Evaluation of the quality and safety of butter with an antioxidant complex of natural origin (birch bark extract and Aloe Vera)

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Abstract. Milk fat, on the one hand, provides high biological characteristics of butter, and on the other hand, it is a labile component susceptible to various types of spoilage under the influence of specific factors (atmospheric oxygen, high temperature, light, heavy metal ions). The processes that occur in the product during its spoilage are irreversible; they indirectly affect the organoleptic characteristics of the oil and, ultimately, lead to pathological changes in the human body. In this aspect, the search for effective antioxidants, especially those of natural origin, is of greatest relevance. However, we shouldn’t forget that the inclusion of a complex of natural antioxidants in the technological cycle of the production of butter will require manufacturers to conduct additional studies confirming the safety of new technological solutions and the stable quality of products. The article presents the results of evaluating the possible toxicity and allergenicity of butter with an antioxidant complex of natural origin (birch bark extract and Aloe Vera). Experimentally, in vivo experiments the absence of changes in the organs and tissues of the control and experimental groups of laboratory animals was proven. Preclinical studies were performed using standard methods on white, sexually mature laboratory mice of the CD-1 line / stock.

1. Introduction

The modern consumer market for food products has increased due to the appearance of goods with desired properties, long shelf life, and functional properties. Creating a new generation of products is impossible without the use of food additives with various functional properties. Within the framework of the Technical Regulation of the Customs Union “On Food Safety” (TR TS 021/2011), special attention should be paid to the quality of the new product and its safety for the consumer. At the same time, the safety of food products means the state of food products, indicating the absence of an unacceptable risk associated with harmful effects on humans and future generations. However, at present, the level of compliance of food products with mandatory requirements continues to raise consumer complaints [1, 2].

Butter is related not only with high-grade food products, but also with strategically important goods for the formation, storage and servicing of state material reserves. Milk fat is well absorbed and
performs energy, reproductive and building functions in the human body. Butter contains vitamins A, D, B12, up to 40% of oleic acid, which has a beneficial effect on blood cholesterol, phospholipids, etc. Therefore, the production of high-quality butter is an important economic task.

The analysis of numerous sources of domestic and foreign literature showed that the priority factor affecting the quality of butter is the quality of the feedstock, which is primarily determined by the state of its fat phase. For the production of good quality butter, it is advisable to use milk with a high fat content, which will not only increase the degree of its use, but also reduce the consumption of raw milk for the production of the product. It should be kept in mind that milk fat, on the one hand, provides high biological characteristics of butter, and on the other hand, it is a labile component susceptible to various types of spoilage under the influence of specific factors (atmospheric oxygen, high temperature, light, heavy metal ions). The processes that occur in the product during its spoilage are irreversible, they indirectly affect the organoleptic characteristics of the oil and, ultimately, lead to pathological changes in the human body [2].

In this aspect, the most urgent issues are, firstly, the regulation of the quality of raw materials taking into account the technological characteristics of milk fat, and secondly, the search for effective inhibitors (antioxidants) of natural origin, which are put directly into the product or in the packaging material. The imperfection of legal and organizational mechanisms regarding the quality of food products leads to the fact that there is a turnover of falsified food products on the Russian market, as well as products that do not meet the needs of the majority of the population. The development of new types of competitive products, in particular butter with guaranteed indicators of quality and safety over the entire shelf life, is relevant for both the manufacturer and the consumer, and for the state as a whole.

Solving the problem of maintaining the quality of butter at the proper level, to prevent possible oxidation, the transport container for butter is lined with package inserts made of polymeric materials, and not parchment. However, this method does not completely solve the problem. So, at not high plus temperatures, a staff appears, which increases the loss of the product by 5-6 times. And at low temperatures, the staff does not form on the surface of the product, but mold can appear during long-term storage, which also negatively affects the quality of butter [3, 4]. Despite the fact that manufacturers of dairy and fat products are increasingly using innovative types of packaging with antibacterial substances and barrier layers, it should be understood that by using modern types of packaging it is impossible to completely solve the problem of increasing the shelf life of butter, as it is necessary to prevent the influence of external negative environmental factors causing various types of damage [5, 6, 7].

In this aspect, the issues of improving the consumer characteristics of butter while increasing its shelf life, which can be solved by the development of new-generation food technology using biologically active additives containing a complex of physiologically functional ingredients that perform various technological functions, are becoming most urgent. Antioxidants occupy a special place among these substances - their spectrum of actions is aimed at slowing down the oxidation process by interacting with atmospheric oxygen, interrupting the oxidation reaction or destroying the already formed peroxides. At the same time, one of the obstacles to the use of antioxidants, especially of synthetic origin, may be the fact that some of them can cause allergic reactions, a violation of the calcium-phosphorus balance in the body, and negatively affect the digestive processes. In this regard, the search for effective inhibitors (antioxidants) of natural origin is relevant.

In Russia and the European Union, the use of antioxidants in products is strictly regulated. Currently, in the dairy and fat industry, mainly synthetic antioxidants are traditionally used as antioxidants - propyl and dodecyl esters of gallic acid, butyloxyanisole and butyloxytoluene, citric acid and its salts, etc. Noting their significant role in extending the shelf life of butter, it is necessary to take into account that the manufacturer should evaluate the need for a specific additive (according to its purpose, functional properties and recommendations for use) in each defined case and choose the most appropriate one to ensure the efficiency and stability of technological processes in a particular company [...]. In addition, according to a number of scientists A.P. Nechaev, M. Nikanorov, L.
Rosival, L.A. Sarafanova, I.E. Kostrovoy, Mon Erich, Lira Martin, it is advisable to add antioxidants at the initial stage of production and observing the necessary concentrations (depending on storage conditions, the degree of bacterial contamination and physico-chemical parameters of the product). Ignoring these recommendations may lead to a loss of activity of their impact [7, 8, 9, 10].

We theoretically substantiated and experimentally confirmed the expediency of using an antioxidant complex of natural origin in butter technology (birch bark extract and Aloe Vera) in quantities \((0.3: 0.8) \times 10^{-3}\) per 1 g of the fat component of the cream, which allows one to increase the time storage of the finished product and get a product with traditional consumer characteristics.

2. Purpose of work
Based on the foregoing, the purpose of these studies was to study the safety and to produce quality assessment of the developed butter with a complex of natural antioxidants in vivo.

3. Material and research methods
Preclinical studies were carried out on white sexually mature laboratory mice of the CD-1 line / stock of both sexes weighing 21-37 grams in accordance with the "Rules for the work using experimental animals" (Appendix to the order of the Ministry of Health of the USSR dated 08/12/1977 No. 755). Weights of organs were determined according to the method proposed by V.N. Tugarinova, G.A. Savonicheva. The hairline scoring of samples of skins of laboratory animals was carried out in accordance with OST 10 10-86 “Farm animals. Fur animals of cell breeding. Zootechnical requirements for appraisal (assessment).” For the experiment, by the method of pair-analogues, 3 groups of animals were formed, 5 animals in each, outwardly healthy and active, having a smooth, dense coat, eyes of bright red color. Laboratory animals were simultaneously involved in the experiment, which excluded the influence of external (microclimate) and other factors on their physiological state. Butter was given to animals orally, at one time, daily in addition to the main diet at the rate of 0.14 g per 1 kg of live weight per day in accordance with the order of the Ministry of Health and Social Development of the Russian Federation of August 2, 2010, N 593 n “About approval of recommendations on rational food consumption standards that meet modern requirements for a healthy diet” [2, 9]. The water met the requirements of Sanitary Rules and Regulations 2.1.4.10749-01 “Drinking water. Hygienic requirements for water quality”. The duration of the experiment was 14 days. At the end of the experiment, the mice were weighed, dissected, internal organs, blood and skins were subjected to further studies.

4. Research results and discussion
Proving the effectiveness of the use of a creamy antioxidant complex of natural origin (birch bark extract and Aloe Vera) in butter technology in amounts \((0.3: 0.8) \times 10^{-3}\) per 1 g of the fat component of cream, it was found that butter is characterized by more low acidity and peroxide value. In terms of "heat resistance" it outdoes the control sample, and, therefore, has good ductility at low temperatures \((5 \pm 3 \, \text{°C})\) and is able to maintain shape at room temperature \((18 - 22 \, \text{°C})\). In addition, the use of a complex of antioxidants can improve the resistance of milk fat to oxidation, including during storage of butter (table 1).

| Product name | AOA, mg / 100g, (standard quercetin) |
|--------------|----------------------------------------|
| Control sample | 1.1                                    |
| Butter with an antioxidant complex of natural origin (birch bark extract and Aloe Vera) | 1.6                                    |
It should be kept in mind that the inclusion of a complex of natural antioxidants in the technological cycle of butter production will require manufacturers to conduct additional studies confirming the safety of new technological solutions.

From literary sources it is known that in case of poisoning the body with toxic food, first of all, the liver and kidneys are damaged, the activity is reduced, convulsions appear and loss of consciousness occurs. In mice, noticeable changes in appearance (tarnishing and reduction of the coat), inhibition of behavioral reactions can be observed. As a result of observations, it was found that all groups of experimental animals were equally active, did not differ in food and water consumption, their behavior was adequate, their safety was 100%.

When examining the hairline, hair length, hair orientation, shine, strength, retention in the skin, and elasticity were determined. During the initial examination of all skin samples, expressed alopecia and hypotrichosis were not found (table 2).

| Animal Group | Hairline Characterization |
|--------------|---------------------------|
| 1st group (control) | The coat is short, smooth, coarse, well-fitting and tight, the density and shine are not expressed, there are a lot of back hair, there is no undercoat on the body, low fluffiness. |
| 2nd group | The coat is short, soft, well-fitting, silky, has a lot of outer hair, undercoat on the body is poorly expressed, average density, density and luster are not expressed, low-fluffy. |
| 3rd group | The coat is short, smooth, well-fitting, shiny, silky, dense, and the hair coat is represented by a large number of straight and strong outer hairs; the undercoat on the body is well defined, not very fluffy. |

The scoring of the hair cover of the samples of skins of laboratory animals is shown in table 3.

**Table 2.** The results of the analysis of the hairline of experimental animals

| Indicator        | Assessment results, points |
|------------------|----------------------------|
| Thickness        | 2                          |
| Hair's breadth   | 3                          |
| Smoothness       | 3                          |
| Fit              | 3                          |
| Undercoat        | 0                          |
| Shine            | 2                          |
| Strength         | 3                          |
| Fuzz             | low                        |
| Rigidity / Softness | coarse, soft, silky     |
| Coat height, cm  | 0.70±0.04, 0.85±0.03<sup>a</sup>, 0.95±0.01<sup>b</sup> |
| Total GPA:       | 2.29, 2.86, 4.57          |

The differences are statistically significant - <sup>a</sup>P<0.05; <sup>b</sup>P<0.01; <sup>c</sup>P<0.001.

Thus, it can be concluded that the hair coat of animals of the 3rd group, when “Useful Breakfast” butter was added to their diet, became more snug, silky, shiny and thick compared to the skins of the control and 2nd group. Also in the 3rd group, the longest outer hair and the presence of a well-defined undercoat were noted. An increase in the number of hair follicles occurred, probably due to the activation of hair growth factor receptors by glycosphingolipids. The duration of active division of productive cells in the follicles has increased.

Studies of the elemental composition of the skins of experimental animals are shown in Figure 1.
Analysis of the results of the study showed that, in comparison with the control group, macro- and microelements accumulated in the skins of experimental animals of the 2nd and 3rd groups due to their entry into the body in the form of an organic form. So, the sodium content increased by 10.1% and 11.8%, potassium - 11.3% and 20.6%, calcium - 6.6% and 12.2%, iron - 2.3% and 7.7%, respectively. In the skin of animals of the second group, the amount of magnesium and copper did not change compared to the control, and in the third group it increased by 1.5% and 7%, respectively. The zinc content remained unchanged.

The obtained experimental data confirmed the well-known fact that under stress and malfunction of the pancreas, the body accelerates the secretion of magnesium, manganese and zinc through the urinary tract.

After an autopsy, the sizes, color, and also the mass of the internal organs of the experimental and control mice were determined (table 4).

| Group of experimental animals | Weight ratios of organs |
|------------------------------|-------------------------|
|                              | heart | kidneys | liver | stomach | spleen |
| 1st (control)                | 0.003 | 0.019   | 0.053 | 0.028   | 0.006  |
| 2nd                          | 0.010 | 0.014   | 0.067 | 0.035   | 0.007  |
| 3rd                          | 0.07  | 0.014   | 0.062 | 0.048   | 0.007  |

*The weight ratios of organs (WRO) determine the ratio of the weight of the organ in grams to the body weight of the animal in grams.

From the data of table 4 it is seen that the weight ratios of the organs of experimental animals increased slightly compared to the control group. In the 2nd and 3rd groups of animals, the weight coefficients of the organs of the heart are higher by 0.007 and 0.004, the spleen is 0.001, the liver is 0.014 and 0.009, the stomach is 0.007 and 0.02, respectively, compared with the control group. The weight coefficient of the kidneys decreased for the 2nd and 3rd experimental groups by 0.005.
5. Conclusion
In order to conduct more in-depth and adequate studies to study the possible toxicity and allergenicity of butter, histological studies of internal organs fixed in formalin and a biochemical blood test were carried out, which fully confirmed the safety of using an antioxidant complex of natural origin (birch bark extract and Aloe Vera) in amounts (0.3: 0.8) * 10^-3 per 1 g of the fat component of cream in the technology of butter.

References
[1] Topnikova E V 2013 Main factors for ensuring the quality of oil products Milk processing 2 24-28
[2] Kuprina A O, Mamaev A V and Simonenkov A P 2013 Butter with antioxidant complex “Aloe vera” and birch bark “Useful breakfast” Technology and merchandising of innovative food products 5 49 - 55
[3] Vyshemirsky F A, Kaneva E F, Gordeeva E Yu and Vyshemirskaya K V 2009 Selection and theoretical substantiation of temperature conditions for storage of butter Storage and processing of agricultural raw materials 2 12-15
[4] Abrosimova S V 2013 Packaging as a tool in ensuring the quality and safety of dairy products Milk processing 1 14-16
[5] Gordeeva E Yu and Ivanova N 2007 Quality and storage capacity of butter and spreads Cheesemaking and butter-making 7 7-8
[6] Golubeva L V, Dolmatova O I and Kuznetsov V S 2017 Application of modified packaging in cream butter technology Bulletin of Voronezh State University of Engineering Technologies 79 (3) 73
[7] Dunchenko N I, Khramtsov A G, Makeev I A, Smirnova I A, Gavrilova N B, Golubeva L V, Kalinina L V and Poznyakovskaya V M 2017 Examination of milk and dairy products. Quality and safety. Textbook (Novosibirsk, Siberian University Publishing House).
[8] Mamaev A V, Kuprina A O and Yarkina M V 2014 Laboratory studies of the safety of butter and cottage cheese with antioxidants in vivo Scientific notes of Oryol State University 7 181 –183
[9] Solovyova A O and Simonenkov A P 2016 Use of dry extracts “Aloe Vera” and birch bark in the production of butter. Butter Safety and quality of agricultural raw materials and food Creation of a national food product quality management system: Collection of scientific papers of the International Scientific and Practical Conference (Moscow RSAU - MTAA) 411-415
[10] Ellen L M 1988 Tearing of light approach to better Dairy Foods 89 6
[11] Lang F and Lang A 1987 New developments in butter and uses of butter fat The Milk Industries 79 4-5
[12] Pitts E 1995 The European market for dairy ingredients Dairy Technol 3 70-86