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THE PROSPECTS OF SAFETY AND MICROBIOLOGICAL STABILITY IMPROVEMENT OF BAKERY PRODUCTS BY USING PHYTO-ADDITIVES

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Abstract: *In this article the main problems of bakery product quality and safety formation have been examined; it has been noted that one of the main objectives of the industry is implementation of measures of bread ropiness and molding prevention. On the basis of analyzing the main methods of product microbiological stability improvement during storage that include the usage of artificial preservatives or sourdough with specifically cultivated fermentative microflora and evaluating their effectiveness, the importance of search for alternative solution for problems of baking industry has been demonstrated. In this aspect especially interesting are aromatic and medicinal plants known for their antiseptic, antioxidant, and other properties capable of stabilizing the product quality. The main goal of this work is the study of a number of phyto-additives and their impact on pathogenic microorganisms and fermentative microflora of semi-products, their biotechnological properties, and finished product quality.*
The usage of phyto-additives as extracts has been proposed. It has been determined that hop, St John's wort, and ashberry extracts have pronounced antiseptic properties towards microbiological pathogens of bakery products. The sage, marigold, peppermint, and chamomile extracts have been effective against mold fungi. All extracts have positive effect on intensity of fermentation process where the best results has been achieved by using cinnamon rose, haw, ashberry, and hop extracts. The consumer properties of wheat bread with extracts added judged by volume, texture, and shape stability for almost all samples have been either improved or identical to check sample. Their microbiological stability has been considerably improved.

Keywords: *extract, aromatic and medicinal plants, bread ropiness, molding*

1. Introduction

The bakery products have a special place in nutrition of population of many countries, and are consumed daily regardless of the customers' age, health state, and lifestyle. They are the constant source of many nutrients and biologically active substances (BAS), have high assimilability, and are capable of regulating the activity of gastrointestinal tract [1]. However the potentially harmful substances of chemical or microbiological nature can enter the human organism with bakery products presenting a serious danger to health. Those are mainly the

compounds accumulated because of violating the rules of growing and storage of raw materials, those used to solve the technological problems of baking industry as well as those produced during bread production and storage. Among these are toxicological agents, radionuclides, polycyclic substances, pesticides, components of mineral fertilizers, products of lipid oxidation and pest vital functions. The problem of raw material contamination by highly toxic, mutagenic, and cancerogenic mycotoxins produced by mold fungi has escalated worldwide [2, 3]. The concern of consumers and nutritionists specializing in product safety has also been caused by the presence of a wide spectrum of additives statistically confirmed to be used by more than 80 % of bakeries in order to improve the quality of bread and pastries [4]. They are used in order to solve the technological problems of forming the product of necessary quality under the condition of considerable fluctuations of raw materials (mainly flour) properties, implementation of accelerated technologies, and introduction of new sources of BAS with low technological properties. Besides, the fact of bread being one of the products with high acrylamide content [5] is a cause of concern since acrylamide formed during baking as a result of a reaction of asparagine with sugar has cancerogenic and genotoxic properties. With the implementation of accelerated technologies of bread production and introduction of high amounts of reducing sugars the acrylamide content of the finished product increases [6]. One of the main problems leading to deterioration of consumer qualities and safety of wheat bakery products and economical losses for manufacturers is a low microbiological stability of finished products during storage. The frequent cases of bread ropiness caused by spore-forming microorganisms *Bacillus subtilis* and *Bacillus mesentericus* and molding caused by the growth of fungi of *Aspergillus, Mucor, Penicillum,* and *Rhizopus* genus [7, 8]. The situation escalation in the last years has been caused by a number of factors. Firstly, the tendency of manufacturers to save time, energy, material, and labor resources leads to implementation of accelerated technologies with shortened duration of technological processes and mainly dough fermentation. This leads to the decrease of growth and fermentative activity of lactic acid bacteria that are the antagonists of undesirable microflora of flour semi-products. Secondly, complying with modern requirements of physiological properties and nutritional value of bread, the manufacturers expand the raw materials base introducing the ingredients potentially more microbiologically contaminated. All of this including the increase of amounts of highly contaminated wheat flour escalates the problem of bakery products microbiological spoilage (fig. 1).

In order to improve the biological stability of bread products during storage the chemical preservatives are mainly used, however, many of them inhibit the fermentative activity of lactic acid bacteria as well. This creates difficulties connected to flavor, aroma, and texture formation of finished product, ensuring the safety of produce, and its staling rate. The efficient technological solution of this problem is the usage of wheat sourdough with specifically cultivated microflora. However, it complicates the technological process considerably due to continuity of sourdough maintenance and its quick contamination by foreign microflora that deteriorates their biotechnological properties due to the lack of aseptic conditions in bakeries.

Thus the pursuit of alternative quick and effective solutions for baking industry

problems including the methods of improving bread products safety and imparting necessary physiological properties is relevant.

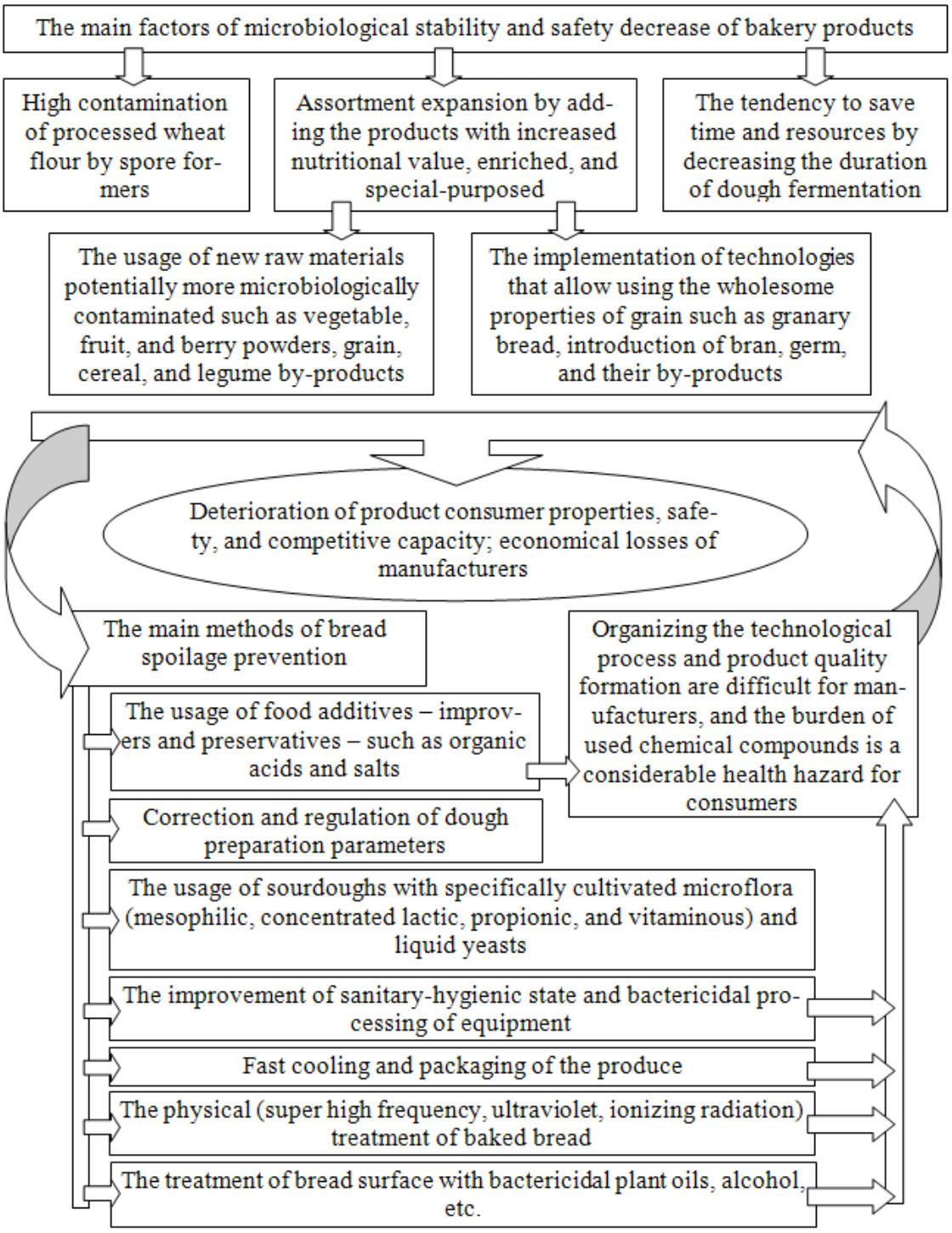

Fig. 1. The problems of formation and improvement of bread microbiological stability [2, 3, 7-10]

In this aspect especially interesting are the aromatic and medicinal plants known for their antiseptic, antioxidant, and other properties capable of stabilizing the product quality as well as their tonic, therapeutic, and prophylactic properties. Nowadays these plants are considered as the main source of parapharmaceuticals – minor components with physiologically active properties, capable of regulating organism functions, and showing immunomodulating, antimutagenic, protective, adaptogenic, and therapeutic effects [11].

The value of aromatic and medicinal plants as raw material for food industry is in the variety of chemical composition and biochemical properties that ensures the complex formation of different technological and physiological properties. The usage of phyto-additives containing phenolic, tanning, pectic agents, phytoncides, vitamins, and wide range of major and minor dietary elements in baking industry is promising in order to improve the biotechnological properties of semi-products, regulate the dough structural-mechanical properties, modify the intensity of technological process, and stabilize the product quality while decreasing the usage of artificial food additives [12, 13]. Besides, nowadays there is a wide search for methods of decreasing the acrylamide content in products including bread showing the perspective of using the phyto-additives with high polyphenol, organic acids, and vitamin B_3 content as well as those intensifying the alcoholic and lactic acid fermentation. Due to their inclusion into the formulation the structure and content of asparagine – the precursor of acrylamide – in baking semi-products changes which leads to lower rate of acrylamide formation and correspondingly to decrease of its content in finished products [3, 14]. Thus the usage of phyto-additives should allow the manufacturers to provide the population with wholesome high-quality produce in increasingly more difficult conditions of industry complying with high demands to assortment, consumer properties, safety, biological activity, and nutritional value.

2. Materials and methods

The goal of our work was the study of antiseptic properties of such phyto-additives as hop (*Humulus lupulus*) cones, pelleted hop of UA-AROMA sort, St John's wort (*Hypericum perforatum*), chamomile (*Matricaria chamomilla*), marigold (*Calendula officinalis*), peppermint (*Mentha* × *piperita*), lemon balm (*Melissa officinalis*), sage (*Salvia officinalis*), redhaw hawthorn (*Craetegus sanguinea*), cinnamon rose (*Rosa majalis*), and mountain ash (*Sorbus aucuparia*), and the research of their effect on biotechnological properties of fermentative microflora and finished product quality.

The technology of extract preparation for fruit phyto-additives (haw, rose, and rowan) includes their grinding, bolting, steeping for 1 hour with material to extractant ratio of 1:10, and extraction with the same ratio at 100 °C for 30 min [15]. Hop extraction conditions were material to extractant ratio – 1:100, temperature – 100 °C, duration – 90 min [16]. The preparation of the other phyto-additives included their grinding, bolting, and extraction with material to extractant ratio of 1:20 at 90-100 °C for 30 min.

The moisture content was determined by the drying process in a drying chamber.

The particle size was determined by sifting through specific sieves.

Total soluble solids (TSS) content was determined by refractometric method.

pH value of the extracts was measured by direct insertion of electrodes.

The activity of chosen extracts towards pathogenic microflora inducing the spoilage of bread products was measured by method of diffusion into agar medium. The test-cultures were the specimen of microflora found in bread semi-products – hay bacillus (*Bac. subtilis*), mold of *Aspergillus* and *Penicillium* genus found in grain, wild yeast, and lactic acid bacteria *Lactobacillus plantarum.* The inoculated medium was cultivated for 48 hours at 30±1 ℃. The antimicrobial activity was determined by the diameter of inhibition zone of test-culture colonies.

The rising power was determined by standard method which included measuring the time a 10 g dough pellet submerged in water at 30±1 ℃ needed to break surface.

Acidity was determined by titrating samples with 0.1N NaOH solution.

The bread quality was determined by the following characteristics: moisture content, titrated acidity, loaf specific volume (volume to weight ratio), shape stability (height to diameter ratio), and porosity (pore to crumb volume ratio).

In order to determine the resistance to microbiological spoilage, bread samples were packed into polythene bags and stored at 30±1 ℃ for 96 hours, controlling the appearance of first signs of microbiological spoilage.

3. Results and discussion

The results of experimental bread making (changes of wheat bread texture, crumb color, and flavor brought by introduction of powdered phyto-additives) indicate the merit of using water extracts instead (table 1). Water was chosen as an extractant because it is a base material for bread industry included in almost all formulations; it is versatile, accessible, and easy to use.

Table 1

Properties of phyto-additives and their extracts

Properties	Hop (cones)	Pelleted hop	St John's wort (herb)	Chamomile (blossoms)	Marigold (blossoms)	Lemon balm (herb)	Peppermint (herb)	Sage (leaves)	Redhaw hawthorn (fruit)	Cinnamon rose (fruit)	Mountain ash (fruit)
Phyto-additive moisture content, %	10.9	9.2	12.2	7.8	9.9	12.4	13.3	12.8	12.6	13.8	16.4
Phyto-additive particle size, less than, mm	2.2	–	2.2	2.2	2.2	2.2	2.2	2.2	0.29	0.329	0.329
Extract TSS, %	0.6	0.7	2.6	1.8	2.1	2.4	2.6	2.9	4.8	5.6	5.1
Extract pH value	5.79	5.90	5.97	6.89	6.55	6.68	6.55	6.77	5.18	4.24	5.32

According to the experimental data (table 2) hop, St John's wort, and ashberry extracts exhibited pronounced antiseptic properties towards pathogenic microorganisms causing bread spoilage. The extract of hop cones was the most effective at inhibiting the foreign microflora while the fermentative microorganisms showed resistance to it. The hop antiseptic properties are due to

high content of bitter acids, essential oils, and tanning agents [17] transferred to extract and isohumulone formed during extraction [16, 18]. Ashberries contain considerable amounts of sorbic (natural preservative) as well as succinic, tartaric, malic, and ascorbic acids [19]. St John's wort due to high content of phytoncides, tanning agents, flavonoids, and essential oils inhibited the growth of pathogenic microorganisms as well as wild yeast that might compete with fermentative microflora and lower the biotechnological properties of baking semi-products [20]. However, ashberry and St John's wort unlike hop extracts had negative effect on lactic acid bacteria metabolism. The sage, peppermint, and chamomile extracts were effective against mold fungi. The haw and rose extracts exhibited no bactericidal or fungicidal effect.

Table 2

Antiseptic activity of phyto-extracts

Extract of:	Diameter of inhibition zone of test-culture colonies, mm				
	Lactobacillus plantarum	**Wild yeast**	***Bacillus subtilis***	***Aspergillus***	***Penicillium***
- hop cones	-	-	13.5±0.5	11,3±0,5	15.8±0.5
- pelleted hop	-	-	11.8±0.5	10,6±0,5	12.3±0.5
- St John's wort	11.8±0.5	11.0±0.5	13.2±0.5	11,1±0,5	14.5±0.5
- chamomile	-	-	-	11,7±0,5	12.6±0.5
- marigold	-	-	-	-	11.8±0.5
- lemon balm	-	-	-	-	-
- peppermint	-	-	-	12,4±0,5	13.6±0.5
- sage	-	-	-	13,7±0,5	13.9±0.5
- haw	-	-	-	-	-
- cinnamon rose	-	-	-	-	-
- ashberry	10.8±0.5	-	14.6±0.5	10.0±0.5	11.4±0.5

Note: "-" – no inhibition zone

The study of antiseptic properties of phyto-extracts showed the necessity of researching their effect on alcoholic and lactic acid fermentation during preparation of bread semi-products, specifically sponges with 70±2 % moisture content. The preliminary studies had determined the rational dosage of phyto-extracts that didn't lower the wheat bread quality. Thus the rose extract content should not be more than 15 % to flour mass; however the full replacement of water (60 % to flour mass) with haw extract showed no negative effect. The maximum dosage of hop extracts was 30 % to flour mass, for sage, St John's wort, peppermint, and lemon balm extracts it was 5-10 %, however on the upper limit bread crumb gained light grey color. Since the weakening effect of chamomile and marigold extracts on gluten had been determined, they may be included in the formulation in the amount of up to 20 % to flour mass, if the flour is strong or medium strong. The changes of rising power (the intensity of alcoholic fermentation) and acidity (the intensity of lactic acid fermentation) of traditional sponge with phyto-extracts included in the formulation in aforesaid dosages have been presented on figure 2.

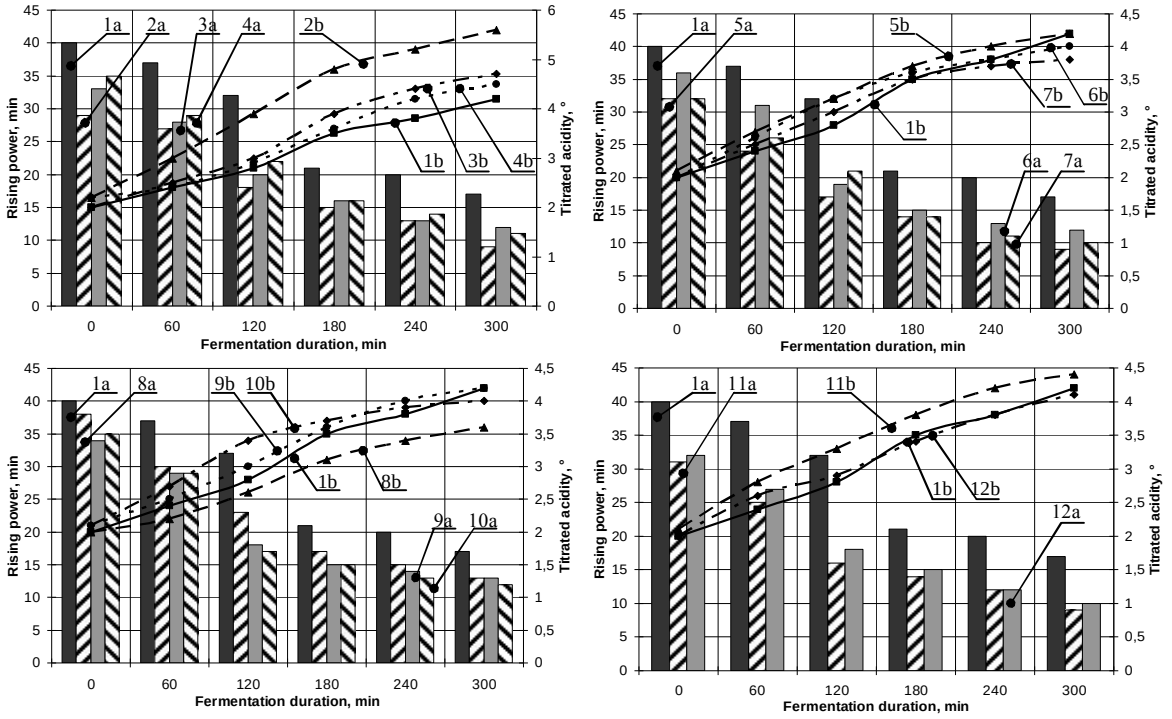

Fig. 2. The effect of phyto-extracts on rising power (a) and acidity (b) of liquid sponges: 1 – control sample; with extracts of: 2 – rose; 3 – haw; 4 – ashberry; 5 – sage; 6 – St John's wort; 7 – chamomile; 8 – marigold; 9 – peppermint; 10 – lemon balm; 11 – hop cones; 12 – pelleted hop

The data analysis determined that water extracts of hop, St John's wort, and ashberry in chosen dosages had selective antiseptic properties namely capacity for inhibiting pathogenic microflora that fermentative microorganisms were resistant to. All phyto-extracts had positive effect on alcoholic fermentation intensity though the best results were reached using hop, rose, haw, and ashberry extracts. This could be explained by their high content of nutrients, organic acids, vitamins, and major and minor dietary elements, deficient in wheat flour semi-products, especially high-grade, and necessary for yeast metabolism.

The complex estimation of phyto-extract technological properties was achieved by experimental baking (table 3).

The obtained results confirmed the effectiveness of using the water phyto-extracts to improve the quality of bread products and their microbiological stability during storage. Thus the consumer qualities of wheat bread (specific volume, shape stability, and porosity) with extracts for almost samples were either improved or analogous to check sample. At the same time their microbiological stability improved. Even the rose and haw extracts that demonstrated no antiseptic properties (table 2) allowed improving the microbiological stability of finished products. Most likely, it was caused by higher amounts of substances inhibiting the growth of foreign microflora produced by lactic acid bacteria as a result of more intense fermentation (fig. 2).

Table 3
The effect of phyto-extracts on bread quality and microbiological stability

Samples	Bread quality properties						
	Moisture content, %	Titrated acidity, °	Specific volume, g/cm^3	Shape stability, H/D	Porosity, %	First signs of molding	First signs of bread ropiness
Rose extract							
Check sample	42.3	2.2	3.03	0.47	70.9	After 72 h	After 48 h
15 % to flour mass	42.5	2.9	3.44	0.50	76.2	–	–
Ashberry extract							
Check sample	42.9	2.2	2.92	0.52	70.0	After 72 h	After 48 h
15 % to flour mass	43.0	2.4	3.13	0.59	77.4	–	–
Haw extract							
Check sample	42.8	2.2	2.93	0.45	73.2	After 72 h	After 48 h
60 % to flour mass	43.0	2.7	3.38	0.50	81.1	After 96 h	–
Sage extract							
Check sample	42.4	2.2	2.84	0.54	70.0	After 72 h	After 48 h
5 % to flour mass	42.4	2.3	3.18	0.58	74.5	–	–
St John's wort extract							
Check sample	42.2	2.2	2.94	0.50	71.4	After 72 h	After 36 h
5 % to flour mass	42.5	2.4	3.46	0.54	75.5	–	–
Pelleted hop extract							
Check sample	43.0	2.8	2.92	0.47	68.0	After 48 h	After 36 h
30 % to flour mass	42.8	3.1	3.30	0.50	73.2	–	–
Hop cone extract							
Check sample	43.0	2.8	2.92	0.47	68.0	After 48 h	After 36 h
30 % to flour mass	43.1	3.3	3.37	0.54	73.7	–	–
Chamomile extract							
Check sample	42.8	2.4	2.90	0.47	67.4	After 72 h	After 48 h
10 % to flour mass	43.1	2.9	2.96	0.41	71.3	–	After 72 h
Marigold extract							
Check sample	42.6	2.3	2.98	0.49	68.1	After 72 h	After 48 h
10 % to flour mass	42.9	2.4	2.95	0.50	67.9	After 96 h	After 96 h
Peppermint extract							
Check sample	42.4	2.2	3.06	0.54	70.0	After 72 h	After 48 h
5 % to flour mass	43.1	2.9	3.29	0.51	75.2	–	–
Lemon balm extract							
Check sample	42.4	2.2	3.06	0.54	70.0	After 72 h	After 48 h
5 % to flour mass	43.0	2.7	3.12	0.53	73.2	–	After 96 h

Note: "-" – no signs of microbiological spoilage

4. Conclusion

Thus the results of studying the antiseptic properties of phyto-extract and their effect on biotechnological properties of semi-products and bread quality demonstrate the merit of their usage in baking industry to solve a number of problems. Thus during the processing of raw materials with high microflora contamination from our point of view the most effective would be hop, sage, and St John's wort extracts. In order

to improve the microbiological stability of buns and pastries containing ingredients with high nutritional value that might need concealment flavor-wise it would be worthwhile to use peppermint and lemon balm extracts. The rose, haw, and ashberry extracts would be useful in accelerated technologies in order to intensify the fermentative process and improve the finished product quality and microbiological stability as a result. In order to control the biotechnological properties of sourdough, liquid yeast, and sponges, the hop extracts might be useful due to their selective antiseptic properties. Their usage should allow controlling the species composition of fermentative microflora in flour semi-products, regulating their biotechnological properties, and improve the bread quality as a result.

The new plant ingredients are interesting from the viewpoint of their usage to improve the nutritional value and physiological properties of bakery products. The high content of pectins, polyphenols, and organic acids is the evidence of their protective, anticarcinogenic, and antioxidant properties. Thus their further study as ingredients for baking industry is important in order to impart the specific physiological properties on finished products as well as lower their acrylamide content.

The diversity of Ukrainian aromatic and medicinal herbs, the growing demands to nutritional properties of food products, and complicated conditions of bread industry demonstrate the merits of researching the natural potential of phyto-additives, their physiological and technological properties in order to find more effective, simple, and safe solution to the problems of baking industry and improve the quality of finished products.

5. References

[1]. TSYPRIAN V.I., Hygiene of nutrition, Book 2, Medicine, Kiev, 544 p, (2007)

[2]. MACHIHINA L.I., ALEKSEIEVA L.V., LVOVA L.S., Ecological safety of grain and grain by-products during storage and processing, *Storage and processing of agricultural raw materials*, 12, 37-40, (2008)

[3]. HOLYNKO O.N., LEVITSKAIA N.P., The problems of safety and quality of bread products, *Nutrition problems*, 1, 16-26, (2013)

[4]. TIURINA YE.B., The market of food additives for baking industry, *Food ingredients. Raw materials and additives*, 1, 19, (2010)

[5]. Acrylamide. The toxicological, epidemiological, and dietary exposure evaluation of compounds on the agenda, *Evaluation of certain food contaminants: sixty-fourth report of the Joint FAO/WHO Expert Committee on Food Additives*, Geneva, 9-20, (2005)

[6]. Code of Practice for the Reduction of Acrylamide in Foods, Codex Alimentarius, CAC/RCP 67-2009, http://www.codexalimentarius.org/download/standards/11258/CXP_067e.pdf. Last accessed: 07.28.2014

[7]. BYKOVCHENKO T.V., KOSTIUCHENKO M.N., VOLOHOVA L.T., The modern system of bread ropiness prevention and ensuring the microbiological purity of bakery products, *Breadmaking of Russia*, 2, 26-27, (2013)

[8]. KOSOVAN A.P., The general idea and systematic ecologization of bread industry, *Storage and processing of agricultural raw materials*, 12, 34-36, (2008)

[9]. SIDOROVA O.A., BERDYSHNIKOVA O.N., The chemical methods of improving the microbiological purity of bakery products, *Baking industry*, 2, 29-37, (2011)

[10]. BERDYSHNIKOVA O.N., SIDOROVA O.A., The influence of sourdoughs cultivated on different nutrient mediums on ensuring the microbiological safety of bread products, *Baking industry*, 5-6, 10-14, (2011)

[11]. KOVALIOV V.M., Pharmacognosy with the basics of plant biochemistry, Prapor, Kiev, 705 p, (2007)

[12]. NIELSEN P.V., RIOS R., Inhibition of fungal growth on bread by volatile components from spices and herbs, and the possible application

in active packaging, with special emphasis on mustard essential oil, *International Journal of Food Microbiology*, 2-3, 219-229, (2000)
[13]. IORGACHEVA YE.H., LEBEDENKO T.YE., Potential of medicinal and aromatic plants to increase the quality of wheat bread, *Eastern-European Journal of Enterprise Technologies*, 2/12(68), 101-108
[14]. BAHRIANTSEVA O.V., SHATROV H.N., KHOTIMCHENKO S.A., About the question of necessary implementation of measures of lowering the acrylamide content in food products, *The materials of XII All-Russian congress of dietitians and nutritionists "Nutrition and health"*, 10-11, (2010)
[15]. LEBEDENKO T., KOZHEVNIKOVA V., NOVICHKOVA T., Prospects of improvement of accelerated bread technologies by usage of dogrose and hawthorn, *Technology audit and production reserves*, 3, 8-11, (2013)
[16]. LEBEDENKO T.YE., SOKOLOVA N.YU., Polyfunctional usage of hop in bread industry, *Food industry: science and technology*, 1, 40-50, (2014)
[17]. LIASHENKO N.I., Biochemistry of hop and hop by-products, Polesie, Zhitomir, 385, (2002)
[18]. KUZNETSOVA YE.A., KORIACHKINA S.A., KHMELEVA YE.V., The usage of hop extract in order to improve the microbiological safety of grain during its pre-processing for preparation of grain bakery products, *Fundamental research*, 12, 100-103, (2010)
[19]. NILOVA L., DUBROVSKAIA N., New additive for bakery products is a powder from graded ashberry, *Bread products*, 11, 52-53, (2008)
[20]. PASCHENKO L.P., KOLOMNIKOVA YA.P., Bread ropiness. New methods of diagnosis and prevention, *Bread business*, 3, 32-35, (2009)