UDC: 663.533

SELECTION OF THE FERMENTATION TECHNOLOGICAL PARAMETERS OF HIGH-CONCENTRATION WORT BY OSMOPHILIC YEAST RACES FOR OBTAINING BIOETHANOL

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DOI: https://doi.org/10.15673/fst.v15i3.2116

Article history

Received 15.04.2021
Reviewed 28.05.2021
Revised 21.07.2021
Approved 31.08.2021

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Cite as Vancouver style citation

Kovalchuk S, Mudrak T. Selection of the technological parameters of fermentation of high-concentration wort with osmophilic yeast races for obtaining bioethanol. Food science and technology. 2021;15(3):15-22. DOI: https://doi.org/10.15673/fst.v15i3.2116

Цитування згідно ДСТУ 8302:2015

Kovalchuk S., Mudrak T. Selection of the technological parameters of fermentation of high-concentration wort with osmophilic yeast races for obtaining bioethanol // Food science and technology. 2021. Vol. 15, Issue 3. P. 15-22. DOI: https://doi.org/10.15673/fst.v15i3.2116

Abstract. Bioethanol production is a key issue that helps meet the growing demand for energy resources and ensure a sustainable economy. A promising direction is producing bioethanol by using the technology of fermentation of grain high-concentration wort. The purpose of this work is researching of the fermentation process of the high concentrations wort by distiller's yeast osmophilic races at high acidity and selection of technological parameters for bioethanol production. Selective breeding of a new strain of the yeast *Saccharomyces cerevisiae* DO-16 has allowed obtaining ethanol producers able to ferment grain wort with the dry matter concentration 24–34% at pH 3.0–6.0, with alcohol accumulation in the fermented wash up to 17% vol. It has been studied how the pH of wort affects the dynamics of yeast cell synthesis by the distiller's yeast races *Saccharomyces cerevisiae* DO-11 and *Saccharomyces cerevisiae* DO-16. It has been established that at the pH values 2.5, 3.0, 3.5, and 4.0, the concentration of yeast cells in the race *Saccharomyces cerevisiae* DO-16 was higher by 2.6, 1.7, 1.5, and 1.4 times respectively, as compared with *Saccharomyces cerevisiae* DO-11. It has been found that culturing industrial yeast of these races at low pH values will provide not only the required sterility of the substrate, but also a high content of yeast cells, which is 250–320 million/cm³. The chemical and technological parameters of the fermented wash obtained by using the yeast races *Saccharomyces cerevisiae* DO-11 and DO-16 at the wort concentration 20–34% DM have been studied. It has been found that under all research conditions, the yeast of the race *Saccharomyces cerevisiae* DO-16 synthesised more ethanol than the strain *Saccharomyces cerevisiae* DO-11 did. The use of a new high-productive strain of *Saccharomyces cerevisiae* DO-16 will allow fermenting wort with a high ethanol concentration in the wash. It will also reduce the consumption of heat expended on isolating alcohol from the wash and of water expended on cooling, and lessen the amount of post-alcohol stillage.

Keywords: *Saccharomyces* ***cerevisiae*****,** highly concentrated wort, bioethanol, fermentation, osmophilic distiller's yeasts.

Introduction. Formulation of the problem

To meet the growing demand for energy resources and ensure a sustainable economy, the key issue is the production of bioethanol [1,2]. Today, there are a number of studies aimed at increasing bioethanol production in the world [2,3]. To manufacture first-generation bioethanol, promising raw materials are cereals, in particular maize, a significant amount of which is processed into bioethanol every year [4]. New directions in the development of the bioethanol technology require increased dry matter concentrations in the wort and fermentation under conditions when the temperatures, acidity, and ethanol concentration in the wash are high. For bioconversion of components of grain raw materials, it is necessary to develop technological parameters and to select highly productive races of alcoholic yeast with thermotolerant and acid-resistant properties.

Analysis of recent research and publications

During the last decades of the twentieth century, there was a huge interest in the production and use of liquid biofuels (biodiesel or bioethanol) as promising alternatives to fossil fuels. Biofuels produced from plant biomass are a renewable energy source. The use of these raw materials would reduce the consumption of fossil fuels and, consequently, the negative impact on the environment [5,6]. Bioethanol production is an integrative and multifunctional concept that uses biomass for sustainable production of various intermediates and final products, as well as for the fullest possible use of all components of raw materials [7-9]. Generally, bioprocessing of grain raw materials usually includes the following stages: pre-treatment and preparation of biomass, separation of biomass components, and subsequent stages of conversion and purification of the product [10-12].

When applying the high-concentration wort fermentation technology, a number of problems arise from the incomplete hydrolysis of raw material components. Selective breeding of highly productive distiller's yeast races is work that never stops. A topical problem is development of rational parameters of fermenting wort from grain raw materials to obtain bioethanol. One of the important tasks is to increase the efficiency of the fermentation department.

Installing additional equipment in the fermentation department in order to increase the capacity of a plant is not profitable. A promising direction is the fermentation of wort with high dry matter concentrations from grain raw materials to obtain bioethanol [13]. Today, enterprises use the filtrate of post-alcoholic stillage to make mash, which causes an acidity increase. The resource- and energy-saving technology of fermentating high-concentration wort involves fermentation at elevated temperatures and an increased initial dry matter concentration in the wort [14]. Under such conditions, it is necessary to use races of distiller's yeast with thermotolerant, osmophilic, alcohol-resistant, and acid-resistant properties.

The physiological and biochemical activity of yeast is of great importance for the fermentation of high-concentration wort. The high concentration of dry matter and ethanol in the stillage and a temperature increase during fermentation inhibit the activity of yeast cells and reduce their fermentation activity [15]. These factors cause a number of disorders in cell metabolism, which leads to inefficient use of carbohydrates of wort, reduced ethanol yield, and increased cost of production, as well as to the low quality of alcohol due to an increasd content of by-products in it (fusel oil, organic acids, aldehydes, etc.) [16-17].

Increasing the fermentation temperature and osmotic pressure of the medium leads to the creation of extreme conditions for the activity of yeast [18]. This can decrease the regenerative and fermentative activity of the yeast, which, in turn, causes instablility in the work of the fermentation department.

To assimilate the nutrients of the medium, the intracellular pressure in a yeast cell must be higher than the osmotic pressure of the medium, because when the osmotic pressure in solutions of salts, sugars, and other substances increases, plasmolysis of yeast cells occurs [19-20].

The pH of the environment significantly affects the viability of yeast cells. Yeast cells can remain viable in a wide range of pH, but the optimal acidity for culturing most strains is 4.0–5.5. The pH of the medium affects the permeability of the shell to individual substances and ions, and accordingly affects the assimilation of nutrients, amino acid metabolism, respiration, the formation of vitamins [17,21]. Changes in the pH of the medium affect the composition and concentration of fermentation by-products. In an alkaline environment, the formation of glycerin and other secondary fermentation products increases [21-23].

For most strains, the optimal value of active acidity is 4.0–5.5. High pH values 6.0–7.0 result in foaming and development of foreign microflora. In turn, at the pH values 8.0–10.0, yeast cells swell, which adversely affects the life processes. To prevent bacterial infection, distilleries use the method based on keeping an aqueous suspension of yeast cells in solutions of hydrochloric and sulphuric acids at the pH values 1.9 and 1.6. It has been found that the effect of organic and inorganic acids on yeast cells is different. Yeast cells are more tolerant to organic acids (lactic, propionic, citric and others) [17,24].

The active acidity of the medium (pH) in alcohol production affects the rate of biosynthesis of a yeast culture and on the synthesis of fermentation products from raw sugars.

Besides, the active acidity of the medium affects the formation of metabolic products. Many researchers have studied how the formation of products of alcoholic yeast fermentation changes depending on the pH of the wort during fermentation [21,24].

During fermentation, an increase in the pH of the medium contributes to the increased accumulation of yeast biomass and glycerol with a simultaneous decrease in the level of alcohol formation, and a decrease in the pH value has the opposite effect [21]. The activity of enzymes, which is the basis of biochemical activity of microbes, depends on the reaction of the medium. For example, it is known that during the fermentation of sugar, the same yeast in an acidic environment forms a lot of ethanol and a small amount of glycerol. On the contrary, in an alkaline substrate, they form a large amount of glycerol and a small amount of ethanol from sugars [25]. It is necessary to investigate how the pH of the culture medium of industrial yeast determines the morphological properties of selectively bred osmophilic, acid-resistant yeast races, and to choose the most practical technological parameters of fermenting high-concentration wort.

The novelty of the work lies in studying and developing practical technological parameters of culturing and fermenting high-concentration wort by osmophilic acid-resistant race of the distiller's yeast *Saccharomyces cerevisiae* DO-16.

The purpose of this work is researching of the fermentation process of the high concentrations wort by distiller's yeast osmophilic races at high acidity and selection of technological parameters for bioethanol production.

Research objectives:

1. To study how the pH of the culture medium of industrial yeast affects the morphological properties of distiller's yeast races.
2. To investigate the chemical and technological parameters of fermented wash depending on different concentrations of dry matter in the wort, the fermentation temperature, and the pH of the wort.

Research materials and methods

The raw material used for the research was milled maize grain containing 69.0% of starch, with particle finess 100% of the undersize from a screen with the mesh diameter 1 mm.

The enzyme preparations used for the research were produced by Danisco. *Amylex 4T* was used as α-amylase, *Diazyme TGA* as glucoamylase, *Alphalase AFP* as the source of protease, *Laminex 750* as the source of cellulose. Enzyme preparations were added by units of activity. The dosage of the enzyme preparations was as follows. Amylolytic: α-amylase (0.8 units AA/g of starch), glucoamylase (7.5 units GA/g of starch); cellulolytic (0.35 units CA/g raw materials); proteolytic (0.05 units PrA/g raw materials).

To prepare the grain mash, a 50–70 g portion of milled grain was placed in a 500 cm³ conical flask, and 150–130 cm³ of water was added. The mixture was stirred, and working solution of the diluent enzyme preparation was added.

Wort was prepared according to the low-temperature cooking scheme at 85–92°C using concentrated enzyme preparations, with 3 hours of exposure. The diluted mass was cooled to 50–55°C and saccharified with glucoamylase for 0.5 h. Fermented wash was obtained after 3 days of fermentation of the wort at 34–37°C.

Osmophilic strains of the yeast *Saccharomyces cerevisiae* DO-11 and *Saccharomyces cerevisiae* DO-16 were selectively bred for microbiological synthesis of ethanol at the Department of Biotechnology of Fermentation Products and Winemaking of the National University of Food Technologies. Technological evaluation of the qualities of *Saccharomyces cerevisiae* DO-11 strains and *Saccharomyces cerevisiae* DO-16 strains was carried out in a laboratory environment. Fermentation of highly concentrated wort results not only in the synthesis of basic products and by-products of fermentation, but of organic acids, too [17,18].

Under industrial conditions, the mash preparation technology involves using stillage filtrate. Its use also causes an increase in the acidity of the wort [18]. It is important that the yeast can withstand not only a high concentration of wort and increased fermentation temperature, but acidity, too.

Yeast was cultured at 30°C, with the concentration of dry matter in the wort 28.0%. The yeast inoculum was introduced in the proportion 20 million/cm³ of wort. In the laboratory, the wort was fermented by the method of "fermentation test" in conical flasks with sulphuric acid seals in an incubator where the temperature was maintained at 30–38°C. The volume of a test sample was 0.2 cm³. Fermentation involved using osmophilic races of *Saccharomyces cerevisiae* IMB Y-5099 (DO-16) [26] and *Saccharomyces cerevisiae* DO-11 [27].

The moisture content of grain was determined by the method of complete desiccation (ISO 712:2007). The particle size distribution of the milled grain was determined by sifting on metal and nylon-6 sieves (DSTU 4525:2006). The starch content of the original grain was determined by the Ewers method [28]. The dry matter concentration was determined on a food laboratory refractometer of the RPL-3 type [28]. In the fermented wash, the pH and active acidity were determined by potentiometry using a pH meter MI. the ethanol content was measured by the pycnometric method and the refractometric method, using an immersion refractometer [28]. The amounts of soluble carbohydrates, insoluble starch, alcohol-soluble carbohydrates, and dextrins were measured photoelectrocolorimetrically (using a photocolorimeter KFK-2) with the anthrone reagent [28]. The total number of yeast cells in 1 cm³ was determined by direct counting in the counting chamber.

All quality indicators were determined in triplicate with subsequent statistical analysis of the results.

Results of the research and their discussion

It has been studied how the pH of wort affects the dynamics of yeast cell synthesis by distiller's yeast races *Saccharomyces cerevisiae* DO-11 (Fig. 1a) and *Saccharomyces cerevisiae* DO-16 (Fig. 1b). The wort was acidified to pH 2.5, 3.0, 3.5, 4.0, 4.5, and 5.0 with sulphuric acid (h. H.).

It has been established that at the pH values 2.5, 3.0, 3.5, and 4.0, in the race *Saccharomyces cerevisiae* DO-16, the concentration of yeast cells was higher by 2.6,1.7, 1.5, and 1.4 times respectively, compared with the races under study (Fig. 1a). This confirms the acid resistance of the selectively bred race of the distiller's yeast *Saccharomyces cerevisiae* DO-16. It is known that under unfavourable conditions, the growth and reproduction of yeast cells stops, which was studied in the works by L. Rymareva and S. Oliinichuk. The pH of the culture medium significantly affects the rate of assimilation of nutrients, the intensity of respiration, the activity of cell enzymes [17,25].

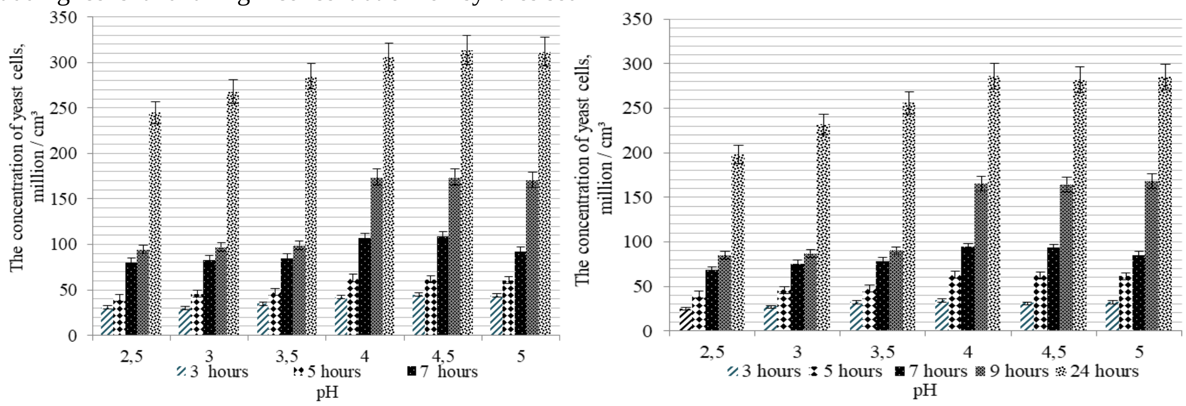
Culturing industrial yeast of the distiller's yeast race *Saccharomyces cerevisiae* DO-16 at low pH values will ensure not only the required sterility of the substrate, but also a high content of physiologically active yeast cells, which is 250–320 million/cm³. The race *Saccharomyces cerevisiae* DO-16 has better acid resistance than the race *Saccharomyces cerevisiae* DO-11. That confirms the practical importance of using this distiller's yeast race industrially for fermentation of high-concentration wort to obtain bioethanol.

Fig. 2a and 2b show how the pH of the culture medium at the values 2.5 affects the morphological characteristics of the osmophilic yeast races under study

A yeast cell is a complex biological system, in which a lot of metabolic processes take place. All biochemical reactions that occur in a living cell are strictly localised. Cytomorphological characteristics of a culture clearly reflect the physiological state [9,15]. The study of the morphological characteristics of races *Saccharomyces cerevisiae* DO-16 and *Saccharomyces cerevisiae* DO-11 shows that at the wort dry matter concentration 28% and the pH values of the culture medium 2.5, the condition of cells of the race *Saccharomyces cerevisiae* DO-16 is satisfactory. This indicates that the race is adaptable to increased pH values of the medium, and confirms its acid-resistant properties. The studies by D. Gomes, M. Cruz, M. de Resende, E. Ribeiro, J. Teixeira, and L. Domingues focus on morphophysiological properties of yeast cells under the conditions of osmotic, acid stress. The works by L. Rymareva and S. Oliinichuk consider the effect of the acidity of the culture medium on the physiological and biochemical properties of distiller's yeast. Yeast cells of *Saccharomyces cerevisiae* DO-16 remain viable at high acidity of the culture medium, and a significant number of budding cells and a high concentration of synthesised cells are observed (Fig. 2b). This is evidence of the acid-resistant properties of the distiller's yeast race *Saccharomyces cerevisiae* DO-16. The studies confirm the effectiveness of the selected race *Saccharomyces cerevisiae* DO-16 for wort fermentation under the conditions of high acidity, as it ensures the microbiological purity of the fermented medium.

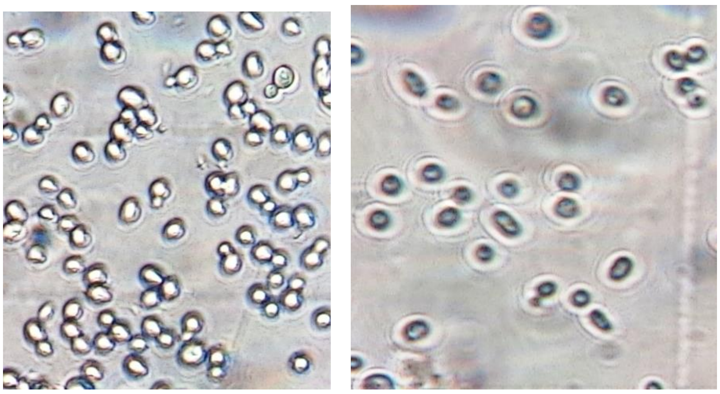
The next stage was studying how the technological parameters of fermenting high-concentration wort by osmophilic races of the yeast *Saccharomyces cerevisiae* DO-11 and *Saccharomyces cerevisiae* DO-16 determined the chemical and technological parameters of fermented washes.

Table 1 shows the content of unfermented carbohydrates and the concentration of alcohol in fermented washes when changing the initial wort concentration from 20 to 34% DM at the fermentation temperatures 34, 36, and 38°C. The data in Table 1 show that when using the yeast races *Saccharomyces cerevisiae* DO-11 and DO-16, an increase in the wort concentration from 20 to 34% DM in the fermented wash resulted in a simultaneous increase in the alcohol concentration from 10.48 to 17.0% vol. However, under all research conditions, the yeast strain *Saccharomyces cerevisiae* DO-16 synthesised more ethanol than the strain *Saccharomyces cerevisiae* DO-11 did.



a *b*

Fig. 1. Effect of the pH of wort on the dynamics of synthesis of yeast cells of the race:
a - *Saccharomyces cerevisiae* DO-11, b - *Saccharomyces cerevisiae* DO-16



a *b*

Fig. 2. Morphological features cells during culturing (pH 2.5, DM 28%) of race:
a - *Saccharomyces cerevisiae* DO-11, *Saccharomyces cerevisiae* DO-16

Table 1 – Parameters of fermented wash during fermentation of wort by the yeast race *Saccharomyces cerevisiae* DO-11 and DO-16 at different values of the dry matter concentration of wort and different fermentation temperatures (n=3, p≤0.95)

No.	Distiller's yeast race *Saccharomyces cerevisiae*	Initial dry matter concentration of the wort, % DM	Content of unfermented carbohydrates, g/100 cm³, at the fermentation temperature, °C:			Concentration of ethanol, % vol., at the fermentation temperature, °C:		
			34	36	38	34	36	38
1.	DO-11	20±0.2	0.31±0.02	0.34±0.02	0.37±0.02	10.48±0.03	10.56±0.03	10.46±0.03
2.	DO-16		0.25±0.02	0.28±0.02	0.29±0.02	10.53±0.03	10.61±0.03	10.51±0.03
3.	DO-11	24±0.2	0.33±0.02	0.33±0.02	0.41±0.02	12.68±0.03	12.79±0.03	12.78±0.03
4.	DO-16		0.27±0.02	0.30±0.02	0.32±0.02	12.88±0.03	12.99±0.03	12.9±0.03
5.	DO-11	28±0.2	0.52±0.02	0.57±0.02	0.61±0.02	15.30±0.03	15.08±0.03	15.07±0.03
6.	DO-16		0.32±0.02	0.40±0.02	0.43±0.02	15.60±0.03	15.49±0.03	15.45±0.03
7.	DO-11	30±0.2	0.52±0.02	0.63±0.02	0.65±0.02	15.96±0.03	16.03±0.03	15.98±0.03
8.	DO-16		0.46±0.02	0.56±0.02	0.58±0.02	16.43±0.03	16.45±0.03	16.35±0.03
9.	DO-11	34±0.2	0.87±0.02	0.90±0.02	0.92±0.02	15.78±0.03	15.88±0.03	15.57±0.03
10.	DO-16		0.53±0.02	0.57±0.02	0.60±0.02	17.00±0.03	16.76±0.03	16.67±0.03

At the initial dry matter concentration 20%, the yeast strain *Saccharomyces cerevisiae* DO-16 synthesised by 0.05% vol. more alcohol than the yeast strain *Saccharomyces cerevisiae* DO-11 did. An increase in the initial concentration of dry matter correlated with the corresponding alcohol content, respectively: at 24% DM – 0.13–0.20% vol., at 28% DM – 0.31–0.42% vol., at 30% DM – 0.38–0.47% vol., at 34% DM – 0.88–1.23% vol.

Yeast of the strain *Saccharomyces cerevisiae* DO-11 synthesised the maximum amount of alcohol (15.96–16.03% vol.) at the wort concentration 30% DM. With an increase in the wort concentration to 34% DM, the concentration of ethanol in its fermented washes decreased to 15.57–15.88% vol., while the test washes with the strain *Saccharomyces cerevisiae* DO-11 had the highest alcohol content – 16.67–17.00% vol. The works by D.Gomes, M. Cruz, M. de Resende, E. Ribeiro, J. Teixeira, L. Domingues, K. M. Sujit, and R. S. Manas investigate the technological parameters of fermentation of high-concentration wort from grain raw materials. The scientists presented various ways of improving the process of bioconversion of raw materials, which is consistent with the presented research results [10,11].

Indicators for the alcohol synthesis mainly coincide with the content of unfermented carbohydrates in fermented washes. In the washes fermented with *Saccharomyces cerevisiae* DO-11 (samples 1, 3, 5 7, 9), their content was always larger, compared with the experimental ones, and the difference increased with increasing concentration of DM in the wort. Under all research conditions, the content of unfermented carbohydrates did not exceed 2% in relation to the carbohydrates introduced for fermentation, which meets the requirements of the effective technological regulations.

The data in Table 1 prove that the selected yeast strain *Saccharomyces cerevisiae* DO-16 is osmophilic and alcohol-resistant, and the normal course of fermentation of the wort at elevated temperatures (34–38ºC) allows classifying it as thermotolerant.

In the next series of experiments, the yeast strain *Saccharomyces cerevisiae* DO-16 fermented wort with the concentrations 20 and 28% DM. For a comparative characteristic of the effect of acidity of the medium under the conditions of high-concentration wort, the studied dry matter concentration 28% is selected. The reference samples are the test samples with the dry matter concentration of wort 20%.

The results of the experiments are given in Table 2. It can be seen that at all pH values, approximately the same amounts of alcohol accumulated in the washes. But the highest alcohol content (15.66% vol.) was in the fermented washes obtained by fermentation of wort with the pH 5.00. At other values of the pH of the wort, this content was lower and tended to decrease with increasing acidity of the wort.

Table 2 – Indicators of fermented wash during fermentation of wort at different pH values and different concentrations of DM (n = 3, p≤0.95)

pH of the wort, units	Content of unfermented carbohydrates, g/100 cm³, at wort concentration, % DM		Concentration of alcohol, % vol., at the concentration of dry matter of the wort, %	
	20.0	28.0	20.0	28.0
6.00±0.02	0.21±0.02	0.36±0.02	10.61±0.03	15.59±0.03
5.00±0.02	0.19±0.02	0.32±0.02	10.69±0.03	15.66±0.03
4.20±0.02	0.26±0.02	0.33±0.02	10.64±0.03	15.58±0.03
3.80±0.02	0.29±0.02	0.36±0.02	10.61±0.03	15.57±0.03
3.60±0.02	0.31±0.02	0.37±0.02	10.60±0.03	15.57±0.03
3.20±0.02	0.31±0.02	0.40±0.02	10.55±0.03	15.54±0.03
3.00±0.02	0.37±0.02	0.42±0.02	10.53±0.03	15.53±0.03

These data correlate with the content of unfermented carbohydrates in fermented washes and are confirmed by other scientists (the works by D. Gomes, M. Cruz, M. de Resende, E. Ribeiro, J. Teixeira, L. Domingues, K. M. Sujit, R. S. Manas, L. Rymareva, S. Olinichuk) [10,11,25].

Probably, the wort pH 5.00 is optimal for the metabolism of the yeast strain *Saccharomyces cerevisiae* DO-16 under conditions of alcoholic fermentation when processing starch-containing raw materials.

The presence of foreign microflora was recorded in none of the samples of fermented washes. Reducing the pH of the wort from 6.00 to 3.00 and increasing the concentration of dry matter of the wort from 20 to 28% at the stage of fermentation allows ensuring not only high sterility of wort and wash, but also the maximum yield of alcohol (up to 15.66% vol.) if stillage filtrate is used at the stage of preparation of grain mashes.

The results of the studies indicate that the new strain of *Saccharomyces cerevisiae* DO-16 has a wide range of action depending on the active acidity of the wort. Thus, the use of the new strain of *Saccharomyces cerevisiae* DO-16 in the production of alcohol from starchy raw materials allows fermenting the wort with the dry matter concentration 34% at pH 6.0–3.0 and obtaining alcohol wash with the ethanol concentration up to 17% vol.

Conclusion

It has been found that culturing industrial yeast of the selected race *Saccharomyces cerevisiae* DO-16 at low pH values will provide not only the required sterility of the substrate, but also a high content of yeast cells (250–320 million/cm³). It has been proved that the selected new strain of yeast *Saccharomyces cerevisiae* DO-16 is able to ferment the wort with the dry matter concentration 24–34% with the ethanol content in the fermented wash up to 17% vol., compared with the race *Saccharomyces cerevisiae* DO-11(16.03% vol.).

The use of the new high-productive osmophilic acid-resistant strain of the distiller's yeast *Saccharomyces cerevisiae* DO-16 will allow fermenting wort with high ethanol concentration in the wash, and reducing the consumption of thermal energy needed to release alcohol from the wash and the expenditure of water for cooling. Besides, it will reduce the amount of post-alcohol stillage on condition of using it to prepare the mash. The rational parameters of fermentating high-concentration wort (24–32%) have been developed for application in industrial bioethanol production.

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ПІДБІР ТЕХНОЛОГІЧНИХ ПАРАМЕТРІВ ЗБРОДЖУВАННЯ СУСЛА ВИСОКИХ КОНЦЕНТРАЦІЙ ОСМОФІЛЬНИМИ РАСАМИ ДРІЖДЖІВ ДЛЯ ОТРИМАННЯ БІОЕТАНОЛУ

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Анотація. Для задоволення зростаючого попиту на енергетичні ресурси і забезпечення стійкої економіки, ключовим питанням є виробництво біоетанолу. Перспективним напрямом для одержання біоетанолу є застосування технології зброджування сусла із зернової сировини з високим вмістом сухих речовин. Мета представленої роботи – дослідження процесу зброджування сусла високих концентрацій осмофільними расами спиртових дріжджів при підвищеній кислотності середовища та підбір технологічних параметрів для отримання біоетанолу. Шляхом селекції нового штаму дріжджів *Saccharomyces cerevisiae* ДО–16 одержано продуценти спирту етилового, які спроможні зброджувати зернове сусло з концентрацією сухих речовин 24–34% при рН 3,0–6,0 з накопиченням спирту в зрілій бражці до 17% об. Проведено дослідження щодо впливу рН сусла на динаміку накопичення дріжджових клітин *Saccharomyces cerevisiae* ДО–11 та *Saccharomyces cerevisiae* ДО–16. Встановлено, що при значеннях рН 2,5; 3,0; 3,5 та 4,0 у раси *Saccharomyces cerevisiae* ДО–16 концентрація дріжджових клітин була вищою в 2,6; 1,7; 1,5 та 1,4 рази відповідно порівнюючи з *Saccharomyces cerevisiae* ДО–11. Встановлено, що культивування виробничих дріжджів досліджуваних рас при низьких значеннях рН забезпечить не тільки необхідну стерильність субстрату, але й високий вміст дріжджових клітин – 250–320 млн/см³. Досліджено хіміко-технологічні показники зрілої бражки при застосування дріжджів раси *Saccharomyces cerevisiae* ДО-11 та ДО-16 при концентрації сусла від 20 до 34% сухих речовин. Встановлено, що за всіх умов досліджень, дріжджі раси *Saccharomyces cerevisiae* ДО-16 синтезували більше етанолу порівняно із расою *Saccharomyces cerevisiae* ДО-11. Застосування нового високопродуктивного штаму *Saccharomyces cerevisiae* ДО–16 дозволить зброджувати сусло з підвищеною концентрацією спирту етилового в бражці, а також знизити витрати теплової енергії на виділення спирту із бражки і води на охолодження, та зменшення кількості післяспиртової барди за умов її використання на приготування замісу.

Ключові слова: *Saccharomyces cerevisiae*, сусло високих концентрацій, біоетанол, зброджування, осмофільні спиртові дріжджі.

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