Effect of different dietary lipid levels on growth performance, slaughter yield, chemical composition, and histology of liver and intestine of pikeperch, *Sander lucioperca*

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Abstract: In this study, 16-month-old pikeperch, Sander lucioperca, (initial body weight 280 g) were fed three diets with different lipid levels with the aim of determining the impact on the growth performance, hepatic and intestinal histological structure, chemical composition, and slaughter yield of this species. The fish were fed isoproteinaceous feeds (450 g protein/kg feed) containing 60 g lipids/kg feed (group F6), 100 g lipids/kg feed (group F10) and 180 g lipids/kg feed (group F18). No significant differences were noted among the treatment groups in body weight gain and in the feeding coefficients of experimental feeds (P > 0.05). In the group of fish administered the diet with the lowest lipid content (group F6), the share of skinned fillet in the whole body weight was the highest (48% vs. 43% in group F18) (P < 0.05). No significant differences among groups were confirmed in the relative values of the viscera weight (4.8-5.8%) (P>0.05). The highest values of the size of hepatocytes and their nuclei, intestinal cells, supranuclear vacuoles of enterocytes, and the degree of vacuolization in hepatocytes were determined in group F18 (P < 0.05), indicating histopathological changes. The highest body and viscera lipid contents were noted in individuals from group F18 (P < 0.05). The high lipid content in the viscera of fish from this group was linked to the significantly lowest content of protein and ash. The levels of lipids, protein, and ash were similar (P > 0.05) in the pikeperch fillets from the three feeding treatments. The levels of n-6 polyunsaturated fatty acids (n-6 PUFA) in the whole fish body, in the viscera and fillets (P < 0.05) were the significantly highest in group F18. Significant differences in n-3 polyunsaturated fatty acids (n-3 PUFA) among the groups were confirmed in the whole fish body and viscera (P < 0.05), while the values in the fillets were similar (P > 0.05). The n-3/n-6 index for the fish fillets ranged from 2.4 (group F18) to 4.7 (group F6) (P < 0.05). The levels of n-3 highly unsaturated fatty acids (n-3 HUFA), arachidonic acid (ARA) and docosahexaenoic acid (DHA) in the fillets of fish from the three dietary treatments were similar (P > 0.05). The fillets of fish from group F6, however, had the lowest levels of linolenic and linoleic acid (ALA and LA) and the highest levels of eicosapentaenoic acid (EPA) (P < 0.05).

Keywords: percid fish; lipid; slaughter yield; histology; body composition

The pikeperch, *Sander lucioperca* L., is a popular freshwater fish species among consumers, especially in communities that promote healthy foods (Dil,

2008). The fillets of this species are rich in polyunsaturated fatty acids (PUFA), including those that are valuable to humans: eicosapentaenoic (C20:5n-3,

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EPA) and docosahexaenoic (C22:6n-3, DHA) (Jankowska et al., 2003). Catches of wild pikeperch have never been significant, and even these have decreased several-fold over the last few decades (FAO, 2007; Dil, 2008). To meet the increased demand for pikeperch production in aquaculture farming, aquaculture systems are needed (Molnár et al., 2006; Philipsen, 2008). The knowledge of dietary requirements of a given species is a key factor in determining the effectiveness of its production in aquaculture. Salmonid artificial feed, often with a high lipid content, has generally been used to date to rear pikeperch (Zakęś et al., 2001; Schulz et al., 2005). Economic factors usually motivate the application of this type of feed since it permits a more rational use of the more expensive protein for somatic growth and lipids as the less expensive energy source. For intensive culture (fattening) of salmonids, feeds with lipid contents as high as 20-30% produce satisfactory results (Einen and Roem, 1997).

It is understood that the optimal shares of protein, lipids, and carbohydrates in diets guarantee appropriate growth rates, condition and health of the fish, and that these parameters are specific to each species (Jobling, 1994; Halver and Hardy, 2002). Fish fed diets that are not tailored to their nutritional requirements exhibit, among other symptoms, halted growth rates and/or anatomical pathology and histopathological changes in the alimentary tract (Parpoura and Alexis, 2001; Ostaszewska et al., 2006). Percid fish of the genus Perca (yellow perch, P. flavescens Mitchill, and European perch, P. fluviatilis L.) fed diets with high lipid contents exhibit unsatisfactory rearing results, and these fish can exhibit visceral fat deposits and internal organ dysfunction (especially in the liver) (Brown et al., 1996; Kestemont et al., 2003). Additionally, diets high in lipid content lead to the deposition of large amounts of fat in the viscera effectively lowering slaughter yield. Therefore, the chemical composition of feed is a determining factor in the market value of reared fish and can impact the yield of the edible parts and the chemical composition of the body, including the fillets (Jobling, 2001).

Nutritional experiments conducted on juvenile pikeperch to date have focused on feeding regime; levels of dietary protein, lipids and carbohydrates; dietary lipid content and type on growth; body composition; and the histological structure of some organs (Zakęś et al., 2001, 2003; Nyna-Wamwiza et al., 2005; Ostaszewska et al., 2005; Molnár et al., 2006; Kowalska et al., 2010). The results of these

studies have not always been unequivocal, and the knowledge of nutritional requirements of this species remains unsatisfactory and far from complete. Some of the aspects of nutrition that have yet to be investigated, included the impact the chemical composition of feed on the values of such significant indexes of pikeperch rearing as slaughter yield and dietary quality of fillets or the histological structure of the liver and intestines. The lipid quantity and quality in feeds can be linked to growth, lipid metabolism, fatty acid content in the tissues, histological changes in the liver, intestine, and resistance of fish (Kalogeropoulos et al., 1992; Rennert et al., 2005; Piedecausa et al., 2007). The lipid level and deficiency of some fatty acids in fish feed have an effect on the degree of hepatocyte vacuolization, supranuclear zone of enterocytes, accumulation of lipid droplets in enterocytes and consequently they impact the size of these cells (Olsen et al., 2000; Caballero et al., 2002; Ostaszewska et al., 2005, Kowalska et al., 2010). Anomalies in hepatocyte and intestine structure can disturb dietary fat metabolism which may affect the immune system and lead to poor fish health (Watanabe et al., 1989; Tacon, 1992; Parpoura and Alexis, 2001; Caballero et al., 2002; Halver and Hardy, 2002).

The aim of the present study was to investigate the impact the administration of feeds with different lipid contents to pikeperch [reared in a recirculating aquaculture system (RAS)] on growth performance, slaughter yield, pathological changes in liver and intestine and in the chemical composition of the fish.

MATERIAL AND METHODS

Fish and rearing

The fish were obtained from induced spawning at the Inland Fisheries Institute in Olsztyn, Poland (IFI Olsztyn; Zakęś and Szczepkowski, 2004). The larvae were reared in an RAS and received initially mixed feed (*Artemia* sp. + formulated feed) and then exclusively commercial trout feed (Szkudlarek and Zakęś, 2007). Juvenile pikeperch of the mean body weight 55 g were placed in three rearing tanks of 0.8 m³ in volume each and fed experimental diets with different lipid contents (60, 100, 180 g/kg feed; Table 1). After 6 months of initial rearing, the proper experiment began. Fish of the mean initial body weight approximately 280 g and total

Table 1. Chemical (g/kg dry weight) and selected fatty acid (FA) composition (g FA/kg of total FA) of experimental diets

	Diets		
	F6	F10	F18
Crude protein	450	450	450
Crude fat	60	100	180
NFE	390	350	270
Crude ash	80	80	80
Gross energy (MJ/kg feed)	19.8	20.7	22.4
Fatty acid composition			
C14:0	37.7	28.3	21.8
C16:0	188.1	165.5	147.2
C18:1 <i>cis</i> 9	189.9	204.5	217.4
C18:2n-6	139.2	239.8	313.1
C18:3n-3	23.8	29.6	8.9
C20:4n-6	0.6	1.1	0.9
C20:5n-3	60.0	42.2	35.0
C22:6n-3	107.7	68.3	52.5
Total n-3	206.2	151.0	105.1
Total n-6	150.1	247.3	318.2
Total n-9	221.7	237.6	251.7
n-3/n-6	1.37	0.61	0.33

F6 - 60 g lipid/kg feed, F10 - 100 g lipid/kg feed, F18 - 180 g lipid/kg feed

NFE – nitrogen free extract, calculated as 1000 – (protein + lipid + ash + fibre) fat/kg feed

Gross energy - calculated as: 24 MJ/kg protein, 39 MJ/kg fat, 17 MJ/kg NFE (Jobling, 1994)

Total n-3 - C18:3n-3, C20:3n-3, C20:4n-3, C20:5n-3, C22:5n-3, C22:6n-3

Total n-6 - C18:2n-6, C20:3n-6, C20:4n-6, C22:5n-6

Total n-9 - C18:1cis9, C20:1n-9, C22:1n-9

length (TL) 31 cm (aged 16 months) were placed in 12 rearing tanks (of $0.2 \,\mathrm{m}^3$ in volume each) in an RAS. Forty-five individuals were chosen from each group receiving feeds with different lipid levels. These were stocked into 3 rearing tanks (n=3) where they continued to receive the experimental feeds. The stocking density was 15 individuals per tank, and the biomass was approximately $20 \,\mathrm{kg/m^3}$. The water flow rate was maintained at $4 \,\mathrm{l/min}$. Water temperature, oxygen content, total ammonia nitrogen (TAN = NH₃-N + NH₄⁺-N) and water pH at the rearing tank outflow were: $22.2 \pm 0.1 \,^{\circ}\mathrm{C}$; $5.6 \pm 0.3 \,\mathrm{mg}\,\mathrm{O}_2/\mathrm{l}$; $0.08 \pm 0.02 \,\mathrm{mg}\,\mathrm{TAN/l}$; 7.65-7.58, respectively. A constant LD 24:0 photoperiod was used. Light intensity measured at the water surface

of rearing tanks was 40–50 lux. The experiment was three months long, which permitted the pikeperch to grow to the size that produced consumer-sized fillets (Summerfelt, 1996).

Feed and feeding

The base feed administered to the fish was Aller Safir (Aller-Aqua, Golub-Dobrzyń, Poland), which is a commercial feed product without the addition of lipids. The main sources of protein in this feed are fish and soy meals. The lipid quantity in the base feed was 60 g/kg feed, and was derived mainly from fish meal. The fish received the base feed (group

F6) or base feed with the addition of fish oil (Peter Möller, Möller's Tran, Oslo, Norway) and soy oil (ZPT Olvit, Gdańsk, Poland) in quantities of 12 and 28 g/kg feed (group F10) or 36 and 84 g/kg feed (group F18). The oils were added to the base feed using a vacuum pump (AGA Labor, Lublin, Poland). Feed was prepared every seven days and stored in a refrigerated unit (+4°C). The chemical composition of the feed is presented in Table 1.

The fish were fed continuously (19 h/day) with a band feeder (4305 FIAP, Fishtechnic GmbH, Germany). For the first three weeks of rearing the daily feed ration was 0.8% of the stock biomass, and in the last nine weeks of rearing it was 0.5% of the stock biomass. The experiment was carried out during a three-month period (from 280 g to the final sampling). No mortality was observed during the experiment.

Experimental procedure and calculation

On the first and on the last day of the experiment, all fish were weighed (W \pm 0.01 g), measured (TL \pm 0.1 cm), and samples were taken for the basic analyses of the chemical composition of the bodies. The fish were anesthetised with a dose of 1.0 ml/l of etomidate (Propiscin, IFI Olsztyn, Kazuń and Siwicki, 2001) before weighing and measuring. The individuals used to determine the body chemical composition and for histological analyses were anesthetized with a dose of 4 ml/l and then decapitated.

The following formulas were used to determine the values of the chosen rearing indexes:

Specific growth rate, SGR (%/day) =
$$100 \times [(\ln W_f - \ln W_i)/T];$$

Daily growth rate, DGR (g/day) = $(W_f - W_i)/T$; Condition factor, CF = $100 \times (W/TL^3)$; Feed conversion ratio, FCR = TFI/(FB – IB); Protein efficiency ratio, PER = (FB – IB)/TFP; Weight of fat in given body parts (whole fish, viscera, fillets), WF_x (g/fish) = $(W_x \times LP_x)/100$; Relative fat content, FC (%) = $(WF_x \times 100)/Wf_t$.

Ten individuals were chosen from each dietary treatment to determine slaughter yield. The fish were anesthetized in a solution of Propiscin (4 ml/l) and then decapitated. They were weighed (\pm 0.01 g), gutted, filleted, and skinned. All body parts were weighed and their shares in the total body weight were determined as follows:

$$WR_x (\%) = (W_x \times 100)/W$$

The values of the hepatosomatic index (HSI, %) were also determined as follows:

 $HSI = 100 \times (LW/W)$

The notation used in the preceding formulas was as follows: W_f and W_i were the final and initial body weight (g); T – time of rearing (days); TL – total length (cm); FB and IB – final and initial fish biomass (g); TFI – total feed intake (g); FBP – final body protein content (%); W_x – weight of a given fish body part (g); WF_x – weight of fat in a given body part (g); WF_t – weight of fat in a given fish body part (%); WF_t – weight of fat in whole fish (g); WF_t – liver weight (g).

Histological analysis of liver and intestines

On the last day of the experiment, the liver and anterior sections of the intestines were excised from 7 individuals from each group (2–3 individuals from each tank). The tissues were fixed in Bouin's fluid, dehydrated in ethanol, cleared in xylene, embedded in paraffin blocks, and then sliced with a rotating microtome (Leica, Bensheim, Germany) into 5 μm sections and stained with H&E. Analyses and histological measurements were performed with a light microscope (Nikon E600, Japan), the MultiScanBase v. 8.08 computer programme (Computer Scanning System Ltd., Warsaw, Poland), and the NIS-Elements F2.30 v. 2.21 programme (Nikon, Japan).

The following analyses were performed: 50 hepatocytes and their nuclei; 50 enterocytes and their nuclei; supranuclear vacuole ($\pm 0.01~\mu m$) (Ostaszewska et al., 2005). Hepatocyte vacuolization was rated on a scale of 0 to 4, which increased as the cell cytoplasm becamed increasingly filled with the lipid vacuole (no lipid vacuolization – 0, low vacuolization – 1, moderate – 2, high – 3, maximum vacuolization – 4; Figueiredo-Silva et al., 2005). The degree of vacuolization in the cells was determined in a field with a surface area of 2500 μm^2 (50 \times 50 μm). Twenty such fields were measured for each individual, and vacuolization on the five-degree scale was determined for each hepatocyte.

Feed and fish chemical analysis

Water content, total protein, crude fat and crude ash (wet weight) and individual fatty acids [g/kg of

total fatty acid (tFA)] were determined in 5 whole fishes, in skinned fillets from 6 fishes, and in the viscera from 10 fishes that were chosen from 2 tanks of each dietary treatment. The fish and body parts of the individuals from a given tank were combined and analysed together (n = 2, for each feeding treatment). Protein content was determined by the Kjeldahl method. Fat content was determined by the Soxhlet method (AOAC, 1975), and the fatty acids were analyzed according to the method of Folch et al. (1957). The fatty acids were methylated by adding a mixture of chloroform, non-aqueous methanol and sulphuric acid (100:100:1) (Peisker, 1964). Chromatographic separation was performed on an Agilent Technologies 6890 N gas chromatograph with a flame ionisation detector (FID). The detector signal was registered by a Philips recorder with 1 mV full-scale sensitivity at a tape speed of 10 mm/min. The fatty acids were identified using standards by Supelco (Bellefonte, PA, USA).

Statistical analysis

All data were subjected to one-way ANOVA and Tukey's test (for the analysis of rearing indexes, cell size structure, fish chemical composition and slaughter yield) or Kruskal-Wallis test (to analyse the degree of vacuolization) using the GraphPad Prism programme (Soft. Inc., USA). Differences were considered significant at $P \le 0.05$. All values expressed as percentages were transformed with *arcsin* before statistical processing.

RESULTS

Growth performance, feed utilization, slaughter yield

The feeds tested did not have a significant impact either on pikeperch growth rate and condition or on the values of the coefficients FCR or PER (P > 0.05, Table 2), but the feeds with different lipid contents influenced the slaughter yields of fish (P < 0.05, Table 3). The highest relative value for skinned fillets, expressed as the per cent of total body weight, was obtained in the group receiving feed with the lowest lipid content (48.1% – group F6 vs. 43.6–42.8% – groups F10 and F18). Among the body parts analyzed, significant differences among groups were noted in the relative values of the weight of the head and fins (P < 0.05), but it did not apply to the weight of the viscera (P > 0.05, Table 3).

Histological observations

Significant differences were determined in the size of hepatocytes and their nuclei, and in the degree of hepatocyte vacuolization in the liver of fish receiving feeds with different lipid contents (Table 4). The largest hepatocytes occurred in individuals administered feed with the highest lipid content (group F18) (P < 0.05, Table 4). The degree of hepatocyte vacuolization increased proportionally to the dietary lipid content (P < 0.05). Small

Table 2. Weight, length and production parameters of pikeperch fed experimental diets (mean \pm SD; n = 3)

	Dietary treatments		
	F6	F10	F18
Body weight (g)	417.23 ± 31.22	451.15 ± 7.85	443.45 ± 11.10
Total length (cm)	36.70 ± 0.45	35.11 ± 0.98	34.98 ± 0.73
Daily growth rate DGR (g/day)	1.90 ± 0.06	1.70 ± 0.03	1.91 ± 0.01
Specific growth rate SGR (%/day)	0.57 ± 0.06	0.45 ± 0.01	0.53 ± 0.01
Condition factor CF	0.96 ± 0.02	0.90 ± 0.01	1.00 ± 0.06
Feed conversion ratio FCR	1.28 ± 0.05	1.26 ± 0.17	1.30 ± 0.02
Protein efficiency ratio PER	1.74 ± 0.08	1.77 ± 0.24	1.70 ± 0.04

Absence of superscripts letters indicates no significant differences between treatments (P > 0.05)

F6 – group fed feed with 60 g lipid/kg feed, F10 – group fed feed with 100 g lipid/kg feed, F18 – group fed feed with 180 g lipid/kg feed

Table 3. Fish body weight (g) and slaughter yield (% based on the whole fish body weight) of pikeperch from three dietary treatments (mean \pm SD; n = 10)

Parameter	Dietary treatments		
	F6	F10	F18
Total fish body weight (g)	483.5 ± 72.1	584.5 ± 83.7	541.5 ± 113.3
Viscera	4.8 ± 0.7	5.8 ± 0.8	5.4 ± 1.1
Gutted fish	94.9 ± 1.6	94.1 ± 0.8	93.9 ± 7.5
Carcass (without head and viscera)	$71.3^{\circ} \pm 1.6$	$68.9^{b} \pm 1.5$	$66.1^{a} \pm 1.4$
Fillet with skin	58.1 ± 2.1	54.3 ± 2.7	53.2 ± 1.0
Skinned fillet	$48.1^{\rm b} \pm 2.7$	$43.6^{a} \pm 3.2$	$42.8^{a} \pm 2.1$
Skin	9.8 ± 1.1	10.2 ± 1.7	10.2 ± 1.6
Fins	$2.6^{a} \pm 0.2$	$2.8^{b} \pm 0.2$	$2.7^{ab} \pm 0.3$
Head	$23.4^{a} \pm 1.2$	$24.3^{ab} \pm 1.3$	$24.9^{b} \pm 1.3$
Spine	9.7 ± 1.8	11.3 ± 2.1	9.9 ± 0.8

Data on the same line with different superscript letters are significantly different (P > 0.05)

F6 – group fed feed with 60 g lipid/kg feed, F10 – group fed feed with 100 g lipid/kg feed, F18 – group fed feed with 180 g lipid/kg feed

lipid vacuoles were observed in the fish from group F6, while fish from groups F10 and F18 had lipid droplets of various sizes (Figure 1). In group F18, the lipid droplets tended to drain and to form large fatty cysts.

The smallest epithelial cells and enterocyte supranuclear vacuoles were confirmed in the fish from group F6, while the largest were in group F18 (P < 0.05, Table 4). Small lipid vacuoles were observed in the individuals from group F6 (Figure 2). Larger and more numerous lipid vacuoles and lipid

droplets in the enterocyte supranuclear vacuoles were confirmed in group F18. In this instance, large lipid droplets distorted the enterocyte cell structure on the surface in some places.

Chemical analysis

The pikeperch receiving feed with 180 g lipids per kg feed had the highest content of fat in the whole fish body and viscera and the lowest levels of

Table 4. Comparison of hepatocyte and enterocyte morphometric characteristics (n = 7) and hepatosomatic index (n = 10) of pikeperch fed experimental diets (mean \pm SD)

	Dietary treatments		
	F6	F10	F18
Hepatosomatic index HSI (%)	0.95 ± 0.14	0.84 ± 0.19	1.06 ± 0.24
Size of hepatocyte (µm)	$11.1^{a} \pm 1.9$	$10.3^{a} \pm 1.8$	$14.3^{b} \pm 1.3$
Size of nuclei (µm)	$3.6^{a} \pm 0.1$	$3.7^{ab} \pm 0.2$	$4.0^{b} \pm 0.1$
Vacuolization degree	$0.7^{a} \pm 0.4$	$1.8^{b} \pm 0.5$	$2.5^{b} \pm 0.7$
Height of enterocyte (μm)	$22.9^{a} \pm 0.9$	$25.7^{ab} \pm 2.1$	$26.5^{b} \pm 2.2$
Size of nuclei (µm)	5.1 ± 0.3	5.1 ± 0.5	5.6 ± 0.7
Height of supranuclear zone (μm)	$12.5^{a} \pm 0.8$	$14.2^{ab} \pm 1.6$	$15.2^{b} \pm 1.7$

Absence of superscripts letters indicates no significant differences between treatments (P > 0.05)

F6 – group fed feed with 60 g lipid/kg feed, F10 – group fed feed with 100 g lipid/kg feed, F18 – group fed feed with 180 g lipid/kg feed

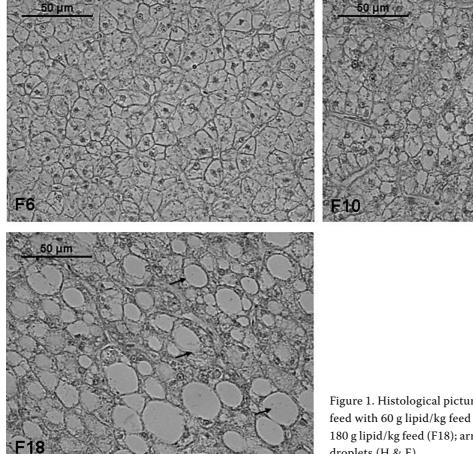


Figure 1. Histological picture of the livers of pikeperch fed feed with 60 g lipid/kg feed (F6), 100 g lipid/kg feed (F10), 180 g lipid/kg feed (F18); arrows indicates hepatocyte lipid droplets (H & E)

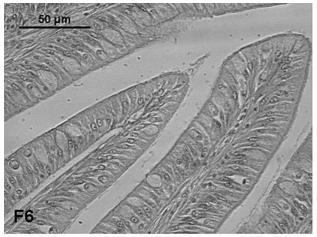
protein and ash in the viscera in comparison with fish administered feed with lower lipid contents (P < 0.05, Tables 5 and 7). The contents of protein, fat and ash in the fillets of fish from the 3 dietary treatment groups were similar (P > 0.05, Table 6). The higher lipid content in the pikeperch feed was also linked to a higher content of fat in the whole fish bodies and viscera (Figure 3). Although it did not influence the amount of fat in the fillets, the relative fat content (FC) in the fillets of fish from group F6 was the highest, while in group F18 it was the lowest. The values of the FC index for the viscera were significantly the highest in the group of fish receiving feed with the highest fat (P < 0.05, Figure 3).

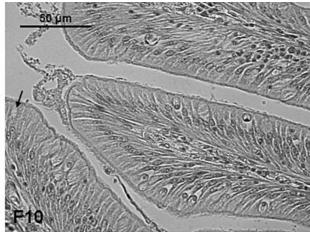
The content of polyenic acids (PUFA) from the n-3 series (total n-3) in the bodies of fish from group F18 was lower than in group F6 (P < 0.05, Table 5), while the fish from group F6 were characterized by the significantly lowest quantity of monoenoic fatty acids (MUFA) of the n-9 series (total n-9) and PUFA from the n-6 series (total n-6) in the whole body as compared to group F18. Significant differences

were noted in the quantities of myristic (C14:0), linoleic (C18:2n-6, LA), linolenic (C18:3n-3, ALA), arachidonic (C20:4n-6, ARA), EPA, and DHA acids in the whole bodies of fish from the different dietary treatments (P < 0.05, Table 5).

The content of n-6 PUFA was significantly the lowest in the fillets of fish receiving feed F6 in comparison with those of fish from the other dietary treatments (P < 0.05, Table 6). The meat of fish from group F6 was characterized by the highest contents of EPA (P < 0.05), but the contents of DHA in the fillets of fish from groups F6, F10, and F18 were similar (P > 0.05). The values of the n3/n6 index were the most advantageous in the fillets of fish from group F6 (4.72 vs. 2.86–2.36 – groups F10 and F18) (P < 0.05, Table 6).

The quantities of n-3 PUFA in the viscera of fish from group F18 were significantly lower than those in groups F6 and F10 (P < 0.05, Table 7). In turn, the content of n-6 PUFA in group F18 was significantly higher (P < 0.05). The administration of feeds with the highest lipid content (group F18) also resulted in the significantly lowest quantities of palmitic





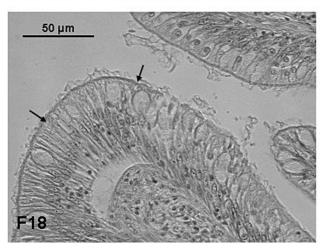


Figure 2. Interior intestine epithelium of fish fed feed supplemented with 60 g lipid/kg feed (F6), 100 g lipid/kg feed (F10), 180 g lipid/kg feed (F18); arrows indicate enterocyte lipid vacuoles (H & E)

acid (C16:0). The viscera of fish from groups F10 and F18 contained more oleic acid (C18:1cis9) and LA, and less ALA, ARA, EPA, and DHA in comparison with the values recorded in group F6 (P < 0.05, Table 7).

DISCUSSION

Growth performance and slaughter yield

The isoproteinaceous diets with different lipid contents did not influence the mean final body weight of t pikeperch. Nyina-Wamwiza et al. (2005) confirmed that when protein supply is appropriate (400–500 g protein/kg feed for percid fish), different lipid contents in feeds do not have an impact on the rearing results of pikeperch. As reported by Zakęś et al. (2004), Nyina-Wamwiza et al. (2005), Molnár et al. (2006) and Schulz et al. (2007), no protein sparing effect was demonstrated. However, Schulz et al. (2007) confirmed significantly fast-

er growth rates in pikeperch administered feeds with higher lipid contents (170 in comparison with 90 and 130 g lipids/kg feed; protein contents 470 to 530 g/kg feed). The value of the FCR coefficient was advantageous (approx. 1.3) in all feeds tested in the present study, and was similar to those reported for the impact of the diets on slaughter yield becamed especially evident when wild and reared individuals of the same species were compared (Jobling, 2001). Lowered slaughter yield values in fish receiving formulated feeds, which resulted mainly from an increase in the weight of the viscera, were confirmed for example in rainbow trout, Oncorhynchus mykiss (Walb.) (Jobling et al., 1998), European sea bass, Dicentrarchus labrax (L.) (Boujard et al., 2004) and European perch (Mathis et al., 2003). However, it was not observed in pikeperch (Jankowska et al., 2003). In the present study, feeds with different lipid contents, and thus energy concentrations, had a significant impact on the slaughter yield of pikeperch. The fillet yield, expressed as the percentage of the total body weight, of individuals fed more

Table 5. Chemical (g/kg of wet weight) and main fatty acid (FA) composition (g FA/kg total FA) of whole bodies of pikeperch fed diets with different lipid contents (mean \pm SD; n = 2)

	Dietary treatments		
	F6	F10	F18
Components			
Crude protein	180.8 ± 0.1	183.0 ± 1.9	180.6 ± 3.6
Crude fat	$40.9^{a} \pm 2.9$	$46.5^{ab} \pm 9.4$	$68.8^{b} \pm 0.4$
Crude ash	40.2 ± 0.6	36.8 ± 0.5	36.5 ± 2.4
Gross energy (MJ/kg)	6.6 ± 0.2	6.8 ± 0.4	7.6 ± 0.1
Fatty acid composition			
C14:0	$48.3^{b} \pm 4.0$	$41.3^{ab} \pm 0.4$	$34.2^{a} \pm 2.6$
C16:0	165.5 ± 11.9	154.9 ± 2.1	140.4 ± 5.6
C18:1cis 9	$193.7^{a} \pm 2.1$	$218.9^{b} \pm 9.2$	$223.9^{b} \pm 1.8$
C18:2n-6	$83.9^{a} \pm 4.0$	$158.7^{b} \pm 8.0$	$211.4^{\circ} \pm 1.3$
C18:3n-3	$14.3^{a} \pm 0.1$	$18.5^{b} \pm 0.1$	$21.2^{c} \pm 1.1$
C20:4n-6	$8.4^{b} \pm 0.6$	$6.4^{a} \pm 0.4$	$4.8^{a} \pm 0.4$
C20:5n-3	$68.7^{\rm b} \pm 4.8$	$47.7^{a} \pm 1.0$	$40.2^{a} \pm 4.2$
C22:6n-3	$162.5^{b} \pm 17.5$	$123.3^{ab} \pm 1.6$	$117.2^{a} \pm 3.2$
Total n-3	$267.1^{\text{b}} \pm 24.9$	$207.5^{ab} \pm 1.5$	$194.2^{a} \pm 3.5$
Total n-6	$98.5^{a} \pm 3.0$	$169.9^{b} \pm 8.3$	$220.4^{c} \pm 2.0$
Total n-9	$446.2^{a} \pm 1.4$	$496.5^{b} \pm 9.0$	$507.4^{b} \pm 3.5$
n-3/n-6	$2.71^{c} \pm 0.34$	$1.22^{b} \pm 0.05$	$0.88^{a} \pm 0.01$

Different letter within a line denotes significant differences (P < 0.05)

F6 – group fed feed with 60 g lipid/kg feed, F10 – group fed feed with 100 g lipid/kg feed, F18 – group fed feed with 180 g lipid/kg feed; for explanation of gross energy calculation and fatty acids groups see Table 1

energetic feeds was significantly lower. It is symptomatic that the relative viscera weight in the experimental groups of fish was comparable and that the relative head and fin weights differed. By com-

parison, in perch it was confirmed that the lowered slaughter yield of individuals receiving formulated feed with higher energy and lipid contents resulted from the higher viscera weight (VSI) (Mathis et al.,

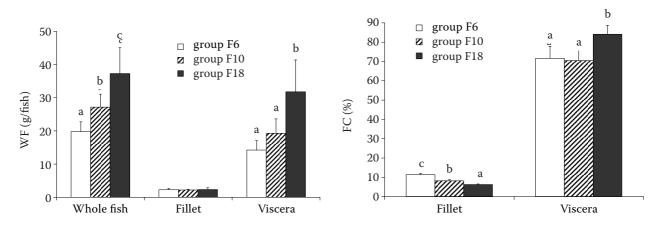


Figure 3. Weight of lipid (WF; g/fish) and relative lipid content (FC; %) in the bodies of pikeperch fed feed with 60 g lipid/kg feed (group F6), 100 g lipid/kg feed (group F10), 180 g lipid/kg feed (group F18)

Table 6. Chemical (g/kg of wet weight) and main fatty acid (FA) composition (g FA/kg total FA) of fillets from pikeperch fed diets with different lipid contents (mean \pm SD; n = 2)

	Dietary treatments		
	F6	F10	F18
Components			
Crude protein	210.5 ± 2.3	202.8 ± 3.6	204.7 ± 3.4
Crude fat	9.7 ± 2.1	8.7 ± 0.1	11.1 ± 3.5
Crude ash	11.9 ± 0.1	11.7 ± 0.1	11.9 ± 0.3
Gross energy (MJ/kg)	5.6 ± 0.1	5.4 ± 0.1	5.5 ± 0.1
Fatty acid composition			
C14:0	$25.4^{b} \pm 1.7$	$17.3^{a} \pm 0.6$	$14.6^{a} \pm 0.1$
C16:0	204.9 ± 9.4	195.9 ± 1.5	196.0 ± 9.0
C18:1 <i>cis</i> 9	131.0 ± 1.46	129.4 ± 8.8	221.8 ± 4.4
C18:2n-6	$58.8^{a} \pm 3.3$	$107.5^{\rm b} \pm 8.5$	$132.8^{b} \pm 7.1$
C18:3n-3	$8.5^{a} \pm 0.2$	$9.9^{ab} \pm 0.7$	$10.6^{b} \pm 0.3$
C20:4n-6	14.6 ± 0.4	13.7 ± 0.4	13.0 ± 0.4
C20:5n-3	$71.5^{\text{b}} \pm 4.7$	$54.9^{a} \pm 3.1$	$50.9^{a} \pm 0.3$
C22:6n-3	282.3 ± 16.3	286.3 ± 2.9	283.4 ± 2.0
Total n-3	380.1 ± 22.1	366.6 ± 24.3	359.1 ± 0.2
Total n-6	$80.5^{a} \pm 3.0$	$128.4^{\rm b} \pm 8.5$	$151.9^{b} \pm 6.9$
Total n-9	299.5 ± 14.6	294.9 ± 9.6	277.3 ± 4.0
n-3/n-6	$4.72^{\rm b} \pm 0.45$	$2.86^{a} \pm 0.38$	$2.36^{a} \pm 0.11$

Different letter within a line denotes significant differences (P < 0.05)

F6 – group fed feed with 60 g lipid/kg feed, F10 – group fed feed with 100 g lipid/kg feed, F18 – group fed feed with 180 g lipid/kg feed; for explanation of gross energy calculation and fatty acids groups see Table 1

2003). It was also confirmed that differences in the filleting yield and VSI were typical not only of wild and reared perch but also of fish reared in different rearing aquaculture systems (extensive, semi-intensive, and intensive rearing). More intensive rearing conditions resulted in lowered slaughter yield and increases in the values of the VSI, with the highest viscera weight recorded in perch reared in recirculating systems (Mairesse et al., 2005).

Histological examination

Differences in the histological structures of liver and intestines were noted among groups of pikeperch reared on feeds with different lipid contents. Similarly like in European perch administered feed containing 60, 120, or 180 g lipids/kg feed (Kestemont et al., 2001), the largest lipid vacuoles and the most pronounced histopathology were

observed in the hepatocytes of fish receiving feed with the highest lipid content. The occurrence of large fatty cysts in the pikeperch from group F18 indicated the beginning of organ degeneration that leads to necrosis and structural distortions in enterocyte membranes which resulted from the high lipid content, including soy oil in quantities of 120 g/kg feed. Bac et al. (1983) and Parpoura and Alexis (2001) also reported necrosis and the degeneration of hepatocyte cell membranes in European seabass and gilthead seabream, Sparus aurata L., which received feeds supplemented with soy oil. The lowest quantities of myristic and palmitic acids (169.0 g/kg tFA) and the highest quantity of LA (313.1 g/kg tFA) in the feed given to group F18 could have led to the cessation of phospholipid synthesis and the accumulation of lipids in the liver of these fish, which concurs with results reported by Kalogeropoulos et al. (1992), Rennert et al. (2005), and Piedecausa et al. (2007). Both Watanabe et al.

Table 7. Chemical (g/kg of wet weight) and main fatty acid (FA) composition (g FA/kg total FA) of viscera from pikeperch fed diets with different lipid contents (mean \pm SD; n = 2)

	Dietary treatments		
	F6	F10	F18
Components			
Crude protein	$67.2^{ab} \pm 3.2$	$81.1^{b} \pm 11.7$	$38.8^{a} \pm 6.6$
Crude fat	$578.8^{a} \pm 31.8$	$623.8^{a} \pm 10.0$	$756.1^{\text{b}} \pm 35.4$
Crude ash	$5.8^{b} \pm 0.1$	$4.1^{a} \pm 0.1$	$3.5^{a} \pm 0.6$
Gross energy (MJ/kg)	$24.3^{a} \pm 1.2$	$26.3^{a} \pm 0.1$	$30.5^{b} \pm 1.2$
Fatty acid composition			
C14:0	$63.9^{b} \pm 3.0$	$45.4^{a} \pm 1.3$	$43.6^{a} \pm 2.8$
C16:0	$161.6^{b} \pm 5.2$	$151.7^{b} \pm 1.1$	$132.0^{a} \pm 2.1$
C18:1 <i>cis</i> 9	$205.7^{a} \pm 2.4$	$241.8^{b} \pm 0.5$	$230.5^{b} \pm 1.6$
C18:2n-6	$90.1^{a} \pm 3.7$	$180.7^{b} \pm 3.9$	$221.8^{b} \pm 19.5$
C18:3n-3	$16.1^{a} \pm 0.1$	$21.3^{b} \pm 0.1$	$23.4^{b} \pm 1.8$
C20:4n-6	$6.0^{a} \pm 0.2$	$4.3^{b} \pm 0.1$	$4.3^{\rm b} \pm 0.4$
C20:5n-3	$69.4^{\rm b} \pm 4.1$	$42.8^{a} \pm 0.4$	$43.6^{a} \pm 3.8$
C22:6n-3	$110.1^{b} \pm 5.3$	$80.8^{a} \pm 1.6$	$81.7^{a} \pm 5.7$
Total n-3	$216.2^{b} \pm 5.7$	$161.1^{a} \pm 6.2$	$165.5^{a} \pm 3.2$
Total n-6	$101.5^{a} \pm 3.9$	$189.4^{b} \pm 4.0$	$230.2^{b} \pm 18.9$
Total n-9	$477.1^{a} \pm 4.4$	$544.5^{b} \pm 0.6$	$527.4^{\rm b} \pm 0.4$
n-3/n-6	$2.13^{b} \pm 0.18$	$0.85^{a} \pm 0.03$	$0.72^{a} \pm 0.10$

Different letter within a line denotes significant differences (P < 0.05)

F6 – group fed feed with 60 g lipid/kg feed, F10 – group fed feed with 100 g lipid/kg feed, F18 – group fed feed with 180 g lipid/kg feed; for explanation of gross energy calculation and fatty acids groups see Table 1

(1989) and Tacon (1992) confirmed the lipid degeneration of hepatic tissues in marine fish species administered diets with low levels of n-3 PUFA. Although pikeperch is capable of bioconverting n-3 HUFA precursors, the level of 8.9 g ALA/kg tFA in the fish diets might be insufficient, especially in the individuals in which pathological changes in the liver are evident. Additionally, the combined content of EPA and DHA in the diet fed to group F18 was just 87.5 g/kg tFA. Liver pathology was noted in European seabass when their diet contained similarly low contents of these acids (69.0 g/kg tFA) (Parpoura and Alexis, 2001). The results above indicate that SFA and PUFA influence significantly hepatic and intestinal pathological changes in percid fish, including the pikeperch. In the present study, no pathological changes were observed in the organs examined at minimal combined quantities of C14:0 and C16:0, EPA and DHA, and ALA of 193.8, 110.5 and 23.8 g/kg tFA, respectively.

Body chemical composition

The share of body fat in pikeperch increased along with increases in the lipid content of feeds, which was also shown in other studies (Zakęś et al., 2004; Nyina-Wamwiza et al., 2005; Schulz et al., 2007). Percid fish store energy reserves mainly in the viscera (intestinal fat), and the chemical composition of the fillets is quite stable (Xu et al., 2001; Zakęś et al., 2004). In the present study, it was estimated that as much as 84% of the fat in the whole bodies of the individuals from group F18 was stored in the viscera. The fat content in the pikeperch fillets in the present study was 8.7 to 11.1 g/kg, in comparison with previously presen-ted < 20 g/kg (Jankowska et al., 2003). The chemical composition of the pikeperch viscera was the most labile (Zakęś et al., 2004; Kowalska et al., 2010; this study). In the present study, significant differences were noted not only in the quantity of fat but also in that of protein. In effect, the energy value of the viscera of fish from group F18 was significantly higher than that in the individuals from groups F6 and F10 while this was not as distinctly apparent with regard to VSI. The reasons for the lack of significant differences among the groups should include, among others, the high individual variation of this index (individual "susceptibility" to fat deposition). The high individual variation in the VSI values was also reported in perch (Mathis et al., 2003).

The lower levels of C14:0 and C16:0 in the fish bodies most probably resulted from supplementing soy oil to the experimental feeds. It was demonstrated that supplementing lipids of vegetable origin to diets leads, among other effects, to lower levels of SFA in pikeperch and in the tissues of other species (Jobling et al., 2001; Caballero et al., 2002; Schulz et al., 2005).

The F6 and F10 diets contained lower quantities of monoenoic acids than did the F18 diet. Nonetheless, the content of MUFA (and oleic acid) in the body of pikeperch might be linked to the tendency of fish to store these acids as an energy source and/or to the synthesis of monoenoic acids in the fish tissues (Karapanagiotidis et al., 2007). However, the apparent selection against metabolism of some fatty acids might also be due to MUFA content in fish tissue (Bell et al., 2003).

The meat of percid fish is usually rich in HUFA (Jankowska et al., 2003; Kowalska et al., 2010). The lipids in pikeperch fillets are mainly phospholipids that are rich in EPA and DHA (Jankowska et al., 2003). Although the quantities of EPA were significantly the highest in group F6, the combined quantity of n-3 HUFA in pikeperch fillets was similar in all dietary treatments. This resulted from the high content of docosapentaenoic acid (C22:5n-3, DPA) and DHA in the fish meat. No differences in the content of DHA in the fish fillets indicate that the amount of DHA and EPA is satisfactory to fulfil the needs for maintaining the phospholipid profile.

To summarize, the results of the present study indicated that the application of low-fat feed is beneficial in the rearing of pikeperch. Although no significant differences were noted in the growth rates of fish or in the feed conversion ratio of diets containing 60, 100, and 180 g lipids/kg feed, the slaughter yield (skinned filet) and the fatty acid profile (n-3/n-6 ratio) were the most advantageous in the pikeperch reared on the diet with the lowest lipid level. It could be connected with lower LA and higher EPA, DHA content in this feed com-

pared to the others experimental diets, with the higher level of lipids partially originating from the vegetable source (high LA and low EPA, DHA content). Additionally, no pathological changes were observed in the hepatic and intestinal histological pictures of the fish in groups F6 or F10, in contrast to those of individuals from group F18.

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