



## Lipid peroxidation in sheep and cow traditional butter obtained from southern Tehran, Iran

Nastaran Akbarieh, Parisa Sadighara, Nabi Shariatifar, Ebrahim Molaee-Aghaee, Ramin Aslani\*

**Division of Food Safety & Hygiene, Department of Environmental Health Engineering, School of Public Health, Tehran University of Medical Sciences, Tehran, Iran.**

### ARTICLE INFO

#### Article history:

Received 16 Aug. 2022

Received in revised form

27 Oct. 2022

Accepted 10 Dec. 2022

#### Keywords:

Butter;

Lipid oxidation;

Free fatty acids;

Hydroperoxide;

Peroxide value;

Storage

### ABSTRACT

Butter is popularly known in dairy products and consists of large amounts of milk fat (at least 80%). The risk of oxidation increases by a higher content of unsaturated fatty acids in milk fat and adversely affects the quality and safety of the product. Primary oxidation products are hydroperoxides, which can decompose to form lower molecular weight compounds such as alcohols, aldehydes, free fatty acids (FFA), and ketones. This process may give rise to butter rancidity. In this study, 20 samples of traditional butter, including 10 samples of cow butter and 10 samples of sheep butter, were purchased from different traditional dairy product supermarkets in southern Tehran and were analyzed to evaluate the amount of fat oxidation. The peroxide levels in all samples of traditional butter were within the permissible limit established by Iran's national standard. According to the results, it was found that the amount of peroxide in cow butter ( $1.08 \pm 0.12$  mEq/kg) is higher than in sheep butter ( $p=0.037$ ). It may be related to the temperature and time of storage. Therefore, strategies such as diminishing temperatures, maintaining products in a dark environment, removing oxygen by creating a vacuum in the packaging, and using antioxidant compounds can prevent butter oxidation.

**Citation:** Akbarieh N, Sadighara P, Shariatifar N, Molaee-Aghaee E, Aslani R. **Lipid peroxidation in sheep and cow traditional butter obtained from southern Tehran, Iran.** J food safe & hyg 2022; 8(4): 237-242

### 1. Introduction

Dairy products with a wide range of ingredients are among the most important components in the human diet and provide many essential nutrients for human health during lifetime (1).

Butter with a high content of fatty acids is one of the most popular dairy products obtained from milk or milk products. It is frequently consumed in different foods such as pastries as an ingredient or in breakfast due to its aromatic and nutritional properties.

\*Corresponding author. Tel.: +982141933075

E-mail address: [ramin.aslani.m@gmail.com](mailto:ramin.aslani.m@gmail.com)



Copyright © 2022 Tehran University of Medical Sciences. Published by Tehran University of Medical Sciences.

This work is licensed under a Creative Commons Attribution-NonCommercial 4.0 International license (<https://creativecommons.org/licenses/by-nc/4.0/>).

Non-commercial uses of the work are permitted, provided the original work is properly cited.

Butter is manufactured industrially, however it is also produced traditionally from yoghurt and cream. It is a water-in-oil emulsion containing a large amount of milk fat (at least 80%). Rancidification is one of the major causes to the butter spoilage which may occur by long time storage, lipolysis and oxidation (2, 3). Milk fat is a complex natural fat composed of over 400 different fatty acids and contributes to other dairy products' physical properties, taste, and nutritional value (4). The quality and taste of butter depend on several factors, including the type of animal (cow, goat, sheep, or buffalo) and the season. Furthermore, the quality of the food the animal eats can impact the final product (5). Traditional butter produced in different regions of Iran is popular because of its good flavor and relatively long shelf life (6).

The main issue in butter storage is rancidity, which can lead to severe problems and damage to the dairy industry and food distribution. This process is caused by lipolysis (the release of free fatty acids) and oxidation of the fatty acids (7). Butter from traditional production methods has higher humidity than industrial butter due to the different production methods. High humidity increases the activity of lipase and stimulates the growth of microorganisms, which in turn promotes the hydrolysis of triglycerides (7). Oxidative spoilage is significant in lipid-containing foods, and lipids containing unsaturated fatty acids are more susceptible to oxidation (8, 21). The risk of oxidation increases by a higher content of unsaturated fatty acids in milk fat and causes undesirable flavors.

Oxidation, especially after storage, can cause metallic or old flavors and discoloration in dairy products with a high content of unsaturated fats. Also, rancidification takes place as a result of oxidation and lipolysis which is contributed to a decreased nutritional quality, off-flavors such as bitter, butyric, soapy in milk and dairy products (9).

Quality of butter is largely dependant on the quality of milk which is produced from. Ability to storage and consumption of butter are effected by the chemical and biological properties that may alter during processing (7).

The oxidation process begins with the loss of hydrogen radicals from a fatty acid in the presence of rare metals, light, or heat and the formation of a lipid-free radical. Lipid free radicals (L<sup>•</sup>) react with oxygen and form peroxy radicals (LOO<sup>•</sup>). In this process called Propagation, LOO reacts with more LH and forms lipid hydroperoxides (LOOH) (10, 11). Hydroperoxides are the primary products of oxidation, which can accumulate and subsequently decompose to form lower molecular weight compounds such as alcohols, aldehydes, free fatty acids (FFA), and ketones. This process leads to autoxidative rancidity (12). The amount of oxidation can be estimated by determining the amount of peroxide and acid value of the fat. Therefore, the lower the amount of peroxide and acid, the better the quality of the lipids (13). According to the high consumption of butter and different conditions of production, storage and supply, the purpose of this study was to investigate the amount of fat oxidation in traditional Iranian animal butter by measuring the peroxide value.

## 2. Materials and Methods

### 2.1. Sample collection

In this study, 20 samples of traditional butter, including 10 samples of cow butter and 10 samples of sheep butter, were purchased from different traditional dairy product supermarkets in the south of Tehran, Iran and were stored at  $-18^{\circ}\text{C}$  until the experiment.

### 2.2. Determination of Peroxide value (PV)

Five grams of butter were dissolved in a 40 mL hexane solution. After stirring for 30 min, the impurities were removed with filter paper. The obtained solution was placed in a rotary evaporator and pure butter solution was obtained by withdrawing hexane. Then, 30 mL of acetic acid and chloroform solution (v/v 2:3) was added to the pure butter solution, followed by the addition of 0.5 mL of a saturated potassium iodide solution. The mixture was maintained in the dark for 1 min. Following the dark stage, 30 mL of distilled water was added. In the next step, 1 mL of starch reagent was added to the sample. The sample turned dark purple, with darker purple indicating higher concentrations of peroxide. The peroxide index was calculated based on the titration results until the sample became colorless.

### 2.3. Statistical analysis

Statistical analyses were performed using SPSS 26 software following one-way ANOVA. Differences were considered statistically significant at the  $p < 0.05$  level.

## 3. Results

Peroxide values of traditional butter samples are presented in Fig. 1 and 2. As shown, the amount of

peroxide in cow butter ( $1.08 \pm 0.12$  mEq/kg) is higher than in sheep butter ( $0.78 \pm 0.11$  mEq/kg) ( $p=0.037$ ). When comparing organoleptic properties, the differences in terms of color, smell, and taste were observed between these two types of butter. Sheep butter is slightly whiter and lighter in color than cow's butter.

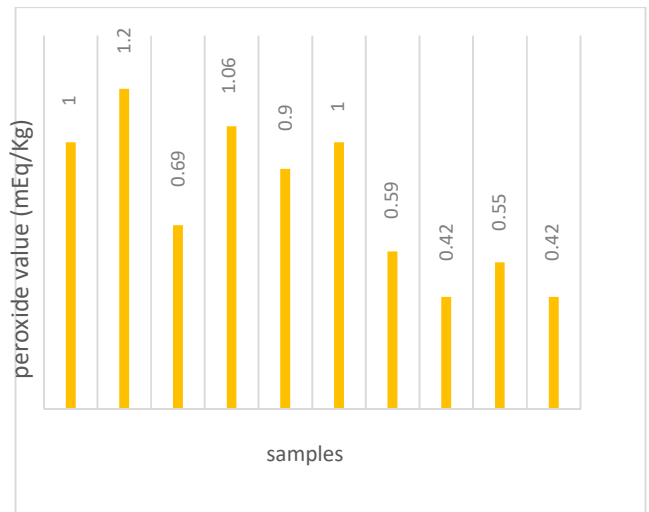


Figure 1. The peroxide value in different sheep butter samples.

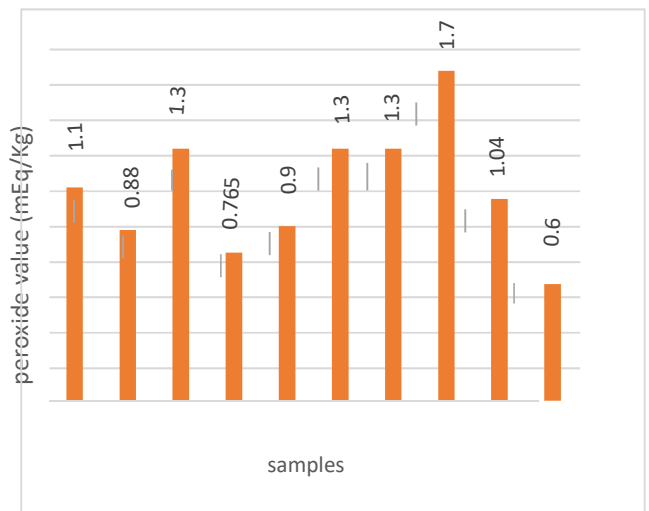


Figure 2. The peroxide value in different cow butter samples.

#### 4. Discussion

In this study, the amount of lipid peroxidation was tested in traditional cows and sheep butter. The level of lipid peroxidation in cow butter was higher than in sheep butter, and a significant difference was observed between these two kinds of butter. The difference in storage time may explain the difference in quality between cow's butter and sheep's butter. Furthermore, the type of feed and the animal-keeping conditions can also be effective. The maximum permissible amount of butter peroxide should not exceed 1.7 mEq of oxygen per kilogram, according to the national standard of Iran No. 4179. The amount of peroxide in all samples of traditional butter was within the permissible limit of Iran's national standard.

Several other studies have also reported similar results. Razm et al. (2013) compared the fatty acid profile and physicochemical indices of traditional sheep and cow butter. The results indicated that the peroxide index of all samples was within standard limits. Furthermore, the highest value of peroxide index was observed in spring cow butter and the lowest value in summer sheep butter (6). Dervisoglu et al. (2013) also reported the peroxide value of all 88 samples within the standard limits after examining the physical and chemical characteristics of 88 different butter samples (14). Another study, by Steward et al. 1980 in Canada on soybeans, found similar results. The results of this research indicate that the amount of malondialdehyde, a peroxidation indicator, increases with storage time (15).

Although Ghasemloy et al., in research on the quality of traditional butter produced in West Azarbaijan province, reported the amount of butter peroxide in seven cities outside the standard range (7). Moreover, Sağdıç et al. research results in 2004 on the peroxide index of butter prepared from goat, sheep, and cow milk did not show a significant difference in the index in the three butter samples (16).

Krause et al. (2008) examined the amount of peroxide in animal butter stored in the refrigerator and the freezer, after some time. Both refrigerated and frozen storage showed a significant increase in PV over 18 months, although the increase was greater in refrigerated storage. Six months later, the PV values of the frozen spheres showed a significant increase, while the PV of the refrigerated spheres did not show a significant increase (17). After 14 weeks of storage at 20°C, the peroxide level in the dairy spread was higher than in butter. This difference could be due to the higher level of unsaturated fatty acids in the dairy spread (9% fatty acids in the dairy spread vs. 2% in butter) (18). Méndez et al. (2017) investigated the effects of temperature and salt addition on the chemical and physical changes of butter during storage. The amount of peroxide significantly increased with increasing storage time (9 months of storage). Fat oxidation increases at higher storage temperatures and the presence of salt, and the oxidation rate is much lower at low temperatures (9). Mokrejs et al. (2012) found that the increase in peroxide levels in uncoated butter was significantly higher than in butter kept inside the coating (19).

This issue could be caused by oxygen and light, since fat oxidation increases in the presence of oxygen and light (16).

It is possible to significantly prevent fat oxidation and peroxide production by reducing the storage temperature and using a coating during butter storage. Also, the use of antioxidants effectively prevents oxidation and increases storage time (20).

### 5. Conclusions

The oxidation of lipids is an important quality concern, because it leads to the development of off-odors and tastes in oily products. The results of this study indicate that the traditional sheep and cow butter available in the south of Tehran is within the acceptable range according to the national standard of Iran. The amount of peroxide in cow butter was higher than in sheep butter, and the possible reason for this difference is probably related to the conditions of keeping and the type of feeding of animals. Methods to prevent oxidation include using lower temperatures, keeping products in a dark environment, removing oxygen by creating a vacuum in the packaging, and using antioxidants. Good personal hygiene, disinfection of equipment, prevention of microbial contamination of samples during production, transportation, storage, and distribution, control of sanitary conditions and storage temperature in retail stores, and maintaining the cold chain before consumption can help to improve the quality of butter.

### Conflict of Interest

The authors declare no conflict of interest.

### Acknowledgments

This study was supported by Tehran University of Medical Sciences.

### References

1. Haug A, Høstmark AT, Harstad OM. Bovine milk in human nutrition—a review. *Lipid Health Dis* 2007; 6: 1-16.
2. Akgül Hİ, Şengül M., Ürkek B, et al. Determination of physicochemical and microbiological properties and fatty acid composition of butter produced in Trabzon, Turkey. *Acta Scientia Technol* 2021; 43: e48905-e48905.
3. Codex alimentarius, Standard for Butter, in CXS 279-1971 amended in 2018.
4. Akalin AS, Gönç S, Ünal G, et al. Determination of some chemical and microbiological characteristics of Kaymak. *Grasas Y Aceites* 2006; 57: 429-32.
5. Alganesh TG, Yetenayet BT. Traditional butter and ghee production, processing and handling in Ethiopia: a review. *Afric J Food Sci* 2017; 11: 95-105.
6. Razm S, Ataye Salehi E, Ghiyasvand R. Comparison of the fatty acid profiles and physicochemical indexes of traditional sheep's and cows's butters. *J Food Sci Technol (Iran)* 2016 13: 35-43.
7. Ghasemloy Incheh KH, Hassanzadazar H, Forouzan SH, et al. A survey on the quality of traditional butters produced in West Azerbaijan province, Iran. *Int Food Res J* 2017; 24: 327-32.
8. Frankel E. Lipid oxidation. *Progress Lipid Res* 1980; 19: 1-22.
9. Méndez-Cid FJ, Centeno JA, Martínez S, et al. Changes in the chemical and physical characteristics of cow's milk butter during storage: Effects of temperature and addition of salt. *J Food Composit Analys* 2017; 63: 121-32.

10. Frankel, EN. Lipid oxidation: mechanisms, products and biological significance. *J Americ Oil Chem Soc* 1984; 61: 1908-17.
11. Min, DB, Lee HO. Chemistry of lipid oxidation. *Flavor Chem* 1999: 175-87.
12. Simsek B. Studies on the storage stability of yayik butter. *J Verbrauch Lebensmittel* 2011; 6: 175-81.
13. Laikoja K, Teder L, Jõudu I. Assessment of chemical and sensory quality of unsalted and salted sweet cream butter during storage at different temperatures and time. *J Agri Sci* 2017; 2: 76-81.
14. Dervisoglu M, Gul O, Guvenc D, et al. Evaluation of chemical and microbiological characteristics and fatty acid profiles of butter samples collected from the black sea region of Turkey. *Asian J Chem* 2013; 25: 10185.
15. Stewart RR, Bewley JD. Lipid peroxidation associated with accelerated aging of soybean axes. *Plant Physiol* 1980; 65: 245-48.
16. Sağdıç O, Dönmez M, Demirci M. Comparison of characteristics and fatty acid profiles of traditional Turkish yayik butters produced from goats', ewes' or cows' milk. *Food Control* 2004; 15: 485-90.
17. Krause AJ, Miracle RE, Sanders TH, et al. The effect of refrigerated and frozen storage on butter flavor and texture. *J Dairy Sci* 2008; 91: 455-65.
18. Christensen T, Hoelmer G. Lipid oxidation determination in butter and dairy spreads by HPLC. *J Food Sci* 1996; 61: 486-89.
19. Mokrejs P, Fenyk J, Janacova D, et al. Study into oxidative changes of butter with protective coat. *Biosci Biotechnol Res Asia* 2016; 9: 199-04.
20. Ozturk S, Cakmakci S. The effect of antioxidants on butter in relation to storage temperature and duration. *Europ J Lipid Sci Technol* 2006; 108: 951-59.
21. Kamkar A, Tooriyan F, Jafari M, et al. Antioxidant activity of methanol and ethanol extracts of *Satureja hortensis* L. in soybean oil. *J Food Qual Hazard Control* 2014; 1: 113-19.