# **Effects of Substituted Blackseed Meal Based Diet on Growth Performance and Nutrient Digestibility of** *Cyprinus carpio* Fingerlings

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#### ABSTRACT

Black seeds (*Nigella sativa*) and their derivatives have certain beneficial consequences for farmed fish. Because of its great adaptability in diet, it is being investigated as a prospective option for commercial aquaculture feed in Asia. To check the effects of black seed meal on common carp (*Cyprinus carpio*) fingerlings, a study trial was performed. The aim of this study was to investigate the effects of replacing the diet of *C. carpio* from soybean meal (SBM) to black seed meal (BSM) on growth rate, nutritional digestibility and hematological profile. Five isonitrogenous and isocaloric meals (labelled BSM-0%, BSM-25%, BSM-50%, BSM-75% and BSM-100%) were used in the experiment, each of which contained varying amounts of BSM in place of SBM (0%, 25%, 50%, 75% and 100%, respectively). Fingerlings of common carp were divided into three groups and fed for 70 days with BSM-based diets at 4% of their body weight. The group fed diet BSM-50% acquired highest rank with respect to the growth rate (final weight 25.67g, weight gain 18.52g and SGR 1.42%), hematological parameters (RBCs2.83×10<sup>6</sup>mm<sup>-3</sup>, PLT68.31×10<sup>6</sup> mm<sup>-3</sup> and Hb 8.12g/100ml) and nutrient digestibility (crude protein 70.24, crude fat 74.54), while the gross energy (74.46) was seen higher in fingerlings fed the BSM-25% diet. This study confirmed that the utilization of 50% BSM in the fish diet in replacement to SBM had a beneficial impact on the growth, nutrients digestibility and hematological parameters in common carp fingerlings.

# INTRODUCTION

The world population has grown significantly over the past few centuries, reaching approximately 8 billion people now (Milner and Boldsen, 2023). Aquaculture is one of the fastest ways to prepare food for a growing population and could become increasingly important in the future (Zhou *et al.*, 2023). The rapidly expanding source of animal protein is aquaculture. According to the State of World Fisheries and Aquaculture (SOFIA, 2018), aquaculture currently accounts for 47% of all fish produced globally and is expected to expand to 52% by 2025. The proportion of fish produced through aquaculture went up from 25.7% to 46% and it is anticipated to reach 50% by 2030 (FAO, 2020).

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#### Authors' Contribution

MMS planned, supervised and provided materials for research. IF conducted the feeding trial and prepared manuscript. MT helped in manuscript preparing. ZH and FY helped in writing, review, and editing. MU helped in editing and reshaping the manuscript.

#### Key words

Common carp (C. carpio), Black seed meal (BSM), Soybean meal (SBM), Growth rate, Nutrient digestibility, Blood profile

In semi-intensive or intensive agricultural operations, proteins constitute most of the costly of dietary source, therefore feed costs often account for 70% of overall operational costs (Hossain *et al.*, 2022). However, due to declining fishery supplies and rising fish meal (FM) demand, FM can no longer satisfy aquaculture's needs and has become a significant impediment to the growth of the industry. Aquaculture has thus long been interested in finding alternatives to FM (FAO, 2022). Because of its accessibility, environmental friendliness, sustainability and affordability, plant protein sources for FM (Latif *et al.*, 2022).

The National Bureau of Statistics of the People's Republic of China (2022) revealed that the cost of SBM in China has increased by a factor due to the high environmental cost of growing terrestrial plants that are high in protein. The cost of SBM, which is the most popular plant protein source for fish, has grown up. The price of SBM has increased by a factor from 2019 to 2022. To formulate fish feed at a reasonable price, this study seeks to identify a substitute for SBM.

For farmed fish, black seeds and its byproducts have some advantageous properties. Regarding nutrients,

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the black seed has a nutritional content of 20.8 % crude protein, 3.7 % ash, 7.0 % moisture, 34.8 % lipids and 33.7 % carbs (Atta, 2003). Studies on the possible impact of *N*. *sativa* on fish health and production have been published. The incorporation of *N*. *sativa* or its active components, such as thymoquinone, in the fish diet is shown to have a good impact on growth performance, feed conversion and meat quality (El-Hack *et al.*, 2021).

According to Dawood and Koshio (2016), common carp represents 71.9% of the freshwater fish produced worldwide and is one of the fish species that is most widely grown 4.08 million tonnes in 2017 compared to 2.41 million tonnes in 2000 (FAO, 2018). According to price structure in the supply chain for fresh carp in Central Europe (CONAPESCA, 2017) 53,421 tonnes of carp were produced in 2017, representing an estimated yearly rise of 7.43%. Inland fisheries rely heavily on common carp, the most popular cyprinid species (Vilizzi, 2012). In 1964, common carp was imported from Thailand to Pakistan for aquaculture purposes (Khan et al., 2016). In 2010, common carp constitute 52% of the average catch from Pakistan's Mangla Reservoir (Mirza et al., 2012). C. carpio has omnivorous feeding habits, a quick development rate and is easy to breed in confined water (Ram et al., 2015). Because of its great adaptability to both environment and diet, it is being investigated as a prospective option for commercial aquaculture in Asia (Rahman, 2015).

The current study was carried out to determine the most inexpensive and ecologically sound feed for fish,

as well as to look at the effects of BSM on common carp fingerlings to figure out nutrient digestibility, the ideal amount of black seed meal-based diets as a substitute protein source and the effect of BSM on the growth rate of *C. carpio* fingerlings.

# **MATERIALS AND METHODS**

#### Experiment design

Fingerlings were accustomed to lab conditions for fifteen days before the trials started. V-shaped tanks were employed, which were created especially for the experiment. To kill the pathogens, *C. carpio* fry were submerged in a container containing saline solution for 1–2 min (Rowland *et al.*, 1991). During the acclimatization phase, fish were provided with a basal diet twice over a 24hour period (Allan *et al.*, 2000). The quality of water like temperature, dissolved oxygen (DO) and pH were checked daily. Throughout the experiment, air was upheld using a capillary system and an oxygen pump.

#### Formulation of experimental diets

All ingredients (Table I) were milled into a fine powder. After weighing the necessary amounts of chromic oxide, ascorbic acid, minerals and vitamin premix using a digital electronic weighing balance, the dry materials were combined in a feed mixer for 5–10 min. Gradually, fish oil was incorporated into the amalgamation of the feed ingredients, with 15 ml of distilled water being

Ingredients %	BSM-0 %	BSM-25 %	BSM-50%	BSM-75%	BSM-100 %
Fish meal	2	5	8	11	14
SBM	52	39	26	13	0
BSM	0	13	26	39	52
Corn flour	22	19	16	13	10
Wheat bran	16	15	14	13	12
Fish oil	5	6	7	8	9
Ascorbic acid	1	-	-	-	-
Chromic oxide	1	-	-	-	-
Mineral/ vitamin	1	-	-	-	-
Total amount	100	100	100	100	100
Nutrients compositio	on in diet				
Crude protein (%)	32.09±0.84	32.11±0.86	32.10±0.94	32.11±0.94	32.09±0.75
Crude fat (%)	5.14±0.25	5.12±0.37	5.14±0.33	5.12±0.35	5.11±0.33
Gross energy (kcal-1)	4.27±0.32	4.26±0.38	4.23±0.25	4.24±0.35	4.27±0.37

Table I. The preparation of diet and the composition of the trial diets nutrients composition in diet.

\*Vit. D3: 3,000,000 IU; Vit. A: 15,000,000 IU; Vit. C: 15,000mg; Vit. B6: 4000 mg; Vit. E: 30000 IU; Vit. B2:7000 mg; Vit. B12: 40 mg; Folic acid: 1500 mg; K3: 8000 mg; Ca pantothenate: 12,000 mg; Nicotinic acid: 60,000 mg. \*\* Mg: 55g; Ca: 155 g; Se: 3 mg; Na: 45 g; P:135 g; Cu: 600 mg; Mn: 2000 mg; Co: 40 mg; Fe: 1000 mg; Zn: 3000 mg; I: 40 mg. Data are means of three replicates (± shows Standard Deviations).

added and stirred to form a dough. This dough was then forced through a 2.0 mm diameter hand-operated pelleting machine. Before formulation, experimental diets were analyzed for the chemical content of each component (Al-Khalifa and Al-Arify, 1999). The resulting pellets were then dried in the sun for 72 h and kept at 4°C until the BSM based diets (BSM 0%-BSM 100%) dried.

### Feeding regime

The feeding session for *C. carpio* fingerlings lasted for 2 h and any uneaten food was subsequently removed from the tanks by opening the valves. Feed was given to the fish twice daily at a rate of 4% of their wet body weight.

#### Straining of fecal matter

The tanks were refilled 20-30 % weekly and fecal matter was collected by using special pipes. To prevent the leaching of any nutrients into the water, the waste matter was collected, dried in an incubator at 70°C for 60 to 90 min each day and then stored for further investigation.

#### Estimation of growth response

Each tank contained *C. carpio* fingerlings weighed an average of 7.12 g. After every 15 days fish were weighed again to analyze the growth performance over the course of the entire experimental period. Weight gain (WG) in g, growth rate (SGR) and feed conversion ratio (FCR) were used to calculate the growth parameters (NRC, 1993).

$$WG\% = \frac{(\text{End weight}) g - (\text{Initial weight})g}{\text{Initial weight g}} \times 100$$
  
FCR =  $\frac{\text{All dry feed ingestion (g)}}{\text{Wet weight gain (g)}}$   
SGR% =  $\frac{(\text{end wteight of fish} - \text{Initial weight of fish})}{\text{Trial day}} \times 100$ 

#### Nutrients digestibility

After being heated to 105 degrees for 12 h, the collected samples were analyzed to determine the humidity content of the experimental meals and feces. The crude protein (CP) concentration was calculated by the Micro Kjeldahl System and the crude fat content (CF) was examined using the Soxhlet apparatus and the petroleum ether extraction (EE) procedure. After being processed with 1.25 NaOH and 1.25 %  $H_2SO_4$ , the crude fiber content of dried lipid-free residue was assessed as a cost on igniting. Ash was weighed after being ignited for 12 h at 650 °C in a Naberthem  $B_{170}$  electric furnace. The contents of  $C_x$  ( $H_2O$ ) were determined using the usual formula.

# Estimation of hematological indices

From each tank, fish were taken and a heparinized

sample needle was used to extract fish blood from the posterior vein. These samples were then sent to the research laboratory in order to analyze hematological indices. The microhematocrit method, which utilizes capillary tubes, was implemented to perform the hematocrit test (Brown, 1980). To count RBCs, a hemocytometer with Neubauer cell counting chamber was used (Blaxhall and Daisley, 1973). Wedemeyer and Yastuke's (1977) description was utilized for the evaluation of hemoglobin (Hb) concentration. MCH (mean/average of corpuscular hemoglobin), MCV (mean/average of corpuscular volume) and MCHC (mean/average of corpuscular hemoglobin content) were computed by employing the related equations.

MCH = Hb / RBC x 10 MCHC = Hb / PCV x 100 MCV = PCV / RBC x 10

ADC of nutrients

The experimental dietary ADC (apparent digestibility coefficient) was estimated by a conventional formula (NRC, 1993).

ADC (%) =  $100 - 100 \times \frac{\% \text{ marker in diet } \times \% \text{ nutrients in feces}}{\% \text{ marker in faeces } \times \% \text{ nutrients in diet}}$ 

# Statistical data of analysis

A one-way analysis of variance (ANOVA) was performed on the data of growth parameters (SGR, FCR, WG), hematological indices (PLT, Hb, HCT) and nutrients digestibility (CF, CP and GE) with results declared important at P $\leq$ 0.05 (Snedecor and Cochran, 1991). The CoStat computer programs (PMB 320, Monterey, CA, Version 6303.93940 USA and IBM SPSS 20) were used to carry out the analysis.

#### RESULTS

#### Growth performance of fish fingerlings

Table II shows that there were variations in the growth parameters of *C. carpio* fingerlings fed on test diets based on BSM, which replaced SBM at 0%, 25%, 50%, 75% and 100%, in terms of WG(g), WG%, FCR, SGR and survival rate. Notably, initial weight was observed to be statistically similar for all five groups.

The present study established that BSM based diets significantly enhanced nutritional quality, as evidenced by a significant (P<0.05) rising in weight gain of *C. carpio* fingerlings at a 50% substitution level. At this level highest WG (18.52g), WG % (259.99%), FCR (1.30), SGR (1.42) and survival rate (97.92%) is observed while the second highest WG (17.40), WG % (244.05), FCR (1.35), SGR (1.37) and survival rate 97.78% is seen at 25 % replacement level.

Diet		IW(g)	FW(g)	WG(g)	Initial wt.*100	Wt. gain/70	FI	FCR	SGR	Survival %
1		7.13±0.44 <sup>a</sup>	$19.81 {\pm} 0.07^{b}$	12.68±0.76°	178.26±13.53b	$0.18 \pm 0.01^{bc}$	$0.27{\pm}0.02^{b}$	$1.49{\pm}0.14^{ab}$	1.14±0.05 <sup>b</sup>	95.69±3.73ª
2		7.14±0.39ª	$24.54{\pm}0.89^{a}$	$17.40{\pm}0.50^{ab}$	244.05±6.46ª	$0.25{\pm}0.01^{\text{a}}$	$0.34{\pm}0.01^{a}$	$1.35{\pm}0.08^{b}$	$1.37{\pm}0.02^{a}$	97.78±3.85ª
3		7.16±0.25ª	25.67±0.81ª	18.52±0.69ª	259.99±19.58ª	$0.26{\pm}0.01^{a}$	$0.34{\pm}0.02^{a}$	$1.30{\pm}0.08^{b}$	1.42±0.06ª	97.92±3.61ª
4		7.13±0.44ª	23.75±0.94ª	16.59±0.70ª	231.82±2.59ª	$0.24{\pm}0.04^{ab}$	$0.31{\pm}0.03^{ab}$	$1.40{\pm}0.14^{b}$	1.33±0.01ª	95.56±3.85ª
5		$7.12{\pm}0.38^{a}$	$18.51{\pm}0.87^{\text{b}}$	11.39±0.50°	$160.11 \pm 2.43^{b}$	0.16±0.01°	$0.28{\pm}0.01^{b}$	1.70±0.05ª	1.06±0.01ª	95.83±7.22ª
St. E		.08399	.77098	.75576	10.64164	.01111	.00902	.04426	.03842	1.05603
Р	С	1.000	.000***	.000***	.000***	.000***	.002**	.006**	.000***	.936
value	L	1.000	.065*	.016*	.038*	.082*	.696	.031*	.020*	.824
	Q	.913	.000***	.000***	.000***	.000***	.000***	.001**	.000***	.558

Table II. Growth performance of C. carpio fingerlings fed on BSM based diets.

Means within columns with various superscripts (a-c) differ considerably at p<0.05Data are three replicates' mean (± symbol shows Standard Deviations); IW, initial weight; WG, weight gain; FW, final weight; FCR, feed conversion ratio; SGR, specific growth rate; St. E, standard error; L-, Linear; Q, Quadratic; C, Combined.

Table III. Analysis of hematological parameters.

Diet	RBC,s	PLT	Hb	PCV (%)	MCHC (%)	MCH (pg.)	MCV(Fl)	Ht %
	(10 <sup>6</sup> mm <sup>-3</sup> )		(g/100ml)					
1	$1.83 \pm 0.23^{b}$	$60.50{\pm}0.78^{b}$	$6.60 \pm 0.41^{bc}$	25.53±0.99 <sup>ab</sup>	32.11±0.85 <sup>b</sup>	$34.54{\pm}0.90^{\text{d}}$	$110.39 \pm 0.84^{d}$	$20.37 \pm 0.73^{d}$
2	$2.38{\pm}0.27^{ab}$	$62.39{\pm}0.84^{b}$	$7.13{\pm}0.36^{ab}$	23.23±0.97 <sup>b</sup>	37.18±0.89ª	$30.46{\pm}0.86^{\circ}$	125.53±0.78°	$23.23{\pm}0.97^{\circ}$
3	2.83±0.32ª	68.31±0.91ª	8.12±0.54ª	27.86±0.81ª	30.54±0.71 <sup>bc</sup>	52.32±0.92°	$159.33{\pm}0.86^{\text{b}}$	30.36±0.84ª
4	$2.42{\pm}0.26^{ab}$	67.43±0.96ª	6.43±0.49 <sup>bc</sup>	23.60±0.72 <sup>b</sup>	29.81±0.97°	58.48±0.77ª	$161.28 \pm 0.89^{b}$	$26.41 \pm 0.77^{b}$
5	$1.78 \pm 0.26^{b}$	$61.47 \pm 0.84^{b}$	5.73±0.46°	20.50±0.95°	$26.48 \pm 0.83^{d}$	55.43±0.82 <sup>b</sup>	164.31±0.93ª	$21.67 \pm 0.96^{cd}$
St. error	.97933	5.87858	3.07011	.95349	.68439	.23498	.87869	.12040
P C	.003**	.000***	.001**	.000***	.000***	.000***	.000***	.000***
value L	.884	.001**	.014*	.000***	.000***	.000***	.000***	.004**
Q	.000***	.000***	.000***	.000***	.000***	.000***	.000***	.000***

Means within columns with various superscripts (a-e) differ considerably at p<0.05. Given data is in the form of three replicates showing mean ( $\pm$  shows Standard Deviations); RBC, red blood cells; PLT, platelets; Hb, hemoglobin; PCV, packed cell volume; MCHC, mean corpuscular hematocrit cells; MCH, mean corpuscular hematocrit; MCV, mean corpuscular volume; Ht, hematocrit; St. E, standard error; L-, Linear; Q, Quadratic; C, Combined.

The minimum WG (11.39g), WG % (160.95%) and minimum survival rate 95% was recorded for *C. carpio* fingerlings fed with a diet containing 100% BSM, Additionally, feed intake was significantly lower when the BSM replacement level was 75% or 100%. Furthermore, the FCR of *C. carpio* fingerlings was most suitable (1.30) with a 50% BSM replacement level diet, as compared to the other diets. The FCR values observed in the fingerlings fed at 100% BSM were the least suitable (1.70) among other groups.

Additionally, this research revealed that by increasing the replacement levels up to 50% growth parameters of *C. carpio* were positive but further increase in replacement started declining at higher levels of BSM. In quadratic and combined tests, the values of FW(g), WG(g), WG(%), FI and SGR were highly significant while in linear test values of FW(g), WG(g), WG(%), FI and SGR values are non-significant. The values of IW(g) and survival rate are highly non-significant in Linear, combined and quadratic test.

#### Effect of BSM hematological indices of fish

Table III showed that there were variations in the blood parameters of *C. carpio* fry fed on BSM based diets, which replaced SBM at 0%, 25%, 50%, 75% and 100%. The analysis of hematological indices of *C. carpio* fingerlings revealed variations between those fed with the experimental diets (BSM 25%-BSM 100%) and the control test diet (BSM 0%).

The maximum levels of RBCs  $(2.83 \times 10^6 \text{ mm}^{-3})$ , platelets  $(68.31 \times 10^6 \text{ mm}^{-3})$  and Hb (8.12g/100 ml) were seen in fingerlings fed BSM 50% diet, followed by

2.42×10 mm for RBCs, 67.43 for platelets, in fingerlings fed BSM 75% while the second highest level of Hb was in fingerlings fed test diet BSM 25%. Conversely, the lowest values of RBCs ( $1.78 \times 10$  mm) and Hb (5.73 g/100ml) were seen in BSM 100% and the lower platelets (60.50) were in fingerlings fed the test diet BSM 0%, with the second minimum values of RBCs ( $1.83 \times 10$ mm) and Hb (6.60g/100ml) found in fingerlings fed control diet and the second lowest platelets.

PCV, which determined the proportion of blood made up of cells, revealed a maximum value of 27.86% in fish fed BSM 50%, followed by 25.53% observed at 0% substitution of SBM. The lowest value of 20.50% was seen in fingerlings fed BSM 100% and the second lowest value of 23.23% was observed in BSM 25%, which had 25% replacement of SBM. The maximum MCHC of 37.18% was observed among the fish that were fed the BSM 25% diet, with 32.11% being seen at 0% replacement of SBM and the lowest value of 26.48% being seen at BSM 100%, with the second lowest being recorded at 75% substitution of BSM. Similarly, the maximum MCV was observed at 100% replacement of SBM with BSM, with a value of 164.31, followed by 161.28 at 75% substitution diet BSM 75%. Conversely, the lowest MCV value of 110.39 was recorded at controlled diet, with a second lowest value of 125.53 being seen at 25% replacement of SBM with BSM. The highest hematocrit (Ht) value of 30.36% was observed in BSM 50%, with 26.41% being observed in BSM 75%, which was 75% substitution of SBM with BSM, while the lowest Ht value of 20.37 was seen in the control diet BSM-0 and the second lowest at 23.23 was observed at BSM 25% Upon comparison of all factors of hematology indices, it was determined that C. carpio fingerlings had the best performance at 50% substitution of SBM with BSM (i.e.

Table IV. Analysis of nutrient digestibility parameter.

BSM 50%). However, performance was also improved at 25% and 75% substitution levels in comparison to the control group. In quadratic and combined tests, the values of RBCs, PLT, Ht and lymphocytes were highly significant while in linear test values of RBCs and Hb, are nonsignificant.

#### Effect of BSM on nutrient digestibility

The study of CF, CP and GE in the BSM diets (Table IV) and in the feces of *C. carpio* fingerlings showed that the CF and CP values of all experimental diets were significantly similar, despite small differences in values being noted. The amount of nutrients discovered to be transmitted via the feces was highest when fingerlings fed diet BSM 50%, while the amount of CP and CF found to be lowest in the feces of fingerlings fed the BSM 100% diet.

The fingerlings fed BSM 50% had the highest quantities of CP (17.24%) and fat (2.36%) in their feces, followed by those fed BSM 0% (16.13% and 2.31%, respectively). The fish who were fed test BSM (50%) had the lowest CP (10.06%) and CF (1.38%) values, whereas the fingerlings that were fed test BSM 75% had lower CP (11.07%) and CF (1.71%) values. The highest value of GE was identified in the feces fed BSM 0%.

This was followed by the fish fed BSM 100% at (1.99%) and the lowest value was discovered in the fish fed BSM 25%.

Conversely, the highest CP digestibility (70.24%) was noted in BSM 50% and the highest GE digestibility (74.46 %) was discovered in fish fed BSM 25%, with 67.51% CP recorded in fingerlings fed BSM 25% and 72.37% GE in fingerlings fed BSM 50%. In contrast, the lowest CP digestibility (49.20%) was noted in fish fed BSM 100%,

Diets			Nutrients in fee	es	Nutrients digestibility			
		СР	CF	GE	СР	CF	GE	
1		16.13±0.53ª	2.31±0.15ª	2.09±0.08ª	52.35±0.90 <sup>d</sup>	57.49±0.8 <sup>d</sup>	53.63±0.87°	
2		11.07±0.4°	$1.87{\pm}0.14^{b}$	1.15±0.1°	67.51±0.99 <sup>b</sup>	65.55±0.84°	74.46±0.97ª	
3		10.06±0.16°	1.38±0.08°	1.23±0.1°	70.24±0.82ª	74.54±0.77ª	72.37±0.96ª	
4		13.42±0.45 <sup>b</sup>	$1.71 \pm 0.15^{bc}$	1.59±0.12 <sup>b</sup>	60.35±0.87°	$68.38 \pm 0.85^{b}$	64.39±0.83 <sup>b</sup>	
5		17.24±0.70ª	2.36±0.14ª	1.99±0.13ª	49.20±0.46e	56.26±0.85 <sup>d</sup>	55.75±0.93°	
St. Error		0.75006	0.10306	0.07346	2.20028	1.83717	2.26103	
P value C		.000***	.000***	.000***	.000***	.006**	.000***	
L		.000***	.825	.243	.000***	.000***	.006**	
Q	)	.000***	.000***	.000***	.000***	.000***	.000***	

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 Means within columns with various superscripts (a-e) differ considerably at p<0.05. P, probability; St.E, standard error; L-, Linear; Q, Quadratic; C, Combined; CP, Crude protein; CF, crude fat; GE, gross energy.</td>
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followed by the second-lowest CP digestibility (52.35%) in fingerlings fed BSM 0% and the lowest. In terms of fat digestibility, fish fed BSM 50% had the highest value (74.54%), followed by fish fed BSM 75% with a value of 68.38%. Fingerlings fed experimental diet BSM 100% had the lowest value (56.26%), with fingerlings fed BSM 0% having lower value (57.49%).

According to our findings, the highest level of gross energy was found in the diet with a 25% replacement of SBM (BSM 25%), but the highest level of protein and fat digestibility was recorded in *C. carpio* fingerlings at 50% replacement of SBM with BSM. On the other hand, the diet with 100% replacement of SBM had the lowest levels of proteins and fat and the diet with no replacement of SBM had the lowest gross energy (BSM 0%). In quadratic, linear and combined tests the values of CP, CF and GE were highly significant in nutrient digestibility while in linear tests values of CF and GE in nutrients in feces are non-significant and in quadratic and combined tests are highly significant.

# DISCUSSION

### Effect of BSM on fish growth

The outcomes of the current study showed that dietary BSM significantly impacts *C. carpio* capacity for growth. Fish fed diets containing 50% replacement showed highest SGR, weight gain, weight gain% and survival rate.

Corresponding to present study, Latif et al. (2021) and Yousefi et al. (2021) studied the effects of dietary N. sativa intake on L. rohita and C. carpio at different inclusion levels and they found similar results. In harmony with current study Ali et al. (2020) evaluated the exchanging fish meal in L. rohita and reached a finding that raising the degree of BSM inclusion in the diet enhanced all growth indices (SGR, WG and survival). Like this work, Al-Shawy and Zaidy (2009) investigated the diet containing 10.0% BSM exhibited the highest FCR (2.54), food conversion efficiency (39.66%), protein efficiency ratio (1.229%) and apparent digestibility (78.3%) among the diet regimens. Khattab (2001) suggested an analogous outcome, showing that the best rates of survival were noted at the 70% inclusion stage, although the greatest average weight increase was observed between 10% and 50%. Similar results were reported by Al-Dubakel et al. (2012) when they investigated the effects of dietary BSM integration at 1 and 3% in C. carpio. They observed that the 1% BSM indicated the highest growth rate and a more ideal FCR value.

In contrast to this work, Sayed *et al.* (2009) observed that there were no significant disparities in final body weight, SGR and BW gain (g/fish) values in comparison

with the control diet, as similarly elucidated by Soliman *et al.* (2000). Nevertheless, increasing the BSM inclusion level led to negative effects on these factors. In dissimilarity to this study Shalaby and Sharaf (2008) found that there were no significant differences in fish growth at low inclusion levels of BSM, yet the higher levels of BSM significantly decreased the fish performance. On the other hand, Aydin (2021) had totally opposite observations to the present study as he found negatively correlated with dietary BSM levels by increasing the inclusion level of BSM. However, FCR values and survival rate did not significantly differ between treatments.

#### Effect of BSM on hematological

This study determined that the replacement of SBM with BSM in the diets of C. carpio fingerlings had a positive effect on their hematological parameters, with the highest values of RBCs, PLT and Hb being recorded from a 50% replacement of SBM with BSM. Likewise, Azarin et al. (2015) experienced an increase in the number of erythrocytes compared to the control fish, similarly, Fadeifard et al. (2018) observed the highest erythrocyte level of 6.29 cells/mm<sup>3</sup>, being recorded when 9% N. sativa seeds were utilized. Similarly, Sa'adah et al. (2015), showed an accelerated increase in Ht levels 19% to 32% when fed with BSM diet. Slightly similar results found by Abouelezz et al. (2020) that increased hemoglobin levels in both BSM 0% and BSM 25% as compared to the control group, ht (PCV %) in fish group fed 2% N. sativa (BSM 25%) when compared to the control group. A similar trend was established by Bektaş et al. (2018) that all parameters statistically significant differences between MCH and MPV values. Furthermore, John et al. (2007) found significant Ht values in O. niloticus fed with 3% N. sativa. Hussein et al. (2020) reported that an increase in Hb levels to 8.81±0.37g/dl and 8.88±0.47g/dl, respectively, in comparison to the control group 7.63±0.43 g/dl. On the other hand, Bektas et al. (2018) found that the number of erythrocytes in O. mykiss did not increase after receiving 0.1-20g/kg of N. sativa seed powder in the meal for 60 days.

## Effect of BSM on nutrients digestibility

In the current study, gross energy is optimal at 25% replacement level (74.97), while nutritional digestibility (protein 70.24, lipids 74.77) is superior at 50% replacement level of SBM with BSM. This study suggests that a diet based on soybeans is not as effective as raising the level of BSM in a *C. carpio* diet as a cheap alternative protein source.

Corresponding to this study, Khattab (2001) analyzed that apparent digestibility was improved by increasing the

replacement of BSM, resulting in a rise from 85.65% to 92.25%. Furthermore, apparent carbohydrate digestibility and apparent gross energy digestibility were observed to be higher in the fish groups fed BSM diets than that of the control test group. Similar to this study, OZ *et al.* (2018) discovered that there was a boost in the protein efficiency scores from 2.77 to 3.07 comparable to the BSM-0 group, as well as a rise in the total amount of fats in the fish. Similar to this study, Wafaa *et al.* (2014) showed that their crude protein content significantly increased when compared to individuals who were fed the control meal. Similar conclusions about how BSM's enhanced protein contents improved its nutritional value were made by Latif *et al.* (2021).

Aydin (2021) in contrast with the current study, discovered that supplementing BSM protein had no appreciable impact on the levels of total protein. However, it proved that the amount of fat in the carp muscle had dropped from 1.39 to 0.56 in contrast to the control group. Ahmed *et al.* (2018) made very different findings, stating that BSM could be used in Nile tilapia meals up to 50% levels (replacing partially SBM) without impacting the amount of protein in the fish's entire body; however, substituting above a 50% level may reduce digestibility and subsequently affect growth performance.

# CONCLUSION

From this study, it is concluded that replacing SBM with BSM resulted in improved growth performance, haematological indices and nutrients digestibility of *C. carpio* fingerlings. So partially substitution of soybean meal from black seed meal as a traditional source of protein in the diet of common carp fish is certainly beneficial and cost effective.

# DECLARATIONS

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# Statement of conflict of interest

The authors have declared no conflict of interest.

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