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EFFECTS OF STARVATION ON THE LIPID COMPOSITION IN MUSCLE TISSUE AND LIVER OF HATCHERY-REARED ARCTIC CHARR, SALVELINUS ALPINUS (L), FROM LAKE TAKVATN

By

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ABSTRACT

Lipid composition in muscle and liver of Arctic charr, *Salvelinus alpinus* (L.), before feeding was stopped were compared with those of fish starved for 25 and 50 days. During the 50 days of starvation period, there were reductions in live weigth and in total muscle- and liver lipid and triacylglycerols (TAG). The proportion of free fatty acids increased during the experiment. The amount of polar lipids was almost constant.

No selective utilization of fatty acids in muscle TAG was detected during starvation in liver TAG however, the (n-9) monoenes 16:1 and 18:1 and 22:1 (n-11) were selectively utilized. The (n-3) polyunsaturated fatty acids were retained during starvation. The (n-3) fatty acid 22:6 was prevalent in the polar lipid of both tissues, but the proportion was not affected by starvation. The monounsaturated fatty acid 18:1 (n-9) was selectively utilized in polar lipid of both muscle and liver.

INTRODUCTION

Many lakes and rivers in Northern Norway contain overpopulated and stunted Arctic charr, *Salvelinus alpinus* (L.). The growth season of the fish may be as short as 3 or 4 summer months, with only small amounts of food, or none at all consumed, through the cold months of the year. Ringø and Burkow (1990) detected a high proportion of free fatty acids (FFA), 15% in muscle

neutral lipids of fish caught in October, and suggested that the high FFA content might be due to the fish having starved for some time. However, no information is available about the effect of starvation on lipid utilization in the charr.

The effect of starvation and energy utilization seem to be dependent on the fish species, gonads maturation, water temperature and length of the starvation period (Cowey and Walton, 1989; Henderson and Tocher, 1987; Sargent er al., 1989). Lipid utilization and preferential mobilization of fatty acids during starvation have been reported in several fish species (Jezierska et al., 1982; Murata and Higashi, 1980; Satoh et al., 1984; Takeuchi and Watanabe, 1982). However, in most of these studies no statistical significance were given.

Arctic charr is well adapted to the rigorous fresh water environment, and the fish may therefore have different mechanisms for utilization of lipid and fatty acids during starvation compared to other fresh water species.

The present paper presents analytical data on fatty acid composition of the lipids in Arctic charr (size c. 40 g) starved for 25 and 50 days, and the selective utilization of fatty acids.

MATERIALS AND METHODS

Fish and experimental conditions

Hatchery-reared Arctic charr, *Salvelinus alpinus* (L.), derived from the Takvatn stock were fed a commercial feed (Skretting LTD) from the initial feeding stage until an average body weight of about 40 g was achieved as described elsewhere (Ringø and Nilsen, 1987). Feed was supplied in excess using automatic disc feeders until starving began. During 50 days of starvation the fish were held in a PVC tank (diameter = 74 cm, h = 54 cm) with a continuous supply of aerated fresh water (2.5 l/min) at 8°C.

Determination of whole body weight

Fish were anaesthetized in 0.3% benzocaine and weighed to the nearest 0.1 g.

Chemical analyses

Muscle tissue and liver were sampled from 4 randomly choosen charr at start of the experiment, and after 25 and 50 days of starvation. Fish were anaesthe-

tized in 0.3% benzocaine, and thereafter killed by a blow to the head. The muscle samples were taken immediately caudal of the dorsal fin. Fat deposits surrounding the gut were not observed.

Muscle and liver tissue were dried for 48 hours at 105°C for the determination of dry matter.

Total lipid was determined by the method of Folch et al. (1957) and samples were stored at -80°C in hexane. Lipid class distribution in the neutral lipid was determined in pooled samples from 4 fish by flame ionization detection (TLC-FID) on a Iactroscan TH-10 mark IV analyzer equipped with Chromarod Slll as described Ringø and Burkow (1990) and by thin layer chromatography (TLC) on Silica Gel 60 plates (Tocher and Sargent, 1984) and thereafter determined gravimetically.

Triacylglycerols (TAG) and polar lipid were separated by TLC. The lipids were saponified and fatty acids esterified in 12% BCl₃ in methanol. The methyl esters of TAG and total polar lipid were determined by gas chromatography (Haug et al., 1988).

Statistical significance

To test possible differences in selective utilization of fatty acids as a results of starvation, a one way variance analysis (Anova) was used. The significance level was accepted at $P \le 0.05$.

RESULTS AND DISCUSSION

Cessation of feeding to fish led to an extensive depletion of lipid from the visceral deposits with only loss of small amounts of lipid from muscle tissue and liver (Henderson and Tocher, 1987). In the present study visceral lipid was not observed.

Whole body weight, dry weight and total lipid content of muscle and liver in fed charr and in fish 25 and 50 days after cessation of feeding are given in Table 1.

Figure 1 shows that TAG, the predominant form of lipid reserves were utilized during starvation in the charr. The amount of polar lipid was almost constant. Our results are in accordance with those cited by Henderson and Tocher (1987).

The increase of free fatty acids (FFA) in muscle and liver (Figure 1) may be due to; a) enhanced lipase activity, b) low utilization of FFA as energy source or c) to low oxidation capacities for FFA in the liver and muscle. Bilinski and Jonas (1970) demonstrated that the liver, kidney and particulary the white

				Starved for					
	Fed fish			25 days			50 days		
	N	Х	Ν	Х	S.t	Ν	Х	S.t	
Live wt (g)	35	38.5	31	36.6	N.S	27	33.5	N.S	
Muscle									
Dry weight	4	21.6	4	21.0	*	4	20.5	*	
Total lipids	4	4.2	4	3.6	*	4	3.1	*	
Liver									
Wet weight (% of									
	4	1.20	4	1.05	*	4	1.00	*	
Dry weight	4	75.8	4	77.3	*	4	78.9	*	
Total lipid	4	8.1	4	6.4	*	4	4.4	*	

Table 1. Whole body weight (g), mean water and total lipid (% of wet weight) content in muscle tissue and liver of fed Arctic charr, *Salvelinus alpinus* (L.), and of fish starved for 25 and 50 days. N = number of fish, x = mean. S.t; statistical test between fed fish and fish starved for 25 days, and between fed fish and fish starved for 50 days.

N.S; not significant differences (P>0.05)

*; significant differences (P<0.05)

muscle of rainbow trout (Salmo gairdneri R.), have only a limited capacity to oxidize fatty acids.

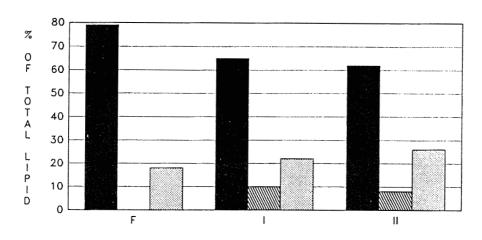
In response to starvation in the charr, no selective utilization (P>0.05) of the fatty acids in muscle TAG occurred (Table 2). However, in the liver TAG a selective utilization (P<0.05) of 16:1 (n-9) 18:1 (n-9) and 22:1 (n-11) was noted (Table 2).

This preferential utilization of fatty acxids in the charr is not consistent with the results from other studies dealing with starvation of freshwater species (Murata and Higasi, 1980; Jezierska et al., 1982; Satoh et al., 1984; Takeuchi and Watanabe, 1982).

Starvation of rainbow trout resulted in significant preferential utilization of both the (n-9) monoenes and (n-6) unsaturates from muscle, but only of the (n-9) monoenes from the liver (Jezierska et al., 1982). However, Takeuchi and Watanabe (1982) demonstrated a utilization of only 18:1 (n-9) from the muscle- and liver TAG of rainbow trout. Satoh et al. (1984) found that no specific changes related to starvation occurred in TAG from whole bodies of *Tilapia nilotica* at 25°C, but that a slight preference for the utilization of 18:2 (n-6) was apparently noted in fish starved at 15°C.

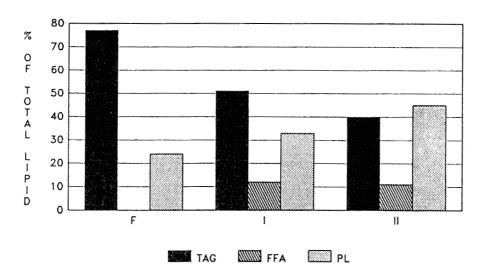
Starvation of carp, *Cyprinus carpio*, has been shown to result in a specific utilization in muscle TAG of 18:1 (n-9) and 18:2 (n-6) (Murata and Higashi, 1980), and 16:0 (Takeuchi and Watanabe, 1982). However, neither in the

Figure 1. Distribution of triacylglycerols (TAG), free fatty acids (FFA) and polar lipid (PL) in muscle tissue and liver of (F) fed Arctic charr, and of fish starved for (I) 25 and (II) 50 days.



MUSCLE





	Muscle TAG			Liver TAG			
	F	25	50	F	25	50	
saturates	21.0	20.2	21.1	17.4	21.1	21.8	
14:0	6.2	5.8	5.8	4.5	5.5	5.3	
16:0	13.4	12.5	13.6	10.7	12.5	13.8	
18:0	1.4	1.9	1.7	2.2	3.1	2.7	
nonoenes	58.7	59.2	57.8	67.8	59.4	53.8	
16:1 (n-9)	11.2	10.3	9.6	12.8	6.7	7.4	
18:1 (n-9)	26.5	27.2	25.8	29.1	28.2	23.6	
20:1 (n-9)	12.0	12.3	13.8	12.3	12.0	13.0	
22:1 (n-11)	9.0	9.4	8.6	13.6	12.5	9.8	
n-6) PUFA	5.1	4.6	5.4	3.9	4.6	5.0	
8:2	4.6	4.3	5.2	3.7	4.6	5.0	
20:4	0.5	0.3	0.2	0.2	0.0	0.0	
n-3) PUFA	11.1	9.6	9.6	7.3	9.0	11.9	
18:3	1.0	0.6	0.4	0.2	0.0	0.0	
20:5	3.5	3.0	3.2	2.9	3.5	5.0	
22:5	1.2	1.0	0.8	1.2	1.5	1.2	
22:6	5.4	5.0	5.2	3.0	4.0	5.7	

Table 2. Fatty acid composition in TAG from muscle and liver of (F) fed Arctic charr and fish starved for 25 and 50 days. Each value represents mean of 4 fish.

studies of Murata and Higashi (1980), and Satoh et al. (1982) nor in those of Takeuchi and Watanabe (1982) statistical tests were used.

Based on the energy content of saturated fatty acids versus unsaturated fatty acids, Jezierska et al. (1982) speculated that factors other than energy content are the main cause of fatty acid utilization in fish. The authors also speculated that the specific retention of polyunsaturated fatty acids (PUFA), may be due to the essentiality of PUFA based upon their unique physical properties in biological membranes. However, in the present study, specific retentions (P<0.05) of 20:5 (n-3) and 22:6 (n-3) were only detected in liver TAG 50 days after cessation of feeding (Table 2). According to Takeuchi and Watanabe (1982) starvation of rainbow trout resulted in a specific retention of 22:6 (n-3) in both body- and liver TAG.

In general, 22:6 (n-3) is perferentially retained by fish subjected to food deprivation, while monounsaturated fatty acids (16:1, 18:1 (n-9) and 20:1 (n-9) are mobilized for use in energy production (Henderson and Tocher, 1987). However, in the present study no utilization of 20:1 (n-9) in TAG occurred (Table 2).

Based on fatty acid composition in polar lipid, our results showed that the monounsaturated fatty acid 18:1 (n-9) was utilized ($P \le 0.05$) in the muscle

Each value represents mean of 4 lish.								
	Muscle			Liver				
	F	25	50	\mathbf{F}	25	50		
saturates	28.0	31.3	31.6	26.6	30.6	31.8		
14:0	2.9	2.9	2.5	1.8	2.8	2.2		
16:0	22.8	25.9	25.6	22.3	25.0	26.6		
18:0	2.3	2.5	3.5	2.5	2.8	3.0		
monoenes	17.7	16.4	12.1	22.4	19.7	17.4		
16:1 (n-9)	2.4	3.0	1.8	3.5	3.5	3.0		
18:1 (n-9)	11.4	10.0	7.8	15.5	13.2	11.6		
20:1 (n-9)	2.7	2.8	2.5	2.7	2.6	2.6		
22:1 (n-11)	1,2	0.6	0.0	0.7	0.4	0,2		
(n-6) PUFA	3.3	2.7	2.5	3.5	2.0	1.9		
18:2	1.8	1.9	2.0	1.3	1.6	1.9		
20:4	1.0	0.6	0.5	1.2	0.2	0.0		
22:5	0.5	0.2	n.d	1.0	0.2	0.0		
(n-3) PUFA	46.1	44.9	45.9	42.1	42.6	44.2		
18:3	0.5	0.2	0.1	0.2	0.2	0.0		
18:4	1.0	0.5	0.4	0.3	0.4	0.0		
20:5	11.4	10.4	10.4	9.0	8.6	8.9		
22:5	1.9	1.8	1.4	1.6	1.8	1.6		
22:6	31.3	32.0	33.6	31.0	31.6	33.7		

and liver after 50 days of starvation (Table 3). The monoene fatty acid 22:1 (n-11) (1.2%) in the muscle, and 20:4 (n-6) (1.2%) and 22:5 (n-6) (1.0%) in the liver of fed fish, was not detected after 50 days of starvation. The proportion of 18:2 (n-6), 20:5 (n-3) and 22:6 (n-3) in the charr polar lipids of both muscle and liver were not affected by starvation.

According to Takeuchi and Watanabe (1982) starvation of rainbow trout led to an increase in the proportion of 22:6 (n-3) in polar lipid of both body and liver. The starvation of carp, *Cyprinus carpio*, also resulted in a specific retention of 22:6 (n-3) in polar lipid but, 18:2 (n-6) was utilized (Murata and Higashi, 1980; Takeuchi and Watanabe, 1982).

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