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Research Paper / Araştırma Makalesi

Effect of Heat Treatment on Storage Stability of Sheep Tail Fat

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ABSTRACT

In this study, the effects of various physical, chemical, and technological properties of sheep tail fat (fresh and ghee) as well as different storage durations, storage temperatures, and the use of additives on the storage stability of thermally processed sheep tail fat (ghee tail fat) were investigated. To prevent lipid oxidation and other degradation factors during use after long-term storage, salt and antioxidant additives were used. Samples were stored in glass jars at 25ºC, +4ºC, and -18ºC for 90 days. The moisture, protein, fat, and ash contents of fresh tail fat was determined as 11.2%, 3.47%, 85.0%, and 0.07%, respectively. In clarified fat, the moisture content was determined as 0.4%, protein content as 2.98%, fat content as 96.0%, and ash content as 0.05%. Additionally, the cholesterol content in tail fat was 60 mg/100 g in fresh tail fat and 58 mg/100 g in ghee tail fat. The pH values of fats were determined as 5.7 in fresh samples and 6.2 in ghee samples. The fatty acid compositions of samples revealed that the dominant fatty acid groups in both fresh and ghee sheep tail fat were palmitic, stearic, and oleic acids. Fresh tail fat stored at room temperature deteriorated in terms of chemical properties (FFA, peroxide, and TBA) within 7 days, while those stored at +4ºC deteriorated within 15 days. However, thermally processed samples were preserved without deterioration for 60 days at these storage temperatures.

Keywords: Sheep tail fat, Tallow, Animal fat, Storage stability

Isıl İşlemin Koyun Kuyruk Yağının Depolama Stabilitesi Üzerine Etkisi

ÖZ

Bu çalışmada, ısıl işlem görmüş koyun kuyruk yağının (sade) depolama stabilitesini belirlemek amacıyla, kuyruk yağının (taze ve sade) çeşitli fiziksel, kimyasal ve teknolojik özellikleri ile farklı depolama sürelerinin, depolama sıcaklıklarının ve katkı maddesi kullanımının bu özellikler üzerindeki etkileri araştırılmıştır. Uzun süreli depolamadan sonra, kullanım sırasında lipit oksidasyonu ve diğer bozulma faktörlerini önlemek için tuz ve antioksidan katkı maddesi kullanılmıştır. Örnekler cam kavanozlarda 25ºC, +4ºC ve -18ºC'de 90 gün boyunca depolanmıştır. Taze kuyruk yağında nem, protein, yağ ve kül içeriği sırasıyla %11.2, 3.47, 85.0 ve 0.07 olarak belirlenmiştir. Berraklaştırılmış yağda nem, protein, yağ ve kül oranları sırasıyla %0.4, 2.98, 96.0 ve 0.05 olarak belirlenmiştir. Ayrıca, kuyruk yağı kolesterol içeriği taze ve sade kuyruk yağlarında sırasıyla 60 ve 58 mg/100g olarak belirlenmiştir. Yağların pH değerleri taze örneklerde 5.7, sade örneklerinde ise 6.2 olarak belirlenmiştir. Örneklerin yağ asidi kompozisyonları incelenmiş ve taze ve sade koyun kuyruk yağında baskın yağ asidi gruplarının palmitik, stearik ve oleik asit olduğu belirlenmiştir. Oda sıcaklığında depolanan taze kuyruk yağları kimyasal özellikler (FFA, peroksit ve TBA) açısından 7 gün içinde bozulurken, +4ºC'de depolananlar ise 15 gün içinde bozuldu. Ancak ısıl işlem görmüş örnekler bu depolama sıcaklıklarında 60 gün boyunca bozulmadan muhafaza edilebilmiştir.

Anahtar Kelimeler: Koyun kuyruk yağı, İç yağ, Hayvansal yağ, Depolama stabilitesi

INTRODUCTION

One of the products obtained from the slaughter of cattle and tailed sheep is tallow and tail fat. Sheep tail fat is the main source of flavoring, especially in the production of kebabs and some dishes. Animal fats are the second most important source of lipid raw materials produced in Turkey [1]. Fat-tailed sheep store fat in their tails for use when natural food sources are scarce [2, 3]. Sheep tail fat is used in kitchens in Turkey and Central Asian countries. In China, sheep tail fat functions as a food seasoning in cooking [4]. However, the use of sheep tail fat in the food industry is limited due to its not widely accepted flavor [5]. Sheep tail fat constitutes a large part of Turkey's animal fat production. It has been reported that as of 2022, there are approximately 44,700,000 sheep in Turkey, and most of them have fat-tails [6, 7]. The weight of sheep tail fat from individual sheep ranges from 3 to 8 kg [5]. It has also been reported that 21,500,000 sheep are slaughtered annually [6], according to a rough calculation, it can be calculated that 100,000 tons of tail tissue can be used as a source of lipid production.

The high ratio of unsaturated fatty acids in their structure causes lipids to be more susceptible to deterioration. Tail fat among animal fats is considered less risky in terms of health compared to tallow fats, since the content of unsaturated fatty acids is higher than other tallow fats [1, 8]. Animal fat can stabilize the three-dimensional network of dissolved myofibrillar proteins that increase the mouthfeel and tenderness of processed meat products. In addition, animal fat increases juiciness and yield by reducing water losses during cooking or ripening [9].

Fats with an average melting point of up to 45ºC are absorbed by the body at a rate of 95% or more. From this point of view, tail fat, which has a melting point below 45ºC, can be digested more easily by the body. Approximately 94% of the tail fat is lipid [10]. In this respect, tail fat can be considered as an important source of fat. The main fatty acids in tail fat are oleic acid (28.37-44.43%), palmitic acid (24.77-31.49%), stearic acid (16.51-30.02%), but also include different fatty acids such as margaric (4.32%), myristic (3.67-3.92%), palmitoleic (3.01-3.14%), linoleic (0.66-2.77%) and lauric acids (0.20%) [7, 11].

In addition to tail fat use as a natural lipid in dishes by melting in Turkey, it is widely used in products such as kebabs, döner, lahmacun, especially in terms of softening meat and providing flavor. Although it is thought to have a high value in terms of cholesterol, it is emphasized that it can prevent joint pain in later ages, so it should be consumed at young ages. However, since it contains omega-6 and omega-3 fatty acids, it is thought to reduce the risk of cardiovascular diseases in people while consumed at a young age, but it is recommended to be consumed occasionally in older ages [12].

Although tail fat, which is used both fresh and as edible fat after long-term storage in Turkey and has a wide range of uses in the food industry, is so important for our country, the number of studies in the literature is very limited. Especially after long-term storage, consumers may experience various problems in quality properties as a result of oxidation of lipids during use. For this reason, determining the oxidation degree and other structural changes of tail fats stored under different conditions and times are important factors in terms of food quality and technology. In this study, the effects of storage time, temperature, salt and antioxidant use on the storage stability of heat-treated tail fat (ghee tail fat) were examined, and some physical and chemical properties of tail fat and storage quality were determined and the data obtained will guide other researchers and the food industry.

MATERIALS and METHODS

Preparation and Storage of Tail Fat Samples

The tail fat of the sheep (Karayaka breed) used in the study was purchased fresh from a private slaughterhouse in Samsun domestic market immediately after slaughtering and then passed through a meat grinder (+4ºC) with a mirror size of 3 mm. A total of 10 sheep were used in the study, with approximately 2 kg of tail fat obtained from each animal. For fresh tail fat samples, 100 g of tail fat taken from the meat grinder was filled into glass bottles with a lid, which were sterilized by keeping them in an oven the day before. For the heat-treated tail fat (ghee tail fat) sample, 30 minutes of heat treatment was applied to the tail fat with an initial temperature of 14ºC, and when the temperature reached 100ºC, heat treatment was continued for another 5 minutes. Then, the tail fat was passed through a muslin cloth, and the cartilage tissue was separated, and the remaining fat part was filled in the same way at about 80 ºC in glass bottles with a lid. In this way, fat was obtained with 62% yield from the tail tissue used in the research. For the heat-treated salt added (salted tail fat) sample type, 2% (17.5 g) salt (sodium chloride, ≥99.8% Merck, USA) was added to 3500 g tail fat, melted in a saucepan and heat treated at 100ºC for 5 minutes. Then, it was passed through a muslin cloth and filled at approximately 80ºC at 75 g per jar. For the sample with heat-treated additive (additived tail fat), 2000 g of weighed 0.2% (2 g) antioxidant was added from the tail fat melted and filtered in the same way as the other samples, mixed and then filled into jars at about 80ºC. Butylhydroxyanisole (BHA) was preferred in the study because it is a widely used antioxidant. Each prepared sample was stored at room temperature (25ºC), +4ºC and -18ºC for 90 days.

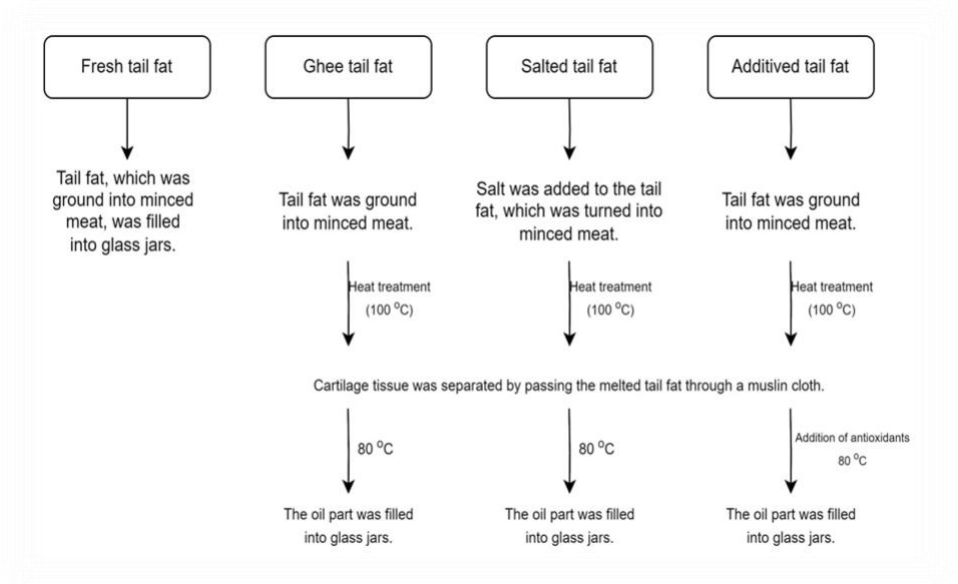

Figure 1. Preparation of tail fat samples

Physicochemical Analysis

Chemical compositions of the samples were determined according to the methods described by AOAC [13], moisture (925.10), crude protein (976.05), ash (923.03) and oil content (920.39). After the samples were diluted by 10%, readings were made using a pH-meter (Starter 2100, Ohaus, USA) and pH values were determined. Water activity was carried out at 25ºC using a measuring device (Aqualab Dew Point Water Activity Meter, USA).

Cholesterol Analysis

Cholesterol analysis was performed using gas chromatography (Varian, 3400, Varian Inc., Netherlands) [14]. 5-a-Cholestane was used as internal standard solution (IS). Cholesterol (CH) standard (2 mg/mL hexane) was prepared into vials. Cholesterol content was calculated with the help of the following Equation (1):

$$\text{Cholesterol content (mg/100g)} = (\text{mg IS} \times \text{area CH}) / (\text{area IS} \times \text{100g sample}) \quad (1)$$

Color Analysis

Color analysis was performed using the Colorflex EZ (Hunter Associates Laboratory Inc., USA) measuring device. In the samples, the International Illumination Commission L*, a*, b* values were measured. Prior to undertaking colour readings, the instrument was calibrated using a white standard plate. For each sample, replicate measurements were taken at randomly selected locations across the surface, and the average value was calculated [15].

Free Fatty Acidity Analysis

Gençcelep [16] method was modified and FFA analysis was performed. After weighing approximately 5 g of the samples taken homogeneously into a 250 mL flask, 50 mL of diethylether:ethanol (1:1,v/v) was added and shaken for 1 minute. 3-4 drops of phenolphthalein indicator were dropped into it and titration was performed with 0.1 N NaOH. The amount of NaOH used was recorded and the % free fatty acidity of the samples was calculated with the following equation (2):

$$\text{FFA (\% in oleic acid)} = (V/m) \times 2.82 \quad (2)$$

where V is the amount of NaOH used (mL), and m is the sample weight (g).

Peroxide Value Analysis

Approximately 2 g of the samples were weighed into flask and 10 mL of chloroform was added to it and then shaken. 15 mL of acetic acid and 1 mL of saturated potassium iodide were added, respectively, then the lid was closed and shaken for 1 min. 75 mL of distilled water and 1 mL of 1% soluble starch solution were added to the bottles, which were kept in a dark place for about 10 minutes, and titration was carried out with 0.01 N sodium thiosulfate solution. The used amount was recorded, and the result was determined as milliequivalents of O_2/kg oil with the help of the following equation (3) [17]:

$$\text{Peroxide value (milliequivalents } O_2\text{/kg oil)} = [\,(a-b) \times N \times 1000] / m \quad (3)$$

where a is the amount of thiosulfate used for the sample in the titration (mL), b is the amount of thiosulfate consumed for the blank in the titration (mL), N is the normality of the thiosulfate, and m is the sample amount (g).

Thiobarbituric Acid Reactive Substance (TBA) Analysis

Approximately 10 g of sample was weighed for analysis. 25 mL of 20% trichloroacetic acid and 20 mL of distilled water were added and homogenized with Ultra Turrax for about 2 minutes. The homogenized sample was passed through Whatman No: 1 filter paper. 5 mL of the obtained filtrate was taken and transferred to tubes. After adding 5 mL of 0.02 M TBA solution, the lids were closed and shaken. The tubes, which were kept in a boiling water bath at 93ºC for about 30 minutes, were cooled in tap water for 10 minutes after being removed. Then, it was transferred to spectrophotometer (T80+, PG Instruments Limited, UK) cuvettes and the absorbance value was read at 532 nm wavelength. TBA value was determined as mg malonaldehyde/kg sample by multiplying the recorded value with the coefficient of 7.8 [18].

FTIR Spectroscopy Analysis

The FTIR spectra of the samples were determined with a (Spectrum Two, Perkin Elmer Inc., Netherlands) device at a wavelength of 4000-450 cm^{-1}. For the measurement, the fat samples were ground and transferred to the instrument plate in solid form.

Differential Scanning Calorimetry Analysis

DSC analysis was performed using the DSC (DSC8000, Perkin Elmer Inc., Netherlands) instrument. An empty aluminum DSC container was used as a reference sample. In the study, melting and crystallization processes were carried out at a speed of 10ºC/minute and between 0-100ºC. The melting and crystallization temperatures of the samples were calculated using the instrument software.

Fatty Acids Composition Analysis

For the analysis of fatty acids composition of fresh and ghee tail fat samples, the samples were derivatized with 1.5 M methanolic HCl and taken into vials and then analyzed in gas chromatography device (Trace 1310, Thermo Scientific, USA). In the analysis, detector temperature and injection temperature are 240°C, detector type is FID (Flame Ionization Detector), helium carrier gas at 1.0 ml/min, oven temperature at 120°C for 0 min, at 210°C for 10 min, 250°C for 1 min parameters were used.

Statistical Analysis

Trials created within the scope of the study were set up in two repetitions according to a completely random trial plan. The obtained data were subjected to ANOVA analysis via SPSS Statistics program (V21, SPSS Inc., Chicago, USA) and the statistical significance limits of the differences were determined and compared by applying Duncan multiple comparison test ($p>0.05$).

RESULTS and DISCUSSION

Compositional Propeties of Fresh and Ghee Tail Fats

The results of moisture, fat, protein, ash and cholesterol values in fresh and heat-treated tail fat (ghee tail fat) are given in Table 1. Composition analysis was not performed in the samples of ghee salted tail fat and ghee additived tail fat obtained by adding salt and antioxidant, because it was predicted that the salt and additive used would not have much effect on the composition.

The moisture content of fresh tail fat is higher than that of ghee tail fat. In the process of obtaining ghee oil as a result of heat treatment, water is separated from the fatty tissue and a tissue with a very high oil content is left. The difference in moisture content between the obtained fats are very important for both technological and storage quality. As a matter of fact, the moisture content of adipose tissues affects the efficiency, and in this case, the yield value of an adipose tissue with a low moisture content is higher than an adipose tissue with a high moisture content. In addition, the hydrolytic degradation potential of water shortens the storage life in adipose tissue with high moisture content [8].

A portion of the protein (3.47%) found in fresh tail fat remained in the cartilage part as a result of melting, and therefore the amount was lower in ghee tail fat (2.98%). There was no statistically significant difference between the amounts of ash and cholesterol in fresh tail fat and ghee tail fat. However, due to the removal of water by the effect of temperature, very small decreases occurred in their amounts. Cholesterol analysis results in fresh and ghee tail fats were determined as 61.10 mg/100 g and 59.02 mg/100 g, respectively.

Table 1. Composition analysis results of fresh and ghee tail fat

	Moisture (%)	Crude fat (%)	Crude protein (%)	Ash (%)	Cholesterol (mg/100 g)
Fresh tail fat	11.2±0.3^{a}	85.0±0.8^{b}	3.47±0.3^{a}	0.07±0.02^{a}	61.10±0.02^{a}
Ghee tail fat	0.4±0.02^{b}	96.0±0.6^{a}	2.98±0.2^{b}	0.05±0.02^{a}	59.02±0.02^{a}

$^{a-b}$Differences between means with different letters in the same column are significant. Results are mean value ± standard deviation.

Physical and Physico-chemical Properties of Fresh and Ghee Tail Fats

The pH, a_w and color values (L*, a* and b*) results of fresh tail fat, ghee tail fat, salted tail fat and additived tail fat are given in Table 2. When Table 2 was examined, the pH value of the fresh tail fat sample was 5.69 lower than the other samples. It is thought that the increase in pH value of 6.17 in ghee tail fats is caused by the loss of buffering properties of the proteins in the composition with the application of heat treatment. When ghee tail fat, salted tail fat and antioxidant added tail fat were

compared among themselves, no statistical difference was found in pH value (p>0.05). However, with the dissolution of the added salt, some pH change was observed in the salted sample due to the Na and Cl elements, but it did not show a statistical difference. It is known that the pH values of animal fats are generally in the range of 6-7. While the water activity value of the fresh tail fat group samples was determined as 0.988, this value was determined as 0.830 in the ghee tail fats with the application of heat treatment. In the salt and antioxidant added samples, the water activity value was determined as 0.728 and 0.780, respectively. It is thought that the heat treatment application in our research may have reduced the water activity value by affecting the amount of water in the tail fat, and it may have had a lowering effect on the water activity due to the water binding property of the salt. It has been determined that BHA, which is used as an antioxidant in the research, also has a water activity-lowering effect. It is known that low water activity has a limiting effect on deterioration such as enzymatic reactions, oxidation and growth of microorganisms. It is thought that the difference in water activity, which is one of the important parameters in the processing and preservation of foods, may be one of the important parameters affecting the storage stability of tail fat.

Table 2. pH, a_w and color analysis results of tail fats with different application processes before storage

	pH	a_w	L*	a*	b*
Fresh tail fat	5.69±0.1^{b}	0.988±0.1^{a}	74.48±0.02^{a}	1.38±0.02^{a}	6.15±0.2^{a}
Ghee tail fat	6.17±0.1^{a}	0.830±0.1^{b}	74.20±0.03^{a}	0.20±0.01^{b}	5.40±0.2^{a}
Salted tail fat	6.26±0.1^{a}	0.728±0.1^{c}	75.45±0.02^{a}	0.05±0.01^{b}	5.20±0.2^{a}
Tail fat with additives	6.19±0.1^{a}	0.780±0.1^{c}	74.55±0.02^{a}	0.62±0.02^{b}	5.83±0.2^{a}

$^{a-c}$Differences between means with different letters in the same column are significant. Results are mean value ± standard deviation. a_w: water activity

The L* values of fresh tail fat, ghee tail fat, salted tail fat, antioxidant added tail fat were determined as 74.48, 74.20, 75.45 and 74.55, respectively, and the difference between them was not statistically significant (p>0.05). However, when the L* gloss values of the samples were examined, it was determined that the gloss values of the samples studied with the addition of salt slightly increased with the effect of salt. While the a* value of the fresh tail fat sample was 1.38, the a* value of ghee tail fat was 0.20, the a* value of salted tail fat was 0.05, and the a* value of additived tail fat was 0.62. It is thought that the heat treatment application that we used to obtain ghee tail fat reduces this value by affecting the a* value. Considering the b* values, the highest value was observed in fresh tail fat samples as 6.15, this value was determined as 5.40 in ghee tail fat, 5.20 in salted tail fat and 5.83 in additived tail fat.

Fatty Acid Composition of Fresh and Ghee Tail Fats

The fatty acid compositions of fresh tail fat and ghee tail fat samples are shown in Table 3. It was determined that the heat treatment application did not have a significant (p>0.05) effect on the fatty acid composition. The difference may be due to the proportional distribution of fatty acids in the ghee tail fat sample with water removal. Three main fatty acids that dominate both samples were determined. Oleic acid, one of the unsaturated fatty acids, was found to be 20.5% and 21.26% in fresh tail fat and ghee tail fat samples, respectively. In saturated fatty acids, palmitic acid was determined as 21.09% in fresh tail fat and 22.02% in ghee tail fat, while stearic acid was determined as 20.61% in fresh tail fat and 20.82% in ghee tail fat. In addition to these, less myristic acid, heptadeconoic acid, linoleic acid, pentadeconoic acid, α-linolenic acid, myristoleic acid were detected. Except for these, all the remaining fatty acids remained below 1%.

When the palmitic acid contents of the samples were evaluated, although there was not statistically significant (p>0.05) difference between them, the palmitic acid content of ghee tail fat (22.02%) was found to be higher than that of fresh tail fat (21.09%). The palmitic acid content of the samples detected in the study is similar to the literature. Mehran and Filsoof [19], in their study, reported that the palmitic acid ratio in the tail fat of Iranian domestic sheep varies between 18.2% and 23.6%. Yılmaz [8] found the palmitic acid content of the tail tissue to be 19.79% on average. Tüfekci et al. [20] in their study on the tail fat of sheep from different breeds bred in the Black Sea Region, they determined the palmitic acid contents of the tail fats of Artlı, Çepni, Karayaka and of sheep as 21.81%, 25.28%, 25.47% and 26.28%, respectively. When the stearic acid contents of the samples were examined, it was found that the stearic acid content of the fresh tail fat sample was 20.61%, and the stearic acid content of the ghee tail fat was 20.82%. These values are partly similar to the literature, partly differing. Mehran and Filsoof [19] reported that the rate of stearic acid in the tail fat of Iranian domestic sheep varies between 7.1 and 22.1%. In the study, the amount of oleic acid in fresh and ghee tail fat samples was determined as 20.50% and 21.26%, respectively. Mehran and Filsoof [19] determined this value as 39.6-53.5% in their study on tail fat of Iranian domestic sheep. Yılmaz [8] determined the amount of oleic acid belonging to sheep tail fat tissues as 35.65%, Ünsal and Aktaş [7] as 28.37%. Total saturated fatty acids were 60.35% in fresh tail fat and 61.97% in ghee tail fat (p>0.05). Total unsaturated fatty acids were 39.65% and 38.03%, respectively (p>0.05). Since the amount of saturated fatty acids is higher, the tail fats are solid at ambient temperature.

Table 3. Fatty acid composition of fresh and ghee tail fat

Fatty acids	% Fatty acids	
	Fresh tail fat	Ghee tail fat
C14:0	6.94±0.23[a]	7.20±1.04[a]
C14:1	1.33±0.05[a]	1.38±0.20[a]
C15:0	6.77±1.16[a]	5.54±0.99[a]
C16:0	21.09±0.61[a]	22.02±3.15[a]
C17:0	6.42±0.21[a]	6.88±0.77[a]
C17:1	4.68±0.11[a]	4.81±0.58[a]
C18:0	20.61±0.65[a]	20.82±0.04[a]
C18:1n9	20.50±0.96[a]	21.26±1.51[a]
C18:2n6	6.64±0.12[a]	7.01±0.75[a]
C18:3n3	2.09±0.07[a]	2.15±0.28[a]
Total Saturated Fatty Acid	60.35±1.78	61.97±5.74
Total Unsaturated Fatty Acid	39.65±1.78	38.03±5.74
Mono Unsaturated Fatty Acids	26.51±0.00	27.45±0.00
Poly Unsaturated Fatty Acids	8.73±0.00	9.16±0.00
PUFA/SFA	0.15±0.00	0.15±0.00
SFA/UFA	1.52±0.00	1.63±0.00

[a-e] Differences between means with different letters in the same column are significant. Results are mean value ± standard deviation.

Although the composition of fatty acids obtained in the study of fat samples is similar to the information in the literature, it has been determined that there are partial differences. These differences may be caused by factors such as sheep type, age, nutritional status. As a matter of fact, the fact that tail fat contains essential fatty acids in its structure can be considered among the features that make tail fat important.

Calorimetric Properties of Fresh and Ghee Tail Fats

DSC is the most widely used thermo-analytical technique to study oils and fats and is a widely accepted method for determining the crystallization and melting properties of fats [21]. DSC thermograms of fresh and ghee tail fat samples are given in Figures 2 and 3. When the DSC thermogram of the fresh tail fat sample was examined (Figure 2), one crystallization peak at 14.77°C and three melting peaks at 10.26°C, 23.34°C and 34.62°C were obtained. One crystallization peak at 19.82°C and three melting peaks at 11.19°C, 26.12°C and 36.88°C were detected in the DSC thermogram of ghee tail fat (Figure 3). Yılmaz and Karakaya [21] conducted thermal analysis of lipids isolated from various tissues of sheep fat and obtained two similar crystallization peaks (31.25-24.69°C and 7.44-3.90°C) and melting peaks (15.36-13.44°C and 45.98-44.60°C) in the DSC thermograms of tallow and intestinal fat. When compared with this information examined in the literature, the results found in the research differ. As a result, although the thermograms of fresh tail fat and ghee tail fat are similar to each other, the differences may be due to small differences in fatty acid composition.

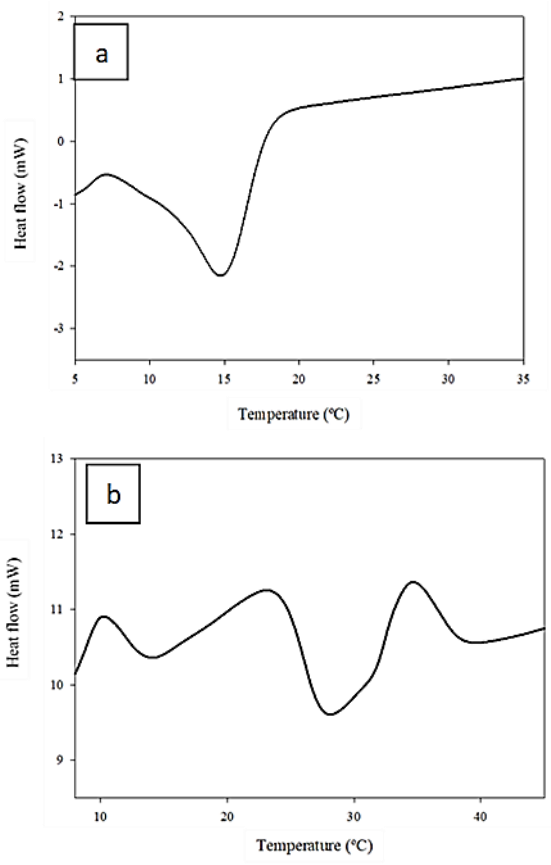

Figure 2. Crystallization (a) and melting (b) DSC thermogram of fresh tail fat sample

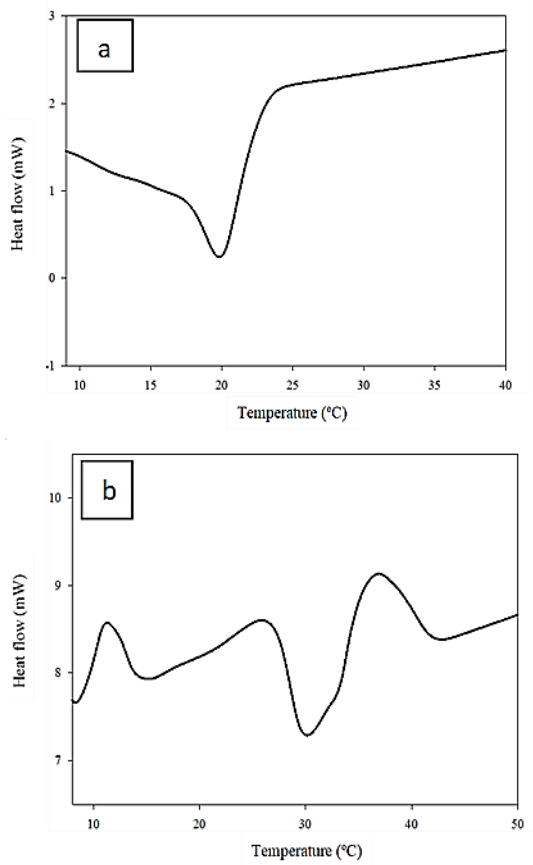

Figure 3. Crystallization (a) and melting (b) DSC thermogram of ghee tail fat sample.

FTIR Spectral Properties of Fresh and Ghee Tail Fats

FTIR spectra of fresh tail fat and ghee tail fat is given in Figures 4 and 5, respectively. 4000-3050 cm^{-1} absorption frequency determines the O-H bond type and the state of water, alcohol, hydroperoxides [22]. When the spectra of fresh tail fat (Figure 4.) and ghee tail fat (Figure 5.) were examined, a difference was found in the frequency range of 4000-3050 cm^{-1}, which is thought to be caused by the decrease in the amount of water in the ghee tail fat samples with the application of heat treatment. It is thought that the absorption frequency of 3025-2850 cm^{-1}, which gives information about C-H stretching, methylene and methylene groups at the end of the fatty acid chain [22], is parallel to the fact that the fatty acid compositions of fresh and ghee tail fat samples are similar. 1870-1550 cm^{-1} absorption frequency C=O stretch gives information about the ester bond between fatty acid and glycerol [22]. When the absorption frequency of 1870-1550 cm^{-1} is examined in both spectra, it is seen that the deterioration in the triglyceride structure is more in the fresh tail fat sample. It has been determined that the FTIR spectrum of sheep tail fat obtained in a study is similar to the spectrum obtained in our study [23].

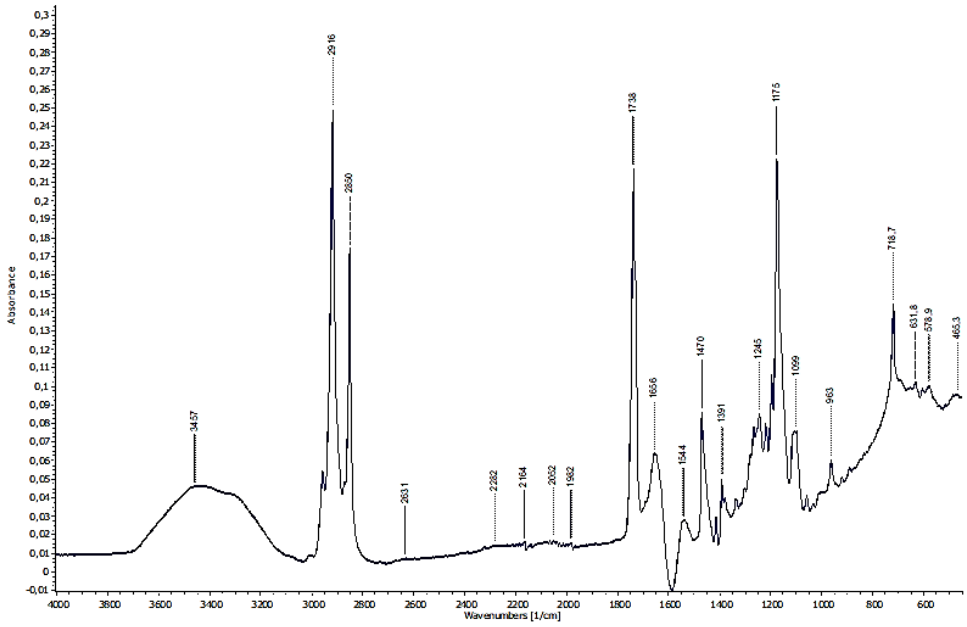

Figure 4. FTIR spectrum of fresh tail fat sample

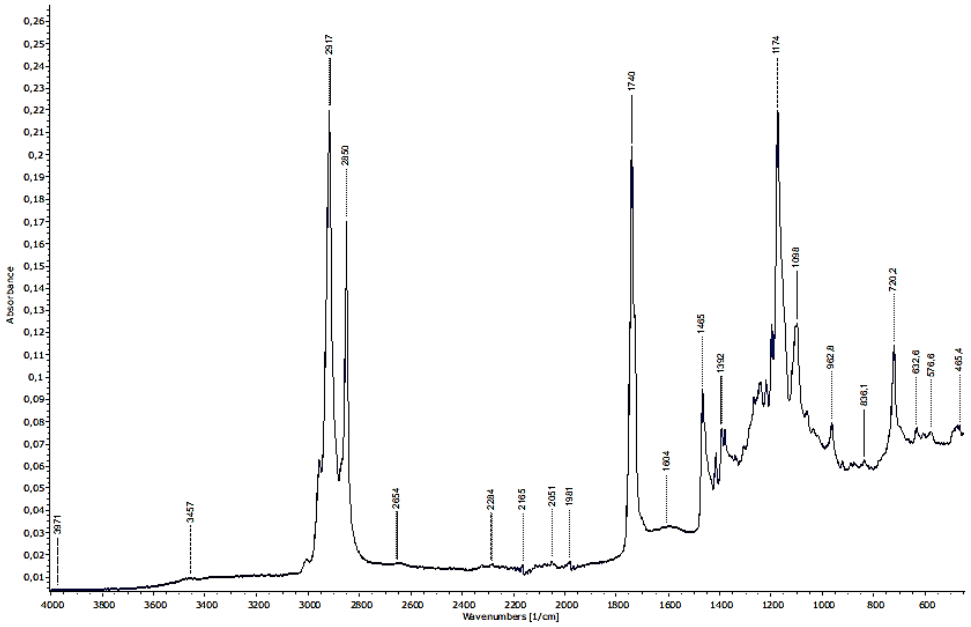

Figure 5. FTIR spectrum of ghee tail fat sample

Results of Analyses in Tail Fats during Storage

Hydrolysis and oxidation are the most important spoilage factors in storage. In this direction, free fatty acidity (FFA) analysis was performed to determine the extent of hydrolysis. In order to determine the degree of oxidation, the primary products of oxidation were determined by the peroxide number, and TBA analysis was performed to determine the secondary products formed in the advanced stages of oxidation. Samples with different applications (fresh tail fat, ghee tail fat, salted tail fat, additived tail fat); The analyzes were made on the 0th, 15th, 30th, 60th and 90th days stored at room temperature (25ºC), +4ºC and -18ºC. In addition, fresh tail fat stored at room temperature was analyzed on the 7th day. According to the results of the 7th day analysis and due to bad odor and deterioration, fresh tail fat stored at room temperature was excluded from the analysis plan.

Free fatty acids, which are not bound to triglyceride structure in the structure of oils, are an important quality parameter for oils. The change in free fatty acidity during storage gives information about the rancidity of the product, and by detecting the change in free fatty acidity, how far the hydrolysis mechanism has progressed can be determined. The FFA values determined during the storage of tail fats with different application processes are given in Table 4. The highest free fatty acidity value was found as 3.28% in the fresh tail fat sample, which was not processed after being minced, while this value was determined as 0.47%, 0.45% and 0.42% in the ghee tail fat, salted tail fat and additived tail fat, respectively. The difference between fresh and ghee tail fat was significant ($p<0.05$). Free fatty acidity values determined in ghee tail fat, salted tail fat and additived tail fat were between the same values on average and were not statistically different ($p>0.05$). A lower percentage of FFA was determined in ghee tail fats compared to fresh tail fat samples. It may be due to the fact that hydrolytic degradation is largely controlled by removing water with the application of heat treatment. FFA values determined at 25, 4 and -18ºC were determined as 1.43, 1.14 and 0.53%, respectively, and it was observed that different temperature values had a significant ($p<0.05$) effect on free fatty acidity. As the temperature value is lowered, the decrease in % FFA value can be said to slow down the activity of the lipase enzyme by limiting the activity of the enzymes in the low temperature application. The effect of storage time on the formation of free fatty acidity is important. The highest % FFA value (2.28) was found in the analysis results performed on the 7th day for the samples stored at 25ºC. A decrease in FFA value occurred after the 15th day and this is expected, as the free fatty acidity formed until the 15th day was broken down into further degradation products as the storage progressed [8, 12]. After being minced, fresh tail fat stored at 25ºC without any treatment showed a significant increase in free fatty acidity until the 15th day, and it was also determined that it could not be used as food due to the bad smell and spoilage. In addition, it is seen that the heat treatment application has a limiting effect on the formation of free fatty acidity.

Table 4. FFA (%), peroxide (meqg O_2/kg) and TBA (mg MDA/kg) values of tail fats stored in different treatments, temperatures and times

Parameters	N	FFA	Peroxide	TBA
Treatment				
Fresh tail fat	50	3.28±3.59^{a}	4.75±5.71^{a}	0.93±0.89^{a}
Ghee tail fat	64	0.47±0.07^{b}	2.67±1.37^{b}	0.72±0.39^{b}
Salted tail fat	64	0.45±0.10^{b}	2.45±1.23^{c}	0.81±0.38^{b}
Additived tail fat	64	0.42±0.09^{b}	2.28±1.11^{d}	0.76±0.36^{b}
Storage temperature (^{o}C)				
4	84	1.14±1.58^{b}	3.73±4.46^{a}	0.94±0.70^{a}
25	84	1.43±2.96^{a}	2.97±2.10^{b}	0.89±0.40^{a}
-18	84	0.53±0.25^{c}	2.11±1.06^{c}	0.55±0.32^{b}
Storage (day)				
0	48	0.49±0.10^{d}	1.03±0.12^{f}	0.31±0.08^{e}
7	16	2.28±3.24^{a}	5.45±2.81^{a}	0.83±0.19^{cd}
15	48	1.54±3.41^{b}	2.44±0.87^{e}	0.90±0.46^{bc}
30	48	0.88±1.11^{c}	4.58±5.74^{b}	0.97±0.60^{ab}
60	48	0.94±1.36^{c}	3.14±1.33^{c}	1.06±0.69^{a}
90	48	0.92±1.37^{c}	2.85±1.06^{d}	0.76±0.31^{d}

$^{a-f}$ Differences between means with different letters in the same column are significant. Results are mean value ± standard deviation. N: Number of samples, FFA: Free fatty acidity, TBA: Thiobarbituric acid reactive substance

The peroxide value, which is an indicator of the progress in lipid oxidation, is obtained by measuring the peroxide (R-OOH) value, which is one of the main reaction products formed in the first stage of oxidation [12]. Table 4. shows the peroxide values determined during the storage of tail fats with different application processes. Peroxide values of fresh tail fat, ghee tail fat, salted tail fat and additived tail fat samples were determined as 4.75, 2.67, 2.45, 2.28 in meqg O_2/kg, respectively, and the differences between them were found to be statistically significant ($p<0.05$). It is thought that the removal of water by heat treatment prevents oxidation, thus causing a lower peroxide value detected in ghee tail fat, and at the same time, peroxides that are not heat-resistant substances may be removed by heat treatment, so the peroxide value is lower in ghee tail fats. Likewise, it can be said that the salt and additive added to the samples reduce the peroxide value in salted and additeved tail fats by inhibiting oxidation. It has been observed that the temperature has a significant effect on the peroxide value in tail fats stored at different temperatures. The peroxide values of tail fats were determined as 2.97 meqg O_2/kg at 25ºC, 3.73 meqg O_2/kg at 4ºC and 2.11 meqg O_2/kg at -18ºC. Lower peroxide values were obtained at -18ºC compared to other temperatures. It is expected that the lowest peroxide value will be detected at -18ºC in the research. An increase was observed in peroxide values until the 30th day of storage, and a decrease was observed in the following periods. It is estimated that this result may be caused by the breakdown of hydroperoxides formed in the initial stage of oxidation to malonaldehydes. It is thought that the changes in peroxide values are due to the fact that the hydroperoxides released in the first stage of oxidation are not stable and can turn into different decomposition products as the degradation progresses [8, 12].

TBA analysis is the most suitable method for the measurement of oxidative rancidity in fat-containing foods, and in this method, malonaldehyde is measured as the secondary oxidation product of polyunsaturated fatty acids [18]. There is an increase in the number of TBA (mg malonaldehyde/kg) in parallel with the accumulation of short carbon chain products that cause rancidity. The TBA values determined during the storage of tail fats with different application processes are presented in Table 4. While the TBA value was found to be 0.93 mg malonaldehyde/kg in the fresh tail fat group samples, it was determined as 0.72 mg malonaldehyde/kg, 0.81 mg malonaldehyde/kg and 0.76 mg malonaldehyde/kg in ghee tail fat, salted tail fat, and additived tail fat, respectively. The difference between fresh and ghee tail fat was found to be statistically significant. The increase in TBA value determined in salted tail fat may be due to the prooxidative effect of salt. TBA values of tail fats stored at 25ºC, 4ºC and -18ºC were determined as 0.89 malonaldehyde/kg, 0.94 malonaldehyde/kg and 0.55 malonaldehyde/kg, respectively. The lowest TBA value was found in the samples stored at -18ºC, and it is thought that freezing at -18ºC delays lipid oxidation in the samples, compared to storage at room temperature and +4ºC, which may be a better method for the preservation of tail fat. There are statistically significant ($p<0.05$) differences between storage times. In parallel with the storage period of tail fat, an increase occurred in TBA values until the 60th day, and then a decrease was observed. It is thought that the decrease in the TBA value observed after the 60th day may be due to the transformation of the released malonaldehydes into further degradation products. Yılmaz (2009) reported that the TBA value in fat samples increased depending on the storage period [8].

CONCLUSION

Within the scope of the study, the effects of heat treatment, temperature, storage time, salt and antioxidant addition on the storage stability of heat-treated tail fat were investigated and some physical and chemical properties of tail fat were investigated. The high amount of fat in the tail tissue has made it an important source of fat and increased demand [24]. It has been determined that fresh tail fat and ghee tail fat have lower cholesterol amounts compared to many

foodstuffs, and it can be said that they are preferable in terms of nutrition thanks to this feature. The amount of water, which affects technological properties of food, has been reduced by the application of heat treatment. In this context, the water activity value was low in ghee tail fat and this improved the technological and storage conditions of tail fat. Color is one of the most important features of acceptability for consumers, and the absence of a negative effect of heat treatment on color can increase the preferability of ghee tail fats. It was determined that the heat treatment did not have a significant effect on the fatty acid composition, and the fact that the saturated fatty acid content of fresh and ghee tail fat was lower than that of other animal fats enabled tail fat to be considered as an important source of fat in terms of nutrition. The fact that the melting temperatures in DSC thermograms were lower than body temperature in fresh tail fat and ghee tail fat showed that the absorption by the body was high and made tail fat preferable. FFA formation was significantly prevented by inactivating the enzymes in the structure with heat treatment. Removal of water by heat treatment prevented hydrolytic rancidity and peroxide values decreased. Fresh tail fat stored at room temperature deteriorated within 7 days, while samples stored at +4ºC deteriorated within 15 days. However, ghee tail fat could be preserved at these storage temperatures for 60 days without deterioration. As a result, heat treatment of tail fat and keeping it at low temperatures affected the storage stability of tail fat and allowed it to remain intact for a longer period of time.

AUTHOR CONTRIBUTIONS

Conceptualization, literature review, organization, analysis was done by MB. Review and editing were done by HG. HG had supervised the entire research works. Literature review, critical analysis of data, manuscript review and editing were written by MOY. All authors of this research read the manuscript and agreed to publish it.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

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