

Research Article

## Different quantities of manganese oxide nanoparticles incorporated feed on the growth and haematological traits of common carp *Cyprinus carpio var. communis*

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### Abstract

In recent times, nanoparticles have been used as raw ingredients for biofertilizers, mineral supplements in animal feed, and pharmaceuticals. Manganese plays a vital role in enhancing fish's growth and biological function. The present research work aimed to analyze the various quantities of manganese oxide nanoparticles on common carp growth and its haematological traits. Synthesized manganese oxide ( $Mn_2O_3$ ) nanoparticles were illustrated using UV-visible Spectroscopy (UV-Vis), Scanning Electron Microscope (SEM), Energy Dispersive X-Ray Spectroscopy (EDAX), X-Ray Diffraction (XRD), and Fourier Transform Infrared Spectroscopy (FTIR). Six different feeds were prepared by incorporating different quantities of manganese oxide nanoparticles (Feed I (control-0mg), Feed II(3mg/100g), Feed III(6mg/100g), Feed IV(9mg/100g), Feed V(12mg/100g), and Feed VI (15mg/100g)) with common ingredients such as groundnut oil cake, fish meal, tapioca flour, and wheat flour. On the completion of 21 days, feed utilization and haematological characteristics of Common carp were assessed. The UV-Vis showed that manganese oxide nanoparticles exhibit strong adsorption peaks at 220nm. SEM image observed at the wavelength range from 9.22 nm to 9.35 nm. The size of the particles was in the 45-55 nm range. The EDAX spectrum recorded two peaks between 0.40 and 6 keV. The XRD graph shows that the diffraction peaks are indexed as 103, 004, 213, 204, 303, and 215. FT-IR spectrum measured at the wavelength range from 500-4000 $cm^{-1}$ . Most of the growth parameters and haematological parameters were higher in feed III, containing 6mg of Manganese oxide nanoparticles. Therefore, results show that manganese oxide incorporated feed enhances the growth and haematological traits in common carp compared to control feed.

**Keywords:** Different quantities, MnO nanoparticles, Growth, Haematology, Common carp

### INTRODUCTION

Nanotechnology provides an extensive array of aquaculture applications that can potentially help industry. Detection and management of diseases, water treatment, pond sterilization, effective delivery of nutrients, and medicine delivery are some of its current applications. The current state of nanotechnology in the area of aquaculture and fisheries is health management and improvement of fish and shellfish growth by dietary

supplements with nanoparticles. (Carlos Fajardo *et al.*, 2022). Nanotechnology is used extensively in many seafood industries in the form of nanoparticles as feed additives, nanofilms, nanofilters, nanobar coding, as well as fish packaging and processing. These methods provide a variety of potential for growth and the sustainability of fisheries resources. (Amir Hussain Dar *et al.*, 2020). Dietary feed supplements of nanomaterials such as zinc, iron, copper, manganese, magnesium, and selenium, and these elements fulfil all fish needs. Na-

noparticle feed supplements enhance the growth muscle composition in fish. The capability of the dietary supplements within the body depends on their size, and chemical structure. Altogether, nanoparticles are associated with aquaculture and nanomedicine due to their chemical, physical, and biological properties. (Vijayaram *et al.*, 2023).

Micronutrients are crucial in improving aquatic animal's immune system health, reproduction, and disease resistance. Every micromineral contains its role in improving the immune systems of cultured animals, but essential trace metals such as Zn, Cu, Mn, Se, and Fe. The trace metals can be incorporated into feed in different forms. From that, metal oxide nanoparticles are utilized in aquaculture as feed additives. (Marippan Yazhinipraba *et al.*, 2022) Among various metal oxide nanoparticles, manganese oxide is vital for growth, reproduction (Arthur Anderson, 1953), and avoidance of skeletal deformities in land-dwelling animals and fish. Mn absorption from water is often inadequate, requiring dietary supplementation with manganese. Additionally, Mn is essential for some fish and crustaceans' improved lifespan, muscular composition, immune system, antioxidant defense, and stress tolerance. (Asaikutti *et al.*, 2016). India has a significant role in the world's aquaculture fish production. More than 10% of the variety of fish in the world is found in India. The nation produces roughly 9.06 million metric tons of fish annually, placing it second in the world regarding overall fish output (FAO, 2020). One of the most prominent cyprinid species, the common carp, contributes to 10% of the world's productivity of freshwater aquaculture. In the current work, freshwater fish, Common carp, is an experimental model organism. To achieve maximum nutritional value from the culture system, the feed used in fish culture must ensure fish development, immunity, and good health. (Ringo *et al.*, 2016). Haematological investigation furnishes an index of corporeal changes in fish and the fish blood, an effective tool for determining alterations in the examined organism (Malgorzata Witeska *et al.*, 2022). This research aimed to analyze the impact of different manganese oxide nanoparticle dosages on the growth and haematological traits of *Cyprinus carpio var. communis*.

## MATERIALS AND METHODS

### Synthesis and characterization of manganese oxide nanoparticles

To synthesize manganese oxide ( $Mn_3O_4$ ) nanoparticles, two distinct manganese salts were used. In 50 mL of water, 2 g of  $MnCl_2 \cdot 4H_2O$  (manganese chloride tetrahydrate) and 2.4 g of  $(CH_3COO)_2Mn \cdot 4H_2O$  (manganese acetate tetrahydrate) were separately liquified. These two solutions are combined and agitated in a magnetic

stirrer with a clean magnetic bead for an hour. Then, using a burette, drops of a 200 mm solution of NaOH are added to the mixture. A brown-coloured precipitate started to form after an hour. This precipitate was gathered in the centrifuge and then rinsed thrice with water. After being dehydrated for a day at 80°C in a forced-air circulating oven, the final product was calcinated at 500°C in the muffle furnace for four hours. The calcinated product was pulverized by a mortar and pestle before further characterized by UV-visible Spectroscopy (UV-Vis), Scanning Electron Microscope (SEM), Energy Dispersive X-Ray Spectroscopy (EDAX), X-Ray Diffraction (XRD), and Fourier Transform Infrared Spectroscopy (FT-IR).

### Characterization of manganese oxide nanoparticles UV-VIS Spectroscopy analysis of manganese oxide nanoparticles

UV-VIS Spectroscopy (GENESYS180 1XX UV-VIS – Double Beam DUV 1100) is a primary technique to measure the light absorbed and scattered by a sample. This technique is used to determine the optical properties of nanoparticles. The UV-visible spectrum was recorded in the wavelength from 200nm to 800nm to confirm the manganese oxide nanoparticles.

### Scanning Electron Microscopy analysis

SEM examined the surface morphology of the nanoparticles—the scanning electron microscopy images of Manganese nanoparticles captured by the VEGA3, TESCAN (Czech Republic).

### Energy Dispersive X-Ray Spectroscopy Analysis

The elemental composition of manganese oxide nanoparticles was determined by energy-dispersive X-ray spectroscopy (Bruker nano, gmbh, D-12489 Germany, accelerating voltage - 0 to 30 keV). The spectrometer records the X-ray, which shows the composition of manganese oxide nanoparticles and the elements' concentration.

### Fourier Transform Infrared Spectroscopy

FTIR Spectroscopy technique was used to identify the functional group present in the nanoparticles. Along with the compound, the intensity of the stretching band was also identified. The intensities were plotted against the wavelength of the FTIR spectrum.

### X-Ray Diffraction Spectroscopy

X-ray diffraction spectroscopy (Panalytical X'pert3) was used to identify the crystalline structure of nanoparticles. XRD is a technique used to characterize both inorganic and organic materials. This technique has been used to determine the phase identification and the crystalline structure of nanoparticles.

### Collection and acclimation of common carp

Common carp fingerlings ( $1\pm 0.25$ g) were procured from K.V.R. Fish Farm in Palani, Tamil Nadu, India and were brought to the wet lab in oxygen-rich water polyethylene sacks. Fish were acclimated in concrete tanks for 15 days at  $28\pm 2^\circ\text{C}$  and were fed ad libitum with trainee feed, which comprised dry pellets of groundnut oil cake, fish meal, rice flour, and wheat flour.

### Experimental feed

The raw materials for feed preparation were selected based on their ability to supply nutrients and are indicated in Table 1. The feed was made by applying the Pearson's Square method of ration formulation (Ali, 1980) after the protein content had been determined by the Micro-Kjeldhal method (Jayaraman, 1992). Protein sources included groundnut oil cake and fish meal; carbohydrates included tapioca flour and wheat flour; lipid sources included vegetable oil (Sunflower) and fish oil; vitamins and minerals included Supplevite mix (Virbac Chelated Agrimin® Forte); and preservatives included sodium chloride (NaCl) and sodium benzoate. The ingredients essential to prepare the feed were powdered, put through a 425-micron sieve, and dried. The main ingredients were Groundnut oil cake, Fish meal, wheat flour, and tapioca flour. They were adequately measured and mixed with 130-150 cc of distilled water. The mixed feed dough was sterilized for 30 minutes at  $121^\circ\text{C}$  & 15 psi pressure) and cooled. Cod liver oil, refined

sunflower oil, supplevite-mix, sodium chloride (NaCl), sodium benzoate, and manganese oxide nanoparticles (3, 6, 9, 12, and 15 mg/90g) were added as minor ingredients after the feed cooled down, and the mixture was then extruded using a pelletizer. The pellets were dried in the shade at room temperature to prevent protein denature. Before usage, the prepared feed was kept in airtight containers at a temperature of  $-20^\circ\text{C}$  to avoid microbial contamination. The containers were labelled feed without nanoparticles as feed I (control), 3mg of manganese oxide nanoparticles marked as feed II, 6mg of manganese oxide nanoparticles marked as feed III, 9mg of manganese oxide nanoparticles marked as feed IV, 12mg of manganese oxide nanoparticles marked as feed V, 15mg of manganese oxide nanoparticles marked as feed VI (Table 2).

### Factorial design for growth studies of common carp

Uniform-sized Common carp *Cyprinus carpio var. communis* ( $1\pm 0.25$ g) was chosen, and the fish were placed in a rectangular polyethylene trough with 45 cm L X 30 cm B X 15 cm H and a 15-litre capacity. The initial measurements of the fish's length and weight were taken when they remained alive and without disturbing the fish. Each trough was provided with five new fish. Triplicates were maintained for every treatment. The fish were fed an ad-libitum diet of the prepared feed twice a day from 9-10 am and 4-5 pm for 1 hour each. After an hour of feeding, the unfed was collected without harming the fish. The unfed were dried and weighed accurately. With as little disturbance to the fish as possible, the faeces were collected every day before changing the water, then dehydrated at  $95^\circ\text{C}$  in a forced air circulating oven. Tap water was used to replace about 70% of the tank's water. After 21 days, the growth parameters were calculated.

### Feed utilization and growth parameters

At the end of the experiment, feed utilization and

**Table 1.** Ingredients used in the feed and protein content

S.No.	Ingredients	Protein Content (%)
1	Fish meal	58
2	Groundnut oil cake	44
3	Wheat flour	11
4	Tapioca	03
5	Fish oil	-
6	Sunflower oil	-
7	Supplevite- mix	-
8	Sodium chloride	-
9	Sodium benzoate	-

**Table 2.** Composition of different components in experimental feed (g/100gm ) of common carp

S.No	Ingredients	Experimental Feeds					
		I (Control)	II	III	IV	V	VI
1	Fish meal	33.75	33.75	33.75	33.75	33.75	33.75
2	GNOC*	33.75	33.75	33.75	33.75	33.75	33.75
3	Wheat flour	11.2	11.2	11.2	11.2	11.2	11.2
4	Tapioca	11.2	11.2	11.2	11.2	11.2	11.2
5	Fish oil	2	2	2	2	2	2
6	Sunflower oil	2	2	2	2	2	2
7	Supplevite mix	2	2	2	2	2	2
8	Sodium chloride	2	2	2	2	2	2
9	Sodium benzoate	2	2	2	2	2	2
10	Manganese oxide nanoparticles	0	3 mg	6 mg	9 mg	12 mg	15 mg

\*GNOC - Groundnut Oil Cake

growth parameters such as condition factor (K) (Weatherly and Gill, 1987) growth, percentage growth, feed consumption (FC), feed conversion efficiency (FCE), feed conversion ratio (FCR), assimilation (A), metabolism (M), and net and gross growth efficiency were all calculated by equations (Thangapandian and Monika, 2019 and Soundharia *et al.*, 2021)

**Condition Factor**

$$\text{Condition Factor (K)} = \frac{W(g)}{L (cm)^2} \quad \text{Eq. 1}$$

**Feed Consumption (FC)**

$$\text{FC} = \frac{\text{Feed given} - \text{unfed}}{\text{Number of Fishes}} \times 100 \quad \text{Eq. 2}$$

**Feed Conversion Efficiency (FCE)**

$$\text{FCE} = \frac{(W1-W0)}{Df} \quad \text{Eq. 3}$$

Where,  
W1 – Final Weight (g); W0 – Initial wet weight (g); Df – Dry feed intake (g)

**Feed Conversion Ratio (FCR)**

$$\text{FCR} = \frac{W1- W0}{\text{Feed Consumption}} \times 100 \quad \text{Eq. 4}$$

**Growth**

$$\text{Growth} = W1- W0 \quad \text{Eq. 5}$$

Where,  
W1 – Final Weight (g); W0 – Initial wet weight (g)

**Specific Growth Rate (SGR)**

$$\text{SGR} = \frac{W1-W0}{W0} \times 100 \quad \text{Eq. 6}$$

**Assimilation (A)**

$$A = \text{FC} - \text{Feacal matter} \quad \text{Eq. 7}$$

**Metabolism (M)**

$$M = A - G \quad \text{Eq. 8}$$

**Gross Growth Efficiency (GGE)**

$$\text{GGE (\%)} = \frac{\text{Growth}}{\text{Feed consumption}} \times 100 \quad \text{Eq. 9}$$

**Net growth efficiency (NGE)**

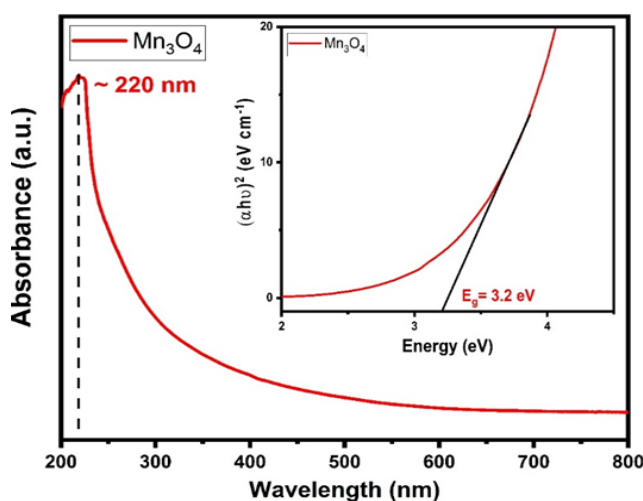
$$\text{NGE (\%)} = \frac{\text{Growth}}{\text{Assimilation}} \times 100 \quad \text{Eq. 10}$$

**Hematological traits**

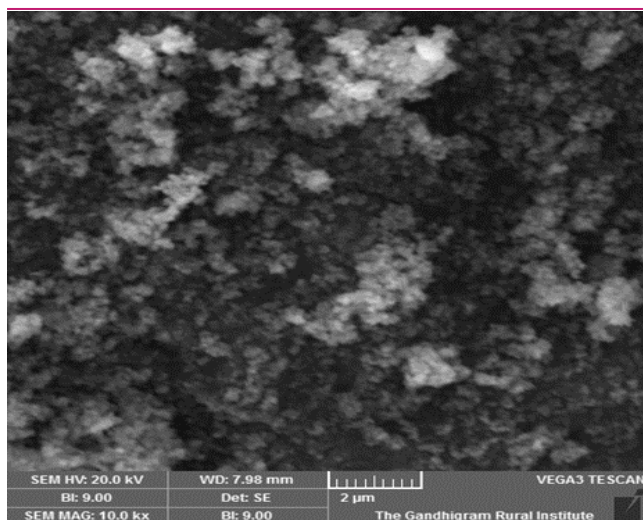
At the end of the experiment, blood samples were obtained from the caudal vein and behind the gill without harming them using an expendable syringe fitted with a tiny needle. The needle and syringe were treated with EDTA before use to prevent the blood samples from coagulation. The collected blood samples were deposited into an Eppendorf tube containing 0.1 N EDTA. When the experiment was completed, haematological traits like RBC (Red Blood Corpuscles), WBC (White Blood Cells), and platelets were determined by the Hemocytometer method (Stevens, 1997) and Hemoglobin (Hb) were analyzed by cyanmethemoglobin method (Richard Lee *et al.*, 1998). The micrometric method is used for the determination of hematocrit (HCT) (Nelson and Morris, 1989).

**RESULTS AND DISCUSSION**

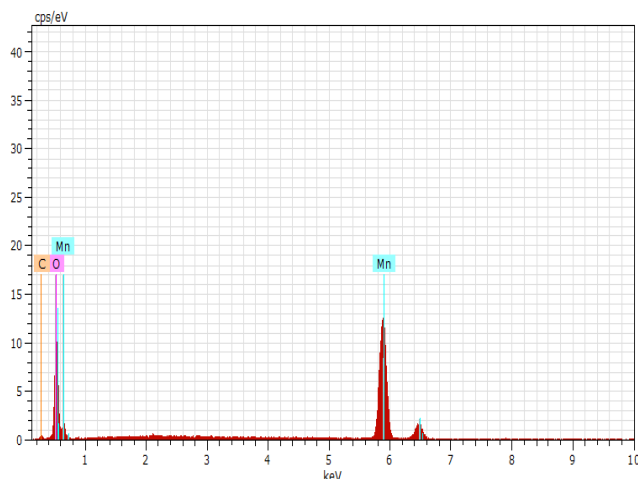
The optical characteristics of the nanosized particle can be examined using the widely used method of UV-visible absorption spectroscopy. The absorbance peak of manganese oxide nanoparticles was at 220 nm (Figure 1). A similar absorption peak(220nm) of manganese oxide nanoparticles was also reported (Mohammed Rafi Shaik *et al.* 2020). The absorption starts to decrease with an increase in the wavelength. The absorbance decreases and tails down to lower values. The energy bandgap was calculated by using the Tauc plot method, as shown in the inset of Fig 1. The Tauc plot plotted using  $(\alpha h\nu)^2$  as a function of energy indicated that the bandgap of the manganese oxide nanoparticles was 3.2 eV. SEM imaging revealed that manganese oxide nanoparticles aggregated into clusters. The particles are spherically shaped, as shown by the dark and light contrast in SEM pictures (Fig.2). The particle's size lies in the 45-55 nm range. EDAX spectrum analysis identified the



**Fig. 1.** UV-Visible Absorption Spectrum of manganese oxide nanoparticles

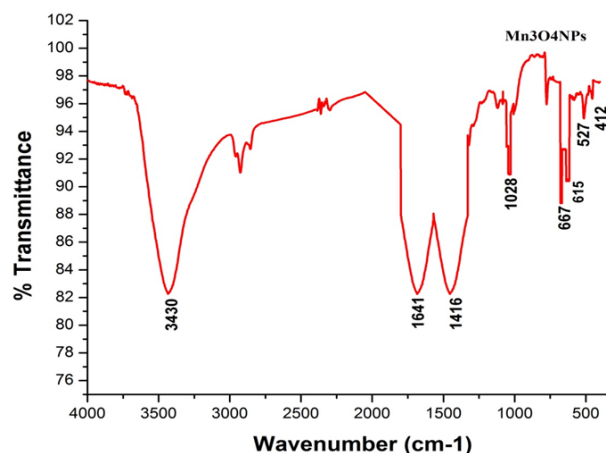


**Fig. 2.** SEM Analysis Images of manganese oxide nanoparticles



**Fig. 3.** EDAX Spectrum Image of manganese oxide nanoparticles

presence of manganese (Mn) and oxygen (O) in synthesised nanoparticles. Two peaks in the EDAX spectrum, which were recorded on the manganese oxide nanoparticles, are visible between 0.40 and 6.0 keV. On the spectrum, two peaks at 0.62 keV and 5.9 keV represented the purity of the manganese oxide nanoparticles, and there was a third peak at 0.46 keV for the element O (Fig.3). According to data from EDX, elemental manganese had a 38.99% composition. A similar finding (Gnana Sundararaj *et al.*, 2015) reported that the SEM image shows that the manganese oxide nanoparticles are spherical, with sizes ranging from 20-50nm. EDX spectrum reveals the presence of manganese and oxygen. On the EDX spectrum, two peaks were observed 0.4 KeV representing oxygen and 5.9 KeV representing manganese. FTIR spectra were examined between 500 and 4000  $\text{cm}^{-1}$ . Functional groups of active components were determined using FT-IR analysis according to the peak intensity in the infrared radiation region. At 527 nm, bands 3430, 1641,

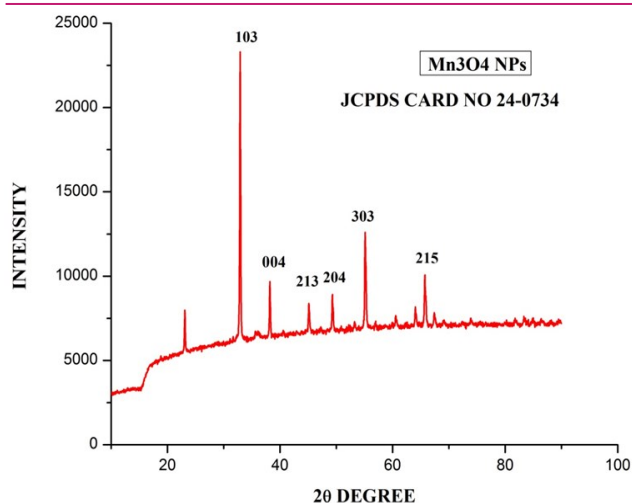


**Fig. 4.** FT-IR Spectrum graph of manganese oxide nanoparticles

1416, 1028, 657, 615, and 412, associated with N-H aliphatic primary amine, N-H amine, O-H alcohol, O-H alcohol, C=C alkene, C-I halogen, and C-O-C ether, respectively, were used for illustrating the synthesis of manganese oxide (Fig. 4). Amaini *et al.*, (2022) report that the peak at  $594 \text{ cm}^{-1}$  is assigned to the stretching mode of manganese oxide of  $\text{Mn}^{2+}$  ions at tetrahedral sites. Whereas the other peak located at  $462 \text{ cm}^{-1}$  represents the tetragonal structure of  $\text{Mn}_3\text{O}_4$ . The band centered at  $1600 \text{ cm}^{-1}$  can be attributed to the stretching and bending of the O-H group. Moreover, the broadband observed at  $3263 \text{ cm}^{-1}$  indicates the stretching vibrations of  $\text{H}_2\text{O}$  molecules on the surface of samples.

The structure and phase analysis of manganese oxide nanoparticles are analysed by the XRD technique, the XRD diffraction pattern of manganese oxide nanoparticles is represented in Fig. 5 indexed as  $32.5^\circ$  (1 0 3),  $38.2^\circ$  (0 0 4),  $50.8^\circ$  (2 1 3),  $53.8^\circ$  (2 0 4),  $55.3^\circ$  (3 0 3),  $64.8^\circ$  (2 1 5), respectively. The prepared manganese oxide nanoparticles were pure, and the distinct and sharp diffraction peaks confirmed a high degree of crystallinity. The obtained diffraction pattern was like the standard JCPDS card no: 24-0734. The synthesized  $\text{Mn}_3\text{O}_4$  NPs unequivocally displayed a tetragonal structure (hausmannite, I41/amd) with consistent lattice values of 'a' = 0.58 nm and 'c' = 0.93 nm. Furthermore, the Scherrer formula was used to estimate the average crystallite size of the synthesized manganese oxide nanoparticle and found to be 40 nm. Saba Jamil *et al.*, (2018) subjected the chemically synthesized manganese oxide NPs to XRD analysis. Also reported that all the peaks are very sharp indicating the product's crystalline nature and no extra peak is present in the XRD pattern which shows that the synthesized product does not contain any impurities.

In Table 3, the condition factor of *Cyprinus carpio* was higher in feed III(9mg/kg). The condition factor(K) is crucial to figuring out the life process of fish species



**Fig. 5.** XRD Analysis Spectrum of manganese oxide nanoparticles

and it helps maintain the ecosystem's equilibrium and proper management of the species (Okonkwo, 2011). All feeds enriched with manganese oxide nanoparticles showed an increase in the common carp's final condition factor. Feed III containing 6mg of manganese oxide, has greater value among all the feeds. According to Srinivasan *et al* (2016) the condition factor of *Macrobrachium rosenbergii*, freshwater prawn post-larvae fed with 40g/kg<sup>-1</sup> of iron oxide nanoparticles in the diet enhanced. Table 4 displays the feed utilization and growth parameters of common carp. Table 5 provides the results of the Analysis of Variance (ANOVA) for the growth parameters (Feed consumption, Growth, Net Growth Efficiency, and Gross Growth Efficiency). In the present study, feed III, which contained 6 mg of manganese oxide nanoparticles, was higher in the amount of feed that Common carp consumed. In feed utilization

**Table 3.** Condition factor of common carp

Feeds	Initial	Final
Experimental feed I (Control)	1.3±0.6	2.4±0.7
Experimental feed II	0.7±0.3	1.6±0.5
Experimental feed III	1.0±0.3	3.6±0.3
Experimental feed IV	2.4±0.2	1.7±0.6
Experimental feed V	3.4±0.4	2.9±0.4
Experimental feed VI	2.7±0.5	3.0±0.6

and growth parameters Common carp reared in feed III had greater values than other feeds. Common carp fed with 3-15 mg of manganese oxide nanoparticles increased growth performance and specific growth rate. Asaikutti *et al.* (2016) showed that the final weight and weight gain (WG), of prawns fed with a diet incorporating 3–18 mg Mn<sub>3</sub>O<sub>4</sub> nanoparticles per kg implied increased growth performance and specific growth rate. Muralisankar *et al.* (2014) reported a high in a diet supplemented with 60 mg ZnO NPs in *M. rosenbergii*. Growth, percentage growth, assimilation and metabolism were higher in feed VI. In feed conversion ratio of common carp was higher in feed III 6mg and lower in the control feed. Thangapandiyan and Monika (2019) conducted a growth study in freshwater fish *Labeo rohita* and reported that the feed conversion ratio was higher in 0.5mg/kg of ZnO nanoparticles and lower in the control feed. The assimilation and metabolism of common carp were higher in feed III containing 6mg of manganese oxide nanoparticles. Rajan and Shakthiprabha (2022) reported that the assimilation and metabolism of mrigal *Cirrhinus mrigala* were higher in feed containing 20mg of iron oxide nanoparticles. In the current work, the net and gross growth efficiency were greater in the common carp fed with 6mg of manganese oxide nanoparticles feed III. Similarly, Rajan and

**Table 4.** Feed utilization and growth parameters of common carp to different quantities of manganese oxide nanoparticles. (Each value is the average ±SD) performance of five individuals in triplicates reared for 21 days

Parameters	Experimental Feeds					
	I (Control)	II (3 mg)	III (6mg)	IV (9mg)	V (12mg)	VI (15mg)
Feed consumption (g/g live wt/21 days)	8.3±0.8	11.4±0.80	16.7±1.0	14.9±1.8	13.5±1.3	12.9±0.4
Feed conversion Efficiency	1.6±0.5	2.2±0.4	4.9±0.3	2.5±0.2	1.9±0.3	1.6±0.08
Feed conversion Ratio	4.3±1.5	5.9±1.6	12.4±1.7	10.8±0.7	9.7±0.8	6.8±0.3
Growth	1.1±0.1	1.2±0.2	2.0±0.3	1.7±0.5	1.4±0.1	1.2±0.5
Specific Growth Rate	22.4±1.4	20±0.5	40.0±0.5	35.0±1.1	25.0±1.1	24.0±1.5
Assimilation (g/g live wt/21 days)	1.3±0.1	1.5±0.1	3.3±0.28	3±0.2	2.3±0.2	1.9±0.05
Metabolism (g/g live wt/21 days)	0.3±0.2	0.4±0.05	1.3±0.2	1.1±0.05	1.2±0.2	0.7±0.1
Gross Growth Efficiency (%)	7.4±1.4	8.0±0.1	18.0±0.5	12.2±0.17	13.0±0.9	10.6±0.5
Net Growth Efficiency (%)	41±1.0	52±1.1	94±0.9	61±1.5	56±0.8	55.3±1.0

**Table 5.** ANOVA (Analysis of Variance) of growth parameters (Feed consumption, growth, gross growth efficiency, net growth efficiency) of common carp (*Cyprinus carpio var. communis*)

Parameters	Source of Variation	Sum of Squares	DF	Mean Squares	F	SIG
Feed Consumption	Between Groups	115.224	5	23.045	17.451	0.0005 S
	Within Groups	15.847	12	1.321		
	Total	131.071	17			
Growth	Between Groups	1.961	5	0.392	26.221	0.0005 S
	Within Groups	0.179	12	0.015		
	Total	2.140	17			
Gross Growth Efficiency	Between Groups	246.461	5	49.292	82.832	0.0005 S
	Within Groups	7.141	12	0.595		
	Total	253.602	17			
Net Growth Efficiency	Between Groups	4706.667	5	941.333	736.969	0.0005 S
	Within Groups	15.333	12	1.278		
	Total	4722.000	17			

**Table 6.** Haematological traits of common carp

Blood Parameters	FEED I	FEED II	FEED III	FEED IV	FEED V	FEED VI
RBC count (millions/cumm)	0.04	0.06	0.3	0.1	0.12	0.14
Hemoglobin (gm/dl)	0.1	0.2	0.7	0.3	0.5	0.6
Haematocrit (PCV) (%)	0.02	0.6	1.4	0.9	1.1	1.2
WBC count (Cells/cumm)	4,700	7,700	10,200	4,900	3,200	1900
Platelets (count Lakhs/cumm)	10,000	12,000	40,000	16,000	23,000	32,000

Roopashree (2022) reported that the Net and gross growth efficiency was higher in zebrafish *Danio rerio* fed with feed III containing 40mg of zinc oxide nanoparticles.

Hematological traits of Common carp are presented in Table 6. The white blood cell count, red blood corpuscles count, haemoglobin, hematocrit and platelets count increased with the increasing quantity of Manganese oxide nanoparticles in the feed (up to feed VI). The haematological analysis is considered the corporeal indicator of the entire fish body, making it crucial in determining the skeletal and functional condition of fish treated with nanoparticles (Rajendra Shejwal *et al.*, 2014). As common carp *Cyprinus carpio var. communis* reared from feed I to feed VI progressed, the WBC and RBC count, haemoglobin, hematocrit and platelets gradually rose. Haematological traits were higher in feed III compared to other feeds. Similar findings are reported by Atif Yaqub *et al.* (2023), who found that when the amount of ZnO NPs in meal D3 was higher than in other meals, Nile tilapia's WBC count, haemoglobin content, hematocrit, and platelets count gradually increased.

## Conclusion

The present study effectively developed to prepare manganese oxide ( $Mn_3O_4$ ) nanoparticles. The study revealed that the manganese oxide nanoparticles were safe and effective supplements for common carp. Feed III, containing six mg of manganese oxide nanoparticles

was suitable for enhancing Common carp's growth and haematological traits.

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## Conflict of interest

The authors declare that they have no conflict of interest.

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