Impact of frozen storage on fatty acid profile in goose meat

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ABSTRACT The objective of this study was to investigate the changes of the fatty acid in breast (**BM**) and leg (**LM**) muscles from 17-wk-old female White Kołuda geese packaged in a vacuum and stored in freezing conditions at $-20°C$. During 17 weeks, the geese were fed ad libitum on the same complete feed. The samples (18 LM and 18 BM) from the right part of the carcasses were stored for 30, 90, 80, 270, and 365 d. The changes in the fatty acid profile were established by gas chromatography. In this work, there were also calculated lipid profile indicators such as Σ PUFA $n-6/\Sigma$ PUFA $n-3$, Σ UFA/Σ SFA, and Σ PUFA/Σ SFA. Time of frozen storage affected the decrease in Σ SFA, Σ MUFA, and Σ PUFA of BM and LM. The statistical analysis of the obtained data shows that the type of muscle also generally affected the fatty acid profile. The BM are characterized higher proportion of Σ SFA, and the LM are defined as containing more Σ MUFA and Σ PUFA. Extending frozen storage time caused only the deterioration of Σ PUFA $n-6/\Sigma$ PUFA $n-3$. The Σ PUFA $n-6/\Sigma$ PUFA $n-3$ were the highest in BM and LM on the 365th day of storage. Although the Σ PUFA $n-6/\Sigma$ PUFA $n-3$ ratio in muscles stored for 180, 270, and 365 d was higher than the recommended values. The lipid profile indicators (Σ UFA/Σ SFA, and Σ PUFA/Σ SFA) were similar in raw meat and in all frozen storage samples. It means that frozen storage didn't affect this index and the BM and LM have the same quality from the dietary point of view. Leg muscles during frozen storage are characterized by higher Σ UFA/Σ SFA and Σ PUFA/Σ SFA than the breast muscles.

Key words: goose, frozen storage, fatty acid profile, lipid indicators, meat

INTRODUCTION

The meat industry and meat processing occupy an important place in the world. For centuries, meat and its products have provided the human body with essential components of our diet. Meat is rich in amino acids, fatty acids, some vitamins, and minerals (Soren and Biswas, 2019; Wołoszyn et al., 2020; Geletu et al., 2021). Meat consumption trends vary from region to region across the world. In some parts of the world, meat consumption might be increasing, while in others, it might be decreasing. It depends on meat consumption trends in different parts of the world (Suleman et al., 2020).

In Poland, goose meat is increasingly popular among consumers because of its good quality. According to statistics, the total geese production in Poland was 1.988 thousand carcasses, and it was about 20.000 tons of goose meat (GUS, 2021). Geese is herbivorous, seasonal poultry and has high dietary quality (Weng et al., 2021). Moreover, in Poland, the basic breed used to produce goose meat is White Kołuda geese, and they are called "Polish oat geese" because the birds are fattened freely with oats in the last 3 wk of rearing. Oat fattening gives unique health-promoting and taste qualities to goose meat and fat (Nowicka, 2018).

Goose meat, among others, is very favorable from a nutritional point of view. It contains all the essential amino acids and the highest amount of unsaturated fatty acids among all kinds of meat (Boz et al., 2019; Guo et al., 2020; Wereńska et al., 2021). Furthermore, waterfowl are characterized by good quality and a good fatty acids composition (Biesek et al., 2020). Goose fat is one of the healthiest animal fats, and it is considered safe for consumers due to its relatively low level of saturated fatty acids (Wołoszyn et al., 2020). The traditional method of fattening birds for oat grain results in the formation of fat in goose meat with a higher content of valuable polyunsaturated fatty acids (**PUFA**s) (Uhlíová et al., 2018; Biesek et al., 2020).

Due to the seasonality of the goose raw material, it must be frozen to maintain its supply throughout the

year. Freezing is one of the most significant strategies for preserving food quality during long-term storage. Unfortunately, many unfavorable changes occur during frozen storage. This kind of storage may alter physical properties, chemical processes, such as protein aggregation, denaturation, and their oxidation, color changes, lipolysis, lipid oxidation, and sensory properties of meat (Li et al., 2022). The changes occurring in the raw meat material during its freezing storage are influenced by exogenous factors, such as oxygen in the atmospheric air, temperature, storage time, as well as the presence of some heavy metal ions (Amaral et al., 2018; Beltrán and Bellés, 2018; Wereńska et al., 2022). Meat lipids are also subject to oxidation processes due to the activity of endogenous factors, that is, tissue enzymes and microbial origin (Arshad et al., 2013; Domínguez et al., 2019). This endogenous factors are inhibited by freezing and not retained. In frozen meat, there are active microorganisms and exogenous enzymes all the time. For this reason freezing does not protect meat against peroxidative processes but only slows them down to a certain extent. Moreover during frozen storage of meat, the ester bonds between the glycerol molecule and fatty acids are hydrolyzed, and fatty acids are oxidized (Contini et al., 2014; Temkov and Mureşan, 2021).

Lipid oxidation processes are one of the most important factors limiting the shelf life of meat. The oxidation dynamics are influenced mainly by the species of animals, the way they are fed, and the profile of fatty acids in lipids (Falowo et al., 2017; Domínguez et al., 2019). Oxidation in meats is influenced by the fatty acids polyunsaturated present in the phospholipids of the cell membranes of the meats, and they are the main targets of oxidative rancidity (Ribeiro et al., 2019). Waterfowl meat contains a high proportion of unsaturated fatty acids (65−75%). Thus, this meat is more susceptible to oxidation than the meat of other poultry species (Banaszak et al., 2020; Wereńska et al., 2021).

That is why, the main aim of the present study was to investigate the changes of fatty acids in BM (breast muscle) and LM (leg muscle) stored in freezing condition at −20°C.

MATERIAL AND METHODS

Meat Samples

The experimental material consisted of breast muscle (n = 18) and leg (thigh) muscles (n = 18) from 17-wk old female White Kołuda geese (W 31), which are called "Polish oat geese." The geese were reared in the same industrial farm and fed on the same complete concentrated diet (Wołoszyn et al., 2020). The birds were slaughtered in a industrial slaughterhouse according to EU regulations. The carcasses were bled, scalded (approximately 1 min, at 63°C), plucked, and eviscerated. The eviscerated carcasses were placed immediately inside a refrigerator at 4°C for 24 h. After that, the BM and LM were cut out from the right side of the carcasses, and then individually packed in a head shrink bag Supravis SHRINK BAG P. The average weight for BM (with skin and subcutaneous fat) was 465 g ± 21 g, and for LM, it was (with skin and subcutaneous fat) 405 g ± 18 g. The packed muscles were frozen in an air tunnel at −20°C, measured at their geometric center. Then, the muscles were placed in a freezer cabinet (HSA29530N, Beko, Warszawa, Poland) and stored for 30, 90, 180, 270, and 365 d at a temperature of −20°C (±1°C). Each time, 18 BM and 18 LM were investigated. Thirty-six (18 breasts + 18 legs) fresh muscles (24 h after slaughter at +4°C) were used for the control (**C**) group, and the results obtained for this group were taken as initial values.

Sample Preparation

To analyze fatty acid profile the muscles were thawed in a refrigerated cabinet for 24 h at +4°C (LG, M600, Seul, South Korea). Next, the skin and subcutaneous fat were separated from the muscles. Each BM and LM has been ground (mesh diameter of 2 mm) in an electric bowl chopper (model MM/1000/887, Zelmer, Rzeszów, Poland). The lipids were extracted using the procedure described by Folch et al. (1957). According to this method, each ground sample (5 g) was separately homogenized using chloroform:methanol (2:1; v/v) solution. The extraction mixture contained 0.001% (w/v) of butylated hydroxytoluene as an antioxidant. The organic solvent was evaporated under a stream of nitrogen. Then the lipid extracts were saponified with 0.5 M KOH solution. Afterward, the methyl esters of fatty acids (FAMEs) were prepared by transesterification according to the AOCS official method Ce 2−66 (AOCS, 1997).

Fatty Acid Analysis

The composition of fatty acids was determined using the gas chromatography technique using a Chromatograph model 7890A (Agilent Tech., Santa Clara, CA) equipped with a flame-ionization detector (FID) and automatic sampler Agilent 7683 (Agilent Tech., Santa Clara, CA). The used column was HP-88 (100 m*0.25 mm ID*0.2 μm). The inlet temperature was carried out at 250°C, injection volume was 1 μL, and a split ratio of 1/50. The gases used to detect the fatty acids were: hydrogen (40 mL/min), air (450 mL/min) and helium make-up gas (30 mL/min). The initial column temperature of 120°C was held for 1 min, increased to 175°C at 10°C/min and then held for 10 min. Then, it was increased to 210°C at 5°C/min, held for 5 min, increased to 230°C at a rate of 5°C/min, and maintained for 5 min. At the end, the detector temperature was at 280°C.

The FAME's peaks were identified by comparing the retention times with those of a mixture of external standard methyl esters (Supelco 37 Component F.A.M.E. Mix, C4−C24, Sigma-Aldrich, St. Louis, MI). The fatty acids were calculated as a percentage (w/w) of total fatty acids with the Agilent ChemStation program (Agilent Tech. Inc.). Each sample was analyzed in triplicates.

Table 1. The share of SFA (saturated fatty acid) in BM and LM of White Koluda geese.

			Time of freezing storage (days)				
			30	90	180	270	365
Parameters [%]	Type of muscle	Control group C			n=18		
C 14:0	BM	0.53 ± 0.08	$^x0.51 \pm 0.05$	0.49 ± 0.07	$^x0.49 \pm 0.04$	0.46 ± 0.07	0.45 ± 0.09
Myristic acid	LM	0.48 ± 0.07	$^y0.44 \pm 0.06$	0.46 ± 0.07	$^y0.43 \pm 0.05$	0.41 ± 0.08	0.41 ± 0.07
C 16:0	BM	$^x21.94^a \pm 0.29$	$^x21.88^a \pm 0.21$	$^x21.85^a \pm 0.22$	$^x21.60^b \pm 0.21$	$^x21.55^b \pm 0.17$	$^x21.36^c \pm 0.18$
Palmitic acid	LM	$^y21.23^a \pm 0.24$	$^y21.07^a \pm 0.20$	$^y20.99^a \pm 0.21$	$^y20.71^b \pm 0.18$	$^y20.60^b \pm 0.19$	$^y20.51^b \pm 0.22$
C 18:0	BM	$8.39^a \pm 0.17$	$8.35^a \pm 0.16$	$8.20^b \pm 0.14$	$7.85^c \pm 0.11$	$7.61^d \pm 0.17$	$7.38^e \pm 0.21$
Stearic acid	LM	$8.37^a \pm 0.16$	$8.31^a \pm 0.14$	$8.15^b \pm 0.13$	$7.92^c \pm 0.19$	$7.65^d \pm 0.18$	$7.34^e \pm 0.20$
Σ SFA	BM	$^x30.86^a \pm 0.45$	$^x30.74^{ab} \pm 0.42$	$^x30.54^{bc} \pm 0.41$	$^x29.94^c \pm 0.48$	$^x29.62^c \pm 0.35$	$^x29.19^d \pm 0.31$
	LM	$^y30.08^a \pm 0.49$	$^y29.82^a \pm 0.39$	$^y29.60^b \pm 0.33$	$^y29.06^c \pm 0.39$	$^y28.66^d \pm 0.32$	$^y28.26^e \pm 0.36$

[a-e]Different letters in a row mean statistically significant differences between group average, including storage time ($P \leq 0.05$).
[x-y]Different letters in columns mean statistically significant differences between the group average, including the type of muscle ($P \leq 0.05$).

Calculation

The sum of SFA, PUFA, PUFA $n-3$, PUFA $n-6$, UFA were calculated by the following:

- Σ PUFA = Σ PUFA $n-3$ + Σ PUFA $n-6$ = α C 18:3 $n-3$ + C 20:5 $n-3$ + C 22:6 $n-3$ + C 18:2 $n-6$ + C 20:3 $n-6$ + C 20:4 $n-6$;
- Σ UFA = Σ PUFA + Σ MUFA = α C 18:3 $n-3$ + C 20:5 $n-3$ + C 22:6 $n-3$ + C 18:2 $n-6$ + C 20:3 $n-6$ + C 20:4 $n-6$ + C 16:1 $n-7$ + C 18:1 $n-9$ + C 20:1 $n-9$ + C 24:1 $n-9$;
- Σ PUFA n-6/Σ $n-3$ = (C 18:2 $n-6$ + C 20:3 $n-6$ + C 20:4 $n-6$)/(α C 18:3 $n-3$ + C 20:5 $n-3$ + C 22:6 $n-3$);
- Σ UFA/Σ SFA = (Σ PUFA + Σ MUFA)/Σ SFA = (α C 18:3 $n-3$ + C 20:5 $n-3$ + C 22:6 $n-3$ + C 18:2 $n-6$ + C 20:3 $n-6$ + C 20:4 $n-6$ + C 16:1 $n-7$ + C 18:1 $n-9$ + C 20:1 $n-9$ + C 24:1 $n-9$)/(C 14:0 + C 16:0 + C 18:0);
- Σ PUFA/Σ SFA = (Σ PUFA $n-3$ + Σ PUFA $n-6$)/Σ SFA = (α C 18:3 $n-3$ + C 20:5 $n-3$ + C 22:6 $n-3$ + C 18:2 $n-6$ + C 20:3 $n-6$ + C 20:4 $n-6$)/(C 14:0 + C 16:0 + C 18:0).

Statistical Analysis

The data were analyzed as a completely randomized design using a 2-way ANOVA concerning the kind of muscles (breast and leg) and time its frozen storage (30, 90, 180, 270, and 360 d) as a factorial design (2 × 5), according to the following linear model: $Y_{ij} = \mu + A_i + B_j + (AB)_{ij} + e_{ij}$, where Y_{ij} = value of trait (the dependent variable); μ = overall mean; A_j = effect of kind of muscle; B_j = effect of time frozen storage of muscles; (AB) = interaction and e_{ij} = random observation error, using Statistica13.3 software (StatSoft Inc., 2019). The statistical significance of the differences between the averages of the groups was calculated using Tukey's test and was at a level of $P \leq 0.05$. The Tables present the average values and their standard deviations.

RESULTS AND DISCUSSION

Saturated Fatty Acid (SFA)

Analysis of the proportion of SFA in both types of muscles stored for 24 h under refrigeration showed that the dominant was palmitic acid (C 16:0). Its average percentage was 21.94% (BM) and 21.23% (LM). On the other hand, the proportion of myristic (C 14:0) and stearic (C 18:0) acids in the lipids of both types of muscles was similar, and the differences were not statistically confirmed ($P \geq 0.05$). The BM were characterized by a higher ($P \leq 0.05$) share of Σ SFA (30.86%) compared to the LM (30.08%) (Table 1).

The share of the C 14:0, C 16:0, and C 18:0 in both types of muscles were similar to the results obtained by Gumułka et al. (2006). They analyzed the fatty acid profile of the intramuscular fat of the White goose Koluda, and Polish geese such as: Kielecka, Lubelska, Suwałki, and Podkarpacka. On the other hand, Biesiada-Drzazga (2006) showed that the share of C 14:0 and C 18:0 acids in breast muscles and leg muscles obtained from geese of the same genotype was lower (about 0.3% and 1.8% respectively) than our results. These differences could result, among others, from the different composition of feed given to birds during their rearing, which, as is well known, significantly affects the fatty acid profile of meat lipids.

It was found that by extending the frozen storage of both types of muscles, the percentage of C 16:0 and C 18:0 in their lipid fraction decreased (Table 1). Significant changes in the C 18:0 share were found in BM and LM from the 90th d, while the C 16:0 from the 180th d of their storage. The decreasing of percent yield of C 18:0 could have been caused synthesis from C 18:0 to the C 18:1 with the aid of delta-9 desaturase. The activity of this enzyme could therefore play an important role to the amount of C 18:0 and C 18:1 in geese tissues during frozen storage. It is possible that the delta-9 desaturase activity was lower than the tempo of oxidation processes taking place. For this reason, the proportion of the C 18:1 in our research did not change until the 180th (BM) and until the 270th (LM) day of storage, and then it decreased due to oxidative processes. These

Table 2. The share of MUFA (monounsaturated fatty acid) in BM and LM of White Koluda geese.

Parameters [%]	Type of muscle	Control group C	Time of freezing storage (days)				
			30	90	180	270	365
			n=18				
C 16:1 $n-7$	BM	$3.00^a \pm 0.10$	$^y2.86^b \pm 0.13$	$^y2.78^b \pm 0.14$	$2.72^b \pm 0.16$	$2.53^c \pm 0.17$	$2.41^c \pm 0.18$
Palmitoleic acid	LM	$3.10^a \pm 0.09$	$^x3.08^a \pm 0.11$	$^x3.03^a \pm 0.10$	$2.88^b \pm 0.12$	$2.60^b \pm 0.16$	$2.57^b \pm 0.19$
C 18:1 $n-9$	BM	$41.16^a \pm 0.98$	$41.13^a \pm 0.64$	$41.10^a \pm 0.77$	$41.13^a \pm 0.93$	$^y40.09^b \pm 0.55$	$^y39.95^b \pm 0.58$
Oleic acid	LM	$41.57^a \pm 0.67$	$41.44^a \pm 0.56$	$41.43^a \pm 0.51$	$40.87^b \pm 0.69$	$^x40.73^b \pm 0.53$	$^x40.54^b \pm 0.53$
C 20:1 $n-9$	BM	0.30 ± 0.04	0.29 ± 0.04	0.29 ± 0.03	0.29 ± 0.04	0.28 ± 0.02	0.27 ± 0.03
Gondoic acid	LM	0.29 ± 0.03	0.29 ± 0.04	$0.28 \pm 0,04$	0.28 ± 0.03	0.27 ± 0.04	0.26 ± 0.04
C 24:1 $n-9$	BM	$0.93^a \pm 0.10$	$0.92^a \pm 0.11$	$0.80^b \pm 0.07$	$0.71^c \pm 0.06$	$0.62^d \pm 0.07$	$0.60^d \pm 0.09$
Nervonic acid	LM	$0.92^a \pm 0.09$	$0.89^{a,b} \pm 0.09$	$0.82^b \pm 0.08$	$0.69^c \pm 0.06$	$0.67^c \pm 0.07$	$0.58^c \pm 0.09$
Σ MUFA	BM	$^y45.39^a \pm 0.43$	$45.20^a \pm 0.51$	$^y44.97^b \pm 0.38$	$44.85^b \pm 0.31$	$^y43.52^c \pm 0.52$	$^y43.23^c \pm 0.50$
	LM	$^x45.88^a \pm 0.53$	$45.70^a \pm 0.62$	$^x45.56^a \pm 0.44$	$44.72^b \pm 0.41$	$^x44.27^c \pm 0.49$	$^x43.95^d \pm 0.37$

[a-d]Different letters in a row mean statistically significant differences between group average, including storage time ($P \le 0.05$).

[x-y]Different letters in columns mean statistically significant differences between the group average, including the type of muscle ($P \le 0.05$).

considerations are consistent with Kouba et al. (1997), which observed a loss of 40% in delta-9 desaturase activity when the enzyme activity is measured in frozen adipose tissue during frozen storage. However, there was no evidence that extending of muscle's frozen storage time affected changes in C 14:0 proportion. The consequence of the observed changes in individual SFA identified in the studied muscles was the decreasing of Σ SFA with the extension of the frozen storage time (Table 1).

The same relationship was demonstrated by Santos-Filho et al. (2005), Popova (2014), Milczarek et al. (2011, 2013) and Sabagh et al. (2016), who analyzed the changes in fatty acid content in intramuscular fat of: goat, pork, lamb and chicken, stored in the freezer for 3, 6, and 10 mo. Santos-Filho et al. (2005) reported that the storage time is the most relevant parameter that contributed to change the fatty acid profile, also in SFA. On the other hand, Ali and Zahran (2010), Chwastowska-Siwiecka et al. (2014), Alonso et al. (2016) found that by extending the frozen storage time of meat of various animals species, caused the increase on total percent of SFA in intramuscular fat.

In our study the lipids of the BM had a higher proportion of Σ SFA than LM in each storage periods. The BM contained more acids such as: C14:0 (on days 30 and 180), and C 16:0 (on days 30, 90, 180, 270, and 365), compared to LM during the same frozen storage periods (Table 1).

Based on the average percentage of SFA, also calculated the changes between their share in the control samples and the values determined at the 30th, 90th, 180th, 270th, and 365th d of frozen storage (relative percent changes). It was shown that with the extension of the frozen storage time of both types of muscles, the relative percentages of Σ SFA (calculated based on Table 1) generally increased, and the differences between the mean groups were statistically confirmed. Moreover, it was found that the LM, compared to the BM, were characterized by significant higher ($P \le 0.05$) relative percent changes in the share of acids: C 16:0, and C 18:0, and Σ SFA at 30th, 90th, and 180th d of frozen storage.

Monounsaturated Fatty Acid (MUFA)

In MUFA profile of both types of control samples dominated oleic acid C 18:1 $n-9$ (41.16% in BM and 41.57% in LM). There was no significant effect (in control group) of the type of muscle on the share of particular kinds of MUFA, but LM characterized a higher share of Σ MUFA compared to BM (Table 2).

The percentage of C 16:1 $n-7$, C 20:1 $n-9$, and C 24:1 $n-9$ were similar to data given by Biesiada-Drzazga (2006) for muscles obtained from 10-wk-old geese broilers of the same genotype. However, the same author stated that the share of the C 18:1 $n-9$ in breast and legs muscles was higher about 13% and 15%, respectively, compared to the our results. The differences could result from the addition of soybean meal and "00" rapeseed meal to the feed mixture in experiment of Biesiada-Drzazga (2006).

The analysis of changes in the percentage of MUFA in examined muscles showed, that the BM stored for 30, 90, and 180 days were characterized by a significantly higher share of C18:1 $n-9$ and Σ MUFA, compared to that stored for 270 and 365 days. On the other hand, LM were characterized by a higher proportion of C 16:1 $n-7$ (on 30th and 90th day), C 18:1 $n-9$ (on 270th and 365th day) and higher proportion of Σ MUFA (on 90th, 270th and 365th day) compared to their percentage determined at the another examined days of storage. Also, the share of the C 24:1 $n-9$ decreased with the extension of the frozen storage time, and the differences were statistically significant ($P \le 0.05$)−Table 2. The decreasing of unsaturated fatty acids (**UFA**) during the storage period was expected because they are more reactive to oxidation processes, mainly those with two or more double bonds. Also Holman et al. (2018) suggested that during frozen storage of meat comes to the processes of oxidation. Lipid oxidation is a highly complex set of free radical reactions between lipid compounds and oxygen. The description of lipid oxidation processes can be difficult to fully explain (Zamuz et al., 2022). There are many sources of oxidants. Some of them are generated from the endogenous sources and others are from the exogenous sources (Huang and Ahn, 2019). To

Table 3. The share of PUFA (polyunsaturated fatty acid) $n-3$ in BM and LM of White Koluda geese.

			Time of freezing storage (days)				
			30	90	180	270	30
Parameters [%]	Type of muscle	Control group C			n = 18		
α C 18:3 $n-3$	BM	$0.95^a \pm 0.04$	$0.90^b \pm 0.03$	$0.90^b \pm 0,02$	$0.83^c \pm 0.05$	$^y0.77^d \pm 0.05$	$^y0.73^d \pm 0.04$
α-linolenic acid	LM	$0.98^a \pm 0.04$	$0.92^b \pm 0.04$	$0.91^b \pm 0.04$	$0.85^c \pm 0.06$	$^x0.82^{cd} \pm 0.04$	$^x0.78^d \pm 0.05$
C 20:5 $n-3$	BM	$0.88^a \pm 0.04$	$0.86^{ab} \pm 0.03$	$0.83^b \pm 0.04$	$0.78^c \pm 0.03$	$0.71^d \pm 0.06$	$0.65^e \pm 0.05$
Eicosapentaenoic acid	LM	$0.92^a \pm 0.04$	$0.89^{ab} \pm 0.05$	$0.87^b \pm 0.03$	$0.80^c \pm 0.04$	$0.73^d \pm 0.05$	$0.68^e \pm 0.03$
C 22:6 $n-3$	BM	$0.28^a \pm 0.03$	$0.26^{ab} \pm 0.03$	$0.23^b \pm 0,04$	$0.17^c \pm 0.04$	$0.14^{cd} \pm 0.03$	$0.12^d \pm 0.04$
Docosahexsaenoic acid	LM	$0.26^a \pm 0.03$	$0.25^a \pm 0.03$	$0.20^b \pm 0.03$	$0.16^c \pm 0.03$	$0.13^{cd} \pm 0.04$	$0.10^d \pm 0.04$
Σ PUFA $n-3$	BM	$2.11^a \pm 0.07$	$2.02^b \pm 0.07$	$1.96^b \pm 0.07$	$^y1.78^c \pm 0.13$	$^y1.62^d \pm 0.10$	$^y1.50^e \pm 0.08$
	LM	$2.16^a \pm 0.07$	$2.06^b \pm 0.08$	$1.98^c \pm 0.14$	$^x1.81^d \pm 0.13$	$^x1.78^e \pm 0.04$	$^x1.56^f \pm 0.07$

[a-f]Different letters in a row mean statistically significant differences between group average, including storage time ($P \le 0.05$);
[x-y]Different letters in columns mean statistically significant differences between the group average, including the type of muscle ($P \le 0.05$).

the exogenous factors include, among others: oxygen contained in atmospheric air, temperature and storage time, as well as the presence of some heavy metals. Lipids contained in meat also undergo oxidative processes due to the activity of endogenous factors, that is, tissue enzymes (Wereńska et al., 2022). Moreover the oxidants or reactive oxygen species are produced from mitochondria as a part of normal metabolic process and as microbicidal products by macrophages. Under normal conditions, about 3 to 4% of the oxygen used in aerobic metabolism of mitochondria are converted to superoxide, which react with MUFA and PUFA (Huang and Ahn, 2019). The process of lipid peroxidation involves the initiation, propagation and termination phases. The interaction between triplet oxygen, light and photosensitizers results in the formation of singlet oxygen. Once activated, singlet oxygen can react with unsaturated fatty acids (MUFA and PUFA), removing a hydrogen atom from the methylene carbon adjacent to the cis double bond of the unsaturated fatty acid, resulting in the formation of free radicals. The formed radicals can attack other fatty acids as well as the products formed at the beginning of the reaction, propagating the oxidation. Once initiated, the reaction goes on in chains and ends only when the reserves of unsaturated fatty acids and oxygen are exhausted. In this way, with the exhaustion of the substrates, the propagation reactions cease and the termination begins, which have as a characteristic the formation of stable or nonreactive final products, which comprise the derivatives of the decomposition of hydroperoxides, such as alcohols, aldehydes, ketones, esters and other hydrocarbons (Ribeiro et al., 2019). Therefore, the initiated pre-freezing oxidation processes also lead to oxidation processes during frozen storage. Of course, the oxidation processes during frozen storage are slower, but they are not completely stopped.

The obtained results are consistent with the results published by Milczarek et al. (2011), who also showed a decrease in the content of MUFA in the lipids of chicken meat stored for 12 mo at $-22°C$. In turn, Popova (2014) in lamb meat stored for 3 and 6 mo at $-20°C$ and Alonso et al. (2016) in pork meat stored for 24 mo at $-22°C$ found that the share of MUFA increased along with the extension of their storage time. On the other hand, Samouris et al. (2011) found no significant effect of the frozen storage time of chicken breast muscles and pork meat on changes in MUFA.

Leg muscles had more proportion of: C 16:1 $n-7$ (on 30 and 90 d) and C 18:1 $n-9$ (on days 270 and 365) compared to the breast muscles at the same frozen storage times. The BM were characterized similar share of C 20:1 and C 24:1 compared to LM (Table 2).

Polyunsaturated Fatty Acid (PUFA)

The study also analyzed changes in the share of $n-3$ and $n-6$ PUFA, taking into account their type of muscle and storage time (Tables 3, 4). It was found that the

Table 4. The share of PUFA (polyunsaturated fatty acid) $n-6$ in BM and LM of White Koluda geese.

			Time of freezing storage (days)				
			30	90	180	270	30
Parameters [%]	Type of muscle	Control group C			n=18		
C 18:2 $n-6$	BM	$^y16.02^a \pm 0.20$	$^y16.04^a \pm 0.21$	$^y15.72^b \pm 0.25$	$^y15.45^c \pm 0.19$	$^y15.20^d \pm 0.18$	$^y15.01^d \pm 0.19$
Linoleic acid	LM	$^x16.36^a \pm 0.18$	$^x16.34^a \pm 0.19$	$^x16.10^b \pm 0.20$	$^x15.84^c \pm 0.24$	$^x15.57^d \pm 0.26$	$^x15.24^d \pm 0.25$
C 20:3 $n-6$	BM	$0.10^a \pm 0.03$	$0.07^b \pm 0.02$	$0.05^b \pm 0.03$	$^x0.04^{b,c} \pm 0.02$	$0.02^c \pm 0.02$	$0.01^c \pm 0.03$
Dihomo-α-linolenic	LM	$0.08^a \pm 0.05$	$0.07^a \pm 0.04$	$0.06^a \pm 0.03$	$^y0.02^b \pm 0.01$	$0.01^b \pm 0.01$	$0.02^b \pm 0.01$
C 20:4 $n-6$	BM	$4.65^a \pm 0.10$	$4.63^a \pm 0.09$	$4.56^a \pm 0.13$	$^y4.34^b \pm 0.12$	$4.19^c \pm 0.14$	$4.18^c \pm 0.16$
Arachidonic acid	LM	$4.69^a \pm 0.11$	$4.66^a \pm 0.11$	$4.67^a \pm 0.12$	$^x4.44^b \pm 0.10$	$4.27^c \pm 0.15$	$4.24^c \pm 0.18$
Σ PUFA $n-6$	BM	$^y20.77^a \pm 0.26$	$^y20.74^a \pm 0.31$	$^y20.33^b \pm 0.34$	$^y19.83^c \pm 0.28$	$^y19.41^d \pm 0.35$	$^y19.20^d \pm 0.22$
	LM	$^x21.13^a \pm 0.22$	$^x21.07^a \pm 0.25$	$^x20.83^b \pm 0.21$	$^x20.30^c \pm 0.27$	$^x19.85^c \pm 0.37$	$^x19.50^d \pm 0.18$

[a-d]Different letters in a row mean statistically significant differences between group average, including storage time ($P \le 0.05$).
[x-y]Different letters in columns mean statistically significant differences between the group average, including the type of muscle ($P \le 0.05$).

dominant fatty acids from both group were: eicosapentaenoic (C 20:5 $n-3$–EPA) and linoleic (C 18:2 $n-6$) acids, the proportion of which were determined in BM and LM during frozen storage, respectively: 0.88% and 0.92% (C 20:5 $n-3$) and 16.02% and 16.36% (C 18:2 $n-6$) (Tables 3, 4). The share of PUFA $n-3$ and $n-6$ were similar to the results published by Okruszek (2012). Author analyzed the fatty acid profile of geese muscles from native conservative flocks.

Considering the type of muscle, it was found that control LM, were characterized by a higher proportion of C 18:2 $n-6$ and total share of PUFA $n-6$, compared to BM (Table 4). Generally, there was no effect of the type of muscle on the share of the individual PUFA $n-3$, and PUFA $n-6$ (except C 18:2) and the Σ PUFA $n-3$ (Tables 3, 4).

The frozen storage time of muscles influenced the proportion of acids from the PUFA $n-3$ and $n-6$ groups. The percentage of: C 18:3, C 20:5, C 22:6, and C 20:4 $n-6$ in both types of muscles decreased from the 180th day, but C 18:2 n−6 and Σ $n-6$ PUFA−from the 90th day of frozen storage time (Tables 3, 4). Our results are opposite to the results presented by Santos-Filho et al. (2005) for goat meat stored at −18°C on 3 and 6 mo, Popova (2014) for lamb stored at −20°C on 3 and 6 mo and Alonso et al. (2016) for pork stored at −22°C on 24 mo. These authors also found that the share of PUFA decreased with the extension of the storage time of frozen. The authors suggested that the progressive processes of their oxidation caused the reduction in the content of PUFA. This is the same case like with monounsaturated fatty acid and their oxidation. Lipids contained in meat undergo oxidative processes due to the activity of endogenous factors, that is, tissue enzymes and microbial origin. During frozen storage of meat, hydrolysis of ester bonds between the glycerol molecule and fatty acids occurs, as well as oxidation of fatty acids. These processes take place through chemical reactions or as a result of the action of endogenous enzymes contained in muscle tissue (Ribeiro et al., 2019).

When examining the influence of the muscle type on changes in PUFA proportion, it was found that LM lipids were characterized by a higher percentage of C 18:3 $n-3$ (on days 270th and 365th), C 18:2 $n-6$ (on days 30th, 90th, 180th, 270th and 365th), C 20:4 $n-6$ (on day 180th), Σ PUFA $n-3$ (on day 180th, 270th and 365th) and Σ PUFA $n-6$ (on −−) compared to the BM (Tables 3, 4).

It was shown that the relative percentage changes (calculated based on Table 3) in the share of: C 18:3 $n-3$ and Σ PUFAs $n-3$ were generally higher ($P \leq 0.05$) in BM than in LM in 180th, 270th and 365th d of frozen storage. However, the relative percentage changes in the proportion of: C 18:2 $n-6$ Σ PUFA $n-6$ were generally higher in LM than in BM (in 180th, 270th and 365th d of frozen storage).

Lipid Profile Indicators

From the nutritional point of view, it is also important to estimate the mutual ratio of Σ PUFA $n-6$ to Σ PUFA $n-3$, the recommended ratio of which should range from 1:1 to 4:1 (Buckland et al., 2022). The Σ PUFA $n-6$ and Σ PUFA $n-3$ and their ratio (Σ PUFA $n-6/n-3$) are the principal fatty acids controlling the hypocholesterolemic index. Values of the Σ PUFA $n-6/n-3$ ratio below 4.0 in a diet indicate desirable quantities for cardiovascular risk prevention. Whereas the Σ PUFA $n-6/n-3$ ratio ranged from 5.0 to 6.0 and can be recognized as close to recommended, suggesting that these species could be categorized as beneficial to human health consumption (Fernandes et al., 2014; Wereńska et al., 2021).

Our research shows that the ratio of Σ PUFA $n-6$ to Σ PUFA $n-3$ in BM and LM increased by extending their frozen storage time. The control BM and LM samples those throughout the entire of frozen storage were far away to the recommended ratio of Σ PUFA $n-6/$Σ PUFA $n-3$. The least favorable ratio of Σ PUFA $n-6$ to Σ PUFA $n-3$ was found in both muscle types on day 365 of their storage (12.80−−in BM and 12.50−−in LM). It was related to a decrease in the share of Σ PUFA $n-3$, especially in EPA and DHA (Table 5). Although the ratio of Σ PUFA $n-6$ to Σ PUFA $n-3$ in muscle lipids stored from 30th to 365th d was higher

Table 5. The lipid indicators in BM and LM of White Koluda geese.

Parameters [%]	Type of muscle	Control group C	Time of freezing storage (days)				
			30	90	180	270	30
			n = 18				
Σ PUFA	BM	[y]22.88[a] ± 0.29	[y]22.75[a] ± 0.32	[y]22.29[b] ± 0.38	[y]21.61[c] ± 0.33	[y]21.03[d] ± 0.49	[y]20.70[e] ± 0,.29
	LM	[x]23.29[a] ± 0.25	[x]23.13[a] ± 0.26	[x]22.81[b] ± 0.47	[x]22.11[c] ± 0.38	[x]21.63[d] ± 0.37	[x]21.06[e] ± 0.29
Σ UFA	BM	68.23[a] ± 1.46	67.96[a,b] ± 1.03	67.25[b] ± 1.90	66.47[c] ± 1.29	64.56[d] ± 0.94	63.94[e] ± 0.64
	LM	69.17[a] ± 1.35	68.83[a,b] ± 1.05	68.40[b] ± 1.33	66.83[c] ± 1.15	65.90[d] ± 0.69	65.04[d] ± 0.68
Σ PUFA n−6/Σ n−3	BM	9.84[d] ± 0.35	10.31[d] ± 0.25	10.37[d] ± 0.47	11.14[c] ± 0.72	11.98[b] ± 0.65	12.80[a] ± 0.96
	LM	9.78[d] ± 0.35	10.23[cd] ± 0.47	10.52[c] ± 0.76	11.22[b] ± 0.95	11.15[b] ± 0.35	12.50[a] ± 0.60
Σ UFA/Σ SFA	BM	[y]2.21 ± 0.08	[y]2.21 ± 0.09	[y]2.20 ± 0.07	[y]2.22 ± 0.07	[y]2.18 ± 0.07	[y]2.19 ± 0.10
	LM	[x]2.30 ± 0.06	[x]2.31 ± 0.06	[x]2.31 ± 0.08	[x]2.30 ± 0.06	[x]2.30 ± 0.10	[x]2.30 ± 0.09
Σ PUFA/Σ SFA	BM	[y]0.74 ± 0.03	[y]0.74 ± 0.03	[y]0.73 ± 0.04	[y]0.72 ± 0.03	[y]0.71 ± 0.04	[y]0.71 ± 0.02
	LM	[x]0.77 ± 0.03	[x]0.78 ± 0.04	[x]0.77 ± 0.03	[x]0.76 ± 0.04	[x]0.75 ± 0.03	[x]0.74 ± 0.03

[a-e]Different letters in a row mean statistically significant differences between group average, including storage time ($P \leq 0.05$).
[x-y]Different letters in columns mean statistically significant differences between the group average, including the type of muscle ($P \leq 0.05$).

than the recommended values, it was also lower than in the so-called "Typical Western diet," in which it is about 20:1 (Husted and Bouzinova, 2016; Greupner et al., 2018).

Therefore, it can be assumed that the calculated ratio of Σ PUFA $n-6$ to ΣPUFA $n-3$ can only be treated as a reasonable compromise between the proportion considered typical for the diet of developed countries and the suggested optimum.

The type of muscle also influenced the calculated values of the Σ PUFA $n-6/\Sigma$ PUFA $n-3$ index. There were no significant differences between Σ PUFA $n-6$ to Σ PUFA $n-3$ ratio for the BM and LM in all their frozen storage time and in control sample (Table 5).

The obtained results concerning the Σ PUFA $n-6$ to Σ PUFA $n-3$ ratio are similar to the results presented by Chwastowska-Siwiecka et al. (2014) and Alonso et al. (2016) They also showed an increase in the proportion of Σ PUFA $n-6$ to Σ PUFA $n-3$ in rabbit meat stored for 3 mo at $-28°$C and pork meat stored 24 mo at $-20°$C.

The Σ UFA to Σ SFA ratio also determines the health-promoting properties of food. The Σ UFA to Σ SFA, and Σ PUFA $n-6$ to Σ PUFA $n-3$ ratios are parameters used to judge the meat nutritional value and the healthiness of meat fat for human consumption (Wereńska et al., 2021). The Σ UFA/Σ SFA assumed that the higher it is, the more beneficial it is for the consumer's health (Attia et al., 2017).

Based on the conducted studies, it was shown that the ratio of Σ UFA to Σ SFA in BM and LM was similar throughout the entire period of their frozen storage, and the differences were not statistically confirmed (Table 5). It was also shown that muscle type influenced the Σ UFA to Σ SFA ratio. Leg muscles were characterized by a higher Σ UFA to Σ SFA ratio ($P \leq 0.05$), compared to the breast muscle, and therefore LM are more beneficial from the nutritional point of view (Table 5).

The parameters used to judge the meat nutritional value and the healthiness of meat fat for human consumption is also Σ PUFA/Σ SFA. A Σ PUFA/Σ SFA above 0.45 is recommended in the human diet to prevent the development of cardiovascular disease and some other diseases, including cancer. Foods with Σ PUFA/Σ SFA below 0.45 have been considered undesirable for the human diet, because of their potential to induce a cholesterol increase in the blood (Mapiye et al., 2011).

Based on the conducted studies, it was shown that Σ PUFA to Σ SFA ratio in BM and LM was similar throughout the entire period of their frozen storage, and the differences were not significant (Table 5). Our research also shows that Σ PUFA to Σ SFA ratio in BM and LM was similar during their frozen storage time and were in range from 0.71 to 0.77 (Table 5). Moreover, the Σ PUFA to Σ SFA ratio was similar in raw meat and in frozen storage muscles. It means that frozen storage didn't affected on this index and the breast and leg muscles have the same quality in the dietary point of view. Lower (0.07−0.17) Σ PUFA to Σ SFA ratio was stated by Santos-Filho et al. (2005) in goat meat during frozen stored on 6 months (at $-18°$C). In turn, Popova (2014) and Alonso et al. (2016) found that the Σ PUFA to Σ SFA ratio in the lamb and pork meat stored for 6 and 24 months at $-20°$C and $-22°$C were in the range of 0.32 to 0.42.

CONCLUSION

The total share of SFA, MUFA, and PUFA in BM and LM did not change significantly until the 90th day of frozen storage. This period was the most optimal for storing goose meat at a temperature of $-20°$C and, at the same time, guarantees that it maintains the proportions of SFA, MUFA, and PUFA similar to those specified in the control samples. The content of PUFA $n-3$ (C 18:3, C 20:5, C 22:6) and C 20:4 $n-6$ acid in BM and LM decreased significantly from day 180, and C 18:2 $n-6$ and Σ PUFA $n-6$ acids−from 90th day of frozen storage. The least favorable ratio of Σ PUFA $n-6$ to Σ PUFA $n-3$ was found in the lipids of both tested types of muscles on day 365 of their storage, which was related to a decrease in the content of Σ PUFA $n-3$, especially EPA and DHA. The changes in muscles during their frozen storage occurred with different intensities depending on their type. Therefore, it is difficult to indicate which of them were characterized by more favorable values of the analyzed parameters, and thus in which of the types of muscles the changes determining the reduction of their quality proceeded faster. Based on lipid profile indicator it can be concluded that the ratios of Σ PUFA $n-6$ to Σ PUFA $n-3$ in muscle lipids were in range 9.78 to 12.80, and were higher than the recommended values, but still were lower than in the so-called "Typical Western diet," in which it is about 20:1. The Σ PUFA to Σ SFA ratios were similar in raw breast and leg muscles and in frozen storage samples, and it means that frozen storage didn't affected on deterioration on their quality in the dietary point of view.

DISCLOSURES

The authors declare no conflicts of interest in publication

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