PERFORMANCE OF RAINBOW TROUT (*Oncorhynchus mykiss*) FED ON DIFFERENT DIETARY PROTEIN WITH FIXED ENERGY RATIO

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Summary

An experiment was conducted on rainbow trout (*Oncorhynchus mykiss*) for eight weeks to investigate the growth performance of the fish fed with different dietary protein with constant diet energy of 20 kJg^{-1}. Four diets containing 25, 30, 35 and 40% crude protein were used. The highest mean final weight was obtained for the fish fed with diet having 35% protein. Grwth performance in terms of Specific Growth Rate (SGR), Food Conversion Ratio (FCR) and Protein Efficiency Ratio (PER) were calculated for each diet. There were no significant differences in SGR but the highest value was exhibited by fish fed with 35% protein diet. Significant differences were found among FCR of different diets. Diets with 35 and 40% crude protein gave better FCR value than that of 25 and 30% crude protein. Although significant differences were not found between PER of different diets but PER of diet with 35% protein was found to be better than PER of both high and low protein diets (diets of 40 and 30% crude protein). It is concluded that diet having 35% protein with protein energy ratio of 17.53 $mgkJ^{-1}$ was suitable for rainbow trout (*O. mykiss*) among the protein spectrum used.

(**Key Words** : Rainbow Trout, Diet, Protein, Energy, Growth)

Introduction

As a result of recent developments of and improvements in the techniques of aquaculture, productions has been increasing dramatically. Continued expansion and improvements in feed utilization are possible through better feed formulation. However, to be successful, feed developments must consider on one hand the nutritional requirements of the cultured species, in terms of their energy, protein and other nutrients and on the other hand, the range of available feed ingredients, their cost, digestibility and quality. In this aspect, the optimization of feeding responses in terms of survival and growth is important (Knights, 1985).

Protein is the most expensive among all the dietary components and fish as a poikilotherm requires more dietary protein levels than terrestrial animals (Lee and Putnam, 1973; Dabrowsky, 1977). Fish preferentially uses protein as an energy source for maintenance and/or growth. However, excess dietary protein than the body can be able to be synthesized into body protein are deaminated and the resultant carbon residue consequently oxidized or stored as fat or carbohydrate (Cowey and Sargent, 1972). This deamination required energy intake which could otherwise be used for growth. The main aim in intensive fish farming is to produce maximum growth in fish and to reduce the operational cost by maximizing utilization of dietary protein for tissue formation rather than for energy generation. Dietary protein requirements are generally expressed in terms of a fixed dietary percentage or a ratio of protein to energy, which is normally calculated using either gross, digestible or metabolizable energy values (Tacon and Coweg, 1985). The dietary protein : energy ratio affects whether protein will be incorporated for tissue formation of catabolized for energy purposes. Protein utilization in fish has been shown to depend on a number of factors, such as the nutritional value of food, relative proportion of dietary protein, lipid, carbohydrate, energy density of diets, species, other environmental factors, such as temperature and salinity (Menton et al., 1986; Lovern, 1988) and the size of the fish (Cowey et al., 1972). Unless the correct

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nutritional balance is achieved, reduced food intake following addition of lipid to the diet may result in an overall reduction in protein intake which might reduce the growth rate of fish (Jauncey, 1982). The balancing of protein and energy requirement in the diets of rainbow trout is of great importance in determining the degree of utilization of formulated diets and hence for a good growth of the fish. Rainbow trout like other carnivorous fishes, requires high levels of good quality dietary protein (Tacon and Jackson, 1985). A positive correlation between protein : energy and protein utilization for rainbow trout was also calculated by Lee and Putnam (1973). The highest Protein Efficiency Ratio (PER) values were obtained if 60% to 70% of the total dietary energy was accounted for by the non-protein fraction. Efficient production and growth of fish depends on feeding the best possible diets at levels not exceeding the dietary needs. The effectiveness of diets formulated on the basis of digestible energy and nutrients can be evaluated by measuring weight gain, protein efficiency ratio (protein : gain) or feed conversion ratio (feed : gain).

The rainbow trout has been one of the most extensively studied fish. It has been harvested to produce thousands of tons of food, it is farmed in many countries around the world and it is an extremely popular experimental fish (Gall and Grandell, 1990; Murai, 1992). Considering this fish as a commercially and experimentally popular the experiment was planned to see the effect of protein level on the growth performance of rainbow trout, *Onchorhynchus mykiss* fed with constant protein energy ratio diets.

Materials and Methods

Proximate analysis of ingredients

Prior to diet formulation, proximate analysis of all ingredients was conducted at the beginning of the study (table 1). Proximate analysis was carried out according to AOAC (1980). Gross energy of diets were calculated from proximate analysis considering equivalent energy for protein, fat and carbohydrate as 23.6, 39.5 and 17.2 kJg^{-1}, respectively (Brafield and Llewellyn, 1982; Jobling, 1983).

Formulation of diets

Four diets with 25, 30, 35 and 40% crude protein were formulated to give four dietary treatments with the energy ratio fixed at 20 kJg^{-1} diet. Table 2 shows the formulation and the inclusion of different ingredients in the diets. Fish meal and fish protein concentrate were obtained from a fish feed manufacturer and wheat and soybean meal were ground to flour in a Scotmec (Ayr) Hammer Mill. The particle's mesh size in the mill was less than 1.0 mm. Fish meal was sieved using a sieve with a mesh size of 250 μm. Then all the milled ingredients (on dry weight basis) were thoroughly mixed in a Hobart

TABLE 1. THE PROXIMATE ANALYSIS OF THE INGREDIENTS USED IN DIETS (% DRY WEIGHT BASIS)

Ingredient	Moisture	Crude protein	Crude fat	Ash
Fish meal	11.10	73.19	8.25	16.11
Soya meal	13.88	38.35	20.62	5.60
Wheat meal	14.12	10.09	0.91	1.48
Fish protein concentrate	5.13	85.05	3.30	6.10

TABLE 2. FORMULATION AND INCLUSION OF DIFFERENT INGREDIENTS (% DRY WT.) FOR DIETS AND THEIR PROXIMATE COMPOSITION

Ingredient	Diets (% of inclusion)			
	1	2	3	4
Fish meal	14.70	16.70	24.80	32.40
Soya bean meal	14.00	14.00	14.00	14.00
Fish oil	15.30	15.30	16.30	14.50
Wheat meal	46.00	39.00	30.00	24.10
Fish protein concentrate	5.00	10.00	10.00	10.00
Vitamin premix	0.30	0.30	0.30	0.30
Mineral premix	0.30	0.30	0.30	0.30
Carboxymethyl methyl cellulose	4.40	4.40	4.40	4.40
Total	100.00	100.00	100.00	100.00
Proximate Composition :				
Moisture	13.83	15.02	16.06	16.93
Crude protein	24.96	30.05	34.79	39.47
Fat	20.48	19.26	21.32	22.17
Ash	5.83	6.04	7.42	8.51
Crude fibre	1.80	1.62	1.74	1.04
Carbohydrate(NFE)	33.10	28.01	18.67	11.88
Calculated				
Dietary energy(KJg^{-1})	19.67	19.52	19.84	20.11
P/E ratio($mgkJ^{-1}$)	12.68	15.40	17.53	19.62

Eq. energy = protein : 23.6 KJg^{-1}
fat : 39.5 KJg^{-1}
carbohydrate : 17.2 KJg^{-1} (Brafield and Llewellyn, 1982)

A 200 industrial mixer for 10-12 minutes. Then the premixes and carboxy methyl cellulose were added to the mixed ingredients and mixed thoroughly. The oil component was added gradually while mixing. The feed mixtures were then pelleted using a California Laboratory Pellet Mill with steam conditioning. The diameter of the holes in the pellet die was 2.0 mm. The diets were allowed to cool on trays at an ambient temperature for 10-15 minutes and then stored in freezer (−20℃) until subsequent use in the experiments.

Fish and experimental system

Rainbow trouts (*Oncorhynchus mykiss*) (80.5 to 98.6 g) were used in this experiment. The fish were collected from College Mill Farm, Almond bank, Perth, Scotland on March 1993. Thirty fish were stocked in each 300 L fibre glass tanks set in a recirculatory water system at a flow rate of 3 $Lmin^{-1}$. Each of the 4 dietary treatments were duplicated. For a period of 4 weeks before commencing the experiment, fish were acclimatized to the system. During this period, the fish were fed initially with a commercial diet containing 40% crude protein for first 2 weeks followed by the experimental diets for another two weeks. During experiments the fishes in each of treatment groups were fed *ad libitum* (measured as about 3% of the body weight) distributed twice daily. From an air pump each tank was aerated using a single airstone in order to maintain a reasonable (saturated) oxygen level. During the experimental period the fish were kept at an average of 13.5℃ (10.5-16.5℃) water temperature and subjected to a natural photoperiod.

Growth performance

At the beginning and at subsequent intervals of 15 days the fish in each tank were weighed individually. From this data initial and final mean weights were calculated and Specific Growth Rate (SGR) was calculated as follows:

$$SGR = \frac{LnW_2 - LnW_1}{T_2 - T_1} \times 100$$

where, L_n = natural logarithm (e-based)
W_1 = mean initial weight
W_2 = mean final weight
$T_2 - T_1$ = No. of days of experiment.

Food Conversion Ratio (FCR) was calculated using the following formula:

$$FCR = \frac{\text{Amount of food fed (g. dry wt.)}}{\text{Live weight gain (g)}}$$

The Protein Efficiency Ratio (PER) was calculated as follows:

$$PER = \frac{\text{Live weight gain}}{\text{Crude protein fed}}$$

Statistical analysis

One-way analysis of variance (ANOVA) was used for data comparison. The means were compared using Duncan's Multiple Range Test (Steal and Torrie, 1980).

Results and Discussion

Daily observation on feeding indicated that food intake by the fish depends on abiotic and biotic factors such as temperature, quality of food and size of the fish (Brett and Groves, 1979). Brett (1971) observed a change in food intake in sockeye salmon (*Oncorhynchus nerka*) under different water temperature. Wootton et al. (1980) also observed that food consumption of three-spine stickleback (*Gasterosteus aculeatus*) is temperature dependent and has been found to increase in growth from 5.6 to 12.2% of body weight with an increase of temperature from 6 to 19℃. During the experimental period it was evident that the food intake in the mornings was double that in the evenings as the time intervals between two meals was shorter in the day time than that in the night. This is in agreement with similar studies by Fange and Grove (1979). The water temperature monitored during the course of the experiment appeared to be within acceptable range (10.5℃-16.5℃).

The growth performances of fish fed different experimental diets are given in table 3. Generally accepted that weight gain could be used as a parameter in evaluating different levels of nutrient intake (protein level) and thus the nutrient reguirement of the fish.

Final mean weight of fish of treatment 1 was significantly different ($p < 0.05$) from that of treatment 3 and 4 and not significantly different ($p > 0.05$) from that of treatment 2. Significant difference ($p < 0.05$) in final mean weight were also found between treatment 2 and 3. It is evident that the highest mean final weight was obtained for fish fed diet 3 (35% protein) followed by diet 4 (40% protein), although there was no significant difference ($p > 0.05$) between them. This is in agreement with other. Cowey (1992) who stated that in current practical diets for rainbow trout, levels of digestible protein range from 33% to 42% depending on the energy density. The dietary protein and energy levels selected for the present study encompassed the range given above.

The specific Growth Rate (SGR) values recorded for each diet showed that there were no significant differences ($p > 0.05$) between diets. However, results revealed that the highest SGR was achieved for diet 3 which contained 35% protein level followed by diet number 4, 1, 2, respectively (table 3).

Growth rates decreased towards the end of the experiment. It might be due to the sudden change in water temperature or may be due to the handling stress. The Specific Growth Rate (SGR) of fishes fed on different diets are presented in table 3. Maximum SGR occurrence was observed within the range of 1.37 to 1.55% per day for all 4 treatments.

TABLE 3. GROWTH PARAMETERS FOR RAINBOW TROUT (*Oncorhynchus mykiss*) FED ON DIFFERENT DIETS FOR 60 DAYS

Parameter	Diet number				
	1	2	3	4	SEM ±
Initial mean weight (g)	$82.02^{a} \pm 1.79$	$83.87^{ab} \pm 1.75$	$92.52^{b} \pm 1.94$	$96.48^{c} \pm 0.23$	1.64
Final mean weight (g)	$192.26^{a} \pm 1.74$	$190.50^{a} \pm 0.38$	$234.60^{b} \pm 2.08$	$227.11^{b} \pm 5.58$	3.11
SGR[1] (% day^{-1})	$1.41^{a} \pm 0.06$	$1.37^{a} \pm 0.06$	$1.55^{a} \pm 0.01$	$1.43^{a} \pm 0.11$	0.06
FCR[2]	$1.48^{a} \pm 0.00$	$1.44^{a} \pm 0.03$	$1.17^{b} \pm 0.02$	$1.15^{b} \pm 0.08$	0.03
PER[3]	$2.31^{a} \pm 0.01$	$2.11^{a} \pm 0.04$	$2.25^{a} \pm 0.05$	$2.00^{a} \pm 0.11$	0.06
Mortality (%)	1.67	10.00	10.00	5.00	3.33

Figures in each row having different superscripts are significantly different ($p < 0.05$).
Means given with ± standard errors of means.
[1] Specific growth rate
[2] Food conversion ratio
[3] Protein efficiency ratio

The FCR calculated for *Oncorhynchus mykiss* is presented in table 3. The best FCR was produced for diet 4 followed by diets 3, 2 and 1, respectively. Significant differences ($p < 0.05$) were found between FCR for diets 3, 4 and 1, 2. In the present study, it was revealed that diets 3 and 4 (35% and 40% protein respectively) gave the best FCR for rainbow trout. The FCR decreased with increasing protein levels (table 3). These results are similar to those cited by Tacon and Cowey (1985) for rainbow trout.

The average PER calculated for each treatment are given in table 3. There was no significant difference ($p > 0.05$) of PER among treatments. The lowest dietary protein level (25%) gave the highest PER. There was a tendency for PER to decrease, except for diet 3, with increasing protein level.

In the present study it was recorded that the maximum growth of rainbow trout obtained at 35% dietary protein level and P:E of 17.53 mg/kJ. The relationship between protein content and energy appears to play an important role in protein conversion efficiency (Mazid et al., 1979).

The mortality varied among the treatment groups (table 3). This high rate of mortality of fish during the experiments in the treatments 2 and 3 were due to a sudden block of water-flow system for few hours caused by mechanical fault. This resulted in a respiratory stress of the fishes.

The present study on the food intake at low to high protein diets with fixed energy content on the performance of rainbow trout (growth, food conversion ratio and protein utilization) have revealed that the best SGR for *Oncorhynchus mykiss* was for the diet having 35% crude protein whereas, the best FCR was at 40% dietary protein level. However, The PER showed no significant difference between diets. From the experiment it reveals that the best growth of rainbow trout requires dietary crude protein level of 35 to 40% with a fixed energy content of 20 kJ/g. Thus, future experiment can be planned by changing the energy content of the diet keeping protein level of 35 to 40% of the diet.

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