

## Growth and survivability of *Labeo rohita* (Hamilton, 1822) spawns in *Moina micrura* augmented biofloc-based nursery system

Chittaranjan Raul<sup>\*1</sup>, J Praveenraj<sup>1</sup>, R Kiruba Sankar<sup>1</sup>, Udipta Roy<sup>2</sup>

<sup>1</sup>ICAR-Central Island Agricultural Research Institute, SriVijaya Puram, Andaman & Nicobar Islands, India

<sup>2</sup>ICAR-Central Institute of Fisheries Education, Mumbai, India

\*Corresponding author's E-mail:- raulcittaranjan339@gmail.com

### Abstract

Biofloc technology is an intensive, water-efficient aquaculture, but it lacks sufficient zooplankton live feed diversity, required for nursery rearing of larvae. The present study aimed to assess the potential of *Moina micrura* augmentation in the biofloc system to improve the growth and survival of *Labeo rohita* spawn to the fry stage. A 30-days study was conducted using different stocking densities of spawn 5000, 6000 and 7000 spawn/m<sup>3</sup> in both biofloc (T1, T3, T5) and zoofloc (T2, T4, T6) treatments. The floc was prepared using jaggery, and a C:N ratio of 10-15:1 was maintained, which enabled the production of floc volume of 8-10 mL/L. High fry survivability (79 to 91%) was achieved, which decreased significantly ( $p \leq 0.05$ ) with an increase in stocking density in both biofloc and zoofloc treatments. The fry length ( $29.7 \pm 1.47$  mm) and weight ( $212.2 \pm 4.49$  mg) of *Labeo rohita* after rearing were significantly higher in T2 treatment compared to others. The proximate composition of biofloc did not vary significantly, whereas the muscle crude protein of fry increased significantly ( $p \leq 0.05$ ) in zoofloc floc-based treatments than biofloc of the same stocking density. The water quality parameters did not vary significantly except TSS and TDS.

**Key words:** Biofloc, Zoofloc, *Labeo rohita*, *Moina micrura*

### Introduction

Global aquaculture production has exceeded capture fisheries, accounting for 60% of total output (FAO, 2024). In India, the freshwater aquaculture sector contributes 88.2% of national aquaculture production, of which Indian major carps constitute 64.5%. Among them, *Labeo rohita* is the most widely cultured species, contributing 3.7% of global finfish production (FAO, 2020), underscoring its significance. For sustainable aquaculture, quality seed is a critical input, representing 5-10% of the total production cost (Ghosh *et al.*, 2019). Conventional nursery-based seed production systems generally show low fry survival rates, with 30-40% in earthen ponds and 50-60% in cemented tanks, even with protein-rich supplemental feed (Jena and Das, 2011; Das *et al.*, 2016). Predation remains major limitation in these carp nursery systems, leading to reduced survival (Jena *et al.*, 1996). Biofloc technology (BFT) provides a bio-secure and water-efficient alternative for both seed production and grow-out culture (Mugwanya *et al.*, 2021; Sayed, 2021). The principle of BFT relies on the assimilation of total ammoniacal nitrogen (TAN) generated from faecal matter

and uneaten feed by heterotrophic bacteria through the maintenance of a carbon-to-nitrogen ratio of 10-20:1 by addition of external carbon sources (Avnimelech, 2012; Panigrahi *et al.*, 2017). Under intense aeration, these bacteria, along with dissolved organic particles, protozoa and planktons, form biofloc that enhances water quality and serves as a natural feed source, allowing culture at high stocking densities (Ahmad *et al.*, 2017; Raza *et al.*, 2022). Initially developed for tilapia and shrimp, biofloc systems have since been standardised for various finfish and shellfish species (Browdy *et al.*, 2012). Beyond grow-out culture, high-density rearing of carp seeds from spawn to fingerling stages can also be effectively practiced in a biofloc system to achieve improved survival (Swain *et al.*, 2025; Dey *et al.*, 2022; Solanki *et al.*, 2023).

The feeding habit of *Labeo rohita* during the spawn to fry stage is omnivorous, with a strong preference for zooplankton such as cladocerans and rotifers (Majumdar *et al.*, 2018). At this stage, fry actively consume easily digestible, protein-rich natural food, as their dietary protein requirement (35-40%) is higher than that of fingerlings and yearlings (Brahmane *et al.*, 2014). Gut

content analysis of *Labeo rohita* fry shows that 60-75% of their diet consists of zooplankton (cladocerans, copepods and rotifers), while 20-40% is composed of phytoplankton such as diatoms and chlorophyceae (Mohapatra et al., 2003). Only 5-15% of the diet is contributed by protozoa, detritus and dissolved organic matter (DOM) (Mohapatra et al., 2003; Majumdar et al., 2018). In freshwater Biofloc systems, zooplankton diversity is largely represented by protozoan ciliates, flagellates and metazoan rotifers such as *keratella* (Chen et al., 2023). Under high C:N ratios, the availability of TAN and DOM in aerated conditions supports the rapid development of protozoa, forming a small microcosm-based food web (Ahmad et al., 2027). During early stages, bacteria and flagellates dominate numerically, quickly converting dissolved organic carbon and ammonia into particulate biomass (single cell protein). As the Biofloc matures, ciliates become more prominent, channelling bacterial biomass to higher trophic levels. Zooplankton such as *Daphnia*, *Moina* and rotifers supply high-quality live protein (40-70%) and are highly suitable for carp fry. Feeding zooplankton to carp fry results in higher specific growth rate (SGR) and weight gain compared to formulated feed (Crab et al., 2012). In addition to being a protein-rich supplement, zooplankton enhances growth and survival by providing essential fatty acids (EFA), vitamins, amino acids, carotenoids and digestive enzymes (Murugan et al., 2022; Grubisic et al., 2012). Several studies have reported that Biofloc-based carp seed rearing yield lower growth compared to pure zooplankton feeding, whereas Biofloc improves survivability due to its biosecure environment, better water quality, probiotic effects and reduced incidence of pathogenic bacterial infections (Lee et al., 2016; Panigrahi et al., 2019; Azim and Little, 2008). Zooplankton feeding is particularly effective for spawn to fry rearing and integrating zooplankton supplement within a Biofloc system can enhance specific growth rates while maintaining high survivability.

*Moina* sp is a freshwater cladoceran characterised by an open brood pouch, distinguishing it from *Daphnia* sp (Rottmann et al., 2003). It is a nutrient rich live feed containing 45- 50 % protein and 20-27 % fat on a dry weight basis, with adult size ranging from 700-1000 µm (Islam et al., 2017). It can be mass cultured, with

phytoplankton species such as *chlorella* sp, and can thrive on various food sources, including phytoplankton, yeast, bacteria, and detritus (Hena et al., 2025). Additionally, *Moina* can utilise a wide range of food resources, including algae, heterotrophic ciliates, and detritus of plant and animal origin, for its growth and reproduction (Rasdi et al., 2020). It can also grow and reproduce in the absence of phytoplankton by consuming bacteria and protozoan ciliates, and flagellates (Wylie and Currie, 1991). To utilise nutrient-rich biofloc and increase growth of *Labeo rohita* fry the augmentation of *Moina micrura* in biofloc based carp seed nursery under different stocking densities were studied in this experiment.

## Materials and Methods

### Experimental setup and stocking of fish

A 30-day nursey rearing experiment was conducted in 18 numbers of circular FRP tank (1mt diameter, 1.2 m height) with a capacity of 1200 L at Marine Hill wet laboratory of ICAR-CIARI, SriVijayaPuram, India. All tanks were filled with water upto 1 mt and chlorinated with bealching powder at 30 ppm chlorine. The water was then dechlorinated through continuous aeration for one week using aerator (Haelia 120 AMP, 95 w). Spawn of *Labeo rohita* ( $5.5 \pm 0.17$  mm,  $2.52 \pm 0.05$  mg), produced from the carp hatchery of ICAR-CIARI, Sri Vijaya Puram, were acclimized and stocked in six treatments (each in triplicate) at three stocking densities: – 5000 (T1, T2), 6000 (T3, T4), and 7000 (T5, T6), individuals m<sup>-3</sup>. Treatments T1, T3, and T5 consisted of a biofloc-based rearing system, whereas in treatments T2, T4, and T6, *Moina micrura* (freshwater caldoceran zooplankton) was added to the biofloc system on alternate days. The experimental design followed a 3 × 2 factorial arrangement. Spawn were fed commercial powdered feed (crude protein 36%, lipid 6%). Feeding was initiated at a daily ration of 400 g per one lakh spawn, split into two meals and with 10% increase in daily increment until.

### Floc preparation and quantification

Initially, biofloc formation in the treatemnt tanks was initiated by adding an external probiotic (CIBA Floc™, chennai, India) containing *Bacilus* strain ( $1 \times 10^9$  CFU) at

a does of  $1 \text{ gm m}^{-3}$  in water, along with a carbon source jaggery. Once the floc volume reached the desired level (8-10 mL/L, measured using an Imhoff cone), the biofloc was maintained through the regular addition of jaggery at a C:N ratio of 10-15:1.

### Production and Addition of *Moina micrura*

*Moina micrura* pure culture inoculum maintained in the algal laboratory of ICAR-CIARI was used for this experiment. This species was cultured in transparent glass fibre rectangular tanks (50 L capacity) under natural light conditions. The water used for culturing *Moina micrura* was treated with bleaching powder (30% chlorine), and aerated continuously, maintaining a pH of approximately 7.4. The culture was fed with baker's yeast, and the *Moina micrura* were harvested using a  $50 \text{ }\mu\text{m}$  mesh plankton net (with adult body size of approximately  $700\text{-}900 \text{ }\mu\text{m}$ ). *Moina micrura* were added to T2, T4, and T6 treatment tanks at a concentration of @ 10 individual  $\text{mL}^{-1}$  on alternate days (Jena et al., 2008).

### Fish sampling and growth assessment

Sampling of fry was carried out at fortnightly intervals to record length, weight, and total biomass. Randomly selected fry were measured for length using laminated graph paper, and weight was recorded using pre-graduated water beaker. A total of 30 fry were taken from each tank for each sampling, and proper care was taken to minimise handling and sampling stress. Furthermore, different growth attributes were calculated using the following formula;

Length gain (%) =  $\{(\text{Final length} - \text{Initial length}) / \text{Initial length}\} * 100$

Weight gain (%) =  $\{(\text{Final Weight} - \text{Initial Weight}) / \text{Initial Weight}\} * 100$

Survivability (%) =  $(\text{Number of Fry harvested} / \text{Number of Spawn stocked}) * 100$

Specific growth rate (SGR) ( $\% \text{ Day}^{-1}$ ) =  $\{(\text{Ln Final length} - \text{Ln Initial length}) / \text{Number of days rearing}\} * 100$

Specific growth rate (SGR) ( $\% \text{ Day}^{-1}$ ) =  $\{(\text{Ln Final Weight} - \text{Ln Initial Weight}) / \text{Number of days rearing}\} * 100$

Apparent Feed Conversion Ratio = Total feed given / weight gain

### Water quality parameters

Water samples from each treatment tank were collected in the morning hours at 6-day intervals to assess important water quality parameters. Temperature and pH were measured using a digital probe (HANNA), while dissolved oxygen was recorded in situ using HACC probe. Total alkalinity and total hardness were measured using titration methods, whereas total ammonical nitrogen (TAN), nitrate and nitrite were analysed using visible-spectrophotometer method (APHA, 2006). Floc volume was measured by imhoff cone, and total suspended solids (TSS) and total dissolved solids (TDS) measured using filterpaper method (APHA, 2006). To check high fluctuation in total alkalinity, need base lime ( $\text{CaCO}_3$ ) was applied whenever necessary.

### Proximate analysis

The proximate composition of *Moina micrura*, feed and fish mussel was analysed following the methods described by AOAC (2006). At the end of the experiment, floc samples were harvested using a  $100 \text{ }\mu\text{m}$  net and dried in a hot air oven at  $60 \text{ }^\circ\text{C}$  for proximate analysis. *Moina micrura* samples were collected using a  $50 \text{ }\mu\text{m}$  plankton net and freeze-dried at  $-80 \text{ }^\circ\text{C}$  for proximate analysis. Crude protein content was estimated using the Kjeldahl method using KELPLUS-CLASSIC DX (Pelican), while crude lipid content was estimated using the ether extraction method with a SCOS PLUS (Pelican) apparatus. Ash content was determined by incineration in a muffle furnace at  $550 \text{ }^\circ\text{C}$  for 6 hours, and moisture percentage was estimated using a hot air oven at constant  $105 \text{ }^\circ\text{C}$ . The carbohydrate percentage content was calculated using the formula given by Wei et al., (2016).

### Statistical Analysis

The data generated from the experiment were first checked for normality and homogeneity of variances. Subsequently, the data were analysed using two-way analysis of variance (ANOVA) followed by Duncan's multiple comparison test, using the statistical package

for social science (SPSS), version 16.0. The level of significance was set at  $p < 0.05$ . All results are presented as the mean  $\pm$  standard error (SE) of the replicates for each treatment.

## Results and Discussion

### Growth parameters

The growth parameters of *Labeo rohita* fry under different treatments are presented in Table 1. The final length of fry was highest in T2 ( $29.7 \pm 1.47$  mm,  $p \leq 0.05$ ), whereas the lengths in T1, T3 and T6 were statistically similar ( $p \geq 0.05$ ). The lowest final length ( $24.7 \pm 1.1$  mm) was recorded in T5, which had the highest rearing density (7000 spawn  $m^{-3}$ ). The final weight of fry was also significantly higher in T2 ( $212.2 \pm 12.5$  mg,  $p \leq 0.05$ ), while the weights in T1, T3, and T5 did not differ significantly. Overall, significantly higher survival ( $p = 0.046$ ), ranging from 79.1 to 91.3 % was observed across the treatments. T2 recorded the highest survival, which was statistically similar ( $p \geq 0.05$ ) to T4. The lowest survival was observed in T5, although it was statistically similar ( $p \geq 0.05$ ) to T1 and T3. The percentage of length gain did not differ significantly among treatments ( $p \geq 0.05$ ). The highest percentage weight gain was obtained in T2, while T4 and T6, did not differ significantly from it. Treatments, T1, T3, and T5 exhibited the lowest weight gain percentages. Specific growth rate (SGR) in both length and weight decreased with increasing stocking density in both biofloc (T1, T3, T5) and zoofloc (T2, T4, T6) based treatments. T2 showed significantly higher SGR in length compared to other treatments, whereas the lowest SGR values in T3 and T5) were statistically similar with T1 and T6. T2 also showed a significantly higher ( $p \leq 0.05$ ) SGR in weight, while no significant differences ( $p \geq 0.05$ ) were observed among the biofloc treatments. There

was no significant difference in condition factor across all treatments. Apparent feed conversion ratio (AFCR) was significantly lower in T2 ( $4.37 \pm 0.12$ ,  $p \leq 0.05$ ), and overall, the zoofloc based treatments showed significantly lower AFCR values than biofloc based treatments. In the present study, the stocking density of *L. rohita* spawn was maintained at 15-20 tme higher than that of the earthen nursery pond syste. As stockig density increased, both growth and survival decreased in the biofloc, and *Moina micrura* augmented biofloc treatments. However, the latter showed significantly higher growth and survival than the former at stocking densities of 5000 and 6000 Nos/ $m^3$ . Several studies have reported simillar trend of reduced growth and survival with increasing stocking density in biofloc systems due to stress genereated by over crowding (Liu *et al.*, 2018; Adineh *et al.*, 2019; Swain *et al.*, 2025). Comparing earlier studies on catla and rohu spawn rearing in biofloc system, the highest length and weight achieved were 20.13 mm and 90.97 mg at 4000 nos/ $m^3$  stocking density (Swain *et al.*, 2025), 16.6 mm and 28.98 mg at 5000 nos  $m^{-3}$  (Dey *et al.*, 2022), and 18.11mm and 54.62 mg at 5000 nos/ $m^3$  (Solanki *et al.*, 2023). In this study, rohu spawn reached significantly higher length and weight values of 24.2 mm and 212 mg at 5000 nos/ $m^3$  stockng density in the T2 treatments of *Moina micrura* enriched biolfoc system. *Moina micrura* is a cladoceran zooplankton rich in protein and lipid (Table 2). It is a most preferred natural food for carp fry and fingerlings, having more than 50% free amino acid, higher  $\omega_3/\omega_6$  fatty acid ratio and source of digestive enzymes (Samat *et al.*, 2020; Singh *et al.*, 2019). By addition of *Moina micrura* into biofloc system it causes increase in specific growth rate in terms of length and and weight than biofloc treatment in the same stocking denisties (Erias *et al.*, 2023). The aparent feed consersion ratio moina Moina-based traetment is significantly lower than that of biofloc-based traetements (de Souza *et al.*, 2025).



**Table 1: Growth attributes of *Labeo rohita* fry reared at three stocking densities under biofloc and zoofloc system for 30 days period.**

Values are expressed as Mean±SE according to two way ANOVA ( $p \leq 0.05$ ) and Duncan's multiple range test.

Treatments	T1	T2	T3	T4	T5	T6	
Stocking density (m <sup>3</sup> )	5000		6000		7000		
Culture type	Biofloc	Zoofloc	Biofloc	Zoofloc	Biofloc	Zoofloc	P value
Initial Length (mm)	5.60±0.058	5.60±0.058	5.60±0.058	5.60±0.058	5.60±0.058	5.60±0.058	-
Final Length (mm)	25.90±1.24 <sup>bc</sup>	29.7±2.84 <sup>a</sup>	25.2±2.34 <sup>bc</sup>	29.1±1.83 <sup>a</sup>	24.7±2.14 <sup>c</sup>	27.5±1.74 <sup>ab</sup>	0.01
Initial Weight (mg)	2.50±0.59	2.56±0.89	2.53±0.79	2.56±0.88	2.50±0.78	2.50±0.96	-
Final Weight(mg)	103.80±7.49 <sup>d</sup>	212.20±8.49 <sup>a</sup>	94.90±7.47 <sup>de</sup>	168.10±8.4 <sup>b</sup>	87.96±6.46 <sup>e</sup>	134.10±4.45 <sup>c</sup>	0.01
Length Gain (%)	364.6±21.8 <sup>bc</sup>	431.9±11.5 <sup>a</sup>	352.3±14.6 <sup>c</sup>	421.7±26.6 <sup>ab</sup>	342.2±16.5 <sup>cd</sup>	389.8±20.3 <sup>b</sup>	
Weight Gain (%)	4051.73±255.82 <sup>d</sup>	8199.71±457.82 <sup>a</sup>	3674.69±375.82 <sup>d</sup>	6462.09±385.82 <sup>b</sup>	3422.35±366.82 <sup>d</sup>	5268.97 ±367.82 <sup>c</sup>	0.01
AFCR	7.87±0.32 <sup>c</sup>	4.37±0.42 <sup>c</sup>	8.79±0.54 <sup>b</sup>	5.34±0.43 <sup>d</sup>	9.77±0.54 <sup>a</sup>	5.98±0.38 <sup>d</sup>	0.01
Condition Factor	0.61±0.08 <sup>b</sup>	0.81±0.07 <sup>a</sup>	0.59±0.07 <sup>b</sup>	0.69±0.07 <sup>ab</sup>	0.59±0.08 <sup>b</sup>	0.66±0.11 <sup>ab</sup>	0.01
SGR in length (% Day <sup>-1</sup> )	67.67±12.91 <sup>bc</sup>	80.33±13.91 <sup>a</sup>	65.33±14.67 <sup>bc</sup>	78.33±11.91 <sup>a</sup>	63.66±14.44 <sup>c</sup>	73.00±12.55 <sup>ab</sup>	0.01
SGR in Weight (% Day <sup>-1</sup> )	337.67±15.03 <sup>d</sup>	698.78±16.33 <sup>a</sup>	307.89±16.33 <sup>de</sup>	551.78±17.31 <sup>b</sup>	284.89±11.33 <sup>c</sup>	438.67±13.11 <sup>c</sup>	0.01
Survival (%)	82.80±3.39 <sup>b</sup>	91.33±4.59 <sup>a</sup>	81.43±4.69 <sup>b</sup>	86.20±4.69 <sup>ab</sup>	79.10±6.33 <sup>b</sup>	84.17±4.42 <sup>ab</sup>	0.048

**Table 2: Proximate composition of feed and *Moina micrura*. Values are expressed as Mean±SE according to two way ANOVA ( $p \leq 0.05$ ) and Duncan's multiple range test.**

Parameters	Feed	<i>Moina micrura</i>
Crude protein (%)	36±4.6 <sup>a</sup>	48.3±2.4 <sup>b</sup>
Crude Lipid (%)	8.2±1.1 <sup>a</sup>	21.4±1.8 <sup>b</sup>
Carbohydrate (%)	39.8±4.5 <sup>b</sup>	22.1±1.7 <sup>a</sup>
Ash (%)	16±1.3 <sup>b</sup>	8.2±0.89 <sup>a</sup>

### Water quality parameters

The trend of water quality parameters that prevailed for 30 days study period was shown in Table.3 and Figure1. A narrow range of variation in water temperature (29.2 to 29.6 °C), pH (6.98 to 7.07), and dissolved oxygen (5.97 to 6.01 mg L<sup>-1</sup>) were recorded among the treatments. The average total alkalinity varied in the range of 66.2-69.7 mg L<sup>-1</sup>, having no significant difference among treatments. The initial total alkalinity was 75.5 mg L<sup>-1</sup>, and after 12 days, it showed a decreasing trend in all treatments till the end of the experiment. The total hardness was 114 to 122 5 mg L<sup>-1</sup>, and there was no significant difference

( $p \geq 0.05$ ) among treatments. The TDS (320 to 404 mg L<sup>-1</sup>) and TSS (45 to 70) values were significantly increased with an increase in stocking density, where no significant difference ( $p \geq 0.05$ ) was observed between biofloc and zoofloc based treatments. The floc volume was significantly highest ( $p \leq 0.05$ ) in T5 than other and increased with an increase in stocking density in all treatments. There was no significant difference between biofloc and zoofloc based treatments in terms of floc volume. TAN (0.31 to 0.46 mg L<sup>-1</sup>), Nitrite (0.93 to 1.14 mg L<sup>-1</sup>) and Nitrate (2.11 to 2.45 mg L<sup>-1</sup>) showed a similar trend and no significant difference ( $p \leq 0.05$ ) in all

treatments. The water quality in a biofloc rearing system is directly influenced by the stocking density, as both feed and carbon inputs increase with rising biomass (Panigrahi *et al.*, 2019). Temperature, pH and dissolved oxygen remained stable across all the treatments with varying stocking densities, without any significant fluctuations, due to the continuous aeration system (Das *et al.*, 2016). Total suspended solids (TSS) and total dissolved solids (TDS) increased with higher stocking densities as floc volume accumulated (Adineh *et al.*, 2022). A decrease in total alkalinity is a common phenomenon in

biofloc systems, and the same trend was observed in all treatments in this study (Azim and Little, 2008). Lower TAN concentration in all treatments with faster reduction after THE 2<sup>nd</sup> week indicated rapid assimilation by heterotrophic bacteria in the biofloc system. Similarly, a gradual increase in nitrite and nitrate levels after the 2<sup>nd</sup> week has been reported in many biofloc studies involving common carp, indian major carp, and shrimp (Adineh *et al.*, 2022; Diatin, 2019). All the water quality parameters remained within suitable ranges without any significant differences between the biofloc and zoofloc treatments.

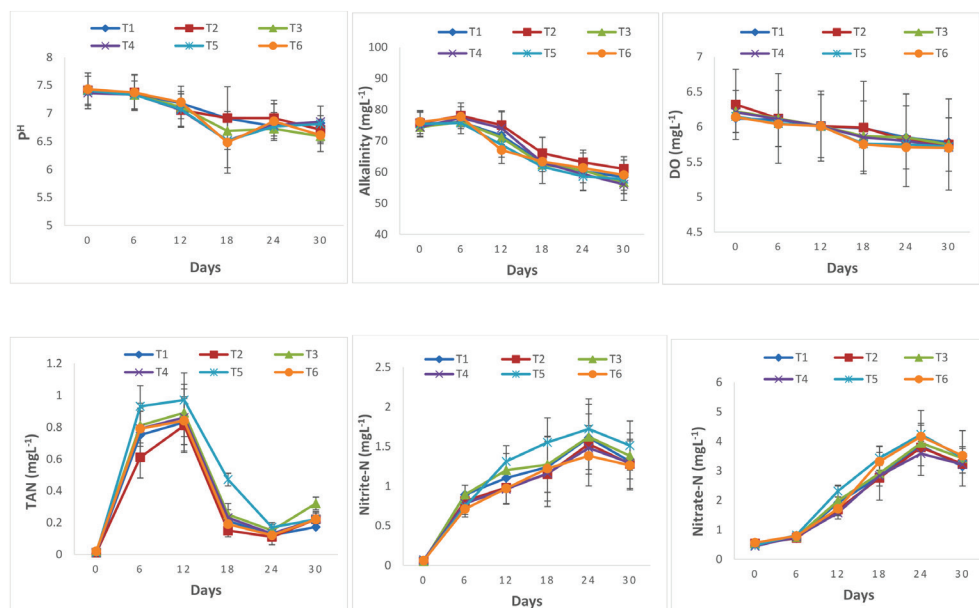


Figure 1: Trend of water quality parameters in different treatments of *Labeo rohita* fry rearing system.

Table 3: Water quality parameters of different treatment of *labeo rohita* rearing water.  
DO- Dissolved oxygen, TDS-Total Dissolved Solids, TSS-Total Suspended Solids.

Water Quality Parameters	T1	T2	T3	T4	T5	T6	Two way ANOVA
Temperature (°C)	29.2±0.15 <sup>a</sup>	29.2±0.1 <sup>a</sup>	29.3±0.11 <sup>a</sup>	29.2±0.15 <sup>a</sup>	29.3±0.12 <sup>a</sup>	29.6±0.14 <sup>a</sup>	NS
pH	7.07±0.11 <sup>a</sup>	7.06±0.11 <sup>a</sup>	6.98±0.14 <sup>a</sup>	6.98±0.13 <sup>a</sup>	6.98±0.14 <sup>a</sup>	6.99±0.16 <sup>a</sup>	NS
DO (mg L <sup>-1</sup> )	5.97±0.55 <sup>a</sup>	6.01±0.84 <sup>a</sup>	5.97±0.72 <sup>a</sup>	5.924±0.78 <sup>a</sup>	5.90±0.75 <sup>a</sup>	5.89±0.79 <sup>a</sup>	NS
TDS(mg L <sup>-1</sup> )	320.5±15.4 <sup>a</sup>	315.5±12.36 <sup>a</sup>	370.8±14.33 <sup>bc</sup>	361.2±18.35 <sup>b</sup>	404.2±17.54 <sup>d</sup>	389.3±15.22 <sup>cd</sup>	S
TSS (mg L <sup>-1</sup> )	45.3±2.08 <sup>a</sup>	42.5±2.26 <sup>a</sup>	56.2±3.35 <sup>b</sup>	55.7±3.9 <sup>b</sup>	68.6±4.31 <sup>c</sup>	70.7±5.25 <sup>c</sup>	S
Hardness (mg L <sup>-1</sup> )	114.6±3.2 <sup>a</sup>	115.2±3.8 <sup>a</sup>	118.5±4.5 <sup>a</sup>	120.8±5.1 <sup>a</sup>	122.1±6.2 <sup>a</sup>	120.5±6.8 <sup>a</sup>	NS
Alkalinity(mg L <sup>-1</sup> )	67.4±3.19 <sup>a</sup>	69.7±2.97 <sup>a</sup>	66.8±3.27 <sup>a</sup>	67.3±3.67 <sup>a</sup>	66.26±3.31 <sup>a</sup>	67.47±3.09 <sup>a</sup>	NS
Floc volume (mL/ L)	6.3±1.1 <sup>a</sup>	6.1±1.3 <sup>a</sup>	6.7±1.5 <sup>ab</sup>	6.8±1.8 <sup>abc</sup>	7.5±1.7 <sup>c</sup>	7.3±2.1 <sup>bc</sup>	S
Ammonia-N (mg L <sup>-1</sup> )	0.34±0.21 <sup>a</sup>	0.31±0.12 <sup>a</sup>	0.4±0.14 <sup>a</sup>	0.37±0.14 <sup>a</sup>	0.46±0.16 <sup>a</sup>	0.35±0.15 <sup>a</sup>	NS

## Proximate Composition of biofloc and fry carcase

The results of the proximate composition of biofloc and fish muscle are presented in Table 4 and Table 5, respectively. The initial proximate composition of the feed and *Moina micrura* is provided in Table 4. The crude protein (36.1 to 39.6%), crude lipid (8.2 to 10.5%) and carbohydrate (36.6 to 39.4%) contents of the biofloc did not vary significantly among the treatments. The crude protein of fish muscle was significantly highest in T2 (53.8%) and lowest in T5 (42.3%), while no significant differences were observed among T1, T3, T4 and T6. The crude lipid, carbohydrate and ash of fish muscle also did not differ significantly across treatments. The proximate composition of biofloc largely depends on the heterotrophic bacteria community and the formulated

feed used in the culture system. Since all treatment tanks received the same feed and were maintained at the same C:N ratio, the proximate composition of biofloc did not vary significantly with varying stocking densities, similar to observations reported in previous studies of Solanki et al., (2023). *Labeo rohita* spawn reared on *Moina micrura* exhibited significantly higher muscle crude protein than those reared solely in biofloc systems. This is because *Moina* contains much higher protein, a superior amino acid profile and higher digestibility compared to microbial biofloc. Feeding of *Moina micrura* also enhances digestive enzyme activity, which improves amino acid assimilation and muscle protein deposition (Woynarovich and Horvath, 1980). In contrast, biofloc typically contains a lower protein and higher ash, reducing its suitability as a complete protein source for carp fry (Azim and Little, 2008).

**Table 4: Proximate composition of biofloc on dry weight basis. Values are expressed as Mean±SE according to two way ANOVA ( $p \leq 0.05$ ) and Duncan's multiple range test.**

	(T1)	(T2)	(T3)	(T4)	(T5)	(T6)	p-value
Crude protein (%)	36.1±2.9 <sup>a</sup>	38.2±2.7 <sup>a</sup>	36.2±1.7 <sup>a</sup>	38.8±3.1 <sup>a</sup>	37.5±3.5 <sup>a</sup>	39.6±4.1 <sup>a</sup>	0.58
Crude Lipid (%)	8.2±0.56 <sup>a</sup>	10.5±0.63 <sup>a</sup>	8.8±0.64 <sup>a</sup>	9.9±0.71 <sup>a</sup>	8.6±0.85 <sup>a</sup>	10.1±1.2 <sup>a</sup>	0.82
Carbohydrate (%)	39.3±4.2 <sup>a</sup>	36.6±3.8 <sup>a</sup>	39.4±3.3 <sup>a</sup>	37.1±3.8 <sup>a</sup>	37.1±4.1 <sup>a</sup>	36.7±3.7 <sup>a</sup>	0.29
Ash (%)	16.3±1.1 <sup>a</sup>	14.7±1.5 <sup>a</sup>	15.8±1.45 <sup>a</sup>	16.3±1.6 <sup>a</sup>	16.1±2.1 <sup>a</sup>	15.7±1.9 <sup>a</sup>	0.42

**Table 5: Proximate composition of fry carcase on dry weight basis. Values are expressed as Mean±SE according to two way ANOVA ( $p \leq 0.05$ ) and Duncan's multiple range test.**

	(T1)	(T2)	(T3)	(T4)	(T5)	(T6)	p-value
Crude protein (%)	47.8±2.8 <sup>bc</sup>	53.8±1.2 <sup>d</sup>	43.6±1.9 <sup>ab</sup>	51.5±1.1 <sup>cd</sup>	42.3±1.32 <sup>a</sup>	49.3±1.62 <sup>cd</sup>	0.19
Crude Lipid (%)	5.8±0.26 <sup>ab</sup>	6.2±0.45 <sup>ab</sup>	5.2±0.44 <sup>a</sup>	7.2±0.51 <sup>b</sup>	5.4±0.61 <sup>a</sup>	6.6±0.56 <sup>ab</sup>	0.62
Carbohydrate (%)	23.1±2.2 <sup>abc</sup>	19.6±2.49 <sup>a</sup>	26.1±2.93 <sup>bc</sup>	19.7±2.26 <sup>a</sup>	27.7±3.3 <sup>c</sup>	20.4±2.3 <sup>ab</sup>	0.29
Ash (%)	23.3±1.41 <sup>a</sup>	20.4±1.62 <sup>a</sup>	25.1±2.2 <sup>a</sup>	21.6±2.4 <sup>a</sup>	24.4±1.9 <sup>a</sup>	23.7±2.6 <sup>a</sup>	0.58

## Conclusion

The study demonstrated that *Moina micrura* augmentation in biofloc significantly improved the growth performance and survival of *Labeo rohita* from spawn to fry stage compared to conventional biofloc. The zoofloc treatment (T2) resulted in the highest fry length, weight, SGR, and survival, particularly at a stocking density of 5000 spawn/m<sup>3</sup>. Increasing stocking

density negatively affected growth and survival in both systems, but the impact was less pronounced in *Moina*-augmented treatments. Zoofloc also showed superior feed utilisation, reflected by lower AFCR values. Proximate composition of biofloc remained stable, while fry reared in zoofloc exhibited significantly higher muscle crude protein content. The enhanced performance in zoofloc is attributed to the rich nutritional profile and digestibility of *Moina micrura*. Overall, *Moina*-fortified biofloc is a

promising strategy to improve nursery rearing of carp fry at high stocking densities. The findings support the integration of zooplankton augmentation to enhance biofloc efficiency for hatchery-scale carp seed production.

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