Replacement of Fish Meal by Alternative Protein Sources in Rainbow Trout Diets^{*1}

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The utilization of some alternative protein sources, i.e. defatted soybean meal (SBM), corn gluten meal (CGM), and meat meal (MM), as a partial replacement for fish meal was studied with rainbow trout having an average body weight of about 3 g. The combination of those protein meals was used to replace 55, 64, 73, 82, and 91% of fish meal in the control diet containing 42% protein. Duplicate tanks of fish were fed to satiation at 15°C for 13 weeks.

Fish fed all the SBM, CGM, and MM combination diets produced a good growth rate, feed efficiency, and voluntary food intake comparable to the control group. Although the PER values of the experimental diets containing alternative protein sources were lower than that of the control, there was no marked difference in protein and energy retention except for the 91% fish meal replaced diet. However, this diet still seemed to be economically beneficial as a relief for a short supply of fish meal. All the test proteins were well digested, assimilation values ranging from 91 to 93%. Hepatosomatic index and chemical composition of the whole body did not vary from the control group.

Soybean meal is one of the most promising replacements for fish meal. However, high levels of soybean meal as a partial or total replacement for fish meal generally reduce feed utilization and result in growth performances lower than those of fish meal diets.^{1–5)} Soybean meal is considerably high in its content of complex carbohydrates which are poorly digested by rainbow trout, leading to a reduced digestible energy (DE) content in the diet.⁵⁾ This protein meal has a poorer balance of essential amino acids (EAA) than fish meal, and is somewhat deficient in some certain EAAs such as methionine. Consequently, some researchers have tried to utilize alternative proteins by complementing them with other protein sources so as to obtain the required EAA profile for fish. The combination of soybean meal and corn gluten meal has been employed with success to replace up to 63% of fish meal in the diet for rainbow trout.⁵⁾ The use of soybean meal with corn also makes an excellent feed for poultry.⁶⁾ These results have indicated that

alternative proteins must be combined with each other for effective utilization in terms of an improved amino acid profile and an elevated dietary protein level.

From these viewpoints, the combination of defatted soybean meal, corn gluten meal, and meat meal were combined as a replacement for fish meal in the diet for rainbow trout.

Materials and Methods

Experimental Diets

Commercial brown fish meal, meat meal (MM), and corn gluten meal (CGM) were obtained from Nippon Nosan Kogyo Co.; defatted soybean meal (SBM) was provided by Sakamoto Fish Feed Company. The proximate composition and amino acid contents of these protein meals are presented in Table 1. The composition and nutrient contents of the experimental diets are given in Table 2. SBM was used in combination with CGM and MM as a protein substitution for

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Table 1. Amino acid and proximate composition of various protein meals

Amino acid (g/100 g protein)	Fish meal	Soybean meal	Corn gluten meal	Meat meal
Arginine	5.35	7.04	3.03	6.45
Lysine	7.63	5.88	1.62	5.07
Histidine	2.90	2.58	1.81	1.93
Phenylalanine	3.80	4.83	6.19	3.24
Tyrosine	3.01	2.96	4.72	2.21
Leucine	7.05	7.31	15.95	5.75
Isoleucine	3.90	4.35	3.89	2.81
Methionine	2.63	1.32	2.37	1.39
Cystine	0.86	1.41	1.72	0.74
Valine	4.91	4.52	4.31	4.31
Alanine	5.93	4.12	8.30	7.81
Glycine	5.99	4.07	2.50	12.40
Proline	4.11	5.04	9.82	7.97
Glutamic acid	11.56	16.72	21.27	10.87
Serine	3.74	4.72	4.94	3.43
Threonine	3.99	3.70	3.28	3.09
Aspartic acid	8.69	10.99	6.07	7.12
Tryptophan	1.03	1.28	0.42	0.65
Proximate composition (%)				
Crude protein	67.64	47.46	63.95	79.65
Crude lipid	9.45	1.97	3.17	11.82
Crude ash	15.14	5.59	1.54	4.38
Moisture	8.16	12.13	10.62	4.30

Table 2. Composition and nutrient analysis of the experimental diets containing alternative protein sources for rainbow trout

Replacement of fish meal (%)	0	55	64	73	82	91
Diet no.						
Ingredient (%)	1	2	3	4	5	6
Brown fish meal	56	25	20	15	10	5
Meat meal	—	—	5	10	15	15
Corn gluten meal	—	15	15	15	15	15
Soybean meal	—	25	25	25	25	30
Feed oil* ¹	15	15	15	15	15	15
α -Starch	15	12	12	12	12	12
Cellulose	6	—	—	—	—	—
Mineral mixture* ²	5	5	5	5	5	5
Vitamin mixture* ²	1.4	1.4	1.4	1.4	1.4	1.4
Choline chloride	0.5	0.5	0.5	0.5	0.5	0.5
Vitamin E (50% purity)	0.1	0.1	0.1	0.1	0.1	0.1
Chromic oxide* ³	1	1	1	1	1	1
Nutrient content (% on dry matter basis)						
Moisture	4.1	5.0	4.5	6.3	7.5	7.9
Crude protein	41.6	43.1	43.4	44.0	44.7	43.5
Crude lipid	22.8	21.3	21.4	22.1	21.7	20.6
Crude ash	13.1	10.0	9.4	8.9	8.5	7.9
Crude starch	14.2	17.7	20.3	21.7	22.7	23.7
Gross energy* ⁴	545.8	564.3	565.6	572.2	580.7	582.5
Digestible energy* ⁴	478.0	486.7	499.8	499.6	508.9	519.6
Protein cost (¥/kg protein)	192.2	137.5	129.6	121.9	114.4	106.5
Protein cost/weight gain (¥ protein cost/kg gain)	68.4	49.6	46.9	44.8	43.1	42.6

*¹ A mixture of pollack liver oil: soybean oil (2: 3).*² The composition was the same as reported previously.⁵⁾*³ Chromic oxide: cellulose (1: 1).*⁴ kcal/100 g dry diet.

Table 3. Amino acid composition of the experimental diets containing alternative protein sources for rainbow trout

Amino acid (g/100 g dry diet)	Diet no.						Requirement* ¹ (g/100 g protein)
	1	2	3	4	5	6	
Arginine	2.21	2.25	2.36	2.38	2.43	2.45	3.1 (1.34/40)
Lysine	3.14	2.39	2.38	2.25	2.19	2.10	4.7 (1.88/40)
Histidine	1.17	1.10	1.11	1.07	1.04	1.03	1.4 (0.46/40)
Phenylalanine	1.60	2.08	2.11	2.06	2.05	2.09	2.8 (1.12/40)
Tyrosine	1.12	1.44	1.44	1.39	1.38	1.38	1.9 (0.76/40)
Leucine	2.93	4.14	4.18	4.08	4.06	4.10	3.9 (1.56/40)
Isoleucine	1.69	1.79	1.81	1.74	1.72	1.71	2.1 (0.84/40)
Methionine	1.13	0.97	0.93	0.89	0.91	0.81	1.6 (0.64/40)
Cystine	0.37	0.55	0.55	0.54	0.58	0.56	0.8 (0.31/40)
Valine	2.08	2.06	2.10	2.06	2.06	2.04	2.8 (1.12/40)
Alanine	2.42	2.59	2.74	2.77	2.87	2.79	
Glycine	2.52	1.99	2.37	2.66	2.99	2.86	
Proline	1.68	2.43	2.64	2.78	2.95	2.99	
Glutamic acid	4.81	6.69	6.85	6.77	6.77	7.02	
Serine	1.52	1.86	1.90	1.84	1.86	1.88	
Threonine	1.64	1.59	1.62	1.54	1.53	1.51	3.0 (1.20/40)
Aspartic acid	3.63	3.77	3.84	3.72	3.69	3.72	
Tryptophan	0.49	0.42	0.42	0.40	0.38	0.38	0.4 (0.16/40)
EAAI* ²	79.07	77.76	77.71	74.92	73.89	74.90	

*¹ In parentheses, the numerators are requirements as percent of diet and the denominators are percent total protein in diet.⁷⁾

*² Essential amino acid index.

fish meal. Diet 1 was the control diet, containing 56% fish meal as its sole protein source. The combination of these protein meals was used to replace 55, 64, 73, 82, and 91% of fish meal in the control diet for diets 2, 3, 4, 5, and 6, respectively. The diets were formulated to contain almost the same levels of protein but a slightly higher energy content (GE) in diets with higher levels of fish meal replacement in order to compensate for undigestible carbohydrates in plant ingredients. The amino acid composition and EAA index of the diets are presented in Table 3. All the experimental diets seemed to contain sufficient amounts of EAA to meet the requirement of this fish. The amino acid content in fish meal replaced diets was almost comparable to that in the control fish meal diet, except for lysine and methionine. However, the lysine level in these diets (>2.1% of dry diet) was over the requirement value of 1.88% of dry diet for rainbow trout reported by Ogino⁷⁾ or 1.3% of dry diet recently reported by Kim *et al.*⁸⁾ Methionine requirement for this fish has been reported to be about 1% of dry diet in the absence of dietary cystine,⁹⁾ or 0.64% when dietary cystine was 0.32%.⁷⁾ Therefore, all the experimental diets contained a sufficient amount of this EAA. EAA index was

calculated on the basis of the EAA of the whole egg protein. The value varied between 74 to 79, being highest in the control diet.

The dietary ingredients were well mixed with a commercial mixer, and oil was gradually added to the dry mix and enough water added to make a stiff dough. The mixture was passed through a meat chopper and noodle-like pellets were cut into a suitable size. Then they were dried in a vacuum dryer at 20°C for 18–24 hours. The diets were kept in the refrigerator (3°C) until use.

Fish and Feeding Methods

The stock rainbow trout were initially fed a commercial rainbow trout diet for 4 weeks. Fish with an average body weight of about 3 g were randomly selected and 30 fish each were placed into 45 l tanks. Water temperature was controlled at 15±1°C and water supply was maintained at 0.6–1.0 l/min. Each diet was fed to fish in duplicate, twice a day at satiation level for 13 weeks. During the feeding period, they were weighed at 4-week intervals in order to record the growth rate.

Analytical Methods

The diets were sampled and analyzed for crude

protein, crude lipid, crude starch, crude ash, moisture, and gross senergy.¹⁰⁾ Feces samples were analyzed for chromic oxide, crude protein, crude starch, and gross energy.¹⁰⁾ The amino acid composition of the experimental diets was determined by Japan Food Research Laboratories. Digestibility was determined using chromic oxide as the inert reference substance. Feces were collected using the same method as mentioned previously.¹⁰⁾

Fifty fish were initially sampled and fifteen from each tank were finally sampled for proximate analysis. Pooled whole body samples from each tank were analyzed for protein, lipid, moisture, and ash using the same methods as described elsewhere.¹⁰⁾

Statistical Analysis

The analysis of variance (ANOVA) and Duncan's multiple range test were applied to compare the differences of treatment means at the 0.05 significance level.

Results and Discussion

Growth and Feed Performances

The growth and feed utilization are given in Table 4. The average body weight of fish fed different diets during a period of 13 weeks is shown in Fig. 1. Fish fed a diet containing 25%SBM and 15%CGM, replacing 55% of fish meal, showed the best growth rate. The replacement of fish meal up to 82% with various combinations of SBM, CGM, and MM produced slightly higher growth than the control fish meal diet. But the growth was slightly reduced when 91% of the fish meal was substituted by 30%SBM, 15%CGM, and 15%MM. However, the final body weight and percent weight gain of fish receiving this diet was not significantly different from

Fig. 1. Growth of rainbow trout fed diets containing soybean meal, corn gluten meal, and meat meal as a substitute for fish meal.

that fed the fish meal diet. Feed efficiency was between 1.1 and 1.2, being lowest in the 91% fish meal replaced diet. The PER of all the experimental diets was high, ranging from 2.5 to 2.8. These values tended to decrease slightly when the fish meal substitution level increased. The lower feed and protein utilization of fish fed the 91% replaced diet may possibly be due to lower amino acid absorption. This was supported by the results of Wilson *et al.*,¹¹⁾ who reported that individual amino acid availabilities within a feed ingredient are variable, although there was reasonable agreement between protein digestibility values and average amino acid availability values. They also noted that the lysine availability value of 90.9% is somewhat higher

Table 4. Growth, feed efficiency, and PER of rainbow trout fed diets containing alternative protein sources

Diet no.	Replacement of fishmeal (%)	Av. body wt. (g)±SD		Weight gain (%)	Feed efficiency	PER
		Initial	Final			
1	0	3.0±0.4	37.5±13.5 ^{a*1}	1166.8 ^a	1.17	2.81
2	55	2.9±0.4	42.3±14.1 ^a	1355.3 ^a	1.19	2.77
3	64	2.9±0.3	41.1±12.1 ^a	1314.7 ^a	1.20	2.76
4	73	2.9±0.3	39.9±10.1 ^a	1287.4 ^a	1.20	2.72
5	82	2.9±0.3	37.9±12.1 ^a	1226.6 ^a	1.19	2.66
6	91	3.0±0.4	35.3±9.7 ^a	1093.3 ^a	1.10	2.50

*1 Values with the same superscript within columns are not significantly different at $P>0.05$.

Table 5. Feed consumption and retention of protein and energy in rainbow trout fed diets with alternative protein sources

Diet no.	Replacement of fishmeal (%)	Feeding rate (%)	Daily intake per fish			Retention (%)	
			Feed (mg)	Protein (mg)	DE*1 (cal)	Protein	Energy
1	0	1.70	344.5	142.9	1674	44.6	47.4
2	55	1.70	384.0	165.2	1930	45.1	48.1
3	64	1.68	369.8	160.5	1878	44.2	47.0
4	73	1.67	358.0	158.3	1830	42.7	45.6
5	82	1.68	342.3	153.6	1769	41.5	44.6
6	91	1.79	342.0	150.5	1782	38.4	41.6

*1 Digestible energy.

than the 80.4% of methionine availability reported for soybean meal in catfish. Dabrowski *et al.*²³ found that amino acid absorption by rainbow trout decreased even when 25% of the fish meal protein was replaced by soybean meal protein (13% of soybean meal in diet), and decreased further at the 50 and 100% substitution levels particularly of methionine, leucine, and threonine. The same authors also found that an average EAA absorption decreased by 9.9% in fish fed a diet containing 25% soybean meal (81.3% EAA absorption) compared to that fed a fish meal diet (91.2% EAA absorption).

The feed consumption and retention of protein and energy are shown in Table 5. Fish fed all the experimental diets exhibited good feeding activity due to a high palatability of the diets. The daily feed intake of all the experimental fish fed various fish meal replaced diets did not differ greatly from that of the control group. The mean value was about 1.7 g/100 g body wt./day. This result indicates that a combination of SBM, CGM, and MM as a replacement for 91% of fish meal could produce feed palatability comparable to the fish meal diet. All the experimental fish exhibited good health, and no mortality was observed throughout the feeding period. Lovell reported that SBM could be included as a major protein source in rainbow trout diets with a satisfactory growth rate, but that if the level of fish meal was reduced below approximately 18%, diet palatability decreased.¹²³ However, the present study found that only about 5% fish meal was enough to provide good feed palatability in rainbow trout when 91% of fish meal was replaced by a combination of 30%SBM, 15%CGM, and 15%MM.

The protein and energy retention in fish fed diets in which fish meal was substituted up to 82% was not apparently different from those

fed the fish meal diet. Meanwhile, fish fed the 91% fish meal replaced diet showed lower protein and energy retention than the control group. The values of protein and energy retention ranged from 38.4 to 45% and from 41.6 to 48.1%, respectively. All nutrient utilization parameters in fish fed diets in which fish meal was replaced up to 82% generally revealed good performances comparable to those fed the fish meal diet.

Although these parameters in fish fed the 91% fish meal replaced diet were inferior to those in fish fed the fish meal diet, the protein cost of feed or the protein cost per kg weight gain of fish was still lower in the former group than in other groups (Table 2.), when calculated on the basis of the price of each protein ingredient in 1992 (fish meal, ¥130/kg; SBM, ¥44.5/kg; CGM, ¥58/kg; MM, ¥85/kg). The replacement of fish meal up to 91% by a combination of 30%SBM, 15%CGM, and 15%MM (with 5% of fish meal in the diet) is therefore economically beneficial.

Apparent Digestibility

The apparent digestibility of dietary nutrients is presented in Table 6. The protein digestibility

Table 6. Apparent digestibility of nutrients in diets with various alternative protein sources in rainbow trout

Diet no.	Replacement of fishmeal (%)	Apparent digestibility (%)		
		Protein	Starch	Energy
1	0	91.2	82.9	87.7
2	55	90.9	77.9	87.7
3	64	91.8	79.3	88.7
4	73	92.3	76.7	88.2
5	82	92.7	78.5	88.6
6	91	93.5	79.1	89.1

Table 7. Carcass composition of the experimental fish fed diets with various alternative protein sources

Diet no.	Replacement of fishmeal (%)	Crude protein	Crude lipid	Moisture	Crude ash	Hepato-somatic index
Initial fish		14.68	5.62	78.36	2.29	
1	0	15.77	14.25	68.74	2.03	1.05
2	55	16.15	14.63	67.90	1.99	0.95
3	64	15.89	14.14	68.65	1.97	1.05
4	73	15.64	13.82	69.16	1.93	1.05
5	82	15.55	13.69	68.78	1.99	1.03
6	91	15.31	14.08	68.51	1.98	1.00

of all the experimental diets was relatively high, ranging from 91 to 93%, and was not greatly affected by the inclusion of these alternative proteins. The high protein digestibility of 91, 89, and 91% for SBM, CGM, and MM has been pointed out previously.^{10,13)*} The digestibility of starch in the fish meal replaced diets was 76–79%, with no great difference between treatments, except for a slightly higher value in the control diet (83%). This could be ascribed to the fact that almost the same amount of plant proteins (25–30%SBM and 15%CGM) and undigestible carbohydrates were included in the 55–91% fish meal replaced diets and that α -starch with high digestibility was contained in the control diet. The energy digestibility values of the diets were all high, ranging between 88 to 89%, and were not markedly different in the dietary treatments. The energy digestibility was also not apparently affected by starch digestibility, due to the low dietary starch level.

Body Composition

The whole body composition and hepatosomatic index (HSI) of all the experimental fish are reported in Table 7. The body protein content of fish fed fish meal replaced diets was not markedly different from fish fed the control diet, except for a slightly lower value in fish fed the 91% fish meal replaced diet. The protein and lipid contents of the initial fish were lower than those of the final fish after the experiment. The moisture content was higher in the initial fish than the final. The body lipid, ash, and moisture contents of fish fed fish meal replaced diets were not apparently different from those on the control diet. There was no marked difference in HSI between dietary groups. It ranged between

0.95 to 1.05, which was in accordance with that in other studies.^{14,15)} This HSI may suggest the good condition of the experimental fish. Satomi and Tanaka, 1973 (cited by Dabrowska and Wojno, 1977),¹⁵⁾ using covariance analysis, showed that there was no direct relationship between the relative weight of the liver of trout fed a diet containing soya bean and that of trout fed commercial pelleted feed.

Therefore, it was concluded that a combination of 30%SBM, 15%CGM, and 15%MM as a replacement for about 90% of fish meal in the diet for rainbow trout produced growth comparable to the fish meal diet at about 44% dietary protein level. The protein and feed utilization of this diet was somewhat lower than that of the fish meal diet, but the diet is still economically beneficial as a relief for a short supply of fish meal. A combination of these alternative proteins might improve their amino acid profiles, leading to improved feed utilization.

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