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# 1. Effect of Probiotic Diet Supplement on Growth, Digestibility and Nutrient Retention in Channa Gachua Fingerlings

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#### Abstract

Five experimental diets with containing varying concentration of lactobacillus spp (probiotics) (0.25,0.5,0.75 and 1.0%) were formulated using processed full fat soybean as the protein source.

Channa gachua fingerling were collected from local fish supplier and kept in glass aquaria (60x30x30cm) with aeration facilities in the laboratory where the temperature was kept as 25+1.c, lighting schedule of LD 12:12.

(Mean body weigh 3.18 ± 4.1 g) were randomly distributed among the aquaria, with 20 fish per aquarium. Each diet treatment was tested in replicate of four (Four aquaria per diet). All fish were fed twice daily, 08.00, at 14:00 h. The feeding rate was at 5% body weight day-1 for the whole rearing period of 70 days, and the amount of feed was adjusted every tenth day following a bulk weighing of each group of fish. The fish were exposed to their respective diet for 4h during each ration, Fish growth, digestibility and nutrient retention result shows that, survival was not affected by the inclusion levels of probiotic. Growth performance [(in terms of live weight gain), growth percent gain in BW and final length). SGR and nutrient retention (PER, GPR, GER and APD)] increased when dietary probiotic level were increased from 0.25g to 0.75g 100g-1 of diet; further increase in dietary probiotic level (>0.75g 100g-1) resulted in a significant (P<0.05) growth depression and nutrient depletion. Apparent protein digestibility was significantly (P<0.05) higher in fish which were fed diets containing probiotic at 0.75g 100g-1 than in fish fed probiotics free diet (Control) or diets containing low or high levels of lactobacillus. FCR values were also significantly (P<0.05) lower in fish fed diet containing Lactobacillus at 0.75g 100g-1 than fish fed other dietary preparations including control diet.

Key words: Probiotic supplement, nutrient retention

#### Introduction

Vaccines are being developed and marketed and they generally cannot be used as a universal disease control measure in aquaculture. Juvenile fish are fully immunocompetent and do not always respond to vaccination. Vaccination by injection, sometimes are the only effective route of administration, is impractical when supplied to small fish or large number of fish. This situation is avoided by an alternative in the production system through the use of beneficial bacteria to fight against pathogenic bacteria i.e., through the use of probiotic which is an acceptable practice in aquaculture. The health of the fish thus can be improved by the elimination of pathogens or at least by minimizing their effects in aquaculture (Patra and Bandyopadhyay, 2003). Due to outbreak of disease in aquaculture industry in the last 20 years, use of antibiotics have lead to the development of drug-resistant strains resulting in reduction of natural defense mechanism in the aqua cultural animals. On the other hand, probiotics can provide better immune response, increase survival and promote growth and nutrient utilization. Its use can thus assure the nutritional security in the next millennium. Probiotics for aquaculture are generally selected by their ability to produce antimicrobial metabolites: however, attachment to intestinal mucus is important in order to remain within the gut of the host (Vine et al., 2004)

#### Materials and Methods

#### Diet preparation

Five experimental diets with containing varying concentrations of Lactobacillus spp (Probiotic) (0.25, 0.5, 0.75 and 1.0%) were formulated using processed full fat soybean as the protein source. The dietary ingredients and proximate composition of the formulated diets are given in table 1

Table 1. Ingredient content (%) and proximate analysis (% dry weight basis) of five experimental diets with different levels of probiotic (g 100g<sup>-1</sup> of diet)

			Diets		
Ingredient	D0	D1	D2	D3	D4
Groundnut oil	65.00	65.00	65.00	65.00	65.00
cake					
Rice bran	3.20	2.95	2.70	2.45	2.20
Wheat flour	3.20	3.20	3.20	3.20	3.20
Processed	26.60	26,60	26.60	26.60	26.60
soybean					

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Chromic	oxide	1.00	1.00	1.00	1.00	1.00
(Cr <sub>2</sub> O <sub>3</sub> )						
Calcium		0.50	0.50	0.50	0.50	0.50
Phosphorus		0.50	0.50	0.50	0.50	0.50
Probiotics	3	_	0.25	0.50	0.75	1.00

Live weight gain (in grams), growth percentage gain, specific growth rate [% body weight (BW) per day], feed conversion ratio (FCR), gross protein retention (GPR) and gross energy retention (GER) were calculated using standard methods (Steffens, 1989). Apparent protein digestibility (APD) of the diets was calculated according to Cho et al. (1982) as follows

	% Cr <sub>2</sub> O <sub>3</sub> in diet	% nutrient in faeces
APD = 100-100 ×		*
	% Cr <sub>2</sub> O <sub>3</sub> in faeces	% nutrient in diet

Gross energy content of the diets and fish were calculated using the average caloric conversion factors of 0.3954, 0.1715 and 0.2364 kJ g<sup>-1</sup> for lipid, carbohydrate and protein, respectively (Henken et al., 1986).

#### **Statistics Analysis**

ANOVA followed by Duncan's multiple range test (Dunan, 1955) and student 't' test (Snedecor and Cochran, 1982) were applied to find out the significant differences between different treatments. Data were further subjected to orthogonal polynomials for trend analysis.

#### Results

Survival was not affected by the inclusion levels of probiotic. Growth performance [(in terms of live weight gain (Fig.7), growth percent gain in BW and final length), SGR (Fig.8) and nutrient retention (PER, GPR, GER and APD)] increased when dietary probiotic level were increased from 0.25g to 0.75g 100 g<sup>-1</sup> of diet; further increase in dietary probiotic level (>0.75g 100g<sup>-1</sup>) resulted in a significant (P<0.05) growth depression and nutrient depletion. Apparent protein digestibility (Fig. 9) was significantly (P<0.05) higher in fish which were fed diets containing probiotic at 0.75g 100 g<sup>-1</sup> than in fish fed probiotic free diet (Control) or diets containing low or high levels of lactobacillus. FCR values were also significantly (P<0.05) lower

in fish fed diet containing Lactobacillus at 0.75g 100g<sup>-1</sup> than fish fed other dietary preparations including control diet (Table 2).

Table 2: Effect of different levels of probiotic supplement on growth performance, digestibility, nutrient retention and excretion of metabolites in Chhana gachua fingerlings under laboratory conditions (LD 12:12 at  $25\pm1^{\circ}$ C) -70 days treatment Diets

Parameters	D0 (control)	D1	D2	D3	D4
Initial	- Look - Look - MARINING				
weight	1.22±0.02a	$1.29\pm0.04a$	1.23±0.01a	1.15±0.03a	1.20±0.02a
(g)					
Initial					
length	4.00±0.05a	4.25±0.06a	4.14±0.07a	4.09±0.06a	4.20±0.06a
(cm)					
Final					
weight	3.02±0.03e	3.53±0.02d	5.44±0.06b	6.79±0. <b>2</b> 4a	4.13±0.05c
(g)	~ ~ ~	Z 10 10 10 1	C 07 : 0 301	<i>m m</i> o . o . t o	6 10 10 00
Final length	5.77±0.07d	6.12±0.10cd	6.87±0.12b	7.72±0.12a	6.19±0.09c
(cm)	1.90±0.02 -	2 22 (0 024	4,21±0,06b	5.64±0.24a	2.93±0.06c
Live weight gain (g)	1.80±0.02e	2.23±0.03d	4.21±0.000	3.04±0.24a	2.93±0.060
	147.19±2.16d	174.20±7.26d	341 63±7 16b	491.21±25.59a	243.61+7.31c
gain in BW)		174.2047.20Q	341.03±7.100	7)1.21±25.57t	243.01±7.31 <b>C</b>
Specific Specific	1,29±0,01e	1.44±0.04d	2.12±0.02a	2.53±0.06b	1.76±0.03c
growth rate		2			
(SGR)					
Feed	2.07±0.08a	1.99±0.05a	1.95±0.05a	1.74±0.08b	2.04±0.08a
conversion					
ratio (FCR)					
Gross	19.97±0.96b	20.76±0.49b	21.47±0.70b	24.65±0.92a	20.49±1.07b
energy					
retention					
(GER)					
Gross	24.86±1.09b	25.93±0.79b	27.33±0.85b	31.81±1.32a	25.46±1.11b
protein					
retention					
(GPR)	1.00.00.001	1 27 ( 0 02)	1 20 : 0 021	1.47.0.07	1.041.0.051
Protein	1.23±0.05b	1.27±0.03b	1.30±0.03b	1.47±0.06a	1.24±0.05b

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efficiency
ratio (PER)
Apparent
            80.07±0.19d
                              81.28±0.26c
                                              83.58±0.20b 85.37±0.36a
                                                                               81.12±0.20c
protein
digestibility
(APD%)
Total
             1349.83±6.31a 1008.75±8.95b 621.46±2.11d 534.55±5.98e
                                                                               799.45 \pm 7.54c
ammonia
excretion
(mg kg<sup>-1</sup>
BW day<sup>-1</sup>)
Total
                             176.99±11.26c 144.84±4.44d 143.28±3.12d 199.77±4.84b
             244.42±0.24a
phosphate
production
     (mg kg<sup>-1</sup> BW day<sup>-1</sup>)
```

All values are mean±SE of mean.

Means bearing different letters in the same row differ significantly (P<0.05)

#### Discussion

The survival of Channa gachua in all different treatments was excellent. The optimum probiotics levels which resulted in high growth in Channa gachua in terms of live weight gain (grams), growth percentage gain, SGR and nutrient retention (PER, GPR, GER and APD) was found to be around 0.75g 100 g<sup>-1</sup> of diet. FCR values decreased with each increase in the dietary probiotic contents of the diet up to 0.75 g 100g<sup>-1</sup> of diet, thereafter, increases in dietary probiotic levels resulted in an increase in FCR and growth depression.

The high APD values for the diet containing lactobacillus at 0.75g 100 g<sup>-1</sup> of diet may be attributed to the probiotic concentration, which was used in diet D3 might be helpful for optimum dietary utilization. Similar results were also reported by Ghosh et al. (2003) using Bacillus circulans as probiotic in Labeo rohita fingerlings and Rengpipat et al. (1998) using Bacillus sp. S11 as probiotic in Penaeus monodon. In the present study, although, all the feeds. Were isonitrogenous but the concentration of probiotic in feed might be helpful for proper nutrient utilization.

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