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FEATURES OF GRAIN GERMINATION WITH THE USE OF AQUEOUS SOLUTIONS OF FRUIT ACIDS

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Abstract. The paper describes some aspects of producing germinated cereal grain with the use of fruits acids of various concentrations. The technological process of grain germination (including washing grain, disinfection, hydration by alternate steeping and air resting, germination, and drying) has been analysed and studied. To intensify germination and disinfection of grain of various crops at the steeping stage, aqueous solutions of organic acids (citric acid, malic acid, and inactive tartaric acid) were used. The results of studying the effect of these organic acids on the energy and germination ability of grain, the flouriness of malt grain, the amino acid composition of malt, the microbiological status of grain have been presented. For each grain crop, concentrations have been determined of active substances in the solutions that activate germination and reduce the time of malt production. A higher flouriness of malt grain, being an important brewing parameter, has been shown. The effect of the considered growth stimulants on the amino acid composition of the finished product has been studied. An increase in the amino acids content indicates splitting the grain endosperm more deeply and obtaining a biologically valuable food product rich in essential amino acids. While studying the microbiological status of grain with the use of the said organic acids, their disinfecting ability towards the pathogenic microflora of the grain was observed. It has been found that the suggested method of intensifying grain germination allows obtaining eco-friendly products in a shorter time, namely, 3–6 days depending on the grain crop. It has been established that, compared to the classical technology, using fruit acids as growth stimulants allows obtaining malt of higher quality, rich in biologically active substances. These grain raw materials can be used both to brew beer and to make highly nutritious healthy food.

Key words: grain, germination, growth stimulant, organic acids, flouriness, amino acid composition.

ОСОБЛИВОСТІ ПРОРОЩУВАННЯ ЗЕРНА З ВИКОРИСТАННЯМ ВОДНИХ РОЗЧИНІВ ФРУКТОВИХ КИСЛОТ

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Анотація. У роботі наведено особливості виробництва пророщеного зерна злаків із використанням фруктових кислот різної концентрації. Проаналізовано та досліджено технологічний процес пророщування зерна, що включає миття, дезінфекцію, почергове повітряно-водяне замочування зерна злакових культур, його пророщування і сушіння. Із метою інтенсифікації процесу пророщування і дезінфекції зерна різних культур на етапі замочування, було використано водні розчини органічних кислот − лимонної, яблучної та виноградної. Показано результати дослідження впливу даних органічних кислот на такі показники як енергія та здатність проростання зерна, борошнистість солодового зерна, амінокислотний склад солоду, мікробіологічний стан зернового матеріалу. Визначено концентрації діючих речовин в розчинах для кожної зернової культури, за яких має місце активація процесу проростання і скорочення часу технологічного процесу виробництва солоду. Показано підвищення борошнистості солодового зерна, що є важливим технологічним параметром для процесу пивоваріння. Вивчено вплив представлених стимуляторів росту на амінокислотний склад готового продукту, збільшення вмісту амінокислот свідчить про більш глибоке розщеплення ендосперму зерна і отримання біологічно цінного продукту харчування багатого незамінними амінокислотами. При дослідженні мікробіологічного стану зернового матеріалу з використанням представлених органічних кислот, відмічено їхню дезинфікувальну здатність по відношенню до патогенної мікрофлори зернового матеріалу. Виявлено, що запропонований спосіб інтенсифікації пророщування зерна дозволяє отримати екологічно чистий продукт в більш короткий термін, а саме, 3–6 діб в залежності від зернової культури. Встановлено, що у порівнянні з класичною технологією, використання фруктових кислот в якості інтенсифікаторів росту дозволяє отримати більш якісний солод, багатий біологічно активними речовинами. Таку зернову сировину доцільно використовувати в приготуванні пива, при виробництві висикопоживних і корисних харчових продуктів оздоровчого призначення.

Ключові слова: зерно, пророщування, стимулятор росту, органічні кислоти, борошнистість, амінокислотний склад.

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Introduction. Formulation of the problem

Germinated grain (malt) is the basic raw material used in production of beer, kvass, alcohol, single-malt and poly-malt extracts. Besides, malt is used in bread-baking, yeast-making and production of special flour, food additives, dairy products, and coffee substitutes. It is due to the fact that malt has valuable nutritional properties and positively effects on the human body. A malt extract contains a number of vital soluble substances and microelements contained in grain. Malt contains phosphorus, magnesium, selenium, calcium, manganese, B group vitamins and vitamin E. Malt has a lot of valuable protein with essential amino acids that stimulate protein metabolism, growth, and development of muscles. Malt is a remedy for gastrointestinal diseases due to insoluble gluten in its composition – it stimulates digestion and cleans the body of toxins.

The traditional technique of malt making includes 2–3 days of steeping and 5–8 days of germination [1-4], in which period many dry substances and active enzymes are lost. This technology, certainly, allows obtaining high-quality malt, but modern industrial production is rapidly growing and constantly requires increase in productivity. All processes involved in malt germination are labour-intensive and time-consuming; therefore, it is advisable to search for innovative and energy-saving technologies of malt production.

Analysis of recent research and publications

The principal methods used to intensify grain germination are: biotechnological, chemical, physical, physicochemical, integrated, etc.

Chemical intensifiers are solutions of organic and inorganic acids of various origin. Biotechnological growth stimulants include phytoregulators, growth regulators, chitosan, Cylloveredin G20H, Disticim P7 [5].

Physical methods of stimulation include vacuum, ultrasonic, ozone treatment, ultraviolet, red, and infrared radiation, cavitation germination, stimulation with microcurrent, sparging [6-7].

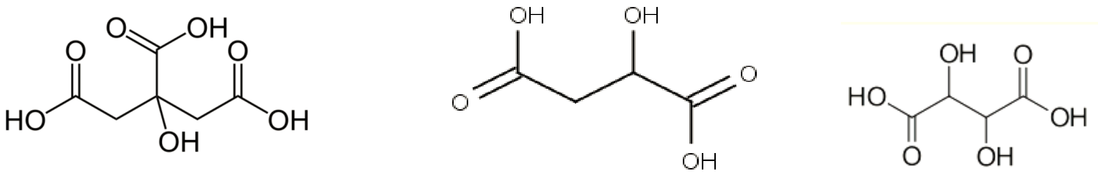
Plasma-chemically activated aqueous solutions [1-2] are used for intensification of the process of malt germination as well.

The most effective method of intensification of grain germination is the use of bio-stimulants along with inhibitors. They accelerate the loosening of endosperm cell walls, promoty the accumulation of gibberellic acid and reducy the time of germination and production of malt [5].

Among the most common organic acids are lactic, gibberellic, succinic, ferulic acids, coumarin, nicotinic and folic acids [8-10].

Much attention is paid by scientists to the products of nanotechnologies – nanopowders of metals. They act as bactericides and as a source of microelements. Besides the above mentioned growth stimulants, alkyl esters of arachidonic, eicosapentaenoic, or jasmine acid are used in the presence of an antioxidant [11-18].

Nevertheless, the above substances have certain disadvantages. Their high cost and insufficient effectiveness make scientists search for a substance that can meet all the requirements. Besides, special attention should be paid to environmental and safety factors of using intensifier. Supposedly, this problem can be solved by using aqueous solutions of citric acid (Fig. 1a) acting as an intermediate carrier of hydrogen atoms at the initial stages of oxidation of carbohydrates and fatty acids, reducing the product' the pH and the development of bacteriological background. It is also involved in metabolism of fats, proteins, amino acids, etc. [19]. Hydroxybutanedioic (malic) acid (Fig. 1b) has hygroscopic properties, acts as an antioxidant, oxidizes carbohydrates, and stimulates metabolism, being responsible for strengthening the immune function. Paratartaric acid (racemic compounds) (Fig. 1c) taking part in cellular respiration, improves elasticity and is capable of expanding water-capillary channels, decelerates decay processes, protects against oxidation of carbohydrates, helps to strengthen proteins, stimulating growth and increasing the rate of metabolism of agricultural crops [20]. Another major advantage of these acids is their comparably low cost.



a b c

Fig. 1. Fruit acids: a – citric acid; b – malic acid; c – inactive tartaric acid

Purpose of the study is the research of peculiarities of obtaining germinated malt with the use of aqueous solutions of citric, malic, and inactive tartaric acids of various concentrations.

Objectives of study:

1. Selecting some concentrations and study the effect of selected fruit acids (citric, malic, and inactive tartaric

acids) on the process of germination of grain and its quality characteristics;
2. Determining the amino acid composition of germinated grain of different crops;
3. Assessing the disinfecting effect of fruit acids on the microbiological characteristics of grain.

Research materials and methods

As a material for research we used the wheat variety Podolyanka (seeds of this variety are red, egg-shaped, with a shallow crease and a medium-sized floccose apex; 1000 grains weigh 43.8–45.7 g), the maize variety Liubava (yellow seeds of a tooth-like flinty shape, 1000 grains weigh 290–300 g), the barley variety Ascold (yellow, elongated-oval seeds, 1000 grains weigh 42–48 g), the rye variety Slavuta (semi-elongated seeds, 1000 grains weigh about 38.2 g, grain contains 15.1–16.5% of protein).

As growth stimulants, citric acid (E 330), malic acid (E 296), and inactive tartaric acid (E 334) were used. These acids had the following characteristics: citric acid – monohydrate, pure, made in China; malic acid – monohydrate, pure, made in China; inactive tartaric acid – monohydrate, pure, made in China. All acids used in the study belong to food acids according to the international classification.

Before the research, the quality parameters of the grain samples were established to meet the requirements of the current normative and technical documentation: sensory characteristics, cup weight, contamination by microorganisms and grain pests, content of impurities, humidity. These were determined according to the requirements of State Standard of Ukraine 4138:2002. The grain germination followed the methods described in this standard; the flouriness of malt grains was determined according to State Standard of Ukraine 4282:2004.

In order to determine how effectively these growth stimulants influence the energy and the germination ability, portions of 500 grains were formed. As the liquid for grain steeping, aqueous solutions of citric, malic, and inactive tartaric acids were used in concentrations ranging 0.25% to 1.5%.

The grain was germinated in the laboratory malt house – in a set of plastic containers covered with a layer of filter paper and soaked in aqueous solutions of acids of the appropriate concentration.

The grain material was treated with aqueous solutions of organic acids as follows. Grain to be germinated was saturated with a solution of acid of specified concentration in two stages. The preliminary steeping was performed during 4 hours at the temperature 18–20°C. Then, the nutrient solution was drained, and the grain was kept for 18 hours with no access of liquid. When repeating the steeping, acids solutions of similar concentrations were used. To prevent acidification of the solutions, at the second stage of steeping, alkali solutions were added. Air and water steeping lasted 26 hours until cereal grains were fully saturated with the preparation. The germination lasted 3–7 days at the temperature 17–21°C, occasionally wetting and stirring 45–55 mm of the grain layer in order to distribute uniformly the liquid and to avoid caking. The final stage of the process is drying of the germinated material to the constant humidity of 5–6%.

After 72 hours since the steeping finished, the germination energy of the grain material was determined, and after 120 hours, the germination ability was established. These characteristics are expressed as the percentage of the total quantity of grains in the weighed portion. The effectiveness of the selected growth stimulants was compared to that of the control (grain not exposed to any chemical treatment). After completion of malt drying, to determine the effect of citric, malic, and inactive tartaric acids on the quality characteristics of malt grain, we studied the consistency of the endosperm (flouriness). Flouriness (glassiness) of malt grain was determined by the standard method on a diaphanoscope DSZ-2M [21]. The amino acid composition was determined by ion-exchange liquid-column chromatography on an amino acids automatic analyser T 339 (made in the Czech Republic, Prague) [22]. Changes in microbiological status of grain material were registered with a microscope MBS-56 by photorecording; the wipe samplings were inoculated on the nutrient medium, with the further counting of microorganism colonies [23].

Results of the research and their discussion

The main indicators of malt quality helpful in establishing the feasibility of using the selected fruit acids are the germination energy and the germination ability of the grain material (Table 1). The acid concentrations were taken as: citric acid 1.25–1.0, malic acid 1.0–0.75, inactive tartaric acid 0.5–0.25. It was conditioned by the fact that in the previous studies, a more pronounced effect of acids had been observed [20]. Besides, these concentrations caused no damage to grain during the experiments or changes in the sensory properties of the finished product.

After analysing the results presented in Table 1, it can be concluded that aqueous solutions of fruit acids in various concentrations act differently on each grain crop. Due to the experimental studies, it has been found that almost in all cases, the best results were achieved with the use of the aqueous solution of inactive tartaric acid. For example, for the maize (concentration – 1.5%), the germination energy and ability increased, respectively, by 8.79 and 7.69%. This solution (with the acid concentration 0.5%) improved the quality characteristics of wheat by 9.88% (germination energy) and 13.95% (germination ability), and of rye, by 15.00% and 7.61%, respectively. A positive effect on the malting process in barley is observed in the aqueous solution of inactive tartaric acid (with the active substance concentration 0.75%); here, an increase in the germination energy reached 12.05%, and the germination ability 11.49%. An exception was the use of aqueous solutions of malic and inactive tartaric acids with the concentration 0.75%. They featured good results of both the germination energy (9.88%) and the germination ability (8.88%). The aqueous solution of citric acid, with the concentration of active substance 1.25%, had a positive effect on rye. So, we can speak of fruit acids acting directly upon the grain of various crops as stimulants. It is explained by active action of acids on grain hulls resulting in more active moistening of seeds, and thus, in activation of the complex of hydrolytic enzymes.

To represent the effect of aqueous fruit aci solutions of on the malt grain endosperm, we took experimental samples with a higher content of floury grains. Flouriness is an important process characteristic since it reflects the degree of dissolution of the malt grain endosperm, being one of the basic criteria of malt assessment for its further use in the brewing process [24]. The effect of aqueous fruit acid solutions on the flouriness of grain is shown in Table 2.

Based on results of the study, we can draw a conclusion that when the aqueous solution of malic acid acts upon the flouriness of wheat grain, the highest effect (47%) can be achieved. With inactive tartaric acid and malic acid used, the flouriness of grain makes, respectively, 43% and 35%. With the use of malic acid, it is possible to achieve the highest result (45%), while due to the use of citric acid and inactive tartaric acid, the effect on the rye grain flouriness is respectively 44% and 42%.

Table 1 – Dependence of the germination energy and the germination ability of grain on the fruit acid concentration

Acid	Acid concentration, %	Germination energy, %	Germination ability, %	Germination energy, %	Germination ability, %
		Wheat		Maize	
Citric acid	1.25	84	93	85	94
	1.0	86	95	84	93
Malic acid	1.0	86	96	90	94
	0.75	87	97	91	98
Inactive tartaric acid	0.5	89	98	93	99
	0.25	88	96	92	98
Control	0	81	86	82	91
		Rye		Barley	
Citric acid	1.25	86	96	86	93
	1.0	87	97	88	96
Malic acid	1.0	87	94	89	97
	0.75	90	98	91	98
Inactive tartaric acid	0.75	89	96	93	97
	0.5	92	99	92	94
Control	0	80	92	83	87

Table 2 – Effect of aqueous fruit acid solutions on the flouriness of grain

Acid	Acid concentration, %	Floury, %	Semi-glassy, %	Glassy, %
Maize				
Citric acid	0.75	22	46	33
Malic acid	0.75	35	46	19
Inactive tartaric acid	1.25	28	25	47
Control	-	32	36	32
Rye				
Citric acid	1.0	45	35	20
Malic acid	1.0	32	47	22
Inactive tartaric acid	0.5	31	43	26
Control	-	35	28	37
Wheat				
Citric acid	1.0	30	44	26
Malic acid	0.75	41	45	14
Inactive tartaric acid	0.5	34	42	24
Control	-	35	26	39
Barley				
Citric acid	1.0	38	40	22
Malic acid	0.75	34	46	20
Inactive tartaric acid	0.5	37	45	18
Control	-	38	28	34

After analysing the experimental data, we can conclude that the best effect on the flouriness of barley is by the aqueous solution of malic acid (46%). When using inactive tartaric acid and malic acid, the figures for barley grain flouriness are 45 % and 40 %.

Changes in the composition of amino acids in malting of cereals are presented in Table 3. The content of amino acids in germinated grains obtained with the use of fruit acids increases (Table 3). It is due to the fact that with the intensification of germination, grain proteins are more actively split into amino acids [24]. Besides, we have shown that the process of germination was reduced, and as a result, amino acids accumulated steadily, but not resynthesized as in the case of long-term germination, i. e. no process of "re-solution" was observed.

So, the usage of fruit acids increases the content of amino acids in the finished product making it more valuable from the biological point of view.

Disinfecting abilities of fruit acids have been studied as well. The microbial population has been calculated by counting the colonies revealed on the standard media. Table 4 includes average data on the use of fruit acids under study.

It should be noted that the disinfecting ability was more pronounced in the samples with higher concentration of fruit acids. It can be explained by the acidic environment adversely affecting the pathogenic microorganisms located on the grain surface [5,24].

Table 3 – Changes in the content of amino acids in malting of cereals with the use of fruit acids, mg/100 g*

Amino acids	Wheat			Barley			Rye			Maize		
	grain	malt (contr.)	malt (experim.)	grain	malt (contr.)	malt (experim.)	grain	malt (contr.)	malt (experim.)	grain	malt (contr.)	malt (experim.)
Lysine	3	11	12	4	25	27	3	19	21	4.5	10	11
Histidine	4	8	9	2	23	24	3	20	19	4	23	24
Arginine	6	25	26	4	52	54	9.5	37	38	5	17	18
Aspartic acid	8	17	18	18	21	22	10	20	21	20	15	16
Threonine	0	5	6	2	19	20	1	14	15	0.9	12	13
Serine	1	7	8	2	21	22	2	16	17	4	20	21
Glutamic acid	8	39	40	27	46	47	29	37	39	28	42	43
Proline	3	50	52	6	229	230	0.9	56	57	21	33	34
Glycine	2	1	2	2	7	8	0.8	3	4	0.9	6	7
Alanine	5	17	19	6	37	38	3	19	20	5	27	28
Cystine	1	2	3	1	6	6	1	1	2	0.1	2	3
Valine	1	11	14	3	46	47	0.5	21	22	0.3	18	19
Methionine	1	2	4	0	8	9	0.2	2	3	0.2	11	13
Isoleucine	0	8	9	1	28	29	0.2	15	16	0.1	9	10
Leucine	1	11	12	2	42	42	0.9	23	24	2	40	41
Tyrosine	1	10	11	2	30	31	3	22	23	4	33	34
Phenylalanine	1	15	17	1	44	45	2	25	26	2	30	31
Glutamine	2	48	51	4	103	104	3	31	30	2	61	62

***** An average index for a crop with the use of fruit acids

Table 4 – Research of the microbiological status of grain during germination

Concentration of fruit acids, mg/l	Microorganisms	
	Control	Experiment
1.5	$3.1*10^{2}$	<10
1.25	$5.2*10^{3}$	$6.1*10^{2}$
1.0	$7.5*10^{4}$	$2.1*10^{3}$
0.75	$8.3*10^{5}$	$3.3*10^{4}$
0.5	$4.7*10^{6}$	$4.3*10^{5}$
0.25	$2.3*10^{7}$	$3.3*10^{6}$

Evaluation of the results of the study. All studies were performed on the basis of the Scientific and industrial laboratory on assessment of grain and grain products quality of the Dnipro State Agrarian and Economic University. The technology of production of malt of cereal grain using aqueous solutions of citric, malic, and inactive tartaric acids is environmentally safe and effective, and this technology can be industrially implemented.

Conclusions

The chemical method of intensifying germination with the use of fruit acids allows obtaining an environmentally friendly product since fruit acids do not leave any unwanted chemical compounds in the grain. Besides, the intensifying effect on the grain during germination allows obtaining the finished product in a shorter time (3–6 days depending on the grain crop) and increasing the flouriness in the malt batch. During the experiments, it has been established that:

1. The most pronounced effect in malt germination can be achieved with the use of inactive tartaric acid for maize – 1.5%, for wheat and rye – 0.5%, for triticale – 1.0%, whereas for oats, both aqueous solutions of inactive tartaric acid and malic acid with the active substance concentration 0.75% can be used.
2. The germination energy and the germination ability of cereals increase on average by 5.7% with citric acid used; by 8–9% with malic acid; by 9–10% with inactive tartaric acid.
3. The amino acid composition of germinated grain has been studied, and it has been found that using fruit acids increases the number of amino acids compared to the control.
4. It has been found that with the increase in the concentration of the active substance, the disinfecting abilities
5. become more active.

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