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Efficiency of feed utilization in Atlantic salmon (*Salmo salar* L.) fed diets with increasing substitution of fish meal with vegetable proteins

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Abstract

Atlantic salmon smolts were fed for 15-week diets where 28% and 55% of the fish meal, respectively, were replaced by a mixture of full-fat soybean meal and maize gluten meal in comparison with a diet where 89% of the protein came from fish meal and the remainder from full-fat soybean meal and maize gluten meal. The diets were equal in gross energy, crude protein, lipids, carbohydrates, lysine and methionine plus cystine. Apparent digestibilities of energy and protein in fish meal, full-fat soybean meal and maize gluten meal were determined as 93.5% (19.5 MJ digestible energy (DE) kg^{-1}) and 89.3%, 69.3% (14.4 MJ DE kg^{-1}) and 76.9%, and 78.6% (16.4 MJ DE kg^{-1}) and 87.0%, respectively. Tested by ANOVA and orthogonal polynomials, a linear reduction that approached significance and with no significant deviation from linearity was found in growth, SGR and TGC with increasing substitution of fish meal. Substitution of fish meal caused a significant linear reduction in the condition factor, while feed consumption was not affected. The effect of substitution on feed conversion ratio (FCR = g feed g^{-1} growth) showed a near significant deviation from linearity, FCR being significantly higher on Diet 3 than Diet 1 and slightly lower on Diet 2 than Diet 1. Apparent protein digestibility of the test diets decreased significantly linearly with increasing substitution, while no significant effects were found on digestibility of lipids and gross energy. Carbohydrate digestibility was low with no significant differences between diets. Fish meal substitution had no significant effect on carcass content of moisture, protein, ash or energy, while content of fat was nearly significantly linearly reduced. Substitution caused a nominal insignificant linear reduction in protein accretion but had no effect

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on digestible protein consumption per gram of protein accretion. Protein efficiency ratio (PER = weight gain/protein consumption) decreased significantly curvilinearly and net protein value (NPV = carcass protein gain/protein consumption) significantly linearly with increasing substitution. Energy utilization for growth decreased significantly linearly with increasing substitution. There was a nominal linear reduction, which approached significance in energy accretion with increasing substitution. Substitution impaired energy utilization for energy accretion (i.e. increased MJ DE consumed/MJ accredited).

Keywords: Fish meal substitution; Full-fat soybean meal; Maize gluten meal; Salmon; Growth performance; Composition

1. Introduction

Periodical high prices of fish meal has led feed producers to replace fish meal with vegetable protein sources in feed for Atlantic salmon in order to keep feed prices down. However, lower feed prices may not reduce the cost of production for the salmon farmer if the growth of the salmon is reduced and the feed conversion ratio is impaired, such that more feed is needed per kilogram of salmon produced.

Previous studies with Atlantic salmon (Anderson et al., 1992) and rainbow trout (Cho and Kaushik, 1990; Hajen et al., 1993; Sanz et al., 1994; Aksnes and Opstvedt, 1998) have shown that the content of digestible energy in vegetable protein sources is lower than that of fish meal. Since utilization of the feed energy for salmon can be predicted from the content of digestible energy (Cho, 1994), it would be expected that substitution of fish meal with vegetable protein will reduce growth rate and impair feed conversion ratio. Several studies (Olli et al., 1994, 1995; Bjerkeng et al., 1997; Storebakken et al., 1998; Carter and Hauler, 2000; Refstie et al., 2001) have explored the effect on growth of replacing fish meal with vegetable protein, mainly soybean products, in diets for Atlantic salmon. In general, these studies have shown that replacement of high-quality fish meal with vegetable protein sources causes a reduction in growth, the magnitude of which depending on the source and level of replacement. There are limited data on the effect of fish meal replacement on the efficiency of feed utilization where a proper technique has been used. We are not aware of studies with Atlantic salmon where fish meal has been replaced by a combination of full-fat soybean meal and maize gluten meal, the most commonly used vegetable protein sources nowadays.

The present study was conducted to reveal the effect on energy and protein utilization of substituting graded amounts of fish meal with full-fat soybean meal and maize gluten meal in feed for Atlantic salmon smolt. The growth study was conducted according to a one-way ANOVA design with equally spaced levels of fish meal, which made it possible to test if effects were linear, i.e. proportional with the level of substitution, or deviating from linearity being curvilinear, i.e. depending on the level of substitution. The salmon were fed under strictly controlled conditions where feed consumption and growth of the fish were closely monitored.

2. Material and methods

The study comprised two trials. Trial 1 was conducted to determine the digestibility of protein and energy in the test ingredients to be used in trial 2, i.e. fish meal, full-fat soybean meal and maize gluten meal. Trial 2 measured growth rate, feed consumption and energy and protein utilization in diets with increasing substitution of fish meal with full-fat soybean meal and maize gluten meal.

2.1. *Trial 1: digestibility of energy, protein and fat in fish meal, full-fat soybean meal and maize gluten meal*

Fish meal (Norse-LT®), full-fat soybean meal (SoyaxAqua®), maize gluten meal and wheat meal were obtained from the commercial market. The chemical composition of the test feed ingredients is shown in Table 1. The trial used the whole-diet substitution method (Cho et al., 1982) for determination of digestibility. The composition of the reference diet is shown in Table 2. In the experimental diets, 45% of the reference diet was substituted with fish meal, full-fat soybean meal and maize gluten meal, respectively.

Atlantic salmon (*Salmo salar* L.) about 12 months post hatching and with an average weight of about 500 g were distributed in blocks comprised of four replicate fiberglass tanks ($2 \times 2 \times 1$ m^3). All replicate tanks within each block contained 80 fish of identical feeding history. The tanks were supplied with 75 l/min seawater taken from a depth of 50 m. Water temperature was about 11 °C and salinity 32–33 ‰. The photoperiod was 24 h light. The fish were fed by automatic feeders at amounts equal to 120% of the growth tables given by Austreng et al. (1987) adjusted according to assumed biomass and

Table 1
Chemical composition and digestibility of energy and protein of test feeds

	Fish meal[a]	Full-fat soybean meal[b]	Maize gluten meal
Protein ($N \times 6.25$), g kg^{-1}	739	379	616
Fat, g kg^{-1}	106	176	14
Ash, g kg^{-1}	103	47	14
Carbohydrates[c], g kg^{-1}	Nil	291	256
Starch, g kg^{-1}	Nil	20	150
Moisture, g kg^{-1}	72	107	100
GE[d], kJ/g	20.84	20.85	20.78
DE[e], kJ/g	19.5 ± 0.3[f]	14.4 ± 0.3	16.4 ± 0.6
Digestibility of GE, %	93.5 ± 1.3	69.3 ± 1.3	78.6 ± 2.7
Digestibility of CP[g]	89.3 ± 1.1	76.9 ± 0.7	87.0 ± 1.8

[a] Norse-LT 94®, Norsildmel AL, N-5141 Fyllingsdalen, Norway.
[b] SoyaxAqua®, Quality guaranty: TIA max 1 m per g, PDI min. 10, Shouten Industries, Burgstraat 12, P.O. Box 1, 4283 Giessen, The Netherlands.
[c] Calculated by difference: 100 − (%protein+%fat+%ash+%moisture) (i.e. N-free extractives (NFE) + crude fiber).
[d] GE = gross energy.
[e] DE = digestible energy.
[f] Standard deviation.
[g] CP = crude protein.

Table 2
Feed and proximate composition of the reference diet

Composition, g kg^{-1}	
Fish meal[a]	750
Fish oil[b]	100
Dextrinised maize starch[c]	125
Vitamin mixture[d]	10
Mineral mixture[e]	5
Chromic oxide (Cr_2O_3)	10
Proximate composition	
Crude protein ($N \times 6.25$), g kg^{-1}	555 (561)[f]
Fat, g kg^{-1}	180 (204)
Carbohydrate, g kg^{-1}[g]	118
Ash, g kg^{-1}	92
Moisture, g kg^{-1}	69 (56)
Gross energy, MJ/kg	222 (221)

[a] Norse-LT 94®, Norsildmel AL.
[b] Norsalmoil, Norsildmel AL.
[c] Pregeflo M. Maize starch. Roquette, 62080 Lestrem Cedex, France.
[d] Provided per kilogram of feed: vitamin D_3, 1.28 mg; vitamin E, 136 mg; thiamin, 8 mg; riboflavin, 16 mg; pyrodoxine-HCl, 16 mg; vitamin C, 80 mg; calcium pantothenate, 17.4 mg; biotin, 0.16 mg; folic acid, 4.8 mg; niacin, 40 mg; vitamin B_{12}, 0.016 mg; menadion bisulphite, 20 mg.
[e] Provided per kilogram of feed: magnesium, 625 mg; potassium, 509 mg; calcium, 465 mg; zinc, 100 mg; iron, 62 mg; manganese, 13 mg; copper, 6 mg.
[f] Analysed values.
[g] Calculated by difference: 100 − (%protein + %fat + %ash + %moisture) (i.e. N-free extractives + crude fiber).

appetite. Wasted feeds were collected and recorded daily. The fish were fed the test diets without a transition period, and the test feeding lasted for 14 days. Feed consumption was monitored throughout the trial using a device that measured wasted feed in the drainage water. The fish were bulk-weighed at the start and end of the trial. At the end of the trial, fish were anaesthetized with benzocain (25 μg kg^{-1}) and gently cleaned with soft tissue, and feces were obtained from all fish by applying gentle pressure from the ventral fin to the anal region as described by Austreng (1978). Ethoxyquin (400 mg ethoxyquin kg^{-1} dry matter (DM)) was added to the feces, mixed, and the mixture immediately frozen and stored at −20 °C pending analysis. Frozen feces were lyophilized (plate temperature 20 °C) prior to chemical analyses. Apparent digestibility of nutrients and energy in test rations was calculated from the formula

$$AD = 100 - 100 \times \frac{Cr_{ed} \times CX_{ef}}{CX_{ed} \times Cr_{ef}}$$

where ed is diet, ef is feces, Cr is chromium content and CX is nutrient or energy content. Digestibilities of test ingredients were calculated from the formulae of Cho et al. (1982):

$$DE_{ti} = \frac{1}{n} DE_{ed} - \frac{1-n}{n} DE_{rd}$$

where n is the inclusion level of test ingredient, ti indicates the test ingredient, ed and rd indicate the experimental diet and the reference diet, respectively, as modified by (Aksnes et al., 1996):

$$AD_x = \frac{1}{n}\left(100 - 100 \times \frac{Cr_{ed} \times CX_{ef}}{CX_{ed} \times Cr_{ef}}\right) - \frac{1-n}{n}\left(100 - 100 \times \frac{Cr_{rd} \times CX_{rf}}{CX_{rd} \times Cr_{rf}}\right)$$

where ef is feces from the test diet and rf is feces from reference diet.

2.2. Trial 2: effects of replacing increasing amounts of fish meal by full-fat soybean meal and maize gluten meal on growth and feed utilization in Atlantic salmon

Atlantic salmon (*S. salar* L.) about 9 months post hatching and 122 g of average weight were obtained from a commercial hatchery and kept for acclimatization for 2 months before the trial started. The fish were distributed randomly to nine fiberglass tanks ($1.5 \times 1.5 \times 1$ m^3) with 80 fish per tank. The tanks were supplied with 75 l/min of seawater taken from a depth of 50 m with a salinity of 32–33 ‰. Water temperature was increased successively by a heat exchanger from 6.5–7.5 to 11.2 °C during the acclimatization period and kept at that level for the experimental period. The photoperiod was 24 h throughout the experiment. The fish were offered feed from automatic feeders. The level of feeding was regulated such that offered feed was 110% of consumption based on daily monitoring of wasted feed. During the acclimatization period, the fish were fed a commercial feed.

At the start of the trial, the different tanks were randomly distributed to three diets and the total fish weight in each determined. Ten fish representing the weight ranges were sampled, killed by a blow on the head, frozen and kept at −20 °C pending chemical analyses at the end of the trials. Feed consumption was monitored daily by a feed waste system, which allowed for ad libitum feeding. At the end of the trial, samples of feces were obtained by stripping as described above and used for determination of nutrient and energy digestibility. The trial period lasted for 104 days.

Ingredient composition and chemical content of the experimental diets are shown in Table 3. The intention was that Diet 1 should resemble a conventional salmon smolt diet in composition with a relatively high content of fish meal and limited amounts of vegetable proteins. Diet 3 was composed with as high content of vegetable protein as was considered safe in order to avoid anorexia. Diet 2 was composed to be intermediate between Diet 1 and Diet 3. To get a reliable comparison between fish meal and the vegetable protein ingredients, the diets were made equal with regard to gross energy, protein, lipids, carbohydrates, lysine and methionine plus cystine. The content of carbohydrate was maintained constant by including different levels of wheat flour. Lysine and methionine plus cystine were maintained constant by adjusting with synthetic lysine and methionine.

2.3. Chemical analysis

In feed ingredients, diets, feces and fish tissue crude protein ($N \times 6.25$) were determined by the Kjeldahl method (ISO 5983-1979) and moisture (ISO 6496-1983)

Table 3
Feed and proximate composition of experimental diets

	Diet 1	Diet 2	Diet 3
Percent of crude protein from fish meal	89	65	40
Ingredient content, g kg^{-1}			
Fish meal[a]	515	373	232
Corn gluten meal	20.0	152	283
Full-fat soybean meal[b]	20.0	88.0	155
NorSalmOil[c]	218	220	220
Wheat flour	212	135	60
L-Lysine HCl	0	7.0	13.9
DL-Methionine	0.8	0.4	0
Vitamin mixture[d]	10	10	10
Mineral mixture[e]	4.0	4.0	4.0
Carophyll Pink[f]	0.8	0.8	0.8
Calcium phosphate ($CaHPO_4 \cdot 2H_2O$)	0	10.4	20.7
Proximate composition, g kg^{-1}			
Moisture	68 (49)[g]	69 (52)	70 (47)
Crude protein ($N \times 6.25$)	428 (438)	427 (439)	427 (446)
Fat	280 (288)	279 (302)	277 (290)
Ash	68 (62)	65 (59)	63 (55)
Carbohydrates[h]	156 (163)	160 (148)	163 (162)
Total starch	150	117	93
Dextrinisation, % of total starch	94	89	89
Total lysine	31.1	31.1	31.1
Total methionine plus cystine	1.69	1.69	1.69
PV[i] start, meq/kg fat	< 1	< 1	< 1
PV end, meq/kg fat	< 1	2.9	7.7
AV[j] start	12	12	9
AV end	< 1	6.9	7.7

[a] See footnote to Table 1.
[b] See footnote to Table 1.
[c] See footnote to Table 1.
[d] Provided per kilogram of feed: vitamin D_3, 1.28 mg; vitamin E, 136 mg; thiamin, 8 mg; riboflavin, 16 mg; pyridoxine-HCl, 16 mg; vitamin C, 80 mg; calcium pantothenate, 17.4 mg; biotin, 0.16 mg; folic acid, 4.8 mg; niacin, 40 mg; vitamin B_{12}, 0.016 mg; menadione bisulphite, 20 mg.
[e] Provided per kilogram of feed: magnesium, 625 mg; potassium, 509 mg; calcium, 465 mg; zinc, 100 mg; iron, 62 mg; manganese, 13 mg; copper, 6 mg.
[f] Astaxanthin.
[g] Analysed figures.
[h] Calculated difference (see footnote g, Table 2).
[i] PV = peroxide value.
[j] AV = anisidine value.

and ash (ISO 5984-1978) gravimetrically after drying for 4 h at 105 °C and after combustion for 16 h at 550 °C, respectively. Peroxide value (PV) (AOCS Official Method Cd 8b-90) and anisidine value (AV) (AOCS Official Method Cd 18-90) were determined in feeds immediately after mixing and after storage for 14 weeks. Gross energy (GE) of feed ingredients, diets and lyophilized feces and fish tissue was determined in a Parr bomb calorimeter. Total starch was determined as glucose by heating in sodium hydroxide followed by enzymatic hydrolysis with glucoamylase. Dextrinised starch was determined by direct hydrolyses by glucoamylase.

2.4. Statistical methods and calculations

Specific growth rate (SGR) was calculated as

$$\mathrm{SGR}\ (\%) = (\ln w_2 - \ln w_1) \times 100/\text{feeding days}$$

where w_1 and w_2 are start and final fish weights, respectively, and ln is the natural logarithm.

Thermal growth coefficient (TGC) was calculated as described by Cho (1992):

$$\mathrm{TGC} = (w_2^{1/3} - w_1^{1/3}) \times 1000/\square(t \times \text{feeding days})$$

where $\square(t \times \text{feeding days})$ is the sum of water temperatures (°C) for every feeding days in the experiment.

The study was conducted according to one-way ANOVA with equally spaced distances between treatments (i.e. levels of fish meal replacement) and 3 replicates. Using orthogonal comparisons to calculate contrasts between treatments (i.e. Diet 1 − Diet 3 (linear effect)) and Diet 1 + Diet 3 − 2 × Diet 2 (quadratic effect) made it possible to test statistically if observed effects were linear, i.e. proportional with the level of replacement, or deviating from linearity (curvilinear), i.e. if the magnitude of the effect was dependent of the level of substitution (Steel and Torrie, 1960). A significant result would appear if the F-test showed that the fitted orthogonal polynomial coefficient was nonzero.

3. Results

3.1. Digestibility of feed ingredients and content of digestible energy

Digestibilities of gross energy (GE) and crude protein are shown in Table 1. It was not possible to test statistically the difference in digestibility of fat and carbohydrates in the test ingredients since they provided small and variable amounts of these nutrients to the test diets. The digestibility of crude protein in full-fat soybean meal was 12.4 percentage units and significantly ($P<0.001$) lower than in fish meal, while it was only 2.3 percentage units and insignificantly ($P>0.05$) lower in maize gluten meal compared with fish meal. The digestibilities of the lipids in the test diets were 92.7 ± 0.4% (average ± S.D.), 87.7 ± 0.2% and 87.2 ± 1.0% for the fish meal-, full-fat soybean meal- and maize gluten meal-based diets, respectively. The addition of full-fat soybean meal and maize gluten meal caused a reduction in the carbohydrate digestibility in the reference diet. The digestibility of GE in fish meal was significantly ($P<0.001$) higher than that of maize gluten meal and full-fat soybean meal by 16% and 26%, respectively. Maize gluten meal had significantly ($P<0.001$) higher GE digestibility than full-fat soybean meal by 12%.

Thus, although the content of GE was about equal in all three feeds, there were significant differences in the digestible energy (DE) content.

3.2. Production performance of Atlantic salmon smolt fed diets with increasing substitution of fish meal for vegetable protein

The test was carried out without unexpected events, the fish were apparently in good health and mortality was low in all groups. At termination, a certain number of fish in all groups were found to have lateral wounds in the front part of the body. This condition is common in tank trials and occurs mainly when the tanks are relatively small for the size of the fish. It is not considered to have had a negative impact on the production performance, and it is unlikely that it has influenced the comparison between the diets. The production performance of the fish is shown in Table 4.

There was a near significant ($P = 0.052-0.059$) linear reduction in g growth, SGR and TGC when the level of fish meal was reduced from 52% in Diet 1 to 37% in Diet 2 and further to 23% in Diet 3. Although the reduction was greater between Diet 2 and Diet 3 than between Diet 1 and Diet 2, the deviation from linearity was insignificant. Further, there was a significant linear decrease with no significant deviation from linearity in condition factor with increasing substitution. There were no significant differences between the diets in feed consumption. Feed conversion ratio was increased at the highest substitution, while it was slightly lower than the control at the lowest. Thus the effect was linearly highly significant, but with a near significant deviation from linearity. At the highest substitution level, feed conversion ratio was about 7% higher on Diet 3 compared to Diet 1. Thus the fish fed the diet with the lowest level of fish meal used 50 g more feed to grow the equivalent of 1 kg compared to those fed the diets with the highest level of fish meal.

Carcass composition of the fish at start and after 14 weeks of experimental feeding is shown in Table 5. The content of protein and fat increased and the content of moisture and ash decreased from the beginning to the end of the trial. Consequently, there was an

Table 4
Production performance of salmon smolts fed diets with increasing substitution of fish meal with vegetable proteins

	Diet 1	Diet 2	Diet 3	S.E.M.[a]	P^b (linear)	Deviation from linearity
Mortality, %	2.9	2.1	0	–		
Body weight at start, g	120	122	122	0.7	0.071	0.34
Body weight at end, g	569	565	528	13	0.071	0.34
Weight gain, g/fish	449	443	407	13	0.059	$F<1^c$
SGR, %	1.50	1.47	1.41	0.024	0.053	$F<1$
TGC, %	2.86	2.81	2.67	0.055	0.052	$F<1$
Condition factor	1.14	1.10	1.09	0.02	0.022	$F<1$
Feed consumption, g/fish	330	324	323	7.7	$F<1$	$F<1$
Feed conversion ratio, g feed/g growth	0.75	0.74	0.80	0.011	0.029	0.079

[a] S.E.M. = standard error of mean.
[b] P = probability of significance.
[c] F = Variance ratio = mean square of sample means/mean square of individuals.

Table 5
Carcass composition and energy content of smolts fed diets with increasing substitution of fish meal for vegetable protein

	Start	Diet 1	Diet 2	Diet 3	S.E.M.[a]	P^{b} (linear)	Deviation from linearity
Moisture, g kg^{-1}	751.0	698.0	703.0	702.3	2.1	0.19	0.41
Protein ($N \times 6.25$), g kg^{-1}	168.8	176.7	175.5	175.9	1.2	$F<1$	$F<1^{c}$
Fat, g kg^{-1}	91.1	142.0	141.0	137.8	1.2	0.051	$F<1$
Ash, g kg^{-1}	24.0	19.8	19.6	20.6	0.14	0.11	0.012
Energy, MJ/kg	7.30	9.45	9.28	9.27	0.09	0.21	$F<1$

[a] S.E.M. = standard error of mean.
[b] P = probability of significance.
[c] See Table 4, footnote [c] for explanation.

increase in the energy content in the course of the trial period. There were no significant differences between the diets in moisture, protein or gross energy. There was a near significant linear decrease in the content of fat with no significant deviation from linearity with increasing substitution of fish meal. The content of ash varied inversely with fat as expected. There was, however, no significant linear effect of substitution but a significant deviation from linearity.

The efficiency of dietary energy and nutrient utilization is shown in Table 6. In line with the protein digestibility of the individual feeds, there were significant differences between diets in protein digestibility, which decreased linearly highly significant with increasing substitution, with no significant deviation from linearity. Protein digestibilities of the experimental diets calculated from the values for fish meal, full-fat soybean meal and maize gluten meal determined in the introductory digestibility trial and a value of 87.0% for wheat meal (Hajen et al., 1993) were 88.6%, 86.0% and 83.5%. Thus protein digestibility was lower than calculated in the fish meal diet, while the opposite was the case for the substituted diets. There were no significant differences between diets in protein accretion or digestible protein consumption per gram of protein accretion. However, there were numerical reductions in protein accretion with increasing substitution. Protein efficiency ratio (PER = weight gain/protein consumption) decreased curvilinearly with increasing substitution. Net protein value (NPV = carcass protein/protein consumption) deceased significantly with increasing substitution. The deviation from linearity approached significance.

There was no significant difference in fat digestibility between diets, but Diet 3 had numerically lower fat digestibility than Diet 1 and Diet 2, which were almost identical in this respect. Carbohydrate digestibility was extremely low on all diets, without significantly differences between diets, although Diet 3 had numerically higher carbohydrate digestibility than Diet 1 and Diet 2.

Despite the fact that the maize gluten meal and full-fat soybean meal had considerable lower content of DE than the fish meal, there were only minor and insignificant differences between the diets in digestibility of GE and content of DE per kilogram of feed. This finding appears surprising. In order to test the validity of the DE values determined in the balance studies, the content of DE was also calculated from the content of digestible protein, fat and carbohydrate and their caloric values (Garret, 1974) of 23.93, 39.75 and

Table 6
Energy and nutrient utilization in Atlantic salmon fed diets with increasing substitution of fish meal for vegetable protein

	Diet 1	Diet 2	Diet 3	S.E.M.[a]	P^b (linear)	Deviation from linearity
Crude protein (CP) consumption, g/fish	147.86	144.29	144.50	3.767	$F<1^h$	$F<1$
Digestibility of CP, %	84.8	83.3	80.2	1.0	0.015	$F<1$
Digestible CP consumption, g/fish	123.68	120.02	115.87	2.636	0.051	$F<1$
CP accretion, g/fish	80.314	78.577	72.421	2.575	0.073	$F<1$
Digestible CP consumption, g/g protein accretion	1.561	1.529	1.585	0.037	$F<1$	$F<1$
PER[c]	3.04	3.07	2.82	0.027	0.014	0.050
NPV[d]	54.33	54.44	50.10	0.863	0.0134	0.080
Fat digestibility, %	92.3	92.4	91.0	0.45	0.081	0.18
Digestibility of carbohydrates[e], %	10.4	10.8	25.5	5.54	0.102	0.337
GE[f], MJ/kg feed	24.26	24.41	24.80	–		
Digestibility of GE, %	81.2	81.2	80.3	1.20	$F<1$	$F<1$
DE[g], MJ/kg feed (± standard deviation)	19.6 ± 0.5	19.8 ± 0.7	19.9 ± 0.2			
Energy consumption, MJ DE/fish	6.612	6.508	6.448	0.171	$F<1$	$F<1$
Energy utilization, MJ DE/g growth	14.710	14.705	15.853	0.247	0.017	0.105
Energy accretion, MJ/fish	4.502	4.355	4.012	0.152	0.063	$F<1$
Energy utilization, MJ DE consumed/MJ accredited	1.469	1.496	1.609	0.026	0.009	0.224

[a] S.E.M. = standard error of mean.
[b] P = probability for significance.
[c] Protein efficiency ratio (PER = weight gain/protein consumption).
[d] Net protein value (NPV = carcass protein gain/protein consumption).
[e] Calculated by difference. See Table 1.
[f] GE = gross energy.
[g] DE = digestible energy.
[h] See Table 4, footnote [c] for explanation.

17.51 MJ/kg, respectively. The two sets of figures are determined independently and show deviations of only around 1%.

There was no significant difference in energy accretion between the diets, but numerically, energy accretion decreased with increasing substitution and was 3% and 11% lower on Diet 2 and Diet 3, respectively, compared with Diet 1. Efficiency of energy utilization (i.e. growth per unit DE consumed) decreased significantly linearly with increasing substitution, and with no significant deviation from linearity. Energy utilization for energy accretion (i.e. energy accretion per unit DE consumed) decreased significantly linearly with increasing substitution, with no significant deviation from linearity.

4. Discussion

The protein digestibility in the fish meal found in this study is higher than that reported by Anderson et al. (1992) for Atlantic salmon, but at the same level as that found by Hajen et al. (1993) for chinook salmon and Aksnes and Opstvedt (1998) for rainbow trout. This study found similar protein digestibility in soybean meal as previously determined in

Atlantic (Anderson et al., 1992) and chinook salmon (Hajen et al., 1993), but considerable lower than found by Cho and Kaushik (1990) for rainbow trout. The digestibility of protein in maize gluten meal in this study is slightly higher than that found for Atlantic salmon by Anderson et al. (1992), but considerably lower than that determined in rainbow trout by Cho and Kaushik (1990). It is possible that the disagreement between the present figures and those reported by Cho and Kaushik (1990) is not only due to the fact that they refer to different fish species but also due to different methods used for feces collection (Anderson et al., 1995). Thus this study used fecal stripping while Cho and Kaushik (1990) obtained the feces by wet collection, which lead to leaching. This would, in particular, have affected the digestibility of the full-fat soybean meal since it was found in this and previous studies (Storebakken et al., 1998) that the feces from the fish fed the full-fat soybean meal have looser consistency than those fed the high fish meal diet. The finding that protein digestibility was lower than would be expected from the digestibility of the feed ingredients for the fish meal diet while the opposite was the case for the substituted diets is of considerable interest since it shows that interactions between feed ingredients may be expected in mixed diets (Aksnes and Opstvedt, 1998). The digestibility of GE in fish meal found in this study is lower than that previously determined in rainbow trout (Aksnes and Opstvedt, 1998) but higher than that determined for chinook salmon (Hajen et al., 1993). The digestibilities of GE determined in full-fat soybean meal and maize gluten in our study are again lower than those reported by Cho and Kaushik (1990) for rainbow trout, which may be due to the reason given above.

The finding that the digestibility of GE was similar in all diets was unexpected since energy digestibility was significantly higher in the fish meal than in the full-fat soybean meal and maize gluten meal. The reason for this anomaly may be that carbohydrate digestibility increased more with increasing substitution of fish meal than protein and fat digestibility was decreased. However, carbohydrate digestibility in this study was low for all diets compared to previously published figures (Arnesen and Krogdahl, 1993, 1996; Aksnes, 1995; Hemre et al., 1995). It has been shown (Arnesen and Krogdahl, 1993) that carbohydrate digestibility in Atlantic salmon is lower in crude compared with heat-treated wheat (e.g. extruded). It has also been shown to decrease with increasing levels of inclusion (Aksnes, 1995; Hemre et al., 1995; Arnesen and Krogdahl, 1996). Thus Aksnes (1995) found starch digestibility to decrease from 81% to 43% when the level was increased from 2% to 17%. Similarly, Hemre et al. (1995) found that starch digestibility decreased from 91% to 76% when the level was increased from 5% to 31%, and Arnesen and Krogdahl (1996), that digestibility were 48%, 41% and 28% for diets which contained 15%, 30% and 45% of extruded wheat meal and 17.8% and 0% of wheat bran. The present study did not determine the digestibility of starch, but that of the combined carbohydrates calculated by difference (i.e. N-free extractives plus crude fiber) and include components with low digestibility. The digestibility found in this study can therefore not be compared directly with those reported for starch. Assuming that starch was the only digestible fraction of the carbohydrates, starch digestibility of 11.3%, 13.6% and 44.4% may be calculated for Diet 1, Diet 2 and Diet 3, respectively (see Table 3). Thus, while the calculated digestibility of starch for Diet 3 is in line with previous reports, those for Diet 1 and Diet 2 are still low. The dextrinisation of the starch was high in all diets, and cannot explain the low carbohydrate digestibility, neither can it be explained by the level of starch in the diets when judged from previous results. This study determined the digestibility in smolt of about 500 g which is not very different from that of the salmon used in previous studies (Aksnes, 1995; Hemre et al., 1995). We are therefore not in the position to give a plausible explanation for the low carbohydrate digestibility found in the present study. However, it cannot be excluded that the particular fish used in this study had a low capacity for carbohydrate digestion. Based on the finding in the present study, it will be prudent to use lower carbohydrate levels than those reported herein in practical feeding.

Under good practical conditions in sea cages, Atlantic salmon of this size and kept at the same water temperature would be expected to have a specific growth rate of about 1.80. Thus the growth rate achieved in the present study is suboptimal to excellent practical condition, but comparable to that expected for fish held under experimental condition in tanks. It is possible that the difference between diets would be greater in faster-growing salmon held under practical conditions. This study, therefore, does not give definite quantitative answers for practical feeding situations; it should be confirmed under practical conditions comprising a whole production period in order to get the quantitative data. The more rapid growth in practical situation may increase between treatment differences.

In general, the graded substitution of fish meal with vegetable proteins gave only minor differences in numerical values of the different criteria between Diet 1 and Diet 2 but more pronounced effects from Diet 2 to Diet 3. Despite this general trend, in most instances when significant differences between diets were found, deviations from linear effects were insignificant, i.e. the effect being proportional with the level of substitution. For each percent of fish meal being replaced, growth decreased by 1.5 g. It appears that part of the growth depression due to the lowest substitution (Diet 2) was compensated by a decrease in body fat content and an increase in the water content. We are not aware of previous studies where a mixture of full-fat soybean meal and maize gluten meal has replaced fish meal in salmon diets. However, several studies (Olli et al., 1994, 1995; Bjerkeng et al., 1997; Storebakken et al., 1998; Carter and Hauler, 2000; Refstie et al., 2001) have explored the effect of replacing fish meal with different soybean products. In agreement with our results, previous studies have shown drastic reductions in growth when 30% or more of the protein in high quality fish meal were replaced with feed grade soybean products (Olli et al., 1994, 1995; Storebakken et al., 1998), while no reductions were observed when purified soybean concentrate was used (Olli et al., 1994; Refstie et al., 2001). Neither did Carter and Hauler (2000) find reduced growth when up to 33% of a nonspecified fish meal was replaced by standard soybean meal. Replacement of smaller amounts of the fish meal by soybean products, e.g. 20% or less, have usually not lead to significant growth depression (Olli et al., 1995; Bjerkeng et al., 1997; Refstie et al., 2001) in agreement with our results. Our data revealed that despite the fact that there was no significant reduction in growth caused by the lowest replacement, the overall effect of replacing up to 56% was linear and proportional with the level of replacement. This finding support the result of the early studies by Olli et al. (1994). It is possible that differences between studies in this regard are due the salmon reacting to the lowest replacements by reduced accretion of tissues. This was supported by the finding that FCR was only affected at the highest substitution level. It is known that nonrefined soybean products contain toxic compounds, which may include lectins that cause diarrhea and pathologic changes (SBM-induced enteritis) in the intestines (Baeverfjord and Krogdahl, 1996). It is unlikely that there is a non-effect level for these toxic compounds, but that the manifestation of the effects may vary between animals.

Substitution of fish meal increased water content and reduced organic matter and energy in the whole fish. This trial lasted for 14 weeks only. It is therefore not possible to draw conclusions with regard to a whole production period. However, Refstie et al. (2001) observed a reduction only in viscera lipids for diets where up to 30% of the fish meal was substituted for a whole production period, while no effect was found on carcass composition.

It is commonly accepted in experimental comparison of feed ingredients that other experimental factors, including nutrient content, should be kept as similar as possible. This may create problems with regard to composing diets with optimal ingredient composition when the ingredients have widely different chemical composition, as is the case for fish meal, full-fat soybean meal and maize gluten meal. In the present study we chose to balance the diets with regard to GE, crude protein, total carbohydrates and fat in addition to lysine and sulfur-containing amino acid. Due to the differences in carbohydrate composition, the content of starch varied between the diets, and may not have been optimal in all diets. Thus Diet 1 (high fish meal) had higher content of starch than would be used for Atlantic salmon smolt of the present size under practical conditions, and may have affected the production performance negatively. Further, the content of lysine and sulfur containing amino acids was above requirement. Although it is unlikely that this has affected production performance negatively (Anderson et al., 1991; Sveier et al., 2001), the need to equate dietary amino acids for experimental purposes would offset the savings resulting from substituting fish meal with vegetable proteins. In practice, lower amino acids contents of substituted diets may be accepted. The results from the present study can therefore not be used directly to calculate feed costs for different substitutions in practical conditions. An indication of cost effects of substitution may be obtained from the ingredient cost of the experimental diets which is based on current raw material prices[1] of £334.22, £336.47 and £329.31 ton^{-1} for Diet 1, Diet 2 and Diet 3, respectively. Although compared to Diet 1, Diet 2 saved 2.0% and Diet 3 4.1% in ingredient cost, feed conversion on Diet 3 was, however, increased by 6.6%, which would have increased feed cost by additional 2.5%. Furthermore, these calculations do not take the savings due to faster growth using Diet 1.

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[1] Scotland, November 2002: herring meal, £475 ton^{-1}; herring oil, £337 ton^{-1}; maize gluten meal, £300 ton^{-1}; wheat £66 ton^{-1}; lysine, £1850 ton^{-1}; methionine, £1800 ton^{-1}.

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