K Int J Health Sci. April 2022;Vol 5(1): 10-18

ORIGINAL ARTICLE

Evaluation of Physico-chemical Properties According to TS 1331 of Butters Sold in Istanbul, Turkey

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ABSTRACT

In our country, the standard for butter is officially regulated according to Turkish Standard (TS) 1331. Butter is an animal fat dairy product with high nutritional value and a distinctive smell and taste. According to the Standard, there are 3 types of butter: breakfast, culinary and plain. Its high price compared to other oils is related to the cost of butter production. This situation causes the use of various imitation-adulteration methods in order to make the product cheaper. Therefore, the aim of the study was to evaluate the physico-chemical properties of butter offered for sale in the Istanbul market according to TS 1331. Reichert-Meissl Index (R.M.I.) value, peroxide number, ratios of milk fat, salt, moisture and acidity were determined in 26 different butter samples. The refractive index was also determined. The most common adulteration in products sold as butter is the addition of margarine to the product which occurs with the determination of the R.M.I. value in samples. The R.M.I. values in 21 samples (81%) and the milk fat ration in 20 samples (77%) were found in accordance with TS 1331. While the acidity in 25 samples (96%) was found to be suitable for TS 1331, it was found to be abnormal in 1 (4%) sample. While the moisture ratio exceeded the maximum value in 2 of the samples (8%), it was found to be among the expected values in 24 (92%) samples. The refractive index, salt and peroxide values are within normal ranges in all of the samples. Considering all the results, 16 butter samples (61.5%) are suitable for TS 1331, while 10 (38.5%) are not. All these results show that there are still imitation-adulteration in the butter offered for sale. It is necessary to increase the inspections on the fact that products that deceive the consumer, put human health at risk and have low nutritional value on the market, and provide education to consumers explaining food safety and the negative effects of these fraudulent foods on health. In addition, it is necessary to carry out food control studies periodically and with a larger sample.

Keywords: *Butter, TS 1331, Imitation and adulteration in food, Food safety*

INTRODUCTION

Butter is a dairy product that is easily digestible, melts at body temperature, contains many nutrients, and has an important place in nutrition because it is a source of energy (1). The reason for the ease of digestion of butter is that milk fat has unique fatty acids and low melting point. Butter is defined as a dairy product that is obtained as a result of processing milk or dairy products in accordance with the

Received 20-02-2022 Accepted 03.04.2022

technique, and can be added to additives, seasonings and other foodstuffs in accordance with the legislation when necessary (2).

The fact that it contains saturated and unsaturated fatty acids as well as low and high molecular fatty acids makes butter different from other oils (3). While capronic, caprylic and butyric acids from the saturated fatty acids in its structure are solid at room temperature; oleic, palmitoleic, linoleic and linolenic acids from unsaturated fatty acids are in liquid form at room temperature. Essential fatty acids such as linoleic and linolenic increase the nutritional value of butter even more. In addition, butyric acid, which is not found in any oil, ensures that butter has a popular taste and smell. Butter contains plenty of vitamin A, β-carotene and essential fatty acids (4). However, due to the fact that its economic value is quite high compared to other oils, the consumer has been deceived and victimized for a long time due to imitation and adulteration (especially adding margarine) by people who want to gain unfairly (5). In our country, although this issue is constantly on the agenda, it has been determined that these concerns have increased especially recently among consumers. This opportunism can also occur because the oil is open to cheating in terms of physical condition. Due to these opportunism, people cannot benefit from butter, which has a high nutritional value and is frequently preferred for breakfast. In order to prevent this imitation and adulteration, it is recommended that people carefully read the labels of the butter they buy. However, the majority of the butter that is sold is openly sold and there is no label on it. It has been observed that there are missing or incorrect information on the labels affixed to the oils that are not wrapped with the appropriate packaging technique. In the determination of these adulterations, the difference of butter from other oils and its qualitative results are examined.

The most commonly used detection method for imitation-adulteration is the determination of R.M.I. which is based on revealing only short-chain fatty acids (such as butyric acid and caproic acid) found in milk fat (6). As a method of identifying imitation and adulteration in butters, the determination of R.M.I. a faster, cheaper and still valid method (5,6). Reliability of this method is directly proportional to the contribution rate. In addition, the formula created by using the R.M.I. works exactly (7). Inexpensive margarines for cooking are a source of adulteration, commonly found in butter. Margarines contain more trans-fatty acids (10-35%) than butter (3-8%) (8). The trans-fatty acids occur mostly during heating, hydrogenation and processing of vegetable oils. In milk-derived fats, trans fatty acids are formed to a lesser extent, but mostly by hydrogenation (9). These compounds increase the level of low-density lipoprotein (LDL) and decrease the level of high-density (HDL) lipoprotein. Therefore, they increase the risk of heart diseases. The presence of trans fatty acid content of more than 10% in butter is an indication of the fact that butter is mixed with margarine (adulteration) (8). The prevalence of imitation and adulteration in butter worries consumers and this situation increases the demand for reliable products (10). Although, it is an important and serious responsibility to offer butter to consumers in accordance with the Standard, labeling, reliability of label information and packaging are very important in this regard. The Standard (TS 1331) consisting of various parameters has been determined to prevent imitation and adulteration in butter. The aim of this study is to determine the peroxide number, R.M.I. value, milk fat content, acidity ratio, moisture ratio and salt ratio as a result of physico-chemical analysis is to investigate the compliance of butter with TS 1331. In addition, the butyro index was also calculated from the refractive index results for the determination of the quality in butter samples.

MATERIALS AND METHODS

This study was carried out on laboratories at The Bezmialem Vakif University, Faculty of

Received 20-02-2022 Accepted 03.04.2022

Health Sciences, Department of Nutrition and Dietetics between October 2019 and January 2021. Ethics committee approval for the study, Bezmialem Vakif University Non-interventional Research Ethics Committee approved 54022451-050.05.04- 15084 dated 18.09.2019.

Determination of Sample Number for Analyzes

Assuming that the inappropriate sample rate varies between 5% and 20%, it was calculated by power analysis that at least 25 samples should be studied, taking into account the 95% confidence level and 80% power coefficients, in order to reveal this frequency approximately. Therefore, 26 butter samples were collected from Istanbul markets, Turkey.

Physico-Chemical Analyzes of Butter Samples

Milk fat in butter should be present minimum 80% and maximum 90% according to

Turkish Food Codex Butter, Other Milk Fat Based Spreads Products and Ghee Communique (Communiqué No: 2005/19) (11). The moisture content can be found in butter at a rate of 16% and other substances at a rate of about 2%. Other substances can be 0.5-0.8% lactose, 0.6-0.7% protein and 0.14% mineral substances (7).

Analyzes for the determination of salt ratio, Reichert-Meisll Index, moisture ratio, milk fat content in ratio, peroxide value, and acidity ratio in butter samples were performed according to TS 1331. In addition, the refractive index including butyro index was also investigated to determine the quality of butters (12).

Determination of Salt Ratio

For the determination of the salinity of the butter samples, 5 g of butter was weighed in a precision balance (Ohaus, USA) and 100 mL of hot distilled water was added to it. After the oil was dissolved, 5% potassium chromate was added and titrated with 0.1 N $AgNO_3$ (Merck, Germany) solution. The amount spent at the end of the titration was recorded from the pipette and the results were calculated as salt ratio (%) in NaCl. The calculation was made using the formula given below.

$$\text{Salt \%} = (V \times 0{,}00585 \times 100 \times F)/m$$

V = Amount spent in sample titration, mL; m = amount of oil sample taken, g; F = Factor of $AgNO_3$ used in the analysis.

Determination of R.M.I. Value

Five grams of the butter sample was weighed on a precision balance and the balloon was centrifuged for 5 minutes (Gerber, Germany). 20 g of glycerin (Merck, Germany) was added to the weighed sample and 2 mL of 44% NaOH (Merck, Germany) solution was added. The contents of the balloon, together with the boiling stone, were swept over a light flame, shaken and saponified. Heating was terminated when the resulting foams disappeared completely and the liquid color became lemon-golden yellow. The mouth of the balloon was closed with a watch glass and allowed to cool down to 90°C. When the desired temperature was reached, 90 mL of distilled water boiled at almost the same temperature was added and mixed. Then, 50 mL of 1 N sulfuric acid was added and as soon as it was added, it was connected to the prepared distillation apparatus and distilled. The temperature of the refrigeration (Lauda Alpha Ra, Germany) is 10°C. After 110 mL distillation, phenolphthalein (Merck, Germany) as an indicator solution was added and titrated with 0.1 N NaOH solution (Merck, Germany). The amount consumed as a result of the titration was recorded from the burette and the results were calculated. The calculation was made using the formula given below.

$$\text{RMI Value} = 1.1 \times F \times V_1$$

F = Factor of 0.1 N NaOH used in analysis; V_1 = The amount of alkali (0.1 N NaOH) consumed in the titration of the sample, mL.

Determination of Moisture Ratio

For all samples, porcelain vessels were left in a pre-set oven (Binder ED 115 E3, Germany) at a temperature of 102 ± 2°C until their

Received 20-02-2022 Accepted 03.04.2022

weight stabilized. After the porcelain cups were taken out of the oven, they were left to cool in the desiccator and the bowl was weighed after cooling. Five grams butter sample was weighed into the porcelain bowl and kept in an oven at 102 ± 2°C for 2 hours. When the time was over, the samples were left to cool in a desiccator and then weighed again. In the gravimetric method, the weighings were made using a precision balance (Ohaus, USA) and the results were calculated as %. The calculation was made using the formula given below.

$$\text{Moisture \%} = 100 \times ((\text{m-m}_1)/\text{m})$$

m = Weight of butter before drying, g; m_1= Weight of butter after drying, g.

Determination of Milk Fat Ratio

The butter sample was filled up to the upper level of the graduated centrifuge tube. The samples were centrifuged for 10 minutes in a heated centrifuge (Gerber, Germany). The centrifuged samples were examined according to the degrees on the tubes, and the milk fat content was calculated as percentage (%).

Determination of Peroxide Value

On a precision balance (Ohaus, USA), 2 g of butter was weighed into the flask and 10 mL of chloroform (Fluka, Switzerland) was added to it. The flask was stirred and shaken until the butter was dissolved. After dissolving, 1 mL of saturated potassium iodide (Fluka, Switzerland) and 15 mL of acetic acid were added and the flask was closed. After the shaking process was completed, it was kept in a dark environment for 5 min. After the waiting process was finished, 75 mL of water and 1 mL of starch (Merck, Germany) solution were added and titrated with 0.01 N sodium thiosulfate (Merck, Germany). The amount consumed as a result of the titration was recorded from the pipette and the results were calculated. The calculation was made using the formula given below.

$$\text{Peroxide Value} = (V/m) \times 10 \times F$$

V = Amount spent in titration of sample, mL; m = amount of oil sample taken, g; F = 0.01 N $Na_2S_2O_3$ factor used in analysis.

Determination of Acidity Ratio

Five grams of butter samples were weighed on a precision balance (Ohaus, USA) for acidity determination. Then, 100 mL of hot distilled water was added to it. After the oil had dissolved sufficiently, phenolflatein (Merck, Germany) was added and titrated with 0.1 N NaOH solution. The amount consumed at the end of the titration was recorded from the pipette and the results were calculated as percentage (%) in terms of lactic acid. The calculation was made using the formula given below.

$$\text{Acidity \%} = (V \times 0.009 \times 100 \times F)/m$$

V = Amount spent in sample titration, mL; m = amount of sample sample taken, g; F = Factor of NaOH used in analysis.

Determination of Refractive Index

One drop of butter samples was placed in the refractometer (Abbe5, United Kingdom) which was centrifuged (Gerber, Germany) with the water converter set to 40°C and the reading was recorded. The refractive index was determined by looking at the equivalent of the reading in the table, and the butyro index was determined (12).

RESULTS

Twenty-six butter samples examined for the compliance to TS 1331 (2). The refractive index of butter samples varies between 1.4531-1.4545. Salt ratio and peroxide values for all samples were among the standard values. The acidity ratio of 25 samples (96%), the moisture ratio of 24 samples (92%), the R.M.I. of 21 samples (81%), the milk fat ratio of 20 samples (77%) were assigned as suitable to the Standard. The findings of the analyzes are shown in Table 1. In terms of all parameters, 16 (61.5%) of the butter samples confirming to the standard and 10 (38.5%) were found to be contrary to the standard.

Received 20-02-2022 Accepted 03.04.2022

Table 1. The results of physico-chemical analyzes of 26 butter samples according to TS 1331 are demonstrated. Samples are not suitable determined are given in bold. Mean values are given with standard deviation.

Sample Number	RMI value	Salt (%)	Acidity (%)	Fat (%)	Moisture (%)	Peroxide Value
TS 1331	24-34	Max. 2%2	Max. 0.27%	Min. 82%	Max. 16%	Max. 5 meq g/kg
1	26.2±1.1	0.13 ± 0.008	0.09 ± 0.000	87 ± 1.41	7.32 ± 0.57	1.77±0.12
2	30.7±1.3	0.10 ± 0.005	0.16 ± 0.001	83 ± 2.83	6.60 ± 2.57	1.80±0.15
3	29.9±0.8	0.09 ± 0.015	0.11 ± 0.007	82 ± 0.00	8.89 ± 1.68	2.12±0.22
4	29.5±0.5	0.06 ± 0.008	0.11 ± 0.000	86 ± 2.83	14.75 ± 0.40	2.30±0.05
5	30.6±0.2	0.07 ± 0.008	0.09 ± 0.000	88 ± 0.00	14.98 ± 0.17	1.78±0.02
6	27.6±0.7	0.18 ± 0.005	0.13 ± 0.001	85 ± 1.41	3.53 ± 1.42	1.56±0.02
7	28.5±1.0	0.10 ± 0.001	0.25 ± 0.002	**72 ± 2.83**	7.36 ± 0.58	0.60±0.00
8	28.0±1.3	0.08 ± 0.007	0.2 ± 0.000	**80 ± 0.00**	10.47 ± 0.40	0.60±0.00
9	26.1±0.8	0.03 ± 0.004	0.07 ± 0.001	85 ± 0.00	6.00 ± 0.82	1.41±0.09
10	26.5±0.7	0.07 ± 0.000	0.15 ± 0.002	84 ± 2.00	6.06 ± 1.06	0.72±0.10
11	28.8±0.5	0.06 ± 0.000	0.05 ± 0.000	87 ± 1.41	**17.9 ± 0.86**	2.07±0.11
12	28.4±1.1	0.07 ± 0.000	0.23 ± 0.000	85 ± 1.41	7.52 ± 0.51	0.93±0.05
13	31.6±0.2	0.16 ± 0.007	0.05 ± 0.000	90 ± 0.00	7.86 ± 1.06	2.38±0.07
14	29.9±0.4	0.03 ± 0.000	0.14 ± 0.014	**80 ± 2.00**	8.47 ± 0.34	1.12±0.02
15	31.6±1.0	0.06 ± 0.000	0.13 ± 0.000	85 ± 1.41	8.22 ± 0.56	0.95±0.00
16	31.8±0.7	0.34 ± 0.006	**0.47 ± 0.000**	84 ± 2.83	7.10 ± 1.25	4.20±0.00
17	28.7±0.0	0.30 ± 0.008	0.09 ± 0.001	85 ± 0.00	9.60 ± 1.03	0.94±0.03
18	28.9±0.5	0.11 ± 0.008	0.08 ± 0.012	85 ± 1.41	15.06 ± 0.26	0.91±0.03
19	24.1±0.0	0.07 ± 0.000	0.07 ± 0.000	85 ± 1.41	15.17 ± 0.08	0.90±0.00
20	**20.6±0.7**	0.04 ± 0.008	0.11 ± 0.000	**77 ± 1.41**	12.35 ± 2.17	0.93±0.00
21	28.3±1.1	0.31 ± 0.002	0.1 ± 0.012	**78 ± 0.00**	**18.7 ± 0.92**	0.92±0.00
22	28.6±0.8	0.36 ± 0.014	0.14 ± 0.013	81 ± 1.41	15.52 ± 0.45	2.36±0.25
23	**17.6±0.5**	0.16 ± 0.000	0.09 ± 0.000	**80 ± 2.00**	9.70 ± 3.20	4.67±0.18
24	**20.6±0.3**	0.04 ± 0.007	0.11 ± 0.001	95 ± 1.41	16.00± 0.00	2.01±0.09
25	**20.4±0.3**	0.23 ± 0.000	0.07 ± 0.001	95 ± 1.41	15.36 ±0.25	1.82±0.11
26	**22.1±0.7**	0.08 ± 0.008	0.15 ± 0.010	87 ± 1.41	12.17 ± 0.08	0.89±0.04

DISCUSSION

The One Health approach to milk and dairy production for the food safety and human health has the potential to improve global health and create best practices for producers to improve milk quality and production (13). Additionally, milk and dairy products industry is very important for human nutrition especially for infants and children, to lead a healthy life. Product prices in the market are affected by differences arising from production and operating costs. Manufacturers who cannot compete with big companies in terms of price and quality are trying to find a place in the markets with cheaper priced products. The desire to increase the profit share to be obtained from the product may bring imitation and adulteration in the products. Milk fat, which is an important and expensive food product for human due to economic reasons, can be adulterated by mixing it with cheap plant and animal based oils (14). There are many different tricks that can be done with butter (5,8,10). Thus, butter consumers are deceived and victimized, and they cannot sufficiently benefit from the benefits of butter (13). There are two reasons for adulteration in the food industry (5). First, it is done to turn the food into a more functional product, to increase the shelf life and to obtain a healthier product for the consumer.

The second cause of adulteration is by mixing low-quality products with high-quality products. Thus, the cost of the product obtained is reduced, and due to its low cost, it is sold at a lower cost, leading to unfair competition (13,14). These products carry a great risk by ignoring consumer health.

Received 20-02-2022 Accepted 03.04.2022

Based on the findings of this study, 26 butter samples were obtained from R.M.I. value between 17.6 and 31.8 and the average R.M.I. value of all butter. The number was determined as 27.13. In a similar study, 15 butter samples were investigated, R.M.I. values were between 22.65-28.40 and the lowest value in our study is lower than this study, and the highest value is higher (15). In another study the R.M.I. average value of butter samples was 26.17, while R.M.I. average value of margarine was 0.42. Wenn the samples of butter and margarine mixture in equal proportions, R.M.I. was measured as 0.55. In our study the lowest R.M.I. value determined as appears to be high (10). In the study conducted in Şanlıurfa, Turkey, the average R.M.I. value was found to be 25.477 ± 1.54 (16). In another study on butter produced in Şanlıurfa, R.M.I. value reported was between 18.9 and 24.14 (6). It is accepted that the R.M.I. value of butter varies between 23 and 33 and butters with values below 23 are mixed with various oils (17). The refractive indices of 26 butter samples were found to be between 1.4531 and 1.4545. Looking at similar studies, a study conducted in Şanlıurfa in 2019 examined the refractive indices of butter obtained from cow and sheep milk. According to the study, it was determined that the refractive index of butter made from cow's milk was between 1.425-1.462 whereas the refractive index of butter made from sheep's milk was 1.456-1.462 (18), which were found in the expected ranges. In another study conducted in Şanlıurfa, the average refractive index of butter was determined to be 1.4624 ± 0.02 (16). and it was stated in the geographical indication document that the refractive index of Şanlıurfa butter was 1.456 - 1.462 (19). Considering the butyro index corresponding to the refractive index in our study, it was observed that all 26 oils were within the reference ranges. The salt rate of butter obtained from sheep milk was determined as 0.02-0.05% (18). In the other study, the average salt in butter was determined as 0.048 ± 0.05% (16). The average salt percentage obtained as a result of our study is 0.13%. Compared to other studies, the average salt percentage was found to be higher in our study. It was stated that the acidity ratio was the lowest 0.02% in butter obtained from sheep milk, the highest 0.05%, and the lowest 0.03% and the highest 0.17% in butter obtained from cow milk (18). According to their properties, butter acidity is 0.27% in I. Class butters, II. 0.56% in class butter, in Class III. butter should be 0.63% (2,11). Investigated butter samples from Şanlıurfa were assigned as I.Class butter. But in our study, 25 of the 26 butter samples were Class I and 1 sample was Class II. In a study conducted on 34 butter samples in Erzurum, it was stated that the fat rate was the lowest 72.5% and the highest 86.0% (5). While only 16 of them are between 80.2-86.0%, 18 of them are between 72.5-79.5%. It was reported that this situation deceives consumers and may lead to malnutrition. In our study, as a result of the analysis performed on 26 butter samples, it was seen that the fat ratio was the lowest 72% and the highest 95%. Three samples showed the fat ratio as below 80%. At the same time, when we look at the fat ratios in clarified butter obtained from sheep milk is 98.00-99.25, while the fat ratio of butter obtained from cow milk is 95.00-99.00 (18). Fat ratio of butter produced in Van provience were determined as 93.85-99.40 (20) whereas butter produced in Şanlıurfa provience were the fat ratio of 98.10 ± 0.72 (16). In the Geographical Signal Document, Şanlıurfa butter should contain at least 99% fat (19). In a study, the highest moisture ratio was reported as 31.63 and the lowest 8.72 in 30 butter samples (21). The moisture ratio of 17 butter samples produced in Şanlıurfa provience were determined as 1.394±0.66 (16). Butter should have a maximum moisture of 16% (2). In our study, the moisture ratio of 2 butters was found to be more than 16% while 24 samples (92%) were compliance with the Standard. The average moisture ratio for all samples was found to be 10.87%. The lowest peroxide value was found 0.8 milliequivalents (meq) g/kg and

Received 20-02-2022 Accepted 03.04.2022

the highest was 1.74 meq g/kg while the average peroxide value was 1.13 meq g/kg while (5). In another study, determined peroxide values were 0.012-0.385 meq g/kg (18). In the present study, peroxide values were among 0.6-4.67 meq g/kg, while the average peroxide value was calculated as 1.64 meq g/kg.

Some limitations of the present study, however, need to be considered when interpreting the results. A convenience sample with a relatively small number of butter samples may limit the generalizability of the results. However, the performing a power analysis before the collection of sample is a considerable strength of the study.

In conclusion, with the widespread use of cheating on butter in recent years, this situation rightly worries consumers and butter producers who do their job correctly and in accordance with ethical rules. Cheats in butter keep its place on the agenda as it reduces the quality of the oil and is a risk factor for health. For this reason, butter attracts suspicion as the most cheated dairy product. That's why consumers prefer butter with guaranteed originality or butter sold by trusted vendors. It should be considered as an important and serious issue to put reliable and health-free butter on the table of consumers. It was determined that more than half of the butter samples analyzed and evaluated in the study were found to be suitable according to the current standard. However, it should not be overlooked that products against the standard are still offered for sale. Based on these results, affordable prices and accessibility to reliable foods should be ensured in order to consume dairy product, which is one of the foundations of a balanced and healthy diet, in an adequate and balanced way. However, people pay attention to the expiry date of the product most when buying dairy products. In addition, in order to increase the consumption of milk and dairy products per capita, not just butter, conferences, panels, trainings, public service announcements and events on nutrition should be organized by both public institutions and organizations and universities at regular intervals. Reliable food research training should be given and the importance of these foods in terms of health should be explained.

Contribution of Authors: Conceptualization, M.G.B., A.G.B.; Investigation, M.G.B., B.Ö., M.İ., B.A., B.Y., H.İ.Y.; Visualization: A.G.B., M.G.B.; Writing—original draft, A.G.B., M.G.B.; Writing-review and editing, A.G.B., B.G.B. All authors have read and agreed to the published version of the manuscript.

Funding: None.

Conflicts of Interest: The authors declare no conflicts of interest.

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Received 20-02-2022 Accepted 03.04.2022