

GROWTH AND FEED CONVERSION IN COD (*GADUS MORHUA*) ON DIFFERENT FEEDS, RETENTION OF SOME TRACE ELEMENTS IN THE LIVER

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ABSTRACT

Young cod were fed for 14 weeks four diets composed to give the average energy distribution in commercial salmon feeds: 46% protein, 40% fat and 14% carbohydrate. A feed based on squid mantle (30% dry matter) gave an exceptional good growth, feed conversion and nutrient retention. A dry pellet (90% dry matter) and a moist pellet from silaged fish (60% dry matter), both typical commercial salmon feeds, gave the same acceptable feed conversions and nutrient retentions, but the dry pellet gave a lower growth. A moist feed (60% dry matter) based on frozen capelin gave inferior growth, feed conversion and nutrient retention. Cod given salmon feeds increase their hepatosomatic index as the liver is the sole storage organ of fat and consequently the fat content should be reduced in cod feeds.

The four feeds had widely varying contents of the trace elements iron, zinc, copper and selenium. The origin of these contents, viz. natural contents in the feed components, mineral fortification of the feeds and processing contamination, are discussed. The liver was found not to be a storage organ for these elements, in contrast to the liver of salmonids. The need for information on trace element requirements of marine species is discussed. The lack of proper experimental design for such information is pointed out.

INTRODUCTION

The important salmon production in Norway (80 000 t in 1987) is backed up by a large-scale fish feed production. Salmon feeds are composed to fill the nutritional needs of the salmon, and are classified according to their contents of dry matter. "Wet" feeds (up to 30% dry matter) contain mostly pelagic fish species as capelin and herring, while "moist" feeds of 60% dry matter may be produced from formic acid silaged fish mixed with a "pellet"

meal of fish meal and extruded wheat. Wet and moist feeds are often produced at the fish farms according to needs. Dry pelleted feeds (> 90% dry matter) are produced by the major mills from fish meal, fish oil and extruded wheat. All three types of fish feed generally provide the following energy distribution: 40–50% from protein, 30–40% from fat and 10–20% from carbohydrate. Mixtures of vitamins and minerals are added in proportion to the dry matter contents, with no regard to the original contents in the different feed components. Salmonids accumulate trace elements in the liver, which may reach toxic concentrations from unintentional dietary overloads.

The successful hatching and mass-production of fry of cod (*Gadus morhua*) (Kvenseth and Øyestad, 1984; Øyestad et al., 1984), has increased the interest in the commercial farming of cod in Norway. There is a need for information on the growth of cod on commercial salmon feed, and on the specific nutrient requirements of cod. The present paper reports on growth, feed conversion and retention of protein and fat in cod given four feeds of different commercial and experimental types, having the energy distribution of salmon feed. Further, contents in the feeds and liver retentions of iron, zinc, copper and selenium were determined.

MATERIAL AND METHODS

Fish and diets

Cod, hatched and reared at the Aquaculture Research Station, Austevoll¹, averaging 45 g, were used in the experiment. The fish were weighed, distributed into four groups (A, B, C and D), each of 40 fish, in 175 L aquaria with the conditions described by Lie et al. (1986) and were fed to satiation three days a week (Monday, Wednesday and Friday). Water temperature was 8 ± 1 °C, and salinity 32 ± 0.2 ‰.

The four feeds used were (A) a commercial dry pellet, (B) a moist feed based on capelin, (C) a commercial silage based on moist pellet and (D) a wet feed based on squid mantle. The composition of the feeds were:

- A: Whole fish meal (58%), extruded starch (24%) and capelin oil (18%).
- B: Frozen capelin (44%), whole fish meal (40%), capelin oil (6%) and extruded starch (10%).
- C: Fish silage (50%) and pellet mix (50%) (consisting of whole fish meal and extruded starch).
- D: Squid mantle (86%), capelin oil (8.4%) and dextrinized potato starch (5.6%).

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Vitamins and minerals were supplied through the carbohydrate source. Analytical values including four trace elements, estimated metabolizable energy and the proportion of energy from protein, fat and carbohydrate in each diet are shown in Table 1. The values used in the calculation of these figures were 17.6 kJ/g protein, 33.6 kJ/g fat and 12.6 kJ/g carbohydrate.

At the start of the experiment 20 fish (mean weight 42.1 g) were taken from the stock for determination of gross composition of whole fish and liver. After a feeding period of 14 weeks the fish were weighed and samples collected for analyses. All samples were stored at -20°C until analysis.

Analytical methods

Feed, whole fish and liver were analysed in pooled samples for water, fat and ash by conventional methods. Protein ($\text{N} \times 6.25$) was determined by the micro-Kjeldahl technique according to Crooke and Simpson (1971) and fat was extracted with ethyl acetate. Carbohydrate was estimated by difference.

All samples for trace element analyses were digested in a mixture of concentrated nitric- and perchloric acid (suprapure quality, 9 : 1) as described by Julshamn et al. (1982), and the elements were then analyzed by atomic absorption spectrophotometry (AAS). Iron, copper and zinc were measured by flame atomic absorption as described by Julshamn and Andersen (1983). Selenium was measured by graphite furnace AAS (GFAAS) and the procedure, apparatus and instrument settings are described in details by Maage et al. (1989). The accuracy of the element analyses was tested in an intercalibration study arranged by International Council for the Exploration of the Sea, ICES (Berman, 1984) as well as by analysing appropriate standard reference material from the National Bureau of Standards, and all methods were found satisfactory with regard to both tests.

RESULTS AND DISCUSSION

Table 1 shows that all four experimental feeds had an energy distribution used in most commercial feeds for salmon farming, with average energy percentages of 46 for protein, 40 for fat and 14 for carbohydrate. The moisture content of the feeds varied, however, from 8% in the dry feed pellet, 31–37% in the two moist feeds, to 60% in the wet feed based on squid mantle. The latter feed (D) was chosen for its high palatability to cod, and therefore used as a reference for growth comparisons. The energy concentrations of the feeds varied with the dry matter contents, from 6 to 17 MJ per kg wet weight.

Table 1. Composition (g/100 g), trace elements (mg/kg wet weight) and calculated energy contents and distributions in the feeds.

	A	B	C	D
Dry matter	91.9	68.7	62.8	30.8
Protein	42.0	37.4	32.0	13.9
Fat	20.8	17.2	11.2	8.1
Ash.....	6.7	6.7	5.0	1.2
Fe.....	102	227	328	224
Zn.....	152	65.2	262	70.4
Cu.....	6.1	7.0	14.8	5.0
Se.....	0.8	1.7	1.0	3.5
MJ/Kg	17.2	13.3	11.2	6.1
Energy distribution (%)				
Protein	43	50	50	40
Fat	41	43	34	44
Carbohydrate	16	7	16	16

A: Dry feed, commercial.

B: Moist feed cont. whole capelin.

C: Moist feed cont. silaged fish.

D: Wet feed cont. squid mantle.

Table 2 shows the growth responses to these feeds. As expected, feed D was well accepted, and gave the highest feed intake, highest growth and also best feed conversion. The moist feed made from silaged fish (C) gave the second best growth response, 76% of feed D, while the dry feed pellets (A) was less well accepted and gave a weight gain of 65% of that of feed D. However, both feed A and C showed a feed conversion factor of 1.1. We have seen in previous experiments with cod and other marine species that dry feed pellets are not well accepted. Feed B gave a surprisingly low growth response of only 51% of that of feed D. This was lower than expected from the feed intake which was equal to that of the fish fed feed C, and gave an inefficient feed conversion of 1.6.

The retention values for the four groups (Table 2), correspond well with the growth responses. Feed D gave the highest retentions of both protein and fat, while feed B gave the lowest, with values lower than acceptable in commercial breeding. Both protein and fat retentions were a little higher in the group on feed A, than in the group on feed C, i.e. the low feed intake of the dry pellet feed did not result in reduced nutrient retention. The hepatosomatic index is included since cod is a "lean" fish which stores its fat exclusively in the liver, and the liver increases in size with increased fat uptake,

Table 2. Growth, liver weight, feed intake and retention of protein and fat in cod given the four feeds for 14 weeks.

	A	B	C	D
Start weight, g	41.5	41.7	44.6	47.5
S.D.	10.5	10.6	12.4	13.6
Final weight, g	86.6	77.6	101.3	126.9
S.D.	34.5	32.5	40.0	56.9
Weight gain, %	109	86	127	167
Energy intake, MJ	1.14	1.45	1.44	1.66
Feed conversion	1.1	1.6	1.1	0.8
Hepatosomatic index, %	11.3	8.9	10.2	12.0
PER	2.1	1.1	1.8	2.7
PPV	0.26	0.15	0.23	0.32
Fat retention				
Whole fish, %	44	20	39	52
Liver, %	39	18	39	47

Feed conversion: g dry feed/live weight gain.

Protein efficiency Ratio (PER): Weight gain/Protein intake.

Productive Protein Value (PPV): Protein gain/Protein intake.

Hepatosomatic index = Liver weight * 100/Final weight, was 7.1% when the feeding experiment started.

as also seen from Table 3. A high liver content is not acceptable in commercial cod breeding, and we have found in earlier experiments that a fat energy concentration not higher than 20% keeps the cod liver at a normal size of 6 to 8% (Lie et al., 1988). The fat contents in the salmon feeds used in the present experiment were obviously too high, giving hepatosomatic index of 10–12% in the fish, with exception of that in group B with a very low nutrient utilization. The gross composition of fish and liver (Table 3) shows that all groups increased their fat reserves during the experiment.

The value PPV, or protein retention, is important in optimizing a cod feed as it indicates a high muscle protein growth, while the PER value increases also with increased liver fat stores. The utilization of dietary protein by fish may be greatly influenced by dietary manipulations of protein and energy (Cho and Kaushik, 1985). Both Lee and Putnam (1973) and Ogino et al. (1976) reported that the PER values of rainbow trout (*Salmo gairdneri*) were negatively correlated to the protein/calorie ratio of the diets. Adron et al. (1976) found that PER of turbot (*Scophthalmus maximus*) increased progressively as the energy levels in the diet rose with constant protein levels i.e. reduced protein/energy ration in the diet.

Table 1 shows a wide range of concentrations of the trace elements iron, zinc, copper and selenium in the four feeds. This contrasts with the major

nutrient distribution in the feeds which varied only slightly. The dry weight concentration of iron in the feeds varied seven times, zinc and copper four times and selenium more than ten times. The feeds A, B and C were all composed of capelin and whole wheat. The natural concentrations (mg/kg dry weight) of these elements in capelin and wheat are for iron 67 and 48, for zinc 60 and 28, for copper 3 and 6 and for selenium 4 and < 0.1, respectively (Julshamn et al., 1978; Lunde, 1968 and 1973). Lunde (1973) found the following average values in commercial fishmeals from capelin: iron 288 mg/kg, zinc 118 mg/kg, copper 4.3 mg/kg and selenium 1.1 mg/kg. The high iron contents in these meals originated from contamination during processing (Søvik et al., 1981). Fishmeals are now produced using indirect drying, and have lower iron contents from processing. The mineral mixture used in the feeds were of a standard composition, supposed to give the same levels of trace elements to the different feeds. It may be assumed that the high levels of iron in feeds B, C and D, as well as the high levels of zinc and copper in feed C arose from processing contamination. A high selenium concentration was found in feed D, based on squid mantle. This was much higher than found in other samples of squid mantle, but occasional high levels of selenium have been found in marine invertebrates (Eisler, 1981).

The liver has often been proposed as a storage organ for trace elements in warm blooded animals as well as in fish (Underwood, 1977, Knox et al., 1982). Liver values of the trace elements iron, zinc, copper and selenium were therefore determined in the present study (Table 4). Previous experiments with rainbow trout and salmon have shown quite high levels of these elements in the liver. Knox et. al. (1982) found 30 mg/kg iron in rainbow

Table 3. Gross composition of whole fish and liver, % wet weight.

	Whole fish			
	A	B	C	D
Dry matter	27.0	25.3	25.3	26.8
Protein	13.1	13.2	13.1	12.5
Fat	8.3	6.9	6.7	9.2
Ash.....	2.6	2.7	2.5	2.3
	Liver			
Dry matter	71.8	73.9	71.1	75.6
Protein	3.8	4.0	4.2	4.1
Fat	60.7	63.7	59.3	64.9
Ash.....	0.5	0.4	0.5	0.4

At the start of the experiment the values were:

Whole fish: Dry matter: (24.5), protein: (13.5), fat: (5.7) and ash (2.8).

Liver: Dry matter: (69.2), protein: (7.3), fat: (57.3) and fish: (0.6).

Table 4. Total intake (mg), concentration (mg/kg w.w.), total increase (μg) and retention in % for iron, copper, zinc and selenium in liver of cod fed the four feeds for 14 weeks.

	A	B	C	D
<i>Iron</i>				
Intake.....	6.8	24.7	42.2	61.0
Concentration.....	2.5	6.3	3.6	4.2
Increase.....	1.4	20.4	12.5	36.9
Retention.....	0.02	0.08	0.03	0.06
<i>Zinc</i>				
Intake.....	10.1	7.1	33.7	19.2
Concentration.....	7.6	9.0	8.2	7.8
Increase.....	43.4	31.7	51.5	84.1
Retention.....	0.4	0.5	0.2	0.4
<i>Copper</i>				
Intake.....	0.40	0.76	1.90	1.36
Concentration.....	3.8	4.2	2.4	2.9
Increase.....	27.4	18.9	13.8	33.1
Retention.....	0.7	2.5	0.7	2.4
<i>Selenium</i>				
Intake.....	0.05	0.19	0.13	0.95
Concentration.....	0.15	0.12	0.19	0.18
Increase.....	0.31	0.12	1.15	1.86
Retention.....	1.3	0.06	0.9	0.2

trout (weighing 150 g) given 300 mg/kg in the diet, and Julshamn et al. (1989) found 200 mg/kg in adult salmon fed a diet also containing 300 mg/kg. According to Knox et al. (1984), Lanno et al. (1985) and Julshamn et al. (1989) the copper contents in the liver of salmonids correlated well with the dietary intakes. Accumulation of selenium in salmonids livers was seen by Poston et al. (1976), Hilton et al. (1980) and Julshamn et al. (1989). Recent analyses of trace elements in livers of adult salmon (4.5 kg) given similar commercial feeds as in the present experiments (Julshamn et al, 1989) gave values around 200 mg/kg for iron, 30 mg/kg for zinc, 65 mg/kg for copper, and 2.5 mg/kg for selenium, all based on wet weight.

Table 4 shows that the cod livers had much lower concentrations of the four elements than found in salmonid livers. Further, little correlation could be found between liver concentrations and feed intakes. Calculated liver retentions in percent of the dietary intakes were extremely small, for iron less than 0.1, for zinc less than 0.5, for copper less than 2.5 and for selenium less than 1.3 (Table 4). It seems therefore that the liver contents of these trace elements are dietary independent in the cod and that the cod liver is

not a storage organ for trace elements. Table 5 gives average values for the same four elements in the livers of wild adult cod of varying size. The values are somewhat higher than those in Table 4 (from young cod of less than 100 g), but far below values recorded for wild salmon (Julshamn et al., 1989).

Feeds are fortified with essential trace elements to avoid deficiency symptoms and reduced growth. However, no experimental data are available about the requirements of these nutrients to marine fish species. Experiments to ascertain trace element requirements to fish are, in general, difficult to design. Among the problems to be considered are the biological availabilities from feed and seawater, interactions between trace elements and other nutrients as well as their chemical form relative to digestion, absorption and possible storage reserves in the body. Even for salmonids feed producers have insufficient knowledge to ensure optimal compositions in their feed recipes. For practical purposes additions of trace elements to fish feeds should be based on knowledge of their natural contents in the feed components.

Table 5. Concentrations of iron, copper, zinc and selenium (mg/kg wet weight) in the liver of wild cod.

	Fe	Cu	Zn	Se
Number anal (N).....	20	52	52	10
Average.....	15	2.4	17	0.55
S.D.....	10	1.3	10	0.20
Min.-max.....	2.3-49	0.5-6.1	6-37	<0.02-0.6
Fish weight, g.....	320-1950	800-3500	800-3500	350-1200

Reduced growth has been shown for low intakes of iron, copper and zinc to salmonids. Still, Zeitoun et al. (1976) found that fish may satisfy their iron requirement through uptake from water and this may be true also for other trace elements for marine fish species. On the other hand, high intakes may be toxic, as Desjardins et al. (1987) found for iron, acting as a lipid oxidation catalyst. High intakes of copper and selenium are also known to be toxic for salmonids (Knox et al., 1982 and Hilton et al., 1980).

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