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Growth Performance of *Labeo rohita* (Ham.) to Livol (IHF-1000), A Herbal Product

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The growth-promoting potential of Livol, a herbal product, was evaluated in the monoculture of Indian major carp, *Labeo rohita* in concrete tanks for a period of 112 days. Livol was incorporated at five levels *viz.*, 0.5, 1, 1.5, 2 and 2.5% by weight into the fish meal based pelleted feed having 35% protein (control). All levels of livol recorded significantly higher growth compared to control. During the fry stage 1% livol promoted better growth while 2% livol produced the final maximum growth both in length and weight. High specific growth rate was associated with 2% livol. The viscerosomatic and cerebrosomatic indices increased in the livol treated fishes while the hepatosomatic index decreased with higher levels of livol. Livol treated fishes showed most efficient feed utilization with high feed conversion efficiency, protein efficiency ratio, protein, carbohydrate and lipid digestibility. The RNA, DNA and RNA/DNA ratio in the muscle and liver were significantly higher ($p<0.01$) in the livol fed fishes with the RNA/DNA ratio showing a direct increase with weight gain. Carcass protein and lipid contents increased in the livol treated fishes with the maximum values in the 2% level. Livol at 2% can be recommended as the optimum level required for maximum growth of *L. rohita* in monoculture.

Key Words : Livol, Growth, Feed utilization, *Labeo rohita*, Proximate composition

Introduction

The most important operational functions in fish culture is supplementary feeding to obtain marketable size in a short time and at less cost. The expenditure on feed can be minimised by using cheaper protein sources or the use of feed additives which enhances growth. Growth promoters are non-nutritive materials which at a very low levels of incorporation in the feed, increase the efficiency of feed utilization (Viola & Arieli 1987). Livol is extensively used as liver tonic in animal husbandry (Patra & Pradhan 1992). Being herbal livol is claimed to be safe and free from any toxic or side effects. Studies on the effect of livol on fish growth and production are rather scarce (Abraham 1992, Shadakshari 1993, Unnikrishnan 1995) and the present study is a step ahead in evaluating its effect on growth, feed utilization, nucleic acid content and carcass composition of fish.

Materials and Methods

Experimental Diet

Livol-IHF 1000, a herbal product, was obtained from the Indian Herbs Research and Supply Co. Pvt. Ltd, Saharanpur, UP. The standard fish meal based pelleted feed with 35% protein was formulated (Table 1) and prepared. The concentration of livol, viz., 0.5, 1, 1.5, 2 and 2.5% were weighed out accurately and added to the cooled dough and mixed thoroughly to ensure uniform dispersal before pelletization.

Experimental Design

The culture experiment was conducted in cement tanks (5L x 4Bx1Hm) without any soil base. The tanks were filled with water upto a level of 75 cm. About 25% of water was changed fortnightly to avoid deterioration of water quality during the culture period. Three replications for each treatment were maintained. The treatments were randomly assigned to the tanks laid out in a completely randomised design (CRD). *Labeo rohita* fry procured from Neyyar Dam fish hatchery were randomly stocked at a density of 20 per tank. The fishes were fed the livol incorporated and control diets at 5% of the body weight twice daily. The feed pellets were placed in the feed trays suspended at four corners of each tank. The fishes were sampled fortnightly. A random sample of 15 fishes were collected and the individual growth in length and weight were measured. The quantity of feed given was re-adjusted after every sampling depending on the weight of fish. The experiment was terminated after 112 days. At the end of the experiment all the surviving fishes were collected by completely draining the tanks and their final length and weight were recorded. Five fishes from each tank were sacrificed for analysing the tissue indices. RNA-DNA contents and carcass proximate composition. Tissue indices, such as viscerosomatic index [VSI], hepatosomatic index [HSI], cerebrosomatic index [CSI] and renosomatic index [RSI] were calculated using the formula :

$$\text{Tissue indices} = \frac{\text{Weight of tissue (g)}}{\text{Total weight of fish (g)}} \times 100$$

The specific growth rate (SGR) was calculated as follows :

Table 1. *Proportion of feed ingredients and the proximate composition* of formulated diet*

Feed ingredients	Proportion (g)	Protein content (%)	Proximate composition of feed	(%)
Ricebran	14.17	0.98	Protein	34.80
Tapioca flour	14.17	0.31	Fat	9.54
Fishmeal	35.83	18.02	Carbohydrate	10.13
Groundnut oil cake	35.83	15.69	Fibre	4.01
			Ash	2.70
Total	100.00	35.00	Moisture	0.08

*Expressed on dry weight basis.

$$SGR (\%) = \frac{\log_e w2 - \log_e w1}{T_2 - T_1} \times 100$$

where w_2 is the weight at time T_2 and w_1 the weight at time T_1.

Water Quality Monitoring

Water samples were collected once in a fortnight and were analysed for dissolved oxygen, free carbon dioxide, ammonia, total alkalinity, total hardness, *p*H, temperature and primary productivity following APHA (1992) procedures.

RNA-DNA and Proximate Composition

RNA and DNA contents in the muscle and liver of the various treatments were estimated by adopting the methods of Curlewis and Stone (1987). The carcass proximate composition of the different treatments was analysed for protein, fat, carbohydrate, moisture, fibre and ash contents following AOAC (1984) procedure.

Feed Utilization

An experiment was conducted in the laboratory in triplicate using fibreglass troughs (50 litres) for 30 days to determine the feed consumption, conversion efficiency and digestibility of protein, fat and carbohydrate. Five individuals were kept in each trough. Three replications for each treatment were maintained and they were fed at 5% body weight level once a day in the morning. The unconsumed feed was collected 6 hrs after feeding. On the subsequent day before feeding, faecal matter accumulated in the tank was siphoned out. The uneaten feed and faecal matter were oven-dried and stored in dessicator for analysis. About three-fourth of the water in the trough was changed daily. Both the feed and faecal matter from the different treatments were analysed for protein, fat, carbohydrate and ash (AOAC 1984). The feed conversion efficiency (FCE), assimilation efficiency (AE), protein efficiency ratio (PER) and nutrient digestibility (protein, fat and carbohydrate) were calculated using the following formulae:

$$FCE (\%) = \frac{\text{Gain in wet weight of fish (g)}}{\text{Dry weight of feed consumed (g)}} \times 100$$

$$AE (\%) = \frac{\text{Assimilation}}{\text{Feed intake (g)}} \times 100$$

$$PER (\%) = \frac{\text{Increment in body weight (g)}}{\text{Protein intake (g)}} \times 100$$

$$\text{Nutrient digestibility (\%)} = \frac{\text{Nutrient intake-Nutrient in faeces}}{\text{Nutrient intake}} \times 100$$

Statistical Analysis

ANOVA method was employed to test the statistical significance of the various parameters in the different treatments. Duncan's multiple range test (Steel & Torrie 1980) was done to determine significant difference between treatment means.

Results

Water Quality Parameters

The water temperature ranged from 24 to 32.5° C, the *p*H from 7.2 to 7.8 (during the culture period), the total hardness of water from 34 to 46 mg $CaCO_3$/L and the total alkalinity from 1 to 2.1 unit equivalent/L.

Growth of L. rohita

The final mean length and weight were highest in 2% level livol fed fishes (Table 2) and the lowest in the control. The pattern of growth was slightly different upto 42nd day of culture. During this period the best mean weight was observed in 1% livol followed by 1.5, 2 and 2.5% respectively. Afterwards from the 84th day onwards (Figure 1) there was a gradual decrease in the growth rate in 2.5% while the fishes in the 2% level began to show superior growth. ANOVA showed significant difference between treatments and control ($p<0.01$) and the maximum growth recorded at 2.5% livol was significantly different from all other groups. The specific growth rate (SGR) was highest in 2% livol while lowest in control. The percentage survival was the same (95%) in all treatments.

Tissue Indices

A significantly high cerebrosomatic ($p<0.01$) index was observed in the 2% livol treatment compared to control. The VSI also increased in all the livol treated groups (Figure 2). HSI was found to decrease from control with the lowest value being recorded in 2% livol treatment except in 1% livol which recorded an increase. RSI did not show any significant variation ($p>0.01$).

Nucleic Acid Content

RNA and DNA contents of muscle and liver of the fishes fed on livol diet were significantly higher than that of the control (Table 3). The RNA/DNA ratio also followed the same trend with highest value being recorded in the 2% level diet which was significantly different from all other treatments ($p<0.01$).

Table 2. *Growth and survival of L. rohita fed different levels of livol*

Parameters		Treatments					
		Control	0.5%	1%	1.5%	2%	2.5%
Initial length (cm)	Mean	2.39	2.33	2.35	2.34	2.25	2.35
	SD±	0.22	0.2	0.24	0.28	0.19	0.25
Final length (cm)**	Mean	12.13^a	12.58^b	13.2^c	13.21^c	13.85^d	13.11^c
	SD±	0.52	0.47	0.33	0.41	0.43	0.38
Percentage increase in length**	Mean	407.6^a	439.9^b	461.8^c	464.5^c	515.5^d	457.9^c
	SD±	7.2314	6.7023	4.5772	5.909	6.4557	5.4218
Initial weight (g)	Mean	0.158	0.153	0.148	0.149	0.141	0.151
	SD±	0.03	0.03	0.04	0.03	0.03	0.04
Final weight (g)**	Mean	20.05^a	23.8^b	30.5^d	30.4^d	33.9^e	29^c
	SD±	0.54	0.35	0.66	0.58	0.54	0.58
Percentage increase in weight**	Mean	1290.1^a	15455.6^b	20508.3^d	20302.8	23942.6^e	19502.7^c
	SD±	114.3378	76.7128	149.0697	128.9206	128.6258	129.5108
Net weight gain (g)		19.892	23.647	30.352	30.251	33.759	29.449
Specific growth rate (SGR)%		4.32	4.51	4.76	4.75	4.90	4.71
Survival (%)		95	95	95	95	95	95

** Significantly different at 1% level ($p<0.01$).

a, b, c, d, e- Means with the same superscript don't differ significantly (Duncan's multiple range test).

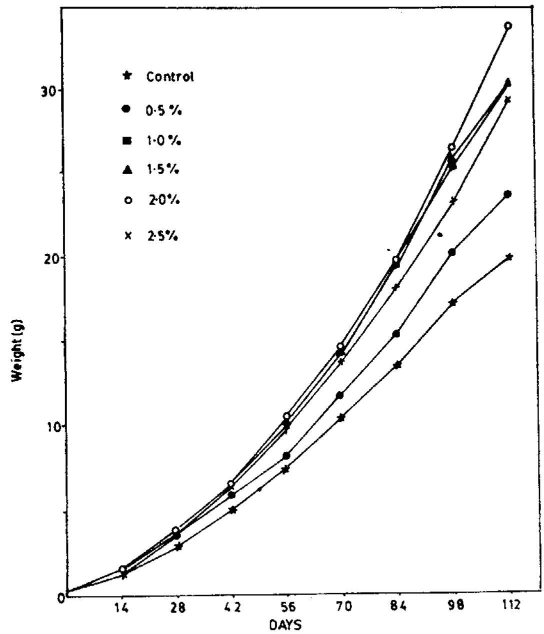

Figure 1. Growth pattern of *Labeo rohita* fed different levels of livol

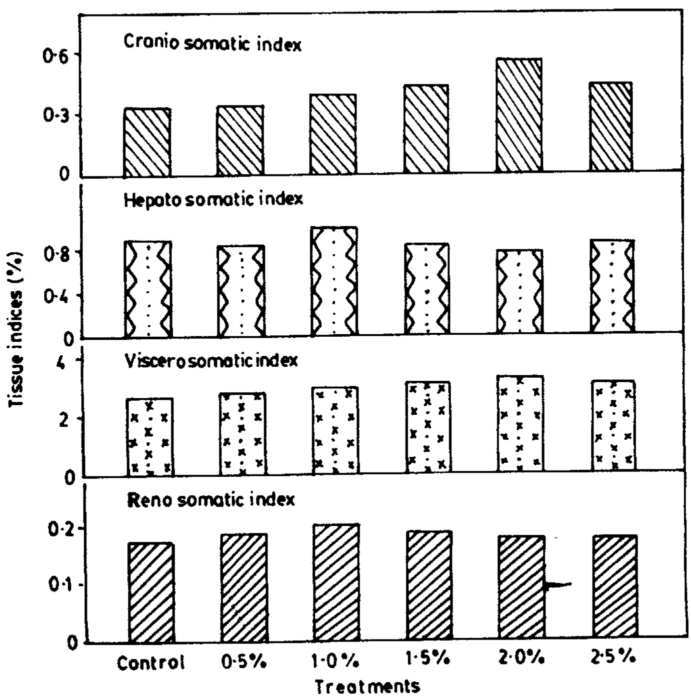

Figure 2. Tissue indices of *Labeo rohita* fed different levels of livol

Proximate Composition

The highest protein content (17.46%) was obtained in 2% livol which showed the maximum growth and the lowest in the control (Figure 3). The fat content also increased in the livol treated fishes while no variation was noticed in the carbohydrate content in the different treatments. The ash content showed a decrease in the livol fed fishes with the maximum in the control.

Feed Utilization

The feed intake and assimilation were higher in the fishes fed with livol incorporated diet compared to that of the control. Better feed conversion efficiency and assimilation efficiency were found due to livol administration (Table 4). Protein efficiency ratio (PER) increased in all livol incorporated diets. High protein digestibility was noticed in 1% livol while high fat digestibility was observed in 2% livol.

Discussion

The positive effect of livol in the efficient feed utilization has led to the possibility of using livol as a growth promoter in fish culture. Very recently, few studies have indicated the growth promoting function of this herbal product in fishes (Abraham 1992, Shadakshari 1993, Unnikrishnan 1995). The present study clearly indicates the positive influence of livol on weight gain of *L. rohita*. The best growth of rohu was observed with 2% level which was also evidenced by high specific growth rate. It is interesting to note that rohu showed better growth

Table 3 *Nucleic acid content in the muscle and liver of L. rohita fed different levels of livol*

Parameters %	Muscle						Liver					
	Control	0.5%	1%	1.5%	2%	2.5%	Control	0.5%	1%	1.5%	2%	2.5%
RNA** M±	68.40^{a}	78.00^{b}	114.00^{cd}	116.4^{d}	153.6^{e}	110.4^{c}	70.00^{a}	98.4^{b}	141.60^{d}	146.40^{e}	183.60^{d}	134.40^{c}
SD±	3.29	2.45	4.24	3.29	3.29	3.29	4.24	3.29	3.29	3.29	3.29	3.29
DNA** M±	17.00^{a}	17.80^{a}	21.40^{b}	21.40^{b}	24.40^{c}	21.00^{b}	19.00^{a}	21.00^{b}	27.40^{c}	28.20^{c}	30.40^{d}	26.80^{c}
SD±	0.71	0.84	1.67	1.67	0.55	2.00	2.00	1.41	1.14	0.84	0.55	0.84
DNA**/M±	4.026^{a}	4.392^{b}	5.354^{b}	5.46^{b}	6.296^{c}	5.298^{b}	4.148^{a}	4.752^{b}	5.172^{c}	5.19^{c}	6.042^{d}	5.034^{cd}
RNA SD±	0.25	0.32	0.47	0.32	0.11	0.59	0.54	0.32	0.22	0.16	0.16	0.20

RNA is expressed as mg RNA 100^{-1} mg tissue (Values are the mean of 5 analysis).
DNA is expressed as mg DNA 100^{-1} mg tissue
**Significantly different at 1% level ($p<0.01$)
a, b, c, d, e- Means with the same superscripts do not differ significantly (Duncan's multiple range test)

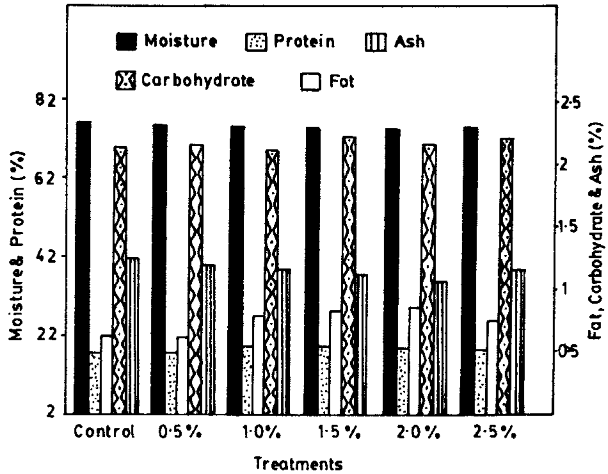

Figure 3. Proximate composition of *Labeo rohita* fed different levels of livol

response to 1% livol during the first half of the culture period, when they were in the fry stage. It was only after 72 days of culture that *L. rohita* began to respond better to 2% level. Thus it appears that the optimum level of livol required for growth promotion varies depending on the age and size of the fish. Abraham (1992) and Shadakshari (1993) reported that 1% livol stimulated higher growth in *C. carpio* while 0.5% and 1.5% livol showed poorer growth. In *C. catla* the growth rate followed a positive increase with higher concentration of livol, the best growth being recorded with highest dosage of 2.5% level (Unnikrishnan 1995). The growth responses of livol in the various fishes indicate that the optimum dosage for promoting maximum growth is species dependent.

A decrease in HSI after livol treatment may be attributed to lipid depletion in the liver consequent to enhanced lipid metabolism. Shadakshari (1993) also reported stimulated fat metabolism of *C. carpio* treated with livol. The increase in the VSI can be attributed to the deposition of fat in viscera. Increased visceral fat deposition has been reported in tilapia (Nair & Gopakumar 1981) and in mahseer (Bazaz & Keshavanath 1993).

Feed conversion efficiency (FCE) can be regarded as an index of food utilization. The highest FCE was obtained with 1% livol where the final growth performance was the second best. Increase in FCE of livol treated fishes was reported by Abraham (1992) in *C. carpio* and Unnikrishnan (1995) in *C. catla*. Protein efficiency ratio (PER) provides better indication of nutritional status and is a measure of the utilization of dietary protein by fish. Higher PER values in 1% livol indicates better utilization of protein from it. Livol incorporation improved protein and fat digestibility in the present study. As the feed utilization experiment was conducted with fry, better performance in feed utilization was obtained in 1% livol treatment. It seems

Table 4. *Feed utilization of* ***L. rohita*** *fed different levels of livol*

Parameters	Treatments											
	Control		0.5%		1%		1.5%		2%		2.5%	
	Mean	±SD	Mean	±SD	Mean	±SD	Mean	±SD	Mean	±SD	Mean	±SD
Initial weight (w1) (g)	2.5	.065	2.3	0.15	2.2	.05	2.4	.07	2.3	.08	2.21	.03
Final weight (w2) (g)	3.7	.24	3.65	0.36	3.9	.30	3.9	.34	3.7	.23	3.69	.36
Production (P=w2 w1) (g)	1.2	.14	1.35	.89	1.7	.46	1.5	.21	1.4	.56	1.48	.25
Feed consumed (C) (g)	5.7	.54	5.8	.62	6.3	.58	6.0	.43	5.8	.64	5.9	.63
Faecal output (F) (g)	3.0	.18	2.9	.22	3.0	.25	2.9	.26	2.8	.12	3.0	.12
Relative growth rate (P/w1)	0.48	.02	0.59	.03	0.77	.02	.625	.03	.61	.01	0.67	.03
Assimilation (A=C-F)	2.7	.33	2.9	.28	3.3	.31	3.1	.36	3.0	.41	2.9	.47
Metabolism (R=A-P)	1.5	.36	1.55	.25	1.6	.19	1.6	.24	1.6	.16	1.42	.28
Net growth efficiency % (K=P/Ax100)	44.44	.65	46.55	.54	51.52	.44	48.39	.80	46.67	.58	51.03	.38
Feed conversion efficiency (%) (P/Cx100)	21.05	.78	23.28	.77	27.42	.32	25.00	.46	24.14	.62	25.08	.56
Assimilation efficiency (%) (A/Cx100)	47.37	1.33	5.00	1.62	52.38	.89	51.67	.95	51.72	1.22	49.15	.73
Protein efficiency ratio (PER)	0.62	.01	0.66	.03	0.78	.03	0.71	.02	.70	.02	.70	.01
Protein digestibility (%)	74.28	.45	76.46	.62	78.92	.47	77.2	.55	77.42	.38	76.8	.62
Fat digestibility (%)	68.96	.32	69.24	.28	73.2	.39	73.1	.23	73.95	.41	72.26	.30
Carbohydrate digestibility (%)	70.84	.64	71.08	.53	71.42	.51	71.23	.48	71.36	.34	71.53	.38

Values are expressed as mean ±SD of three replicates that in the fry stage, fishes may respond better to lower concentration of livol. Growth in fish is associated with maximum deposition of protein. A significant increase was obtained in the level of RNA, DNA and RNA/DNA ratio in the muscle and liver of livol fed fishes. Wilder and Stanley (1993) reported that rapidly growing organisms synthesise and accumulate RNA needed for protein synthesis and RNA and the ratio of RNA to DNA are useful as indices to growth. According to Mathers et al (1992), RNA concentration is directly linked to capacity for protein synthesis. In *L. rohita,* the highest growth rate in 2% livol may be correlated with the high levels of RNA and RNA/DNA ratio leading to enhanced protein synthesis. Since fish growth is largely due to protein synthesis, higher protein content of livol fed fishes indicates the growth promoting potential of livol.

The present study demonstrates an increase in the growth rate of *L. rohita* following administration of livol supplemented diets. There is an increase in feed consumption, conversion efficiency, digestibility, RNA/DNA ratio and protein content of body. Livol can be recommended for use as a safe growth promoter in aquaculture industry. Nevertheless, further detailed studies are necessary to elaborate the specific role of livol, its mode of action in stimulating growth and the economics of culture operation.

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