UDC 641.13:664.87:355.65:579:621.796

MICROBIOTA OF INSTANT CEREALS AND ITS CHANGE DURING STORAGE

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Abstract. The paper presents the results of the study of changes in the microbiological parameters of instant cereals for military personnel during storage. The purpose of the microbiological studies of instant cereals was assessing whether the products were safe for human health immediately after manufacturing and during their further storage at a certain temperature and in various packaging. Special attention was paid to detecting certain microorganisms: pathogenic (including Salmonella), opportunistic pathogenic, sanitary indicator microorganisms (coliform bacteria), some microbial spoilage agents (yeast and mould fungi). Various types of packaging materials having been analysed, it has been studied how flexible packaging, foil-sealed plastic cups, and metallised packages of biaxially oriented propylene film (BOPF), GM-200, change the consumer properties of new instant cereals during storage. The studies have helped establish that natural enriching supplements of plant and animal origin in the composition of new instant cereals prolong the storage of finished products. It has been established that enriching supplements prevent the development of microbiota due to a bactericidal effect of supplements on microorganisms. Studying how packaging changes the consumer properties of new instant cereals during storage has shown that bacteria decreased in number in all samples during storage, regardless of the type of packaging. However, the most significant decrease (by an average of 1.5 times) was observed when metallised BOPF bags were used. Based on the experimental studies, it has been found that instant cereals packed in metallised BOPF bags belong to long-term storage products and can be stored for 12 months at room temperature and the relative humidity 60–75%.

Key words: microbiota, instant cereals, military, storage.

МІКРОБІОТА КАШ МИТТЄВОГО ПРИГОТУВАННЯ ТА ЇЇ ЗМІНА ПРИ ЗБЕРІГАННІ

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Анотація. У статті наведено результати дослідження зміни мікробіологічних показників каш миттєвого приготування для військовослужбовців у процесі зберігання. Мікробіологічні дослідження каш миттєвого приготування проводили з метою оцінки безпечності продукції для здоров'я людини, безпосередньо після виготовлення і в процесі їхнього подальшого зберігання за відповідних температурних режимів і в різних упаковках. Особливу увагу приділено визначенню наявності патогенних мікроорганізмів, у тому числі сальмонел, умовно-патогенних, санітарно-показникових мікроорганізмів (БГКП), окремих збудників мікробного псування продукту (дріжджів та пліснявих грибів). Проаналізовано різні види пакувальних матеріалів, у результаті обрано та досліджено вплив гнучкої упаковки, пластикових стаканчиків запаяних фольгою і металізованих пакетів із біоксальноорієнтованої пропіленової плівки (БОПП) GM-200, зміну споживних властивостей нових каш миттєвого приготування у процесі зберігання. На основі досліджень встановлено, що включення до складу нових каш миттєвого приготування збагачувальних натуральних добавок рослинного та тваринного походження, дозволяє подовжити строки зберігання готової продукції. Встановлено, що збагачувальні добавки перешкоджають розвитку мікробіоти, що пов'язано з бактерицидною дією добавок. Дослідження щодо впливу пакування на зміну споживних властивостей нових каш миттєвого приготування у процесі зберігання показало, що у всіх досліджуваних зразках, незалежно від виду пакування, кількість бактерій зменшувалася. Але, найбільш значне зниження спостерігалося при використанні металізованих пакетів із БОПП у середньому в 1,5 разів. На основі досліджень встановлено, що каші миттєвого приготування, упаковані у металізовані пакети із БОПП відносяться до продуктів тривалого зберігання і можуть зберігатися протягом 12 місяців при кімнатній температурі та відносної вологості повітря 60–75%.

Ключові слова: мікробіота, каші миттєвого приготування, військовослужбовці, зберігання.

DOI: http://dx.doi.org/10.15673/fst.v13i1.1338

Introduction. Formulation of the problem

Consumer properties of a food product formed during its production should be kept in the same qualitative and quantitative proportion until the product is consumed, so storage is quite an important stage of the life cycle of products. During storage, a number of different processes take place in food: microbiological, biochemical, and others. In the case of inappropriate storage, these processes can result in a decrease in the consumer properties of products, particularly, in deterioration of its physical and chemical properties, as well as a decrease in nutritional value [1,2].

The environmental, social, and economic deterioration and the extreme conditions of work are affecting military personnel's health. That is why, there is a need for new food products (including instant cereals) with improved consumer properties based on natural ingredients. However, along with developing and manufacturing products with improved consumer properties, it is important to preserve these properties at all stages of product distribution.

Analysis of recent research and publications

Nowadays, people are more than ever interested in healthy nutrition, that is why they focus on products that can adequately meet human physiological needs. At present, a wide range of such dry breakfasts as instant cereals (in the form of grain flakes and their mixtures, muesli, puffed grain, granola, couscous, etc.) can be found in retail chains [3].

Cooking instant cereals takes five to ten minutes. Their production requires not whole grain, but flakes. The thinner the flakes, the faster the dish is cooked. The thickness of flakes for instant cereals usually does not exceed 0.5–1.0 mm [4].

Instant cereals do not require boiling. They have the form of a powdery mixture or flakes, usually as thick as 0.3–0.5 mm. A prerequisite for this type of cereal is hydrothermal treatment. Cereals are exposed to high temperature and pressure, then dried [5,6].

Extrusion helps to obtain a product with high sterility and a low mass fraction of moisture, which makes it possible to prolong the storage. However, the extended specific surface area and high hygroscopy adversely affect the product's quality parameters which can change during storage [7].

The microbiological characteristics of instant cereals are an important criterion for safety assessment and a mandatory indicator in the quality control system [8-9].

Some instant cereals can be a favourable environment for the existence and reproduction of microorganisms. Non-specific microbiota (microorganisms that accidentally entered instant cereals from the environment) makes products inedible or even harmful.

The development of microorganisms, that is, their growth rate, depends on the product's composition and properties, the environmental conditions, the proper selection of packaging, and compliance with sanitary and hygienic standards and with transportation and storage regulations. First, the growth rate of microorganisms depends on the moisture content of the product and the relative humidity of the air. Instant cereals are low humidity products, with the suppressed activity of microorganisms [10,11]. Under favourable conditions, such as high humidity, cereals can increase the content of free moisture thus giving rise to the development of bacteria and fungi.

Another factor affecting the growth and development of microorganisms is the presence or absence of oxygen. The dependence of the growth rate of microorganisms on the composition of the gas environment is widely used by applying vacuum packaging or packaging in an inert gas environment [12].

Recently, the maximum genuineness and naturalness, the absence of preservatives, flavouring agents and other additives have become the most important trends in the instant food market. Products companies bring to the market have new properties and cost more, but are within the consumer trend towards healthy nutrition [13].

The shelf life of instant cereals for military personnel can be increased by using modern packaging materials, which are a mandatory component of industrial development. Packaging materials, if used appropriately, help against mechanical damage, pollution, sunlight, and heat, prevent sticking and damping, preserve the taste and aroma, reduce losses, prolong the shelf life (twice or more), and make the product look bright and attractive [14,15].

Manufacturers impose the following requirements for instant cereal packaging: it should have high barrier properties (particularly, it should not allow access of oxygen, moisture, and ultraviolet rays), weigh little, protect products from foreign odours, extend the shelf life of products, and be an advertising medium of brand attributes [16,17].

It is packaging that promotes the product and, no doubt, significantly influences a consumer's preference of a product or a manufacturer. Besides, the packaging is not only an advertising means, but also a powerful tool for branding products and protecting them from counterfeiting. A unique package allows the

manufacturer to distinguish products among the similar ones, to make them recognisable and memorable [18].

The packaging is also an important source of information about the manufacturer, the product composition, the conditions and terms of storage and consumption. The domestic consumer is becoming more demanding and choosier, so this function is very important as well [18,19].

Most instant cereals are packed in portion packets, some are sold in boxes out of which a customer can take as much as he or she needs. The achievements of chemical industry in the field of polymeric materials are more and more widely used in packaging. Today, there are films that are easy to use and make it possible to introduce systems allowing instant cereals manufacturers to make polymer pads and even plastic bags, fill them, and heat-seal them simultaneously.

Nowadays, the main global trends in the development of packaging are its efficiency, environmental friendliness, convenience, and information value. The package of flexible multi-layer materials meets these demands best. Layers of polymer films, paper, metal foil, and other materials do not only make a package strong enough, but also protect from moisture, oxygen, gases, and foreign aromatic substances. Moreover, these materials have high fat resistance, are effectively heat-sealed, and reflect light and heat. Various methods of manufacturing flexible packaging films (extrusion layering, co-extrusion of polymer melts, lamination) make it possible to obtain materials with predetermined properties [20-21].

An economical packaging means is corrugated cardboard boxes with an inner vacuum bag. This package protects the product from external impacts and mechanical damage. However, products after unpacking have a short shelf life, quickly absorbs moisture, thus losing its consumer qualities [22].

Polypropylene bags are resistant to fats and temperatures, environmentally safe, but very sensitive to moisture [23]. Therefore, instant cereals packed in polypropylene packages should be stored in rooms with low relative humidity of the air, which is not always possible when selling the products.

Foil-sealed plastic cups protect instant cereals from mechanical damage and exposure to light quite well, and foil is good against pollution, as it makes the product fat and water resistant [22].

Metallised packages of biaxially oriented polypropylene film (GM-200, GM-201, GM-210, GM-212, 20–35 μm thick) are widely used for instant cereals. This film has a number of advantages: it does not change the taste of products, does not cause mutual migration of the material and the product components, prevents the ingress of moisture, and protects products from sunlight. It should be noted that this film does not contain harmful additives and impurities, and its packaging waste is easily disposed [20].

Packaging materials for new instant cereals were selected taking into account their protective properties, the cost, and the possibility of use with these products.

After analysing various types of packaging materials and learning their properties, we selected flexible packaging, foil-sealed plastic cups, and metallised packages of biaxially oriented polypropylene film (BOPF), GM-200, and studied how they changed the consumer properties of new instant cereals during storage.

During the storage of instant cereals, various processes take place in them, which lead to deterioration of quality. At the same time, the quantitative characteristics of microbiota change as well. These phenomena cause the microbial spoilage of instant cereals.

The **purpose** of the study is to learn the microbiota of instant cereals for military personnel since the rate of microbiological processes and the intensity of accumulation of pathogenic microorganisms are among the critical factors that regulate the shelf life of food and make it consumable.

To achieve this goal, the following **objectives** were set:

1. to analyse the materials used for instant cereal packaging;
2. to study how the microbiological parameters of instant cereals for military personnel change depending on the conditions and terms of storage;
3. to determine the ideal shelf life of instant cereals.

Research materials and methods

The objects of the study were instant cereals. A floury mixture of spelt and oats (80:20) and that of spelt and buckwheat (75:25) were selected as the main components of their recipe. To improve the consumer properties of the finished product (so that it was of the appropriate taste, aroma, and colour) and to enrich it with vitamins, mineral components, flavonoids, phytoncides, and glycosides, the following components were added to the formulation: pieces of dried apple, of dried blueberries, of dried chokeberry, of dried carrots, of dried onions, of dried Jerusalem artichoke, of dried celery, dried pumpkin powder, dried parsnip powder, dried horseradish root, dried parsley and dill, cinnamon, whey powder, stevioside, salt, and black pepper. For protein enrichment, a 10% mass fraction of dried beef pieces was added.

Thus, we obtained and studied six samples of instant cereals:

– sample 1 – instant cereal without supplements (spelt + oats in the ratio 80:20, the control);

– sample 2 – instant cereal with apple, cinnamon, and stevioside;

– sample 3 – instant cereal with the inclusion of blueberries and chokeberry;

– sample 4 – instant cereal without supplements (spelt + buckwheat in the ratio 75: 25, the control);

– sample 5 – instant cereal with beef meat, pieces of dried carrots, onions, parsnip powder, dried dill and parsley, and horseradish root;

– sample 6 – instant cereal with vegetables: pieces of dried Jerusalem artichoke, celery, carrots, onions, and parsnip powder.

After instant cereals are manufactured, the next important task is preserving their nutritional and biological value and delivering them to the consumer with no loss in quality.

One of the most important indicators of the quality of any food, especially when it is obtained with the addition of components of plant and animal origin, is its microbiological characteristics.

We have studied the qualitative and quantitative composition of instant cereal microbiota, and the changes in it depending on the composition, conditions, and duration of storage.

Instant cereals are supposed to be introduced into the army dry ration, thus their storage has been adjusted to field conditions. All samples studied were stored indoors, at a temperature of 16–27°C and the relative humidity 60–75%. The temperature and relative humidity varied depending on the season. The samples of instant cereals were studied prior to storage, and then, every 2 months of storage during 12 months.

The packaging used was:

– foil-sealed plastic cup;

– flexible packaging;

– metallised bag with biaxially oriented polypropylene film (BOPF), GM-200.

The qualitative and quantitative composition of microbiota was determined both by classic techniques and by using the modern microbiological rapid-response analyser BacTrac 4300 (Austria). Its work is based on registering the changes in the electrical resistance (impedance) of the culture medium that result from the activity of microorganisms. The main advantage of this method is facilitating a microbiologist's work and reducing the research time from 1–7 days (as it is with the classic methods) to 24 hours to determine mesophilic aerobic and facultative anaerobic microorganisms (QMAFAnM), and to 48 hours to determine micromycetes.

The samples were placed in germ-proof glassware under aseptic conditions, excluding microbial contamination of the samples from the environment. The composition of the microbiota in the samples was determined by microbiological and sanitary parameters, which include the quantity of mesophilic aerobic and facultative anaerobic microorganisms (QMAFAnM), of micromycetes (mould and yeast fungi), of coliform bacteria, with further determining potentially pathogenic Escherichia coli and Staphylococcus aureus, and pathogenic microorganisms, including Salmonella and sulphite-reducing clostridia. The determining was carried out by inoculating the microorganisms on special culture media with the subsequent cultivation and characterising by State Standard 10444.9, 10444.12, and 10444.15.

The total number of bacteria was determined by inoculating the wipe-samples of various degrees of dilution in meat-and-peptone agar, and mould and yeast fungi in wort agar, followed by cultivation at a temperature of 30±1°C for 24–48 hours, and 28±1°C for 5–7 days, respectively. Spore forms of bacteria were identified in pasteurised wipe-samples that had been inoculated on a complex nutrient medium of meat-and-peptone agar and wort agar in the ratio 1:1. To identify clostridia, the inoculation was carried out in liver broth and in the Kitt-Tarozzi medium; to identify Staphylococci, in the milk-salt agar; to identify Salmonella, in the bismuth sulphite agar; and to identify Proteus vulgaris, in the condensing water of freshly-cut meat-and-peptone agar. The presence of *E. coli* in the Kessler medium was determined by the turbidity of the medium, the appearance of gas bubbles in the floats, and changes in the colour of the nutrient medium, which are obligatory signs of acid and gas formation and are formed when these bacteria ferment sugars. Then, opportunistic pathogenic *Escherichia coli* was identified by transferring it to Endo's differential diagnostic nutrient medium. The cultivation was carried out at a temperature of 30±1°C for 24–48 hours.

Results of the research and their discussion

The results of the microbiological behaviour of the studied samples depending on the type of the sample are shown in Fig. 1.

The total quantity of bacteria in all samples taken as 100%, the contamination of each of them is 10.3% to 24.2%, depending on the type of the sample.

The diagram shows that the lowest microbial contamination level is observed in sample 2 (10.3%); the highest is 24.2% in sample 6.

The characteristics of the changes in instant cereals microbiota during storage vary depending on the composition and storage conditions (Fig. 2, a, b, c).

In all instant cereals controls (samples 1 and 4) stored in foil-sealed plastic cups, the total number of bacteria increased by 20.7– 26.7% during 12 months of storage, respectively. The increase was mainly in the coliform bacteria (by 2.5–3 times), while the spore-forming bacteria remained unchanged. There was almost no development of micromycetes, but a change in their species composition. For up to 12 months of storage, field fungi were not detected. The fungal microbiota in instant cereals was permanently represented by the fungi *Aspergillus* and *Penicillum*. During storage in a plastic cup, the proportion of micromycetes in the total amount of microorganisms increased slightly, mainly that of *Aspergillus* and *Penicillum*.

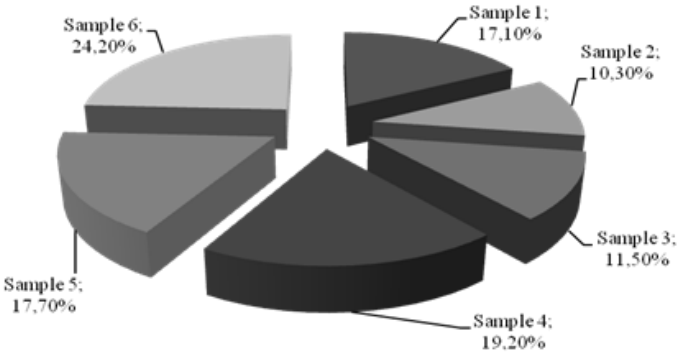

Fig. 1. Total bacterial contamination of instant cereals prior to the storage, % of the total microbiota of the samples

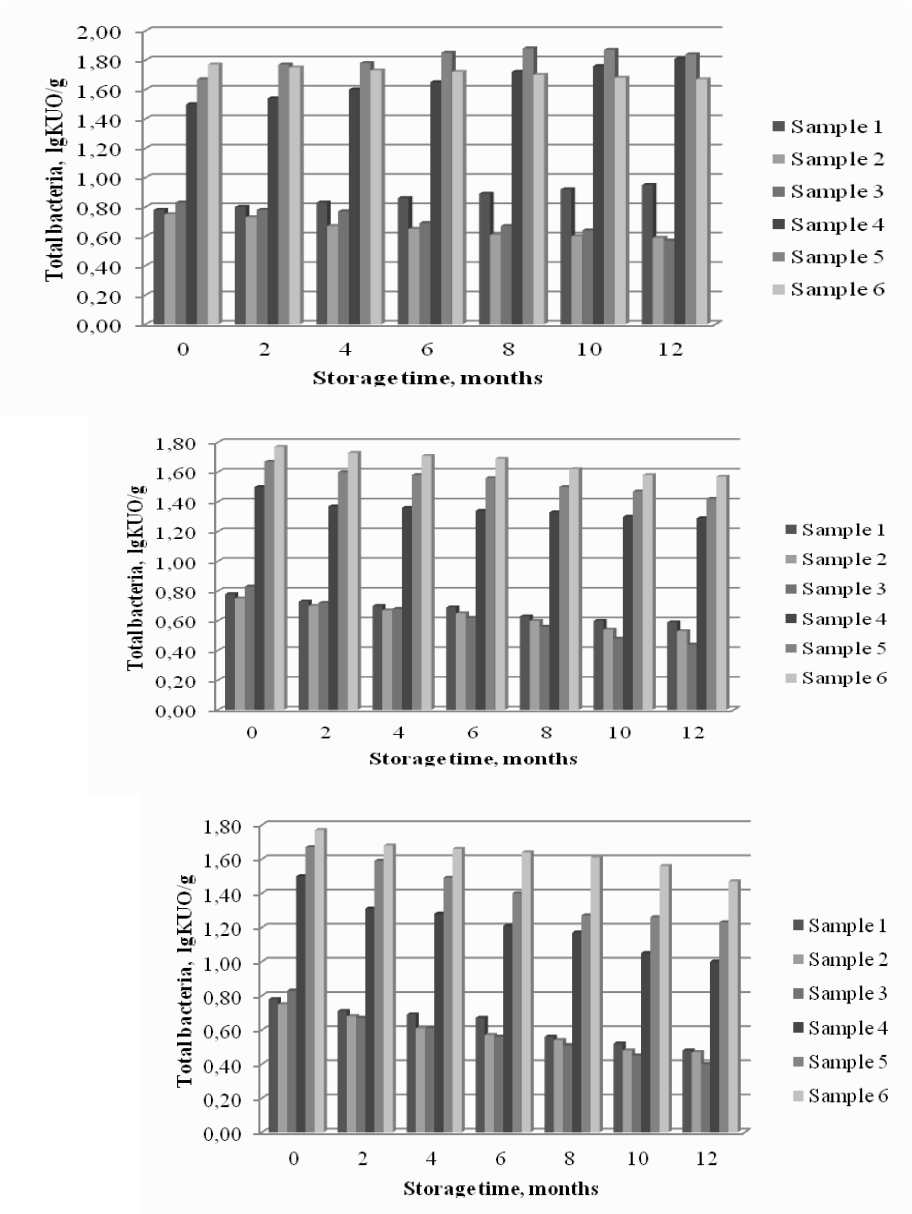

Fig. 2. The change in the quantity of mesophilic aerobic and facultative anaerobic microorganisms depending on the duration of storage of instant cereals: a) foil-sealed plastic cup; b) flexible packaging; and c) metallised BOPF bag

The characteristics of the changes in the microbiota of instant cereals with plant and animal supplements indicate that during storage, in all samples, regardless of the type of packaging, the number of bacteria decreased. The most significant decrease was observed when using metallised BOPF bags in sample 2 (1.6 times), sample 3 (2 times), sample 5 (1.3 times), and sample 6 (1.2 times) (Fig. 3).

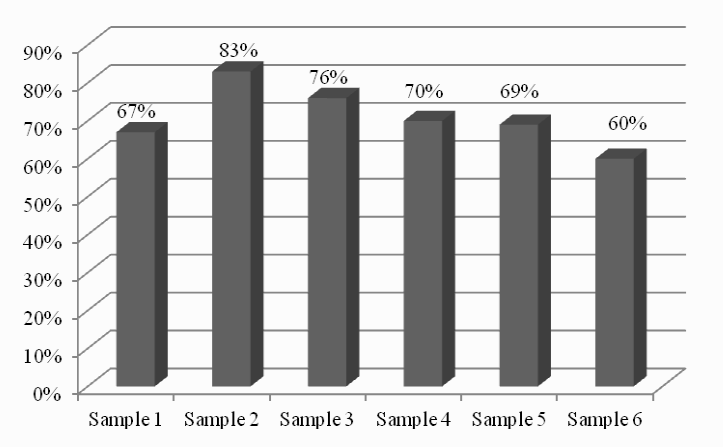

Fig. 3. Decrease in the microbial contamination level of instant cereals after 12 months of storage in metallised BOPF bags

The lowest is the quantity of mesophilic aerobic and facultative anaerobic microorganisms in instant cereals with apple and cinnamon (sample 2) and with berries (sample 3). This can be explained by the replacement of sugar with stevioside. Stevioside is known to have antiseptic and antifungal properties. It starts absorbing moisture at the relative atmospheric humidity 45%, thus spoilage of products with stevioside takes a longer time, and there is a decrease in microbiological parameters compared to sugar-containing products. As for the effect of root crops in the composition of instant cereals (samples 5, 6) on mesophilic aerobic and facultative anaerobic microorganisms, it can be noted that they also have antibacterial and antioxidant properties.

In all the studied samples, irrespective of the type of packaging and the inclusion of supplements of plant and animal origin, *E. coli, Salmonella,* sulphite-reducing *Clostridia, Staphylococcus aureus, and Proteus vulgaris* were not detected. The presence of bacteria and micromycetes was within regular limits. This indicates that the instant cereals were manufactured in proper sanitary and hygienic conditions.

In today's market conditions, the optimum shelf life of instant cereals adds much to their competitiveness. The use of new technological processes and modern packaging materials can protect finished products from bacterial spoilage and negative organoleptic changes, and to some extent prolong their shelf life.

Conclusion

1. Various types of packaging materials used by manufacturers for instant cereals have been analysed. The analysis has helped select the forms of packaging (foil-sealed plastic cups, flexible packaging, metallised packages of biaxially oriented polypropylene film, GM-200) and study how they influence the change in consumer properties of new instant cereals during storage.

2. It has been established that plant supplements in instant cereals reduce their contamination by microorganisms, which is due to the bactericidal action of the supplements on microorganisms. Thus, the number of bacteria decreased in all samples during storage, regardless of the type of packaging. The most significant decreases were observed when using metallised BOPF bags in sample 2 (1.6 times), sample 3 (2 times), sample 5 (1.3 times), and sample 6 (1.2 times). As for the effect of supplements, it has been found that the lowest is the quantity of mesophilic aerobic and facultative anaerobic microorganisms in instant cereals with apple and cinnamon (sample 2) and with berries (sample 3), which can be explained by the replacement of sugar with stevioside. When assessing the changes that root crops (samples 5, 6) cause in mesophilic aerobic and facultative anaerobic microorganisms, it can be noted that they also have antibacterial and antioxidant properties. The presence of bacteria and micromycetes is within regular limits.

3. Instant cereals with improved consumer properties, packed in flexible packaging and metallised bags of BOPF are long-term storage products and can be stored for 12 months at room temperature and the relative humidity 60–75%.

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Отримано в редакцію 22.09.2018 Received 22.09.2018
Прийнято до друку 06.03.2019 Approved 06.03.2019

Цитування згідно ДСТУ 8302:2015

Mardar M., Stateva M., Yegorova A., Evdokimova G., Ustenko I., Masanski S. Microbiota of instant cereals and its change during storage // Food science and technology. 2019. Vol. 13, Issue 1. P. 112-121. DOI: http://dx.doi.org/ 10.15673/fst.v13i1.1336

Cite as Vancuver ctyle citation

Mardar M, Stateva M, Yegorova A, Evdokimova G, Ustenko I, Masanski S. Microbiota of instant cereals and its change during storage. Food science and technology. 2019; 13(1): 112-121. DOI: http://dx.doi.org/ 10.15673/fst.v13i1.1336