

UDC: 54-44:57.03:663:664

THE INFLUENCE OF NANOPARTICLES OF BIOGENIC METALS ON CULTURING THE YEAST *SACCHAROMYCES CEREVISIAE*

DOI: <https://doi.org/10.15673/fst.v14i3.1610>

Article history

Received 21.02.2020
 Reviewed 12.04.2020
 Revised 05.08.2020
 Approved 17.09.2020

Correspondence:

K. Danilova
 E-mail: dankoe77@gmail.com

Cite as Vancouver style citation

Sychevsky M., Danilova K., Oliynichuk S. The influence of nanoparticles of biogenic metals on culturing the yeast *Saccharomyces Cerevisiae*. Food science and technology. 2020;14(3):37-43. DOI: <https://doi.org/10.15673/fst.v14i3.1610>

Цитування згідно ДСТУ 8302:2015

Sychevsky M., Danilova K., Oliynichuk S. The influence of nanoparticles of biogenic metals on culturing the yeast *Saccharomyces Cerevisiae* // Food science and technology. 2020. Vol. 14, Issue 3. P. 37-43 DOI: <https://doi.org/10.15673/fst.v14i3.1610>

Copyright © 2015 by author and the journal "Food Science and Technology".

This work is licensed under the Creative Commons Attribution International License (CC BY).
<http://creativecommons.org/licenses/by/4.0>



Introduction. Formulation of the problem

The physicochemical, biological, biochemical, pharmacological, and toxicological mechanisms of action of nanoscale materials are now intensively studied throughout the world.

By convention, nanomaterials is the term for disperse and bulk materials containing structural elements with at least one geometrical dimension not exceeding 100nm, and with entirely new properties, functional and operational characteristics [1]. By their size, nanomaterials are in between individual atoms/molecules and microorganisms. Transition from macro- and microsized to those in the range 1–10 nm leads to qualitative changes in the physical and chemical properties of these materials (heat resistance, strength, electrical conductivity) and to an increase in the reactivity and catalytic properties never

M. Sychevsky¹, Doctor of Science, Professor, Academician of the NAAS, Director

K. Danilova², Ph.D., Senior Researcher

S.Oliynichuk², Doctor of Science, Senior Researcher

¹Institute of Food Resources of NAAS

²Department of fermentation technology

Institute of Food Resources of NAAS Ukraine

4A, Ye. Sverstiyka Str., Kyiv, Ukraine, 02002

Abstract. The effect of nanoparticles of biogenic metals (Fe, Mg, Zn, Mn) and their combinations on culturing the yeast *Saccharomyces cerevisiae* and on fermentation of sugar-containing raw materials into ethanol have been investigated. The research involved the use of nanometal preparations obtained by volumetric electric-spark dispersion. It has proved effective to add nanozinc and a preparation containing nanomanganese and nanomagnesium, prior to yeast cultivation, to the growth media agar and wort agar in the concentrations 0.5µg/cm³ and 11µg/cm³ respectively. The experimental yeast grown on the medium containing these preparations increased the alcohol concentration in the distiller's wort by 0.2%, whereas the content of unfermented carbohydrates remained within the prescribed limits 0.32–0.39g/100cm³. The yeast biomass increased by 1.2–1.4 times. Zinc and manganese/magnesium nanopreparations increased the maltase and invertase activities of the yeast under study by 40–25%. The nanoiron preparation contributed to inhibiting the fermentation activity of the yeast biomass. Biogenic metal nanocomplexes are used by yeast as an additional nutrient source. They form organometallic and intracomplex active compounds with yeast cell enzymes, primarily with hexokinase, aldolase, enolase. This intensifies synthesis of enzymes and increases their catalytic effect. The results obtained prove the effectiveness of biogenic metal nanopreparations as catalysts for biochemical transformations in a yeast cell. Using nanometals increases the productivity of bakery yeast, improves the technological process of alcoholic fermentation, and offers ample opportunity to increase the activity of enzyme preparations in the course of their production.

Keywords: nanometal preparations, yeast, growth medium, molasses wort, fermentation, alcohol.

observed in macro- and microscopic materials of the same chemical nature. The specific properties of nanomaterials are due to the well-developed surface of the particles that form them and to the electronic and quantum effects characteristic of these nanoparticles [2]. That makes it likelier that various processes will develop inside certain cellular structures: organelles, biological membranes. This means that the contact of the nanomaterial with the cell nucleus and DNA becomes stronger [3,4]. A big variety of nanomaterials, their specific functional properties that can enhance or inhibit biotechnological processes allow controlling the activity of microbial biocatalysts in food technologies.

So, the presence of nanoparticles in industrial raw materials can effect on the course of technological processes based on using biological objects [5]. This

has been confirmed by studies in this area recently conducted in many countries.

Analysis of recent research and publications

It is known that different concentrations of biogenic metal nanopreparations can have a positive as well as a negative effect on biotechnological processes.

In some publications [6,7], there are data on how various nanopreparations (metal nanoparticles, nanofibres, nanotubes) are used to increase the productivity of biotechnological processes of producing biofuels. It is noted that immobilised nanoparticles have a positive effect on bioethanol production, in particular, that nanopreparations can effectively inhibit formation of inhibitory compounds under certain conditions, depending on their concentration and the methods they are obtained by. It is known that salts of biogenic metals acting upon yeast increase its productivity and stability [8]. Accordingly, the use of nanopreparations of these metals will also have an effect on the activity of microbial biocatalysts, in particular, enzymes and yeast.

Enzymes immobilised by nanopreparations are characterised by higher enzymatic activity, thermal stability, and wider ranges of effective pH. All this reduced the cost of the biotechnological process. Thus, the properties of cellulase immobilised on manganese oxide nanoparticles were higher than those for the free enzyme, and bioethanol could be produced from agricultural waste in a wider range of temperatures and pH values [9].

Nanopreparations NiO and Fe₃O₄ added in the mass concentration 0.01% increase by 1.6 and 1.13 times, respectively, the production of bioethanol by the strain BY4743 of the yeast *Saccharomyces cerevisiae*. When the concentration is more than 0.02% by weight, the yield of bioethanol decreases in all the nanopreparations under study [7].

In [10], it was studied how nanoparticles of silver, copper, zinc, and titanium oxide effected on the development of top and bottom-fermented brewer's yeast population, and on how the yeast accumulated ethanol during beer wort fermentation. It was noted that silver nanopreparations inhibited yeast cultivation, while zinc and zinc oxide nanoparticles enhanced the brewer's yeast biomass and increased the ethanol content in the "immature" beer, depending on their concentration. Low concentrations of nanozinc activated the development of brewer's yeast, but their increase slightly inhibited the yeast population [11,12]. Other indications of an increase in the fermentation energy of yeast are a 31.5% increase in the yield of carbon dioxide and their higher maltase and zymase activity [13].

Yeast is known to contain no fewer than 50 enzymes, and their synthesis increases by several times depending on the substrate added to the growth medium [14]. Magnesium ions catalyse the action of

hexokinase that makes glucose transform into glucopyranose-6-phosphate. Aldolase is activated by zinc, cobalt and calcium ions, and acts upon fructose-1,6-diphosphate. As a result, the latter breaks down into two phosphotrioses: 3-phosphoglycerol aldehyde and phosphodioxyacetone. Conversion of 2-phosphoglyceric acid into phosphoenolpyruvic acid is catalysed by enolase activated by the ions Mg²⁺, Mn²⁺, Zn²⁺ [15].

The research [16] investigated the effect of a zinc oxide nanopreparation on the formation of β-glucosidase (BGL) in the yeast *Saccharomyces cerevisiae* under various conditions. The findings showed that the yeast culture, after the addition of 5 mmol/mol of the ZnO nanopreparation, increased the intracellular activity of β-glucosidase up to 28%, which was accompanied by an increase in the number of yeast cells. However, higher doses of the ZnO nanopreparation (10 and 15mmol/mol) caused a decrease in the activity of the enzyme and in the yeast growth. Adding ethyl alcohol to the growth medium till its content was 2, 5, and 10% inhibited the yeast biomass growth. However, in the presence of the zinc oxide nanopreparation, the biological activity of the enzyme in the cells increased regardless of the ethanol concentration, as evidenced by a higher yield of β-glucosidase [16].

So far, the effect of nanopreparations on various biotechnological processes has not been fully determined. However, many publications point out that nanoparticles of biogenic metals can be used to stimulate the growth, development, and synthesis of microorganisms [7,10,12,16]. Such studies, though, are still in their infancy, so it is very important to continue them. They are necessary both in theoretical and in practical aspects, since enzymes of microorganisms are widely used in biotechnological processes to develop the practical areas of agriculture, the food, chemical, and pharmaceutical industries, etc.

The purpose of the research was to study how biogenic metal nanoparticles (Fe, Mg, Zn, Mn) could be used to culture the yeast *Saccharomyces cerevisiae* during fermentation of sugar-containing raw materials into ethanol.

The objectives:

1. To determine the effect of nanometals (Fe, Mg, Zn, Mn) on molasses wort fermentation.
2. To study cultivation of the yeast *Saccharomyces cerevisiae* in the presence of nanometal preparations of zinc, magnesium, and manganese.
3. To determine the enzymatic activity of yeast grown on a medium with nanometals added.

Research materials and methods

For the research, we used preparations of nanometals (PNM) obtained by volumetric electric-spark dispersion [17]. The distribution of particle sizes and of the zeta potential was determined with an analyser Zetasizer Nano ZS (Malvern Instruments Ltd,

United Kingdom). We used PNM with particle sizes from 119 to 229 nm, containing deionised water as the basis of their composition. The detailed characteristics of the preparations studied are given in Table 1.

Table 1 – Characteristics of the preparations of metal nanoparticles under study

No.	Name	Metal concentration, mg/dm ³	Zeta potential, mV	Size, nm
1	PNM Zn	300	-8.46	229
2	PNM Fe	100	-14	180
3	PNM Mn/Mg	5450	-11.5	119

In the first series of laboratory experiments, beet molasses fermentation was reproduced. For this, into the flask with 12% molasses wort, we added 5 N hydrochloric acid solution (to achieve the pH value 4.85) and 1.7g of dry yeast *Saccharomyces cerevisiae* of the race K-7, with the DM concentration 94%. The source of nutrition was urea (2.8cm³, 5:100) and orthophosphoric acid (2.1cm³, 5:100), and besides, 1cm³ of the antiseptic *Bactrilon* was added. The molasses was fermented in a number of ways: with no additives (control), with addition of a mixture of the nanopreparations PNM Zn (3.5µg/cm³), PNM Mn/Mg (0.08g/cm³), and PNM Fe (5.6µg/cm³), and using the cellulolytic-action enzyme preparation *Tegazym RT 75 L* (0.3cm³).

At the second stage, it was studied how the nanometallic preparations directly effected on the yeast cell *Saccharomices cerevisiae*. The solution of nanometals was added directly into the malted wort at the yeast culturing stage, prior to using the yeast for fermentation. The concentration of nanometals was 5 times lower than in the first series of experiments. To check the fermentation activity of this yeast, the prepared molasses wort was fermented in the same way as in the previous experiment. The control was the yeast grown in pure malt wort, with no preparations added.

At the third stage of the study, the nanopreparations were introduced directly into the growth medium on which a pure yeast culture was grown. In Variant 1, the agar and wort agar medium was prepared with addition of 0.08cm³ of nanozinc in the concentration 3.5µg/cm³. In Variant 2, 0.12cm³ of the mixture of nanomanganese and nanomagnesium (in the concentration 0.08 mg/cm³) was added to the medium. In Variant 3, the growth media was prepared using the same amounts of nanozinc, nanomanganese, and nanomagnesium as in the previous variants. The yeast was generated on the pure malt wort, and fermentation was performed on the molasses wort with nutrients added (2.8cm³ of urea (5:100), 2.1cm³ of orthophosphoric acid (5:100), 1cm³ of the antiseptic *Bactrilon*). The physicochemical parameters of wort fermentation were studied by the standard and commonly used methods [18]: the alcohol concentration in the distiller's wort was determined by the standard

dichromate-iodometric method, the mass concentration of unfermented carbohydrates was determined by the photoelectric colourimetric anthrone method. The amounts of carbon dioxide released and of the yeast biomass were determined by gravimetric analysis [18]. The fermentation rate was determined by the amount of carbon dioxide released during the fermentation process. The fermentation process was considered complete when the amount of carbon dioxide released in the last 2 hours did not exceed 0.1g.

The maltase activity of yeast was determined polarimetrically [19]. The maltase activity of the yeast (Ma) was calculated by the formula 1:

$$M_a = \frac{0.0438 \cdot \Delta P \cdot 1000000}{60 \cdot a \cdot 342} = \frac{20.14 \cdot \Delta P}{a} \quad (1)$$

where 0.0438 is the amount of glucose formed as a result of maltose hydrolysis in 1cm³ of the incubation mixture, which corresponds to a change in polarisation of 1, g;

ΔP is the change in the angle of the polarisation plane equal to the difference of the polarisation of the control solution and that of the experimental one, °;

60 is the time of the enzyme's action, min;

342 is the molecular mass of maltose, g;

a is the amount of yeast in the incubation mixture, g.

The invertase activity of the yeast was determined by colourimetry [19].

The experimental data were processed by the methods of mathematical statistics using the Microsoft Excel 2010 and STATISTICA 10 software. The analysis of variance consisted in determining the total and factorial variances using the coefficient of determination R² when dealing with the null hypothesis. To determine the validity of the results, the p-level was used characterising the probability of error. At p<0.05, error is not more than 5%, and the result is reliable.

Results of the research and their discussion

The main parameters characterising the effectiveness of wort fermentation process is the content of alcohol, unfermented sugars, the amount of yeast biomass, and the content of metabolic products, which are determined in distiller's wort. The fermentation rate is characterised by the amount of carbon dioxide released during fermentation.

To solve the problem of intensifying alcohol production, the biochemical activity and physiological state of yeast are very important. They directly depend on the composition of the substrate and the conditions of yeast generation. The previous studies established that enriching the growth medium with metal ions at the cultivation stage increased the biomass concentration and the fermentation activity of yeast [10,12].

It is known that particles of zinc, manganese and magnesium activate the enzymes of yeast cells so

we have studied the influence of nanoparticles of these metals on the processes of yeast cultivation. To determine the effect of biogenic metal nanoparticles on the fermentation process, beet molasses fermentation was reproduced in the laboratory. The nanometal preparations of zinc, manganese and magnesium mixture, iron, and the cellulolytic-action enzymatic preparation were added to the prepared wort (Table 2).

In the experiments on fermentation of the molasses wort, adding a mixture of biogenic metal nanopreparations of zinc, manganese, and magnesium intensified alcoholic fermentation. This is evidenced by an increase in the released CO₂ compared to the control (by 0.5g), and a decrease in the total content of unfermented sugars (by 0.2g/100cm³). At the same time, the decrease in the yeast biomass amount and in the volume fraction of alcohol in the distiller's wort can indicate an inhibition of the development of the yeast population, probably due to the high concentration of nanopreparations. Adding nanoiron and a cellulolytic enzyme preparation did not have a visible effect: the parameters of the distiller's wort were the same as in the control, and in the experiment with nanoiron, inhibition of yeast biomass development was observed.

The productivity of biological processes directly depends on the type of the nanopreparation used and its concentration [20]. Thus, zinc nanooxide, in a concentration of up to 5mmol/mol, promotes an increase in saccharomycetes and in their intracellular enzymatic activity. Besides, it contributes to ethanol accumulation during wort fermentation [16]. Based on this, we made the assumption that biogenic metal nanopreparations can be consumed by yeast as an additional source of nutrition. To confirm or refute this conclusion, a research was made on the fermentation process in the molasses wort. The latter was fermented with yeast that had been previously grown on malt wort, with addition of the zinc nanopreparations and

the mixture of manganese and magnesium, as these metals increased yeast fermentation energy (table 3).

The data obtained indicate that addition of nanometallic preparations led to an increase in the yeast biomass by almost 2.5 times, in comparison with the control. However, other parameters of the distiller's wort were within the experimental error. Thus, nanometallic preparations are only effective if added at the yeast cultivation stage.

The yeast was reproduced from a pure culture, and the preparations of nanometals were added to the growth media: to the Petri dish with wort agar and to the test tube with malt wort. In the control experiment, a pure yeast culture was grown in a medium with no nanopreparations added.

The fermentation activity of yeast cells was evaluated by the amount of carbon dioxide produced during fermentation (Fig. 1).

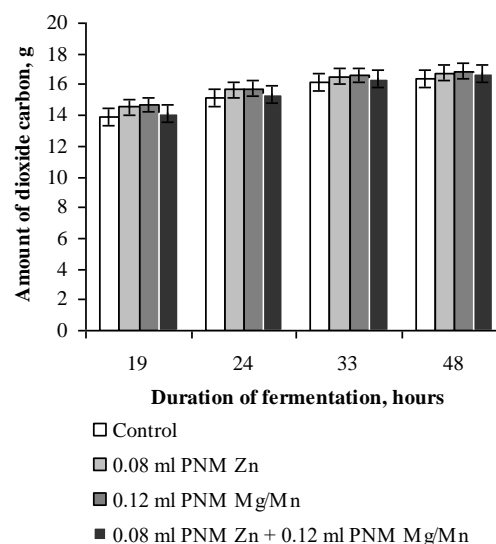


Fig. 1. Carbon dioxide production depending on the duration of molasses wort fermentation

Table 2 – Characteristics of beet molasses wort fermentation

Parameter	Control	Variant 1 (PNM Zn, Mn/Mg)	Variant 2 (PNM Fe)	Variant 3 (EP RT 75 L)
Amount of CO ₂ released, g/50 g	19.49±0.03	19.95±0.03	19.37±0.03	19.49±0.03
Alcohol content, % (by volume)	9.45±0.03	9.4±0.03	9.4±0.03	9.45±0.03
Amount of yeast biomass, g/dm ³	9.6±0.05	7.8±0.05	6.5±0.05	8.6±0.02
Total content of unfermented carbohydrates, g/100cm ³	1.16±0.02	0.91±0.02	1.18±0.02	1.14±0.02
True DM concentration, %	13.0±0.05	13.0±0.05	13.0±0.05	13.0±0.05

Table 3 – Parameters of the distiller's wort obtained by fermentation with yeast grown on wort with nanopreparations added

Parameter	Control	Variant 1 0.7µg/cm ³ of PNM Zn	Variant 2 16µg/cm ³ of PNM Mn/Mg	Variant 3 0.5µg/cm ³ of PNM Zn+16 µg/cm ³ of PNM Mn/Mg
Amount of CO ₂ released, g/50 g	20.18±0.03	20.17±0.03	20.25±0.03	20.22±0.03
Alcohol content, % (by volume)	9.95±0.03	9.90±0.03	10.00±0.03	10.00±0.03
Amount of yeast biomass, g/dm ³	6.9±0.05	10.20±0.05	15.1±0.05	18.2±0.05
Total content of unfermented carbohydrates, g/100cm ³	0.38±0.02	0.34±0.02	0.32±0.02	0.31±0.02

As can be seen from Fig. 1, on the first day of fermentation, in all variants, the yeast with the nanopreparations added had higher fermentation activity compared to the control. When the mixture of nanopreparations was used, there was by 0.23 g more CO₂ released than in the control, but less carbon dioxide was produced than in the variants when only one nanopreparation was used. By the end of fermentation, the amount of carbon dioxide released in the experiments exceeded the control sample by 0.3–0.4 g, which confirmed the assumption that the fermentation activity of yeast cells would increase with addition of the nanometal preparations of Zn, Mn, Mg [10,12].

The parameters of the distiller's wort fermented by this yeast are shown in Table 4. As can be seen from the data in Table 4, addition of nanometal preparations to the growth medium at the yeast cultivation stage intensified the fermentation of the molasses wort. This is evident from the 0.2% increase in the proportion of alcohol by volume, from the 0.3–0.4 g increase in CO₂ produced, and from the reduced total content of unfermented sugars.

It is known that when sugar-containing raw materials are fermented at the first stage, under the action of invertase, sucrose hydrolyses to form fructose and glucose. That is why, at the beginning of fermentation, the activity of maltase (alpha-glucosidase) and invertase (beta-fructafuranosidase) is very important. Metal nanoparticles affect the activity of enzymes produced by *Saccharomyces cerevisiae* cells [6,16]. The catalytic activity level of these enzymes was determined in the saccharomycetes grown with and without the nanometal preparations added. The findings on the maltase and invertase activities in the yeast

Saccharomyces cerevisiae (race K-7) are shown in Table 5.

As can be seen from Table 5, zinc and magnesium/manganese nanopreparations increased the maltase and invertase activity of the yeast under study by 40 and 25%, compared with the control. This is confirmed by other researchers' data [6,9,16].

Thus, nanopreparations of biogenic metals are only effective if used at the yeast cultivation stage, which is confirmed by the data in Tables 3 and 4. The studies [16,20] describe the mechanism of action of nanopreparations on yeast growth and enzyme activity. A study of the growth of yeast cells based on N-acetylcysteine showed that cells died not only because of reactive oxygen species, which indicates another mechanism of action of biogenic metal nanoparticles [21,22]. In our opinion, metal nanoparticles from the growth medium form organometallic and intracomplex compounds with yeast cell enzymes, primarily with hexokinase, aldolase, enolase. This intensifies synthesis of yeast cell enzymes and increases their catalytic effect (Table 5). The results obtained prove the effectiveness of biogenic metal nanopreparations as catalysts for biochemical transformations in a yeast cell. The greatest catalysing effect on yeast enzymes was produced by the magnesium and manganese nanopreparations. Due to addition of nanozinc, the yeast biomass increased by 1.7 times compared with the control, but this increase was smaller than it was after using nanomanganese and nanomagnesium. Using manganese and magnesium nanometals and their combinations with nanozinc at the yeast cultivation stage improves the technological process of alcoholic fermentation and increases the productivity of bakery yeast.

Table 4 – Parameters of the distiller's wort

Parameter	Control	Variant 1 0.7 µg/cm ³ of PNM Zn	Variant 2 16 µg/cm ³ of PNM Mn/Mg	Variant 3 0.5 µg/cm ³ of PNM Zn + 16 µg/cm ³ of PNM Mn/Mg
Amount of CO ₂ released, g/50 g	16.54±0.03	16.84±0.03	16.92±0.03	16.75±0.03
Alcohol content, % (by volume).	7.9±0.03	8.1±0.03	8.0±0.03	8.2±0.03
Amount of yeast biomass, g/dm ³	10.1±0.05	17.0±0.05	18.3±0.05	21.6±0.05
Total content of unfermented carbohydrates, g/100cm ³	0.43±0.02	0.4±0.02	0.39±0.02	0.32±0.02

Table 5 – Enzymatic activity of the yeast

Parameter	Control	Variant 1 0.7 µg/cm ³ of PNM Zn	Variant 2 16 µg/cm ³ of PNM Mn/Mg	Variant 3 0.5 µg/cm ³ of PNM Zn + 16 µg/cm ³ of PNM Mn/Mg
Maltase activity, un./g of ADM of yeast	37.4	61.3	65.8	58.2
Invertase activity, un./g of ADM of yeast	310.2	412.5	415.8	407.6

Conclusion

Biogenic metal nanocomplexes are an effective factor that allows regulating alcoholic fermentation. They increase the catalysis of biochemical transformations. In the experiments on molasses wort fermentation, adding a mixture of biogenic metal nanopreparations of zinc, manganese, and magnesium led to an increase in the yeast biomass by almost 2.5 times, in comparison with the control. However, other parameters of the distiller's wort were within the experimental error. In the experiment with nanoiron, inhibition of yeast biomass development was observed.

The nanometal preparations of manganese, magnesium, zinc added to the medium at the yeast growth stage made the yeast biomass increase by 1.2–1.4 times. The experimental yeast obtained in this way increased the alcohol concentration in the distiller's wort by 0.2%, while the content of unfermented carbohydrates remained within the prescribed limits 0.32–0.39g/100cm³ (the maximum loss and yield value permitted by the standard being 0.45g/100cm³).

The catalytic activity has been determined for the enzymes alpha-glucosidase and beta-fructafuranosidase of the yeast *Saccharomyces cerevisiae* (race K-7) cultivated on a medium with biogenic nanometal preparations. This determination has shown that their maltase and invertase activity was higher by, respectively, 1.7 and 1.3 times, compared with the control. This confirms the assumption that biogenic metal preparations are used by yeast as an additional nutrition source and form active intracomplex compounds with yeast cell enzymes.

Thus, when fermenting sugar-containing raw materials, it is recommended to use nanometal preparations of Zn and Mn/Mg at the yeast cultivation stage. The use of nanometals of metals opens up wide opportunities for increasing the activity of enzyme preparations in the process of their production, and promotes increasing overall productivity. Using biogenic nanometals in biotechnological processes offers ample opportunity to increase the activity of enzyme preparations in the course of their production and the overall productivity.

References:

- Baloian BM, Kolmakov AG, Alymov MI, Krotov AM. Nanomaterialy. Klassifikatsiia, osobennosti svoistv, primeneniie i tehnologii polucheniia. Mezhdunarodnyj universitet prirody, obschestva i cheloveka "Dubna": Filial "Ugresha"; 2007.
- Shtykov SN, Rusanova TU. Nanomaterialy i nanotehnologii v himicheskikh i biohimicheskikh sensorah: vozmozhnosti i oblasti primeneniia. Rossiiskii himicheskii zhurnal. 2008;LII(2):92-100.
- Maltsev PP. Nanotehnologii. Nanomaterialy. Nanosistemaia tehnika: Mirovye dostizheniia. Moskva: Technosfera; 2008.
- Yaroslavtsev AB. Nanomaterialy: svoistva i perspektivnye prilozheniia. Moskva: Nauchnyi mir; 2014.
- Chekman IS, Ulberg ZR, Malanchuk VO. Nanonauka, nanobiologiia, nanopharmatsiia. Kyiv: Poligraph plus; 2012.
- Sekoai PT, Ouma CNM, du Preez SP, Modisha P, Engelbrecht N, et al. Application of nanoparticles in biofuels: An overview. Fuel. 2019;237:380-397. <https://doi.org/10.1016/j.fuel.2018.10.030>.
- Sanusi IA, Faloye FD, Kana EBG. Impact of Various Metallic Oxide Nanoparticles on Ethanol Production by *Saccharomyces cerevisiae* BY4743: Screening, Kinetic Study and Validation on Potato Waste. Catalysis Letters. 2019;149(7):2015-2031. <https://doi.org/10.1007/s10562-019-02796-6>.
- Shyshkov YuI, izobretatel'; Shyshkov YuI, patentoobladatel'. Akvahelat, sposob polucheniia akvahelata, sposob modelirovaniia harakteristiki kultury kletok, kultury tkani, odnokletochnogo organisma ili mnogokletochnogo organisma i transportnaia sistema. Patent RF 2115657. 1998 July 20.
- Cherian E, Dharmendirakumar M, Baskar G. Immobilization of cellulase onto MnO₂ nanoparticles for bioethanol production by enhanced hydrolysis of agricultural waste. Chinese journal of catalysis. 2015;36(8):1223-1229. [https://doi.org/10.1016/S1872-2067\(15\)60906-8](https://doi.org/10.1016/S1872-2067(15)60906-8).
- Karpenko DV, Gernet MV. Effect of nanopreparations on development of the populations of *Saccharomyces brewer's* yeasts. Microbiology (Russian Federation). 2017;86(5):596-601. <https://doi.org/10.1134/S0026261717050113>.
- Wahab R, Mishra A, Yun SI, Kim YS, Shin HS. Antibacterial activity of ZnO nanoparticles prepared via non-hydrolytic solution route. Appl. Microbiol. Biotechnol. 2010;87(5):1917-25. <https://doi.org/10.1007/s00253-010-2692-2>.
- Karpenko DV, Vartanova UV, Borisova NO, Shirokova MA. Vliianie nanozinka na aktivnost proteoliticheskikh fermentov. Pivo i napitki. 2016;5:16-19.
- Shyshkov YuI, Plahov SA. Uvelicheniie fiziologo-biohimicheskoi aktivnosti posevnykh droggei. Pivo i napitki. 2002;3:14-19.
- Konovalov SA. Biohimiia droggei. Moskva: Pishchevaya promyshlennost; 1980.
- Marynchenko VO. Tehnologiiia spyrtu. Vinnytsia: Podillia-2000; 2003.
- Deependra Kumar Ban, Subhankar Paul. Zinc oxide nanoparticles modulates the production of β-glucosidase and protects its functional state under alcoholic condition in *Saccharomyces cerevisiae*. Applied Biochemistry and Biotechnology. 2014;173(1):155-166. <https://doi.org/10.1007/s12010-014-0825-2>.
- Shcherba AA, Zakharchenko SM, Lopatko KG, Aftandyliants EG. Primeneniie ob'emnogo elektroiskrovogo dispergirovaniia dlia polucheniia sedimentatsionno ustoiichivnykh gidrozolei biologicheskii aktivnykh metallov. Praci Instytutu elektrodynamiky NAN Ukrainy. Kyiv: IED NANU. 2009;22:74-79.
- Polygalina GV. Tehnohimicheskij kontrol spirtovogo i likero-vodochnogo proizvodstva. Moskva: Kolos; 1999
- Rukhlyadeva AP, Polygalina GV. Metody opredeleniia aktivnosti gidroliticheskikh fermentov. Moskva: Legkaya i pishchevaya promyshlennost; 1981.
- Stankic S, Suman S, Haque F, Vidic J. Pure and multi metal oxide nanoparticles: synthesis, antibacterial and cytotoxic properties. J. Nanobiotechnol. 2016;14:73-78. <https://doi.org/10.1186/s12951-016-0225-6>.
- Eriksen DT, Hsieh PC, Lynn P, Zhao H. Directed evolution of a cellobiose utilization pathway in *Saccharomyces cerevisiae* by simultaneously engineering multiple proteins. Microbial Cell Factories. 2013;12:1-11. <https://doi.org/10.1186/1475-2859-12-61>.
- Chakraborti S, Joshi P, Chakravarty D, Shanker V, Ansari ZA, et al. Interaction of polyethyleneimine-functionalized ZnO nanoparticles with bovine serum albumin. Langmuir. 2012;28(30):11142-11152. <https://doi.org/10.1021/la3007603>.

ВПЛИВ НАНОЧАСТИНОК БІОГЕННИХ МЕТАЛІВ НА ПРОЦЕС КУЛЬТИВУВАННЯ ДРІЖДЖІВ *SACCHAROMYCES CEREVISIAE*

М.П.Сичевський, д.е.н., проф., академік НААН, директор, E-mail: ipr_2018@ukr.net

К.О. Данілова, к.т.н., с.н.с., E-mail: dankoek77@gmail.com

С.Т. Олійнічук, д.т.н., с.н.с., E-mail: oliynichukst@ukr.net

Відділ технології продуктів бродіння

Інститут продовольчих ресурсів НААН України, вул. С.Сверстюка, 4А, м. Київ, 03190

Анотація. Досліджено вплив наночастинок біогенних металів (Fe, Mg, Zn, Mn) та їхніх комбінацій на процеси культивування дріжджів *Saccharomyces cerevisiae* та зброджування цукровмісної сировини на етиловий спирт. Для досліджень використовували препарати нанометалів, отримані способом об'ємного електроіскрового диспергування. Внесення нанозинку та препарату, що містить наномарганець і наномагній, в поживне середовище агар і сусло-агар до початку культивування дріжджів в концентрації 0,5мкг/см³ та 11 мкг/см³ відповідно, показало свою ефективність. Експериментальні дріжджі, вирощені на поживному середовищі з цими препаратами, забезпечували підвищення концентрації спирту в зрілій бражці на 0,2%, при цьому вміст незброджених вуглеводів залишався в межах регламентованих значень 0,32–0,39г/100 см³. Біомаса дріжджів збільшилась в 1,2–1,4 рази. Нанопрепарати цинку і магнію/марганцю на 40 і 25% підвищували мальтазну і інвертазну активності дослідних дріжджів. Препарат нанозаліза сприяв інгібуванню бродильної активності дріжджової біомаси. Наноконплеси біогенних металів використовуються дріжджами як додаткове джерело живлення і утворюють з ферментами дріжджової клітини, перш за все гексокіназою, альдолазою, ендолазою, металоорганічні і внутрішньоконплесні активні сполуки, що сприяє інтенсивному синтезу ферментів та підвищенню їх каталітичної дії. Одержані результати свідчать про ефективність використання нанопрепаратів біогенних металів як каталізатора біохімічних перетворень, що відбуваються в дріжджовій клітині. Застосування нанометалів сприяє підвищенню продуктивності хлібопекарських дріжджів, удосконалює технологічний процес спиртового бродіння та відкриває широкі можливості для збільшення активності ферментних препаратів в процесі їх виробництва.

Ключові слова: препарати нанометалів, дріжджі, поживне середовище, мелясне сусло, бродіння, спирт.

Список літератури:

1. Наноматериалы. Классификация особенностей свойств, применение и технологии получения / Балоян Б.М., и др. // Международный ун-т природы, общества и человека «Дубна»: филиал «Угреша», 2007. 145 с.
2. Штыков С.Н., Русанова Т.Ю. Наноматериалы и нанотехнологии в химических и биохимических сенсорах: возможности и области применения // Российский химический журнал. 2008. т. LI, № 2. С. 92-100.
3. Нанотехнологии. Наноматериалы. Наносистемная техника: мировые достижения / под ред. П.П.Мальцева. Москва: Техносфера, 2008. 432 с.
4. Наноматериалы: свойства и перспективные приложения / Ярославцев А.Б. и др. Москва: Научный мир, 2014. 449 с.
5. Чекман І.С., Ульберг З.Р., Маланчук В.О. Нанонаука, нанобіологія, нанофармація: монографія. Київ: Поліграф плюс, 2012. 328с.
6. Application of nanoparticles in biofuels: An overview / Sekoai P.T., et al. // Fuel. 2019. N 237. P. 380-397. <https://doi.org/10.1016/j.fuel.2018.10.030>.
7. Sanusi I.A., Faloye F.D., Kana E.B.G. Impact of Various Metallic Oxide Nanoparticles on Ethanol Production by *Saccharomyces cerevisiae* BY4743: Screening, Kinetic Study and Validation on Potato Waste // Catalysis Letters. 2019. N 149 (7). P. 2015-2031. <https://doi.org/10.1007/s10562-019-02796-6>.
8. Аквахелат, способ получения аквахелата, способ моделирования характеристики культуры клеток, культуры ткани, одноклеточного организма или многоклеточного организма и транспортная система: пат. на изобр. 2115657 РФ: МПК C07 F19/00, C12 N1/20 / Шишков Ю.И. изобретатель; патентособственник Шишков Ю.И. № 96102066А; заявл. 01.02.1996; опубл. 20.07.1998.
9. Cherian E., Dharmendrakumar M., Baskar G. Immobilization of cellulase onto MnO₂ nanoparticles for bioethanol production by enhanced hydrolysis of agricultural waste // Chinese journal of catalysis. 2015. N. 36 (8). P. 1223-1229. [https://doi.org/10.1016/S1872-2067\(15\)60906-8](https://doi.org/10.1016/S1872-2067(15)60906-8).
10. Karpenko D.V., Gernet M.V. Effect of nanopreparations on development of the populations of *Saccharomyces brewer's yeasts* // Microbiology (Russian Federation). 2017. № 86 (5). P. 596-601. <https://doi.org/10.1021/nl301934w>.
11. Antibacterial activity of ZnO nanoparticles prepared via non-hydrolytic solution route / Wahab R., et al // Appl. Microbiol. Biotechnol. 2010. N. 87 (5). P. 1917-25. <https://doi.org/10.1007/s00253-010-2692-2>.
12. Влияние нанозинка на активность протеолитических ферментов / Карпенко Д.В., и др. // Пиво и напитки. 2016. № 5. С.16-19.
13. Шишков Ю.И., Плахов С.А. Увеличение физиолого-биохимической активности посевных дрожжей // Пиво и напитки. 2002. № 3. С.14-19.
14. Коновалов С.А. Биохимия дрожжей. Москва: Пищевая промышленность, 1980. 310 с.
15. Технологія спирту / під ред. Маринченка В.О. Вінниця: Поділля-2000, 2003. 496 с.
16. Deependra K. B., Subhankar P. Zinc oxide nanoparticles modulates the production of β -glucosidase and protects its functional state under alcoholic condition in *Saccharomyces cerevisiae* // Applied Biochemistry and Biotechnology. 2014. № 173(1). P.155-166. <https://doi.org/10.1007/s12010-014-0825-2>.
17. Применение объемного электроискрового диспергирования для получения седиментационно устойчивых гидрозолей биологически активных металлов / Щерба А.А., и др. // Праці Інституту електродинаміки НАН України / ІЕД НАНУ. Київ, 2009. № 22. С.74-79.
18. Полюгалина Г.В. Технохимический контроль спиртового и ликеро-водочного производства. Москва: Колос, 1999. 334 с.
19. Рухлядева А.П., Полюгалина Г.В. Методы определения активности гидролитических ферментов. Москва: Легкая и пищевая промышленность, 1981. 288 с.
20. Pure and multi metal oxide nanoparticles: synthesis, antibacterial and cytotoxic properties / Stankic S., et al // J. Nanobiotechnol. 2016. № 14. P.73-78. <https://doi.org/10.1186/s12951-016-0225-6>.
21. Directed evolution of a cellobiose utilization pathway in *Saccharomyces cerevisiae* by simultaneously engineering multiple proteins / Eriksen D.T., et al // Microbial Cell Factories. 2013. № 12. P.1-11. <https://doi.org/10.1186/1475-2859-12-61>.
22. Interaction of polyethyleneimine-functionalized ZnO nanoparticles with bovine serum albumin / Chakraborti S, et al. // Langmuir. 2012. № 28(30). P.11142-11152. <https://doi.org/10.1021/la3007603>.