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ANTIMICROBIAL ACTIVITY OF CHROMIUM CITRATE AND ITS EFFECT ON THE GROWTH OF *LENTINULA EDODES*

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Abstract. The creation of a dietary food product by growing shiitake mushrooms (*Lentinula edodes*) on a wood substrate enriched with three-chromium is relevant and scientifically substantiated. The antimicrobial activity of chromium citrate is not known from the literature, but it can be assumed that it inhibits the growth of microorganisms and in certain concentrations can be toxic to *Lentinula edodes*. Therefore, the purpose of this work is to determine the antimicrobial activity of chromium citrate solutions of different concentrations (antibacterial and fungicidal) against test cultures and to study the effect of these solutions on the growth of *L. Edodes* fungi. Trivalent chromium citrate solution, shiitake mushroom mycelium (*Lentinulaedodes* 3790, Mycelia, Belgium), museum reference strains of pathogenic staphylococcus (*Staphylococcusaureus* ATCC 25923), Escherichia coli (*Esherichiacoli* ATCC 25922) and mold fungi (*Aspergillusniger* ATCC 16404), nutrient medium were used for research When determining the antimicrobial effect of a solution of chromium citrate at a concentration of 9.0 mg/l of chromium on test cultures of *E. coli*, *S. aureus* and *A. niger*, a bactericidal effect was established (100% for all tested strains). The bacteriostatic effect of the tested solutions of chromium citrate at a concentration of 6.0 mg/l was established for *E. coli* (72%), *S.aureus* (68%) and *A. niger* (62%). When studying the effect of chromium citrate solutions on *E. coli* ATCC 25922 in concentrations of 3.0 and 0.3 mg/l, it was established that the solutions did not exhibit antimicrobial properties, but, on the contrary, enhanced the growth of this culture. Chromium citrate solutions in the same concentrations (3.0 and 0.3 mg/l) did not show antimicrobial properties against the pathogenic strain of staphylococcus *S.aureus* ATCC 25923, and fungicidal activity against mold fungi *A. niger* ATCC 16404.Solutions with a high concentration of chromium (more than 9 mg/l) showed fungicidal activity and prevented the cultivation of shiitake mushrooms. Instead, solutions with a low chromium content (up to 3 mg/l) enhanced the growth of *L. Edodes*, acting as mycopromoters. It can be assumed that chromium in small amounts and in a bioavailable form is necessary for fungi to synthesize biologically active substances that stimulate their development. And chromium citrate in this case acts not as a disinfectant, but as a micro-fertilizer. So, chromium citrate solutions in concentrations up to 3.0 mg/l of chromium can be used in the cultivation of shiitake mushrooms (*L. edodes*) to increase the yield and enrich the mushrooms with chromium.

Keywords: *L. edodes*, cultivation, chromium citrate, antimicrobial effect, fungicidal activity.

Introduction. Formulation of the problem

Mushrooms are a valuable food product, as they can not only be an alternative source of protein, but also contain a large number of trace elements in a bioavailable form [1-4].

It is known that mushrooms absorb and accumulate various chemical elements, but the effectiveness of this process depends on the species of mushroom, the concentration of the element, and the chemical form in which it is bound [5,6]. Enrichment of mushrooms with trace elements necessary for vital activity can significantly increase the nutritional and pharmaceutical value of the final product and expand the scope of its application. Recently, the concept appeared that mushrooms enriched with various trace elements, such as selenium, lithium, zinc, copper, can be used as functional food products [7-9].

It is promising to use the chromium salts for enriching mushroom substrate due to the wide range of its biological value [10], but it is known that this substances can inhibits the growth of microorganisms and in certain concentrations can have an inhibitory effect on the growth of fungi [11].

Therefore, in order to solve this problem, it was decided to conduct research on establishing the antimicrobial activity of chromium citrate solutions of different concentrations in relation to test cultures and to study the effect of these solutions on the growth of *Lentinula edodes* fungi.

Analysis of recent research and publications

Mushroom production in Ukraine is mostly focused on the domestic market, with 90% of the cultivated mushroom market being mushrooms (Agaricus bisporus) [12]. The production of Agaricus bisporus also ranks first in the world market (almost 70%) [13], and the cultivation of common oyster mushroom (Pleurotus ostreatus) and shiitake (Lentinula edodes) is promising. Shiitake is the second largest mushroom in the world's production of mushrooms after mushrooms due to its high nutritional value, excellent taste and a wide range of medicinal properties [14-16]. The growing popularity of shiitake is evidenced by the fact that its global production has increased more than 30 times over the past 40 years [13].

Lentinula edodes is a delicacy mushroom from Southeast Asia that has been used for thousands of years in traditional Oriental medicine to restore the body's defenses. Due to its natural balanced complex of unique biologically active substances, *Lentinus edodes* has a pronounced oncostatic, immunomodulatory, hepatoprotective, and antiviral effect [14-16].

Lentinula edodes is used in the treatment of immunodeficiency states, cancer and cardiovascular diseases: hypertension, coronary heart disease, atherosclerosis of the arteries, in the treatment of the consequences of heart attacks and strokes and other diseases [14-16].

Since the popularity of shiitakes is growing every year, there is an interest in studying the possibility of their enrichment with biologically active substances, in particular bioavailable chromium. It is advisable to enrich the substrate used for growing *Lentinula edodes*.

Recent studies indicate the important role of trivalent chromium in trace element homeostasis of the body [10].

Chromium, being part of the corresponding enzymes, acts as a regulator of the assimilation and transformation of sugars in the body. Chromium enhances the action of insulin, a hormone that is critically important for the metabolism and storage of carbohydrates, fats and proteins in the body. Lack of chromium, or its poor absorption, is one of the reasons for the development of diabetes [10,17].

Along with the biological activity of chromium compounds, their antimicrobial effect is known.

The work [18] described that chromium(III) complexes are bactericidal for *S. aureus* and *E. coli*.

Reddy et al. [19] studied the antibacterial activities of macrocyclic Cr (III) *complexes against gram-positive bacteria (Bacillus subtilis, Staphylococcus aureus),* gram-negative *(Escherichia coli, Klebsiella pneumonia*) and antifungal activity toward the yeast *S. cerevisiae* and molds *A. niger* and *A. fumigatus*.

The antibacterial activity of chromium oxide nanoparticles at different concentrations was investigated against *Escherichia coli* and *Pseudomonas aeruginosa*. The antibacterial investigation of as prepared nanoparticle showed that chromium oxide nanoparticles (Cr_3O_4 NPs) can be used as antibacterial agents depending upon the concentrations used [20].

The antibacterial activities of Cr_2O_3 nanoparticles (NCs) have been studied using Gram-negative *E. coli* and Gram-positive *S. aureus*. The results showed that Cr_2O_3 NCs have excellent antibacterial activities against *E. coli* and *S. aureus* [21].

The Cr (III) complexes (derived from benzimidazole) exhibited potent antibacterial activity against a panel of strains of Gram negative bacterial and Gram positive species [22].

A chromium (III) coordination compound of 2-amino-3-carbomethoxy-4,5,6,7-tetrahydrobenzo[b]thiophene (ACTT) (3) with $Cr(NO_3)_3 \cdot 9H_2O$ were screened for their antimicrobial activities against several strains of bacteria (*Staphylococcus aureus* ATCC25923, *Escherichia coli* S2 (1), *Bacillus subtilis*, *Staphylococcus aureus, Pseudomonas aeruginosa* ATCC27853, *Shigella flexneri* SDINT) and fungi (*Candida albicans* ATCC10231, *Candida tropicalis* PK233, *Cryptococcus neoformans* H99). The chromium complex showed promising antimicrobial activity against the microorganisms with minimum inhibitory values between 4 and 16 μg/mL [23].

The aim of study [11] was to investigate the biocidal activity of new chromium based coordination complexes against Gram-positive and -negative

bacteria, fungi and brine shrimp nauplii. The complexes S1, S3, S4, S6 showed good antibacterial activity at the concentration of 200 μg disc-1 and gave MIC values between 16-64 μg ml-1 against the tested bacteria.

There is no information on the microbiological activity of chromium citrate. It can only be assumed that it will suppress the growth of microorganisms and in certain concentrations will be toxic to L. Edodes fungi. On the other hand, its disinfecting properties can be useful when preparing a sterile substrate for growing shiitake mushrooms.

Therefore, the **purpose** of this work was to study the effect of chromium citrate solutions of different concentrations on the growth of *L. edodes* mushrooms and to compare their bioactivity (antibacterial and fungicidal) in relation to test cultures.

Objectives of the study

1. To trace the antibacterial and fungicidal activity of different concentrations of chromium citrate on reference test cultures.

2. Investigate the possibility of growing *L. edodes* on substrates enriched with chromium (in the form of citrate) in order to further create a functional food products.

3. Set the limit value of the concentration of a solution of chromium citrate that does not interfere with the growth of *L. edodes*.

Research materials and methods

Materials. Trivalent chromium citrate solution, concentration 600 mg/l, shiitake mushroom mycelium (*Lentinulaedodes* 3790, Mycelia, Belgium), museum reference strains of pathogenic staphylococcus (*Staphylococcusaureus* ATCC 25923), Escherichia coli (*Esherichiacoli* ATCC 25922) and mold fungi (*Aspergillusniger* ATCC 16404).

Nutrient medium: Endo medium, meat-peptone broth (MPB), Sabouraud broth, Sabouraud medium, salt egg yolk agar (SEYA), tryptone soya agar (TSA) (Merck Company), potato glucose agar (PGA).

Sterile distilled H_2O.

1% complex neutralizer. The neutralizer contained Tween-80–3.0%, saponin–3.0%, histidine–0.1%, cysteine–0.1% in 100 ml of sterile distilled water.

Preparation of chromium citrate working solutions. Control aqueous solutions were prepared from primary by multiple dilution. Chromium citrate solutions with concentrations of: 0.03, 0.3, 3.0, 6.0, 9.0, 12.0, 15.0, 20.0, 30.0 and 60.0 mg/l chromium were used.

Inspection of cultures for qualitative properties. Before using for research purposes, cultures were checked for quality properties. Cultures of *Escherichia coli* and *Staphylococcus* were incubated on elective media at a temperature of 37+1°C for the purpose of nutrition, recovery and accumulation. MPB and Endo medium were used for *Escherichia coli*, MPB and SEAY were used for *Staphylococcus*. Mold fungi A.niger were incubated at 25+1°C for 3–5 days using Sabouraud broth and Sabouraud agar.

Accumulation of *L. Edodes* culture. Sabouraud broth and Sabouraud agar were used for feeding, recovery and accumulation of the mycelial culture of *L. edodes*. The culture of *L. edodes* was incubated for 5–10 days at a temperature of 25+1°C.

Preparation of workingsuspensions of test cultures. To prepare working suspensions of test cultures, daily cultures of *E. coli* and staphylococcus *S. aureus* on mowed TSA grown at a temperature of 37+1°C were used. To prepare a suspension of mold fungi, *A. niger* culture was used, which was kept at a thermostat for up to 3 days at a temperature of 25+1°C on a slanted Sabouraud agar.

The concentration of test strains of bacteria *S.aureus* and *E. coli* (10^8 μ/ml) and mold fungi *A. niger* (10^7 μ/ml) were used in the work.

To prepare a bacterial suspension, the culture was washed off the agar with sterile water. The resulting suspension of microbes was diluted with sterile water to a concentration of $1x10^9$ μ/ml according to the optical standard of turbidity ("State research institute of standardization and control of medical biologicals named after L.A. Tarasevich") №10, which corresponds to $1x10^9$ microbial bodies per 1 ml of suspension, after which successive 10-fold dilutions were made to the required concentration. To confirm the necessary concentration of experimental cultures, a densitometer "Laboratory densitometer DEN–1" was used.

Study of the activity of working solutions of chromium citrate in relation to test cultures. 1.0 ml of microbial suspension of the required concentration was sterilely introduced into a test tube with 9.0 ml of working solution of chromium citrate. The contents of the test tubes were mixed. Exposure to the interaction of test microorganisms with chromium solutions was 60 minutes. Next, 1.0 ml of the solution (chromium citrate solution with test culture) was added to 9 ml of the neutralizer, and then 1 ml of the neutralizer was added to 9 ml of sterile water.

The research was carried out using a quantitative in-depth method.

1 ml of the experimental suspension of the microorganism with a working solution of chromium citrate was sown in a Petri dish and 15 ml of nutrient medium (PGA), heated and cooled to 42–45 °C, was poured. Bacterial cultures were incubated for up to 48 hours, and mold cultures – up to 5 days.

In parallel, the following were monitored: the concentration of introduced bacteria on the medium without chromium citrate solution, the effectiveness and toxicity of the neutralizer, the sterility of nutrient media. Dilution of both cultures and solutions was carried out in laboratory conditions at an air temperature of +20+2°C. The study was performed in five-fold repetition.

Assessment of antimicrobial effect. The assessment of the antimicrobial effect was carried out

by counting the number of formed colonies of microorganisms in 1 cm³, which grew on nutrient medium in comparison with the control of this culture [10] and expressed in colony-forming units (CFU/cm^3). The effectiveness of disinfection was determined in the presence of reduction of microflora (absence of growth) on dense medium. The results of the experimental data were given in absolute numbers and in % reduction.

Evaluation of the effect of working solutions of chromium citrate on *L. edodes.* When conducting research on the effect of working solutions of chromium citrate on the growth activity of shiitake mushrooms (*L. edodes*) on PGA, solutions of chromium citrate in different concentrations were added to the nutrient medium. The research was conducted using a qualitative surface method. The culture of *L. edodes* at a concentration of 108 μ/ml was sown 0.1 ml on the surface of PGA and rubbed with a spatula. The culture of *L. edodes* was grown for 24–48 hours at a temperature of 25°C, controlling the growth of the culture after 3, 6, 9, 18, 24 and 48 hours. Such a control was not chosen by chance, since solutions with a concentration of 30 and 60 mg/l of chromium change the structure of the dense medium of PGA, which makes it impossible to fix the result.

To determine the true values of experimental values and conduct correlation analysis, mathematical and statistical processing of experimental data was carried out. Optimization was carried out by the mathematical method of least squares. Mathematical data processing of experimental data was carried out using data of statistical processing in the Microsoft Exel.

Results of the research and their discussion

Antimicrobial effect of working solutions of chromium citrate on *E. coli*. It was established the bactericidal (100%) and bacteriostatic (72%) effect of the tested solutions of chromium citrate in concentrations of 9.0 and 6.0 mg/l, respectively, on *E. coli* ATCC 25922 when grown on TSA medium (exposure – 60 min) (Table 1).

As a result of studies of the antimicrobial effect of working solutions of chromium citrate on *E. coli* in concentrations of 3.0 and 0.3 mg/l, it was shown that chromium solutions in such concentrations did not exhibit antimicrobial properties, but, on the contrary, enhanced the growth of *E. coli* ATCC 25922 culture (Table 1).

Antimicrobial effect of working solutions of chromium citrate on *S. Aureus*. Studies of the antimicrobial effect of working solutions of chromium citrate on pathogenic staphylococcus *S. aureus* ATCC 25923 showed both 100% bactericidal effect of solutions at a concentration of 9.0 mg/l, and 68% bacteriostatic effect of a solution with a concentration of 6.0 mg/l (exposure – 60 min) (Table 2).

The use of working solutions of chromium citrate in concentrations of 3.0 and 0.3 mg/l did not show antimicrobial properties against the pathogenic strain of staphylococcus *S. aureus* ATCC 25923 (Table 2).

Fungicidal effect of working solutions of chromium citrate on A. Niger. As a result of studies conducted to study the fungicidal effect of working solutions of chromium citrate against mold fungi *A. niger* ATSS 16404, 100% fungicidal effect of the solution at a concentration of 9.0 mg/l and fungiostatic effect (62%) of a solution at a concentration of 6.0 mg/l was established (exposure – 60 min) (Table 3).

Working solutions of chromium citrate in concentrations of 3.0 and 0.3 mg/l also, as in similar previous studies, did not show a fungicidal effect against mold fungi *A. niger* ATCC 16404 (Table 3).

Table 1 – Study of the antimicrobial effect of working solutions of chromium citrate on *E. Coli*

№	Chromium concentration in the working solution, mg/l	Exposition min	*E. coli*		Culture control	Testing error (number of colonies)
			ABC number	%		
1	9.0	60	0	100	1212	±22
2	6.0	60	872	72		
3	3.0	60	Continuous growth	–		
4	0.3	60	Continuous growth	–		

Table 2 – **Investigation of the antimicrobial effect of working solutions of chromium citrate on *S. Aureus***

№	Chromium concentration in the working solution, mg/l	Exposition, min	*S. aureus*		Culture control	Testing error (number of colonies)
			ABC number	%		
1	9.0	60	0	100	1012	±24
2	6.0	60	688	68		
3	3.0	60	Continuous growth	–		
4	0.3	60	Continuous growth	–		

Study of the effect of working solutions of chromium citrate on *L. edodes*

A study of the growth intensity of *L. edodes* on PGA medium in the presence of chromium citrate working solutions of different concentrations (Table 4) at certain time intervals for 48 hours showed the absolute fungicidal activity of solutions with concentrations of more than 15 mg/l of chromium inclusive.

The growth of visible colonies in the size of 1–2 mm after 9 hours of incubation was observed on PGA medium with the addition of 3.0; 0.3 and 0.03 mg/l solutions of chromium citrate.

Quantitative assessment of the effect of chromium solutions of different concentrations on the growth activity of *L. edodes* for 24 and 48 hours is shown in Table 5.

It was established that the fungicidal effect of chromium citrate solutions is manifested at concentrations of 9.0, 12.0, 15.0, 20.0, 30.0 and 60.0 mg/l of chromium.

The effect of solutions of chromium citrate with a concentration of 30.0 and 60.0 mg/l was determined only after 24 hours, because they change the structure of the medium (dilute the PGA).

When studying the influence of chromium citrate solutions in concentrations of 9.0 and 12.0 mg/l on the growth activity of *L. edodes* on PGA, the lack of growth of this culture was recorded during the entire observation period (24–48 h) (Fig. 1, b, c; Table 5), while the solution at a concentration of 6.0 mg/l when added to the nutrient medium with PGA inhibited the growth activity of *L. edodes* by 12.9% relative to the control (Table 5).

Chromium solutions in concentrations of 0.03, 0.3, and 3.0 mg/l on PGA did not show fungicidal activity, but, on the contrary, enhanced the growth of *L. edodes* culture (Table 5).

So, as a result of the research, it was established that solutions of chromium citrate in concentrations of 9.0, 12.0, 15.0, 20.0, 30.0 and 60.0 mg/l on PGA inhibit the growth of *L. edodes*.

Table 3 – Study of the fungicidal effect of working solutions of chromium citrate on mold fungi *A. Niger*

№	Chromium concentration in the working solution, mg/l	Exposition, min	*A. niger*		Culture control	Testing error (number of colonies)
			ABC number	%		
1	9.0	60	0	100	784	±18
2	6.0	60	486	62		
3	3.0	60	Continuous growth	–		
4	0.3	60	Continuous growth	–		

Table 4 – Study of the intensity of growth of *L. edodes* on PGA medium with the addition of chromium citrate solutions of different concentrations

№	Chromium concentration in the working solution, mg/l	Growth of *L.edodes*, h					
		3	6	9	18	24	48
1	60.0	No growth	No growth	No growth	No growth	The process of rarefaction of the medium	The process of rarefaction of the medium
2	30.0	No growth	No growth	No growth	No growth	No growth	The process of rarefaction of the medium
3	20.0	No growth	No growth	No growth	No growth	No growth	No growth
4	15.0	No growth	No growth	No growth	No growth	No growth	No growth
5	3.0	No growth	No growth	Growth of visible culture colonies	Continuous growth	Continuous growth	Continuous growth
6	0.3	No growth	In the form of plaque	Growth of visible culture colonies	Continuous growth	Continuous growth	Continuous growth
7	0.03	No growth	Growth of visible culture colonies	Growth of visible culture colonies	Continuous growth	Continuous growth	Continuous growth
8	Culture control	No growth	No growth	In the form of plaque	Growth of visible culture colonies	Continuous growth	Continuous growth
9	Control of medium	No growth	No growth	No growth	No growth	No growth	No growth

Table 5 – **Effect of chromium citrate solutions of different concentrations on growth activity of *L. Edodes***

№	Chromium concentration in the working solution, mg/l	Cultivation of *L.edodes*, h	*L.edodes*		Culture control	Testing error
			ABC number	%		
1	60.0	24	0	100	522	±22
2	30.0	24	0	100	522	±22
3	20.0	48	0	100	584	±24
4	15.0	48	0	100	584	±24
5	12.0	48	0	100	558	±19
6	9.0	48	0	100	558	±19
7	6.0	48	122	12.9	558	±19
8	3.0	48	1196	–	584	±24
9	0.3	48	Continuous growth	–	584	±24
10	0.03	48	Continuous growth	–	584	±24

Addition of chromium citrate solution to the medium with PGA at a concentration of 6.0 mg/l also inhibits the growth of *L. edodes* (by 12.9%). Solutions in concentrations of 0.03–3.0 mg/l on PGA do not show fungicidal activity, but, on the contrary, enhance the growth of *L. edodes* culture.

As a result of the conducted studies, the antimicrobial and fungicidal activity of trivalent chromium solutions (in the form of citrates) were established.

Chromium, as a heavy transition metal, exhibits toxic properties to living organisms, its six-charged form Cr^{6+} is particularly dangerous.

Most of the world's research on the influence of chromium on the development of bacteria and fungi is aimed at finding microorganisms resistant to the action of heavy metals (including chromium) for the purpose of biological treatment of wastewater that poisons the environment, water bodies, soils and plants.

In these studies, it was found that the effect of a heavy metal depends on its type, concentration, properties of the environment (temperature, acidity) and the microorganism that is cultivated in this environment. The overwhelming majority of heavy metals inhibit the growth of bacteria. But there are resistant strains among bacteria that have developed mechanisms for the biodegradation of heavy metals. Thus, the authors [24] set limit concentrations for cadmium, chromium, nickel, lead,which inhibited the development of *E. Cloacae* strain MC9. For chromium, this value was 150 mg/l.

The authors [25] studied different strains of bacteria, which were coded as S11, S13, S17, S18, S30, S35 and S48, in order to establish their resistance to different concentrations of chromium (100-1500 mg/l). Two strains of bacteria S35 and S48 showed maximum resistance to chromium (1500 mg/l). They were identified by RNA sequencing and showed 99% identity with *Bacillus parathracis* and *Bacillus paramycoides*.

In work [26], the toxic effect of an excess of copper and zinc on ground bacteria was established.

But the situation with fungi was not so unambiguous. Thus, the same authors [26] observed a 5–7 times acceleration of the growth of soil fungi in a medium with a high content of copper and zinc. The work [27] investigated the resistance of strains of *Aspergillus niger, Aspergillus foetidus* and *Penicillium simplicissimum* to heavy metals (nickel, cobalt, molybdenum, tungsten, manganese, iron, and zinc). Certain adaptation mechanisms of each of the strains to various heavy metals were established, tungsten and vanadium did not affect the development of these fungi at all, while nickel turned out to be the most toxic.

In work [28] strains of Penicillium citrinum, Trichoderma viride, Penicillium sp. showed resistance to Cr^{6+} in concentrations up to 100 mg/l. The same strains showed significant growth at chromium concentrations up to 250 mg/l. At higher concentrations of chromium (1000 mg/l), only T. viride could demonstrate growth, and in the acceleration phase. It was established that the most significant response to an increase in chromium concentration is the synthesis of certain enzymes by the fungus.

When we determined the antimicrobial effect of a solution of chromium citrate at a concentration of 9.0 mg/l of chromium on test cultures of *E. coli, S. aureus,* and *A. niger*, we established a bactericidal effect (100% for all tested strains). The bacteriostatic effect of the tested solutions of chromium citrate at a concentration of 6.0 mg/l was established for *E. coli* (72%), *S.aureus* (68%) and *A. niger* (62%). That is, these concentrations are significantly lower than those given in the above-mentioned works, which is caused by different different habitats and the fact that the microorganisms studied by various authors were purposefully selected for biodecontamination of wastewater.

We also confirmed that solutions of chromium citrate of small concentrations do not show antimicrobial and fungicidal activity, and sometimes can stimulate the growth of cultures.

Thus, when studying the effect of working solutions of chromium citrate on *E. coli* ATCC 25922 in concentrations of 3.0 and 0.3 mg/l, it was established that the solutions did not exhibit antimicrobial properties, but, on the contrary, enhanced the growth of this culture. Working solutions of chromium citrate in the same concentrations (3.0 and 0.3 mg/l) did not show antimicrobial properties against the pathogenic strain of

S.aureus ATCC 25923, and fungicidal activity against mold fungi *A. niger* ATCC 16404.

Works devoted to the study of the elemental composition of edible mushrooms primarily determine the content of metals in the final product and their bioaccumulation.

Thus, the authors [29] studied the accumulation of molybdenum, germanium, and selenium in varieties of oyster mushrooms. In [30], they determined the effect of heavy metals on the growth of oyster mushrooms. Among other things, it was established that an excess of iron will prevent the development of these mushrooms.

A systematic study of the content of metals in wild mushrooms was carried out in [3]. The authors [6] conducted the same study for cultivated mushrooms and, among other things, established that shiitake mushrooms are most prone to bioaccumulation of cadmium.

Most often, shiitake mushrooms are tried to enrich with selenium. Thus, the authors of the work [31] investigated the possibility of enriching shiitake mushrooms with selenium in order to create a food supplement.

They came to the conclusion that the addition of selenium salt Na_2SeO_3 to the substrate proportionally increases its content in mushrooms, does not impair their attractiveness, taste and nutritional value, but high selenium concentrations of 0.96 mmol inhibit the growth of mushrooms. The authors [32], in addition to selenium, investigated the possibility of enriching shiitake mushrooms with lithium. The paper [33] shows the results of the influence of heavy metals on the development of shiitake mushrooms. It is noted that the accumulation of heavy metals occurs in the sequence of cadmium, chromium, manganese, copper, zinc, iron. At the same time, there is an increase in the yield of mushrooms and an improvement in their nutritional quality.

In our studies of the effect of chromium citrate solutions of different concentrations on growth activity on the PGA of higher fungi *L. edodes*, the situation turned out to be similar. Solutions with a high concentration of chromium (more than 9 mg/l) showed fungicidal activity and prevented the cultivation of shiitake mushrooms.

Instead, solutions with a low chromium content (up to 3 mg/l) unexpectedly enhanced the growth of *L. edodes*, acting as mycopromoters.

Since it was not possible to find similar studies in the literature, we can only assume that chromium in small amounts and in a bioavailable form is necessary for mushrooms to synthesize biologically active substances that stimulate their development. And chromium citrate in this case acts not as a disinfectant, but as a micro-fertilizer.

Conclusion

1. The fungicidal activity of chromium citrate solutions in concentrations higher than 6 mg/l of chromium has been established, which makes it possible, after further research, to expand the range of preparations used as disinfectants.

2. The phenomenon of promotion of chromium citrate solutions in small concentrations of *E. coli* and *L. edodes* growth was discovered.

3. Chromium citrate solutions in concentrations up to 3.0 mg/l of chromium can be used in the cultivation of shiitake mushrooms (*L. edodes*) to increase the yield and enrich the mushrooms with chromium, with the aim of creating functional food products in the future.

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АНТИМІКРОБНА АКТИВНІСТЬ ХРОМУ ЦИТРАТУ ТА ЙОГО ВПЛИВ НА РІСТ *LENTINULA EDODES*

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Анотація. Актуальним і науково обґрунтованим є створення продукту дієтичного харчування шляхом вирощування грибів шиітаке (*Lentinula edodes)* на збагаченому тризарядним хромом деревному субстраті. Із літературних джерел не відома антимікробна активність хрому цитрату, але можна припустити, що він пригнічує ріст мікроорганізмів і в певних концентраціях може виявитися токсичним до *Lentinula edodes*. Тому метою даної роботи є встановлення антимікробної активності розчинів хрому цитрату різної концентрації (антибактеріальної та фунгіцидної) по відношенню до тест-культур та дослідження впливу цих розчинів на ріст грибів *L. Edodes*. Для досліджень використовували розчин тривалентного хрому цитрату, міцелій гриба шиітаке (*Lentinulaedodes* 3790, Mycelia, Belgium), музейні еталонні штами патогенного стафілококу (*Staphylococcusaureus* ATCC 25923*),* кишкової палички *(Esherichiacoli*ATCC 25922) та плісеневих грибів (*Aspergillusniger* ATCC 16404), поживні середовища. При визначенні антимікробної дії розчину хрому цитрату в концентрації 9.0 мг/л на тест-культури *E. coli*, *S.aureus* і *A. niger* встановлено бактерицидну дію (100% – для всіх досліджуваних штамів). Бактеріостатичну дію досліджуваних розчинів хрому цитрату в концентрації 6.0 мг/л встановлено для *E. Coli* (72%), *S. aureus* (68%) і *A. Niger* (62%). При дослідженні дії робочих розчинів цитрату хрому на кишкову паличку *E. coli* ATCC 25922 в концентраціях 3.0 і 0.3 мг/л показано, що розчини не проявляли антимікробних властивостей, а, навпаки підсилювали ріст цієї культури. Робочі розчини хрому цитрату в цих же концентраціях (3.0 і 0.3 мг/л) не проявляли антимікробних властивостей до патогенного штаму стафілокока *S.aureus* ATCC 25923, і фунгіцидної активності до плісеневих грибів *A. niger* ATCC 16404. Розчини з високою концентрацією хрому (більше 9 мг/л) проявляли фунгіцидну активність і унеможливлювали культивацію грибів шиітаке. Натомість, розчини з низьким вмістом хрому (до 3 мг/л) підсилювали рост *L. Edodes*, виступаючи мікопромоутерами. Можна припустити, що хром у невеликих кількостях і у біодоступній формі необхідний грибам для синтезу біологічно-активних речовин, що стимулюють їх розвиток. А хрому цитрат в даному випадку виступає не як дизенфіктант, а як мікродобриво. Отже, розчини цитрату хрому у концентраціях до 3.0 мг/л хрому можуть бути використані при вирощуванні грибів шиітаке (*L. edodes*) для збільшення урожаю та збагачення грибів на хром.

Ключові слова: *L. edodes*, культивування, хрому цитрат, антимікробна дія, фунгіцидна дія