

## APPARENT AVAILABILITY OF FAT AND INDIVIDUAL FATTY ACIDS IN ATLANTIC COD (*Gadus morhua*)

By

EINAR LIED and GEORG LAMBERTSEN

Institute of Vitamin Research

Directorate of Fisheries

N-5013 Nygårdstangen/Bergen, Norway

### ABSTRACT

Apparent availability values for fat and the major saturated and monoene fatty acids were determined by the indirect indicator method in the digestive tract of cod, divided into the stomach, the pyloric caeca region, the anterior, the middle and posterior ileum and the rectum. In two experiments, cod were given saithe fillet with 7% capelin oil and containing chromium(III)-oxide and whole sprat containing titanium(IV)-oxide. Further, cod, which had preyed on capelin were caught in the Barents Sea, and availability values were measured using calcium as an internal indicator, and the stomach contents as reference. Fat digestion started in the pyloric caeca region, and only minor amounts of fatty acids were present in the stomach. The fatty acids formed during the fat digestion were found partly as free ether extractable, and partly as «bound» fat, probably salts of fatty acids with divalent ions, such as calcium and magnesium. The bound fat was ether extractable after treatment of the intestinal content with hydrochloric acid. Omitting the acid treatment before fat extraction results in overestimation of the apparent availability values. Most of the fat was absorbed in the pyloric and anterior ileal region from cods on a mixed diet, whereas the fat absorption took place over a longer intestinal distance in cod fed whole fish. Reliable samples for the determination of apparent availability of fat in fish should be collected from the posterior ileum. Apparent nutritional availability values for fat of 87%, 70% and 85% were found for cod fed a mixed diet, whole sprat and on a natural feed, respectively. The corresponding available energy values were 8.27, 6.60 and 8.08 kcal/g fat. Evidently these values vary with the form of feed taken. The long chain monoene fatty acids 20:1 and 22:1, particularly the latter, accumulated in both extractable and bound fat in the contents of the last segments of the intestine. Palmitic and stearic acids accumulated in the soaps in the first three segments of the intestine.

### INTRODUCTION

Lipids constitute the principal non-protein energy source in natural diets of carnivorous as well as omnivorous fish. The nutritional value of lipids, however, depends on their availability to the fish. Although several studies have been made to establish the lipolytic processes in the digestive tract of fish as reviewed by COWEY and SARGENT (1972, 1977, 1979) and KAPOOR et al. (1975), comparatively few studies concern the nutritional availability of fat in fish. Such studies have been carried out mainly on rainbow trout (*Salmo gairdneri*) (TRYAMKINA and SHCHERBINA, 1974, STRELTSOVA and OLSHANSKAYA,

1974, CHO and SLINGER, 1979, WINDELL et al, 1978, TAKEUCHI et al., 1979, AUSTRENG et al., 1979), but also on carp (*Cyprinus carpio*) (TAKEUCHI et al., 1979) and channel catfish (*Ictalurus punctatus*) (ANDREWS et al., 1978) using chromium(III)-oxide as the indigestible indicator substance. By means of the balance-sheet method BEAMISH (1972) studied the lipid digestion in large-mouth bass (*Micropterus salmoides*). Similarly SARGENT et al. (1979) studied the assimilation of lipids in herring (*Clupea harengus*) and rainbow trout.

No information could be found on the fat availability in cod (*Gadus morhua*). The results presented here were obtained from a study on the use of chromium(III)-oxide and titanium(IV)-oxide as external indicator substances, and further the use of calcium as an internal indicator substance in nutrition experiments on captive and wild cod (LIED et al., 1982). In a previous paper further results from the same study were given on the nutritional availability of protein and amino acids (LIED and NJAA, 1982).

## METHODS

### *Experimental*

Details regarding the fish obtained for the experiments and their treatment are given in the two preceding papers of this study (LIED et al., 1982, LIED and NJAA, 1982). Only pertinent information on the three experiments are repeated here.

Expt 1 consisted of 70 cod obtained at different times from different localities off the western and northern coast of Norway. The fish weighed from 1.1 to 1.8 kg, and were force-fed moist pellets with 86% minced fish fillet from saithe (*Pollachius virens*), 7% capelin oil and 7% dextrin, and with 0.1% chromium(III)-oxide added to the mixture. Each fish was fed 16 g per kg body weight. The gastric contents were collected at 4, 8 and 12 hrs., whereas the full gastrointestinal contents were sampled at 24, 36, 48 and 72 hrs. after feeding. Each sample was pooled from 10 randomly selected fish.

Expt 2 was based on cod averaging 1.5 kg, given whole sprat (*Sprattus sprattus*) each weighing on an average 15 g. Immediately before feeding 0.1 ml of a suspension of titanium(IV)-oxide was injected into the stomach of each feed fish giving a wet-weight concentration of the indicator substance of 0.5%. The cod were fed *ad lib.* twice daily for 9 days.

Finally 15 cods caught at three trawling stations on the Skolpen Bank (Barents Sea) were sampled aboard the R/V «G. O. Sars». All fish had fed on capelin (*Mallotus villosus*) as their sole food source. Immediately upon the catch the fish were gutted and the alimentary tract ligated into segments, removed from the fish and frozen. The frozen samples were brought to the laboratory, thawed and analysed as described below.

The gastrointestinal tract was divided into six segments by ligations at the oesophagus, immediately before the pyloric ceca, at the first cranial, the first caudal, the second cranial bend of the intestine, at the ileorectal valve and at the anus. Starting with the stomach, the segments were numbered from one to six, representing 12, 6, 32, 30, 12 and 8% of the total length, respectively. Pooled samples were prepared and freeze dried as described by LIED et al., (1982).

### *Analyses*

Aliquots were digested for the determination of nitrogen, chromium and titanium by the micro-Kjeldahl technique in a thermostat-regulated heating block (TECATOR 40). Titanium (IV)-oxide was determined in the digest according to NJAA (1961).

Chromium was determined by atomic absorption spectrophotometry as described by LIED et al., (1982). Calcium was determined after nitric acid/perchloric acid digestion by AAS according to JULSHAMN and BRAEKKAN (1975). Fat was extracted by diethyl ether from 0.3–0.5 g of the pooled, freeze-dried samples. After three extractions the combined ether extracts were evaporated *in vacuo* and the fat residue weighed. A further aliquot as well as the sample residue after fat extraction was digested with 2M hydrochloric acid for 1 hr. at 90°C and again extracted with diethyl ether as above. The three fat extracts (total, before and after the HCl treatment) were converted into fatty acid methyl esters by a conventional method, using boron trifluoride as a catalyst (LAMBERTSEN et al., 1966). The fatty acid methyl esters were separated on a PERKIN-ELMER 900 gas chromatograph using a 6', 1/4" glass column with 10% EGSS-X in chromosorb as the stationary phase. The peaks were identified by comparison of retention times with standard series of fatty acid methyl esters, and integrated in a computer system (WANG 2200B).

The apparent nutrient recovery (ANR) relative to the indicator concentration either for fat or fatty acids was taken as a measure for apparent nutrient availability (ANA). The relationship between ANR and ANA is given by:

$$ANA = 100 - ANR$$

Percentage ANA was calculated by the formula:

$$ANA = 100 - 100 (S_q/I_q)$$

were  $I_q$  and  $S_q$  are the ratios between nutrient concentration and indicator concentration in the feed and the gastrointestinal segment in question, respectively. In wild fish the calculation of availability was related to the gastric content of nutrients and calcium (LIED et al., 1982). The ratio  $I_q$  was replaced by  $G_q$ , which is the ratio nutrient concentration over calcium concentration in the stomach.

## RESULTS AND DISCUSSION

A digestive lipolytic activity has been demonstrated in the intestine of several fish species (BROCKERHOFF and HOYLE, 1965, BROCKERHOFF, 1966, SASTRY, 1975, PATTON et al., 1975). BROCKERHOFF (1966) in studies on Atlantic cod, found that dietary fat was hydrolysed to diglycerides and monoglycerides by a digestive lipase which preferentially attacked the primary alcoholic esters, but was not stereospecific. This enzyme corresponded in its action to the pancreatic lipase of mammals. Investigations carried out by PATTON et al. (1975), however, indicated that digestive lipase in fish intestine was non-specific in its activity and hydrolysed both primary and secondary alcoholic esters, cleaving fatty acids from all three positions of the glycerol. This enzyme possibly compete with pancreatic lipase (glycerol ester hydrolase) as a major fat digesting enzyme in fish intestine, producing free fatty acids and glycerol as the bulk of triacyl digestion in fish.

Lipase activity in stomach extracts has been observed by NAGASE (1964) for tilapia (*Tilapia mossambica*) and by MACKAY (1929) for eel-pout (*Zoarces anguillaris*) suggesting a possible gastric digestion of lipids in fish. Thin layer chromatographic fractionation of the lipids from the gastrointestinal contents in Expt 2 showed only triglycerides to be present in the stomach (Fig. 1). Lipolytic activity in the stomach of cod therefore was negligible in our experiments. It is likely that the presence of gastric lipolytic activity in fish, if any, is due to regurgitation of digest from the intestine rather than to gastric secretion of a digestive lipase. The lipid fractions found in samples of chyme collected from the pyloric ceca region contained among other components triglycerides as well as fatty acids, the latter being present partly as extractable acids and partly in the «bound» fat fraction, ether extractable after acid treatment. In subsequent intestinal segments the lipid material was found mainly as free fatty acids, both extractable and in bound form. Thus, in cod the digestion of fat starts in the pyloric ceca region, probably mainly by the secretion into the intestinal lumen of a non-specific lipase producing fatty acids and glycerol.

A digest rich in such divalent ions as calcium and magnesium originating from a feed of whole fish as well as from the sea water, will favor the intestinal hydrolysis of fatty acids. Cod, feeding mainly on other fish species, prawns and euphausiids has a large intake of minerals. LIED et al. (1982) found that the elements calcium, iron and zinc accumulated in the gastrointestinal tract of cod fed whole sprat. Lipid determination in feed and feces in digestibility studies has been carried out mainly by extraction with diethyl ether or a mixture of methanol and chloroform according to FOLCH et al. (1957) or BLIGH and DYER (1959). Experiments with rat, mink, and suckling pigs has shown, however, that a considerable part of the fat fraction in feces may be bound in the form of salts, probably of calcium and magnesium (CHENG et al., 1949,

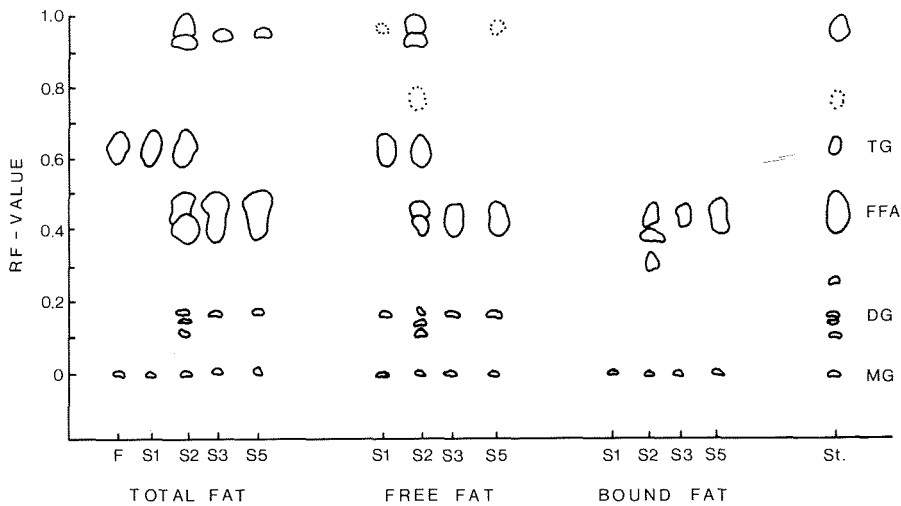


Fig. 1. Thin-layer chromatograms of the lipids in total, free and bound fat from feed and four sections of the intestinal contents from Expt 2. Plate: DC-alufolie, silicagel 60, Merck. Eluent: 20% diethylether in hexane. TG = triglycerid, FFA = free fatty acids, DG = diglyceride, MG = monoglyceride. Standard (St.): partly hydrolyzed soy bean oil.

CARROL and RICHARDS, 1958, UTNE et al., 1966, THOMSEN, 1969, NJAA et al., 1971). Such salts are only slightly soluble in diethyl ether, but may be partly soluble in mixtures of methanol and chloroform depending on the concentration of methanol in the solvent. The presence of such «soaps» therefore necessitate an acid digestion of the samples prior to the extraction with organic solvents to determine free and bound fat in feces.

The amounts of extractable and total fat in dry matter of the feed and gastrointestinal contents of captive cod fed a mixed diet containing capelin oil (Expt 1), whole sprat (Expt 2) and of wild cod on a natural diet of capelin are given in Table 1. In all feeds the total fat fraction was in the free, ether-extractable form. The percentage level of fat was slightly increased in the gastric contents as compared with the feeds, and the ratio free fat to total fat varied from 0.94 to 0.99, indicating that only a negligible part of the fat fraction was in the bound form. In the succeeding intestinal segments the ratio of free fat to total fat decreased. In fish fed a mixed, easily digestive diet, soaps were formed mainly in the pyloric caeca region, with a ratio of extractable fat to total fat of 0.29. The ratios were between 0.24 and 0.26 in the successive ileal segments and the rectum. In cod fed whole sprat the ratio of extractable fat to total fat decreased gradually from 0.95 in the stomach through 0.90 in the pyloric caeca region and 0.36 in the anterior ileum to a level of 0.16 to 0.20 in the successive segments of the digestive tract. Similarly, in wild fish the ratio

Table 1. Total fat and ether extractable fat in the feed and the gastrointestinal contents of cod on diets of saithe fillet (Expt 1), of whole sprat (Expt 2) and on a natural diet of capelin (wild cod). The values are expressed as percentage fat of dry matter.

	Feed	Gastrointestinal segment <sup>1)</sup>					
		1	2	3	4	5	6
<i>Expt 1</i>							
Ether extr. fat	19.8	22.0	4.5	2.2	2.3	2.3	3.6
Total fat	20.0	23.4	15.5	9.0	8.7	9.3	14.7
<i>Expt 2</i>							
Ether extr. fat	42.4	43.3	28.3	11.5	5.5	5.2	6.5
Total fat	42.6	45.8	31.3	31.7	28.3	32.4	32.3
<i>Wild cod</i>							
Ether extr. fat	42.0	41.9	26.5	22.3	20.5	16.3	14.9
Total fat	41.8	42.4	40.1	38.4	33.3	42.7	45.8

- <sup>1)</sup> 1 Stomach  
 2 Pyloric caeca  
 3 Anterior ileum  
 4 Middle ileum  
 5 Posterior ileum  
 6 Rectum

extractable fat/total fat decreased gradually from 0.99 in the stomach to 0.33 in feces collected from the rectum.

The experimental background for the availability values given in Table 2 were discussed by LIED et al. (1982) who concluded that based on the methods given, the use of chromium(III)-oxide and titanium(IV)-oxide as external indicators and calcium as an internal indicator were reliable in nutritional availability studies on fish. The passage of chyme through the pyloric caeca region and into the anterior ileum reduced the concentration of total fat considerably compared to that of the feed and gastric content (Table 1). In cod fed an easily digestible diet including capelin oil (Expt 1) the total fat fraction in the dry matter decreased from 23.4% in the stomach through 15.5% in the pyloric caeca region to an anterior ileal concentration of 9.0%. Relative to chromium(III)-oxide, 17.9% of the total feed fat were recovered in segment 3 (Table 2), showing that 82% of the fat absorption took place in the pyloric and anterior ileal region of the digestive tract. The apparent availability of fat was further increased by 5% to 87% in the sample collected in the posterior ileum. In cod ingesting whole sprat (Expt 2) the content of total fat in gastrointestinal samples was reduced from 45.8% in the dry matter in stomach to 32.4% in segment 5, leaving 30.5% of the fat unabsorbed relative

Table 2 A. Apparent recovery (ANR) of fat in different segments of the intestinal tract in cod fed saithe fillet (Expt 1), whole sprat (Expt 2) and in wild cod. The values are based on the total fat fraction.

Intestinal segment		Apparent recovery (%)		
		Expt 1 (Cr <sub>2</sub> O <sub>3</sub> )	Expt 2 <sup>d</sup> (TiO <sub>2</sub> )	Wild fish <sup>c</sup> (Ca)
Pyloric ceca	(S2)	54.0±4.7 <sup>a</sup>		63.5
Anterior ileum	(S3)	17.9±1.2 <sup>a</sup>	64.3	42.1
Middle ileum	(S4)	15.7±1.7 <sup>b</sup>	28.1	18.3
Posterior ileum	(S5)	13.0±1.0 <sup>b</sup>	30.5	14.9
Rectum	(S6)	12.0 <sup>c</sup>	31.0	15.0

Table 2 B. Apparent availability (ANA) of fat in cod. The values are based on the dry matter extractable fat and total fat in the intestinal content of segment 5. (Posterior ileum).

	Apparent availability (%)		
	Expt 1 <sup>b</sup> (Cr <sub>2</sub> O <sub>3</sub> )	Expt 2 <sup>d</sup> (TiO <sub>2</sub> )	Wild fish <sup>c</sup> (Ca)
Ether extractable fat	95.6±0.3	96.0	93.0
Total fat	87.0±1.0	69.5	85.1

<sup>a</sup> Mean value ± SEM of 4 samples, each consisting of pooled digest from 10 fish.

<sup>b</sup> Mean value ± SEM of 3 samples, each consisting of pooled digest from 10 fish.

<sup>c</sup> Pooled samples from 10 fish collected at 72 h.

<sup>d</sup> Pooled samples from 8 fish collected at 36 h.

<sup>e</sup> Pooled samples from 15 fish.

to titanium(IV)-oxide (Table 1 and 2). In samples from wild fish the total fat fraction in stomach and posterior ileum both accounted for 42% of dry matter. Relative to gastric levels and using calcium as the reference substance 15% of the fat was recovered in the posterior ileum.

Although the fat was absorbed mainly in the pyloric and anterior ileum region, the ileal part of the intestine was of significance for the absorption of fat in cod ingesting whole fish. Thus, in cod fed whole sprat and in wild cod only 36% and 58%, respectively, of the fat were absorbed in the pyloric ceca and anterior ileum, while additional 34% and 27%, respectively, were absorbed in the successive ileal segments. The absorption processes were terminated in the posterior ileum in all experiments. Reliable samples for the determination of the apparent availability of fat in cod fed whole feed fish should therefore preferably be collected from the posterior ileum, but pooled samples from the middle and posterior part of the ileum may be collected for fish fed man-made mixed diets.

Overestimation of the apparent availability of fat in diets for cod results when using values for the fat content obtained by diethyl ether extraction (Table 2). An acid treatment of the intestinal content is necessary to hydrolyse the fatty acid soaps followed by an extraction to obtain values for total fat. The availability based on extractable fat was  $95.6 \pm 0.3\%$  in Expt 1, significantly different ( $P < 0.05$ ) from the value of  $87.0 \pm 1.0\%$  obtained for the total value. The difference between availability values based on free and total fat was even more pronounced in cod fed whole sprat, from which the availability value of 96.0% was obtained from free fat whereas the availability value calculated from the total fat was reduced to 69.5%. The available energy from fat must be calculated from its gross energy value (9.50 kcal/g of fat) modified by its availability. Available energy values of 9.08, 9.12 and 8.84 kcal per gram of fat are obtained using the fat availability values found from the extractable fat of Expt 1, Expt 2 and of wild cod, respectively. The corresponding values based on total fat were 8.27, 6.60 and 8.08 kcal per gram of fat. The latter values from Expt 1 and from wild fish are in accordance with the caloric values of 8.0 kcal/g usually assigned to available fat in fish nutrition (BRETT and GROVES, 1979). We believe that this value is not generally applicable since the type of the diet may complicate the digestibility of fat as shown by the value of 6.60 kcal/g of fat in Expt 2.

The marine lipids available to the cod in the northern Atlantic and the Barents Sea have an easily recognisable fatty acid pattern, originating mainly from the *Calanus* copepod species (LAMBERTSEN, 1973). The compositions of the fatty acids in the feed fats of Expt 1 and 2 and in the capelin taken by wild cod, are given in Table 3. 5–10% of 14:0, myristic acid and high percentages of the long chain monoene acids 20:1 and 22:1, 5 to 20% of each, are typical for these pelagic fish species. The winter capelin oil has low contents of polyenoic acids, whereas the total lipids of the fish, including phospholipids, show higher contents of these acids.

The composition of the major saturated and monoene fatty acids in the extractable and bound lipids in the feed and the gastrointestinal contents from the cod given whole sprat and the wild cod are given in tables 4 and 5. The values for polyene fatty acids have been disregarded, as the treatment of the samples, i.e. freeze drying and for bound fat, HCl-digestion, makes the values unreliable. The seven acids given in the two tables add up to  $87\% \pm 2.5\%$  (SEM) with the exception of those from segment 2, extractable and bound of Expt. 2, which for unknown reasons were analytically unsuccessful.

It has been generally recognized that the digestibility of fats and thus fatty acids in fish are related to their melting point (LEE and SINNHUBER, 1972). In experiments with rainbow trout (TAKEUCHI et al., 1979, AUSTRENG et al., 1979) and with carp (TAKEUCHI et al., 1979) the apparent availability of fat was found to increase as the melting point decreased. AUSTRENG et al. (1979) found



Table 3. The fatty acid composition of the feed fats. (Taken from earlier analyses).

Fatty acid	Capelin oil <sup>1)</sup> (Expt 1)	Sprats <sup>2)</sup> Expt 2)	Capelin <sup>3)</sup> (Wild fish)
14:0	8.6	4.9	6.5
16:0	12.5	16.3	10.2
18:0	1.2	3.2	1.3
16:1	7.8	6.5	9.3
18:1	21.4	18.2	14.0
20:1	16.3	5.9	12.7
22:1	13.1	9.5	10.6
Sum major sat. + monoene	80.9	64.5	64.6
18:4	3.2	1.7	5.3
20:5	3.3	9.5	10.0
22:6	3.4	13.7	9.6
Other minor fatty acids	9.2	10.6	10.5

<sup>1)</sup> Winter capelin oil, unpublished analyses 1969

<sup>2)</sup> General composition, «Fiskets Gang», no. 36, 1976

<sup>3)</sup> » » » , «Fiskets Gang», no. 33, 1976

in their experiments on trout and mink an inverse relationship between the apparent availability values and chain length of the 14–18 fatty acids, while a further increase in chain length up to 22 increased the availability. In experiments on channel catfish fed animal fat ANDREWS et al. (1978) demonstrated poorer fat availability in fish kept at 23°C than in fish kept at 28°C.

The saturated fatty acids 14:0, 16:0 and 18:0 have melting points of 58°, 63° and 72°C, respectively, and the monoene acids 16:1, 18:1, 20:1 melt at 0°, 14°, 24° and 34°C, respectively. The present experiments were carried out in a water temperature of 8°C, and the wild cod was caught in waters of even lower temperatures. The low ambient temperature in the cod must reduce the availability of the free fatty acids and enhance the formation of fatty acids salts with divalent cations.

Table 4 shows a decrease of the acids 14:0 and 16:1, and an increase of the long chain acid 22:1 in the extractable fat, as compared to the feed fat composition. In the bound fat (soaps) there was a decrease in the acids 16:1 and 18:1, and increases in the three saturated acids and in 22:1, i.e. the four highest melting fatty acids. An accumulation of the two acids 14:0 and 16:0 was seen in the stomach and anterior ileum.

Table 4. The contents of saturated and monoene fatty acids in the ether extractable fat and in the bound fat in the feed and gastrointestinal contents from cod fed whole sprat (Expt 2)

Fatty acid	Feed	Gastrointestinal segment <sup>1)</sup>				
		1	2	3	4	5
<i>Ether extract</i>						
14:0	10.3	12.2	7.3	7.8	6.4	5.8
16:0	16.1	19.4	13.8	11.4	12.2	15.4
18:0	1.9	1.8	2.5	1.2	1.7	3.1
16:1	9.5	10.1	8.9	9.1	7.0	6.9
18:1	11.1	11.7	13.3	11.9	10.6	9.4
20:1	12.8	13.2	11.6	15.7	15.2	13.3
22:1	21.9	23.5	16.0	31.0	32.0	28.4
<i>Bound fat (soaps)</i>						
14:0		3.5	5.1	17.7	13.5	11.5
16:0		39.3	22.0	39.6	28.8	23.8
18:0		10.0	6.0	4.8	3.8	3.0
16:1		5.3	5.5	2.9	2.8	3.7
18:1		11.8	12.9	4.3	5.0	6.3
20:1		7.0	7.1	7.2	10.2	12.3
22:1		11.2	7.9	12.6	24.8	28.8

<sup>1)</sup> See footnote Table 1.

Similar, but more pronounced changes were found in the fatty acid compositions of the samples from wild cod (Table 5). The extractable fat from segment 5 had about half the concentration of the saturated fatty acids compared to the feed fat, and significant increases of the long chain monoenes; 20:1 and 22:1. Again, the soaps showed accumulation of the saturated fatty acids and a doubling of 22:1, whereas the two shorter monoene acids 16:1 and 18:1 decreased as compared to the feed fatty acid composition. As for Expt 2, there was an accumulation of the two highest-melting acids, 16:0 and 18:0 in the three first segments of the gastrointestinal tract.

The relative accumulation in the intestinal contents of long chain monoene fatty acids 20:1 and 22:1 as free, extractable fatty acids, may be due both to the lower solubility and to a reduced emulsifying property of these acids. The accumulation of saturated fatty acids and long chain monoene fatty acids in the bound fat parallels their melting points. The accumulation in the soaps in the upper segments of the acids 14:0 and 16:0 may possible point to a preferential first step hydrolysis of these acids.

Table 5. The contents of saturated and monoene fatty acids in the ether extractable fat and in the bound fat in the feed and gastrointestinal contents from wild cod on a natural diet of capelin.

Fatty acid	Feed	Gastrointestinal segment <sup>1)</sup>				
		1	2	3	4	5
<i>Ether extract</i>						
14:0	7.6	9.6	7.5	7.2	5.9	4.1
16:0	13.2	16.6	11.8	10.2	8.7	7.2
18:0	1.5	1.9	1.6	1.0	1.1	0.8
16:1	13.3	11.5	15.1	13.3	10.5	10.0
18:1	16.0	17.1	20.4	18.2	16.9	17.3
20:1	16.5	17.0	16.2	17.7	21.3	24.2
22:1	10.7	13.8	16.0	18.0	21.6	24.0
<i>Bound fat (soaps)</i>						
14:0		5.7	12.0	14.0	13.7	10.2
16:0		36.6	33.5	32.7	26.6	20.1
18:0		4.8	4.5	3.7	2.8	2.3
16:1		6.2	5.9	5.0	4.0	4.1
18:1		18.9	11.0	9.5	7.8	8.7
20:1		5.2	10.1	10.7	14.5	19.0
22:1		5.5	10.3	11.0	15.5	21.5

<sup>1)</sup> See footnote, Table 1.

Differences in the availability of individual fatty acids have been demonstrated in pigs (FREEMAN et al., 1968), rats (CARROL, 1958), chickens (RENNER and HILL, 1961; OPSTVEDT, 1973) mink and trout (AUSTRENG et al., 1979).

Calculated on preliminary fatty acid determinations on the samples of Expt 1, apparent availability values between 84 and 96% were found for the saturated and monoenoic acids, with a clear trend of decreasing availability with increasing melting points of the fatty acids (Fig. 2).

From Expt 2, apparent availability values were calculated from the recovered fatty acids in the total fat fractions. The values obtained were 68, 62 and 56% for the saturated acids 14:0, 16:0 and 18:0 respectively, and 86, 80, 67 and 58% for the monoene acids 16:1, 18:1, 20:1 and 22:1, respectively. The availability values for cod fed whole sprat were significantly lower than those for cod fed an easily digestible blend. Fig. 2 shows the decreasing availability values with the melting points of the two series of fatty acids, and also that the saturated acids give higher ANA-values than might be expected from their

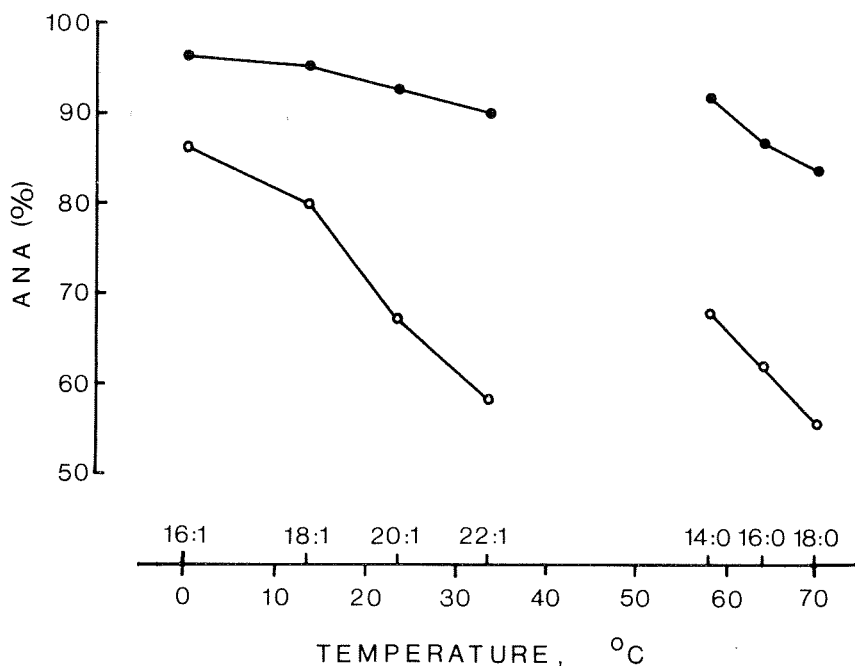


Fig. 2. Apparent nutritional availability (ANA) of single saturated and monoene fatty acids relative to their melting points. Filled rings: values from Expt 1, open rings: values from Expt 2.

high melting points, a fact possibly related to a specific earlier hydrolysis. Comparison of three experiments shows that ingestion of whole fish may delay decomposition and hydrolysis, thereby reducing the availability of fats and fatty acids.

#### REFERENCES

- ANDREWS, J. W., M. W. MURRAY and J. M. DAVIS, 1978. *J. Nutr.* 108, 749-752.  
 AUSTRENG, E., A. SKREDE and A. ELDEGARD, 1979. *Acta Agric. Scand.* 29, 119-126.  
 BEAMISH, F. W. H., 1972. *Can. J. Zool.* 50, 153-164.  
 BLIGH, E. G. and W. J. DYER, 1959. *Can. J. Biochem. Physiol.* 37, 911-917.  
 BRETT, J. R. and T. D. D. GROVES, 1979. In «Fish Physiology» (Hoar, W. S., D. J. Randall and J. R. Brett, eds.). Vol 8, 279-352. Academic Press. London.  
 BROCHERHOFF, H. and R. J. HOYLE, 1965. *Biochim. Biophys. Acta* 98, 435-436.  
 BROCKERHOFF, H., 1966. *J. Fish. Res. Bd. Canada*, 23, 1835-1839.  
 CARROL, K. K., 1958. *J. Nutr.* 64, 399-410.  
 CARROL, K. K. and J. F. RICHARDS, 1958. *J. Nutr.* 64, 411-424.  
 CHENG, A. L. S., M. G. MOREHOUSE and H. J. DEUEL, Jr., 1949. *J. Nutr.* 37, 237-250.

- CHO, C. Y. and S.J. SLINGER, 1979. In «Finfish Nutrition and Finfish Technology» (Halver, J. E. and K. Tiews, eds.). Vol. 2, 239–247. Berlin.
- COWEY, C. B. and J. R. SARGENT, 1972. *Adv. Mar. Biol.* 10, 383–492.
- COWER, C. B. and J. R. SARGENT, 1977. *Comp. Biochem. Physiol.* 57B, 269–273.
- COWEY, C. B. and J. R. SARGENT, 1979. In «Fish Physiology» (Hoar, W. S., D. J. Randall and J. R. Brett, eds.). Vol 8, 1–69. Academic Press, London.
- FOLCH, J., R. LEES and G. H. SLOANE STANLEY, 1957. *J. Biol. Chem.* 226, 497–509.
- FREEMAN, C. P., D. W. HOLME and E. F. ARMISON, 1968. *Br. J. Nutr.* 22, 651–660.
- JULSHAMN, K. and O. R. BRAEKKAN, 1975. *At. Absorp. Newsl.* 14, 49–52.
- KAPOOR, B. G., H. SMIT and I. A. VERIGHINA, 1975. *Adv. Mar. Biol.* 13, 109–239.
- LAMBERTSEN, G., 1973. *Wiss. Veröff. Deutschen Gesell. Ernährung*, 24, 25–31.
- LAMBERTSEN, G., H. MYKLESTAD and O. R. BRAEKKAN, 1966. *J. Food Sci.* 31, 48–52.
- LEE, D. J. and O. R. SINNHUBER, 1972. In «Fish Nutrition» (J. E. Halver, ed.). 145–180, Academic Press, London.
- LIED, E., K. JULSHAMN and O. R. BRAEKKAN, 1982. *Can. J. Fish. Aquat. Sci.* 39, 854–861.
- LIED, E. and L. R. NJAA, 1982. *Fisk. Dir. Skr., Ser. Ernæring* 2, 53–62.
- MACKAY, M. E., 1929. *Biological Bulletin* 56, 8–23.
- NAGASE, G. J., 1964. *Zeitschr. für vergleichende Physiologie* 49, 270–284.
- NJAA, L. R., 1961. *Acta Agric. Scand.* 11, 227–241.
- NJAA, L. R., O. R. BRAEKKAN, G. LAMBERTSEN and F. UTNE, 1971. *Nutr. Metabol.* 13, 207–221.
- OPSTVEDT, J., 1973. *Acta Agric. Scand.* 23, 217–224.
- PATTON, J. S., J. C. NEVENZEL and A. A. BENSON, 1975. *Lipids* 10, 575–583.
- RENNER, R. and F. W. HILL, 1961. *J. Nutr.* 74, 259–264.
- SARGENT, J. R., R. MCINTOSCH, A. BAUERMEISTER and J. H. S. BLAXTER, 1979. *Mar. Biol.* 51, 203–207.
- SASTRY, K. V., 1975. *Zoologica Poloniae* 25, 23–30.
- STRELTSOVA, S. V. and L. Y. OLSHANSKAYA, 1974. *Izvestiya GOSNIORKH*, 97, 23–28. Translation Series No. 3475, Fisheries Service, Department of Environment, Vancouver B.C. 1975.
- TAKEUCHI, T., T. WATANABE and C. OGINO, 1979. *Bull. Jap. Soc. Sci. Fish.* 45, 1521–1525.
- THOMSEN, K. V., 1969. *Bilag til Landsøkonomisk Forsøkslaboratoriums efterårsmøde*, København, pp 252–255.
- TRYAMKINA, S. P. and M. A. SHCHERBINA, 1974. *Izvestiya GOSNIORKH*, 97, 55–61, Translation Series No. 3597, Fisheries Service, Department of Environment, Vancouver, B.C., 1975.
- UTNE, F., L. R. NJAA and O. R. BRAEKKAN, 1966. *Norsk pelsdyrblad* 40, 539–541.
- WINDELL, J. T., J. W. FOLTZ and J. A. SAROKON, 1978. *Prog. Fish Cult.* 40, 51–55.

#### ACKNOWLEDGMENTS

We thank Mr. O. M. Smedstad of the Institute of Marine Research, Dir. of Fisheries, for collecting samples from wild cod, and Mr. R. Solbakken of the Institute of Vitamin Research for technical assistance. This research was supported by the Norwegian Fisheries Research Council grant I 711.10.