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Effects of Formulated Fish Feed on Water Quality, Growth Performance, and Nutritional Properties of Catla Fish, *Catla catla*

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Abstract

The study was to investigate the comparative effects of commercial fish feed (CFF) and formulated fish feed (FFF) on pond water quality, growth performance, meat quality, survival rate, and antioxidant properties of fish. The FFF greatly reduced (from 565 to 505 µS/cm) water conductivity. The weight and length of fish were significantly ($p < 0.05$) increased by 94.1% and 42.5% in FFF treatment after 90 days of freeing as compared to CFF, indicating the higher growth performance of fish. Similarly, the highest survival growth rate was observed in FFF treatment after 60 days of release. However, the lowest food conversion ratio (0.28 g/g) was found in FFF at 90 days of freeing, representing the FFF as a quality feed. No mortality was found in both feed treatment. The protein (from 24.5 to 31.5%), and fat (from 0.8 to 1.8%) contents were significantly ($p < 0.05$) increased with FFF, however, the energy and mineral contents were nonsignificantly ($p > 0.05$) different as compared to CFF. In addition, inducing glycine (0.45 µg/g) and alanine (0.18 µg/g) in fish muscle indicates the improved quality of fish meat with the supply of FFF feed. The both total phenolic (from 1.6 to 2.8 mg/100 g) and total flavonoid (from 0.76 to 3.9 mg/100 g) contents were significantly ($p < 0.05$) increased with FFF treatment as compared to CFF treatment. Finally, FFF has the potential to improve the water quality, growth performance, nutritional properties, and bioactive compounds of Catla fish as a potential alternative to CFF feed in aquaculture.

Keywords Fish feed · Water quality · Feed composition · Fish growth · Nutritional properties

Introduction

Aquaculture is one of the world's fastest-growing food-producing industries with an average annual growth rate of 2.3% and an estimated 96.4 million tons of fish produced in 2020 (FAO 2020). In addition, commercial aquaculture produced more than half of the world's fish consumption in the last few decades (Naderi et al. 2017). According to World Fish Organization (WFO), commercial fish farming became one of the most reliable industries that meet up the global human nutritional requirements and played a pivotal role in the world economy (Ude et al. 2020). Food and Agriculture Organization (FAO) have also observed that, the global aquaculture production is around ~ 160 million tons in 2020 (FAO 2020). To maintain this high productivity, fish farmers are indiscriminately using the commercial fish feed (CFF) and that resulted in overexploitation. This exploitation greatly lowered water quality and reduced fish production as well as fish diversity (Behera et al. 2014). Besides, fish farmers in developing country often prefer traditional, lower-cost feeds over nutritionally rich, but higher-cost feeds that causes poor fish appetite, slow growth, high feed conversion ratio and low survival rate (Khan et al. 2020). It was also reported that increasing the temperature and pH level of the farming water reduces the length, survivability, diversity, and growth of fish as well as the dissolved oxygen in water (Mansour et al. 2017). Fish cultivated with CFF are also remained underdeveloped and also showing protein deficiencies. However, fish feed plays an important role in

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the quantity and quality of the production of the fish. To have a significant impact on farm net gain, feed ingredients should ensure growth, immunity, and health-promoting factors.

Nanotechnology has grown rapidly around the world and has proven to be an excellent technique for addressing a wide range of aquaculture issues including fish nutrition, water quality management, and disease treatment (Tawfik et al. 2017). Different feed additives were found to have a variety of effects such as improving growth and immunity. Therefore, the addition of novel feed ingredients integrated with nanoparticles (NPs) could have a great impact on aquaculture production. In the last few years, metal NPs showed great potential for controlling pathogenicity in aquaculture such as ZnO-NPs, FeO-NPs, and CuO-NPs (Tawfik et al. 2017; Khan et al. 2020). ZnO-NPs played a vital role in survivability, immune response, and stress tolerance (Izquierdo et al. 2017). Literature also revealed that the incorporation of FeO-NPs into fish feed influenced antioxidant activities, hematology, immune response, tissue deposition, and growth performance (Khan et al. 2020). Similarly, the CuO-NPs played a great role in enhancing the immune response and the biological as well as the physiological connection of aquatic animals (Muralisankar et al. 2016). Vitamins and minerals are essential nutrients for the growth and normal body functions in fish. They repair cellular damages and convert food into energy. A recent study investigated the impact of vitamin C supplementation integrated with SeO_2-NPs on the evaluation of somatostatin, a hormone that helps to regulate a variety of physiological movements and metabolism (Khan et al. 2017). In addition, the effect of mixed nanoparticles (MNPs) feed on the growth, histopathology and some serum biochemical alteration were studied (Fadl et al. 2020). However, to the best of our knowledge, no study on the incorporation of novel ingredients with synthesized NPs for formulated fish feed (FFF) has yet been conducted.

In this work, a novel fish feed was formulated with the inclusion of NPs. Besides, some novel ingredients like prawn powder, mustard oil, egg, and vitamin premixes were also mixed with the synthesized NPs to form a novel FFF. This feed was then supplied to the fish as their daily feed. Eventually, a comparative study of the effect of FFF and CFF on the growth performance, survivability, proximate composition, amino acid composition, and antioxidant properties of Catla fish was performed.

Materials and Methods

Materials

All the chemicals and reagents for nanoparticle preparation were purchased of analytical grade either from Sigma Aldrich, China, or Merck, India. The other materials: corn powder, prawn powder, oils, and vitamin premixes were purchased from the local market in Jashore Sadar, Jashore, Bangladesh.

Formulation of Novel Fish Feed

The FFF was prepared by using locally available feed ingredients. The ingredients and proximate composition of FFF were analyzed and summarized in Table 1. To prepare the FFF, corn powder, prawn powder, oils (soybean and mustard), and vitamin premixes were used as the major source of carbohydrate, protein, lipid, and vitamins, respectively. A homogeneous mixture of egg powder was also used as a binder for this formulation. The Fe nanoparticles (FeO-NPs), Zn nanoparticles (ZnO-NPs), and Cu nanoparticles (CuO-NPs) were synthesized. In distilled water, NaOH (1.0 M) was dissolved and the solution was agitated at 90 °C temperature. For 26 min, a 0.5 M $CuCl_2$ solution was added dropwise to the NaOH solution. Then the mixture was stirred for 2 h at 90 °C and kept overnight to form a precipitate. The suspension was then filtered and rinsed multiple times with deionized water. Then the suspension was washed and dried at 70 °C temperature to obtain Cu nanoparticles. Similarly, using $FeCl_2$ and $ZnCl_2$, chemical precipitation was used to synthesis Fe and Zn nanoparticles (Tarafder et al. 2020). To confirm, NPs were subjected to characterize the surface morphology, structure, crystallinity and phase formation using different instruments like scanning electronic microscopy (SEM), energy dispersive X-Ray (EDX), and X-ray diffractometer (XRD). Then the FeO-NPs, ZnO-NPs, and CuO-NPs were gradually mixed with the FFF ingredients. The required amount of deionized water was gradually added to the mixture of different ingredients for the formation of a dough-like structure. Then, it was compressed into pellets by using a pellet maker. Finally, the wet pellets were dried

Table 1 Proximate composition of the FFF with the supplemented amount

Ingredients	Amount	Proximate compositions	
Corn powder	61.42%	Dry matter	8.98%
Prawn powder	17.55%	Crude protein	43.2%
Soybean oil	2.19%	Lipid	17.6%
Mustard oil	2.19%	Ash	11.5%
Egg	16.23%	Fat	10.9%
Vitamin premixes	0.39%	Carbohydrate [a]	27.6%
FeNPs	0.002%	Nanoparticle	0.0054%
ZnNPs	0.002%	Energy [b] (MJ/Kg)	22.245
CuNPs	0.0005%		

[a] Carbohydrates were calculated as 1000—(protein (g/ Kg) + lipid (g / Kg) + ash (g /Kg))

[b] Energy was calculated based on 24.3, 39.7, and 17.2 g/KJ of protein, lipid, and carbohydrates, respectively

under sunlight until the moisture is less than 10% and stored at room temperature in a plastic container for further use.

Fish Sampling and Feeding

As a part of experimental design, 50 kg Catla fish (average weight and length of per fish were ~ 273.5 g and ~ 8.7 inches, respectively) was collected from a local Hatchery in Churamonkathi, Jashore Sadar, Jashore, Bangladesh. These fishes were acclimatized in a concrete pond for seven days. The acclimatized fish sample was then divided into two equal parts (25 kg in each part) and released into the respective experimental ponds. The pond water and soil were evaluated before and after the application of feeds. The CFF and FFF feed were applied to the pond at the rate of 3% and 0.3% of the body weight of fish, respectively. A daily routine was followed to maintain the schedule of fish feeding in every morning at 8.0 am. As previously reported, the provided fish feed was sunk properly and fishes were able to take the feed easily (Wang and Li 2011). The water conductivity, pH level, temperature, and the dissolved oxygen (DO) in the pond water were also measured once in a week. The water quality of the pond treated with CFF and FFF was presented in Table 2.

Growth Performance Analysis

To evaluate the growth performance, the initial weight (g), final weight (g), weight gain (%), initial length (inch) and length gain (%) of catla fish were calculated and presented in Table 3. The specific growth rate (SGR, %) and feed conversion ratio (FCR) were calculated using the following formulas:

1. SGR (% per day) = 100 × ln (final weight) − ln (initial weight) / days
2. WG (%) = 100 × (final weight − initial weight) / initial weight
3. FCR = (total feed casting − total fish residue) / (total fish final weight—total fish initial weight + total fish mortality weight)
4. Survival rate (%) = 100 × (final number of fish) / (initial number of fish)
5. Length gain (%) = 100 × (final length – initial length) / initial length

Proximate Composition of Catla Fish

The fish samples treated with CFF and FFF were subjected to proximate analysis, where the standard AOAC (Association of Official Analytical Chemists) methods for moisture,

Table 2 Effects of CFF and FFF on water quality of the pond before and after feeding the Catla fish (*Catla catla*)

Feed treatment	Water quality				Mineral contents (mg/g)					
	pH	Temperature (°C)	Conductivity (mS/cm)	DO (mg/L)	Pond water			Pond soil		
					Fe	Cu	Zn	Fe	Cu	Zn
CFF_b	7.4 ± 0.05b	26.4 ± 0.2a	565 ± 0.5b	6.51 ± 0.02a	0.43 ± 0.02b	0.05 ± 0.01b	0.11 ± 0.01 cd	1.16 ± 0.05b	0.44 ± 0.01b	1.82 ± 0.08a
CFF_a	7.9 ± 0.03a	26.5 ± 0.3a	575 ± 0.7a	6.95 ± 0.03a	0.45 ± 0.03b	0.07 ± 0.02b	0.46 ± 0.03a	0.69 ± 0.02c	0.08 ± 0.02c	0.33 ± 0.02b
FFF_b	7.4 ± 0.02b	26.3 ± 0.2a	565 ± 0.5b	6.59 ± 0.02a	0.47 ± 0.02b	0.09 ± 0.01a	0.14 ± 0.01c	1.29 ± 0.04a	0.65 ± 0.02a	0.15 ± 0.01c
FFF_a	7.7 ± 0.04ab	25.5 ± 0.4a	505 ± 0.6c	6.85 ± 0.01a	0.65 ± 0.03a	0.10 ± 0.01a	0.25 ± 0.02b	1.18 ± 0.06b	0.49 ± 0.03b	1.82 ± 0.07a

Values were expressed as mean (n = 3) ± SD. The column with different subscript letters is significantly different at $p < 0.05$

CFF_b commercial fish feed before supplying into the pond, CFF_a commercial fish feed after supplying into the pond, FFF_b formulated fish feed before supplying into the pond, FFF_a formulated fish feed after supplying into the pond, *DO* dissolved oxygen, *SD* standard deviation.

Table 3 Effect of CFF and FFF on growth performance and survival rate of Catla fish (*Catla catla*)

Growth parameters	Initial (20 days)	After 30 days		After 60 days		After 90 days		After120 days	
		CFF	FFF	CFF	FFF	CFF	FFF	CFF	FFF
Weight (g)	273.3 ± 1.5i	285.4 ± 1.35 h	370.3 ± 1.23 g	386.7 ± 1.30f	459.7 ± 1.03c	393.3 ± 1.15e	483.6 ± 1.05b	443.3 ± 1.27d	530.5 ± 1.10a
Weight gain (%)	8.7 ± 0.6e	4.4 ± 0.10 g	35.5 ± 0.80 g	41.5 ± 0.76f	68.2 ± 0.98c	43.9 ± 0.93e	76.9 ± 0.95b	62.2 ± 0.93d	94.1 ± 0.95a
Length (inch)	-	10.3 ± 0.56d	12.2 ± 0.34a	11.7 ± 0.43bc	12.3 ± 0.14a	11.8 ± 0.18bc	12.4 ± 0.30a	11.9 ± 0.26b	12.5 ± 0.23a
Length gain (%)	-	18.4 ± 0.45	40.2 ± 0.24	34.4 ± 0.56	41.3 ± 0.65	35.6 ± 0.67	42.5 ± 0.74	36.7 ± 0.36	43.6 ± 0.66
SGR(%day-^{1})		0.14 ± 0.02 g	1.01 ± 0.05a	0.58 ± 0.04d	0.87 ± 0.07b	0.40 ± 0.09f	0.63 ± 0.06c	0.40 ± 0.07f	0.55 ± 0.12de
FCR (g/g)		33.47 ± 0.58a	0.42 ± 0.03f	3.77 ± 0.28d	0.57 ± 0.05e	5.11 ± 0.32b	0.31 ± 0.02 g	4.92 ± 0.34bc	0.28 ± 0.05gh
% Survivality	100	100	100	100	100	100	100	100	100

CFF commercial fish feed, *FFF* formulated fish feed, *SGR* survival growth rate, *FCR* feed conversion ratio

protein (AOAC 992.15), fat (AOAC 922.06), and ash (AOAC 923.03) were used. Carbohydrate content was calculated as the difference (Carbohydrate = [100 − moisture% – protein% – fat% – ash%]). While the energy was also calculated using the following general formula:

$$\text{Energy(MJ/100gm)} = (\text{Protein} \times 24.3) + (\text{fat} \times 39.7) + (\text{carbohydrate} \times 17.2) \quad (1)$$

Amino Acid Analysis of Catla Fish

To analyze the amino acid composition, fish sample was washed properly with ultrapure water. The muscle of the fish was removed and oven-dried for 48 h at 60 °C temperature. Then, the oven dried sample was grounded to a powder using a grinder. A 2 g of the fish powder was washed with absolute ethanol and then hydrolyzed with 10 ml (6 N) of HCl for 23 h in a scaled beaker in an electric oven at 110 °C temperature. Finally, an automatic amino acid analyzer (Hitachi L-8900, USA) was used to determine the amino acid compositions in the fish sample (Liu et al. 2017).

Determination of Minerals

For minerals detection, 2 g of different parts of the raw fish (dorsal, ventral part, and gill) was collected in a 50 ml beaker. Prepared sample was individually dissolved in 10 ml of concentrated nitric acid (HNO_3) for 6 h at 140 °C temperature. Then, 2 ml hydrogen peroxide (H_2O_2) was added and the mixture was filtered with filter paper (Whatman no.1). The solution of mixture was diluted with 50 ml of water (H_2O) and stored for further analysis. Heavy metals (like Fe, Cu, and Zn) concentrations in fish samples were then determined using the ignite atomic absorption spectrophotometer (AAS) (Model: AA-6300 Shimadzu, Japan). The standard solutions for all the metals were used according to the previously reported study (Ashoka et al. 2009). The actual metal concentration was measured using the following mathematical equation:

$$\text{Actual mineral content} = \frac{\text{Ac} \times \text{A1}}{\text{A}} \quad (2)$$

Where Ac is the concentration of the sample (mg/L).

A_1 is the volume of the sample (ml)
A is the weight of the sample (mg)
Finally, the results were expressed in mg/g

Antioxidant Properties Analysis

Total Phenolic Content

Total phenolic content (TPC) was measured according to the method used by Sumczynski et al. (2015) with slight modification. The gallic acid was used as the phenolic standard to determine the TPC present in the fish samples. A 100 µl of the extracted fish sample was mixed with 3.16 ml distilled water and 1 ml of absolute methanol to produce a stock solution. 10% (w/v) of sodium carbonate (Na_2CO_3) and 2 mM of Folin–Ciocalteu reagent were prepared. The stock solution was first added into 96-well microplates and then 200 µl of Folin–Ciocalteu reagent was added. The mixture was allowed to react for 8 min at room temperature. Then, 600 µl of the 10% Na_2CO_3 was added to the reacted mixture. Microplates were placed in the dark at 40 °C temperature for 1 h and the absorbance was taken at 765 nm against blank. The calibration curve of the gallic acid was expressed in mg of gallic acid equivalent (GAE mg/100 g extract). The TPC value was calculated using the following formula:

$$y = 5.4092x + 0.0271 \quad (3)$$

where, x is the concentration of the gallic acid from the calibration curve (mg/mL) and y is the absorbance of the sample at 765 nm.

Total Flavonoid (TF) Content

TF content was determined using a method reported by Sokamte et al. (2019) with minor modification. 1 ml of extracted fish sample was placed into a test tube and then 1 ml of 30% methanol was added to produce a stock solution of 1 mg/ml. A 300 µl of this stock solution was mixed with 1.5 ml distilled water and 300 µl of sodium nitrite (5% w/v) in a test tube. After 10 min of incubation, a 300 µl of $AlCl_3$ (10% w/v) solution and 1 ml of sodium hydroxide (1 M) solution were added to the incubated mixture and it was further incubated for 10 min at a dark place. Then, the absorbance of the incubated mixture was taken at 506 nm against the blank. The TF content was expressed as mg catechin equivalent/100 g dry extract (mg CE/100 g dry extract). The TF value was calculated using the following formula:

$$y = 0.0154x + 0.0301 \quad (4)$$

where, x is the concentration of catechin from the calibration curve (mg/mL) and y is the absorbance of the sample at 506 nm.

DPPH Radical Scavenging Activity

The antioxidant activity of the fish sample was measured in terms of DPPH radical scavenging activity following the method previously reported by Ismail et al. (Ismail et al. 2017). Firstly, a working solution containing 12 mg of pure DPPH and 50 ml of methanol was prepared. Then, 50 µl of extracted solution of the fish sample was dissolved in 1.5 ml of the working DPPH solution. The reaction mixture was vortexed to a well-mixed solution and then incubated for 40 min in a dark place at room temperature. The resultant absorbance was spectrophotometrically determined at 517 nm against the blank solution. Lastly, the DPPH free radical scavenging percentage was calculated using the following formula:

$$\text{DPPH scavenging effect}(\%) = \frac{\text{Ab} - \text{As}}{\text{Ab}} \times 100 \quad (5)$$

where A_b is the absorbance of the control sample,

As is the absorbance of the test sample.

$ABTS^{++}$ Scavenging Effect

$ABTS^{++}$ radical scavenging test was done by using the method reported by Sokamte et al. (2019) with slight modifications. The ABTS stock solution was prepared by mixing 9 ml of water with a 7 mM of ABTS concentration. The radical cation of ABTS was produced by showing the reaction with the ABTS stock solution in the presence of 100 mM potassium persulfate. Then, the mixture was kept in a dark chamber at room temperature for 15 h, and then it was diluted with 0.1 M of phosphate buffer saline (pH 7.4). A 2.99 ml of ABTS working solution was added to a 10 µl of extracted fish sample in a test tube and the mixed solution was incubated for 6 min at room temperature. While buffer saline was mixed with 2.9 ml of ABTS working solution for the control. Then, the absorbance of both the mixed solution and the control were recorded at 734 nm. The ABTS scavenging assay was expressed as a percentage and measured using the following equation:

$$\text{ABTS Radical scavenging}(\%) = \frac{\text{Ac} - \text{As}}{\text{Ac}} \quad (6)$$

where Ac and As is the absorbance of the control and the experimental sample, respectively.

Statistical Analysis

The obtained data were calculated as the mean (n = 3) ± SD (standard deviation) and analyzed by one-way of variance (ANOVA). The level of significance was at $p < 0.05$. Statistical analysis was performed using SPSS (software version 11.5). Duncan's multiple range test (DMRT) was used to compare the means of the fish fed with CFF and FFF.

Results and Discussion

Characterization of Nano-nutrients

The surface morphology, microstructure, and particle size allocation of the prepared NPs were assessed and presented in Fig. 1A–C. The average NPs size were about 17 nm, 72 nm, and 35 for the Fe, Zn, and Cu NPs, respectively. The EDX spectra (Fig. 1D) and elemental mapping of the same NPs (Fig. 1E–G) confirmed the distribution of NPs in the prepared nanocomposite. Figure 1H represents the peak points observed at different degrees (31.52, 33.50, 37.20, 48.81, 57.25, 63.40, 68.53, 77.58, and 81.7°) confirmed the presence of face-centered cubic shaped Fe, Cu, and Zn NPs (Wakisaka et al. 2020). Similar confirmation for Fe, Cu, and Zn NPs were observed in mixed nano fertilizer applied in tomato plants (Rahman et al. 2021).

Effects of CFF and FFF on Ponds Water Quality

The characteristics of the water and soil of the ponds were evaluated and the results are summarized in Table 2. Results showed that the CFF and FFF had no significant ($p < 0.05$) effects on the pH level, temperature, and dissolved oxygen in the pond water. The initial pH of the water was approximately 7.4 in both ponds. The supplying of CFF significantly

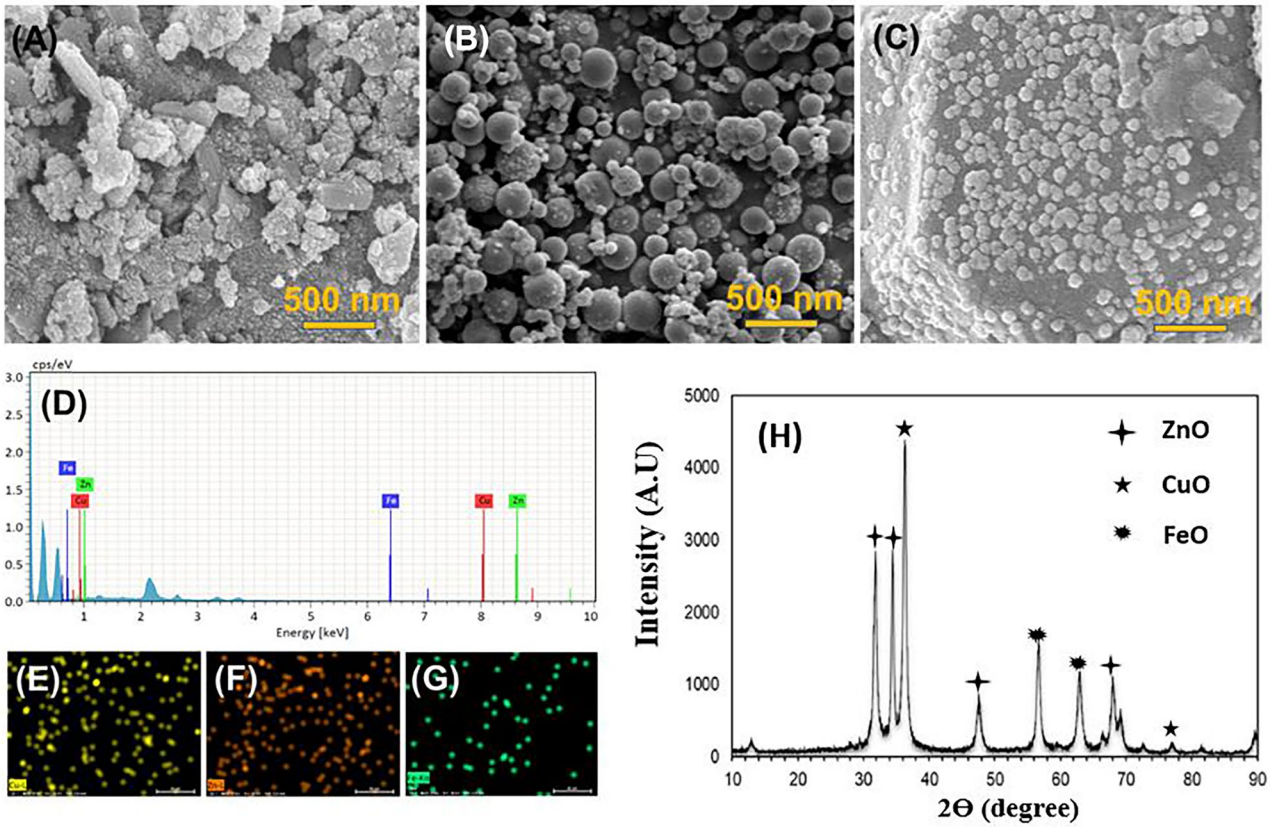

Fig. 1 SEM image of synthesized (**A**) Fe NPs; (**B**) Zn NPs; and (**C**) Cu NPs. EDX spectra of the NPs (**D**) and their corresponding elemental mapping (**E–G**). Powder XRD spectra of the prepared Fe, Zn and Cu NPs (H)

increased the pH of water, whereas it was non-significant ($p > 0.05$) in the FFF fed pond. However, the pH level in each pond was an acceptable range of 6.5–9.0. This pH range was reported to promote the growth and survival rate of fish (Mustapha and Atolagbe 2018). In addition, both feeds had no effect on the water quality and temperature in both ponds as noted in Table 2. However, the water conductivity was significantly increased (from 565 to 575 µS/cm) as well as decreased (from 565 to 505 µS/cm) with the supply of CFF and FFF respectively. The literature revealed that increasing water conductivity decreased fish growth (Makori et al. 2017). Therefore, FFF feed may lead to high growth performance and survival rate of catla fish as shown in Table 3. Additionally, CFF had no significant effects on Fe and Cu in pond water, however, the soil has significantly ($p < 0.05$) lost its Fe, Cu, and Zn (Table 2). Contrarily, supplying the water with FFF containing Fe, Cu, and Zn NPs prevented any loss of NPs from water and soil.

Effects of CFF and FFF Feed on Growth Performance of Catla Fish

The results of the comparative growth performance and survival rate of catla fish fed with CFF and FFF were summarized in Table 3. Results showed that the weight of catla fish was significantly ($p < 0.05$) increased by 35.5, 68.3 76.9, and 94.1% with the supply of FFF feed. However, the weight of fish fed with CFF increased by 4.4, 41.5, 43.9 and 62.2% after 30, 60, 90 and 120 days of releasing, respectively, which is comparatively lower than FFF. Similarly, the length

Table 4 Effect of FFF fed on proximate composition of Catla fish over CFF

Feed treatments	Proximate compositions (%)				Carbohydrate (g/100 g)	Energy (M J/Kg)
	Moisture	Ash	Protein	Fat		
CFF	34.9 ± 0.59b	6.5 ± 0.22a	24.5 ± 0.86b	0.8 ± 0.04b	33.3 ± 0.59a	1.19 ± 0.22a
FFF	37.5 ± 0.73a	5.5 ± 0.08a	31.5 ± 0.14a	1.8 ± 0.03a	23.7 ± 0.73b	1.24 ± 0.08a

CFF commercial fish feed, *FFF* formulated fish feed

of fish was also increased by 40.2, 41.3, 42.5, and 43.6% after 30, 60, 90, and 120 days of releasing, respectively, with the supply of FFF, however, the CFF fed fish had comparatively a lower increase of length as presented in Table 3. This implies the positive effect of the FFF suggesting, a balanced diet having a great combination of protein, fat, vitamins, and minerals that influenced the growth and development of fish. In addition, the SGR per day (%) was significantly ($p < 0.05$) higher in the fish fed with FFF, whereas, the FCR was significantly ($p < 0.05$) decreased after 120 days of releasing as compared to CFF. These results indicates that the FFF had no adverse effect on growth performance and feed intake as quality feed. These are an agreement with the findings previously reported by Wang and Li (Wang and Li 2011).

Effects of CFF and FFF Feed on Proximate Composition of Catla Fish

The proximate composition of Catla fish was greatly influenced by FFF over the CFF feed as shown in Table 4. The results showed that the fish fed with FFF had significantly ($p < 0.05$) increased the moisture, protein, and fat content by 2.6, 7.0, and 1.0%, respectively. In addition, the ash and carbohydrate contents were decreased by 1.01 and 9.5%. This decrease in carbohydrate contents has resulted from the increase in moisture, protein, and fat content. However, the increase of moisture, protein, and fat might be due to the supplementation of egg, vitamin premixes, and NPs in FFF feed, suggesting the higher growth performance as presented in Table 3. The increase of protein level were an agreement with the results reported by Oushani et al. (2020) in rainbow trout fish fed with dietary chitosan and nano-chitosan-loaded clinoptilolite. Similar findings were also observed in the red sea bream (Pagrus major) fish fed with Cu NPs and Cu-sulfate (El Basuini et al. 2016), blunt snout bream fish (*Megalobrama amblycephala*), and common carp (*Cyprinus carpio*) fed with dietary selenium (SeO) as nano feed.

Effects of CFF and FFF Feed on Mineral Contents in Catla Fish

The results of the comparative study on the mineral contents in Catla fish fed with FFF and CFF were summarized in Table 5. Results showed that the Cu and Zn content were not significantly ($p > 0.05$) affected by the FFF feed as compared to CFF feed. However, the Fe content was significantly ($p < 0.05$) increased in fish fed with FFF as compared to CFF feed. In addition, the lower Cu content in the fish might be due to the higher deposition of Cu into the pond soil as seen in Table 2. Zn content in fish fed with FFF was numerically higher as compared to CFF, however, statistically non-significant ($p > 0.05$). Besides, the Fe content was greatly improved by FFF feed and that might greatly influence growth performance as shown in Table 3. Similar findings were observed in the survival rate, growth performance, and nutrition of fish by Fe, Cu, and Zn NPs based diet supplementation (Behera et al. 2014; El Basuini et al. 2016; Muralisankar et al. 2014).

Table 5 Effect of CFF and FFF feed on mineral contents in Catla fish after feed treatment

Feed treatments	Mineral contents in catla fish (mg/g)		
	Fe	Cu	Zn
CFF	0.33 ± 0.02b	0.03 ± 0.0a	0.25 ± 0.01a
FFF	1.08 ± 0.05a	0.02 ± 0.0a	0.27 ± 0.01a

CFF commercial fish feed, *FFF* formulated fish feed

Effects of CFF and FFF Feed on Amino Acid Composition

Amino acids are well-known for their importance as protein building blocks, nutrient transporters, and intermediates in animal metabolic pathways. In addition, humans need dietary amino acids found in high-quality proteins for essential body functions. However, the amino acid composition of Catla fish fed with CFF and FFF was evaluated and results were shown in Table 6. The result shows that tyrosine, phenylalanine, lysine, histidine, and arginine were decreased in fish fed with FFF feed as compared to the CFF

Table 6 Effect of CFF and FFF on the amino acid composition of Catla fish (*Catla* catla)

Amino acids (µg/g fish)	Feed treatment	
	CFF	FFF
Asparagine	nd	nd
Glycine	nd	0.45
Alanine	nd	0.18
Threonine	nd	nd
Serine	nd	nd
Glutamine	nd	nd
Cysteine	nd	nd
Valine	nd	nd
Methionine	3.28	0.51
Isoleucine	nd	nd
Leucine	nd	nd
Tyrosine	0.72	0.36
Phenylalanine	7.50	5.70
Lysine	7.87	4.70
NH3	0.89	0.78
Histidine	4.80	4.79
Arginine	12.49	11.04

CFF commercial fish feed, *FFF* formulated fish feed, nd: not detected

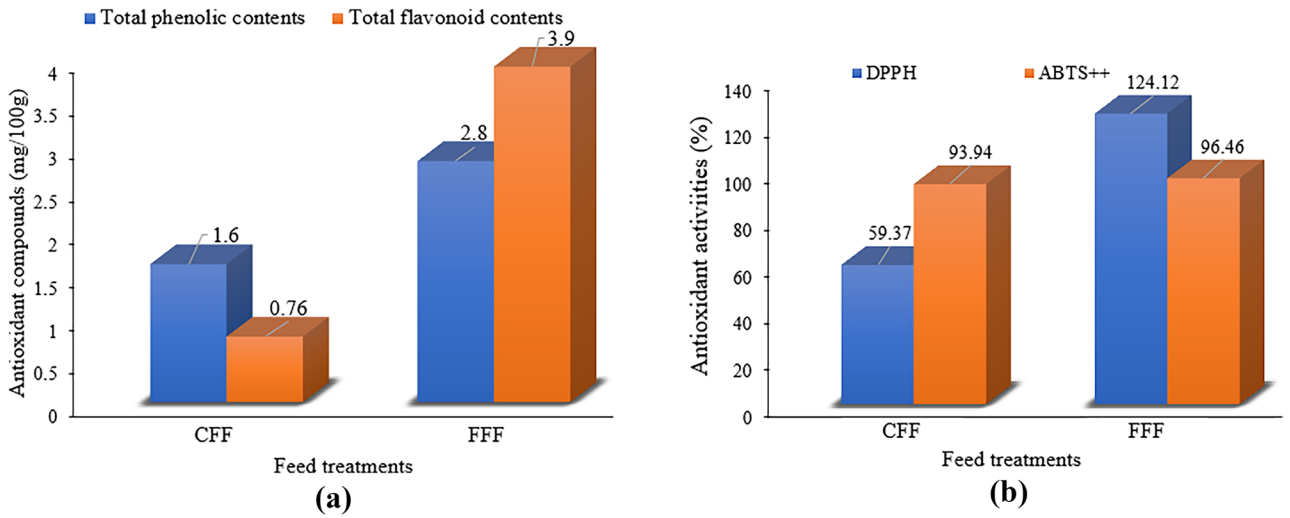

Fig. 2 **a** Antioxidant compounds in Catla fish fed with CFF and FFF (CFF: commercial fish feed, FFF: formulated fish feed). **b** Antioxidant activities of Catla fish fed with CFF and FFF (CFF: commercial fish feed, FFF: formulated fish feed)

feed. However, glycine and alanine were detected in the fish fed with FFF. Glycine is an amino acid with a long list of health advantages. It is required by the body for the production of essential compounds such as glutathione, creatine, and collagen. This amino acid can also help to protect the human liver from the negative effects of alcohol as well as to improve sleep and heart health. Xie et al. (2014), found that glycine plays a great role in weight gain and increase specific growth rate of white shrimp, *Litopenaeus vannamei*. Moreover, alanine, an amino acid, is used for the production of proteins. It aids in the absorption of tryptophan and vitamin B-6. This is a source of energy for the central nervous system and muscles. It helps the body to use sugars by strengthening the immune system. Therefore, the FFF could enhance the amino acid composition of the fish body as well as play a diverse role in the human body with other amino acids like cysteine, leucine, methionine, and lysine essential (Khan et al. 2020).

Effects of CFF and FFF Feed on Antioxidant Properties

The antioxidant properties in terms of antioxidant compounds and antioxidant activities of Catla fish fed with CFF and FFF were investigated and the results were shown in Fig. 2a, b, respectively. The total phenolic contents (TPC) and total flavonoid contents (TFC) were significantly ($p < 0.05$) increased in Catla fish fed with FFF as shown in Fig. 2a. However, the CFF feed showed lower TPC and TFC as compared to FFF. This indicates that FFF can greatly enhance the antioxidant compounds in Catla fish. Similarly, the antioxidant activities were also significantly ($p < 0.05$) increased in the fish fed with FFF indicating a significant increase of DPPH and $ABTS^{++}$ scavenging activity as shown in Fig. 2b. The increase of antioxidant properties in Catla fish fed with FFF might be due to the incorporation of NPs, vitamin premixes and minerals that improves bioactive compounds. Similar findings were found in Nile tilapia fed with nanohybrid of [GO@Se.ZnO] and Oreochromis niloticus fed with chitosan NPs and thymol (Abd El-Naby et al. 2020).

Conclusion

The novel FFF greatly influenced the maximum growth and nutritional composition of Catla fish without affecting the quality of pond water. The proposed FFF feed significantly increased the growth performance as well as the protein, fat, amino acid composition, and energy suggesting an improved meat quality in Catla fish as compared to CFF feed. The incorporation of various NPs had synergistic interaction with other supplemented ingredients, which enhances the Fe content in fish as a great source of minerals. In addition, FFF significantly increased the TPC and TFC indicating higher antioxidant properties. This study demonstrated that the addition of NPs with corn powder, prawn powder, oils, and vitamin premixes could greatly improve the growth rate, survivability and induced the physiological attributes of Catla fish.

Author Contributions MHR performed data analysis, interpretation as well as executed original draft writing, MNH & MS Carried out the experimental work including data collection and chemical test in the laboratory together, SN & MASK reviewed the article and approved the final version to be submitted, MZHK Conceptualized the work and supplied all resources required for the experiment.

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Availability of Data and Material Data are not shared publicly.

Code Availability Not applicable.

Declarations

Ethics Approval All fish experiment procedures were performed under the Guidelines and approved by the Animal Ethics Committee of Jashore University of Science and Technology.

Consent to Participate All participants are notified.

Consent for Publication All authors are informed.

Conflicts of Interest The authors declare that there is no conflict of interest.

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