respective stage are as follows.

- a. Zygote stage: The newly fertilized eggs developed into zygote and formed blastodisc (region of yolk-free cytoplasm at the peripheral end of the egg). Progression of this stage (2–32 cells) took duration of 1:26 h, 1:06 h, 1:40 h and 1:50 h for control (0), T1 (5),T2 (10) and T3 (15 ppt), respectively.
- a) Morula stage: In this stage, cell number was increased from 32 to 64. The average time taken to reach this stage after fertilization is 1:26 h, 1:06 h, 1:40 h and 1:50 h for control (0), T1 (5),T2 (10) and T3 (15 ppt), respectively.
- b) Blastula stage: This stage was characterized by appearance of cells in

circular shape with epiboly. After fertilization, the duration to progress through this stage was 2:50–3:24 h, 3:45–4:00 h, 3.50–4:50 h and 4:20–5:20 h at 0, 5, 10, and 15 ppt, respectively.

- c) Gastrula stage: This stage showed multi-layered gastrula movement towards the animal ring and blastoderm coverage of most part of yolk. The zygote reached this stage after duration of 3:24–5:50 h, 4:00–6:00 h, 4:50–7:30 h and 5:20–8:50 h after fertilization at 0, 5, 10, and 15 ppt, respectively.
- d) Neurella stage: Embryo was formed/appeared in the stage after duration of 5:50–7:45 h, 6:00–6:45 h, 7:30–8:45 h and 8:50–10:30 h after fertilization of egg cells at 0, 5, 10, and 15 ppt, respectively.
- e) Appearance of melanophores in the eye: Melanophores appeared in the eyes of the embryo at 7:45–16:25 h, 6:25–13:12 h, 8:45–22:30 h and 10:30–28:03 h after fertilization of egg cells at 0, 5, 10, and 15 ppt, respectively.
- f) Pharyngula stage: This stage is characterized by the development of brain, optic vesicles, small tail, and rudimentary heart. The duration to reach this stage was 16:25–24:25 h, 13:12–21:00 h, 22:30–34:15 h and 28:03–43:03 h after fertilization of egg cells at 0, 5, 10, and 15 ppt, respectively.
- g) Hatching stage: At this stage, embryos showed movement with differentiated development of eyes and tail. Hatching occurred at 24:25–31:00 h, 21:00–27:00 h, 34:15–47:50 h and 43:03–69:00 h after fertilization of egg cells at 0, 5, 10, and 15 ppt, respectively.
- h) Juvenile stage: Larvae looked as adult fish where fins rays, scales, anal fins developed. It happened after 24–30 days, 24–28 days, 36–45 days, and 42–60 days after fertilization of egg cells at 0, 5, 10, and 15 ppt, respectively.

4. Discussion

Development of suitable technologies for the utilization of ISGW and Inland saline soils is one of the national priorities of India (Lakra et al., 2014). The ionic composition of ISGW is significantly different from marine or brackish water. Especially, ISGW is deficient in potassium (K⁺) and marine species could not show optimum growth in ISGW without potassium fortification. In recent times, culture practices for different euryhaline fish species have been developed by fortifying the potassium in ISFW (Rahman et al., 2005; Jahan et al., 2018). Likewise, the ISGW and freshwater also differ in the composition of minerals/ions, especially hardness (Ca^{2+} and Mg^{2+}) are quite high in ISGW compared to freshwater. However, potassium (K⁺) ion concentration is quite similar to ISGW (Lakra et al., 2014). Few freshwater stenohaline species such as C. carpio could able to tolerate moderate fluctuations in water parameters including pH, salinity and temperature. It is essential to record the optimal salinity for growth, breeding and larval development of the C. carpio.

Generally, water parameters such as salinity, temperature, pH, and alkalinity can affect the gonadal development and spawning (Sapkale et al., 2011; Hui et al., 2014; Okamoto et al., 2009). At elevated salinity levels as the concentration of Na²⁺, Ca²⁺ and Mg²⁺ increases, most of the available energy would be spent for osmoregulation and it causes reduced development of gonads (Kirschner, 1993). Accordingly, in the present study, gonad development of *C. carpio* was delayed at higher salinities as manifested by less gonad weight at treatment T3 (15 ppt). Previous studies on *Cirrhinus mrigala* also showed the adverse effect of higher salinities on gonadal development (Rajender, 2000).

The present study showed the feasibility of C. carpio breeding up-to salinity of 15 ppt. However, the fertilization rate was reduced drastically from 95 to 61% at 15 ppt (T3) while no significant difference was observed between salinities 5 (T1: 95%) and 10 ppt (T2:91%). It could be due to the adverse effect of increased salinity on ovulatory response and egg quality (Boeuf and Payan, 2001). Hui et al. (2014) reported fertilization rate of 87.77% in *Oreochromis niloticus* (Nile tilapia) reared at a salinity of ~10 ppt.

Sperm motility is an indicator of milt quality and acts as a key factor

for successful fertilization of the eggs (Griffin et al., 1998). Martins et al. (2015) have reported that sperm quality greatly affected by variation in environmental factors such as salinity. The freshwater fish exhibits sperm motility approximately for 30–40 s in hypo-osmotic medium, but in brackish water sperm activity last for 200 s (Griffin et al., 1998). Similarly, in the present study, *C. carpio* showed significantly highest sperm motility at 0–5 ppt (94–96 s) later it was reduced (30 s) as salinity increases.

In the present study, *C. carpio* displayed uniform fecundity values across salinity range of 0–5 ppt. However, the fecundity value was significantly reduced at 15 ppt (50% of the value observed at 0–5 ppt). Malik et al. (2018) reported considerable spawning rate of *C. carpio* at various salinity levels of artificial seawater. Previous studies also demonstrated successful spawning and egg development of *Oreochromis niloticus* (Fridman et al., 2012), *Heterobranchus longifilisi* (Fashina-Bombata and Busari, 2003), *Brachydanio rerio* (Sawant et al., 2001) and *Cirrhinus mrigala* (Rajender, 2000) at lower salinities.

Senoo (2003) reported that delayed hatching period due to salinity causes high mortality of fish embryo/larvae. Accordingly, in the present study, hatching duration was increased as salinity rises and led to higher mortality and larvae deformation (> 70%) at 15 ppt. At low salinities (0–5 ppt), hatching period was 21–27 h while increased hatching period of 43–69 h was evident at 15 ppt. Therefore, it indicates that higher salinity (ISGW) of > 10 ppt causes long hatching time and subsequently more deformation rate of larvae.

The reports on effect of ISGW on culture and breeding of freshwater fish are still scanty and are limited to cichlids (Uchida et al., 2000). Nguang et al. (2012) studied effects of salinity (0-30 ppt) on hatching and larval deformation in freshwater fish Marble Goby (Oxyeleotris marmoratus) and showed highest hatching and lowest deformation of larvae up-to salinity of 10 ppt. Hernandez-Rubio and Lucero (2013) suggested that the salinity could highly affect the embryonic development of freshwater endemic Mexican aterinopsids (Chirostoma humboldtianum and Chirostoma riojai), and reported the survival of both species up-to salinity of 12 ppt. In the present study, C. carpio had reached the juvenile stage within 24-60 days as per the salinity level. Specifically, development from eggs to juvenile was delayed at 10 ppt (36-45 days) and 15 ppt (42-60 days) compared to 0 and 5 ppt (24-30 days). Recently, several studies on C. carpio have reported the duration for reaching the juvenile stage after fertilization is 30--35 days (Mojer, 2015; Park et al., 2017).

5. Conclusion

Fish breeding is affected/influenced by many environmental conditions that could provide a conducive environment or stimulating factors. Each fish could tolerate either narrow or wide range fluctuations in its habitat as per their physiological plasticity. Precise identification of tolerance levels of salinity could provide an opportunity to culture the freshwater fish in brackish water also. The present study recorded a considerable success rate in gonadal development, breeding, fertilization and larvae development of *C. carpio* at a salinity range of 0–10 ppt. *C. carpio* could yield optimum production at salinity of 5 ppt and it could also breed successfully this salinity. It will assist in sustainable utilization of non-arable inland saline water to provide livelihood and nutritional security to the local communities.

Declaration of competing interest

The authors report no conflict of interest.

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