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Research Article

Assessment of the Growth and Survival of the Major Carp, Mrigal (*Cirrhinus cirrhosus*) Larvae Raised on Microalgae and Enriched Zooplankton

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Aquaculture is an emerging sector in meeting global food demands, with significant potential to accelerate fish production. However, a major bottleneck in this industry remains the challenge of fulfilling the complex nutritional needs of fish larvae, often limited by the high cost and scarce availability of appropriate feed sources. In this study, we explored the potential of live microalga *Desmodesmus* sp. and nutritionally enriched copepods, administered at different concentrations, as alternative dietary supplements to enhance the growth performance and survival rate of the mrigal, *Cirrhinus cirrhosus* larvae. The fish larvae were reared for a period of 30 days under six feeding regimes (treatments): T_1 ($10–13 \times 10^4$ cells mL^{-1} live *Desmodesmus* sp.), T_2 ($7.5–9.5 \times 10^4$ cells mL^{-1} live *Desmodesmus* sp. + 5–10 individuals mL^{-1} enriched live copepods), T_3 ($5–6.5 \times 10^4$ cells mL^{-1} live *Desmodesmus* sp. + 10–20 individuals mL^{-1} enriched live copepods), T_4 ($2.5–3.25 \times 10^4$ cells mL^{-1} live *Desmodesmus* sp. + 15–30 individuals mL^{-1} enriched live copepods), T_5 (20–40 individuals mL^{-1} enriched live copepods), and T_6 commercial feed (0.5–1 gL^{-1}) as a control diet under laboratory environments. The T_3 diet exhibited the highest protein (17.87%) and fat (2.89%) content in the fish larvae, significantly higher than the control diet T_6 (16.63% protein, 2.28% fat). The T_3 diet significantly improved the larval growth in terms of gain in length (17.34 mm), gain in weight (118.81 mg), percent gain in length (299.69%), percent gain in weight (10,069.49%), and specific growth rate (15.83%). The highest survival rate was observed in T_3 (92.55%), followed by T_4 (90.15%), T_5 (85.48%), T_6 (84.21%), T_2 (82.44%), and T_1 (69.43%). This study highlights the potential of the combined *Desmodesmus* sp. and enriched copepods as diets for the rearing of mrigal larvae with an aim of sustainable aquaculture development.

Keywords: aquaculture; *Desmodesmus* sp.; enriched copepods; fish larval feed; larval growth; sustainable fish farming

1. Introduction

Aquaculture is the fastest-developing sector in recent times for offering global food and economic amenities in response to the rising population need [1]. Fish farmers are encountering several difficulties in the expansion of aquaculture, primarily associated with the dietary and nutritional requirements of the fish larvae [2, 3]. In general, commercial fishmeal is the vital source of protein and other balanced nutrients typically fed to aquaculture species for their growth and healthy advancement. Nevertheless, the usage of fishmeal is severely limited in aquafeeds due to its excessive cost and reduced availability [4]. In addition to the high cost and limited availability of fishmeal, formulated feeds pose

challenges in fish larval rearing due to the underdeveloped digestive systems of the larvae, which often struggle to process these diets effectively, leading to reduced growth and survival rates [2]. Hence, the identification of alternative protein sources for use in aquaculture presents a crucial challenge [5]. Consequently, the live feeds, including microalgae and zooplankton, may be regarded as dependable protein-rich feed sources to achieve remarkable success in the larval rearing of different species of fish and shellfish.

Microalgae, the powerhouse of nutrients, are single-celled microscopic organisms that efficiently liberate oxygen and produce biomass through photosynthesis, mostly utilized as a source of food, feed, biofuel, and other pharmaceutical and nutraceutical compounds [6, 7]. These verdant living entities are also recognized as the natural food of many aquatic species [8, 9]. Microalgae serve as live feed, playing a pivotal role in nurturing zooplankton and supporting the growth of fish and shellfish during their early life stages [10]. Furthermore, they provide the most undertaking and sustainable repositories of essential amino acid profiles, e.g., leucine, isoleucine, lysine, arginine, valine, methionine, and phenylalanine [11, 12], and polyunsaturated fatty acids, especially eicosapentaenoic acid (EPA), arachidonic acid (AA), and docosahexaenoic acid (DHA). These features, along with their content of bioactive compounds [13, 14], pigments, and vitamins, make them a suitable alternative to fishmeal [15–17]. They impart an alluring effect to the flesh of salmonids, shrimp, lobster, and crayfish, as well as enhancing their development and overall health [18, 19]. Moreover, microalgae play a pivotal role in larval nutrition during a short timeframe, serving as a diet component for live prey provided to small fish larvae [20]. In addition, plausible functions are also noticed in water quality management through wastewater remediation in aquaculture systems with microalgae in larval rearing [21, 22]. Microalgae, the Green Gold, have a flourishing effect on the larval stages of cultured fish, crustaceans, and abalone [23, 24], as well as providing not only high survivability but also high weight gain (WG), high specific growth rates (SGR), an increase in length, a specific growth response, tolerance to stress, efficient FCR, and other biorefinery activities [25, 26]. Significant advancements in zooplankton, fish, and shellfish production can be attained through the utilization of various species of microalgae as feed [27].

Desmodesmus sp., *a* green microalga, is commonly found in both freshwater and marine water, drawing the attention of aquaculture researchers for its easy cultivation and biomass production [28]. It has been shown to be a valuable source of essential fatty acids, particularly omega-3 fatty acids, such as alpha-linolenic acid, and can provide up to 50% protein depending on different culture media [29–31]. According to certain research, replacing over 20% of a typical diet with *Desmodesmus* sp. can be a dependable protein and lipid source for larval rearing [32, 33]. Furthermore, it offers a comprehensive set of essential amino acid profiles, supporting the growth and digestibility of various fish species, including prawns, flounders, grunters, and salmonids. Additionally, it functions as a biorefinery mechanism due to its capacity to withdraw nutrients, such as nitrogen and phosphorus, from wastewater during biomass production [34]. Consequently, incorporating live feed, such as *Desmodesmus* sp. microalga, and enriched zooplankton in fish diets offers an innovative solution for aquaculture. This approach represents a highly effective and compelling advancement in the larval rearing of aquaculture species, ultimately enhancing profitability and safety.

Zooplankton, serving as living capsules, perform a crucial ecological role within the aquatic food web by facilitating the transfer of chemicals and energy to higher trophic levels [35]. They hold paramount significance in meeting nutritional demands as live feed for the preliminary stages of several commercial fish species. Thereby, they are a prospective candidate enriched with essential amino acids, polyunsaturated fatty acids, micro- and macronutrients, vitamins, antibodies, and several gastric enzymes that support larval growth, optimum survivability, and good palatability [36]. Microalgae are widely recognized as the most potent nutritional source for zooplankton, commonly employed in aquaculture hatcheries [37]. Therefore, before being ingested by fish larvae, they might boost their nutritional value by ingesting the microalgae in the green water [38, 39]. The most promising zooplankton is copepods, which are employed as the initial feed in many hatcheries. Copepods are thought to provide more nutrients than other traditional live feeds and are fortified with higher concentrations of DHA, EPA, free amino acids, and other key nutrients [40–42]. Highly nutritious copepods can be an excellent choice as live feed organisms to boost the growth and survival of fish and shellfish larvae [43, 44]. Feeding copepods exclusively with nutritious microalgae, such as *Desmodesmus* sp., enhances their suitability as a food source for fish larvae due to their high protein and unsaturated fatty acid content. Thus, nutrient enhanced copepods indicate a variety of opportunities for promoting the development and longevity of freshwater Indian major carp, *Cirrhinus cirrhosus* larvae.

Carps constitute the largest segment of global aquaculture, accounting for 20%–25% of total production. Mrigal is a significant species within this group, contributing approximately 1.91% to global carp production [45]. As a major component of the carp polyculture system, *C. cirrhosus* is predominantly cultivated in South Asian countries, mostly in Bangladesh and India. Though the mrigal is a bottom feeder in advanced juvenile and adult stages but in the early stages of life history, it is plankton feeder in the nature [46]. The limited availability of high-quality fingerlings is a constraint for its enhanced production [47]. In the

context of rearing fish larvae throughout their early stages of development, the size, digestibility, and nutritional value of the meal are the basic considerations. Consequently, adequate nutrition is a crucial factor for promoting the digestion, growth, and survival of fish and shellfish larvae in hatcheries. While there is limited information on rearing larvae with nutrient-rich microalgae, it is necessary to evaluate the nutritional potential of *Desmodesmus* sp. Therefore, this research was aimed to investigate the optimal combination of diets incorporating live *Desmodesmus* sp. along with enriched copepods to ameliorate the growth performance and survival rates of *C. cirrhosus* larvae for sustainable aquaculture practices.

2. Materials and Methods

2.1. Culture of Desmodesmus sp. for Use as Feed in Rearing of C. cirrhosus Larvae. Pure algal strains of *Desmodesmus* sp. were obtained from the Laboratory of Plankton Research, Department of Fisheries Management, Bangladesh Agricultural University, Bangladesh. *Desmodesmus* sp. was cultured in Bold Basal Medium (BBM) at the ambient temperature of $26 \pm 2^{\circ}C$ with constant stirring under controlled lighting conditions of 60 μmol m^{-2} s^{-1} at a photoperiod of 12:12 h, L:D. The microalgal strains were inoculated in sterilized BBM in a 1-L conical flask. After 5–7 days, the cultured microalgae were transferred to BBM in 10-L carboys for mass culture. *Desmodesmus* sp. was harvested during its exponential growth phase and centrifuged to get the required cells for feeding the larvae of *C. cirrhosus.* Cell density of *Desmodesmus* sp. was determined using a Sedgewick–Rafter counting chamber under a compound microscope (B-510BT OPTICA, Italy), as illustrated in Figure 1.

2.2. Enrichment of Copepods Feeding on Desmodesmus sp. Zooplankton were collected using a 50 μm mesh-sized net from the ponds at Bangladesh Agricultural University campus, Bangladesh. The collected zooplankton were thoroughly rinsed with filtered fresh water to remove any contaminants before being transferred to a petri dish. The petri dish was then placed under a stereomicroscope (Stemi 305, Carl Zeiss, Suzhou, China), where individual copepods were carefully identified and isolated using droppers (Figure 2(a)). The isolated specimens were subsequently stocked in three 20-L buckets at an initial density of 2 individuals mL^{-1}. For enrichment, 5×10^5 cells mL^{-1} of *Desmodesmus* sp. were centrifuged and subsequently utilized as nourishing feed three times daily. Following that, the clonal culture was subsequently established and sustained using *Desmodesmus* sp. Throughout the cultivation period, copepods were harvested at 8-day intervals and transferred to fresh culture containers to maintain optimal growth conditions (Figure 2(b)).

2.3. Rearing of the Indian Major Carp, C. cirrhosus Larvae. The experimental setup used to rear the major carp *C. cirrhosus* larvae is shown in Figure 3. The carp larvae were collected from the hatchery of the Fish Seed Multiplication Farm, Maskanda, Mymensingh, operated by the Department of Fisheries, Government of the People's Republic of Bangladesh. The 3-day-old larvae were carefully brought to the laboratory in sealed plastic bags infused with oxygen. In the laboratory, the larvae were acclimatized for 18 h before the experimental procedures commenced. The study was conducted in eighteen 15-L plastic buckets, each containing 10 L of water. The larvae were stocked at a density of 20 individuals per liter (mean length: 5.76 ± 0.004 mm; weight: 1.18 ± 0.02 mg). Six distinct feeding treatments were evaluated, including a control group that received a commercial powdered feed (Mega Feed, Spectra Hexa Feeds Ltd). Each treatment consisted of three replicates, as detailed in Table 1. We followed rigorous care and monitoring to maintain uniform environmental conditions in our effort to raise healthy *C. cirrhosus* larvae. The procedure started with tap water that was kept overnight in a 50-L container to aerate it before being applied. This technique made sure that the dissolved oxygen (DO) level was optimized, and the temperature was carefully regulated to match the ambient room conditions. Additionally, a stone fragment was placed at the bottom of each bucket to offer shelter, simulating a natural environment. Air stones were used for continuous, gentle aeration throughout the experimental period to maintain uniform oxygenation and feeding conditions. The larvae were fed three times a day at 6:00 a.m., 1:00 p.m., and 8:00 p.m. Ahead of each feeding session, cultured copepods were collected with great care, filtered through a 50 μm screen, and promptly rinsed with freshwater. Requisite cell densities were accurately measured with a Sedgewick–Rafter cell before introduction to the fish larvae. Each morning, before feeding, about 30% of the water was exchanged, and any remaining feed or fecal matter was removed from each bucket. This holistic approach to larval care played a pivotal role in fostering healthy growth and development.

2.4. Investigation of Growth Performance and Survival Rate of C. cirrhosus Larvae. Larval weight and length in different replicates of each treatment were measured at the end of the experiment. To estimate overall length (mm), nine larvae were randomly selected from each replicate, and all of the survived larvae were sampled to determine the weight (mg). The growth performance in terms of gain in length (GL), gain in weight (GW), percent weight gain (WG %), percent length gain (LG %), SGR of the body weight (% day^{-1}), and the survival rate was calculated using the following equations [48–51]:

FIGURE 1: Culture of the green microalga *Desmodesmus* sp.: (a) pure cells of *Desmodesmus* sp. (scale bar: 50 µm; 40 × magnification; B-510BT OPTIKA, Italy), (b) pure stocks of *Desmodesmus* sp., (c) initiation of the primary culture of *Desmodesmus* sp., (d) mass culture of *Desmodesmus* sp., (e) centrifugation of *Desmodesmus* sp., and (f) concentrated biomass of *Desmodesmus* sp.

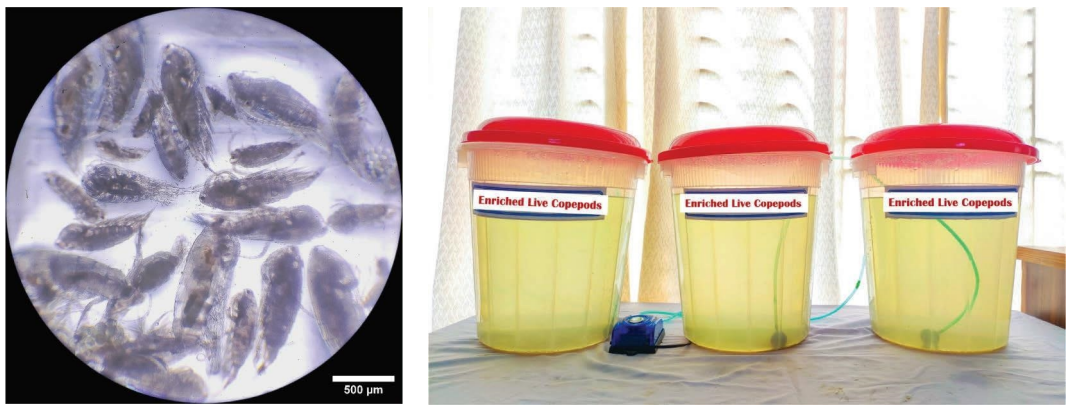

FIGURE 2: (a) Collected copepods and their culture: *Diaptomus* sp., and *Cyclops* sp. (scale bar: 500 µm; 10 × magnification; B-510BT OPTIKA, Italy), (b) mixed culture of copepods feeding on *Desmodesmus* sp.

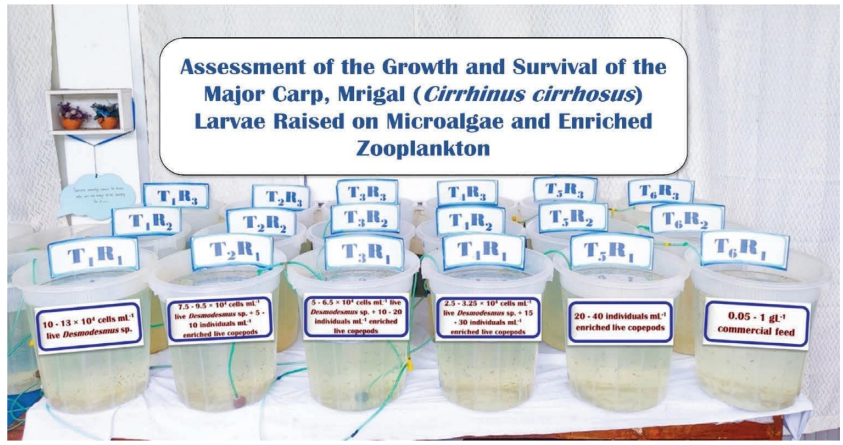

FIGURE 3: Experimental framework for rearing *Cirrhinus cirrhosus* larvae under six different dietary treatments.

$$
\begin{aligned}
\text{gain in weight (GW)} &= (\text{final weight} - \text{initial weight}),\\
\text{gain in length (GL)} &= (\text{final length} - \text{initial length}),\\
\text{percent weight gain (WG \%)} &= \left\{\frac{(\text{final weight} - \text{initial weight})}{(\text{initial weight})}\right\} \times 100,\\
\text{percent length gain (LG \%)} &= \left\{\frac{(\text{final length} - \text{initial length})}{(\text{initial length})}\right\} \times 100,\\
\text{specific growth rate (SGR)}\left(\%\,\text{day}^{-1}\right) &= \left\{\frac{(\text{Ln final weight} - \text{Ln initial weight})}{(\text{number of days})}\right\} \times 100,\\
\text{larval survival rate (S) (\%)} &= \left\{\frac{(\text{number of remaining healthy larvae})}{(\text{total number of stocked larvae})}\right\} \times 100.
\end{aligned}
\tag{1}
$$

2.5. Maintenance of Water Quality Parameters. To ensure a congenial environment for the development of the larvae, some water quality parameters, such as pH, temperature, DO, and ammonia level, were observed on every day throughout the raising phase. DO was measured using an oxygen meter (DO-5509, LUTRON, Taiwan), and pH and temperature were measured using a combined pH and temperature meter (HI98108, HANNA, Romania). The ammonia level was determined using an ammonia test kit (HANNA instrument test kit).

2.6. Biochemical Analysis. The proximate composition analysis was carried out following the standard methods of Horwitz [52]. To determine moisture content, samples were carefully collected and placed in dried porcelain basins and then heated to a steady 105°C in an oven. Subsequently, ash content was determined by burning the dried samples in a furnace at temperatures between 550°C and 600°C for 6 h until they turned white. For assessing crude protein content, dried samples were analyzed using the micro-Kjeldahl nitrogen method. The procedure was conducted with specialized analytical instruments manufactured by Labor-Technik GmbH, Dusseldorf, Germany. Before titration with 0.2 N HCl, the distillate containing ammonia was trapped in a 4% boric acid solution to calculate crude protein content, using conversion factors of 5.85 and 6.25 for plant- and animal-based samples, respectively. Total fat content was determined using the Soxhlet extraction technique, followed by defatting with petroleum ether to assess crude fiber content. Total carbohydrate was calculated by subtracting the cumulative content of protein, fat, moisture, ash, and fiber from 100. Results were expressed as percentages, representing the proximate content of the samples. Finally, the moisture, protein, fat, and ash content percentages were calculated using the following formulas:

(a) % moisture = [(Weight loss/ Original weight of the sample taken) × 100]

(b) Percent (%)Nitrogen = [{Milliequivalent of , N_2,×, Strength of HCl} × Titrant used (mL)/ Weight of sample (g)] × 100

Where, milliequivalent of $N_2 = 0.014$; Strength of HCl = 0.2 N.

The % protein in the sample was calculated by multiplying the % N_2 with an empirical factor of 5.85

Table 1: Dietary variation of *C. cirrhosus* larvae throughout the study period.

Treatments	Feeding schedule		
	Day (1–10)	Day (11–20)	Day (21–30)
T_1	10×10^4 cells mL^{-1}	11.5×10^4 cells mL^{-1}	13×10^4 cells mL^{-1}
T_2	7.5×10^4 cells mL^{-1} + 5 individuals mL^{-1}	8.62×10^4 cells mL^{-1} + 8 individuals mL^{-1}	9.75×10^4 cells mL^{-1} + 10 individuals mL^{-1}
T_3	5×10^4 cells mL^{-1} + 10 individuals mL^{-1}	5.75×10^4 cells mL^{-1} + 15 individuals mL^{-1}	6.5×10^4 cells mL^{-1} + 20 individuals mL^{-1}
T_4	2.5×10^4 cells mL^{-1} + 15 individuals mL^{-1}	2.87×10^4 cells mL^{-1} + 23 individuals mL^{-1}	3.25×10^4 cells mL^{-1} + 30 individuals mL^{-1}
T_5	20 individuals mL^{-1}	30 individuals mL^{-1}	40 individuals mL^{-1}
T_6	0.5 gL^{-1}	0.7 gL^{-1}	1 gL^{-1}

Note: The experimental diets consisted of the microalga *Desmodesmus* sp., enriched copepods, and a commercial feed.

for the phytoplankton and 6.25 for the zooplankton and fish.

$$\%\ \text{protein} = \%\ \text{total}\ N_2\ \text{x}\ 5.85\ \text{or}\ 6.25\,. \quad (2)$$

(c) Percent (%) crude lipid content = $[(W_3 - W_2)/W_1] \times 100$

Where, W_1 = Weight of sample, W_2 = Weight of empty flask, W_3 = Weight of flask containing extracted crude lipid.

(d) % ash = [{(wt. of crucible and ash – wt. of crucible)/(wt. of crucible and sample – wt. of crucible)} × 100]

2.7. Data Analysis. Results from the triplicate samples are presented as the mean alongside the corresponding standard deviation. One-way ANOVA (SPSS Version 25.0) was used to analyze the data of the six treatments' GL, GW, LG%, WG %, SGR, survival rates, and proximate composition (protein, lipid, ash, and carbohydrate) of the larvae. Before commencing the variance analysis, the data were evaluated for normal distribution and homogeneity of variance using a significance level of $p > 0.05$; in cases where these criteria were not met, the Johnson Transformation test was employed for further analysis. The significant variations among the means were determined by Duncan's multiple range test (DMRT) for paired comparison [53]. Statistical significance for all analyses was considered at $p < 0.05$.

3. Results

3.1. Proximate Composition of Dietary Items Used in Larval Rearing. Each experimental diet exhibited distinct nutrient compositions that reflected the unique nutritional contributions of the different feed types (Table 2). Protein content was notably highest in enriched zooplankton (70.15 ± 0.52%), highlighting its superior protein profile compared to *Desmodesmus* sp. (48.50 ± 0.39%) and commercial feed (40.27 ± 0.36%). Lipid content differed significantly, with *Desmodesmus* sp. exhibiting the highest amount (23.56 ± 0.58%), while both enriched zooplankton (16.21 ± 0.25%) and commercial feed (8.30 ± 0.42%) contained substantially lower amounts. Carbohydrate levels peaked in commercially formulated feed (38.52 ± 0.41%), contrasting sharply with the lower concentrations observed in *Desmodesmus* sp. (14.54 ± 0.49%) and enriched zooplankton (6.12 ± 0.27%). Ash content was lowest in enriched zooplankton (4.13 ± 0.38%), followed by *Desmodesmus* sp. (9.64 ± 0.10%) and commercial feed (9.29 ± 0.57%). Fiber content remained relatively uniform across all diets, regardless of the dietary source.

3.2. Biochemical Composition of the Indian Major Carp Mrigal, C. cirrhosus Larvae With Different Treatment Regimens. Protein content varied significantly ($p < 0.05$) across treatments with the peak value observed in T_3 (17.87 ± 0.15%), subsequently T_4 (17.07 ± 0.29%), and T_5 (16.51 ± 0.20%) (Table 3). The minimal protein levels were recorded in T_1 (15.20 ± 0.17%) and T_2 (15.07 ± 0.26%), indicating a limited contribution of *Desmodesmus* sp. alone to protein enhancement in the fish larvae. Fat content was significantly higher in T_3 (2.89 ± 0.31%) and T_4 (2.68 ± 0.13%) compared to T_1 (1.49 ± 0.10%) and T_2 (1.62 ± 0.13%) ($p < 0.05$). Carbohydrate levels remained statistically consistent among treatments with no evident influence of the diet composition. Ash content reached its maximum in T_3 (1.78 ± 0.19%) and its minimum in T_4 (1.14 ± 0.06%), reflecting differences in mineral retention. Moisture content was lowest in T_3 (75.75 ± 1.17%) and highest in T_1 (79.85 ± 0.59%), indicating an inverse relationship between moisture and protein accumulation. The findings evidenced that an optimal combination of *Desmodesmus* sp. and enriched copepods, particularly in T_3, enhanced protein and lipid deposition while maintaining balanced ash and moisture levels in *C. cirrhosus* larvae under controlled rearing conditions (Table 3).

3.3. Variations in Water Quality Parameters During the Culture Period. During the study, water quality parameters, including temperature (°C), pH, DO (mg L^{-1}), and ammonia levels (mg L^{-1}), were meticulously measured daily to ensure accuracy and consistency (Table 4). All water quality parameters remained remarkably stable, exhibiting minimal fluctuations throughout the study period.

3.4. The Influence of Different Dietary Treatments on the Growth Parameters of the Indian Major Carp, Mrigal (C. cirrhosus) Larvae. Table 5 illustrates the distinct variations in growth performance among the mrigal larvae (*C. cirrhosus*) under different treatments. The most significant length increase was recorded in T_3 (17.34 mm) ($p < 0.05$), whereas T_1 exhibited the least growth (9.65 mm). Intermediate values were observed in T_4 (14.85 mm), T_5 (13.43 mm), and T_6 (12.57 mm) (Figure 4(a)). A similar trend appeared in WG, with T_3 achieving the highest increase (118.81 mg), followed by T_4 (90.64 mg), T_5 (80.18 mg), and T_6 (75.64 mg), while T_1 displayed the lowest gain as 31.21 mg ($p < 0.05$) (Figure 4(b)). Regarding percentage length gain (LG), T_3 led with 299.69%, with T_4 (257.93%), T_5 (233.23%), and T_6 (218.40%) following sequentially, whereas T_1 showed the lowest increase (168.54%). A corresponding pattern was observed in percentage WG, where T_3 showed the most pronounced rise (10,069.49%) ($p < 0.05$), succeeded by T_4 (7681.44%), T_5 (6794.47%), and T_6 (6410.25%), with T_1 at the lowest (2630.51%) (Figure 4(c)). The growth parameters exhibited consistent trends across all indices, with T_3 performing the best, followed by T_4, T_5, and T_6. In contrast, T_1 remained the weakest performer in all aspects, reinforcing the considerable differences in larval development across treatments and highlighting significant variations in both absolute and relative increases in length and weight.

3.5. Impact of Diverse Experimental Diets on the SGR % of C. cirrhosus Larvae. In the T_3 diet, the SGR of *C. cirrhosus* larvae increased by 15.84% per day ($p < 0.05$), demonstrating

Table 2: Proximate composition of different diets used in the larval rearing (% dry matter basis).

Essential nutrients	*Desmodesmus* sp. (%)	Enriched copepods (%)	Commercial feed (%)
Protein	48.50 ± 0.39	70.15 ± 0.52	40.27 ± 0.36
Lipid	23.56 ± 0.58	16.21 ± 0.25	8.30 ± 0.42
Carbohydrate	14.54 ± 0.49	6.12 ± 0.27	38.52 ± 0.41
Ash	9.64 ± 0.10	4.13 ± 0.38	9.29 ± 0.57
Fiber	3.76 ± 0.60	3.39 ± 0.37	3.62 ± 0.24

Note: The data are presented as mean value ± SD.

Table 3: Biochemical composition of mrigal, *Cirrhinus cirrhosus* larvae under different treatments (% dry matter basis).

Proximate composition	Various experimental treatments					
	T_1	T_2	T_3	T_4	T_5	T_6
Moisture (%)	79.85 ± 0.59^{a}	79.77 ± 0.68^{a}	75.75 ± 1.17^{c}	77.36 ± 1.38^{ab}	78.31 ± 0.61^{ab}	77.8 ± 0.38^{ab}
Dry matter (%)	20.15 ± 0.56^{d}	20.24 ± 0.20^{d}	24.25 ± 0.42^{a}	22.64 ± 0.30^{b}	21.69 ± 0.17^{c}	22.2 ± 0.24^{bc}
Protein (%)	15.20 ± 0.17^{c}	15.07 ± 0.26^{c}	17.87 ± 0.15^{a}	17.07 ± 0.29^{b}	16.51 ± 0.20^{b}	16.63 ± 0.30^{b}
Lipid (%)	1.49 ± 0.10^{d}	1.62 ± 0.13^{d}	2.89 ± 0.31^{a}	2.68 ± 0.13^{ab}	1.92 ± 0.15^{cd}	2.28 ± 0.1^{bc}
Carbohydrate (%)	2.14 ± 0.17^{a}	2.26 ± 0.11^{a}	1.7 ± 0.09^{a}	1.75 ± 0.15^{a}	1.98 ± 0.24^{a}	1.92 ± 0.47^{a}
Ash (%)	1.32 ± 0.07^{ab}	1.29 ± 0.09^{ab}	1.78 ± 0.19^{a}	1.14 ± 0.06^{b}	1.28 ± 0.11^{ab}	1.37 ± 0.3^{ab}

Note: The data are presented as mean value ± SD. Means with different letters in the same row are significantly different from one another ($p < 0.05$). [T_1 ($10–13 \times 10^4$ cells mL^{-1} live *Desmodesmus* sp.), T_2 ($7.5–9.5 \times 10^4$ cells mL^{-1} live *Desmodesmus* sp. + 5–10 individuals mL^{-1} enriched live copepods), T_3 ($5–6.5 \times 10^4$ cells mL^{-1} live *Desmodesmus* sp. + 10–20 individuals mL^{-1} enriched live copepods), T_4 ($2.5–3.25 \times 10^4$ cells mL^{-1} live *Desmodesmus* sp. + 15–30 individuals mL^{-1} enriched live copepods), T_5 (20–40 individuals mL^{-1} enriched live copepods), and T_6 (0.5–1.0 gL^{-1} commercial feed)]. The alphabetical superscripts "a, b, c, d, and e" given on mean values in a row indicate the statistically significant difference from one another. "a" indicates the highest value, and then "b, c, d, and e" indicate the subsequent values.

Table 4: Water quality parameters observed in different treatments during the culture period.

Diets	Water quality parameters			
	pH	Temperature (°C)	DO (mg L^{-1})	Ammonia (mg L^{-1})
T_1	8.42 ± 0.05	27.73 ± 0.09	6.91 ± 0.02	1.01 ± 0.06
T_2	8.35 ± 0.06	27.68 ± 0.01	6.95 ± 0.01	0.98 ± 0.03
T_3	7.91 ± 0.07	27.63 ± 0.10	6.82 ± 0.03	0.94 ± 0.01
T_4	8.12 ± 0.07	27.62 ± 0.04	6.78 ± 0.02	1.03 ± 0.02
T_5	8.32 ± 0.06	27.53 ± 0.01	6.75 ± 0.02	1.07 ± 0.01
T_6	8.64 ± 0.06	27.41 ± 0.01	6.67 ± 0.02	1.17 ± 0.05

Note: The data are presented as mean value ± SD. (T_1 [$10–13 \times 10^4$ cells mL^{-1} live *Desmodesmus* sp.], T_2 [$7.5–9.5 \times 10^4$ cells mL^{-1} live *Desmodesmus* sp. + 5–10 individuals mL^{-1} enriched live copepods], T_3 [$5–6.5 \times 10^4$ cells mL^{-1} live *Desmodesmus* sp. + 10–20 individuals mL^{-1} enriched live copepods], T_4 [$2.5–3.25 \times 10^4$ cells mL^{-1} live *Desmodesmus* sp. + 15–30 individuals mL^{-1} enriched live copepods], T_5 [20–40 individuals mL^{-1} enriched live copepods], and T_6 [0.5–1 gL^{-1} commercial feed]).

Table 5: Assessment of growth characteristics in *C. cirrhosus* larvae under various dietary conditions.

Factors	Different treatments of experimental diets					
	T_1	T_2	T_3	T_4	T_5	T_6
GL (mm)	9.65 ± 0.40^{d}	9.80 ± 0.30^{d}	17.34 ± 0.29^{a}	14.85 ± 0.42^{b}	13.43 ± 0.76^{c}	12.57 ± 0.37^{c}
GW (mg)	31.21 ± 0.28^{f}	39.80 ± 0.30^{e}	118.81 ± 0.37^{a}	90.64 ± 1.86^{b}	80.16 ± 0.19^{c}	75.64 ± 1.39^{d}
LG %	168.54 ± 0.41^{e}	172.50 ± 0.48^{e}	299.69 ± 0.66^{a}	257.93 ± 3.55^{b}	233.23 ± 2.19^{c}	218.40 ± 1.90^{d}
WG %	2630.51 ± 34.88^{f}	3383.9 ± 20.40^{e}	$10{,}069.49 \pm 41.55^{a}$	7681.44 ± 45.54^{b}	6794.47 ± 45.74^{c}	6410.25 ± 22.68^{d}

Note: The data are expressed as mean ± standard deviation for three replicates. Distinct superscript letters within the same row indicate significant differences ($p < 0.05$). LG% = percent length gain; WG% = percent weight gain. (T_1 [$10–13 \times 10^4$ cells mL^{-1} live *Desmodesmus* sp.], T_2 [$7.5–9.5 \times 10^4$ cells mL^{-1} live *Desmodesmus* sp. + 5–10 individuals mL^{-1} enriched live copepods], T_3 [$5–6.5 \times 10^4$ cells mL^{-1} live *Desmodesmus* sp. + 10–20 individuals mL^{-1} enriched live copepods], T_4 [$2.5–3.25 \times 10^4$ cells mL^{-1} live *Desmodesmus* sp. + 15–30 individuals mL^{-1} enriched live copepods], T_5 [20–40 individuals mL^{-1} enriched live copepods], and T_6 [0.5–1.0 gL^{-1} commercial feed]). The alphabetical superscripts "a, b, c, d, and e" given on mean values in a row indicate the statistically significant differences among the means.
Abbreviations: GL = gain in length; GW = gain in weight.

a rapid growth rate (Figure 5). T_4 (14.52% per day), T_5 (14.11% per day), and T_6 (13.92% per day) showed significantly higher SGR than T_1 (11.29% per day) and T_2 (11.85% per day) but remained lower than T_3 ($p < 0.05$). The introduction of enriched copepods alongside *Desmodesmus* sp. in T_3 resulted in superior growth performance. A decline in SGR was observed in T_4, T_5, and T_6 as the proportion of *Desmodesmus* sp. decreased. The lowest SGR was recorded in T_1, where larvae were fed exclusively on *Desmodesmus* sp., suggesting that only a single-species microalgal live diet is

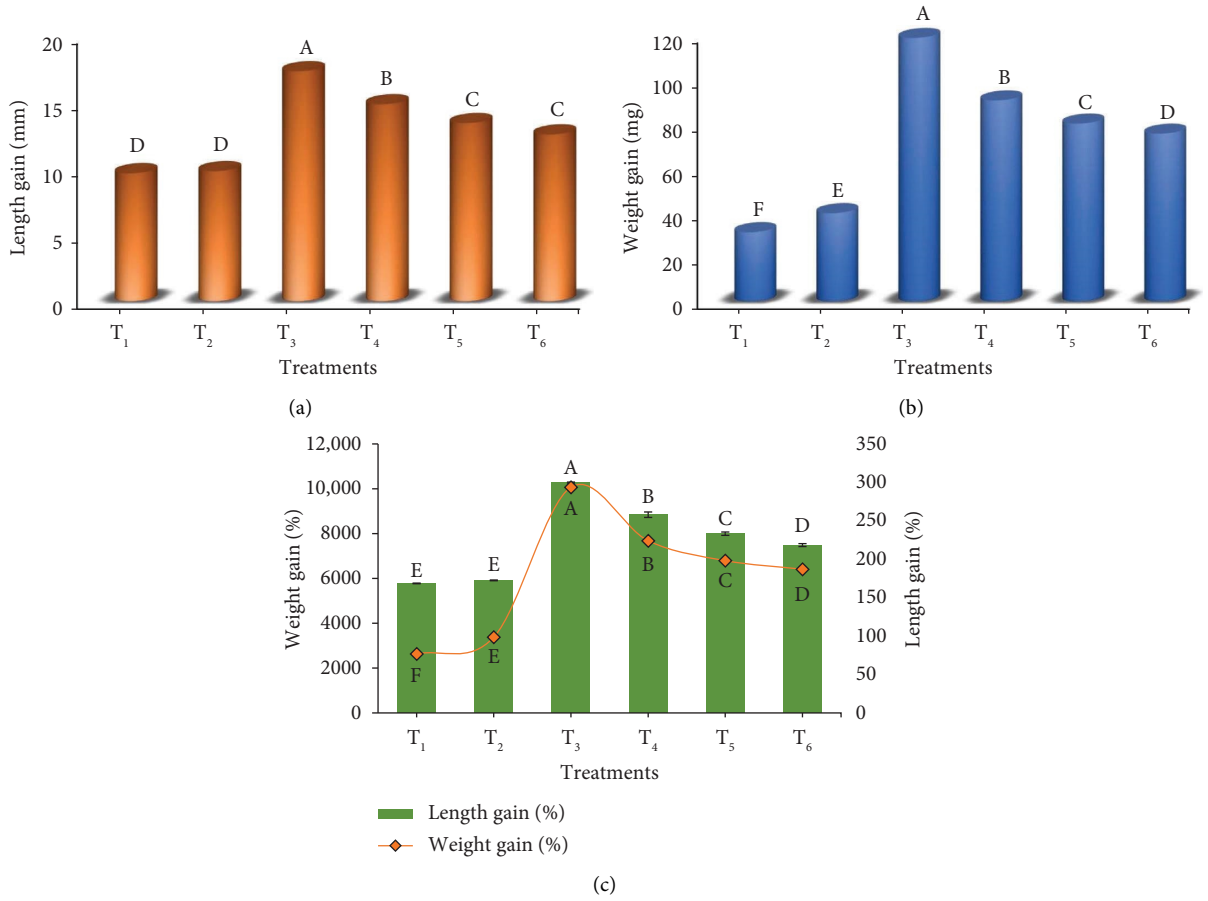

FIGURE 4: Growth development of *Cirrhinus cirrhosus* larvae in terms of (a) gain in length, (b) gain in weight, and (c) percent length gain and percent weight gain on the 30th day of the rearing period. (T_1 [10–13 × 10^4 cells mL^{-1} live *Desmodesmus* sp.], T_2 [7.5–9.5 × 10^4 cells mL^{-1} live *Desmodesmus* sp. + 5–10 individuals mL^{-1} enriched live copepods], T_3 [5–6.5 × 10^4 cells mL^{-1} live *Desmodesmus* sp. + 10–20 individuals mL^{-1} enriched live copepods], T_4 [2.5–3.25 × 10^4 cells mL^{-1} live *Desmodesmus* sp. + 15–30 individuals mL^{-1} enriched live copepods], T_5 [20–40 individuals mL^{-1} enriched live copepods], and T_6 [0.5–1 gL^{-1} commercial feed]). Means with different letters are significantly different from one another ($p < 0.05$).

less effective than mixed diets (microalgae and zooplankton) or commercially formulated feed.

3.6. Effects of Various Experimental Diets on the Survival Rate of the Indian Major Carp, Mrigal (C. cirrhosus) Larvae. The survival rate of *C. cirrhosus* larvae differed significantly among the treatments (Figure 6). The highest survival was recorded in T_3 (92.55%), followed by T_4 (90.15%), with both treatments showing significant ($p < 0.05$) differences from T_1 (69.43%). T_5 (85.48%), T_6 (84.21%), and T_2 (82.44%) exhibited moderate survival rates significantly higher than T_1 but lower than T_3 and T_4. The combination of *Desmodesmus* sp. and copepods in T_3 resulted in the highest survival, emphasizing the benefits of mixed live feed. T_4 also demonstrated a high survival rate, indicating the effectiveness of the increased copepod density. T_6 showed comparable survival to live feed treatments except T_3 and T_4. The results highlighted the critical role of live feed combinations in optimizing *C. cirrhosus* larval survival.

3.7. Insights Into Larval Development After the Experimental Period. Differential effects on larval growth and development were observed across the treatments, with T_3 demonstrating superior performance among all the treatments. In Figure 7(c), the initial larval length measured 5.76 mm, notably increased to 23.02 mm in the T_3 treatment, which exhibited the most substantial growth ($p < 0.05$). In contrast, T_4, T_5, and T_6 displayed respective increases of 20.61, 19.18, and 18.33 mm. Conversely, T_1 and T_2 exhibited the least growth, evident from their attained lengths of 15.47 and 15.67 mm, as depicted in Figure 7. A similar trend was observed in weight measurements, with T_3, T_4, and T_6 registering 120, 91.82 and 76.82 mg per individual, respectively. At the same time, T_1 and T_2 resulted in the lowest values as 32.22 and 41.11 mg per individual. These disparities in growth (GL and GW) are attributed to variations in the nutrient compositions of the diets provided. Notably, the T_3 diet comprising (5–6.5) × 10^4 cells mL^{-1} live *Desmodesmus* sp. and 10–20 individuals mL^{-1} enriched live copepods yielded the most favorable outcomes.

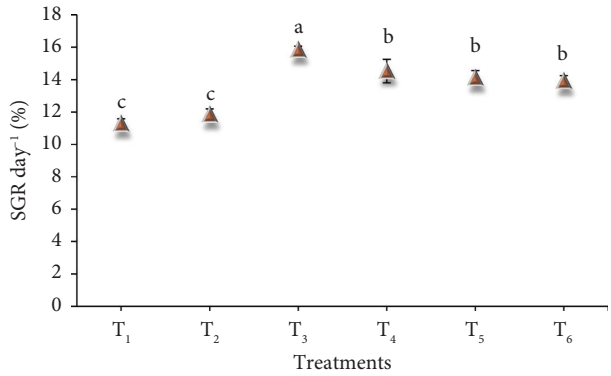

FIGURE 5: Growth performance of *Cirrhinus cirrhosus* larvae in terms of specific growth rate (%). (T_1 [10–13 × 10^4 cells mL^{-1} live *Desmodesmus* sp.], T_2 [7.5–9.5 × 10^4 cells mL^{-1} live *Desmodesmus* sp. + 5–10 individuals mL^{-1} enriched live copepods], T_3 [5–6.5 × 10^4 cells mL^{-1} live *Desmodesmus* sp. + 10–20 individuals mL^{-1} enriched live copepods], T_4 [2.5–3.25 × 10^4 cells mL^{-1} live *Desmodesmus* sp. + 15–30 individuals mL^{-1} enriched live copepods], T_5 [20–40 individuals mL^{-1} enriched live copepods], and T_6 [0.5–1.0 gL^{-1} commercial feed]). Means with different letters are significantly different from one another ($p < 0.05$).

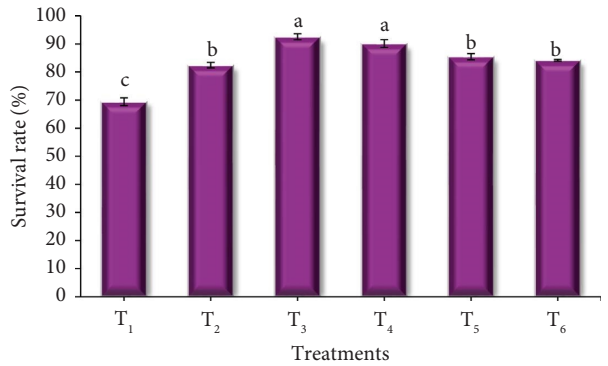

FIGURE 6: Larval survival trends observed under different experimental diets. (T_1 [10–13 × 10^4 cells mL^{-1} live *Desmodesmus* sp.], T_2 [7.5–9.5 × 10^4 cells mL^{-1} live *Desmodesmus* sp. + 5–10 individuals mL^{-1} enriched live copepods], T_3 [5–6.5 × 10^4 cells mL^{-1} live *Desmodesmus* sp. + 10–20 individuals mL^{-1} enriched live copepods], T_4 [2.5–3.25 × 10^4 cells mL^{-1} live *Desmodesmus* sp. + 15–30 individuals mL^{-1} enriched live copepods], T_5 [20–40 individuals mL^{-1} enriched live copepods], and T_6 [0.5–1.0 gL^{-1} commercial feed]). Means with different letters are significantly different from one another ($p < 0.05$).

4. Discussion

4.1. Biochemical Composition of the Major Carp Mrigal, C. cirrhosus Larvae With Different Feeding Regimes. The biochemical composition of fish serves as a critical determinant of their physiological condition and overall health [54]. In this study, dietary supplementation with the green microalga *Desmodesmus* sp. led to notable increase in the protein, lipid, and ash contents, while the moisture content declined. The highest protein and lipid concentrations were observed in the fish larvae receiving a combined diet of *Desmodesmus* sp. and enriched copepods (T_3), demonstrating the superior nutritional quality of this dietary approach. Live feed is inherently rich in essential proteins and lipids, contributing to enhanced muscle development and reduced moisture retention compared to commercially formulated diets. Additionally, copepods serve as an abundant source of energy-dense lipids and reinforce their role in optimizing fish nutrition [55]. Similarly, Jaseera et al. [56] also suggested that the cofeeding of microalgae and zooplankton promotes lipid deposition more effectively in *Penaeus monodon* than in single-feed diets. In contrast, carbohydrate levels remained relatively stable across all treatments, indicating limited dietary influence on this parameter. This study highlights the potential of integrating live *Desmodesmus* sp. (5.0–6.5 × 10^4 cells mL^{-1}) with enriched copepods (10–20 individuals mL^{-1}) to enhance the nutritional quality of aquaculture species [57]. Future investigations should focus on evaluating the long-term implications of these dietary strategies on growth performance, metabolic efficiency, and overall health outcomes.

4.2. Influence of Water Quality Parameters on the Mrigal (C. cirrhosus) Larvae Under Different Treatment Conditions. Water temperature is a critical physical parameter that directly affects the chemical composition, physical properties, and biological processes within an aquatic system. Warm water fish thrive at temperatures between 17°C and 32°C, depending on the species, with optimal growth observed between 26.7°C and 30.0°C [58], which aligns with the conditions in this study. pH plays a significant role in determining the productivity of aquatic environments. The recorded pH levels in this study fall within the acceptable limits for fish culture. Bhatnagar and Devi [59] reported that a pH range of 6.5–8.5 is ideal for aquaculture, while values exceeding 9.5 are unsuitable. The stability of pH within the optimal range throughout the study period indicates favorable conditions for fish development. DO concentration is another vital parameter influencing fish survival and productivity. Boyd and Tucker [60] established that DO concentrations between 5.0 and 7.0 mg L^{-1} promote optimal fish growth, whereas levels below 3 mg L^{-1} significantly impair productivity. Similarly, Akter et al. [61] reported a DO range of 5–8 mg L^{-1} for aquaculture, confirming that the observed values were within the suitable limits. Ammonia primarily originates from fish metabolism and excretion, necessitating effective management to prevent toxicity. In this study, water exchange and cleaning of precipitated wastes were performed to maintain ammonia concentrations below 1.0 mg L^{-1}. Boyd and Tucker [60] emphasized that maintaining ammonia within this threshold prevents physiological stress and enhances fish survival. Overall, the monitored physicochemical parameters remained within optimal ranges, fostering an environment conducive to the growth and survival of mrigal larvae.

4.3. Impact of Various Dietary Treatments Utilizing Desmodesmus sp. and Enriched Zooplankton on Larval Development of Mrigal (C. cirrhosus). The transition from endogenous to exogenous feeding represents a crucial developmental phase

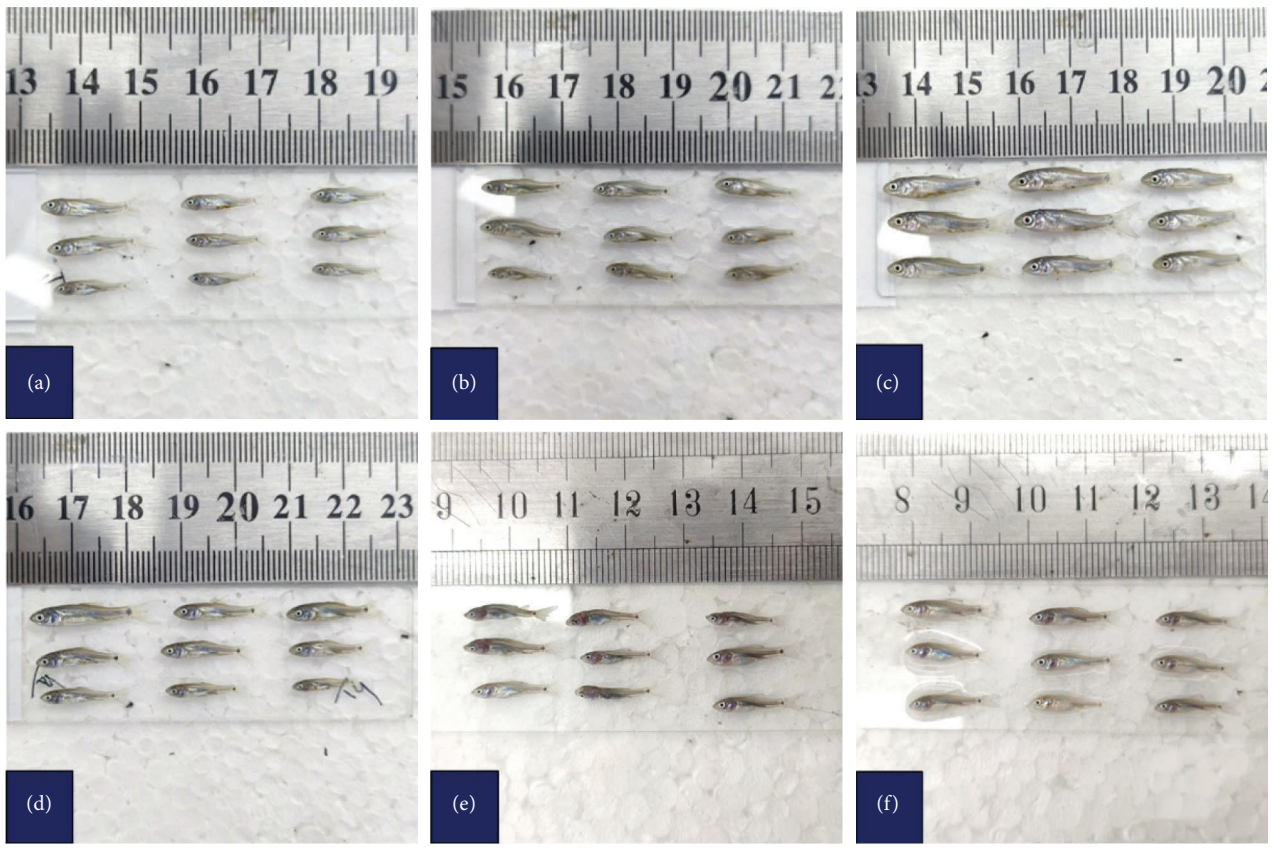

FIGURE 7: Larval development at the end of the experiment: (a) T_1 ($10–13 \times 10^4$ cells mL^{-1} live *Desmodesmus* sp.), (b) T_2 ($7.5–9.5 \times 10^4$ cells mL^{-1} live *Desmodesmus* sp. + 5–10 individuals mL^{-1} enriched live copepods), (c) T_3 ($5–6.5 \times 10^4$ cells mL^{-1} live *Desmodesmus* sp. + 10–20 individuals mL^{-1} enriched live zooplankton), (d) T_4 ($2.5–3.25 \times 10^4$ cells mL^{-1} live *Desmodesmus* sp. + 15–30 individuals mL^{-1} enriched live copepods), (e) T_5 (20–40 individuals mL^{-1} enriched live copepods), and (f) T_6 (0.5–1 gL^{-1} commercial feed).

in fish larvae, where external nutritional intake becomes essential for survival and growth [62]. Several factors influence the initial feeding success of fish, including grazing capacity, food availability, prey size, and palatability. As fish grow and mature, their dietary needs evolve in response to these factors [63, 64]. Notably, live feed resources, such as microalgae and zooplankton, play a fundamental role during this sensitive phase of many marine and freshwater fish species by providing highly digestible proteins, essential fatty acids, and bioavailable nutrients that are crucial for successful larval development and aquaculture sustainability. Researchers have consistently demonstrated the superior benefits of live feeds for rearing fish larvae and fingerlings due to their ease of digestion and nutrient assimilation [65, 66]. Radhakrishnan et al. [67] further emphasized that live food outperforms formulated diets in terms of promoting better growth and survival rates in fish larvae. While several studies have investigated the use of microalgae and copepods as dietary replacements for shrimp larvae [68–70], the potential benefits of cofeeding live feeds for freshwater fish larvae remain underexplored.

Addressing this gap, our study investigates the impact of a cofeeding strategy incorporating live *Desmodesmus* sp. and enriched copepods on the growth performance and survival rate of *C. cirrhosus* larvae. Like other fish larvae, Indian major carp larvae rely on live feeds, such as microalgae and zooplankton [63, 71]. Our study corroborates the findings of Khan and Siddiqui [63], underscoring the essential role of live feeds in the early developmental stages of mrigal carp larvae compared to commercial fish feed. Notably, the T_3 diet, consisting of live *Desmodesmus* sp. ($5–6.5 \times 10^4$ cells mL^{-1}) and enriched copepods (10–20 individuals mL^{-1}), markedly improved GL, GW, LG%, WG %, and SGR of *C. cirrhosus* larvae. The superior performance of the T_3 diet can be attributed to the provision of essential growth factors, balanced nutrients, and elevated digestive enzyme activity, which facilitate enhanced feed utilization and metabolism. These findings are consistent with previous research highlighting the role of microalgae in providing essential nutrients during early larval development [72]. These results are consistent with the findings of Zhang et al. [73], who observed improved growth performance in largemouth bass when the diets were supplemented with up to 50% *Chlorella* sp. in conjunction with other meals. Additionally, the incorporation of copepods, known for their active swimming behavior, high levels of phospholipids, essential fatty acids, and elevated digestive enzymes may further stimulate larval appetite, enhancing growth and survival [23, 36, 74]. In contrast, larvae fed formulated diets exhibited significantly lower growth performance, likely due

to reduced palatability and feed intake, as mentioned by Kissinger et al. [75]. A significant increase in survival rates was observed in larvae fed the T_3 diet compared to those receiving commercially formulated feed. This improved survival may result from the inclusion of *Desmodesmus* sp. that made the copepods more nutritious and active to act as the efficient live feed of mrigal larvae. Similar trends have been observed in other aquatic species. For instance, the survival rate of *Macrobrachium rosenbergii* postlarvae was higher when fed copepods enriched with mixed algae compared to other enrichments and diets, including *Tetraselmis* sp., *Nannochloropsis* sp., and yeast. The given diets positively influenced the SGR of the postlarvae of *M. rosenbergii* [76]. Likewise, copepod supplementation as a live feed has shown to improve the growth and survival of *Lates calcarifer* (Asian seabass) larvae [40]. Similarly, studies involving *Pampus argenteus* larvae have shown enhanced survival when multiple microalgal species were combined with zooplankton [77], which supports these findings.

Throughout the culture period, the mrigal larvae in the T_3 treatment might have experienced a favorable environment that supported their advancement in terms of growth and development. Larvae exhibit greater selective feeding behavior in T_3, adjusting their diet based on their life cycle stages and transitioning to new food sources at specific times. Kamal [78] found that mrigal larvae strongly prefer zooplankton, particularly copepods, during the first 8 days of their life cycle stages. With the development of the gill filaments and the rakers, the fish larvae progressively incorporate microalgae into their diet alongside zooplankton. Jhingran and Khan [79] observed that the alimentary canal of mrigal larvae remains relatively short in early life stages but elongates with age, reflecting a gradual dietary transition from carnivorous to herbivorous. This shift in feeding preference closely aligns with the findings of our study, further emphasizing the understanding of ontogenetic dietary changes in mrigal larvae. Consequently, larvae initially showed high survivability and overall development in the T_5 diet-fed treatment but experienced a decline due to the insufficient availability of the microalgae, which became increasingly important as their dietary preference evolved. Abdel-Tawwab et al. [80] found that *Cyprinus carpio* exhibited superior growth and survival rates when fed a combination of zooplankton and other dietary sources rather than zooplankton alone. Similarly, while the presence of both microalgae and enriched zooplankton in the T_4 feeding treatment supported good survival and growth rates from the beginning, the overall performance remained lower than in the T_3 treatment. The lower production in the T_4 treatment could be explained by inappropriate cell densities of both microalgae and zooplankton. In T_2 treatment, the larval survival rate slightly improved owing to the availability of certain enriched zooplankton that supported the early-stage growth of the larvae. Ultimately, the scarcity of proper nutrients in larval diets led to a poor survival rate, growth rate, and lower SGR in T_2 compared to the T_3-fed diet.

While Gbadamosi and Lupatsch [81] demonstrated that *Nannochloropsis salina* outperformed fish meal in the rearing of Nile tilapia (*Oreochromis niloticus*) fingerlings, our study found that the mrigal, *C. cirrhosus* larvae fed exclusively on *Desmodesmus* sp. (T_1) exhibited buoyancy issues, floating at the water surface, which led to high mortality [82]. According to Alikunhi [83], at the age of 6 days, the gill rakers of mrigal larvae are underdeveloped for filtering feed from the water. The absence of functional gill rakers may compromise their ability to filter microalgae, potentially jeopardizing survival in T_1. While they may subsist on their initial feeding, a high concentration of microalgae presents significant risks to their survival rate. The microbial degradation of uneaten cells likely contributed to deteriorating water quality, as indicated by the presence of excess bubbles despite regular water exchange in T_1. As noted by Ritu et al. [82], an excessive concentration of microalgae can reduce productivity, emphasizing the need for optimal concentrations to support larval survival. Consequently, the concentration of $(10–13) \times 10^4$ cells mL^{-1} of live *Desmodesmus* sp. (T_1) was found unsuitable for promoting larval survival rate, as well as growth indicators, such as GL, GW, and SGR. Al-Abdul-Elah et al. [77] highlighted the importance of dietary diversity, demonstrating that combining microalgae, such as *Nannochloropsis* sp., *Chlorella* sp., and *Isochrysis* sp., with rotifers yielded superior outcomes compared to microalgae alone, a finding that strongly resonates with the observations of this study with *C. cirrhosus* larvae. This trend corresponds with the findings of Fermin and Recometa [84], which demonstrated that incorporating algae with zooplankton significantly enhances live body weight and SGR compared to a 100% microalgae diet. Additionally, microalgae play a crucial role in water quality management, particularly in mitigating toxic ammonia levels. This is consistent with the observations made on other fish: bighead carp (*Aristichthys nobilis*) [84], silver pomfret (*Pampus argenteus*) [77], and shrimp (*Litopenaeus vannamei*) [85].

The use of commercially formulated feed (T_6) resulted in markedly higher mortality rates during the early stages of the experiment compared to T_3. This increase in mortality is associated with the underdeveloped digestive systems and limited enzymatic activity of the larvae at this stage, as reported in similar studies [86]. Akbary et al. [87] further noted that offering commercially formulated diets to *Oncorhynchus mykiss* larvae during their first week was associated with significantly higher mortality rates than live feed. Although *C. cirrhosus* larvae showed good acceptance of commercially formulated feed over time, noticeable disparities in size distribution emerged by the end of the experiment. Larger individuals appeared to benefit from aggressive dominance during feeding interactions, contributing to uneven growth. Despite the poor growth performance in terms of WG and LG observed in T_6-fed larvae, their SGR values were comparable to those of T_4 and T_5 throughout the study. This finding suggests that while commercially formulated feed may support minimal growth, it is unable to provide the balanced nutrition necessary for sustained and healthy development. These findings align with previous research findings of Akbary et al. [87], which demonstrated that while formulated diets can enhance growth response, they often lead to physiological imbalances, thereby limiting their effectiveness in the early larval

stages. Additionally, assessing the role of live feed, whether used alone or in combination, remains crucial in optimizing fish larval-rearing strategies.

Thus, integrating live food production techniques of both microalgae with enriched zooplankton into aquaculture can generate enhanced fish larval growth and survivability, resource-efficient and environment-friendly value chains by supplementing nutritious diets, as well as ecosystem services for the progression of the sustainable aquaculture industry and society more broadly.

5. Conclusion

This study highlights the effectiveness of combining *Desmodesmus* sp. and enriched copepods as a superior live feed for the Indian major carp mrigal, *C. cirrhosus* larvae. The T_3 diet ($5–6.5 \times 10^4$ cells mL^{-1} live *Desmodesmus* sp. + 10–20 individuals mL^{-1} enriched copepods) significantly improved growth and survival ($p < 0.05$) compared to other treatments. In contrast, *Desmodesmus* sp. alone (T_1) suppressed larval growth and survival, while enriched zooplankton alone (T_5) yielded suboptimal results. The T_6 control diet (commercial formulated feed) exhibited the lowest performance. Commercial formulated feed often fails to meet the nutritional needs of early-stage larvae, leading to poor digestibility and environmental concerns. These findings provide insights into sustainable aquaculture practices, emphasizing the effectiveness of microalgae and copepod-based feeds in optimizing larval development and bolstering fish population viability. Such approaches show promise in addressing the nutritional demands of fish larvae and advancing aquaculture as a solution for global food security concerns. Thus, these promising live feed sources could be recommended as initial feed for mrigal larvae in commercial sectors, promoting sustainable aquaculture nationally and globally.

Data Availability Statement

The data generated in this study are available upon request from the corresponding author.

Ethics Statement

This research work was conducted following the ethical standard protocols outlined by the Ethical Standard Research Monitoring Committee of the Bangladesh Agricultural University Research System (BAURES), and it has been approved by the Committee (approval letter number: BAURES/ESRC/54/2024).

Conflicts of Interest

The authors declare no conflicts of interest.

Author Contributions

Naushin Fatima and Mohammad Ariful Islam Sumon contributed to the methodology, investigation, data curation, formal analysis, and writing of the original draft. Naushin Fatima, Sadia Momota, Most. Sanjida Sultana, and Md. Mahfuzul Haque contributed to writing, review, and editing. Saleha Khan contributed to conceptualization, design, supervision, resources, validation, writing–review, editing, and funding acquisition. Naushin Fatima and Mohammad Ariful Islam Sumon contributed equally to this work.

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