

SYNERGISTIC EFFECTS OF DIETARY GLUCOSAMINE AND PLANT/ANIMAL PROTEINS ON THE GROWTH PERFORMANCE OF ASIAN CATFISH (*Clarias Batrachus*) JUVENILES

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ABSTRACT: A 84-days feeding trials was conducted to evaluate the use of animal and plant protein, in combination with glucosamine source for Asian Catfish, *Clarias batrachus* (av. wt. 0.22±0.01 to 0.24±0.07g). Six (31.18 to 43.51% crude protein, 369 to 399 kcal/100g, and crude lipid 0.0 to 6.69%) practical feeds were formulated. The animal and plant protein component of the feeds was progressively added with glucosamine 0.0, 0.5, 5.0 and 10.0% with fish meal, silkworm pupae, soybean meal and casein (F-1, PAG 0:100:0.5; F-2, PAG 0:100:5.0; F-3, PAG 0:100:10.0; F-4, PAG 100:0:0.5; F-5, PAG 100:0:5.0; F-6, PAG 100:0:10.0; The experimental feeds were fed to triplicate groups of fingerlings at 10% body weight per day and results were compared with control feed. Growth performance and feed utilization efficiency of catfish, fed with animal proteins are better than those of plant protein. The best growth among the animal protein group (F-1 to F-3) was recorded in F-2 followed by F-3 and F1 containing glucosamine @5.0, 10.0 and 0.05%. Amongst the plant protein fed fishes showed best in F6 followed by F5 and F4. The survival was improved in glucosamine supplemented feeds ranging from 49±3.2 to 85±1.7 whereas the control showed 41±1.8%. Results indicate that animal protein rich feeds were much acceptable than alternative plant protein sources for the Asian catfish, *Clarias batrachus* and the potential for replacing animal protein with soybean meal in the feeds of fish need more evaluation along with synergistic effects of growth promoter like glucosamine.

Key words: *Clarias batrachus*, glucosamine, animal protein, plant protein, growth

INTRODUCTION

Fishmeal as raw material is the first choice in aquaculture production due to high quality protein with balanced amino acid profile (Gatlin III et al., 2007). Since last twenty years the production of fishmeal is relatively stable and the increasing requirement could not be matched in present scenario due to increased aquaculture requirement (Goitortua-bores et al., 2006). Moreover the cost of fishmeal is increasing day by day therefore there is an urgent need to evaluate the other ingredients as well find alternative protein source to make up for the shortage of fish meal and fulfill the requirement and secure the supply for commercial feed (Hardy, 2006). In this context soybean meal (SBM) regarded as an economical and nutritionally rich food ingredient which contain higher protein content in comparison to other plant ingredient (Gatlin III et al., 2007). The nutritional evaluation of Soybean meal to replace fishmeal has been a long standing priority in fish nutritional research (Hardy, 1999). Due to evermore research data a considerable success has achieved in supplement of FM with SBM plant proteins in aquatic animals (Dersjant-Li, 2002; Kaushik et al., 1995). However, at higher rate of replacement of the fishmeal with SBM encouraged growth retardation may be due to imbalance nutrition in carnivorous fishes (Kaushik et al., 1995; Toma's et al., 2005; Wang et al., 2006; Martinez-Llorens et al., 2009; Ye et al., 2011) and/or higher ammonia excretion (Ballestrazzi et al., 1994; Tantikitti et al., 2005). The reduced growth may be due to anti-nutritional factors (Francis et al., 2001; Leenhonwers et al., 2006; Ye et al., 2011). The histological changes in intestine can also

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reduce growth performance on feeding plant proteins (Boonyaratpalin et al., 1998; Krogdahl et al., 2003; Heikkinen et al., 2006; Wang et al., 2006).

Air-breeding Catfish, *Clarias batrachus* (Family: Clariidae), locally known as Magur, is a fish of great demand and attracts the attention of farmers for its high market value. Feed management determines the viability of aquaculture as it accounts for at least 40-60% of the cost of fish production (Jamu and Ayinla, 2003). Reducing the feeding costs could be key factor for successful development of aquaculture. Protein is the most expensive component in fish feeds hence it is known to require in relatively large amount by several fishes (Delong et al., 1958; Ogino and Saito, 1970; Nose and Arai, 1972; Anderson et al., 1981; Mazid et al., 1979; Wee and Tacon, 1982), the exact level of its requirement for formulation of well-balanced feed and also the most important factor affecting growth performances of fish and feed cost (Lovell, 1989). So it is important to accurately determine the protein requirements for each species and size of cultured fish. Level of dietary protein is of fundamental important because it significantly influences growth, survival and yield of fish as well as economics of a farming industry by determining the feed cost which is typically the largest operational cost. Glucosamine a amino sugar and a prominent precursor in the biochemical synthesis of glycosylated proteins and lipids synthesizes chitin, is one of the most abundant monosaccharide (Horton and Wander 1980, Roseman 2001, Muzzarelli 1977) which composes the exoskeletons of crustaceans and other arthropods. It has been well established that animal protein performs better than plant protein in the growth and nutritive value of cultivable fish (Rao and Kumar 2006). Silkworm pupa is one of the unconventional top class animal proteins (65-67%). Recycling of these wastes into an acceptable source of animal protein in the feed of fish is a big challenge in the pursuit of sustained procedure of inexpensive catfish, *Clarias batrachus* feed. Silkworm pupae (*Bombyx mori*) is a low cost animal protein source, rich in both protein and lipid (Bhuiyan et al., 1989). This study was taken up as huge mortality is recorded at fry stage of this fish in natural condition. Therefore, this experiment was carried out to study the synergistic effects of dietary glucosamine in combinations with Plant/animal proteins on the survival and growth performance of *Clarias batrachus* fry.

MATERIALS AND METHODS

Fish and feeding trial

Newly hatched larvae of catfish, *Clarias batrachus* obtained from a single batch of hatchery bred spawned broodstock were used in the experiment after acclimation for one week. In the wet laboratory the experimental fish, *Clarias batrachus* fry (av. wt. 0.22 ± 0.01 to 0.24 ± 0.07 g) were subsequently segregated and stocked in separate specially designed plastic pool (capacity 300 l, containing 50 l of tap water with continuous aeration), in a groups of 100 fry in each pool. The experiment consisted of two replicates for each feed and continued for 84 days. The experimental feeds were hand-fed @ 10% of the total body weight. Each scheduled daily ration per batch of fish was divided into two equal proportions and distributed to the fish at 11:00 hr and 17:00 hr respectively. Initial and subsequent fortnightly weight gains (g) were recorded on electronic balance (make: Sartorius). At the end of the experiment 6-8 fish from each treatment were sacrificed and analyzed for proximate composition of the muscles. The water quality parameters were recorded for water temp, pH, dissolved oxygen and total alkalinity.

Analytical methods and analysis of data

Proximate compositions of feeds and fish carcasses were analyzed following AOAC (1990) methods. All samples were analysed in triplicate. Dry matter was estimated after drying in oven at 105°C for 24 hours; crude protein (N x 6.25) by the Kjeldahl method after acid digestion; Crude lipid by di-ethyl ether extraction method using Soxhlet apparatus. The performance of the feeds, in terms of the weight gain (%), Specific growth rate (SGR), feed conversion ratio (FCR), Protein efficiency ratio (PER). The growth in length and weight and the survival data were analysed using One-way ANOVA. Duncan's multiple Range test was used to determine which treatment means differed significantly (P<0.05) using SPSS version 16.0.

Weight Gain (%) = $\frac{\text{(Final body weight)} - \text{(Initial body weight)}}{\text{(Initial body weight)}} \times 100$

Specific Growth Rate (SGR; % day⁻¹) = $\frac{\text{(Final body weight)} - \text{(Initial body weight)}}{\text{(experimental days)}} \times 100$

Survival (%) = $100 \times \frac{\text{No. of total fish} - \text{No. of dead fish}}{\text{Number of total fish}}$

Biomass = Final average weight x Total no. of fish

Experimental feeds and feed preparation

Six feeds were prepared by using plant and animal protein in combination with glucosamine source for Asian catfish, *Clarias batrachus*. Ingredients and proximate composition of the experimental feeds are given in Table 1. The natural live feed serves as control. In the experiment six (31.18 to 43.51 % crude protein, 369 to 399 kcal/100g, and crude lipid 0.0 to 6.69%) practical feeds were formulated and their composition is given in Table 2. The animal and plant protein component of the feeds was progressively added with glucosamine 0.0, 0.5, 5.0 and 10.0 % with basic ingredients like fish meal, silkworm pupae, soybean meal and casein (F-1, PAG 0:100:0.5; F-2, PAG 0:100:5.0; F-3, PAG 0:100:10.0; F-4, PAG 100:0:0.5; F-5, PAG 100:0:5.0; F-6, PAG 100:0:10.0. Fishmeal was freshly prepared from in lab from dried trash fishes mainly *Mystus vittatus*, *Puntius sophore*, etc. Live silkworm pupae were procured from Department of Applied Animal Science, Babasaheb Bhimrao Ambedkar University, Raebareilly Road, Lucknow, cultured upto VIth Instar larvae and then de-oiled in the lab by di-ethyl-ether (Merck).



The de-oiled pupae was dried in oven at 60°C for an hour and powdered and used for feed preparation. The feeds were prepared by thoroughly mixing of the dry ingredients in a mixer and water was added to make stiff dough. Each feed was cooked in a pressure cooker for 15 minutes for the proper gelatinization of the ingredients. Finally cooked moist feeds were stored in plastic zipped polybags in a freezer (-20°C) until used.

Table - 1 Ingredients composition (w/w) of feeds for *Clarias batrachus* Fry

Feeds Ingredients	F1	F2	F3	F4	F5	F6	Control
	PAG 0:100:0.5	PAG 0:100:5.0	PAG 0:100:10.0	PAG 100:0:0.5	PAG 100:0:5.0	PAG 100:0:10.0	NATFO
Soybean meal ¹	0.0	0.0	0.0	60.8	60.8	60.8	-
Silkworm Pupae	20.3	20.3	20.3	0.0	0.0	0.0	-
Fish Meal	20.3	20.3	20.3	0.0	0.0	0.0	-
Casein ²	20.2	20.2	20.2	0.0	0.0	0.0	-
Glucosamine (Chitosamine -HCl) ³	0.5	5.0	10.0	0.5	5.0	10.0	-
Starch ⁴	32.0	27.5	22.5	32.0	27.5	22.5	-
CMC ⁵	2.2	2.2	2.2	2.2	2.2	2.2	-
Papain ⁶	2.0	2.0	2.0	2.0	2.0	2.0	-
VM + MM ⁷	2.5	2.5	2.5	2.5	2.5	2.5	-
Natural -Live food	-	-	-	-	-	-	100.0
Total	100	100	100	100	100	100	100

P:A:G= Plant Protein : Animal protein : Glucosamine; CMC=Carboxy-methyl-cellulose. ¹HiMedia, Mumbai Lot No: 0000013648; ² HiMedia, Mumbai Lot No: 0000016171; ³HiMedia, Mumbai, Lot No: 0000028805; ⁴HiMedia, Mumbai, Lot No: 0000028340; ⁵HiMedia, Mumbai, Lot No. 0000014218; ⁶HiMedia, Mumbai, Lot No. 0000003862; Each kg of Vitamin and mineral mixture named 'Agrimin Forte' contains Vit. A 700000 IU, Vit. D₃ 70000 IU, Vit. E 250mg, Nicotinamide 1000mg, Co 150mg, Cu 1200mg, I 325mg, Fe 1500mg, Mg 6000mg, Mn 1500mg, K 100mg, Se 10mg, Na 5.9mg, S 0.72%, Zn 9600mg, Ca 25.5%, P 12.75% Manufacturer Brindavan Phosphates Pvt. Ltd, 48N, Doddaballpur Ind. Area, Doddaballapur - 561 203, India Batch No. BFA-61.

Table 2 - Calculated values of Protein, carbohydrate, fat and energy composition of feeds

Total Protein	43.52	43.52	43.52	31.18	31.18	31.18
Carbohydrate	34.70	34.70	34.70	55.98	55.98	55.98
Total Fat	6.70	6.70	6.70	0.00	0.00	0.00
GE/ kg	3990.48	3990.48	3990.48	3698.46	3698.46	3698.46
KJ.g ⁻¹	16.76	16.76	16.76	15.53	15.53	15.53

RESULTS AND DISCUSSION

Various water quality parameters: water temperature, pH and dissolved oxygen (DO), total alkalinity were observed and found to be least affected by different treatment feeds. The values of all the parameters of ambient water, i.e. temperature, pH, DO and alkalinity were almost similar for all the feeding treatments during the experimental period and were well within the optimal range. The water quality recorded for water temp, pH, dissolved oxygen and total alkalinity as 20 - 24 °C, 6.8 - 7.5, 6.9 - 7.4 ppm and 130 - 138 ppm, respectively.

Table 3 - Survival Percentage of *Clarias batrachus* fry reared for 12 weeks

Feed	Stocking Nos. (N=100 X 2 replicates)	4 th Week	8 th Week	12 th Week
F-1	200	87 ± 2.5 ^a	84 ± 2.4 ^a	78 ± 1.9 ^b
F-2	200	90 ± 2.8 ^a	84 ± 3.3 ^b	80 ± 1.6 ^c
F-3	200	92 ± 1.4 ^a	90 ± 2.2 ^a	85 ± 1.7 ^b
F-4	200	65 ± 3.1 ^a	52 ± 2.2 ^b	49 ± 3.2 ^b
F-5	200	68 ± 2.5 ^a	62 ± 3.7 ^b	59 ± 3.2 ^c
F-6	200	69 ± 1.4 ^a	66 ± 1.7 ^a	63 ± 5.3 ^b
F-7 (control)	200	52 ± 1.9 ^a	46 ± 2.4 ^b	41 ± 1.8 ^c

Same alphabet in superscript in a row represents no significant difference in weight gain * = p < 0.05. The results are of duplicate sets of feeding trial.

The survival and average fish weight gain shown graphically in Tables 3 and 4 respectively. The best growth was recorded in fish fed F3 among the animal protein group feeding regime (F1 to F3) as 85±1.7% in F3 followed by F2, 80±1.7% and F1, 78±1.9%. The results are showing dose-dependent effects of glucosamine on the survival. The plant protein based feeds with graded level of glucosamine @ 0.5, 5.0 and 10.0 resulted in an overall poor survival for the *Clarias batrachus* fry and recorded as 49±3.2%, 59±3.2% and 63±5.3% in F4, F5 and F6 respectively. The control feed showing 41±1.8% survival. The growth of the fry was recorded better in animal



protein fed fishes but there is no sign of dose-dependent effects on growth performance. The best growth recorded in F2 which contains 5.0% glucosamine with 100% animal protein. The plant protein diets showed poor growth in comparison to animal protein diets. The control showed poor growth after 12th week. The results are shown in Table 4. The results of FCR, SGR, PER, Feed intake and Protein intake are shown in Table 5. The FCR, SGR, PER, feed intake, protein intake ranged between 1.6 ± 0.2 to 2.4 ± 0.2 ; 36.64 to 170.5%; 0.94 ± 0.1 to 1.75 ± 0.03 ; 129 ± 11.0 to 600.0 ± 31 ; 87 ± 6 to 187.6 ± 11 . The synergistic growth on supplementing protein and glucosamine showed significant variation ($p < 0.05$) in case of weight gain, FCR, SGR, PER, However there is no change ($p > 0.05$) in feed intake and protein intake in all the treatments.

Table 4 - Growth of *Clarias batrachus* fry reared for 12 weeks

Feeds	In	4 th week	8 th week	12 th week
F1	0.24 ± 0.01^a	0.28 ± 0.04^b	0.29 ± 0.04^b	0.38 ± 0.02^c
F2	0.23 ± 0.02^a	0.21 ± 0.03^a	0.37 ± 0.02^b	0.56 ± 0.04^c
F3	0.23 ± 0.01^a	0.31 ± 0.02^b	0.37 ± 0.01^c	0.46 ± 0.02^d
F4	0.22 ± 0.02^a	0.29 ± 0.01^b	0.33 ± 0.02^c	0.29 ± 0.01^b
F5	0.28 ± 0.03^a	0.20 ± 0.04^b	0.26 ± 0.02^a	0.31 ± 0.03^c
F6	0.24 ± 0.02^a	0.26 ± 0.02^b	0.30 ± 0.01^c	0.36 ± 0.01^d
F7 (control)	0.22 ± 0.01^a	0.29 ± 0.02^b	0.33 ± 0.02^c	0.30 ± 0.01^b

Same alphabet in superscript in a row represents no significant difference in weight gain; * = $p < 0.05$. The results are of duplicate sets (n = 2) of feeding trial.

In the present study, the experimental feeds were formulations with different protein are based on previous reports (Kikuchi, 1999; Kim et al., 2002, 2006; Cho et al., 2006 and Ye et al., 2011). In the study, the differences observed in the performance of the dietary animal and plant protein feeds in combination with graded level of glucosamine (0.5, 5.0, 10.0). The experimental feeds F1, F2 and F3 with animal protein along with glucosamine (0.5, 5.0, 10.0), performed better than the plant proteins based feeds F4, F5 and F6. Dietary proteins dietary protein plays a dominant role in fish growth (Cowey et al., 1972; Satia, 1974; Cho et al., 1976). On the basis of average specific growth rate and % live weight gain, an improvement in growth response was noticed with increase in dietary protein level up to maximum of 35% animal protein (casein) content and thereafter a decrease with further increase in dietary protein concentration (Das and Ray, 1991). The present study showed that different protein types (plant or animal) significantly affected the growth and feed utilization of Asian catfish, *Clarias batrachus*. The negative effects of weight gain, FCR, PER in response to dietary plant protein suggesting that dietary plant protein type is poorly suitable than animal protein. Similar reports are made by Ye et al. (2011) in Japanese Flounder using soybean meal more than 16% and Kikuchi (1999), who found that 43% of fishmeal protein could be replaced by soybean meal (25%) in combination with bloodmeal (10%) or corn gluten meal (10%) and blue murels meat (5%). The data in present study on *Clarias batrachus* indicated that tolerance to animal protein substitution by plant protein in combination with glucosamine was somewhat low. According to Rao and Kumar (2006), experiment conducted to know the effect of animal protein incorporated formulated feeds on the growth and nutritive value of Rohu fingerlings, the test feeds containing 35% dietary protein level, showed better performance in growth and fertilization than the control feed having only plant protein and also the test feeds having higher protein levels. This infers that the plant protein (GOC) can be replaced by squillameal (an animal protein), which is very much similar to our results. Fish meal has superior nutritive values over other animal proteins (Seenappa and Devraj, 1995) and plant proteins (Eyo, 1991), because of its well balanced amino acid compositions and their bioavailability (Moon and Gatlin, 1994), which influenced the performance of animal (Gaylord and Gatlin, 1996). 0.5 glucosamine with animal protein gives better results than 5.0 or 10.0 % glucosamine with animal protein which shows that 0.5% levels of glucosamine good for the health of fish. Similar results have been reported by Mollah and Alam (1990), who obtained value of 15% carbohydrate (glucosamine 5.0, 10.0) in the feed showed retardation of growth. Further, the foregoing results agree and extend the findings of Chakraborty et al. (1973) by showing that silkworm pupae (animal protein), groundnut and wheat bran was better utilized by fry *Labeo rohita* and *Cirrhinus mrigala* than that of mustard oilcake and rice bran. Prawn shell waste protein is rich in essential amino acids (Forster 1975; Penafiorida, 1989). Dietary glucosamine was found to be a growth promoting factor in shrimp (Kitabayashi et al., 1971) and the shell (chitin) in shrimp waste growth promoting agents for the prawn *Penaeus indicus* (Vaitheswaran et al., 1986). The effect of dietary chitin on the growth and survival of juvenile *P. monodon* was studied by various workers (Lan and Pan 1993; Sudaryono et al., 1996). In the present experiment, conducted to know the effect of animal and/or plant protein incorporated with glucosamine (at graded levels of 0.5, 5.0, 10.0), the test feed F1 (100% animal protein with 0.5 % glucosamine) showed better performance in survival and growth than the other feeds containing plant proteins. Growth performance and feed utilization efficiency of this catfish, fed feeds with animal protein are better than those of plant protein. Results indicate that animal protein rich feeds were much acceptable than alternative plant protein sources for the Asian catfish, *Clarias batrachus* and the potential for replacing animal protein with soybean meal in the feeds of fish need more evaluation along with synergistic effects of growth promoter like glucosamine. Results indicate that animal protein rich feeds with glucosamine were much acceptable than natural feeds for Asian catfish, *Clarias batrachus*.



Table 5 - Growth performance, nutrient utilization in *Clarias batrachus* fry reared for 12 weeks

Items Feed	Dietary Glucosamine	Animal : Plant Protein Ratio	In wt (g)	4 th week wt. gain % (up to 4 wk)	8 th week wt. gain % (up to 8 wk)	12 th week wt. gain % (up to 12 wk)	FCR%	SGR%	PER %	Feed Intake (mg)	Protein Intake (mg)
F1	0.5	100:0	0.24±0.07 ^a	16.6±1.2 ^a	20.8±1.3 ^a	58.3± 4.7 ^a	2.4±2.0 ^a	69.11	1.31±0.04 ^a	342±20 ^a	106±4 ^a
F2	5.0	100:0	0.23±0.02 ^a	-8.7±0.9 ^b	60.8±4.5 ^b	143.5± 8.3 ^b	1.9±0.1 ^b	170.5	1.75±0.03 ^b	600±31 ^b	187.6±11 ^b
F3	10.0	100:0	0.23±0.01 ^a	34.78±3.1 ^c	60.6±3.1 ^b	100.1±7.6 ^c	1.9±0.2 ^b	118.8	1.74±0.05 ^b	420±18 ^c	130.9±8 ^c
F4	0.5	0:100	0.22±0.02 ^a	31.8± 2.1 ^c	50.0±4.1 ^c	31.0±2.7 ^d	2.4±0.03 ^a	36.64	0.94±0.01 ^d	170±10 ^d	73.9±5 ^d
F5	5.0	0:100	0.22±0.03 ^a	-9.1±1.3 ^b	18.1±2.0 ^a	40.8±3.2 ^e	2.1±0.1 ^c	48.30	1.03±0.04 ^d	201±20 ^e	87.0±6 ^e
F6	10.0	0:100	0.24±0.02 ^a	8.33±0.41 ^d	25.0±3.3 ^d	50.1±4.8 ^f	2.0±0.3 ^c	59.6	1.20±0.02 ^a	230±28 ^f	100.0±8 ^a
F7	-	-	0.22±0.01 ^a	31.7±1.8 ^c	50.6±2.7 ^c	36.4±2.8 ^d	1.6±0.2 ^d	43.07	-	129±11 ^g	-
	0.5	-	0.23±0.01 ^a	24.2±1.8 ^d	35.4±2.8 ^e		2.4±0.1 ^a	52.82	1.13±0.07 ^d	254±22.7 ^d	90.4±6.1 ^e
	5.0	-	0.22±0.03 ^a	-8.9±0.8 ^b	39.5±3.7 ^e		2.0±0.2 ^c	52.83	1.41±0.10 ^a	401±30.2 ^d	137.3±8.7 ^c
	10.0	-	0.24±0.02 ^a	21.5±2.5 ^d	42.8±2.9 ^f	44.6±3.6 ^e	1.95±0.3 ^c	52.80	1.48±0.11 ^e	325±21.6 ^d	115.4±8.2 ^c
Animal Protein 100 %	-	100:0	0.23±0.04 ^a	14.3±1.1 ^a	47.4±5.1 ^c	92.1±6.3 ^c	2.06±0.2 ^c	109.36	1.61±0.14 ^d	454±28.4 ^d	141.7±10.1 ^c
Plant protein %100	-	0:100	0.23 ± 0.03 ^a	10.3±0.9 ^d	31.0±2.2 ^g	75.2±4.1 ^g	2.2±0.1 ^d	89.25	1.06±0.08 ^d	203±13.1 ^e	86.9±6.7 ^e

Mean Values in same column with different superscript letters are significantly different (P <0.05). Values are mean ± SE of duplicate determinations (n=2); In = Initial weight of fish before feeding; SGR=Specific Growth Ratio; FCR = Feed Conversion Ratio; PER = Protein Efficiency Ratio. FI = Feed Intake; PI = Protein Intake

Table 6 - Whole body proximate composition (g.kg⁻¹ DM) of *Clarias batrachus* fry fed feeds containing different proteins (animal or plant protein origin) for 12th week

Parameters (g.kg ⁻¹)	In Wt	F1	F2	F3	F4	F5	F6	F7(Control)
Moisture	72.2 ± 2.1 ^a	70.3 ± 2.0 ^b	70.8 ± 2.8 ^b	71.2 ± 2.2 ^b	70.8 ± 1.8 ^b	70.7 ± 2.7 ^b	73.1 ± 2.5 ^a	70.3 ± 1.9 ^b
Crude Fat	6.8 ± 0.3 ^a	8.7 ± 0.4 ^b	8.5 ± 0.3 ^b	8.3 ± 0.2 ^b	7.0 ± 0.5 ^a	6.9 ± 0.3 ^a	6.5 ± 0.2 ^a	7.2 ± 0.3 ^c
Crude Protein	56.3 ± 2.1 ^a	57.6 ± 2.9 ^b	59.7 ± 1.8 ^c	58.1 ± 2.6 ^b	56.2 ± 1.8 ^a	56.9 ± 2.9 ^a	56.0 ± 3.2 ^a	57.5 ± 2.4 ^b
Dry Matter	24.2 ± 1.5 ^a	26.1 ± 1.3 ^b	25.6 ± 1.2 ^b	25.8 ± 1.2 ^b	28.1 ± 1.7 ^c	27.5 ± 1.6 ^c	23.2 ± 1.1 ^a	26.9 ± 1.4 ^b

Mean Values in same column with different superscript letters are significantly different (P <0.05)

The results suggest that the feeding habit of the fish with small crustaceans is met by the addition of glucosamine therefore, it is confirmed that glucosamine has impact on growth promotion in this fish. And the potential for replacing animal protein with soybean meal in the feeds of fish need more evaluation along with synergistic approach of incorporating glucosamine. Inclusion of plant protein blend affected growth performance and reduced growth performances and even not compensated by increased feed intake.

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