Fishery Technology
2010, Vol. 47(2) pp: 133 - 138

Effects of Varying Levels of Dietary Protein on Growth and Reproductive Potential in *Barilius bakeri*

Jomol Antony[1] and P. Natarajan[2]
Department of Aquatic Biology and Fisheries, University of Kerala, Trivandrum, India

A 90 day experiment was conducted to observe the influence of varying levels of dietary protein on the growth and reproductive potential of *Barilius bakeri*. Diets were prepared with fish meal as the single source of protein. A total of twelve fishes weighing 1.6 ± 0.2 g were stocked in glass tanks in triplicate for each of the treatment. Fishes were fed with 20, 30, 40 and 50% crude protein, twice daily. There was no significant weight increment between the different protein groups. Gonadosomatic index was significantly higher ($p<0.05$) for 30% and lowest for 20% group. Fecundity was highest for 30% ($p<0.05$) and least for 20%. For *Barilius bakeri*, a dietary protein level between 30 and 40% was found to be optimum with regard to growth and reproduction.

Key words: Feed, protein level, growth, *Barilius bakeri*

Export of ornamental fishes is assuming exceptional significance in recent years. About 85% of fishes exported from India are indigenous ornamental fishes (Kurup & Radhakrishnan, 2006). Eventhough the breeding aspects of indigenous ornamental fishes have been studied (Mercy, 2006), nutritional requirements of the same have not been well understood. Dietary protein is the main component that assists growth in fishes. Protein content usually constitutes the single largest cost factor in fin fish feeds (Watanabe, 2002). Protein is the most relevant parameter to express growth effect of specific feed formula (Satpathy *et al.*, 2003). There is an optimal protein level for reproductive success and this level is related to the growth of the concerned species (De Silva & Anderson, 1995). The present study was undertaken to investigate the effects of varying levels of protein on the growth and reproductive potential of *Barilius bakeri*, an indigenous ornamental fish with good export market.

Materials and Methods

Juveniles of *B. bakeri* weighing (1.6 ± 0.2 g) were collected from river Kallar, a hill stream near Ponmudi hills, Kerala and were stocked in large fibre glass tanks for a week for acclimatization. The fishes were then transferred to aged tapwater in glass tanks of 180 litre capacity connected to a biological filter. A total of twelve fishes were stocked in each tank and a triplicate was maintained for each treatment. Fishes were fed at 5% level of body weight twice daily at 10.00 and 16.00 h. Half of the water in the tank was exchanged weekly and water quality parameters were monitored daily throughout the experimental period. Water temperature was recorded with a centigrade mercury thermometer and pH using a digital pH meter. Dissolved oxygen, total alkalinity, ammonia, nitrite and nitrate were estimated following standard methods of APHA (1992) and were monitored throughout the experimental period.

[1] Corresponding author; email: jomolz@gmail.com
[2] Rajiv Gandhi Chair in Contemporary Studies, Cochin University of Science and Technology, Cochin - 682 022, India

Dried and powdered anchovy (fish meal), groundnut oil cake (GOC), tapioca powder (TP) and rice bran (RB) procured from a local market, were used for preparation of the feed. Vitamins and mineral premix were added to the feed. Experimental diets were formulated according to the method of Hardy (1980) to contain varying levels *viz.,* 20, 30, 40 and 50% of crude protein.

Proximate composition of feed ingredients and experimental diets were analyzed using standard methods (AOAC, 1995). Moisture was determined by drying the sample at 105^{0}C to constant weight. Total nitrogen was estimated using Kjeldahl method and the protein was computed by multiplying the nitrogen value with 6.25. Crude fat was estimated by Soxhlet exhaustive extraction technique using petroleum ether (40-60^{0}C, BP) as solvent. Ash was determined by incinerating the dried sample at 550^{0}C for 12 h. At the termination of the experiment, the body weight, gonad weight and fecundity of the fishes were recorded. Fecundity and gonadosomatic index (GSI) were determined by following the method of Biswas (1993).

Data from triplicate glass tanks were analyzed to find out the difference between different dietary treatments by ANOVA using SPSS version 10.1. The results were compared using Duncan's multiple range test at 5% significance level.

Results and Discussion

The mean water quality parameters estimated during the experiment are given in Table 1. The proximate composition of experimental diets and the proportion of protein in feed ingredients of the various diets, are given in Table 2 and 3 respectively.

After 90 days of experiment, significant weight increment could be detected between different protein levels ($p< 0.05$).

Table 1. Mean water quality parameters recorded

Parameters	Mean ± SD
Temperature (°C)	28.20 ± 0.45
pH	7.00 ± 0.08
Dissolved oxygen (mg/l)	5.07 ± 0.35
Total alkalinity (mg/l)	29.20 ± 1.79
Ammonia (mg/l)	0.27 ± 0.14
Nitrite (mg/l)	2.23 ± 0.28
Nitrate (mg/l)	1.36 ± 0.13

Weight gain, GSI and fecundity of *B. bakeri* fed with varying levels of dietary protein are given in Table 4. Fig. 1 shows the growth pattern of *B. bakeri* fed with experimental diets during the 90 day study period. There was no significant weight increment between different protein levels, though 40% group showed the highest mean followed by 30, 50 and 20% group. GSI was highest for 30% group and least for 20% group. There was no significant difference in GSI between 30% and 40% group. GSI was significantly lower for 20% and 50% group, ($p<0.05$). Higher fecundity was noticeable for the 30% and 40% group, 30% showing the highest value. The 20% and 50% groups showed the least fecundity with no significant difference between the two.

There was significant difference in the survival between the different groups ($p< 0.01$).

Growth in fishes is affected by dietary protein level. Gunasekara & Lam (1997) reported that brood stock of *Oreochromis niloticus* fed with 10% protein diet attained less weight than those fed with 20 and 30% diets. They found no difference in growth of fish receiving 20 and 30% protein diet. In the present work on *B. Bakeri,* weight increment was noticeable up to 40% dietary protein beyond which growth showed slight decline. According to Jauncey (1982), protein levels above optimum may result in decreased growth rates because of a reduction in dietary energy due to the energy requirement to deaminate and excrete amino acids

Table 2. Proximate composition of feed ingredients (% dry weight basis)

Feed ingredients	Moisture (%)	Protein (%)	Carbohydrate (%)	Lipid (%)	Ash (%)	Fibre (%)
TF	12.05 ± 0.60	5.82 ± 0.29	80.00 ± 3.75	2.05 ± 0.10	0.53 ± 0.03	1.52 ± 0.08
RB	7.04 ± 0.35	10.55 ± 0.53	37.47 ± 1.57	3.35 ± 0.17	20.62 ± 1.03	21.92 ± 1.10
GOC	4.97 ± 0.24	47.91 ± 1.45	30.40 ± 1.52	6.92 ± 0.96	9.58 ± 0.48	3.62 ± 0.18
FM	5.73 ± 0.64	54.03 ± 3.63	5.72 ± 0.59	6.72 ± 0.57	25.65 ± 0.67	6.60 ± 0.48

TF-Tapioca flour, RB-Rice bran, GOC-Groundnut oil cake, FM-Fishmeal

absorbed in excess. Reduced feed intake was observed in the 50% group compared to other groups. This has been reported for other fishes like *Colossoma macropomum* (Castagnolli, 1991., Vander Meer *et al.,* 1995) and *Ictalurus punctatus* (Page & Andrews, 1973, Mangalik, 1986). It has also been reported that crude protein content in carps is about 40% and a protein level in excess of 45% is undesirable as it will not be utilized by them (Watanabe *et al.,* 1987). Further, Ogino (1980) reported an optimum protein content in the range of 35-50% in the diets of carp and rainbow trout.

Cumaratunga & Mallika (1991) reported that GSI is not affected by variation in dietary protein in *Oreochromis niloticus*. In contrast, Kiron Vasudevan (1996) reported higher GSI in *O. mossambicus* fed with 30% protein feed and then a decline at 40% and 50% levels. The results of the present study also indicated that dietary protein influenced GSI of fish as reported by Dahlgren (1980). GSI in *Rhamdia quelen* juveniles was highest for 35% protein level group compared to 20 and 30% groups (Reidal *et al.,* 2009). Higher GSI was also reported in *Leptobarbus hoevenii* fed with 32% and 42% protein diets (Pathmasothy,1983). Lower GSI in 20% group, as observed in the present study, may be due to lower protein content in the diet leading to decreased growth and eventually lower GSI compared to other groups. On the contrary, a noticeable decline in GSI value in 50% group can be attributed to reduced feed intake. Further, if a diet is not balanced with an adequate supply of non protein energy, the dietary protein may be catabolised for energy, resulting in decreased growth and reduced GSI (Kaushik & Luquet, 1984; Lovel, 1988).

The diets containing 30 and 40% of protein produced higher fecundity (Table 4). Fecundity is found to be affected by nutritional level in brood stock diet (Izquierdo *et al.,* 2001). Manissery *et al.* (2001) reported maximum fecundity in *Cyprinus carpio* at 35% protein. Similarly, a 45% protein feed enhanced the total number of eggs per Nile Tilapia (Siddiqui *et al.,*1998). Santiago *et al.* (1991) reported higher fecundity in *Arischthys nobilus* fed with 40% protein than with 20% dietary protein level. It was suggested by Chong *et al.* (2004) that a minimum of 30% protein level was required for an effective fecundity in *Xiphophorus helleri.* Higher fecundity in *O. nilotius* and *O. mossambicus* have also been reported when fed with less than 30% protein feed (De Silva & Radampola, 1990; Kiron Vasudevan, 1996). In contrast, dietary protein level did not affect relative fecundity in *O. niloticus* and *Poecilia reticulata*

Table 3. The proportion of protein in various ingredients in the experimental diets

Feed Ingredients	Protein level (%)			
	20	30	40	50
RB	40.14 ± 2.01	35.04 ± 1.75	29.93 ± 1.50	24.82 ± 1.24
TF	40.14 ± 2.01	35.04 ± 1.75	29.93 ± 1.50	24.82 ± 1.24
GOC	9.85 ± 0.49	14.96 ± 0.75	20.07 ± 1.00	25.18 ± 1.26
FM	9.85 ± 0.49	14.96 ± 0.75	20.07 ± 1.00	25.18 ± 1.26
Crude protein	19.60 ± 0.98	30.00 ± 1.50	39.80 ± 1.00	50.00 ± 2.50

TF-Tapioca flour, RB-Rice bran, GOC-Groundnut oil cake, FM-Fishmeal

Table 4. Mean values of weight, GSI, fecundity and survival estimated in experiments with various formulated diets

Parameters	Treatments				
	F ratio	20% feed	30% feed	40% feed	50% feed
Initial weight (g)		1.69 ± 0.19	1.69 ± 0.15	1.69 ± 0.11	1.69 ± 0.03
Final weight (g)		3.70 ± 0.16	3.80 ± 0.22	3.90 ± 0.32	3.70 ± 0.15
Percentage gain in weight	2.432	124.24 ± 3.68	130.30 ± 3.86	136.36 ± 4.04	124.24 ± 3.68
GSI (%)	26.979**	3.80 ± 1.20^{a}	5.30 ± 0.20^{c}	5.10 ± 1.00^{c}	4.30 ± 1.10^{b}
Fecundity	6.660**	300 ± 17^{a}	348 ± 33^{b}	338 ± 40^{b}	304 ±15^{a}
Survival (%)	29.339**	87b	96a	96^{a}	79^{c}

a, b, c, d – Means with the same superscript do not differ significantly (Duncan's multiple range test)
** p<0.01

(Dahlgren, 1980; Santiago *et al.* 1983; Gunasekara *et al.*,1996). Dietary protein requirement for favourable reproductive performance in fresh water species was reported to be at 30-40% levels (Shim *et al*., 1989; Gunasekara *et al*., 1997; Al Hafedth *et al*., 1999; El Sayed *et al*., 2003; Chong *et al*., 2004). The present study also corroborate with their findings and it can be concluded that for *Barilius bakeri*, a dietary protein level between 30 and 40% was optimum with regard to growth and reproduction.

The authors are grateful to Prof. (Dr.) Tresa Radhakrishnan, Head, Department of Aquatic Biology and Fisheries, University of Kerala, Trivandrum for providing the necessary facilities to carry out the experiments.

References

AOAC (1995) *Official Methods of Analysis*, Association of Official Analytical Chemists Arlington, VA, USA

APHA (1992) *Standard Methods for Examination of Water and Waste Water,* 16th edn., American Public Health Association, Washington, DC, USA

Al Hafedth, Y. S., Siddiqui, A. Q. and Al-Saidy, M. Y. (1999) Effects of dietary protein levels on gonad maturisation, size and age at first maturity, fecundity and growth of Nile Tilapia, *Aquac. Int*. **7**, pp 319-332

Biswas, S. P. (1993) *Manual of Methods in Fish Biology*, South Asian Publishers Pvt Ltd, New Delhi, India

Castagnolli, N. (1991) Brazilian finfish, Tambaqui, Pacu and Matrinxa. In: *Handbook of Nutrient Requirements of Finfish* (Wilson, R.P., Ed), pp 31-34, CRC Press, Boca Raton. FL

Chong, A. S. C., DahlianisIshak, S., Osman, Z. and Hashim, R. (2004) Effect of dietary protein level on the reproductive performance of female swordtail *Xiphophorus helleri* (Poeciliidae) *Aquaculture*, **234**, pp 381-392

Cumaratunga, P. R. T. and Mallika, K. L. G. P. (1991) Effects of different levels of dietary protein and a legume *Vigna catiang* on gonadal development in *Oreochromis niloticus* (L.). In: *Fish Nutrition Research in Asia* (De Silva, S. S., Ed), pp 125-133, Proceedings of the Fourth Asian Fish Nutrition Workshop, Asian Fisheries Society Special Publication 5. Asian Fisheries Society, Manila, Philippines

Dahlgren, B. T. (1980) The effects of three different dietary protein levels on the fecundity in the guppy *Poecilia reticulate* (Peters) *J. Fish Biol*. **16**, pp 83-97

De Silva, S. S. and Radampola, K. (1990) Effect of dietary protein level on the reproductive performance of *Oreochromis niloticus*. In: *The Second Asian Fisheries Forum* (Hirano, R. and Hanyu, I., Eds), pp

559-563, Asian Fisheries Society, Manila, Philippines

De Silva, S. S. and Anderson, T. A. (1995) *Fish Nutrition Aquaculture*, Chapman and Hall Aquaculture Series 1, Chapman and Hall, London, UK 49 p

El-Sayed, A. M., Mansour, C. R. and Ezzat, A. A. (2003) Effects of dietary protein level on spawning performance of Nile Tilapia (*Oreochromis niloticus*) broodstock reared at different water salinities, *Aquaculture* **220**, pp 619-632

Gunasekhara, R. M. and Lam, T. J. (1997) Influence of dietary protein level on ovarian recrudescence in Nile tilapia, *Oreochromis niloticus* (L), *Aquaculture* **149**, pp 57-69

Gunasekhara, R. M., Shim, K. F. and Lam, T. J. (1996) Effect of dietary protein level on spawning performance and amino acid composition of eggs of Nile tilapia, *Oreochromis niloticus*, *Aquaculture* **146**, pp 121-134

Hardy, R. (1980) *Fish feed formulation*. Paper presented at the FAO/UNDP Training Course in Fish Feed Technology, Seattle, W A, USA., ADCP/REP/80, 11, pp, 8

Izquierdo, M. S., Fernandez Palacios, H. and Tacon, A. G. J. (2001) Effect of broodstock nutrition on reproductive performance of fish, *Aquaculture*. **197**, pp 25-42

Juancey, K. (1982) The effects of varying dietary protein level on the growth, food conversion, protein utilization and body composition of juvenile tilapias (*Sarotherodon mossambicus*), *Aquaculture* **27**, pp 43-54

Kaushik, S. J. and Luquet, P. (1984) Relationship between protein intake and voluntary energy intake as affected by body weight with an estimation of maintenance needs in rainbow trout, *Z. Tier-Physiol, Tierernähr. U. Futtermittlekd.* **51**, pp 57-69

Kiron Vasudevan (1996) *Studies on some aspects of the culture of Mozambique Tilapia*, University of Kerala, Trivandrum, Kerala

Kurup, B. M. and Radhahrishnan K. V. (2006) Indigenous ornamental fish resources of Western Ghats with special reference to Kerala. In: *Souvenir Ornamentals Kerala 2006* (Kurup, B. M., Pillai, N. G. K., Dileep, M. P., Boopendranath, M. R., Prasadachandrapillai, K., Nair, K. S., Mercy, T. V. A., Banu, S., Kurien, M. J., Sajeev, U. S., Thomas, J. V. and Radhakrishnan, K.V., Eds), pp 37-57, Department of Fisheries (Government of Kerala and Ministry of Agriculture, Govt of India), Thiruvananthapuram

Lovel, R. T. (1988) *Nutrition and Feeding of Fish*,Van Nostrand Reinhold, New York. pp 11-161

Mangalik, A. (1986) *Dietary Energy Requirements of channel catfish*. Auburn University, Auburn, Alabama, USA

Manissery, J. K., Krishnamurthy, D., Gangadhara. B. and Nandheesa, M. C. (2001) Effect of varied levels of dietary protein on the breeding performance of common carp *Cyprinus carpio, Asian Fish Sci*. **4**, pp 317-322

Mercy, T. V. A. (2006) Status of development of captive breeding technology for the indigenous ornamental fishes of the western ghats of India. In: *Souvenir Ornamentals Kerala 2006* (Kurup, B. M., Pillai, N. G. K., Dileep, M. P., Boopendranath, M.R., Prasadachandrapillai, K., Nair, K. S., Mercy, T. V. A., Banu, S., Kurien, M. J., Sajeev, U. S., Thomas, J. V. and Radhakrishnan, K.V., Eds), pp 71-75, Department of Fisheries (Government of Kerala and Ministry of Agriculture, Govt of India), Thiruvananthapuram

Ogino, C. (1980) Protein requirement of carp and rainbow trout, *Bull. Japan. Soc. Sci. Fish*. **46**, pp 385-391

Page J. W. and Andrews J. W. (1973) Interaction of dietary protein and energy in channel catfish *Ictalarus punctactus*, *J. Nutrition*, **103**, pp 1333-1346

Pathmasothy, S. (1983) A review of feed, their processing and feeding techniques in

Malaysian aquaculture system. In: *Proceedings of the international Conference on Development and Management of Tropical Living Aquatic Resources* (Chan, H. H., Ang., K .J, Ibrahim H. M, and Ambak M. A., Eds), pp 126-131, Universiti Pertanian Malaysia, Serdang, Selangor, Malaysia

Reidal, A., Boscolo, W.R., Feiden, A. and Romagosa, E. (2009) The effects of diets with different levels of protein and energy on the process of final maturation of the gametes of Rhamdia quelen stocked in cages, *Aquaculture* **298,** pp 354-359

Santiago, C. B., Aldaba, M. B. and Laron, M. A. (1983) Effect of varying dietary crude protein levels on spawning frequency and growth of *Sarotherodon niloticus* breeders, *Fish. Res. J. Philippines*. **8,** pp 9-18

Santiago, C. B., Camacho, A. S. and Laron, M. A. (1991) Growth and reproductive performance of bighead carp (*Aristichthys nobilis*) reared with or without feeding in floating cages, *Aquaculture* **96**, pp 109-117

Satpathy, B. B., Mukherjee, D. and. Ray, A. K. (2003) Effects of dietary protein and lipid levels on growth, feed conversion and body composition in rohu *Labeo rohita* (Hamilton) fingerlings, *Aquacult. Nutr*. **9**, pp 17-24

Shim, K. F., Landesman, L. and Lam, T. J. (1989) Effect of dietary protein on growth, ovarian development and fecundity in the dwarf gourami *Colisa lalia* (Hamilton) *J. Aquacult. Trop*. **4**, pp 111-123

Siddiqui, A. Q., Al-Hafedh,Y. S. and Ali, S. A. (1998) Effect of dietary protein level on the reproductive performance of Nile tilapia, *Orechrominiloticus (*L), *Aquacult. Res*. **29**, pp 349-358

Van der Meer, M., Machiels, M. A. M. and Verdegem, M. C. J. (1995) The effect of dietary protein level on growth, protein utilization and body composition of *Colossoma macropomum* (Cuvier), *Aquacult. Res.* **26**, pp 901-909

Watanabe, T., Takeuchi, T., Satoh, S., Ida T. and Yaguchi, M. (1987) Development of low protein high energy for practical carp culture with special reference to reduction of total nitrogen excretion, *Nippon Susan Gakkaishi.* **53**, pp 1513-1423

Watanabe, T. (2002) Strategies for further development of aquatic feeds, *Fish. Sci*. **68**, pp 242-252