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RESEARCH ARTICLE

Impact of sex and size range on fat, cholesterol content, and fatty acid profiles in edible tissues of spiny-cheek crayfish (*Orconectes limosus* Raf.) from Lake Gopło (Poland)

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Abstract. The aim of the present work was to determine the fatty acid profile, the total cholesterol content, and the percentage of fat in the meat of the abdominal section of spiny-cheek crayfish *Oreonectes limosus* (Raf.) caught in Lake Gopło. A total of 138 crayfish, including males, was collected for the study in spring (May 2012). Analyses indicated that fat content was higher in the meat of females (1.31%) than in that of males (1.14%), and there were no statistically significant differences in the fat content between females and males. Differences between the mean fat content in the muscle of males from groups I, II, and III were not statistically significant ($P > 0.05$). The same results were observed for both groups of females. The total cholesterol content was higher in the meat of female crayfish (95.65 mg 100 g^{-1}) than in that of males (88.53 mg 100 g^{-1}). The differences between the mean cholesterol content in the muscles of the three male groups were not statistically significant. The same results were observed for the groups of females. In all the crayfish groups, the main SFA was C16:0, MUFAs were dominated by C18:1 n-6, and the highest percentage of PUFA was of C20:5 n-3.

Keywords: crayfish, sex, size range, fat, cholesterol fatty acids

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Introduction

Several recent studies have focused on the nutritional benefits of seafood, and there are growing numbers of papers promoting the consumption of crustaceans. The white meat of crab, shrimp, crayfish, and lobster is regarded as gourmet seafood. Other valuable products include the meats of Antarctic krill and crayfish. The edible parts of crustaceans are the meat of the claws, legs, and abdomen in crab, the abdomen of shrimp, lobster, and krill, and the abdomen and claws of crayfish. Seafood is rich in protein, liposoluble vitamins, and essential minerals, and it contains low levels of cholesterol (Barrento et al. 2009). Crab meat is a rich source of minerals, especially calcium, iron, zinc, potassium, and phosphorus (Chen et al. 2007). Shellfish meat is rich in protein (25%) and fat (2.5%). The total cholesterol content in the edible parts of shellfish body depends on the species, harvesting period, and the type of

tissue analyzed. Cholesterol content ranges from 40 to 140 mg 100 g^{-1} in crab and from 50 to 170 mg 100 g^{-1} in lobster, while the amount of this sterol in squid tentacles can be as high as 450 mg 100 g^{-1} (Sikorski 2004). Shrimp is a high cholesterol food, although it is a rich source of protein. On the other hand, marine animal food, including shrimp are rich in polyunsaturated fatty acids (PUFA) which are considered anticholesterolemic (Bragagnolo and Rodriguez-Amaya 2001). Numerous data are available regarding the chemical composition and nutritional value of meat from crustaceans, for example, brown crab *Cancer pagurus* L. (Barrento et al. 2010), Chinese mitten crab *Eriocheir sinensis* (Milne Edwards) (Chen et al. 2007), and European lobster *Homarus gammarus* (L.), and American lobsters, *Homarus americanus,* Milne Edwards (Barrento et al. 2009) or Antarctic krill, *Euphausia superba* Dana (Gigliotti et al. 2011, Martin 2007).Crayfish meat is juicy, low-fat, low calorie, and rich in protein, and many consumers consider it to be a delicacy comparable to caviar (Konieczny et al. 2004). Four species of crayfish inhabit the fresh waters of Poland: the noble crayfish, *Astacus astacus* (L.); the pond crayfish, *Astacus leptodactylus* Eschscholtz; the signal crayfish, *Pacifastacus leniusculus* (Dana) and the spiny-cheek crayfish, *Orconectes limosus* (Raf.) Their meat contains 18-20% protein and 0.8-2.8% fat. The meat from the claws is marketed as a canned dish called "crayfish neck". The characteristics of the crayfish body structure make their meat yield relatively low (Walkowiak 1979). It ranges, depending on individual size and species, from 12 to 18% (Krzywosz et al. 2002). The main edible part of spiny-cheek crayfish is meat obtained from the abdomen. Many recipes for dishes with crayfish meat are presented by Mastyński and Andrzejewski (2005).

The aim of the present paper was to compare the fat, total cholesterol content, and fatty acid profile in the meat of the abdominal section of females (two size groups) and males (three size groups) caught in spring in Lake Gopło. The results obtained were compared with the meat quality of other crayfish species, other crustaceans, and the same species analyzed in earlier research.

Study area

Lake Gopło is located in the southern part of Kujawsko-Pomorski Voivodeship. The western part of this lake is in a nature reserve. Lake Gopło is a eutrophic reservoir according to limnological classification, and a pikeperch type lake based on fisheries classification. The zooplankton of Lake Gopło includes 65 species of Rotifera, 34 species of Copepoda, and 8 species of Cladocera. The main benthic group was Chironomidae (dominated by *Chironomus plumosus*). The ichthyofauna is dominated by white bream, *Abramis bjoerkna* (L.). The significant number of the predatory fish in this lake include eel, *Anguilla anguilla* (L.), and pikeperch, *Sander lucioperca* (L.). The quality of Lake Gopło waters is unclassified (Raport WIOŚ 2008).

Materials and methods

Spiny-cheek crayfish were caught in spring (May 2012) in Lake Gopło using pond tools. Crayfish with damaged claws were not analyzed. A total of 138 females and males were collected for the study. The total length of the specimens ranged from from 6.5 to 11.0 cm (Table 1). Total length was measured from the rostrum to the end of the telson. Because of the relatively small amounts of meat obtained from the abdomens of individual crayfish, the material from individuals with similar body lengths (about 8-10 individuals) was combined. As a result, five meat samples were obtained from females and 11 from males. The muscle samples (about 12 g) were freeze dried, and then the fatty acid composition (% of the fatty acids determined), fat content (%), and total cholesterol concentration (mg 100 g^{-1}) were determined. The analyzed meat samples were freeze dried in a Lyovac GT2 freeze-drier by Finn-Aqua (Finland).

About 2 g of freeze-dried tissue was weighed to determine the percentage content of fat in the crayfish meat using the modified method by Folch et al. (1957). The total fat was extracted in duplicate from the muscle using 60 cm^3 (2 × 30 cm^3) of chloroform-methanol

Table 1
Total length (cm), size range, and sex of spiny-cheek crayfish (*Orconectes limosus*) caught in Lake Gopło. n_i, n – number of collected individuals and samples, respectively

	Total length (cm)				
Group	range	mean value	Sex	n_i	n
Group I (small)	6.51-8.00	7.60	-	-	-
			males	21	3
Group II (medium)	8.01-9.50	8.95	females	15	2
			males	50	5
Group III (large)	9.51-11.00	10.55	females	20	3
			males	32	3

n_i, n – number of the collected individuals and samples, respectively

(2:1). After shaking, filtering, and solvent removal, the percentage content of the fat in the tissue was determined (% wet weight).

The cholesterol content was determined with the modified Liebermann-Burchardt colorimetric method (Strzeżek and Wołos 1997) using a Shimadzu (Japan) spectrophotometer. The cholesterol was extracted from 0.25 g samples of freeze-dried tissue with 15 cm^3 of chloroform. After filtration, the solution was supplemented with chloroform in the measurement container to a volume of 25 cm^3. One cm^3 of acetic anhydride and 0.25 cm^3 of sulfuric acid (VI) were added to 2 cm^3 of the filtrate obtained. After 5 minutes, the absorption value was measured in a blind test at a wavelength of 620 nm. The results are presented in mg 100 g^{-1} of wet weight.

The fatty acid profile was determined with an HP 6890N gas chromatograph with a flame-ionization detector (Hewlett-Packard, USA). The temperature of the injector was 225°C, and that of the detector was 250°C, while the column temperature was 180°C. The volume of the injected sample was 1 µl (split 1:50). The analysis was performed on a Supelcowax10 (30 m × 0.32 mm × 0.25 µm) column. The carrier gas was helium at a flow rate of 1 cm^3 min^{-1}. The fat was extracted with the method by Folch et al. (1957). The fatty acid methyl esters were prepared from total lipid with the Peisker method and a mixture of chloroform:methanol:sulfuric acid (100: 100: 1 v/v) (Żegarska et al. 1991). The group of the fatty acids analyzed included saturated acids (SFA), monounsaturated acids (MUFA), and polyunsaturated acids (PUFA). The fatty acid methyl esters were identified using the model and the Supelco 37 component FAME Mix (Supelco, USA).

Atherogenic and thrombogenicity indexes

Lipid quality indexes (atherogenic index – AI and thrombogenicity index – TI), were calculated according to Ulbricht and Southgate (1991). AI = [12:0 + (4×14:0) + 16:0]/[(PUFA n-6 + n-3) + 18:1 + other MUFA]; TI = [14:0 + 16:0 + 18:0]/[0.5×18:1 + 0.5×other MUFA + 0.5×n-6 PUFA + 3×n-3PUFA + (n-3 PUFA/n-6 PUFA)].

Statistical analysis

Data analyses were performed with Statistica 8.0 software (StatSoft, USA). All the data regarding fat, cholesterol, and fatty acids (SFA, MUFA, PUFA, n-3, n-6, n-3/n-6, n-6/n-3 and AI, TI) were analyzed statistically with Tukey's test to compare the mean values between the three groups of males. The significance of differences in average contents were calculated with one-way analysis of variance (ANOVA). Differences in fat and cholesterol content and fatty acid profiles between the two groups of females were analyzed with Student's test. The normality of the data was tested using the Shapiro-Wilk's test, and the homogeneity of variance was tested with Levene's test.

Results

The total fat content of spiny-cheek muscle ranged from 1.00 to 1.35% (Tables 2, 3, 4, and 5). There were no statistically significant differences between these values, which were below 5% and are generally considered to be characteristic of low-fat foods. There were no statistically significant differences (P = 0.830) in fat content between female groups II and III at 1.33 and 1.31%, respectively (Table 4). The mean fat content in the meat of male groups I, II, and III was 1.00, 1.11 and 1.35%, respectively, (Table 5). These differences were not statistically significant (P > 0.05). There were no statistically significant differences in the fat content between females and males (Table 2) or between medium and large individuals (Table 3). The total cholesterol content in the meat of the crayfish caught in Lake Gopło ranged from 87.69 to 96.68 mg 100 g^{-1}. The cholesterol content in the meat of the spiny-cheek males was the highest in group II (89.98 mg 100 g^{-1}) and the lowest in group III (86.96 mg 100 g^{-1}; Table 5), but there were no statistically significant differences (P > 0.05) between these values. The total cholesterol content was higher in the meat of group II females (96.68 mg 100 g^{-1}), and there were no significant differences (P = 0.607) in comparison with the data obtained for group III females (94.61 mg 100 g^{-1}; Table 4). Statistically significant differences were noted in the total cholesterol content in the meat from females (95.65 mg 100 g^{-1}) and males (88.53 mg 100 g^{-1}) caught in spring from Lake Gopło (Table 2) and between medium and large individuals (Table 3). The highest percentage from the SFA group was noted for C16:0 in all groups of the samples obtained from crayfish caught in Lake Gopło. The lowest percentages from the SFA group were for C14:0, C15:0, and C22:0. The average percentage shares of C16:0 and C18:0 differed significantly among the males from the three groups (Table 5). There were no statistically significant differences in SFA content between the medium and large females (Table 4).

The highest MUFA percentage was noted for C18:1 9c, which ranged from 16.88 to 17.38% in the meat from the males, and from 18.52 to 19.32% in that of the females (Tables 4 and 5). The average percentage contents of C14:1 and C18:1 9c differed significantly between female groups II and III. The lowest MUFA percentage was noted for C14:1 and C20:1 n7 in the meat of males and females.

The fatty acid profiles of the spiny-cheek crayfish analyzed were dominated by PUFA, which comprised from 49.15 to 50.18% of the total fatty acids for males and from 47.66 to 49.16% for females. Statistically significant differences were noted for C18:2, C18:3, C20:2, C20:4 n6 and C20:3 n3 between female groups II and III (Table 4). Differences were noted in the male groups for C18:3, C20:2, C20:4 n6, and C22:6 (Table 5). The differences in the fatty acid profiles between females and males are presented in Table 2.

The n3/n6 ratio was 1.14 in female group II and 1.16 in female group III, while that in the meat of the males from groups I, II, and III was 1.36, 1.31, and 1.17, respectively (Tables 4 and 5). The AI index ranged from 0.21 to 0.23 in the meat from males, and was 0.21 in the meat from two groups of females. The mean value of the TI index was 0.19 in the meat from females, and it ranged from 0.19 to 0.20 in that from the males (Tables 4 and 5).

Discussion

The total fat extracted from the crayfish comprised triglycerides, phospholipids, and polar non-phospholipids. As Gigliotti et al. (2011) report, the major lipid classes in all of the fats extracted from Antarctic krill were polar non-phospholipids (> 60% of lipid class in total fat), followed by phospholipids (20-33% of lipid class in total fat), and triglycerides (1-3% of lipid class in total fat). The polar non-phospholipid classes comprised cholesterol, mono- and diglycerides, and red pigment. Total cholesterol included low density lipoproteins (LDL), high density lipoproteins (HDL), and very low density lipoproteins (VLDL). The phospholipids were the major lipid class containing fatty acids (predomi-

Table 2
Fatty acid profile (% of total acid; mean ± SD), total cholesterol (mg 100 g^{-1}), and fat content (%) in the meat of females and males of spiny-cheek crayfish (*Orconectes limosus*) from Lake Gopło. AI – atherogenic index, TI – thrombogenicity index. Values in the same row with different letter indexes differ significantly statistically (t-test, $P < 0.05$)

Fatty acid	females (n=5)	males (n=11)
SFA		
C14:0	0.53 ± 0.05[a]	0.51 ± 0.08[a]
C15:0	0.50 ± 0.04[a]	0.45 ± 0.03[b]
C16:0	14.14 ± 0.43[a]	14.73 ± 0.39[b]
C17:0	0.77 ± 0.06[a]	0.84 ± 0.03[b]
C18:0	5.09 ± 0.18[a]	5.85 ± 0.16[b]
C20:0	0.72 ± 0.05[a]	0.68 ± 0.04[a]
C22:0	0.53 ± 0.06[a]	0.46 ± 0.06[a]
Σ SFA	22.27 ± 0.42[a]	23.56 ± 0.53[b]
MUFA		
C14:1	0.34 ± 0.03[a]	0.24 ± 0.05[b]
C16:1	3.45 ± 0.15[a]	2.83 ± 0.21[b]
C17:1	0.75 ± 0.09[a]	0.86 ± 0.23[a]
C18:1 9c	18.84 ± 0.48[a]	17.12 ± 0.44[b]
C18:1 11c	4.40 ± 0.40[a]	4.16 ± 0.21[a]
C20:1 n9	1.07 ± 0.04[a]	1.08 ± 0.06[a]
C20:1 n7	0.31 ± 0.01[a]	0.29 ± 0.03[a]
Σ MUFA	29.16 ± 0.91[a]	26.61 ± 0.63[b]
PUFA		
C18:2 n6	6.15 ± 0.81[a]	5.76 ± 0.59[a]
C18:3 n3	2.71 ± 0.31[a]	2.01 ± 0.54[b]
C20:2 n6	2.34 ± 0.17[a]	2.16 ± 0.10[b]
C20:3 n6	0.21 ± 0.03[a]	0.20 ± 0.02[a]
C20:4 n6	10.74 ± 1.51[a]	11.32 ± 1.69[a]
C20:3 n3	0.68 ± 0.03[a]	0.66 ± 0.12[a]
C20:4 n3	0.35 ± 0.07[a]	0.33 ± 0.06[a]
C20:5 n3	20.81 ± 0.59[a]	21.34 ± 1.41[a]
C22:5 n6	0.47 ± 0.01[a]	0.41 ± 0.07[a]
C22:5 n3	0.54 ± 0.02[a]	0.65 ± 0.06[b]
C22:6 n3	3.57 ± 0.12[a]	5.00 ± 0.40[b]
Σ PUFA	48.56 ± 1.02[a]	49.83 ± 1.03[a]
Σ n-3	25.94 ± 0.63[a]	27.99 ± 1.46[b]
Σ n-6	22.62 ± 0.55[a]	21.86 ± 1.18[a]
n-3/n-6	1.15 ± 0.03[a]	1.29 ± 0.13[b]
n-6/n-3	0.87 ± 0.02[a]	0.78 ± 0.07[b]
AI	0.21 ± 0.01[a]	0.22 ± 0.01[a]
TI	0.19 ± 0.01[a]	0.19 ± 0.01[a]
total cholesterol	95.65 ± 3.34[a]	88.53 ± 5.63[b]
total fat	1.31 ± 0.08[a]	1.14 ± 0.23[a]

nantly EPA and DHA), and the ratios of n6/n3 and saturated fatty acids/unsaturated fatty acids were higher in the triglycerides (Gigliotti et al. 2011). Konieczny et al. (2004) report that the fat contents in the meat of different crayfish species are similar and do not exceed 1%. Stanek et al. (2011) report that the total fat content in the meat of spiny-cheek crayfish caught in spring and summer in Lake Gopło, ranged

Table 3
Fatty acid profile (% of total acid; mean ± SD), total cholesterol (mg 100 g^{-1}), and total fat content (%) in the meat of spiny-cheek crayfish (*Orconectes limosus*) groups II and III from Lake Gopło. AI – atherogenic index, TI – thrombogenicity index. Values in a row with different letters differ statistically significantly (t-test, $P < 0.05$)

Fatty acid	group II (n=7)	group III (n=6)
SFA		
C14:0	0.51 ± 0.05[a]	0.48 ± 0.05[a]
C15:0	0.45 ± 0.03[a]	0.48 ± 0.04[a]
C16:0	14.56 ± 0.23[a]	14.29 ± 0.49[a]
C17:0	0.80 ± 0.06[a]	0.83 ± 0.05[a]
C18:0	5.59 ± 0.47[a]	5.59 ± 0.44[a]
C20:0	0.70 ± 0.03[a]	0.71 ± 0.04[a]
C22:0	0.48 ± 0.03[a]	0.49 ± 0.07[a]
Σ SFA	23.10 ± 0.58[a]	22.92 ± 0.82[a]
MUFA		
C14:1	0.27 ± 0.07[a]	0.28 ± 0.05[a]
C16:1	3.00 ± 0.46[a]	3.08 ± 0.32[a]
C17:1	0.92 ± 0.19[a]	0.73 ± 0.08[b]
C18:1 9c	17.58 ± 1.24[a]	17.95 ± 0.64[a]
C18:1 11c	4.32 ± 0.27[a]	4.11 ± 0.35[a]
C20:1 n9	1.05 ± 0.05[a]	1.10 ± 0.03[a]
C20:1 n7	0.30 ± 0.02[a]	0.31 ± 0.01[a]
Σ MUFA	27.44 ± 1.80[a]	27.53 ± 1.23[a]
PUFA		
C18:2 n6	6.07 ± 0.73[a]	5.46 ± 0.13[a]
C18:3 n3	2.18 ± 0.61[a]	2.02 ± 0.52[a]
C20:2 n6	2.15 ± 0.07[a]	2.36 ± 0.11[b]
C20:3 n6	0.22 ± 0.02[a]	0.19 ± 0.02[b]
C20:4 n6	10.74 ± 1.38[a]	12.55 ± 0.82[b]
C20:3 n3	0.70 ± 0.14[a]	0.61 ± 0.05[a]
C20:4 n3	0.36 ± 0.05[a]	0.31 ± 0.08[a]
C20:5 n3	21.20 ± 1.02[a]	20.65 ± 0.62[a]
C22:5 n6	0.46 ± 0.06[a]	0.43 ± 0.06[a]
C22:5 n3	0.64 ± 0.07[a]	0.61 ± 0.09[a]
C22:6 n3	4.74 ± 0.87[a]	4.39 ± 0.87[a]
Σ PUFA	49.46 ± 1.27[a]	49.53 ± 0.68[a]
Σ n-3	27.64 ± 1.67[a]	26.56 ± 0.64[a]
Σ n-6	21.82 ± 0.86[a]	23.02 ± 0.16[b]
n-3/n-6	1.27 ± 0.12[a]	1.15 ± 0.03[b]
n-6/n-3	0.79 ± 0.07[a]	0.88 ± 0.02[b]
AI	0.22 ± 0.00[a]	0.21 ± 0.01[a]
TI	0.19 ± 0.00[a]	0.19 ± 0.01[a]
total cholesterol	91.90 ± 7.35[a]	90.78 ± 5.21[a]
total fat	1.17 ± 0.25[a]	1.33 ± 0.06[a]

from 0.92 to 1.10% wet weight. As earlier research shows, the average fat contents in crayfish meat in individuals from the Brda River and Lake Gopło were 0.43 and 0.44% wet weight, respectively (Stanek et al. 2010). Walkowiak (1979) reports that the amount of fat in crayfish necks ranged from 0.4 to 0.9%. Reports by Własow et al. (2002) show that the percentage content of fat in the meat of crayfish caught in the Mazurian Lake District ranged from 0.15 to 0.30%. Similar values are reported for crayfish

Table 4
Fatty acid profile (% of total acid; mean ± SD), total cholesterol (mg 100 g^{-1}), and total fat content (%) in the meat of two groups of females of spiny-cheek crayfish (*Orconectes limosus*) from Lake Gopło. AI – atherogenic index, TI – thrombogenicity index. Values in a row with different letters differ statistically significantly (t-test, $P < 0.05$)

Fatty acid	group II (n=2)	group III (n=3)
SFA		
C14:0	0.56 ± 0.04[a]	0.51 ± 0.05[a]
C15:0	0.48 ± 0.04[a]	0.51 ± 0.04[a]
C16:0	14.44 ± 0.17[a]	13.94 ± 0.46[a]
C17:0	0.73 ± 0.06[a]	0.80 ± 0.05[a]
C18:0	4.92 ± 0.07[a]	5.20 ± 0.13[a]
C20:0	0.70 ± 0.06[a]	0.73 ± 0.04[a]
C22:0	0.50 ± 0.06[a]	0.55 ± 0.06[a]
Σ SFA	22.33 ± 0.42[a]	22.24 ± 0.51[a]
MUFA		
C14:1	0.37 ± 0.01[a]	0.32 ± 0.02[b]
C16:1	3.60 ± 0.12[a]	3.36 ± 0.08[a]
C17:1	0.79 ± 0.11[a]	0.73 ± 0.10[a]
C18:1 9c	19.32 ± 0.27[a]	18.52 ± 0.17[b]
C18:1 11c	4.61 ± 0.11[a]	4.26 ± 0.48[a]
C20:1 n9	1.04 ± 0.06[a]	1.09 ± 0.02[a]
C20:1 n7	0.31 ± 0.02[a]	0.32 ± 0.00[a]
Σ MUFA	30.02 ± 0.11[a]	28.59 ± 0.65[a]
PUFA		
C18:2 n6	7.04 ± 0.15[a]	5.56 ± 0.07[b]
C18:3 n3	3.04 ± 0.10[a]	2.49 ± 0.11[b]
C20:2 n6	2.17 ± 0.05[a]	2.45 ± 0.09[b]
C20:3 n6	0.24 ± 0.01[a]	0.19 ± 0.02[a]
C20:4 n6	9.11 ± 0.17[a]	11.83 ± 0.34[b]
C20:3 n3	0.71 ± 0.00[a]	0.66 ± 0.01[b]
C20:4 n3	0.41 ± 0.06[a]	0.31 ± 0.03[a]
C20:5 n3	20.46 ± 0.49[a]	21.04 ± 0.61[a]
C22:5 n6	0.47 ± 0.01[a]	0.47 ± 0.02[a]
C22:5 n3	0.54 ± 0.03[a]	0.53 ± 0.02[a]
C22:6 n3	3.50 ± 0.10[a]	3.61 ± 0.14[a]
Σ PUFA	47.66 ± 0.30[a]	49.16 ± 0.81[a]
Σ n-3	25.61 ± 0.43[a]	26.16 ± 0.72[a]
Σ n-6	22.05 ± 0.13[a]	23.01 ± 0.24[b]
n-3/n-6	1.16 ± 0.03[a]	1.14 ± 0.03[a]
n-6/n-3	0.86 ± 0.02[a]	0.88 ± 0.02[a]
AI	0.21 ± 0.00[a]	0.21 ± 0.01[a]
TI	0.19 ± 0.00[a]	0.19 ± 0.01[a]
total cholesterol	96.68 ± 1.94[a]	94.61 ± 4.66[a]
total fat	1.33 ± 0.13[a]	1.31 ± 0.07[a]

caught in lakes Dgał, Harsz, and Pobłędzie (0.24-0.30%; Własow et al. 2005). These values indicate that the fat content in crayfish meat is similar to that of lean fish, which contains up to 2% fat (Sikorski 2004). The fat content of krill was low, but it was rich in EPA and DHA (Martin 2007). Analyses of shrimp conducted by Krzynowek and Panunzio (1989) indicate that these crustaceans have low fat contents at about 1%. Several studies have shown that muscles are the principal sites of lipid storage in

Table 5
Fatty acid profile (% of total acid; mean ± SD), total cholesterol (mg 100 g^{-1}), and total fat content (%) in the meat of three groups of males of spiny-cheek crayfish (*Orconectes limosus*) from Lake Gopło. AI – atherogenic index, TI – thrombogenicity index. Values in a row with different letters differ statistically significantly (Tukey's test, $P < 0.05$)

Fatty acid	group I (n=3)	group II (n=5)	group III (n=3)
SFA			
C14:0	0.61 ± 0.08[a]	0.49 ± 0.04[b]	0.44 ± 0.01[b]
C15:0	0.48 ± 0.06[a]	0.44 ± 0.01[a]	0.45 ± 0.01[a]
C16:0	15.01 ± 0.66[a]	14.60 ± 0.25[a]	14.64 ± 0.12[a]
C17:0	0.82 ± 0.03[a]	0.83 ± 0.03[a]	0.87 ± 0.01[a]
C18:0	5.69 ± 0.21[a]	5.86 ± 0.08[a,b]	5.99 ± 0.05[b]
C20:0	0.67 ± 0.09[a]	0.70 ± 0.02[a]	0.69 ± 0.02[a]
C22:0	0.47 ± 0.12[a]	0.48 ± 0.02[a]	0.43 ± 0.01[a]
Σ SFA	23.76 ± 1.08[a]	23.41 ± 0.20[a]	23.62 ± 0.11[a]
MUFA			
C14:1	0.28 ± 0.09[a]	0.23 ± 0.01[a]	0.23 ± 0.01[a]
C16:1	2.98 ± 0.21[a]	2.76 ± 0.24[a]	2.80 ± 0.10[a]
C17:1	0.89 ± 0.33[a]	0.98 ± 0.19[a]	0.72 ± 0.08[a]
C18:1 9c	17.28 ± 0.54[a]	16.88 ± 0.41[a]	17.38± 0.17[a]
C18:1 11c	4.29 ± 0.16[a]	4.21 ± 0.23[a]	3.95 ± 0.02[a]
C20:1 n9	1.09 ± 0.10[a]	1.05 ± 0.05[a]	1.10 ± 0.04[a]
C20:1 n7	0.28 ± 0.05[a]	0.30 ± 0.03[a]	0.30 ± 0.01[a]
Σ MUFA	27.09 ± 1.02[a]	26.41 ± 0.43[a]	26.48 ± 0.25[a]
PUFA			
C18:2 n6	6.30 ± 0.86[a]	5.68 ± 0.38[a]	5.36 ± 0.06[a]
C18:3 n3	2.73 ± 0.47[a]	1.84 ± 0.18[b]	1.56 ± 0.09[b]
C20:2 n6	2.07 ± 0.05[a]	2.15 ± 0.08[a,b]	2.27 ± 0.03[b]
C20:3 n6	0.20 ± 0.02[a]	0.21 ± 0.02[a]	0.18 ± 0.01[a]
C20:4 n6	9.24 ± 0.43[a]	11.40 ± 1.00[b]	13.27 ± 0.11c
C20:3 n3	0.69 ± 0.05[a]	0.69 ± 0.17[a]	0.57 ± 0.01[a]
C20:4 n3	0.34 ± 0.04[a]	0.34 ± 0.03[a]	0.31 ± 0.13[a]
C20:5 n3	22.18 ± 2.16[a]	21.50 ± 1.06[a]	20.25 ± 0.33[a]
C22:5 n6	0.37 ± 0.05[a]	0.45 ± 0.07[a]	0.39 ± 0.04[a]
C22:5 n3	0.59 ± 0.06[a]	0.68 ± 0.03[a]	0.68 ± 0.07[a]
C22:6 n3	4.44 ± 0.18[a]	5.24 ± 0.18[b]	5.17 ± 0.21[b]
Σ PUFA	49.15 ± 2.00[a]	50.18 ± 0.34[a]	49.90 ± 0.29[a]
Σ n-3	28.23 ± 2.45[a]	28.45 ± 1.12[a]	26.97 ± 0.03[a]
Σ n-6	20.92 ± 1.18[a]	21.73 ± 1.04[a]	23.03 ± 0.09[a]
n-3/n-6	1.36 ± 0.18[a]	1.31 ± 0.11[a]	1.17 ± 0.00[a]
n-6/n-3	0.75 ± 0.09[a]	0.77 ± 0.07[a]	0.85 ± 0.00[a]
AI	0.23 ± 0.01[a]	0.22 ± 0.01[a]	0.21 ± 0.00[a]
TI	0.19 ± 0.02[a]	0.19 ± 0.00[a]	0.20 ± 0.00[a]
total cholesterol	87.69 ± 4.29[a]	89.98 ± 8.00[a]	86.96 ± 1.48[a]
total fat	1.00 ± 0.12[a]	1.11 ± 0.26[a]	1.35 ± 0.05[a]

crustaceans. The gonads in females also seem to be a lipid storage site, and decreased lipids in female meat in spring and summer could be related to the reproductive period. The total fat and cholesterol levels in ovaries increased in the period prior to reproduction (Silva-Castiglioni et al. 2007). During periods with high energy demands, such as mottling and gametogenesis, lipids are mobilized mainly from the hepatopancreas to the gonads.

Cholesterol is a very important component of food, and it is an essential biological compound that is distributed widely in foods. Moderate values of daily human intake of cholesterol is considered to be 140 mg (Barrento et al. 2009). Cholesterol is a structural component of cell membranes and a forerunner of the sex hormones involved in reproductive control in crustaceans (Buckup et al. 2008). Stanek et al. (2011) report that the total cholesterol content in the meat of crayfish caught in Lake Gopło in 2010 ranged from 64.84 to 72.11 mg 100 g^{-1}. The total cholesterol content in male brown crab meat ranged from 37.0 to 40.7 mg 100 g^{-1} wet weight (Barrento et al. 2010). The cholesterol concentration in the leg and claw meat of green crab, *Carcinus maenas* (L.), ranged from 57.2 to 64.8 mg 100 g^{-1} (Skonberg and Perkins 2002). The lipid content and its composition in krill depend on the species, age, and time elapsed between capture and freezing. The cholesterol content in krill meat is higher than in fish meat, but lower than in shrimp. It is significant that two-thirds of the sterols in shellfish are non-cholesterol sterols (Gigliotti et al. 2011). Krzynowek and Panunzio (1989) report that the mean content of total cholesterol in the meat of several shrimp species was 152±15 mg 100 g^{-1}. The shrimp meat analyzed by Sampaio et al. (2006) had a cholesterol content range of 142 to 191 mg 100 g^{-1}. This variation was probably associated to several factors: species, available feeding, age, sex, water temperature, geographical location, and collection season (Sampaio et al. 2006). Analyses of the concentration of metabolic substances in the muscle tissue of the crayfish *Parastacus varicosus* Faxon indicated that there were no statistically significant differences in the total cholesterol content between the sexes (Silva-Castiglioni et al. 2007). These authors analyzed the differential response in cholesterol reserves in muscle tissues in females in spring and summer, which could have been related to the reproduction, because cholesterol levels in the ovaries increased in this period. Buckup et al. (2008) analyzed seasonal variations in the biochemical composition of the crayfish *Parastacus defossus* Faxon in its natural environment, and they report no significant differences in total cholesterol content in the muscle tissue of the two sexes over four seasons. They also report that the total cholesterol content in the muscle tissue of males was the highest in summer (0.2 mg g^{-1}), while that in females was the highest in spring (0.8 mg g^{-1}). Barrento et al. (2009) report no statistically significant differences in the cholesterol content between the sexes in European lobster meat (36.6 mg 100 g^{-1} – females; 31.2 mg 100 g^{-1} – males) and American lobster (43.2 mg 100 g^{-1} – females; 37.2 mg 100 g^{-1} – males). Differences between the sexes are observed in the hepatopancreas cholesterol content, which confirms that cholesterol is important in crustacean ovary maturation (Barrento et al. 2009). In a study of shrimp from Santa Catarina, Bragagnolo and Rodriguez-Amaya (2001) report that size did not significantly influence cholesterol content in *Penaeus schimitti* (Burkenroad), but it was significant among *Penaeus brasiliensis* (Latreille) since the cholesterol content is significantly lower in large shrimp.

The fatty acid composition of shellfish varies with species, the water temperature they inhabit and from which they are caught, their food, habitat, and sex (Barrento et al. 2009). Differences in fatty acid profiles between the female and male crayfish analyzed presented in Table 2 are probably related to the cumulative effects of diet, molting cycle, and reproductive metabolism. In all the groups of samples obtained from crayfish caught in Lake Gopło, the highest percentage share of the SFA analyzed was of C16:0 (Tables 2-4). In an earlier study, Stanek et al. (2010) report the highest percentage share of C16:0 in the meat of crayfish caught in the Brda River (21.33% of total acids) and Lake Gopło (15.36% of total acids). The mean percentage share of all SFA ranged from 21.26 to 22.56% of the total acids in the meat of 3- and 4-year-old males caught in Lake Gopło in 2010 (Stanek et al. 2011).

The mean percentage share of all MUFA in the analyzed crayfish ranged from 26.41 to 27.09% for males and from 28.59 to 30.02% for females. The highest percentage of the MUFA group was of C18:1 9c. The same results were observed in an earlier studies by Stanek et al. (2010, 2011). Walkowiak (1979) also reports that the highest share of the

MUFA group in crayfish carapace extract was of C18:1 9c. The same results were obtained for brown crab meat (Barrento et al. 2010), and the highest amounts of total fatty acids were reported for C18:1 9c (30.96%) in Chinese mitten crab meat (Chen et al. 2007).

The fatty acid profiles of the spiny-cheek crayfish analyzed in the current study were dominated by PUFA (Tables 2-5). The importance of a balanced PUFA intake has been recognized by health organizations throughout the world in the past decade, and there is now a consensus that PUFA should comprise at least 3%, and preferably 8-23%, of the total lipid intake (Latyshev et al. 2009). The same results were observed by Stanek et al. (2011) for 3- and 4-year old male spiny-cheek crayfish caught in Lake Gopło in 2010 (39.18%) and in the Brda River (37.76%), by Naczk et al. (2004) for green crab (47.1-50.5%), by Barrento et al. (2010) for brown crab (48.4-50.7%), and by Barrento et al. (2009) for European (1.9-2.0 mg 100 g^{-1}) and American (2.0-2.2 mg 100 g^{-1}) lobsters. Barrento et al. (2010) report that PUFA content in brown crab meat did not differ between females and males. The analyses of PUFA in the meat of spiny-cheek crayfish from Lake Gopło indicated that the highest percentage share was of C20:5 n-3 (EPA), and the lowest amounts were of C20:3 n-6 (Tables 2-5). Statistical analyses indicated that there were statistically significant differences between two groups of females for C18:2, C18:3, C20:2, C20:4 n6 and C20:3 n3 (Table 4). Among the males, differences were detected for C18:3, C20:2, C20:4 n6 and C22:6 (Table 5). Walkowiak (1979) demonstrates that the sum of PUFA in crayfish carapace extract was 34.7%. PUFA from the n-3 series, especially DHA and EPA, have been identified in the past few decades as essential nutrients for marine animals in general. EPA and DHA comprised about 30% of the total fatty acids in shrimp meat (Krzynowek and Panunzio 1989). EPA concentrations in the meat of various shellfish species analyzed by Mahaffey (2004) ranged from 0.01 to 1.5 g 100 g^{-1} and DHA was from 0.01 to 2.00 g 100 g^{-1}. The dominant PUFA in green crab meat were EPA (22.3-26.5%) and DHA (9.38-13.4%) (Naczk et al. 2004). DHA and EPA were also the most abundant (% of the total fatty acids) fatty acids in langoustine *Nephrops norvegicus* (L.) (EPA-15.30, DHA-18.45), lobster *Palinurus vulgaris* Latreille (EPA-11.17, DHA-10.79), and shrimp *Peanaeus kerathurus* (ForsskÍl) (EPA-17.28, DHA-13.05) (Tsape et al. 2010). The main PUFA in the lipids of the organs of crab from the northwest Pacific were EPA and DHA, and they also comprised the majority of the sum of total acids in muscle tissues. EPA percentages of total fatty acids ranged from 13.5% in *Chionoecetes angulatus* Rathbun to 31.8% in *Chionoecetes opilio* (O. Frabricius), while those for DHA ranged from 5.92% in *Ch. angulatus* to 19.2% in *Chionoecetes japonicas* Rathbun (Latyshev et al. 2009).

The n3/n6 ratios in the meat of the three male groups of spiny-cheek crayfish from Lake Gopło was 1.36, 1.31, and 1.17, respectively. The same ratio in the meat from female groups II and III was 1.14 and 1.16, respectively (Tables 3 and 4). These values did not differ statistically significantly between females and males. The same results were observed for crayfish caught in Lake Gopło in 2010, and in which the n3/n6 ratio ranged from 0.92 to 1.04. The lowest value was determined in the meat of 4-year-old males, which differed statistically significantly from those of the other crayfish groups (Stanek et al. 2011). Stanek et al. (2010) reports that the n3/n6 PUFA ratios in the meat of crayfish from the Brda River and Lake Gopło were 0.72 and 0.70, respectively. The ratio of n3/n6 in lobster meat was higher at 4.2 in the females and 4.1 in males (Barrento et al. 2009). This ratio in male crab muscle was 3.5 and 4.0 (Barrento et al. 2010). This coefficient was within the range for freshwater fish (0.5-3.8) (Steffens and Wirth 2005). Increased values of the n3/n6 PUFA ratio increases the availability of n-3 PUFA, which are beneficial for human health. FAO experts recommend that the n6/n3 PUFA ratio in the diet should be between 5:1 and 10:1 (Chen et al. 2007). The n6/n3 ratio in the meat of spiny-cheek crayfish from Lake Gopło ranged from 0.92 to 1.04 (Table 3 and 4). The United Kingdom Department of Health recommends an n6/n3 ratio of 0.25 as the ideal for human diets (Barrento et al. 2010).

SFA, MUFA, and n-6 PUFA are dietary indicators of meat quality. The atherogenic index (AI) and thrombogenicity index (TI) are lipid quality indicators, and they are determined based on the relative contents of particular groups of fatty acids These indexes indicate the overall dietary quality of lipids and their potential effects on the development of coronary disease (Jankowska et al. 2010). The AI ranged from 0.21 to 0.23 in the male meat, and was 0.21 in the female meat from groups I and II. The mean value of TI was 0.19 in the female meat and ranged from 0.19 to 0.20 in that of the males (Tables 3, 4). These values were lower than those obtained for the other food products such as lamb, beef, pork, rabbit, and chicken. The AI ranged from 0.22 to 0.26, and TI was from 0.14 to 0.17 in lobster muscle (Barrento et al. 2009), and the values for crab meat were 0.17 and 0.12, respectively (Barrento et al. 2010).

The results of this study concur with the common knowledge that seafood is a rich source of essential PUFAs, especially those from the n-3 group, and that cholesterol contents are low. Seafood is considered a low-fat food like many fish species, and almost all shellfish contains less than 2.5% total fat in muscles with only a small number of species containing more than 15% (Barrento et al. 2010). These values are considered to be characteristic of low-fat food.

Conclusions

1. The total fat content of spiny-cheek crayfish muscle were independent of sex.
2. Analyses indicated that there were differences in the total cholesterol content between females and males.
3. The fatty acid profile of the analyzed crayfish lipids was dominated by PUFA. There were significant differences in the contents of SFA, MUFA, and PUFA between females and males.
4. There were no differences in the contents of most of the analyzed fatty acids, total fat, or cholesterol in the meat of small and large individuals. Differences were noted for C17:1, C20:2, C20:3 n6, and C20:4 n6.

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