UDC 001.8:577.114.7
DOI: 10.15587/2312-8372.2017.119420

**Dubinina A.,**
**Letuta T.,**
**Frolova T.,**
**Savinova H.,**
**Bolshakova G.,**
**Novikova V.**

# RESEARCH OF TOXICITY OF CHITOSAN-BASED FILM-FORMING COMPOSITIONS

*Із розглянутих композицій на основі хітозану та на основі аналізу характеристики лікарських відварів було обрано 20 видів для подальшого складання композиції з хітозаном 2 %. Для встановлення їх безпечного використання в плодоовочевій продукції всі зразки досліджені на токсичність та зони гемолізу. При проведенні досліджень отримані позитивні результати – не було виявлено токсичності та зони гемолізу.*

**Ключові слова:** *композиції на основі хітозана, відвари лікарських трав, токсичність композицій, зони гемолізу на відварах, кров'яний агар.*

## 1. Introduction

Food spoilage is inevitable – it's only a matter of time and the influence of external factors. The problem of preserving quality through the use of effective methods and storage technologies is especially relevant for perishable products that have a high moisture content, such as fruit and vegetable crops.

The main reason for the spoilage of fruits and vegetables during storage is the spread of microorganisms that cause various diseases. The development of pathogens can be prevented or slowed down by the creation and use of food packages that have a complex of antimicrobial properties – bactericidal and fungicidal.

Over the past decades, the evolution of technology has evolved from the large-scale storage of fruits and vegetables (under artificial cooling, a controlled gas environment) to small-party (in a modified gas environment, a biological vacuum) and individual. Expansion in the market has received «active» packaging of fruit and vegetable products, which, in addition to traditional mechanical and barrier protective functions, can have a targeted effect on packaged products, including antimicrobial ones. As a consequence, their storage periods are significantly increased.

Today, the problem of preserving fresh fruits and vegetables to ensure their population during the year can be solved by developing and introducing storage technology by treating them with film-forming solutions with antimicrobial properties.

## 2. The object of research and its technological audit

Chitosan (Fig. 1) is a high molecular weight glucosamine polymer derived from chitin. Chitosan is one of the most common polysaccharides. As the most common organic compound, it occupies the second position after cellulose.

The unique properties of chitosan and inexhaustible supplies of raw materials cause considerable interest in its production and practical application (Fig. 2) [1].

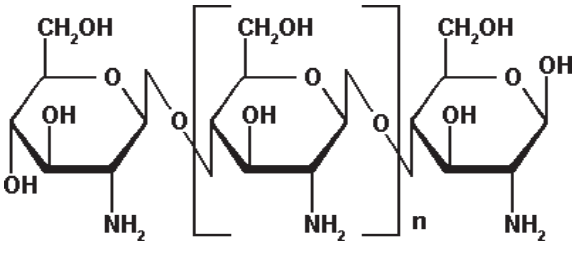


**Fig. 1.** Structural formula of chitosan

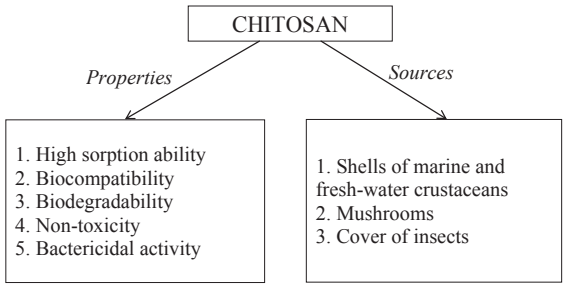


**Fig. 2.** Properties and sources of chitosan

To improve the antimicrobial properties of chitosan-based film-forming compositions, in order to enhance its preservative effect on microorganisms, it has been proposed to add decoctions from medicinal plants.

Medicinal herbs were chosen from those used in medicine as anti-inflammatory and wound-healing preparations, hence they have pronounced antibacterial (bactericidal and bacteriostatic) and antifungal (fungicidal and fungistatic) properties.

It is known that the antimicrobial effect of medicinal plants is due to their chemical composition, namely the content of basic BAS (biologically active substances) [2]. Based on the above factors, the broths of medicinal herbs were investigated. As *objects of the study*, 20 types of film-forming compositions based on chitosan and herbal decoctions were selected (Table 1).

ISSN 2226-3780

**Table 1**

Film-forming compositions based on chitosan and herbal decoctions

| No. | Name of herbal decoctions | Chemical composition | | |
|---|---|---|---|---|
| | | Biologically active substances | Macro and micro elements | Other |
| 1 | 2 | 3 | 4 | 5 |
| 1 | Sweet flag | Essential oil (up to 48 %): camphene, acarone, azarone, borneol; tannins; ascorbic acid (up to 150 mg); Acetic and valeric acids; phytoncides; slime; gum | Mn, Cu, Zn | Bitter glycoside of acorin (0.2 %); starch (up to 20 %); alkaloid calamine; resins |
| 2 | Senegalia catechu | Tannins: catechin, epicatechin and their dimers; flavonoids; essential oil; organic acids; slime; tryptamine alkaloids and DMT | Ca, P, Mg, Na, K, Cl, S, Fe, Zn, I, F | Glycosides; sugars; tannins |
| 3 | Ledum | Essential oil (aboveground part – 1.5 %, leaves at the flowering stage – 14.04–9.23 %): ice (6.09–8.87 %), palustrol, cymene, geranilacetate; tannins; flavonoids (quercitrin); organic acids; vitamins; gum; phytoncides | K, Ca, Mn, Fe, Mg, Cu, Zn, Al, Ba, Cr | Pectin substances; glycosides: arbutin, erivolin; glycosides; flavonoids; arbutin (20–23 %); sugar; coumarins; hydroquinone (2–5 %); resins |
| 4 | Bergenia crassifolia | Tannins of the group of gallotanins (tannin 8–12 %); gallic acid; ascorbic acid | K, Ca, Mn, Fe, Mg, Cu, Cr, Al, Pb | Arbutin; isocoumarin, flavonoids; hydroquinone; sugar; starch; phenols; ash; resins; carotenoids |
| 5 | Common yarrow | Essential oil (0.6–0.8 %): chamazulene, azulene, camphor, alpha- and beta-pinenes, borneol, cineole thujone, caryophyllene; esters, cineole; glycosides: apigenin and luteolin; tannins; resins; formic, isovaleric, ascorbic and acetic acids; vitamin K | Mg, Zn, Mo, Cr, Al, Se, Ni, Sr, Pb, B | Carotene; sesquiterpenes: millefolide, matrix; flavonoids: rutin, luteolin-7-glucoside; alkaloids: stachidrine, achillein, betonycin; resins |
| 6 | Eucalyptus globulus | Etheric (eucalyptus) oil (0.7–2 %); tannins | K (14.1 mg/g), Ca (16.1 mg/g), Mg (2.4 mg/g), Fe (0.3 mg/g), Mn (4.16 mg/g), Cu (0.81 mg/g), Zn (0.66 mg/g), Co (0.11 mg/g), Mo (0.27 mg/g), Cr (0.3 mg/g), Al (0.31 mg/g), Ba (0.68 mg/g), Se (7.5 mg/g), Ni (0.73 mg/g), Sr (1.34 mg/g), Pb (0.12–0.25 mg/g), B (7.6 mg/g) | Terpenic compounds; aldehydes; ketones; free and esterified alcohols; carbonyl compounds; resins |
| 7 | Eleutherococcus senticosus | Essential oil; alkaloid aramine; resin (up to 18 %); tannins; vitamins C, E | Mg, Cu, Zn, Co, Cr, Mo, Al, B, Ni, S, I, Ba, V, Se | Flavonoids; Coumarin derivatives; derivatives of flavonoids; glucose; sugar; starch; polysaccharides; wax; pectin substances; Eleutherosides A, B, C, D, E |
| 8 | Hypericum perforatum | Tannins (13 %); essential oil (0.1–1.25 %): α-pinene, myrcene, cineole, geraniol; resinous substances (17 %); vitamins P and PP; ascorbic acid; nicotinic acid | K (16.8 μg/g), Ca (7.3 μg/g), Mg (2.2 μg/g), Fe (0.11 μg/g), Mn (0.25 μg/g), Cu (0.34 μg/g), Zn (0.71 μg/g), Co (0.21 μg/g), Mo (5.6 μg/g), Cr (0.01 μg/g), Al (0.02 μg/g), Se (5.0 μg/g), Ni (0.18 μg/g), B (40.4 μg/g) | Hyperin; hypericin; Hyperoside (0.3–1.1 %); azulene; carotene; anthocyanins (up to 6 %); saponins; choline |
| 9 | Chamaenerion angustifolium | Tannins (10–20 %); vitamins A (180 μg), B1 (0.033 mg), B2 (0.137 mg), B3 (1.356 mg), B6 (0.632 mg), B9 (112 μg), C (2.2 mg), PP (4.674 mg ); organic acids | Ca (429 mg), Mg (156 mg), Na (34 mg), K (494 mg), P (108 mg); Fe (2.4 mg), Zn (2.66 mg), Cu (320 μg), Mn (6.704 mg), Se (0.9 μg) | Pectin |
| 10 | Iceland moss | Mucus (70 %); organic acids; carbohydrates; vitamins (A, B1, B12); fats; gum | I, Zn, Sn, Cd, Pb, Si | Enzymes; wax |
| 11 | Calendula officinalis | Carotenoids and flavonoids: carotene, lycopene, violaxanthin, citraxanthin, rubixanthin, flavoxanthin, flavochrome; mucus (2.5 %); nitrogen-containing mucus (1.5 %); organic acids: malic, ascorbic and traces of salicylic | K (28.80 μg/g), Ca (11.40 μg/g), Mg (2.50 μg/g), Fe (0.15 μg/g), Mn (0.20 μg/g), Cu (0.86 μg/g), Zn (1.31 μg/g), Co (0.03 μg/g), Mo (1.47 μg/g), Cr (0.09 μg/g), Al (0.05 μg/g), Se (4.20 μg/g), Ni (0.5 μg/g), Sr (0.10 μg/g), Pb (0.03 μg/g), I (0.05 μg/g), B (48.40 μg/g) | Resins (about 3.4 %); polysaccharides; polyphenols |
| 12 | Urtica | Tannins and protein substances; formic acid; carbohydrates; vitamins: ascorbic (up to 700 mg %), vitamin K; pantothenic acid; carotenoids (in fresh leaves 13–14 %, in dry leaves up to 50 mg %) | K, Mg, P, Fe, I, Cu, Cu, Mg | Urticin glycoside; chlorophyll (2–5 %); ash |
| 13 | Peppermint | Essential oil (2.5–4.5 %): menthol, α-pinene, limonene, cineole, dipentene, pulegone, β-phellandrene; ascorbic acid; organic acids; tannins | Cu, Mg, Sr | Rutin; carotene; flavonoids; betaine; hesperidinum |
| 14 | Potentilla alba | Phenolcarboxylic acids; tannins; flavonoids: rutin, cyanidine, kaempferol, quercetin | Zn, Mn, Si, Cu, Ce, Al, Fe, Co, I | Gallotanine; saponins; starch; iridoids |

**Continuation of Table 1**

| 1 | 2 | 3 | 4 | 5 |
|---|---|---|---|---|
| 15 | Common tansy | Essential oil; organic acids; tannins; vitamins of group B; carotene; vitamin C | K (32.5 mg/g), Ca (6.5 mg/g), Mg (3.8 mg/g), Fe (0.1 mg/g), Mn (0.22 mg/g), Cu (0.55 mg/g), Zn (0.71 mg/g), Co (0.01 mg/g), Mo (88.0 mg/g), Cr (0.02 mg/g), Al (0.02 mg/g), Se (8.5 mg/g), Ni (0.51 mg/g), Sr (0.02 mg/g), Pb (0.01 mg/g), B (68.8 mg/g) | Flavonoids; alkaloids; tanacetin; resins; sugar; gum |
| 16 | Plantago major | Carbohydrate mannitol; vitamin K; ascorbic (289 mg %), citric and oleic acids; tannins | K (44.60 mg/g), Ca (39.30 mg/g), Mn (3.60 mg/g), Fe (0.70 mg/g), Mg (0.25 BAC), Cu (0.92 BAC), Co (0.25 BAC), Mo (2.67 BAC), Cr (0.12 BAC), Ba (22.05 BAC), V (0.13 BAC), Se (0.70 BAC), Ni (0.17 BAC), Sr (1.82 BAC), Pb (0.13 BAC) | Nitrogen (20 %) and nitrogen-free (10 %) extractives; cellulose (10 %); fats (0.5 %); glycoside aucubine; flavonoids; ash (15.79 %) |
| 17 | Artemisia absinthium | Essential oil (0.5–2 %): thujone, pinene, cadinene, bizabolon, chamazulenogen, selenin; phytoncides; alkaloids; vitamins: ascorbic acid and provitamin A; organic acids: malic, amber; tannins | K, Ca, Co, Fe, Na, Zn, Ag, Ba, Br, Cr, Sr, As, Sb, Th, U | Bitter glycosides: absintine (up to 0.25 %), anabsinthin (0.03 %); flavonoids; capillin; saponins (4.22 %); carotene; mineral salts |
| 18 | Motherwort | Essential oil (up to 0,05 %); tannic, bitter and sugary substances; organic acids P-coumarinic, tartaric, malic, ursolic, vanillinic, citric, vitamins A, E, C | Co, Mg, Cu | Amine stachidrine; falavonoids: quercetin, rutin, quinqueloside; flavonoid glycoside; saponins; alkaloids (0.4 %): leonurin, leopuridine |
| 19 | Sage | Tannins and tarry substances; organic acids: oleanolic, ursolic, chlorogenic; vitamins P and PP; essential oil: pinene, cineole, thujone, borneol, salven | K, Na,Ca, P, Zn, Fe, Cu, Mn, J | Flavonoids; alkaloids (up to 0.4 %); phytoncides |
| 20 | Common sunflower | Resinous substances; fatty oil (40 %); carbohydrates (up to 25 %); Vitamin PP and E | Fe, K, Ca, Mg, Cu, Na, P, Zn | Flavonoids quercimer-trine); coumarinic scopolin glycoside; triterpene saponins; sterols (glycoside sitosterolin); carotenoids: β-carotene, cryptoxanthin, taraxanthin; phenolic carboxylic acids: chlorogenic, ochlorogenic, coffee; anthocyanins |

**Note:** data on the chemical composition are taken from [2, 3].

Medicinal herbs have biological activity (toxicity), which accompanies the main therapeutic effect and depends on the dose. Toxicity is not observed in all medicinal herbs, but among them there are strong and even poisonous (aconite, autumn crocodile, belladonna, hemlock, milestone, wolfish, datura, lily of the valley, nightshade black, veratrum). Some plants cause poisoning, dizziness and headaches – magnolia, lily, bird cherry.

Presented in Table 1 herbal composition based on 2 % chitosan was not studied for toxicity, therefore, to establish the safety of their use as a packaging for fruit and vegetables, this must be done.

## 3. The aim and objectives of research

*The aim of research* is an experimental study of the safety of film-forming compositions for the treatment of fresh fruits and vegetables to extend shelf life.

In order to study the set aim, it is necessary to:

1. Create 20 types of chitosan-based film-forming compositions.

2. Conduct an analysis of the compositions for toxicity and hemolysis zones to confirm their safety and for further use in the coating of fruit and vegetable products.

## 4. Research of existing solutions of the problem

Analysis of scientific literature and patents shows that one of the generally recognized effective ways of storing fruit and vegetable products are: refrigerated storage and storage in conditions of changing the external gas environment – regulated and modified (RGE and MGE).

For many years, the technology of the controlled environment, well known to specialists as an atmosphere control system inside the chamber, is used to increase the storage period of fruits and vegetables with a high quality standard.

So, in [4] the way of storage of fruit and vegetable products is patented in conditions of a controlled atmosphere of storage in a room with a water-air heat exchanger.

In work [5] questions related to the development of technology for the long-term preservation of fruit quality of some apple varieties are investigated: Antonovka vulgaris, varieties Martovskiy, Zhigulevsky, Severny Sinap. These studies indicate that in order to increase the efficiency of storage of fruits, the following conditions must be observed:

1) timely harvesting of fruits;

2) selection and formation of batches of fruits for storage should be carried out taking into account the forecast of keeping quality.

Researchers [6] propose the use of a synchronous humidification and cooling system for fruit and vegetable products. This system includes special devices for water spraying, humidity controller, microcomputer and air cooler, which will promote optimal storage of products.

The authors [7, 8] study gas methods for increasing the storage of fruit and vegetable products. For such studies, gas distribution systems are used, where the gas medium in the chambers was maintained.

ISSN 2226-3780

Some researchers [9–11] propose a method of cold storage. This method of storage provides special conditions inside the storage and equipment. In some works it is recommended to use additionally vacuum packages for products.

In paper [12], it is suggested to use the pre-cooling pressure drop during the storage of fruit and vegetable products. In this way, losses can be significantly reduced, and the quality of fruits and vegetables can be improved by optimizing and controlling the fan speed through a variable speed controller.

In [13, 14], methods for forming a microclimate in storage facilities using artificially created moisture and a disinfected environment with cooling to preserve the quality of fruits and vegetables are proposed. Such methods allow storing fruits and vegetables for a long time.

At the present stage of the development of storage technologies for fruits and vegetables, membrane technologies and membrane plants are widely used in various industries for processing both liquid and gaseous mixtures. The principle of operation of the membrane element of the plant is based on the different rate of gas penetration through the polymeric hollow fiber membrane under the influence of pressure drop on the membrane. The practical focus of [15] is based on an in-depth analysis of the theoretical issues of the distribution of complex gas mixtures on hollow fiber membranes.

The author of [16] studies the modes and parameters of ozonation of vegetable storage during the storage of eggplants, depending on the dynamic characteristics of the electro-ozonizer and the parameters of the discharge device.

According to the results of the research, a technological process has been developed that allows increasing the shelf life of eggplants, which is characterized by the fact that certain rational regimes and parameters of electro-ozone decoration of premises and vegetables allow to increase the yield of standard products by 8.4 %.

In turn, the author of the works [17, 18] offers a method for storing vegetables, which includes loading them into special containers for preliminary treatment with ozone of concentration of 45–50 mg/m$^3$ for a given time.

Some patents [19, 20] suggest storage methods in special boxes with sterilization, which reduces pesticide residues. In such boxes there is a small-size ozone generator capable of continuously producing ozone, which has the function of sterilization, which slows the course of chemical processes in fruits and vegetables.

The high coefficient of heat transfer during refrigeration treatment of products in various types of liquids makes it possible to significantly accelerate the process and maximize the preservation of product quality [21].

The authors of [22, 23] recommend the regime of storing fruits of apricot in the cooled state:

– at a temperature of 1 to 2 °C for 10–20 days, depending on the characteristics of the variety [22];
– at a temperature of –0.5 °C for 7–14 days [23].

Storage of fruits in a chilled way does not completely solve the problem of reducing losses and preserving the quality of products. According to the data of [24], losses of fruits during storage in the cooled state reach 50 %.

According to the authors of the works [25, 26], the prolongation of the storage of fruit of stone fruit crops with the maximum preservation of quality is possible with the use of fast freezing technology.

These methods of storage, in addition to significant financial costs, are technologically complex processes, since they require working out and strict adherence to the temperature-humidity-gas regime, and also are not environmentally friendly.

The modern direction of improving storage methods is the processing of fruits and vegetables with film-forming compositions. It is proposed in [27] to use a solution of chitosan as a preservative for the treatment of plant products prior to storage.

The method proposed in [28] is interesting, which discloses a method of obtaining a preservative film for short-term storage of fruits and vegetables. The film has a sufficient adsorption capacity for ethylene, so the rate of ripening of fruits and vegetables wrapped in film is reduced.

In [29], antibacterial properties of a polyolefin film for the storage of ecologically pure fruits and vegetables are investigated. These studies indicate that the film can improve the antibacterial effect, and also reduce the use of chemical preservative, secondary chemical contamination, as well as the impact on the environment and products. In addition, the film improves the appearance of fruits and vegetables, increases the product's product appeal.

To increase the effectiveness of the use of polyethylene packages in the film, bactericidal components (potassium sorbate, imunocytophyte, lauryl thiodipropionate, etc.) are added during its production.

Such polymer bactericidal container reduces the level of pathogenic microflora by a factor of 2, slows the physiological processes in fruits, which allows preserving the turgor of fruits and their presentation [30].

In the United States, a package for the storage of fruit in the MGE, consisting of a copolymer of ethylene, α-olefin, acrylic and methacrylic acids, has been patented [31, 32].

The prospects for creating film-forming compositions are recognized by scientists all over the world. In economically developed countries, despite the existence of a continuous refrigeration chain and a network of processing enterprises, research is constantly being carried out in this area.

It should be noted that the developed and patented film-forming compositions have not been properly applied in Ukraine and in connection with this there are a number of unresolved problems:

– lack of necessary harmlessness;
– technological complexity;
– lack of sufficient efficiency or the need to comply with certain storage regimes.

Thus, the development of material-saving effective environmentally friendly and simple in the implementation of storage technologies for fresh fruit and vegetable products using film-forming compositions and the scientific basis for its preservation has great theoretical and practical significance.

## 5. Methods of research

The developed compositions based on 2 % chitosan are analyzed (Fig. 3).

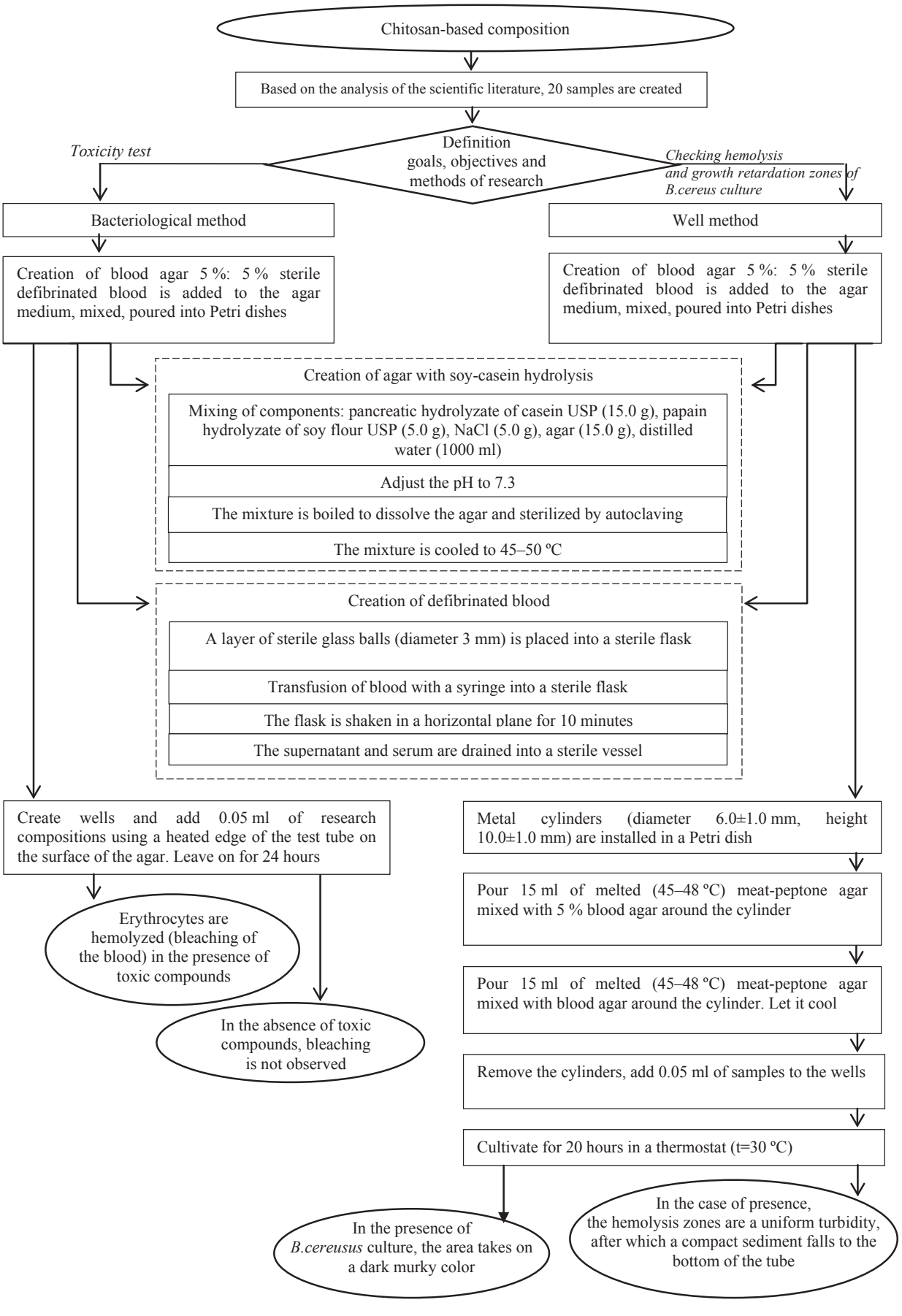


**Fig. 3.** Algorithm of research on sanitary bacteriology methods

ISSN 2226-3780

Blood that is used to create blood agar, should not contain antibiotics and chemotherapeutic drugs. When creating defibrinated blood, fibrin is formed, which remains on the balls, and supernatant fluid.

To check the toxicity, hemolysis zones and propagation of *B.cereus* culture, time must pass. Lysis of red blood cells is used to detect toxic compounds. In this case, the method implies an incubation of 24–48 h at $t=37$ °C. In the presence of toxic compounds, an enlightenment is observed at the site of application of the sample. In the study of hemolysis zones, the culture is carried out in a thermostat for 20 h at $t=30$ °C, along with this, an increase in the culture of *B.cereus* can be detected. Hemolysis zones have the appearance of turbidity, as well as precipitation on the bottom of the tube, the growth of *B.cereus* culture forms a dark murky color [33].

## 6. Research results

The solutions are tested for toxicity using bacteriological methods by inoculating with 5 % blood agar. To do this, on the surface of the blood agar, the wells are made by applying the heated edge of the tube to the agar surface for a few seconds. To the center of the well, 0.05 ml of research compositions are added.

Toxic compounds hemolize red blood cells and there is a bleaching of the blood at the site of the sample (lysis of erythrocytes).

After 1 day, samples of the solutions are checked. In all 20 samples, lysis on blood agar is not detected, that is, when evaluating the results, it can be concluded that the samples are not toxic.

Along with this, a study is carried out by the method of diffusion into agar (well method), based on the ability of medicinal substances to penetrate into the thickness of the agar and to manifest hemolytic activity on 5 % blood agar. For this purpose, metal cylinders (inner diameter 6.0±1.0 mm, height 10.0±1.0 mm) are installed in Petri dishes.

Around the cylinders, 15 ml of melted and cooled to 45–48 °C meat-peptone agar mixed with blood (5 % blood agar) are poured. When the agar in the plates solidified – the cylinders are gently removed with sterile forceps, 0.05 ml of test samples of solutions are added to the wells. After culturing for 20 hours at $t=30$ °C in the thermostat, the results are evaluated according to the guidelines [34].

The hemolysis zone is not detected, however, the growth inhibition of *B.cereus* culture, which grew on the surface of blood agar, is suppressed.

Bacillus cereus is a kind of gram-positive, spore-forming soil bacteria. This bacterium causes a person to have a toxic infection (vomiting and diarrheal syndrome). This kind of bacteria can get on the fruit and vegetable products with fetal growth or transportation. Like any kind of bacteria, it can remain on the surface of the fetus after washing or other treatment, and during storage *B.cereus* has the ability to multiply.

Zones of growth retardation of *B.cereus* are presented in Table 2 and in Fig. 4.

The study show that compositions based on chitosan 2 % inhibit the growth of *B.cereus*, which allows the coated products to be safe for consumption.

**Table 2**

The sensitivity of *B.cereus* to the test samples

| No. | Test samples | Observation data (diameter of growth arrest zones of *B.cereus*, mm) |
|---|---|---|
| 1 | Sweet flag | 34 |
| 2 | Senegalia catechu | 32 |
| 3 | Ledum | 30 |
| 4 | Bergenia crassifolia | 28 |
| 5 | Common yarrow | 30 |
| 6 | Eucalyptus globulus | 32 |
| 7 | Eleutherococcus senticosus | 30 |
| 8 | Hypericum perforatum | 0 |
| 9 | Chamaenerion angustifolium | 24 |
| 10 | Iceland moss | 34 |
| 11 | Calendula officinalis | 34 |
| 12 | Urtica | 30 |
| 13 | Peppermint | 32 |
| 14 | Potentilla alba | 34 |
| 15 | Common tansy | 0 |
| 16 | Plantago major | 24 |
| 17 | Artemisia absinthium | 30 |
| 18 | Motherwort | 0 |
| 19 | Sage | 24 |
| 20 | Common sunflower | 32 |

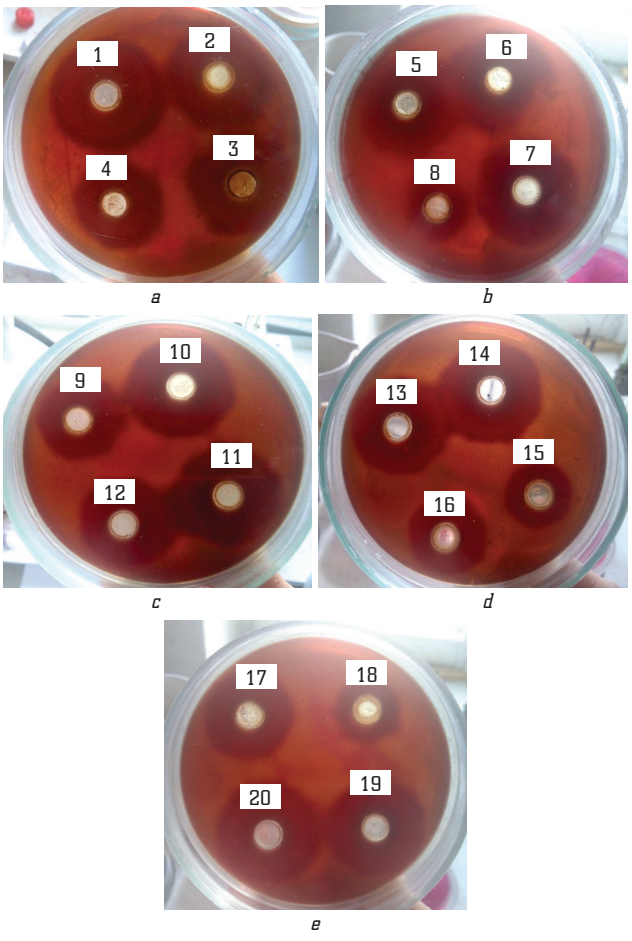


**Fig. 4.** Delays in the growth of *B.cereus* culture: *a* – Sweet flag, Senegalia catechu, Ledum, Bergenia crassifolia; *b* – Common yarrow, Eucalyptus globulus, Eleutherococcus senticosus, Hypericum perforatum; *c* – Chamaenerion angustifolium, Iceland moss, Calendula officinalis, Urtica; *d* – Peppermint, Potentilla alba, Common tansy, Plantago major; *e* – Artemisia absinthium, motherwort, sage, common sunflower

## 7. SWOT analysis of research results

*Strengths*. Among the strengths of this research, it is necessary to note the nontoxicity of film-forming compositions, as well as the inhibition of growth of *B.cereus* culture. In favor of this, the results of the studies cited above testify. There are no analogues of such compositions.

*Weaknesses*. The weaknesses of this work are related to the fact that the use of such compositions requires further study of them, since the effect of the film-forming composition on fruit and vegetable products has not been studied. Therefore, in order to prevent this drawback, an additional experiment should be carried out to apply the test components to different types of fruit and vegetable products during storage and to investigate the slowing of the loss of product quality.

*Opportunities*. The study of chitosan is very promising, as the product has high physiological activity and is an environmentally friendly product. Chitin is a source of chitosan, which is widely distributed in nature. In combination with herbs that have bactericidal properties, it is not toxic. Components of the compositions are inexpensive, and to create the composition itself, no special equipment and knowledge is required. When the method of storing fruit and vegetable products covered with a composition of chitosan is introduced into the enterprise, it will be possible to protect the production from contamination with the culture of *B.cereus* and preserving the quality of fruits and vegetables.

*Threats*. Taking into account that natural and bactericidal products were used in the preparation of the chitosan-based composition, no threats are identified. Also the constituent components of the finished compositions are all available and not costly. Previously, a solution of chitosan as a preservative was used to treat products of plant origin before storage, but with other constituents. There are no analogues of this kind in the world.

## 8. Conclusions

1. Based on the planning of experimental works, the initial stages of the study are determined, namely the creation of 20 samples from herbs, to which chitosan 2 % is added. All samples were examined for toxicity and hemolysis zones.

2. It has been established that the samples are not toxic and there are no hemolysis zones. The fact of inhibition of the growth of *B.cereus* culture, which grew on the surface of blood agar, and the zones of growth retardation of *B.cereus* (mm):

- Sweet flag – 34;
- Senegalia catechu – 32;
- Ledum – 30;
- Bergenia crassifolia – 28;
- Common yarrow – 30;
- Eucalyptus globulus – 32;
- Eleutherococcus senticosus – 30;
- Hypericum perforatum – 0;
- Chamaenerion angustifolium – 24;
- Iceland moss – 34;
- Calendula officinalis – 34;
- Urtica – 30;
- Peppermint – 32;
- Potentilla alba – 34;
- Common tansy – 0;
- Plantago major – 24;
- Artemisia absinthium – 30;
- Motherwort – 0;
- Sage – 24;
- Common sunflower – 32.

### References


1. Dubinina, A. A. Upakovka kharchovykh produktiv iz polimernykh materialiv [Text]: Monograph / A. A. Dubinina, H. A. Synytsyna, O. H. Myshnyk et al. – Kharkiv: Fakt, 2011. – 399 p.
2. Maznev, N. I. Entsiklopediia lekarstvennyh rastenii [Text] / N. I. Maznev. – Ed. 3. – Moscow: Martin, 2004. – 496 p.
3. Lavrenov, V. K. Sovremennaia entsiklopediia lekarstvennyh rastenii [Text] / V. K. Lavrenov, G. V. Lavrenova. – Moscow: ZAO «OLMA Media Grupp», 2007. – 272 p.
4. Method for preserving fruits and vegetables by using controlled atmosphere storage house with water-air heat exchanger [Electronic resource]: Patent CN 104642513 A / Jiang Fen. – Appl. No. CN 201510060513; Filed February 5, 2015; Publ. May 27, 2015. – Available at: \www/URL: https://encrypted.google.com/patents/CN104642513A?cl
5. Gudkovskii, V. A. Progressivnye tehnologii hraneniia plodov [Text] / V. A. Gudkovskii, A. A. Klad, L. V. Kozhina, A. E. Balakirev, Yu. B. Nazarov // Dostizhenie nauki i tehniki APK. – 2009. – No. 2. – P. 66–68.
6. Air cooler synchronous humidification system for fruit and vegetable cold storage storeroom [Text]: Patent CN 203949317 U, MPK A23L3/36; F24F11/02; F24F6/12; F25D13/00 / Feng Zhihong, Wang Chunsheng, Song Zhuojun, Li Chao, Wang Liang, Chen Jia, Zhao Yingli. – Filed March 12, 2014; Publ. November 19, 2014. – 1 p.
7. Ilinskii, A. S. Razvitie tehnologii hraneniia v reguliruemoi atmosfere (obzor) [Text] / A. S. Ilinskii, A. V. Dmitriev, V. Yu. Pugachiov, A. M. Kuznetsov // Hranenie i pererabotka sel'hozsyr'ia. – 2003. – No. 8. – P. 52–56.
8. Gas regulation control method of spontaneous modified atmosphere storage for fruit and vegetable preservation [Electronic resource]: Patent CN 102609011 B / Juan Wang, Xiangyou Wang, Peijuan Xiang, Miao Huang, Xin Han. – Appl. No. CN 201210075848; Filed March 21, 2012; Publ. July 25, 2012. – Available at: \www/URL: https://www.google.com/patents/CN102609011A?cl
9. Cold storage method of vegetables in agricultural and sideline products [Text]: Patent CN 105123897 A, MPK A23B7/04, A23B7/152, A23B7/154, A61L2/18 / Zhang Guoqian, Zheng Yang, Zhang Jian. – Filed August 25, 2015; Publ. December 9, 2015. – 1 p.
10. Cold storage method for fruit and vegetable [Text]: Patent TW 201117726 A, MPK A23B7/04, B65D81/02 / Cheng Sung-Chi. – Filed November 27, 2009; Publ. June 1, 2011. – 1 p.
11. Kolodiazhaia, V. S. Primenenie ais slarri dlia zamorazhivaniia rastitel'nyh produktov [Text] / V. S. Kolodiazhaia, V. N. Sokolov // Proizvodstvo i realizatsiia morozhenogo i bystrozamorozhennyh produktov. – 2003. – No. 5. – P. 24–28.
12. Integrated fruit and vegetable differential pressure precooling storage [Electronic resource]: Patent CN 202184088 U / Sheng Liu, Jinpeilou. – Appl. No. CN 201120249678; Filed July 15, 2011; Publ. April 11, 2012. – Available at: \www/URL: https://www.google.com/patents/CN202184088U?cl
13. Universal'nyi i prostoi sposob sohraneniia tovarnogo vida, vkusovyh i poleznyh kachestv ovoshchei i fruktov pri ih prodolzhitel'nom hranenii [Text]: Patent RU 2013156913 A, MPK A23L 3/00 / Milevskii V. B. – Appl. No. CN 201510060513; Filed December 20, 2013; Publ. April 10, 2016. – 2 p.
14. Taurit, V. R. Formirovanie temperaturnogo rezhima v ventiliruemyh ovoshchehranilishchah [Text] / V. R. Taurit, C. B. Taurit // Sel'skohoziaistvennye vesti. – 1997. – No. 4–5 (27–28). – P. 48–49.
15. Kriukov, A. M. Hranenie sel'skohoziaistvennogo syr'ia v kontroliruemoi gazovoi srede s ispol'zovaniem kompressorno-membrannogo oborudovaniia [Text] / A. M. Kriukov, E. P. Koshevoi // Materialy II mezhdunarodnoi nauchno-tehnicheskoi konferentsii. – Voronezh, 2004. – Part 1. – P. 249–251.

ISSN 2226-3780

16. Normov, D. A. Vliianie obrabotki ozono-vozdushnoi smes'iu na liozhkost' baklazhanov [Tex] / D. A. Normov, E. A. Fedorenko // Gavrish. – 2009. – No. 1. – P. 32–34.
17. Sposob hraneniia ovoshchei [Text]: Patent RU 2000104186 A, MPK A23B 7/14, A23B 7/00, A01F 25/00 / Pavlov A. M. – Filed February 21, 2000; Publ. March 10, 2002. – 2 p.
18. Sposob hraneniia ovoshchei [Text]: Patent RU 2000112978 A, MPK A23B 7/00, A23B 7/14, A23L 3/3409, A01F 25/00 / Pavlov A. M. – Filed May 24, 2000; Publ. May 27, 2003. – 2 p.
19. Fruit and vegetable storage-transportation fresh-keeping box with functions of sterilizing and degrading pesticide residue [Electronic resource]: Patent CN 201849811 U / Cunkun Chen, Wensheng Wang, Ning Jia. – Appl. No. CN 201020602303; Filed November 11, 2010; Publ. June 1, 2011. – Available at: \www/ URL: http://www.google.com.pg/patents/CN201849811U?cl=en
20. Fruit and vegetable storage and transportation fresh-keeping box with functions of sterilization and pesticide residue degradation [Electronic resource]: Patent CN 102001490 A / Cunkun Chen, Wensheng Wang, Ning Jia. – Appl. No. CN 201010540376; Filed November 11, 2010; Publ. April 6, 2011. – Available at: \www/ URL: http://www.google.sr/patents/CN102001490A?cl=en
21. Chachin, K. The quality preservation of agricultural products at refrigeration storage [Text] / K. Chachin // Reito Refrigeration. – 2000. – No. 867. – P. 20–23.
22. Sumnu, G. Quality control charts for storage of apricots [Text] / G. Sumnu, L. Bayindirh, M. Ozilgen // Zeitschrift fur Lebensmittel-Untersuchung und Forschung. – 1994. – Vol. 199, No. 3. – P. 201–205. doi:10.1007/bf01193444
23. Shoruhin, K. Questions of fruits and vegetables storage quality [Text] / K. Shoruhin // Machinery and Equipment Food Industry. – 1998. – No. 3. – P. 60–72.
24. Paronian, V. H. Progressivnye sposoby obrabotki plodoovoshchnoi produktsii pered zakladkoi na hranenie [Text] / V. H. Paronian, G. P. Kiuregian, N. V. Komarov // Hranenie i pererabotka sel'hozsyr'ia. – 2003. – No. 7. – P. 23.
25. Turbin, V. A. Prognozirovanie himicheskogo sostava zamorozhennyh plodov abrikosa pri dlitel'nom hranenii [Text] / V. A. Turbin, G. I. Glushko // Pishchevoi tehnologii. – 2002. – No. 1. – P. 36–38.
26. Belenko, E. L. Nizkotemperaturnoe zamorazhivanie plodov kostochkovyh kul'tur [Text] / E. L. Belenko, E. L. Jeneeva, S. V. Levchenko, N. L. Studennikova, P. L. Parfenova, V. N. Kuzmenko // Sadovodstvo i vinogradarstvo. – 1997. – No. 2. – P. 10–12.
27. Use of water solution of chitosan as a conserving agent for treatment of food products of vegetable origin before storage [Text]: Patent UA 20183 U, MPK A23B 4/00, A01F 25/00, C08B 37/00 / Kavyrshyn O. P., Fedorov S. A. – Filed July 10, 2006; Publ. January 15, 2007, Bull. No. 1. – 5 p.
28. Preservative film for short-term storage of fruits and vegetables, preparation method of preservative film and prepared preservative bag [Electronic resource]: Patent CN 104309903 A / Shi Dixing. – Appl. No. CN 201410499541; Filed September 25, 2014; Publ. January 28, 2015. – Available at: \www/URL: https://www.google.com/patents/CN104309903A?cl=en
29. High-transparency physical antibacterial polyolefin fruit and vegetable storage and transportation preservative film [Electronic resource]: Patent CN 101643567 A / Yanwen Zhou, Shijun Wang, Ping Zhang, Jiazheng Li. – Appl. No. CN 200910306633; Filed September 7, 2009; Publ. February 10, 2010. – Available at: \www/ URL: https://www.google.com/patents/CN101643567A?cl=en
30. Dobias, J. Emballage des denrees alimentaries en atmospheres contrôlèes au modifièes [Text] / J. Dobias, M. Voldrich, J. Philippen // Rev. Gen. Froid. – 1994. – No. 7. – P. 55–61.
31. Multilayered film [Electronic resource]: Patent US 5169728 A / Murphy M., Jennergren B. C. G.; assignee: The Dow Chemical Company. – Appl. No. US 07/663,216; Filed February 28, 1991; Publ. December 8, 1992. – Available at: \www/URL: https://www.google.com/patents/US5169728
32. Oxygen-permeable multilayer film [Electronic resource]: Patent US 5491019 A / Kuo B. P.; assignee: W. R. Grace & Co.-Conn. – Appl. No. US 08/218,776; Filed March 28, 1994; Publ. February 13, 1996. – Available at: \www/URL: http://www.google.tl/patents/US5491019
33. Jones, D. Manual of Methods for General Bacteriology [Text] / D. Jones // Journal of Clinical Pathology. – 1981. – Vol. 34, No. 9. – P. 1069–1069. doi:10.1136/jcp.34.9.1069-c
34. Volianskyi, Yu. L. Vyvchennia spetsyfichnoi aktyvnosti protymikrobnykh likarskykh zasobiv [Text]: Handbook / Yu. L. Volianskyi, I. V. Hryshchenko, V. P. Shyrobokov et al. – Kyiv, 2004. – 38 p.

## ИССЛЕДОВАНИЕ ТОКСИЧНОСТИ ПЛЕНКООБРАЗУЮЩИХ КОМПОЗИЦИЙ НА ОСНОВЕ ХИТОЗАНА

Из рассмотренных композиций на основе хитозана и на основе анализа характеристики лекарственных отваров было выбрано 20 видов для дальнейшего составления композиции с хитозаном 2 %. Для установления их безопасносного использования в плодоовощной продукции все образцы исследованы на токсичность и зоны гемолиза. При проведении исследований получены положительные результаты – не было обнаружено токсичности и зоны гемолиза.

**Ключевые слова:** композиции на основе хитозана, отвары лекарственных трав, токсичность композиций, зоны гемолиза, кровяной агар.

***Dubinina Antonina,*** *Doctor of Technical Sciences, Professor, Department of Commodity Research and Expertise of Goods, Kharkiv State University of Food Technology and Trade, Ukraine, e-mail: tovaroved206@ukr.net, ORCID: http://orcid.org/0000-0003-3132-1918*

***Letuta Tatiana,*** *PhD, Associate Professor, Department of Commodity Research and Expertise of Goods, Kharkiv State University of Food Technology and Trade, Ukraine, e-mail: lettanya@ukr.net, ORCID: http://orcid.org/0000-0002-0110-2047*

***Frolova Tatiana,*** *Postgraduate Student, Department of Commodity Research and Expertise of Goods, Kharkiv State University of Food Technology and Trade, Ukraine, e-mail: tetfrol70@ukr.net, ORCID: http://orcid.org/0000-0001-7188-8236*

***Savinova Helena,*** *Doctor of Medical Sciences, Professor, Department of Clinical Immunology and Microbiology, Kharkiv Medical Academy of Postgraduate Education, Ukraine, ORCID: http://orcid.org/0000-0003-2524-2287*

***Bolshakova Galina,*** *PhD, Associate Professor, Department of Clinical Immunology and Microbiology, Kharkiv Medical Academy of Postgraduate Education, Ukraine, e-mail: gal.bolsha@gmail.com, ORCID: http://orcid.org/0000-0002-1151-0562*

***Novikova Vira,*** *Postgraduate Student, Department of Commodity Research and Expertise of Goods, Kharkiv State University of Food Technology and Trade, Ukraine, e-mail: novikova_vera@ukr.net, ORCID: http://orcid.org/0000-0003-3153-2619*