# МЕЖДИСЦИПЛИНАРНЫЕ ИССЛЕДОВАНИЯ

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Original article

# STUDY OF THE EFFECT OF GAMMA RADIATION ON THE ANTIBIOTIC ACTIVITY OF OSMOTIC MICROBIOTA IN SOME TYPES OF URBECHS

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***Abstract***

**Background.** “Urbech” – traditional national product of Dagestan peoples recently found increasing popularity among adherents of a healthy diet. Urbech made according to traditional recipes retains its properties when stored for more than 1-2 years. Nowadays urbech range is constantly expanding. Some of the new types of urbech can spoil in 5-7 days.

**Materials and methods.** The samples of urbech made from coconut flakes and dried mulberries spoiled most quickly, peanut urbech with grated cocoa beans and date syrup, and sesame urbech with honey. The urbech has been treated with antioxidants and mild preservatives and has been exposed to gamma radiation. Using standard methods, the authors have determined humidity, pH, acid number of fat, viscosity, the number of pathogenic microorganisms, including salmonella, QMAFAnM, coliform bacteria, yeast and moldy fungi, osmophilic yeast.

**Results.** The introduction of antioxidants has increased the best before date of urbech by 7-14 days. Mild preservatives have had no effect on increasing the best before date of nut butter. The drug Polybiom has increased the best before date of urbech by 21-28 days. The study of the urbech microbiological indicators with obvious signs of spoilage has shown that the number of pathogenic microorganisms, including salmonella, QMAFAnM, coliform bacteria, molds and yeasts does not exceed the values regulated in TR CU 021/2011. Treatment of the urbech with gamma radiation has shown that the radiation dose of more than 2 kGy leads to the change in its organoleptic properties. Osmoresistant microorganisms are present in all the variants. Compared to the control samples, with an increase in the radiation dose, the osmophilic microflora decreases from 10 to 55 times.

**Conclusion.** During the storage of newly developed types of urbech, it has been found out that its spoilage is not associated with the natural processes of fat oxidation. No microorganisms above the values regulated in TR CU 021 have been found in the urbech. Osmophilic microorganisms develop in the experimental samples of the urbech. Gamma irradiation of urbech at a dosage permissible for food up to 10 kGy reduces the amount of osmophilic microflora up to 55 times. When treated with radiation at a dose of up to 2 kGy, the urbech organoleptic properties are preserved. Accordingly, the treatment with ionizing radiation at the doses up to 2 kGy is effective for preserving organoleptic and microbiological parameters urbechs, including osmophilic yeast, which is not regulated by the regulatory documents in the Russian Federation, for a certain period of storage. To study the causes of the urbech spoilage, further detailed studies of microorganisms and their metabolic products are required.

**Keywords:** urbech; microorganisms; osmophilic yeast; antioxidants; Mild preservatives; gamma irradiation; kGy; doses of ionizing radiation

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Научная статья

# ИЗУЧЕНИЕ ВЛИЯНИЯ ГАММА-ИЗЛУЧЕНИЯ НА АНТИБИОТИЧЕСКУЮ АКТИВНОСТЬ ОСМОУСТОЙЧИВОЙ МИКРОБИОТЫ В НЕКОТОРЫХ ВИДАХ УРБЕЧЕЙ

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***Аннотация***

**Обоснование.** Урбеч – традиционный национальный продукт народов Дагестана в последнее время находит все большее распространение среди приверженцев здорового питания. Урбеч, изготовленный по традиционным рецептурам, сохраняет свои свойства при хранении более 1-2 лет. Ассортимент урбечей постоянно расширяется. При этом некоторые из новых видов урбечей портятся в течение 5-7 сут.

**Материалы и методы.** Наиболее быстро портились образцы урбечей из кокосовой стружки и вяленых ягод шелковицы, арахисовый урбеч с тертыми какао-бобами и финиковым сиропом, а также кунжутный урбеч с медом. Урбечи были обработаны антиоксидантами и консервантами мягкого действия, обработаны гамма-излучением. Определяли влажность, pH, кислотное число жира, вязкость по стандартным методикам, количество патогенных микроорганизмов, в т.ч. сальмонелл, БГКП, дрожжей и плесневелых грибов, и осмофильных дрожжей.

**Результаты.** Введение антиокислителей увеличило сроки годности урбечей до 7-14 сут. Консерванты мягкого действия не оказали влияния на увеличение сроков годности урбечей. Препарат Полибиом увеличил сроки годности урбечей до 21-28 сут. Исследование микробиологических показателей урбечей с явными признаками порчи показало, что количество патогенных микроорганизмов, в т.ч. сальмонелл, КМАФАнМ, БГКП, плесеней и дрожжей не превышает показаний, регламентированных в ТР ТС 021/2011 значений. Обработка урбечей гамма-излучением показала, что доза излучения свыше 2 кГр приводит к изменению органолептических свойств. Во всех образцах урбечей присутствовали осмоустойчивые микроорганизмы. По сравнению с контрольными образцами при увеличении дозы излучения снижение осмофильной микрофлоры происходит в 10-55 раз.

**Заключение.** В процессе хранения новых разработанных видов урбечей установлено, что порча не связана с естественными процессами окисления жиров. В урбечах не обнаружены микроорганизмы, свыше регламентируемых в ТР ТС 021 значений. В опытных образцах урбечей развиваются осмофильные микроорганизмы. Гамма-облучение урбечей при допустимой для продуктов питания дозировке до 10 кГр снижает количество осмофильной микрофлоры от 10 до 55 раз. При обработке излучением дозой до 2 кГр сохраняются органолептические свойства урбечей. Соответственно, обработка ионизирующим излучением дозами до 2 кГр эффективна для сохранения органолептических и микробиологических показателей урбечей, в том числе нерегламентируемых нормативными документами в РФ осмофильных дрожжей, на определенном периоде хранения. Для исследования причин порчи урбечей требуется проведение дальнейших углубленных исследований микроорганизмов и продуктов их жизнедеятельности.

**Ключевые слова:** урбеч; микроорганизмы; осмофильные дрожжи; антиоксиданты; консерванты; гамма-излучение; кГр; доза ионизирующего излучения

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**Introduction**

The article discusses the issues related to ensuring the urbech microbiological safety, osmoresistant microorganisms in particular, as a result of its treatment with different doses of ionizing radiation.

Numerous ethnographic studies conducted at various times on the territory of Dagestan indicate that urbech is a traditional national dish of this region inhabitants [6]. Many peoples of the world use oilseeds, nuts, and honey as food, but only in Dagestan a dish from curly flax seeds ground on the stone millstones of a mill with the addition of melted butter and honey has appeared. Flax seed contains antioxidants (selenium and vitamin E), which prevent lipid oxidation, which affects the storage capacity of nut butter. The secret of urbech modern popularity is determined by traditional technologies, natural ingredients and minimal heat treatment, which has been highly appreciated by supporters of the healthy diet. Initially, urbech was prepared by craft and referred to food products of non-industrial manufacture. Nowadays, the urbech assortment is expanded due to the use of pumpkin seeds, stirrups, coconut flakes, stone fruits, oilseeds, amaranth, etc. Instead of honey, other ingredients for functional purposes are added to its composition: mango, cranberry, banana, strawberry, date syrup, etc.

In addition to urbech, dishes made from tahini (sesame butter) and hummus (chickpea flour) are widespread all over the world. The name “tahini” comes from the Arabic “tanina”, from “tanin” - flour, from “tanana” - to grind. To prepare tahini, one uses sesame seed butter, to which lemon juice, oil or water, salt, and spices are added [20]. Tahini has grown in popularity around the world, especially in using in vegetarian recipes, as it adds rich flavour to dishes [34]. Hummus is a traditional Arabic dish made from chickpea flour and served as a side dish or as an appetizer.

The humus popularity is growing around the world now. Its value is determined by chickpeas, a high-protein plant from the legume family, which is used for food by animal meat quitters. Reducing beef consumption helps reducing a number of lifestyle diseases such as cancer, diabetes, oncology and hypertension. The production of 1 kg of chickpeas requires 350 liters of water, and the production of 1 kg of beef requires 15400 liters [16].

Urbech, tahini and hummus are made from the ground fruits of various plants. However, grinding technologies in different parts of the world differ in their national characteristics. So, the technology for producing urbech is that the raw material is ground not on metal millstones, but on the millstones made exclusively of stone. It is believed that grinding seeds by using stone protects their energy, preserving their beneficial properties.

Before grinding, contaminants are removed from the raw material, the peeled seeds and nut kernels are fried, which leads to the acquisition of characteristic taste and technological properties [2]. When deviating from the set temperature during frying, changes occur in the taste and technological properties of the raw material. The resulting paste is mixed with sweet syrups, honey, ghee, and then dried and crushed fruits, berries and candied fruits are added. Along with it, the introduction of new ingredients into the traditional recipe with new food combinatorics creation can lead to different atypical reactions. Thus, mixing sesame butter with honey almost always gives sour taste after 2-3 weeks of storage.

Expanding the distribution geography and increasing the complexity of logistics requires research to ensure microbiological safety indicators and to determine the best before date and storage conditions of the finished product. Not all ingredients can be subjected to significant heat treatment to destroy surface microflora while preserving their natural color, taste and smell.

At the same time, insufficiently high temperature and duration of raw materials heating while preparing urbech can cause food poisoning. Gram-negative, motile, non-spore-forming, facultative anabolism Cronobacter spp., classified as a member of the Enterobacteriacea family, is often found on the surface of spices and dried fruits [21].

In recent years, there has been an increase in the nut production in the world. Nuts are very susceptible to contamination by pathogenic microflora, including Salmonella, which has led to the significant change in the approach to these products processing and handling using modern thermal and non-thermal methods to combat pathogens and the factors affecting their effectiveness [27]. Bacteria of the Cronobacter species, which pose the great danger to human health, can also be found in dry food mixtures [19]. It is known that honey is a shelf-stable product. At the same time, honey contains the great amount of undesirable microflora in its inactive state - mainly these are osmophilic representatives of unicellular fungi and fungus-like bacteria [4; 15; 31]. After mixing honey with pastes from seeds and nuts, the preservative properties of honey may deteriorate and cause spoilage of urbech.

Fat-containing products, which include urbech with the fat content of more than 20%, may not be subjected to ozonation for disinfection, despite the fact that such a technique is compatible with organic food processing. Ozonation causes the oxidation of fatty acids, enzymes and proteins in cell membranes, envelopes and cytoplasm. Ultraviolet radiation is not effective because it has low penetrating power and can cause changes in genomic material.

To suppress the causative agent of salmonellosis in tahini and hummus, the addition of acetic and citric acids in dosages of up to 0.5% has been effective [25]. To suppress Salmonella spp. and Listeria spp. the scientists have proposed to use the antagonistic effect of the Lactiplantibacillus plantarum and Compani Lactobacillus alimentarius strains [18; 23].

It has been established that when sesame seeds are roasted, some microorganisms also retain their activity. To decontaminate them, microwave radiation has been used, which has resulted in almost complete suppression of microbiological activity [29]. The literature contains information on the inactivation of Salmonella a temperature of 110...150°C [33].

It is known that the salmonella activity in tahini can be suppressed by adding the extracts of essential oils of various plants. However, these extracts have also changed the organoleptic properties of tahini [25]. Plant extracts having antioxidant properties can have the certain effect on extending the best before date of vegetable oils [8], but to ensure their microbiological safety research is required.

Foods and ingredients with low water activity are often the sources of pathogenic microorganisms that cause food poisoning. Such microorganisms stay alive for several months and even years in food products and their production workshops [28].

Dangerous microorganisms such as Salmonella enterica and Listeria monocytogenes can easily survive in date paste and syrup at a temperature of +4°C for more than 90 days. Consequently, the possibility of contamination of the finished product during its manufacturing must be completely excluded [32].

To destroy osmophilic microorganisms that can appear in the food products with low water activity, traditional types of processing in the form of thermal heating, drying, baking and frying in oil are not effective ways to deactivate pathogenic microflora. To destroy such microflora, it is required to observe more strict processing conditions using traditional methods or other methods, for example, steam treatment, treatment with moist hot air, since temperature in combination with moisture significantly reduces the thermal resistance of microorganisms [24]. This category of non-traditional processing methods also includes extrusion processing, irradiation, and radio frequency heating. The choice of one or another method of processing food products and raw materials for them depends on the chemical composition, recipe and other factors [17].

The use of low-frequency ultrasound increases the efficiency of drying plant raw materials and inactivation of microorganisms maintaining food properties such as texture and nutritional value at the same time. The use of ultrasonic and osmotic dehydration reduces water activity, increases the ability to rehydrate, improves product color and reduces nutrient losses [14].

The principle of food resources preservation plays an important role in food security governance in accordance with the Sustainable Development Goals (SDGs) [11]. According to FAO, annual losses for oilseeds, because of microbial spoilage in particular, amount to up to 20%. Microorganisms enter raw materials mainly from the environment (as the surface of plants has its own epiphytic microflora) and partly through technological equipment.

The Russian Federation monitors compliance with the law in the field of ensuring the safety of food resources and agricultural raw materials, taking into account generally accepted international standards, which contributes to the harmonious development of the national food safety control system [10; 11].

Traditionally, perishable food products, agricultural food raw materials are subjected to radiation treatment in order to ensure their microbiological safety, to destroy insects and pests, to inhibit germination and to extend the best before date of plant products and for quarantine phytosanitary disinfection [5]. The effectiveness of ionizing radiation exposure depends on the radiation dose, the type and strain of the microorganism and the radiosensitivity of the microorganisms and the type and condition of the product itself [1; 9]. Thus, a dose of 0.10-0.97 kGy leads to a 10-fold decrease in the number of Escherichia coli, a dose of 1.0-2.3 kGy leads to complete inactivation, the number of Salmonella decreases respectively at a dose 0.15-0.80 kGy and they become inactive at a dose of 3 ,7-4.8 kGy. As for mold, a dose of 0.06-0.60 kGy leads to its 10-fold decrease, and a dose of 1.3-11.0 kGy leads to its complete inactivation. As for Clostridium, doses of 0.8-2.5 kGy lead to its 10-fold decrease, and a dose 19.0-37.0 kGy results in its complete inactivation, etc. This proves that it`s necessary to perform practical testing of the radiation use at different doses for various types of food products and agricultural raw materials.

One of the most effective ways to ensure the microbiological safety of food products is the use of radiation treatment. Foodborne infections are caused by consuming food products contaminated with pathogenic and opportunistic microflora; consequently, such processing is the most optimal way to ensure safe and high-quality products, for consumers with immunodeficiencies (cancer patients) in particular [7].

Although low moisture foods (LMF) have been defined as foods with water activity (aw) of less than 0.85, and they are generally considered to be less susceptible to microbial spoilage and the foodborne pathogens growth, (e.g. in tahini and hummus), new inactivation methods are required, including radiation treatment, which can disrupt DNA or RNA or other bacterial structures, and which is effective for foods with varying moisture levels, resulting in subtle changes in texture, flavour and nutritional properties of products [31].

They have established the gamma irradiation effect on the Escherichia coli O157:H7 strain and microbiota inactivation in tahini: the number of E. coli O157:H7 decreased 10 times when treated with gamma radiation at the doses from 0.31 to 0.39 kGy. In unfavorable situations for microorganisms, the radiation resistance of E. coli O157:H7 decreases ($P < 0.05$). The irradiation dose of 1 kGy reduces the number of E. coli O157:H7 and microbiota in tahini by 2.6–3.2 • 10 1 CFU/g and 1.6 • 10 1 CFU/g, respectively ($P > 0.05$) [30].

When treated with gamma radiation at the doses up to 4 kGy of tahini halva or its main ingredient tahini, previously inoculated with fresh or dried cultures of the tested microorganisms separately Salmonella spp., Listeria monocytogenes and Escherichia coli O157:H7, it has been found out that when the microorganisms are dried, the resistance coefficient decreases Salmonella and L. monocytogenes to gamma radiation up to 0.85 and 0.73 respectively, compared to the cells not subjected to desiccation, as it has not affected the resistance of E. coli O157:H7. Gamma irradiation at the doses ranging from 0.1 kGy to 0.6 kGy is effective in suppressing Salmonella enterica, Listeria monocytogenes or Escherichia coli in hummus [17]. Treatment with gamma radiation doses of 1.5 kGy and 2.5 kGy ensures the extension of the best before date of the hummus packed in laminated bags when stored at a temperature of +4°C and compliance with microbiological indicators [26]. Irradiation of nuts used as the raw material with low doses (< 5 kGy) is effective in destroying pathogenic microorganisms [22].

The open information base of literary sources does not contain information concerning the methods of destroying the microbial environment in urbech. However, problems with microbiological contamination are similar for nuts, hummus and tahini.

One of the effective ways to inactivate pathogenic microflora is to treat the finished product with different doses of gamma radiation, which has been permitted for using in the Russian Federation since 2017 for a number of food products.

Ionizing radiation is used in the food industry to inactivate microorganisms in food products [3].

**Problematics**

Nowadays, in addition to traditional urbech recipes, manufacturers try to expand their range by introducing new ingredients. The introduction of new raw materials requires the comprehensive study of the combined effects of different raw materials and production technologies. Various types of food products are produced based on coconut paste, but coconut in combination with honey is subject to quick spoilage, which is manifested in the coconut fat rancidity. But when combined with the date syrup and cocoa beans, peanut butter becomes sour and rancid. Sesame paste - tahini in combination with honey becomes sour after 2-3 weeks. In case of further storage, acidity increases. It is known that honey contains various enzymes from the bees` digestive juice. Grated cocoa beans may contain lipase, which is formed as a result of the fruit fermentation for their better separation. Dried mulberries may contain various microorganisms on the surface that survive even after heat treatment. At the same time, the enzymatic activity of microorganisms is very high.

In scientific research works in the public domain, there is no work concerning variable approach to selecting the effective method for preventing microbial spoilage of the finished product - urbech, which has determined the *purpose of the experimental research* that is to assess the effect of different doses of ionizing radiation on the microbiological parameters of some types of urbech, in particular on the viability of osmoresistant microorganisms. At the same time, the authors show the sequence of searching the effective way to prevent spoilage of urbech by studying the influence of various processing methods (by antioxidants, preservatives or radiation) - on microbiological parameters changes.

**Materials and methods**

The research objects include 3 types of urbech: the urbech from coconut pulp with the addition of dried mulberries (CM), the urbech from blanched peanuts with the addition of grated cocoa beans and date syrup (PCD) and the urbech from white sesame seeds with honey (SH). For urbech production, raw materials from different manufacturers have been used.

At the first stage, antioxidants were added to the finished urbech during its storage in order to prevent fats oxidation and to prevent lipases activation, the one of microbiological origin in particular. So, the drugs were administered in the maximum recommended dosage in accordance with the specifications in terms of the fat amount in the product under study. The enzyme preparation Long Way (0.15%) from the company "Grein Ingredient", tocopherol (1.5%), preparations Antioxidine (1.0%) and Unicons Eco (2.0%), sodium erythorbate

(1.0%), butyloxytoluene (0.1%), butylhydroxyanisole (0.5%) from the Unicons Group were tested.

At the second stage, to suppress microflora in urbech, preservatives were used. Manufacturers classify the used preservatives as “a clean label”: Deztin (1.0 %), Biatis (0.2 %), Milekons (0.7 %), Glycyrfit (2.0 %), Lysozyme (0.5 %) and Polybiome (0.5 %). The preservatives effect in urbech was tested in the thermostat. For this purpose, a preservative in the maximum recommended dosage was added to the experimental samples of urbech weighing 600 g. Then it was mixed and put in the thermostat at a temperature of +40°C.

At the third stage, the finished urbech was treated with different doses of gamma radiation at OOO RCOT “Era” using the RTU-3000 installation. Gamma irradiation source is 60 Co, its type is GIK-A6. The process of controlling the γ-radiation treatment was carried out automatically using the computer technology. Operational monitoring of the absorbed dose was carried out using CVID (manufactured by state scientific centre VNIIFTRI).

Choosing doses of ionizing radiation is determined by the results of studies made by WHO, which confirm the safety of an absorbed dose of ionizing radiation up to 10 kGy [10, 12] and the same is proved by a number of researchers [1; 5; 9].

In each species group of urbech samples (CM, PCD and SH), the researchers have formed control and experimental groups depending on the irradiation intensity (Table 1).

*Table 1.*

**Symbol of control and experimental groups of urbech treated with different doses of gamma radiation**

| Dose of Radiation, kGy | Species Groups | | |
|---|---|---|---|
| | CM | PCD | SH |
| Control Groups | | | |
| 0 | CM-0 | PCD-0 | SH-0 |
| Experimental Groups | | | |
| 2; 4; 6; 8; 10 | CM-2; CM-4; CM-6; CM-8; CM-10 | PCD-2; PCD-4; PCD-6; PCD-8; PCD-10 | SH-2; SH-4; SH-6; SH-8; SH-10 |

(compiled by the authors)

Microbiological tests were carried out in an accredited laboratory in accordance with GOST 31659-2012 to identify pathogenic microorganisms, including salmonella in 25 g; QMAFAnM - according to GOST 10444.15-94 article 6; coli bacteria (coliform bacteria) - according to GOST 31747-2012; the amount of yeast and moldy fungi - according to GOST 1044.12-2013 article 9; osmophilic microflora and yeast - according to GOST ISO 21527-2-2013.

The pH of urbech samples was measured using a pH meter 150M. The urbech viscosity was measured using a rotational viscometer at a temperature of 28°C. Humidity in accordance with GOST 5900-2014, acid number of fat - according to GOST R 50457-92.

Experimental studies**.** The urbech production scheme: rawmaterial purification; roasting of seeds and nuts (105-120°C); cooling of seeds and nuts (20-25°C); grinding seeds and nuts on stone millstones (less than 0.1 mm); tempering honey, cocoa beans (40-43°C); mixing of ingredients (3-5 minutes); packaging and capping.

After the initial tasting assessment, the nutritional value in the control samples of each species group was calculated in accordance with the standard methodology for defining the nutritional and biological value of food products [13] (Table 2).

*Table 2.*

**Urbech nutritional value, 1g/100g of the product (p≤0,05)**

| Product name | Protein, g | Fat, g | Carbohydrates, g | kcal |
|---|---|---|---|---|
| 1st species group – Urbech SH-0-0 | 12.5±0.4 | 33.6±0.6 | 35.8±0.3 | 495 |
| 2st species group – Urbech CM-0 | 12.0±0.4 | 52.5±0.8 | 24.2±0.2 | 617 |
| 3st species group – Urbech PCD-0 | 22.0±0.5 | 42.2±0.5 | 17.6±0.1 | 538 |

(compiled by the authors)

In the control samples of the 1st species group of the urbech (SH-0) the highest carbohydrate content was found, that was equal up to 35.8±0.3 g/100g compared with the samples of the 2nd species group of the urbech (CM-0) and the 3rd species group of the urbech (PCD-0). This fact can be explained by the addition of honey to the urbech composition. In the samples of the 2nd (CM) and 3rd (PCD) species groups, there was the increased fat content up to 52.5±0.8 g/100g and protein -up to 22.0±0.5 g/100g, respectively, due to introducing the raw material component of coconut and peanuts into the urbech recipe.

Traditionally, urbech is not subjected to heat treatment and it is stored in terms of positive temperatures no higher than +25°C for up to 1 year. The control samples of urbech with developed unconventional recipes (SH-0, CM-0, PCD-0) were placed under provocative background conditions for up to 30 days: temperature +40°C, humidity 75 %, exposure for 30 days, dark place. The control samples of Urbech SH-0 and CM-0 were stored for up to 5 days without changing the primary signs of organoleptic indicators, the control samples of Urbech PCD-0 were stored for up to 7 days.

**Research results and discussions**

Humidity determination has shown that in the control samples of Urbech type SH-0 the moisture content is 5.1±0.3 %, in the control samples of Urbech type CM-0 it is 4.8±0.3 %, in the control samples of type SH-0 it is 5.1 %, in the control samples of Urbech type PCD-0 it is 5.3±0.3 %. At this moisture content, microbiological activity is minimal.

At the first stage of the study, the antioxidants influence on the urbech preservation was studied based on the results of organoleptic assessment. The addition of sodium erythorbate, butyloxytoluene and butyloxyanisol in the maximum recommended dosages led to a slight increase in the best before date to 7 days in the urbech SH and CM; and up to 14 days in the urbech PCD. During its keeping, it was found out that the urbech samples deteriorated due to these antioxidants presence, which resulted in sourness in the urbech (SH); in rancidity in the urbech CM; and in sourness and bitterness in the urbech PCD. At the same time, the presented antioxidants are successfully used on an industrial scale in other food products. Accordingly, the most probable assumptions are that the reasons for the studied urbech samples spoilage are not due to the fats rancidity as a result of their oxidation. Microbiological indicators in accordance with the regulated standards TR CU 021/2011 in all the samples of urbech.

At the next stage, the preservatives effect (rowan extract, rosemary extract, lingonberry extract, glycyrrhite, lysozyme) in urbech samples was studied. The first signs of spoilage began to appear after 7 days in the urbech CM (rancidity), and after 14 days - in the urbech SH and PCD - the appearance of sourness and sourness with bitterness, respectively. When adding the Polybiome preservative, the best before date of the urbech CM and the urbech SH and PCD increased to 21-28 calendar days, respectively. For more effective search of pathogenic microflora at this stage, microbiological studies were carried out in the urbech with obvious signs of spoilage in the form of decomposition odors and rancid taste after 21 days (Table 3).

*Table 3.*

**Results of microbiological studies of the urbech samples with the Polybiome preservative addition**

| Indicators | Standard according to TR CU 021/2011 | CM | PCD | SH |
|---|---|---|---|---|
| Pathogenic microorganisms, including salmonella in 25 g | Not allowed | Not detected | Not detected | Not detected |
| QMAFAnM, CFU/g (cm3) | No more $5 \cdot 10^3$ | $1,3 \cdot 10^2$ | $4,4 \cdot 10^1$ | $3,8 \cdot 10^2$ |

| | | | | |
|---|---|---|---|---|
| Coliforms, not allowed in the product weight, g | 0,01 | Not detected | Not detected | Not detected |
| Mold, CFU/g (cm3), no more | 100 | 11…18 | 10…14 | 10…17 |
| Yeast, CFU/g (cm3), no more | 50 | 5…8 | 5…8 | 5…8 |

(compiled by the authors)

The research results have shown that in all the samples of the urbech with the preservative Polybiome addition, despite obvious signs of spoilage in organoleptic indicators, microbiological indicators correspond to the regulated indicators in accordance with the requirements of TR CU 021/2011. It has been assumed that the studied urbech samples contain osmoresistant, non-gas-forming microflora, which can survive at a substrate humidity of about 5 %.

At the third stage of the research, the reseachers decided to treat the studied urbech samples with different doses of gamma radiation (according to the data in Table 1) and to perform the detailed study of microbiological safety indicators, including indicators regulated by TR CU 021/2011, as well as osmophilic microorganisms. This approach depends on the fact that when studying microbiological safety indicators using standard methods, undesirable microflora is not detected, and at the same time, the unsuitability signs for human consumption have been established in the urbech: the taste and smell of spoiled products.

The organoleptic properties (colour, smell) have been studied in the control samples of the urbech (SH-0, PCD-0, CM-0), not treated with ionizing radiation, and in the experimental samples of the urbech species groups (SH, PCD, CM), treated with different doses of ionizing gamma radiation: 2, 4, 6, 8 and 10 kGy (Tables 4 and 5).

*Table 4.*

**The influence of treating the urbech with different doses of gamma radiation on its smell**

| Radiation dose, kGy | Samples the urbech | | |
|---|---|---|---|
| | CM | PCD | SH |
| 0 | Coconut, sweetish | Peanut chocolate | Sesame-honey |
| 2 | | | |
| 4, 6, 8, 10 | Unusual for the urbech species | Unusual for the urbech species, faint smell of cocoa | Unusual for the urbech species, with bitterness |

(compiled by the authors)

It has been experimentally established that the odor of the urbech of the experimental groups changes to the unusual odor for a certain type of urbech when it`s treated with doses of 4 kGy and higher

*Table 5.*

**The influence of treating the urbech with different doses of gamma radiation on its taste**

| Radiation dose, kGy | Samples the urbech | | |
|---|---|---|---|
| | CM | PCD | SH |
| 0 | Coconut-mulberry | Peanut with chocolate, sweet | Sesame-honey, sweet |
| 2 | | | |
| 4 | Unusual for the urbech species | Peanut with chocolate, sweet with slight bitterness. Unusual for the urbech species | Sesame-honey, sweet with slight bitterness. Unusual for the urbech species, poor taste |
| 6, 8, 10 | Unusual for the urbech species | Unusual for the urbech species. One can taste only cocoa | Sesame, poor taste |

(compiled by the authors)

With an increase in the gamma radiation dose, the urbech taste has changed, becoming poor, unusual, and weakly expressed. It has been established that the permissible dose of gamma radiation, which does not affect the change in taste, for all the groups of urbech experimental samples is 2 kGy; with an increase in dose of 4 kGy and above, a change in the urbech taste is noted.

Next, the urbech color of control and experimental samples (SH, PCD, CM) has been studied (Figures 1-3).

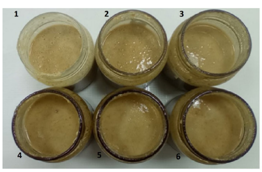


**Figure 1.** Urbech samples from sesame and honey (SH)


**Figure 2.** Urbech samples made from peanuts, cocoa beans and date syrup (PCD)


**Figure 3**. Urbech samples from coconut and mulberry (CM)

1 – the control sample (0 kGy), 2 – the experimental sample of the urbech, treated with a dose of 2 kGy, 3 – the experimental sample of the urbech, treated with a dose of 4 kGy, 4 – the sample of the urbech, treated with a dose of 6 kGy, 5 – the sample of the urbech, treated with a dose of 8 kGy, 6 – the sample of the urbech, treated with a dose of 10 kGy

The samples of the light-colored urbech (SH and CM) show that as a result of processing, the urbech has darkened. So, when irradiated with 2 kGy, the mass has darkened by half a tone in comparison with the control sample, which does not worsen the urbech appearance. It has been established that every subsequent 2 kGy darkens urbech by half a tone in comparison with the previous version. In dark urbech samples (PCD), slight darkening can also be noted.

In addition to organoleptic studies, the urbech viscosity has been studied (Table 6).

*Table 6.*

**The influence of treating the urbech with different doses of gamma radiation on its viscosity, conventional units of the device**

| Radiation dose, kGy | Samples the urbech | | |
|---|---|---|---|
| | CM | PCD | SH |
| 0 | 20±1.2 | 30±1.6 | 35±1.5 |
| 2 | 21±1.4 | 34±1.7 | 41±1.4 |
| 4 | 22±1.3 | 37±1.8 | 46±1.6 |
| 6 | 20±1.5 | 42±1.9 | 53±2.1 |
| 8 | 22±1.4 | 45±1.4 | 60±2.4 |
| 10 | 23±1.4 | 48±1.8 | 77±2.5 |

The study of changes in the urbech viscosity of experimental groups as a result of treating them with different doses of gamma radiation has shown that the urbech viscosity of experimental groups CM has remained practically unchanged (from 20±1.2 to 23±1.4 conventional units of the device). In the urbech samples of the PCD experimental groups, the viscosity has increased 1.6 times, these samples have been treated with a dose of 10 kGy compared to the untreated samples. The increase in thickening has been approximately 3-5 conventional units of the device for every 2 kGy of exposure. The samples of the experimental groups of the urbech SH has turned out to be the most resistant to irradiation. The increase in viscosity when treated with a dose of ionizing radiation of 10 kGy has been about 71 %. The viscosity of the urbech with a maximum irradiation dose of 10 kGy - 77 conventional units of a device.

At the intermediate stage, it can be noted that when urbech is treated with doses up to 2 kGy inclusive, in all the tested samples CM-2, PCD-2 and SH-2, the organoleptic characteristics and viscosity have remained unchanged compared to the control samples of the urbech CM-0, PCD-0 and SH-0 for up to 30-40 days.

The organoleptic assessment data is confirmed by the data obtained when studying the acid number of urbech fat of control and experimental samples of the urbech SH, PCD and CM (Figure 4).

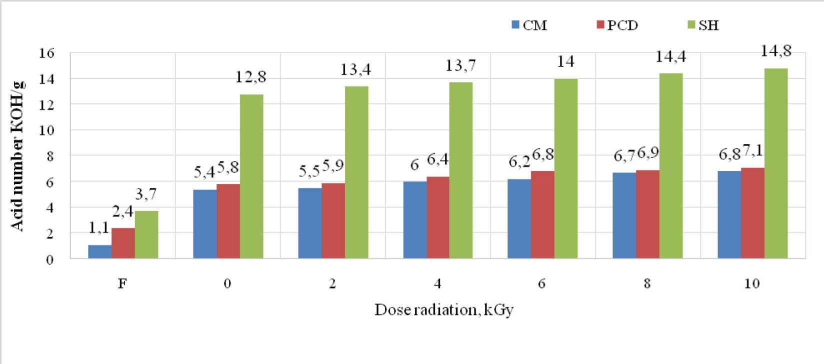


**Figure 4**. The influence of treating the urbech with different doses of gamma radiation on the acid number (AN) of urbech samples (F - freshly prepared Urbech samples; 0 kGy, 2 kGy, 4 kGy, 6 kGy, 8 kGy, 10 kGy - Urbech samples treated with different doses of radiation , CM, PCD, SH during their storage over 100 days)

Figure 4 shows that freshly prepared samples (F) have the minimum acid number (AN); when stored for more than 100 days, the acid number has increased in the control and tested samples of urbech. The AN of the urbech SH and the urbech PCD (0 kGy and 2 kGy) during their storage for over 100 days is comparable to the AN of the second class sunflower oil. In the samples of the urbech CM, the acid number has a high value of 3.7 immediately after the urbech production and subsequently during their storage for more than 100 days such urbech is not suitable for consumption.

Due to the fact that the urbech (after 100 days after treatment with different doses of gamma radiation and storage in room conditions) has had signs of spoilage, expressed in the unpleasant odor and taste and a high acid number, microbiological studies have been carried out. All indicators meet the requirements of TR CU 021/2011. No pathogenic microorganisms have been found in all the samples, including salmonella, and coliforms; QMAFAnM in the experimental samples of the urbechs SH, PCD and CM, depending on the radiation dose, has been in the range of 1.3·101 - 2.8·103 CFU/g, 1.1·101 - 7.5·101 CFU/g and 3.1·102 - 5.2·102 CFU/g accordingly; mold - in the experimental samples of the urbechs SH, PCD and CM, depending on the radiation dose, has been in the range of 8 - 15 CFU/g, 7 - 15 CFU/g, and 11 - 19 CFU/g G accordingly; yeast - in the experimental samples of the urbechs SH, PCD and CM, depending on the radiation dose, has been within 3 - 4 CFU/g, 3 - 5 CFU/g and CFU/g G accordingly.

Since organoleptic signs of spoilage have been established previously in the samples of the experimental groups of the urbech SH, PCD and CM, and at the same time pathogenic microorganisms have not been identified using standard methods for food products, additional studies have been carried out to identify microorganisms not regulated by the relevant technical regulations, in particular to identify osmophilic microflora (Table 8).

*Table 8.*

**The influence of treating the urbech with different doses of gamma radiation on the amount of osmophilic yeast in it, CFU/g**

| Radiation dose, kGy | Samples the urbech | | |
|---|---|---|---|
| | CM | PCD | SH |
| 0 | 310 | 521 | Not detected |
| 2 | 150 | 178 | Not detected |
| 4 | 133 | 98 | Not detected |
| 6 | 117 | 90 | Not detected |
| 8 | 106 | 84 | Not detected |
| 10 | 80 | 77 | Not detected |

In the Russian Federation, microbiological safety requirements for food products are not regulated in terms of rationing the amount of osmophilic yeast, including in honey and the products with low water activity. The permissible amount of these microorganisms in honey is regulated in the standard NMX-036-NORMEX-2006 "ALIMENTOS-MIEL-ESPECIFICACIONES Y METODOS DE PRUEBA" (Food. Honey. Technical characteristics and testing methods) (Mexico) and it is no more than 100 CFU/g . In accordance with the Chinese standard GB 14963 - 2011 "State Food Safety Standard. Honey" the amount of osmophilic yeast is allowed no more than 200 CFU/g. Consequently, we can focus on foreign standards and determine the safe amount of osmophilic yeast in urbech as the products with low water content and high osmotic pressure up to 100 - 200 CFU/g.

Osmophilic yeasts have not been found in the control and experimental samples of the urbech SH. The experimental samples of the urbech CM after treatment with gamma radiation contain 80...150 CFU/g of osmophilic yeast. In the experimental samples of the urbech PCD, the number of osmophilic yeasts is 100 CFU/g or less, only at the irradiation dose of 4 kGy or more. The reduction in the number of osmophilic yeasts in the samples of urbech CM treated with a dose of 10 kGy of gamma radiation has been 3.9 times compared to the untreated samples, in the samples of the PCD – 6.8 times, respectively. Consequently, treatment of the urbech of different composition with the doses of gamma radiation from 2 kGy to 10 kGy is effective in ensuring microbiological safety in relation to osmophilic yeasts.

After 100 days of storage of the different types of urbech pH value haws been determined as an indicator of quality and safety (Figure 5).

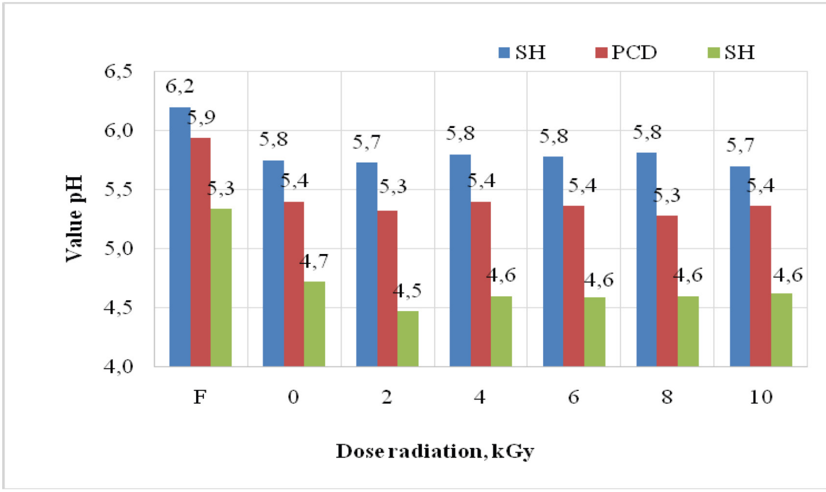


**Figure 5.** The influence of treating the urbech with different doses of gamma radiation on its pH (F - freshly prepared urbech samples; 0 kGy, 2 kGy, 4 kGy, 6 kGy, 8 kGy, 10 kGy – the urbech samples treated with different doses of radiation during their storage over 100 days)

In all the urbech samples the pH value has decreased compared to freshly prepared urbech (F) after irradiation. Thus, in the experimental samples of the urbech CM, the pH decreases from 6.2 to 5.8 - 5.7; in the tested samples PCD - from 5.9 to 5.4 - 5.3; in the tested samples SH - from 5.3 to 4.72 - 4.8. These values are stable in all the experimental samples of the corresponding species, but they are lower compared to the freshly prepared urbech.

**Conclusions**

In the process of storing the newly developed urbech types the auhors have noted the fact of rapid spoilage (up to 5 - 7 days) for the urbech from coconut pulp with the addition of dried mulberries (CM), from blanched peanuts with the addition of grated cocoa beans and date syrup (PCD) and from white sesame seeds with honey (SH). It has been experimentally revealed that the cause of the urbech damage has not been the processes of natural fat oxidation. Consequently, the antioxidants and preservatives of various groups cannot slow down the spoilage process. At the same time, the effect of the preservative - the drug Polybiom - has given a prolonged effect up to 21-28 calendar days to prevent the urbech spoilage. The humidity of the studied urbech species has been in the range of 4.8-5.3%, which is an important factor in the microorganisms inactivation and ensuring the requirements of technical regulations for microbiological safety. Thus, with the standard methods for detecting microorganisms, urbech contains 5 – 8 CFU/g of yeast cells, 10 – 18 CFU/g of molds and 4.4·101 – 3.8·102 of QMAFAnM at a rate of 50 CFU/g, 100 CFU/g and no more than 5·103 CFU/g, respectively, no pathogenic microorganisms, including salmonella, have been detected.

One of the effective ways to ensure the urbech microbiological safety is treatment with different doses of gamma radiation. The urbech tested samples have been treated with the appropriate doses: 2 kGy, 4 kGy, 6 kGy, 8 kGy and 10 kGy. When the dose increases above 2 kGy, an unusual smell and odor appear in the rbech samples, which determines the need to limit the radiation dose to 2 kGy. The organoleptic assessment data is confirmed by the data obtained from studying the acid number of urbech fat during its storage period of up to 30-40 days. The viscosity of the urbech CM, treated with different doses of radiation, has not practically changed, in contrast to the samples of the urbech PCD and the samples of the urbech SH. In the experimental samples of the urbech, as a result of treatment with ionizing radiation, after 100 days all the regulated indicators in the tested samples are within the regulated indicators, in contrast to the control samples: QMAFAnM - 5.2·103 CFU/g, mold - 102 CFU/g, yeast - 54 CFU/g. The study of the species composition of osmo-resistant microflora, not regulated by technical regulations in the Russian Federation, has shown that urbech contains osmo-resistant yeast in the amount of 310 and 521 CFU/g in the control samples of the urbech CM and the urbech PCD, respectively, and they are absent in the samples of the urbech SH. The samples of the experimental group of the urbech CM after treatment with gamma radiation have contained 110...150 CFU/g of osmophilic yeast. In the urbech PCD the amount of osmophilic yeast of 100 CFU/g or less has been observed only at irradiation doses of 4 kGy and 6 kGy. The decrease in the number of osmoresistant yeasts in the urbech samples treated with gamma radiation has occurred by 10...55 times. Therefore, the use of gamma radiation is effective in reducing the osmophilic yeast population.

Thus, the treatment with ionizing radiation at the doses up to 2 kGy is effective for preserving organoleptic and microbiological parameters, including osmophilic yeast, which is not regulated by the regulatory documents in the Russian Federation, for a certain period of storage. To study the causes of the urbech spoilage, further detailed studies of microorganisms and their metabolic products are required, as well as studying the storage conditions and possible changes in technology and recipe composition with the exclusion of ingredients with high humidity.

**Conflict of interest information.** The authors declare no conflict of interest.
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**Elena S. Lybenko**: conducted theoretical research, participated in experimental studies and in drawing conclusions based on the research results.

**Sergey O. Nikitin**: organized and participated in experimental research on radiation treatment and subsequent analysis of research results.

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