

Possibilities of Utilizing Dietary Supplements in Semi-intensive Culture of Deccan Mahseer, *Tor khudree* (Sykes)

Y. BASADE^{1*}, M.P.S. KOHLI² AND S.N. OGALÉ³

¹Directorate of Coldwater Fisheries Research
(Formerly National Research Centre on Coldwater Fisheries)
Bhimtal – 263 136, District Nainital (Uttarakhand), India

²Aquaculture Division
Central Institute of Fisheries Education (Deemed University)
Mumbai, India

³Tata Power Company Limited
Lonavla, District Pune, India

Abstract

Effectiveness of dietary supplements on the growth performance, survival, nutritional efficiency, body indices and biochemical composition of deccan mahseer (*Tor khudree*) was evaluated in a semi-intensive culture system. The ponds were fertilized with raw cow dung and single super phosphate. Fish (1.10 ± 0.03 g) at a stocking density of three per square meter were fed for a period of 180 days with the formulated practical diets. The test diet D1 was un-supplemented basal diet, which served as the control while in four of the test diets, D2, D3, D4 and D5, dietary supplements viz. cholecalciferol ($1800 \text{ IU} \cdot \text{kg}^{-1}$), soylecithin (3.5%), thyroxine (0.05ppm) and betaine (0.5%), respectively, were added. The inclusion level of the dietary supplement was based on the best results obtained, in terms of growth performance, feed utilization and survival, in previous investigations carried out for the same fish species under laboratory conditions.

Feeding soylecithin supplemented diet (D3) resulted in significantly ($P < 0.05$) higher percentage weight gain and specific growth rate (SGR) than was observed with other diets. Survival was independent of the treatments. Feed conversion ratio (FCR), feed conversion efficiency (FCE) and protein efficiency ratio (PER) were significantly better ($P < 0.05$) in the fish fed with the diet D3 compared to fish fed with all other diets. The crude protein efficiency (CPE), crude fat efficiency (CFE) and gross energy efficiency

* Corresponding author. Tel.: +91 5942 247280, Fax: +91 5942 247693
E-mail address: yasmeenbasade@yahoo.co.in

(GEE) in relation to all the treatments were significantly higher ($P < 0.05$) in D3. Among the body indices, viscero-somatic index (VSI) was significantly ($P < 0.05$) higher in D3, cranio-somatic index (CSI) was significantly higher ($P < 0.05$) in the control diet (D1), while hepato-somatic index (HSI) and reno-somatic index (RSI) exhibited no significant differences ($P > 0.05$) between the test diets except for D4 in which HSI and RSI were significantly lower ($P < 0.05$). Body protein, lipid, ash, energy contents and RNA/DNA ratio were significantly ($P < 0.05$) higher in fish fed with the diet D3 and significantly lower in fish fed with diet D1 compared to fish fed with all other diets. Environmental variables, particularly water temperature and the natural productivity of the pond ecosystem were within the optimum range as required for the growth of deccan mahseer under culture systems.

Introduction

Aquaculture is undergoing an exponential development due to overexploitation of capture fisheries, coupled with an increasing demand for food fish. The world aquaculture production has increased by more than 85% in the past decade. Freshwater aquaculture represents approximately half of the total world aquaculture production, with an annual yield of about 25 million tonnes of finfish (FAO 2007). Finfish culture is dominated by semi-intensive practices (De Silva 1993) and is mostly pond based in developing countries (De Silva and Davy 1992). Fish yield in semi-intensive aquaculture depends to varying extent on natural food production and to a considerable extent on supplementary feeding.

Mahseer, *Tor* spp. (Order Cypriniformes; Family Cyprinidae) are widely distributed in India, Pakistan, Afghanistan, Bangladesh, Nepal, China, Sri Lanka and Burma, occurring abundantly in mountain and rocky streams (Nautiyal 1994). They show excellent aquaculture characteristics, such as amenability to culture in captivity, capacity to accept supplementary feed and ability to tolerate wide range of environmental parameters (Cordington 1939; Jhingran and Sehgal 1978; Chauhan et al. 2007).

Mahseer are considered as the most valuable game and food fishes. But in the past there has been a great decline in the fishery of mahseer and are feared to be endangered (Anon. 1976). Reduction in natural populations of mahseer has been reported from India, Pakistan, Nepal and Bangladesh (Mirza 1994; Mirza and Khan 1994; Mirza et al. 1994; Shrestha 1994; Islam 2002; Chauhan et al. 2007). The need for scientific management, conservation of fishery resources of mahseer and the development of fisheries of this fishes in impoundment waters has been stressed (Sehgal and Malik 1992; Singh 1992; Sunder et al. 1995).

Sen and Jayaram (1982) described six species of mahseer from India (*T. putitora*, *T. tor*, *T. mosal*, *T. mussullah*, *T. khudree*, *T. progeneius*) of which, the deccan mahseer, *Tor khudree* (Sykes), is widely distributed in all the Peninsular rivers. Fortunately, biological investigations had already commenced towards conservation of deccan mahseer. A commendable success on the production of stocking material through artificial spawning has also been achieved and attempts have been made to assess their culture feasibility (Kulkarni 2000; Kohli et al. 2005). However, studies on the nutritional aspects from the point of view of culture, though important, are very limited (Keshavanath 2000; Basade and Kohli 2004). The success of any culture practice depends on the availability of well-balanced cheaper supplementary diets (Keshavanath et al. 2007). The recent advances in aquaculture research have revealed that dietary supplements like hormones, feeding stimulants, other nutritive and non-nutritive feed additives not only increase voluntary feed intake but also increase growth, feed efficiency, and survival of fishes (Lone and Matty 1982; 1983; Fredette et al. 2000; Papatryphon and Soares 2000). Most of the research with dietary supplements is limited only to the laboratory conditions. The efficacy of using such compounds in practical feeding under field conditions needs to be evaluated. Therefore, the objective of the present study was to evaluate the possibilities of including certain dietary supplements viz., cholecalciferol, soylecithin, thyroxine and betaine in the feeds of deccan mahseer being reared under semi-intensive culture system.

Materials and Methods

Experimental design

Deccan mahseer, *Tor khudree* juveniles were obtained from the Mahseer Fish Farm of Tata Power Company Limited, Lonavla, District Pune and transported to Central Institute of Fisheries Education (Deemed University), Mumbai. Experiments were conducted in cement tanks, each having a water holding capacity of 14.73 m³ (4.96 m x 2.97 m x 1 m). Prior to starting the experiments all the tanks were drained, cleaned, sun dried, limed at the rate of 200 kg•ha⁻¹ (300 g•cistern⁻¹) using quick lime and allowed to dry for about a week. Then they were filled with ground water to a depth of 1 m. This level of water was maintained throughout the experimental period. After filling water, initially the tanks were fertilized with cowdung at the rate of 2000 kg•ha⁻¹ (3 kg•cistern⁻¹) on wet weight

basis and single super phosphate at the rate of $300 \text{ kg}\cdot\text{ha}^{-1}$ ($40 \text{ g}\cdot\text{cisterns}^{-1}$). Subsequently, fertilization was done at monthly intervals with half of the initial amounts till the conclusion of the trial. A week after initial fertilization, uniform sized fingerlings of deccan mahseer were randomly stocked in ten cement tanks after recording their initial total length and body weight at the rate of $3 \text{ fish}\cdot\text{m}^{-3}$ ($45 \text{ fish}\cdot\text{tank}^{-1}$). The experimental feeds were fed to duplicate groups of fish at 5% of the body weight in plastic trays (26 cm x 21 cm x 5 cm) kept suspended in the cisterns at a level of about 30 cm above from the bottom. The quantity of feed given was adjusted based on the increase in weight of fish as measured during the monthly samplings.

Experimental diets

All experimental diets were formulated to contain a basal mixture of fish meal, groundnut oil cake, soybean oil cake, wheat bran, wheat flour, vegetable oil, mineral mixture (Bernhart Tommarelli- a modified NRC salt mixture) and vitamin mixture (Cebexin, Vitamin A capsules, Evion 600 and Arachitol 3L). In addition to the above ingredients the feed additives Eltroxin (Glaxo India Ltd., Mumbai) as thyroxine source, soylecithin (Alpine Solvex Ltd., Mumbai) as a source of phospholipid, Arachitol 3L (Duphar-interfran Ltd., Vapi, Gujarat) as cholicalciferol source and Betaine hydrochloride (E. Merck India Ltd., Mumbai) as feed attractant were used as dietary supplements.

Five experimental diets (D1 to D5) were formulated, out of which four diets (D2 to D5) were formulated by supplementing the basal diet with the dietary supplements viz., cholicalciferol at $1800 \text{ IU}\cdot\text{kg}^{-1}$ level, soylecithin at 3.5% level, thyroxine at 0.05 ppm level and betaine at 0.5% level, respectively, based on the best results obtained, in terms of growth performance, feed utilization and survival, in previous investigations carried out for the same fish species under laboratory conditions (Basade 2001). One diet without dietary supplement having only the basal feed was used as a control (D1). Diet formulations and their respective proximate compositions are presented in [table 1](#).

Experimental diets were prepared as sinking pellets. All the dry feed ingredients were separately ground, weighed in desired proportions and mixed thoroughly. The requisite amounts of vegetable oil, vitamin mixture, mineral mixture and dietary supplements were mixed thoroughly with water and then added to the mixture of dry ingredients to produce dough. The dough was extruded through twin screw extruder having sieve of 2.0 mm diameter. The pellets were dried in a hot-air oven at 40°C till

the moisture content was reduced to less than 10%. The feeds were cooled to room temperature, crumbled and packed in plastic bags and stored in cool dry place until use.

Table 1. Ingredient and proximate composition of the experimental diets

| Ingredients (g•kg ⁻¹ diet) | Diets | | | | |
|--|-------|-------|-------|-------|-------|
| | D1 | D2 | D3 | D4 | D5 |
| Fish meal | 360 | 360 | 360 | 360 | 360 |
| Groundnut oil cake | 210 | 210 | 210 | 210 | 210 |
| Soybean oil cake | 120 | 120 | 120 | 120 | 120 |
| Wheat bran | 120 | 120 | 120 | 120 | 120 |
| Wheat flour | 90 | 90 | 90 | 90 | 85 |
| Vegetable oil | 60 | 60 | 25 | 60 | 60 |
| Vitamin mix ¹ | 10 | 10 | 10 | 10 | 10 |
| Mineral mix ² | 30 | 30 | 30 | 30 | 30 |
| Cholicalciferol (IU) ³ | - | 1800 | - | - | - |
| Soylecithin (g) ⁴ | - | - | 35 | - | - |
| Thyroxine (ppm) ⁵ | - | - | - | 0.05 | - |
| Betaine (g) ⁶ | - | - | - | - | 5 |
| Proximate composition (g•100g ⁻¹): | | | | | |
| Dry matter | 96.96 | 96.33 | 97.47 | 96.52 | 96.62 |
| Crude protein | 37.86 | 37.78 | 37.13 | 37.13 | 37.36 |
| Crude lipid | 10.31 | 10.68 | 10.69 | 10.10 | 10.42 |
| Crude fiber | 7.65 | 7.70 | 7.80 | 7.81 | 7.86 |
| Ash | 10.09 | 12.44 | 13.65 | 13.16 | 13.08 |
| NFE | 31.03 | 27.73 | 28.21 | 28.32 | 27.56 |
| Gross energy (kJ•g ⁻¹) | 17.91 | 17.46 | 17.40 | 17.19 | 17.23 |

Diets: D1-Control; D2-Cholicalciferol supplemented; D3-Soy lecithin supplemented; D4-Thyroxine supplemented; D5- Betaine supplemented

¹Vitamin mix: Each 100g contains thiamine mononitrate (3.52g), riboflavin (3.52g), pyridoxine hydrochloride (1.05g), cyanocobalamin (0.004g), folic acid (0.53g), nicotinamide (35.15g), calcium pantothenate (3.52g) and ascorbic acid (52.72g), vitamin A (500 IU), vitamin D₃ (30 IU), vitamin E (81.6 IU).

²Mineral mix: Each 100g contains calcium phosphate dibasic (73.5g), potassium phosphate dibasic (8.1g), potassium sulphate (6.8g), sodium chloride (3.06g), calcium carbonate (2.1g), sodium phosphate dibasic (2.14g), magnesium oxide (2.21g), ferric citrate (0.558g), zinc carbonate (0.081g), manganese carbonate (0.421g), cupric carbonate (0.333g), potassium iodide (0.00072g), citric acid (0.702g).

³ Cholicalciferol: Arachitol 3L, Duphar-Interfran, Ltd., Vapi, Gujarat, India

⁴ Soylecithin: Alpine Slovex, Ltd., Mumbai, India

⁵ Thyroxine: Eltroxin, Glaxo India Ltd., Mumbai, India

⁶ Betaine: Betaine hydrochloride, E. Merck (India), Ltd., Mumbai, India

Diet and fish analysis

Proximate analysis of the formulated diets and test fish was performed according to AOAC (1995) methods. The dry matter was estimated

by heating the samples at 105°C in an oven till a constant weight was obtained. The crude protein content was determined by estimating nitrogen using micro-kjeldhal method. The nitrogen value obtained was multiplied by the factor 6.25 to get crude protein value. Crude fat was extracted using petroleum ether (40-60°C boiling point) in Soxhlet extraction apparatus. Samples were heated at 600°C for 6 hrs in a muffle furnace to determine the ash content. Crude fiber was estimated by digesting the samples in acid and alkali in Fibretec apparatus (Tecator, Fibretec system M, 1017). The nitrogen free extract (NFE) was calculated as $NFE = 100 - (\% \text{ moisture} + \% \text{ crude protein} + \% \text{ crude lipid} + \% \text{ crude fiber} + \% \text{ ash})$ (Hardy 1989). The gross energy values were calculated in terms of kilo Joules (kJ) using energy values of 38.9 kJ•g⁻¹ for lipid, 22.6 kJ•g⁻¹ for protein and 17.2 kJ•g⁻¹ for carbohydrates (Mayes 1990).

The biochemical composition of deccan mahseer was analyzed at the onset and on the conclusion of trial in terms of moisture, protein, lipid, ash, energy contents and muscle nucleic acids. Ribonucleic acid (RNA) and deoxyribonucleic acid (DNA) were extracted from fish muscle by trichloroacetic acid and quantified by means of colorimetric reaction, using diphenylamine reagent for DNA and orcinol reagent for RNA taking degraded free acid herring sperm DNA (SISCO Research Laboratories Pvt. Ltd., Mumbai) and extra pure RNA (SISCO Research laboratories Pvt. Ltd., Mumbai) as DNA and RNA standards, respectively, as per the methodology recommended by Schneider (1966).

Growth and nutritional utilization

The average total length and body weight of fish was recorded initially and then subsequently at monthly intervals till conclusion of the trial. Fish were sampled with dragnets of suitable mesh size and 40 fish were collected randomly for assessment of growth and biomass. On termination of the trial for each of the experimental unit various growth and nutritional parameters were calculated, namely, average total length and body weight of fish, percent weight gain, specific growth rate (SGR), percentage survival, feed conversion ratio (FCR), feed conversion efficiency (FCE), protein efficiency ratio (PER), body indices viz., hepato-somatic index (HSI), viscero-somatic index (VSI), reno-somatic index (RSI), and cranio-somatic index (CSI) and the crude protein, crude fat and gross energy efficiencies (Jover et al. 1999).

Water quality management

Physico - chemical and the biological parameters of water were analyzed at fortnightly intervals in each experimental tank. Water temperature was recorded twice daily in the morning and in the evening. Dissolved oxygen, free carbon dioxide, total alkalinity, hardness, nitrate and phosphate were analyzed following the [APHA \(1998\)](#) standard procedures. pH was measured using digital pH meter (Electronic Corporation of India Limited (ECIL), series 416 PH 5652).

Plankton samples were collected by filtering 50l of water from each tank through bolting silk cloth (No. 25, mesh size 64 μ), preserved in 4% formalin for qualitative and quantitative analysis. Samples were analysed qualitatively by identification up to the generic level and quantitatively by counting phytoplankton and zooplankton using Sedgewick Rafter plankton counting cell ([Adoni 1985](#)) and by determining the dry weight.

Statistical analysis

One-way analysis of variance (ANOVA) technique ([Snedecor and Cochran 1994](#)) was employed to test the difference between treatment means for the various parameters studied and when significant difference ($P<0.05$) was observed between treatments, further analysis was carried out employing Duncan's multiple range test at 5% level of significance ([Montgomery 1991](#)).

Results

Water quality

The physico-chemical and biological parameters of water observed during the trial are presented treatment wise in [table 2](#). During the experimental period water temperature in the ponds averaged from 27.62 \pm 0.22 to 29.04 \pm 0.75 $^{\circ}$ C in the morning and 29.73 \pm 0.25 to 32.85 \pm 0.47 $^{\circ}$ C in the evening. The pH was alkaline throughout the experimental period in all the ponds varying from 7.95 \pm 0.10 to 8.12 \pm 0.04. Among the treatments the average values of dissolved oxygen were 3.77 \pm 0.23 to 9.66 \pm 0.23 mg \cdot L $^{-1}$, free carbon dioxide 0.01 \pm 0.01 to 0.08 \pm 0.04 mg \cdot L $^{-1}$, total alkalinity 139.23 \pm 6.73 to 191.23 \pm 7.03 mg \cdot L $^{-1}$, total hardness 164.46 \pm 9.09 to 242.15 \pm 9.24 mg \cdot L $^{-1}$, nitrate 0.24 \pm 0.01 to 0.29 \pm 0.01 mg \cdot L $^{-1}$ and phosphate 0.18 \pm 0.01 to 0.20 \pm 0.01 mg \cdot L $^{-1}$.

Table 2. Water quality parameters in different treatment ponds. Symbols of experimental diets are as given in Table 1.

| | Diets | | | | |
|--|-------------------------------|-------------------------------|-------------------------------|-------------------------------|------------------------------|
| | D1 | D2 | D3 | D4 | D5 |
| Temperature (°C; a.m.) | 27.62 ±0.22 ^a | 28.38 ±0.29 ^a | 28.31 ±0.27 ^a | 27.96 ±0.23 ^a | 29.04 ±0.75 ^a |
| Temperature (°C; p.m.) | 29.73 ±0.25 ^a | 32.85 ±0.47 ^a | 32.19 ±0.43 ^a | 30.65 ±0.41 ^a | 31.08 ±0.32 ^a |
| pH | 7.95 ±0.10 ^a | 8.12 ±0.04 ^a | 8.06 ±0.06 ^a | 8.00 ±0.08 ^a | 8.08 ±0.07 ^a |
| Dissolved oxygen (mg•L ⁻¹) | 3.77 ±0.23 ^a | 9.66 ±0.23 ^b | 8.82 ±0.38 ^c | 6.14 ±0.60 ^d | 8.17 ±0.47 ^e |
| Free carbon dioxide (mg•L ⁻¹) | 0.08 ±0.04 ^a | 0.01 ±0.01 ^a | 0.02 ±0.01 ^a | 0.06 ±0.04 ^a | 0.04 ±0.03 ^a |
| Total alkalinity (mg•L ⁻¹) | 180.31 ±14.64 ^a | 139.23 ±6.73 ^b | 139.85 ±13.49 ^b | 179.38 ±11.87 ^a | 191.23 ±7.03 ^c |
| Total hardness (mg•L ⁻¹) | 217.85 ±17.38 ^a | 164.46 ±9.09 ^b | 181.85 ±16.51 ^c | 217.08 ±15.92 ^a | 242.15 ±9.24 ^d |
| Nitrate (mg•L ⁻¹) | 0.28 ±0.01 ^a | 0.24 ±0.01 ^a | 0.24 ±0.02 ^a | 0.26 ±0.01 ^a | 0.29 ±0.01 ^a |
| Phosphate (mg•L ⁻¹) | 0.20 ±0.01 ^a | 0.18 ±0.01 ^a | 0.18 ±0.01 ^a | 0.19 ±0.01 ^a | 0.19 ±0.01 ^a |
| Plankton biomass (mg•L ⁻¹) | 0.86 ±0.10 ^a | 1.87 ±0.07 ^b | 1.56 ±0.11 ^c | 1.68 ±0.11 ^d | 1.28 ±0.13 ^e |
| Phytoplankton (nos•L ⁻¹) | 14354 ±8134 ^a | 145115 ±51660 ^b | 84745 ±36008 ^c | 95810 ±36884 ^d | 38529 ±13655 ^e |
| Zooplankton (nos•L ⁻¹) | 1643 ±313 ^a | 7545 ±1948 ^b | 7229 ±1846 ^b | 4662 ±898 ^c | 2038 ±266 ^d |

Values are means (\pm SE); means within a row having different superscripts are significantly different ($P < 0.05$).

The average dry weight of plankton ranged from 0.86 ± 0.10 to 1.87 ± 0.07 mg•L⁻¹ between the treatments. Phytoplankton encountered in the various treatments belonged to three main groups, viz., Cyanophyceae, Chlorophyceae and Bacillariophyceae. Chlorophyceae were generally the dominant group in all the treatments, followed by Cyanophyceae and Bacillariophyceae. Chlorophyceae were represented mainly by *Closterium* sp., *Coelastrum* sp., *Pediastrum* sp., *Pandorina* sp., *Mougeotia* sp., *Scenedesmus* sp., *Eudorina* sp., *Ulothrix* sp. and *Volvox* sp., while Cyanophyceae consisted of *Anabaena* sp., *Microcystis* sp., and *Oscillatoria* sp. *Synedra* sp., *Melosira* sp., *Navicula* sp. and *Fragilaria* sp. were recorded among Bacillariophyceae. The average values of phytoplankton count for the different treatments ranged from 1.44×10^4 to 1.45×10^5 No.•L⁻¹.

Rotifers, copepods, cladocerans, ostracods and crustacean larvae were the important groups of zooplankton that were present in the different treatments during the experimental period. Rotifers were mainly represented by *Brachionus* sp., *Keratella* sp., *Hexarthra* sp., *Ascomorpha* sp., *Asplanchna* sp. and *Monostyla* sp. Copepods consisted of *Cyclops* sp. and *Diaptomus* sp., while cladocerans were mainly represented by *Moina* sp. *Cypria* sp. and *Cyclocypris* sp. belonging to ostracods were also observed. The average values of zooplankton count ranged from 1.64×10^3 to 7.55×10^3 No. \bullet L⁻¹ under different treatments.

Growth performance, nutritional utilization and survival

On termination of the trial, the average total length and body weight recorded for the treatments were observed to be highest in D3 averaging 199.25 ± 0.25 mm and 70.80 ± 0.69 g (Table 3). The percent weight gain, specific growth rate (SGR), feed conversion ratio (FCR), feed conversion efficiency (FCE) and protein efficiency ratio (PER) were significantly better ($P < 0.05$) in D3 being $6672.20 \pm 774.26\%$, $2.34 \pm 0.01\% \bullet \text{day}^{-1}$, 1.89 ± 0.03 , $52.98 \pm 0.81\%$ and 1.43 ± 0.02 , respectively, compared to all other treatments. Among treatments the crude protein efficiency (CPE), crude fat efficiency (CFE) and gross energy efficiency (GEE) were significantly higher ($P < 0.05$) in D3 with the respective values of 31.21 ± 0.49 , 59.83 ± 0.94 and $29.35 \pm 0.46\%$. The percentage survival ranged from 95.56 to $98.89 \pm 1.11\%$ and showed no significant difference ($P > 0.05$) between the treatments (Table 3).

Body indices

Viscero-somatic index (VSI) was significantly higher ($P < 0.05$) in D3 treatment ($5.07 \pm 0.06\%$) compared to all other treatments. Hepato-somatic index (HSI) and reno-somatic index (RSI) were not significantly different ($P > 0.05$) among the treatments except for D4 in which HSI and RSI were significantly less ($P < 0.05$). In comparison to all the treatments carnio-somatic index (CSI) was significantly ($P < 0.05$) higher in D1 treatment, $0.55 \pm 0.03\%$ (Table 4).

Body composition and muscle nucleic acids

The whole body protein, lipid and energy contents were significantly higher ($P < 0.05$) in fish fed on diet D3 with the respective values $21.79 \pm 0.07\%$, $12.03 \pm 0.04\%$ and $9.60 \pm 0.02 \text{kJ} \bullet \text{g}^{-1}$ compared to fish fed all other diets (Table 5). Consequently, among all the treatments moisture content was significantly less ($P < 0.05$) in treatment D3 to the order of

62.29±0.14%. Of all the treatments highest muscle RNA value of 1.03±0.01 mg•g⁻¹ was observed in fish from D3 treatment, while DNA value of 0.25±0.01 mg•g⁻¹ in D3 treatment was the minimum. Hence, the ratio of RNA to DNA was significantly higher (P<0.05) in D3 treatment with the value of 4.13±0.05.

Table 3. Growth performance, nutritional utilization and survival of deccan mahseer fed on experimental diets. Symbols of experimental diets are as given in Table 1.

| | Diets | | | | |
|-------------------------------|---------------------------------|---------------------------------|---------------------------------|--------------------------------|--------------------------------|
| | D1 | D2 | D3 | D4 | D5 |
| Initial length (mm) | 51.00 ±1.00 | 50.00 ±1.00 | 48.00 ±0.00 | 51.50 ±0.50 | 52.00 ±1.00 |
| Final length (mm) | 136.25 ±1.25 | 181.85 ±0.35 | 199.25 ±0.25 | 147.75 ±5.37 | 159.54 ±0.54 |
| Initial weight (g) | 1.10 ±0.09 | 1.03 ±0.08 | 1.06 ±0.11 | 1.22 ±0.02 | 1.10 ±0.01 |
| Final weight (g) | 30.96 ±0.36 ^a | 54.30 ±0.97 ^b | 70.80 ±0.69 ^c | 36.58 ±0.45 ^d | 41.82 ±0.46 ^c |
| Weight gain (%) | 2721.25 ±192.64 ^a | 5168.37 ±290.89 ^b | 6672.20 ±774.26 ^c | 2892.80 ±96.38 ^a | 3694.82 ±13.51 ^d |
| SGR (%•day ⁻¹) | 1.85 ±0.02 ^a | 2.20 ±0.01 ^b | 2.34 ±0.01 ^c | 1.89 ±0.02 ^a | 2.02 ±0.001 ^d |
| FCR | 2.22 ±0.02 ^a | 2.19 ±0.02 ^a | 1.89 ±0.03 ^b | 2.40 ±0.04 ^c | 2.33 ±0.02 ^d |
| FCE (%) | 44.99 ±0.41 ^a | 45.67 ±0.36 ^a | 52.98 ±0.81 ^b | 41.69 ±0.63 ^c | 42.91 ±0.28 ^c |
| PER | 1.19 ±0.01 ^{ad} | 1.21 ±0.01 ^a | 1.43 ±0.02 ^b | 1.12 ±0.02 ^c | 1.15 ±0.01 ^{cd} |
| CPE (%) | 21.78 ±0.19 ^a | 23.17 ±0.19 ^b | 31.21 ±0.49 ^c | 21.05 ±0.32 ^a | 21.86 ±0.14 ^a |
| CFE (%) | 39.48 ±0.36 ^a | 45.13 ±0.36 ^b | 59.83 ±0.94 ^c | 38.35 ±0.58 ^a | 40.10 ±0.26 ^a |
| GEE (%) | 19.25 ±0.17 ^{ad} | 22.07 ±0.18 ^b | 29.35 ±0.46 ^c | 19.04 ±0.29 ^d | 20.14 ±0.13 ^a |
| Survival (%) | 97.78 ±0.00 ^a | 97.78 ±2.22 ^a | 98.89 ±1.11 ^a | 96.67 ±1.11 ^a | 95.56 ±0.00 ^a |

Values are means (± SE); means within a row having different superscripts are significantly different (P<0.05).

SGR: Specific Growth Rate = (In Final weight – In Initial weight)/days X 100. FCR: Feed Conversion Ratio = Feed intake/ Weight gain. FCE: Feed Conversion Efficiency = (Weight gain/ Feed intake) X 100. PER: Protein Efficiency Ratio = Weight gain/ Protein intake. CPE: Crude Protein Efficiency = (Fish protein gain/ Protein intake) x 100. CFE: Crude Fat Efficiency = (Fish fat gain/Fat intake) x 100. GEE: Gross Energy Efficiency = (Fish gross energy gain/ gross energy intake) x 100

Table 4. Body indices of deccan mahseer fed on experimental diets. Symbols of experimental diets are as given in Table 1.

| | Diets | | | | |
|---------|------------------------|------------------------|------------------------|------------------------|-------------------------|
| | D1 | D2 | D3 | D4 | D5 |
| HSI (%) | 1.09±0.03 ^a | 1.10±0.03 ^a | 1.08±0.06 ^a | 0.89±0.04 ^b | 1.00±0.04 ^{ab} |
| VSI (%) | 3.88±0.07 ^a | 4.46±0.06 ^b | 5.07±0.06 ^c | 3.73±0.09 ^a | 4.14±0.06 ^d |
| RSI (%) | 0.54±0.02 ^a | 0.57±0.04 ^a | 0.55±0.02 ^a | 0.39±0.03 ^b | 0.52±0.02 ^a |
| CSI (%) | 0.55±0.03 ^a | 0.34±0.01 ^b | 0.35±0.02 ^b | 0.45±0.03 ^c | 0.44±0.01 ^c |

Values are means (\pm SE); means within a row having different superscripts are significantly different ($P < 0.05$).

HSI: Hepato-somatic index = (Weight of liver/weight of fish) x 100. VSI: Viscero-somatic index = (Weight of viscera/Weight of fish) x 100. RSI: Reno-somatic index = (Weight of kidney/Weight of fish) x 100. CSI: Cranio-somatic index = (Weight of brain/ Weight of fish) x 100.

Table 5. Body composition and muscle RNA/DNA ratio of deccan mahseer fed on experimental diets. Symbols of experimental diets are as given in Table 1.

| | Diets | | | | |
|------------------------------|-------------------------|-------------------------|-------------------------|-------------------------|--------------------------|
| | D1 | D2 | D3 | D4 | D5 |
| Moisture (%) | 69.42±0.15 ^a | 66.69±0.11 ^b | 62.29±0.14 ^c | 68.62±0.17 ^d | 67.84±0.24 ^e |
| Protein (%) | 18.52±0.11 ^a | 19.11±0.09 ^b | 21.79±0.07 ^c | 18.66±0.15 ^d | 18.96±0.09 ^{bd} |
| Lipid (%) | 9.05±0.06 ^a | 10.52±0.05 ^b | 12.03±0.04 ^c | 9.28±0.08 ^d | 9.72±0.04 ^e |
| Energy (kJ·g ⁻¹) | 7.64±0.01 ^a | 8.41±0.03 ^b | 9.60±0.02 ^c | 7.83±0.01 ^d | 8.06±0.02 ^e |
| RNA (mg·g ⁻¹) | 0.61±0.01 ^a | 0.98±0.04 ^{bd} | 1.03±0.01 ^b | 0.80±0.004 ^c | 0.93±0.01 ^d |
| DNA (mg·g ⁻¹) | 0.38±0.004 ^a | 0.27±0.01 ^b | 0.25±0.01 ^b | 0.34±0.01 ^c | 0.32±0.01 ^d |
| RNA/DNA | 1.59±0.01 ^a | 3.68±0.05 ^b | 4.13±0.05 ^c | 2.37±0.03 ^d | 2.96±0.05 ^e |

Values are means (\pm SE); means within a row having different superscripts are significantly different ($P < 0.05$).

Discussion

The supplementary feed of appropriate quality and slow growth rate of the species are recorded as major constraints in the culture of the indigenous commercially important deccan mahseer. In this context the present study was undertaken to evaluate some potential dietary supplements-cholicalciferol, soylecithin, thyroxine and betaine which can potentiate the overall growth of deccan mahseer in presence of natural food at on-farm level. The results of the present study indicate that, for the different treat-

ments, growth and survival were highest in fish fed on diet supplemented with soylecithin followed by fish fed on diet supplemented with cholicalciferol, and betaine. Weight gain and SGR were also significantly higher in fish fed with soylecithin supplemented diet than that of fish fed on all other diets. In fish fed with thyroxine supplemented diet there was no significant difference in weight gain and SGR compared to that of fish fed with the control diet. It has been reported that dietary supplementation of soylecithin enhanced body weight gain and survival in *Oncorhynchus mykiss* (Poston 1990a), *Salmo salar* (Poston 1990b), *Oplegnathus fasciatus* and *Paralichthys olivaceus* (Kanazawa 1993), *Cyprinus carpio* (Geurden et al. 1995), *Sciaenops ocellatus* (Craig and Gatlin 1997) and *Scophthalmus maximus* (Geurden et al. 1998). Andrews et al. (1980) observed that in *Ictalurus punctatus* there was supplemental requirement of cholicalciferol for maximal growth and cholicalciferol deficiency caused marked decrease in weight gain in *Sciaenops ocellatus* (Craig and Gatlin 1997). Contrarily, Horvli et al. (1998) and Vielma et al. (1999) were of the view that growth of fish was not influenced by dietary cholicalciferol. Clarke et al. (1994), Castro et al. (1998) and Fredette et al. (2000) observed that supplementation of FinnStim, a betaine-amino acid mixture successfully enhanced growth and also survival of several cultured marine finfish species. However, thyroxine was reported to be effective in reducing the hatching period, accelerating the yolk absorption, improving the growth and survival of post embryonic and larval stages in *Brachydanio rerio*, *Cyprinus carpio*, *Labeo rohita*, *Cirrhinus mrigala*, *Catla catla*, *Oreochromis mossambicus* and *Oreochromis niloticus* (Lam 1980; Nacario 1983; Sawant and Belsare 1994).

Feed supplements are being used for growth promotion in fishes and are found to have profound influence on digestion and assimilation of feed and consequently FCR, FCE and PER. In deccan mahseer FCR, FCE and PER were significantly better for soylecithin supplemented diet and were significantly poor for thyroxine supplemented diet compared to all other diets. However, there was no significant difference in these parameters of the fish fed with control and cholicalciferol supplemented diets, but for fish fed with betaine supplemented diet these parameters were lower than those of fish fed with control diet and diets supplemented with cholicalciferol and soylecithin. The results of the present study were found to be in agreement with the results of other investigators. Supplemental soylecithin exhibited better feed conversion in *Oncorhynchus mykiss* (Poston 1990a), *Salmo salar* (Poston 1990b), *Oplegnathus fasciatus* (Kanazawa 1993) and *Sciaenops ocellatus* (Craig and Gatlin 1997). Cholicalciferol

deficiency caused marked decrease in feed efficiency of *Oncorhynchus mykiss* (Barnett et al. 1979), *Ictalurus punctatus* (Andrews et al. 1980) and hybrid tilapia, *Oreochromis niloticus* x *O. aureus* (Shiau and Hwang 1993). Betaine was effective in eliciting enhance feed intake in several cultured marine fin fish species (Clarke et al. 1994; Castro et al. 1998; Fredette et al. 2000). James and Sampath (1994) found that thyroxine gradually reduced total feed intake in *Oreochromis mossambicus*.

Growth signifies change in magnitude. Hence, the organ's weights are related to growth in fish (Adams and McLean 1985). HSI of fish did not vary significantly with treatments, while VSI was significantly higher in fish fed soylecithin supplemented diet. Further, VSI was significantly higher in fish fed cholicalciferol and betaine supplemented diets compared to fish fed control diet. However, Craig and Gatlin (1997) reported that *Sciaenops ocellatus* fed with diet containing lecithin had significantly higher HSI and Andrews et al. (1980) found that *Ictalurus punctatus* fed with diet lacking in supplemental vitamin D had poor HSI values. Moreover, HSI were found to be good predictors of growth rate in *Gadus morhua* (Holdway and Beamish 1984) and *Micropterus salmoides* (Adams and McLean 1985). In fish fed diets supplemented with thyroxine HSI and VSI were lower. These findings are in agreement with Kang and Chang (1997) study on juvenile *Aathopagus schlegeli*. Mobilization of fat from liver and viscera to muscle reduced HSI and VSI (Lone and Matty 1982). RSI of treated fish was not significantly different from that of control fish, except for fish fed thyroxine supplemented diet, in which RSI was significantly lower than that of the fish fed all other feeds. The lower values of RSI may be due to the mobilization of mesentric and perinephric lipids (Lanari et al. 1999). CSI was found to be inversely related to the growth of the fish (Lanari et al. 1999). CSI was higher in fish of smaller size and decreased with increase in size of fish. CSI was significantly lower in fish fed on supplemented diets compared to fish fed with control diet. Moreover, CSI was significantly lower in fish fed cholicalciferol and soylecithin supplemented diets than the fish fed thyroxine and betaine supplemented diets. Similarly in *Cyprinus carpio* the CSI declined consistently in response to a dietary growth promoter, which was in turn found to enhance overall growth of the fish (Lone and Matty 1983).

In the current trials the dietary supplements were found to be responsible for the assimilation of protein, lipid, ash and gross energy in fish because the proximate composition and gross energy contents of the different test diets were similar and the experimental conditions were also same. The whole body moisture content of deccan mahseer for all the treatments

exhibited inverse relationship with the whole body lipid and protein contents (Love 1970; Lanari et al. 1999). The whole body protein, lipid, ash and gross energy contents were significantly higher in fish fed with soylecithin supplemented diet compared to the fish fed with all other diets. Fish fed with choliciferol supplemented diet had next higher levels of these biochemical parameters followed by fish fed diets supplemented with betaine and thyroxine. These biochemical parameters were lowest in fish fed with control diet. Poston (1990a; 1990b) and Hung et al. (1997) also reported that supplemental soylecithin significantly increases whole body lipid, protein and dry matter in *Oncorhynchus mykiss* and *Salmo salar*.

Muscle the most representative tissue of growth, shows the highest efficiency of protein deposition for growth (Peragon et al. 1999). The total quantity of DNA per cell is constant in normal somatic tissue within a given species and this amount is apparently not altered by starvation or other stress (Bulow 1987). The quantity of RNA varies directly with the activity of protein synthesis, and therefore, it is expected to be more concentrated in tissues undergoing faster growth or protein synthesis. Since the amount of DNA per cell is constant within a species, the ratio of RNA/DNA is usually considered a more accurate index of protein synthesis activity than RNA concentration alone, because the ratio is not affected by differences in cell number (Bulow 1987). The muscle RNA/DNA ratio in the present study showed a direct relationship with fish growth. The RNA/DNA ratio was significantly higher in fish fed soylecithin supplemented diet followed by fish fed choliciferol, betaine and thyroxine supplemented diets compared to fish fed control diet.

The nutrient and energy efficiencies of fish were found to be influenced by the dietary micronutrients and feed additives, however, they were independent of the dietary fat, protein and NFE contents (Jover et al. 1999; Lanari et al. 1999; Nordrum et al. 2000). The crude protein, crude fat and gross energy efficiencies were significantly higher in fish fed soylecithin supplemented diet followed by fish fed diet supplemented with choliciferol than fish fed all other diets. This brings forth that dietary supplements though added in minor quantities enhances the nutrient and gross energy efficiencies of fishes to a greater extent than those achieved by increasing the levels of macronutrients in feed.

The feeding trials conducted to evaluate the possibilities of including dietary supplements viz., choliciferol, soylecithin, thyroxine and betaine in deccan mahseer diet, inferred that the growth performance and feed efficiency of deccan mahseer under semi-intensive culture system was

significantly affected by the type of dietary supplement and hence they can be of interest to the farming industry.

Acknowledgement

The authors are thankful to the Director, National Research Centre on Coldwater Fisheries, Bhimtal for granting permission to carry out the study and Director, Central Institute of Fisheries Education (Deemed University), Mumbai for providing all the facilities to conduct the experiments. The authors also acknowledge with gratitude the assistance rendered by the staff of Central Institute of Fisheries Education (Deemed University), Mumbai for the study.

References

- Adams, S.M. and R.B. McLean. 1985. Estimation of largemouth bass, *Micropterus salmoides* Lacepede, growth using the liver somatic index and physiological variables. *Journal of Fish Biology* 26:111-126.
- Adoni, A.D. 1985. Workbook on limnology. Indian MAB Committee, Department of Environment, Government of India. Pratibha Publishers, Sagar, India. 216 p.
- Andrews, J.W., T. Murai and J.W. Page. 1980. Effects of dietary cholecalciferol and ergocalciferol on catfish. *Aquaculture* 19:49-54.
- Anon. 1976. Report of the National Commission on Agriculture (Fisheries). Government of India, New Delhi, pp. 8-10.
- AOAC. 1995. Official Methods of Analysis, 16th ed. Association of Official Analytical Chemists, Washington, DC, USA.
- APHA. 1998. Standard Methods for the Examination of Water and Wastewater, 20th ed. American Public Health Association, Washington, DC, USA.
- Barnett, B.J., C.Y. Cho and S.J. Slinger. 1979. The essentiality of cholecalciferol in the diets of rainbow trout (*Salmo gairdneri*). *Comparative Biochemistry and Physiology* 63A:291-297.
- Basade, Y. 2001. Evaluation of the nutritional role of some growth promoters and possibilities of their utilization in mahseer culture. Ph.D. Thesis. Central Institute of Fisheries Education (Deemed University), Mumbai, India. 142 p.
- Basade, Y. and M.P.S. Kohli. 2004. Influence of dietary cholecalciferol levels on its digestibility in deccan mahseer, *Tor khudree* (Sykes). *Indian Journal of Fisheries* 51(2):123-131.
- Bulow, F.J. 1987. RNA-DNA ratios as indicators of growth in fish: A review. In: Age and Growth of Fish (ed. R.C. Summerfelt and G.E. Hall), pp. 45-64. Iowa State University Press, Ames, Iowa.
- Castro, H., J. Battaglia and E. Virtanen. 1998. Effects of FinnStim on growth and seawater adaptation of coho salmon. *Aquaculture* 168:423-429.

- Chauhan, D.P.S., R.S. Chauhan, P.V. Dehadrai, G.P. Dubey, K. Kumar, M. Mohan, P.C. Mahanta and D.N. Sarangi. 2007. Conservation and management. In: Art and Science of Mahseer Conservation and Management, Indian Fisheries Association and Central Institute of Fisheries Education, Mumbai, India, pp. 5-29.
- Clarke, W.C., E. Virtanen, J. Blackburn and D.A. Higgs. 1994. Effects of a dietary betaine/ amino acid additive on growth and seawater adaptation in yearling chinook salmon. *Aquaculture* 121:137-145.
- Cordington, K.D.B. 1939. Notes on Indian mahseer. *Journal of Bombay Natural History Society* 46:336-344.
- Craig, S.R. and D.M. Gatlin III. 1997. Growth and body composition of juvenile red drum (*Sciaenops ocellatus*) fed diets containing lecithin and supplemental choline. *Aquaculture* 151:259-267.
- De Silva, S.S. 1993. Supplementary feeding in semi-intensive aquaculture system. In: Farm-made Aquafeeds (ed. M.B. New, A.G.J. Tacon and I. Csavas), pp. 24-60. AADCP Regional Expert Consultative Consultation on Aquafeeds, 14-18 December 1992, Bangkok, Thailand. FAO-RAPA/AADCP, Bangkok, Thailand.
- De Silva, S.S. and F.B. Davy. 1992. Fish nutrition research for semi-intensive culture system in Asia. *Asian Fisheries Science* 5:129-144.
- FAO. 2007. Year book. Fishery Statistic, Aquaculture Production. Food and Agriculture Organization of the United Nations, Rome.
- Fredette, M., J. Batt and J. Castell. 2000. Feeding stimulant for juvenile winter flounders. *North American Journal of Aquaculture* 62:157-160.
- Geurden, I., J. Radunz-Neto and P. Bergot. 1995. Essentiality of dietary phospholipids for carp (*Cyprinus carpio* L.) larvae. *Aquaculture* 131:303-314.
- Geurden, I., O.S. Reyes, P. Bergot, P. Coutteau and P. Sorgeloss. 1998. Incorporation of fatty acids from dietary neutral lipid in eye, brain and muscle of postlarval turbot fed diets with different types of phosphatidylcholine. *Fish Physiology and Biochemistry* 19:365-375.
- Hardy, R.W. 1989. Diet preparation. In: *Fish Nutrition*, 2nd ed. (ed. J.E. Halver), pp. 475-548. Academic Press, Inc., San Diego, California.
- Holdway, D.A. and F.W.H. Beamish. 1984. Specific growth rate and proximate body composition of Atlantic cod (*Gadus morhua* L.). *Journal of Experimental Marine Biology and Ecology* 81:147-170.
- Horvli, O., O. Lie and L. Aksnes. 1998. Tissue distribution of vitamin D₃ in Atlantic salmon *Salmo salar*: effect of dietary level. *Aquaculture Nutrition* 4:127-131.
- Hung, S.S.O., G.M. Berge and T. Storebakken. 1997. Growth and digestibility effects of soylecithin and choline chloride on juvenile Atlantic salmon. *Aquaculture Nutrition* 3:141-144.
- Islam, M.S. 2002. Evaluation of supplementary feeds for semi-intensive pond culture of mahseer, *Tor putitora* (Hamilton). *Aquaculture* 212:263-276.
- James, R. and K. Sampath. 1994. Effect of L. thyroxin on food consumption and conversion in *Oreochromis mossambicus*. *Indian Journal of Fisheries* 41:76-79.
- Jhingran, V.G. and K.L. Sehgal. 1978. *Coldwater Fisheries of India*. Inland Fisheries Society of India, Barrackpore, West Bengal, India. 239 p.
- Jover, M., A. Garcia-Gomez, A. Tomas, F. de la Gandara and L. Perez. 1999. Growth of Mediterranean yellowtail (*Seriola dumerilli*) fed extruded diets containing different levels of protein and lipid. *Aquaculture* 179:25-33.
- Kanazawa, A. 1993. Essential phospholipids of fish and crustaceans. In: *Fish Nutrition in Practice* (ed. S.J. Kaushik and P. Luquet), pp. 519-530. Proceedings of the IVth

- International Symposium on Fish Nutrition and Feeding, Les Colloques No.61, INRA, Paris.
- Kang, D.Y. and Y.J. Chang. 1997. Effects of exogenous thyroid hormone (T₃) on skeletal development and physiological conditions of juvenile black seabream (*Acanthopagrus schlegeli*). Journal of Korean Fisheries Society 30:305-312.
- Keshavanath, P. 2000. Nutritional studies on mahseer, *Tor khudree* (Sykes). In: Coldwater Aquaculture and Fisheries (ed. H.R. Singh and W.S. Lakra), p. 219-228. Narendra Publishing House, Delhi, India.
- Keshavanath, P., M. Mohan and Y. Basade. 2007. Nutrition and feeding. In: Art and Science of Mahseer Conservation and Management, Indian Fisheries Association and Central Institute of Fisheries Education, Mumbai, India, pp. 79-93.
- Kohli, M.P.S., S. Ayyappan, R.K. Langer, S.N. Ogale, K. Dube, C. Prakash and A.K. Reddy. 2005. Cage culture of mahseer, *Tor khudree* (Sykes) and *Tor putitora* (Ham.) at Walwan reservoir, Lonavla, Maharashtra. In: Proceedings National Seminar on Aquatic Resource Management in Hills (ed. K.K. Vass, S.A.H. Abidi and V.P. Agrawal), pp. 171-178. National Research centre on Coldwater Fisheries, Bhimtal, India.
- Kulkarni, C.V. 2000. Artificial propagation of *Tor khudree* (Sykes) and *Tor tor* (Ham.). In: Coldwater Aquaculture and Fisheries (ed. H.R. Singh and W.S. Lakra), pp. 203-218. Narendra Publishing House, Delhi, India.
- Lam, T.J. 1980. Thyroxine enhances larval development and survival in *Sarotherodon* (*Tilapia*) *mossambicus* Rupell. Aquaculture 21:287-291.
- Lanari, D., B.M. Poli, R. Ballestrazzi, P. Lupi, E. D'Agaro and M. Mecatti. 1999. The effect of dietary fat and NFE levels on growing European sea bass (*Dicentrarchus labrax* L.). Growth rate, body and fillet composition, carcass traits and nutrient retention efficiency. Aquaculture 179:351-364.
- Lone, K.P. and A.J. Matty. 1982. The effect of feeding 11-ketotestosterone on the food conversion efficiency and tissue protein and nucleic acid contents of juvenile carp, *Cyprinus carpio* L. Journal of Fish Biology 20:93-104.
- Lone, K.P. and A.J. Matty. 1983. The effect of ethylestrenol on the growth, food conversion and tissue chemistry of the carp, *Cyprinus carpio*. Aquaculture 32:39-55.
- Love, R.M. 1970. The Chemical Biology of Fishes. Academic Press, Inc. Ltd., London, 547 p.
- Mayes, P.A. 1990. Nutrition. In: Harper's Biochemistry. 22nd ed. (ed. R.K. Murray, D. K. Granner, P. A. Mayes and V.W. Rodwell), pp. 571-579. Prentice – Hall International Inc., USA.
- Mirza, M.R. 1994. A note on the fishes of the river Hingol, Pakistan. Pakistan Journal of Biology 26(2):181.
- Mirza, M.R. and M.S. Khan. 1994. A note on the fishes of the Badri stream near Swabi, Northwest Frontier Province, Pakistan. Pakistan Journal of Zoology 26(4):361.
- Mirza, M.R., M.N. Javed and M. Tariq. 1994. A note on the fish fauna of the river Zhob, Pakistan. Pakistan Journal of Biology 26(2):189.
- Montgomery, D.C. 1991. Design and analysis of experiments, 3rd ed. John Wiley and Sons, Inc., Singapore. 649 p.
- Nacario, J.F. 1983. The effect of thyroxine on the larvae and fry of *Sarotherodon niloticus* (*Tilapia nilotica*). Aquaculture 34:73-83.
- Nautiyal, P. 1994. Mahseer the Game Fish (Natural history, status and conservation practices in India and Nepal). Published by Rachna, Srinagar (Garhwal), U.P.

- Nordrum, S., A. Krogdahl, C. Rosjo, J.J. Olli and H. Holm. 2000. Effects of methionine, cysteine medium chain triglycerides on nutrient digestibility, absorption of amino acids along the intestinal tract and nutrient retention in Atlantic salmon (*Salmo salar* L.) under pair-feeding regime. *Aquaculture* 186:341-360.
- Papatryphon, E. and J.H. Soares Jr. 2000. The effect of dietary feeding stimulants on growth performance of striped bass, *Morone saxatilis*, fed-a-plant feedstuff based diet. *Aquaculture* 185:329-338.
- Peragon, J., J.B. Barroso, L. Garcia-Salguero, M. de La Higuera and J.A. Lupianez. 1999. Carbohydrate affect protein-turnover rates, growth, and nucleic acid content in the white muscle of rainbow trout (*Oncorhynchus mykiss*). *Aquaculture* 179:425-437.
- Poston, H.A. 1990a. Performance of rainbow trout fry fed supplemental soylecithin and choline. *Progressive Fish-Culturist* 52:218-225.
- Poston, H.A. 1990b. Effect of body size on growth, survival and chemical composition of Atlantic salmon fed soylecithin and choline. *Progressive Fish-Culturist* 52:226-230.
- Sawant, N.H. and S.G. Belsare. 1994. Effect of thyroxine on hatching and on postembryonic growth in *Brachydanio rerio*, *Cyprinus carpio*, *Labeo rohita*, *Cirrhinus mrigala* and *Catla catla*. *Journal of Indian Fisheries Association* 24:133-137.
- Schneider, W.C. 1966. Determination of nucleic acids in tissues by pentose analysis. In: *Methods in Enzymology*, Vol.III (ed. S.P. Colowick and N.O. Kaplan), pp. 680-684. Academic Press, Inc., New York.
- Sehgal, K.L. and D.S. Malik. 1992. *Aquaculture of Tor putitora vis a vis* stock enhancement and management of fisheries in the Himalayas. *Punjab Fisheries Bulletin* 16:55-63.
- Sen, T.K. and K.C. Jayaram. 1982. Mahseer Fishes of India-A Review Record of Zoological Survey of India, Occupational Paper 39:1-34
- Shiau, S.Y. and J.Y. Hwang. 1993. Vitamin D requirement of juvenile hybrid tilapia *Oreochromis niloticus* X *O. aureus*. *Nippon Suisan Gakkaishi* 59:553-558.
- Shrestha, T.K. 1994. Migration and spawning of golden mahseer in Himalayan waters of Nepal. *Journal of Freshwater Biology* 6(1):71-77.
- Singh, C.S. 1992. Golden mahseer, *Tor putitora* (Ham.) holds promises in Tarai waters. *Punjab Fisheries Bulletin* 16:17-20.
- Snedecor, G.W. and W.G. Cochran. 1994. *Statistical Methods*, 8th ed. Affiliated East-West Press Pvt. Ltd., New Delhi. 503 p.
- Sunder, S., H.S. Raina, M. Mohan and C.B. Joshi. 1995. Cultural possibilities of golden mahseer *Tor putitora* (Ham.) in Himalayan uplands. *Uttar Pradesh Journal of Zoology* 15:177-181.
- Vielma, J., S.P. Lall, J. Koskela and P. Mattila. 1999. Influence of low dietary cholecalciferol intake on phosphorus and trace element metabolism by rainbow trout (*Oncorhynchus mykiss*, Walbaum). *Comparative Biochemistry and Physiology* 122A:117-125.