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ETHYLTHIOSULFANILATE EFFECT ON *Candida tropicalis*

*L. B. ORIABINSKA[1], S. O. STAROVOITOVA[2], S. V. VASYLYUK[3], V. P. NOVIKOV[3], V. I. LUBENETS[3]**

[1]National Technical University of Ukraine "Igor Sikorsky Kyiv Polytechnic Institute";
[2]National University of Food Technologies, Kyiv, Ukraine;
[3]Lviv Polytechnic National University, Ukraine;
e-mail: vlubenets@gmail.com

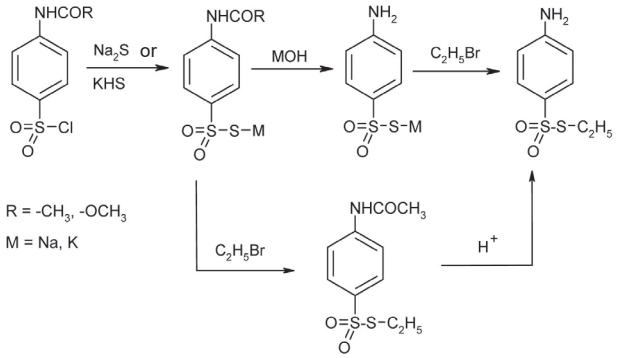
The problem of resistance of microorganisms to antibiotics stimulates the search for new chemotherapy drugs. Ethylthiosulfanilate (ETS, S-ethyl 4-amino-benzenesulfonothioate, S-ethyl ester of 4-aminobenzenethiosulfoacid) as a structural analog of a number of phytoncides was synthesized based on thiosulfoacid salts. ETS effect on opportunistic yeast fungi Candida tropicalis was studied. It was shown that ETS in subfungicidal concentration (125 μg/ml) affected some aspects of a constructive and energy metabolism of fungi: inhibited endogenous respiration and reduced the pool of nucleic acids. Additionaly the significant changes in lipogenesis of C. tropicalis were detected. It was established that ETS in fungistatic and subfungicidal concentrations possessed membranotropic effect and provided a high degree of cooperativity of structural transitions of membranes.

K e y w o r d s: Candida tropicalis, antimycotic action, ethylthiosulfanilate effect.

The antimicrobial resistance to known drugs is one of the challenges to modern medical practice. Thus search for new substances and development on their basis effective antimicrobial and antiviral drugs is an important task of the modern pharmaceutical chemistry. Synthesis of structural analogs of biologically active compounds of natural origin is one of the known approaches that allows creating new efficient agents for various purposes [1, 2]. S-Esters of thiosulfoacids are structural analogs of volatile compounds from garlic (*Allium sativum* L.) [2], onion (*Allium cepa* L.) [3], various types of cabbage, especially cauliflower [1], deep-sea urchin *Echinocardium cordatum* [4], etc. The treatment with these plants has been considered as a promising approach for treatment of atherosclerosis, coronary thrombosis, asthma as well as bacterial infections. It is known that synthetic esters of thiosulfoacids exhibit a wide range of biological effects, and their antimicrobial activity often exceeds the effectiveness of natural analogs. They are offered as drug-candidates, preservatives of fruit and vegetables, effective pesticides, growth regulators, biocide additives, insecticides, radioprotectors [5].

Aryl esters of arylthiosulfoacids (*p*-tolyl-*p*-toluenethiosulfonate and *p*-methoxybenzene-*p*-methoxy-benzenethiosulfonate) possess high insecticidal activity towards larvae *Anagasta kuehniella* [6]. Some thiosulfoacid esters are promising antithrombotic agents [7]. Alkyl esters of some 4-acylaminobenzene- and heterocyclic thiosulfoacids exhibit antifungal effect towards different types of fungi (*Candida albicans*, *Candida tropicalis*, *Candida parapsilosis*, *Candida krusei*, *Candida inconspicua*, *Candida norvegensis*, *Candida kefyr*, *Candida lusitaniae*, *Candida glabrata*, *Cryptococcus neoformans*, *Trichosporon asahii*, *Geotrichum capitatum*, *Blastoschizomyces capitatus*) in low concentrations [8].

Alkylthiosulfanilates disrupt the normal physiological functions of bacteria and fungi, especially in combination with rhamnolipid biosurfactants. Bactericidal and fungicidal effect of thiosulfoesters in combination with rhamnolipids were



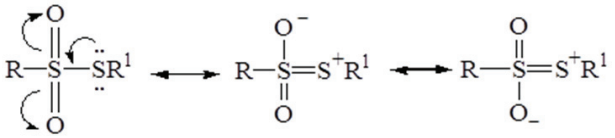
Scheme 1

demonstrated on strains *Pseudomonas aeruginosa*, *Bacillus subtilis*, *Alcaligenes faecalis* and *Rhizopus nigricans* [9].

Several synthesis protocols for ethylthiosulfanilate were developed in Lviv Polytechnic National University (Ukraine) as outlined on *Scheme 1* [9].

ETS stands out for its high fungicidal activity and low toxicity. It is an active substance of antifungal ointment "Esulan"™, which antimicrobial activity against various diseases of the skin, hair and nails was studied by G. N. Pershin and others [10]. Studies of biological action of ETS have shown a wide spectrum of antimicrobial activity [9, 10], it is active not only towards micromycetes, yeast cultures, but also against gram-positive and gram-negative bacteria, including pathogenic, opportunistic and phytopathogenic microorganisms. For instance clinical studies revealed high efficiency of antifungal ointment [10].

Thiosulfoacid esters are highly reactive compounds that interact with nucleophiles, electrophiles, and radicals. Nucleophilic substitution reactions occur with breaking of -S-S-bond due to the redistribution of electron density in thiosulfogroup that determines the direction of nucleophile attack [5].



ETS affects the metabolism of RNA and DNA, which contain amino groups, as well as proteins and aminoacids with disulfide and sulfide fragments, which are components of plant cells and microorganisms. Pretreatment of seeds with ETS at low concentrations improves germination of plants, increases weight of seedlings, reducing the number of diseased plants [11]. The impact of ETS on plants is closely related with its protective action, participation in redox processes, contributing to disruption of respiratory system of phytopathogenic microorganisms. ETS shows a stimulating effect on the growth of field pea and sorghum, and also enhances adaptive capacity of plants to adverse conditions. This proves the prospects of use of ETS as efficient and environmentally safe agent to intensify the process of phytoremediation of contaminated soils [11]. Following the wide range of possible applications and low toxicity of ethylthiosulfanilate an in-depth study of the mechanism of action of ETS on microorganisms, especially on fungi is reasonable.

Thus, the aim of this work was to study effect ethylthiosulfanilate action on the model of yeast *Candida tropicalis.*

Materials and Methods

Methods of obtaining the ethylthiosulfanilate were presented in the previous work [9].

Yeast fungus *C. tropicalis*, obtained from the Museum of Cultures of Department of Industrial Biotechnology, Faculty of Biotechnology and Biotechnics NTUU "KPI" was used as a test culture. To study the effect of ethylthiosulfanilate on the metabolism of *C. tropicalis*, the cells were cultured on nutrient medium (beef-extract broth) with 2% glucose in the presence of subfungicidal concentrations of the test drug for 48 h at 37 °C [12].

Determination of nucleic acids levels in yeast cells under the influence of ethylthiosulfanilate was conducted spectrophotometrically using Spirin's method, separation of DNAs and RNAs was performed by Schmidt's and Tannhauser's method [13].

Yeast lipid composition was analyzed by thin layer chromatography. To determine the total lipids we used the eluent system: benzene : ethylacetate : acetic acid (85 : 15 : 1), and to determine phospholipids – system chloroform : methanol : water (65 : 25 : 4). Solution of sulfuric acid in methanol (10%) was used for the spots detection [14]. Percentage of distribution of total lipid fractions of yeast *C. tropicalis* influenced by subfungicidal concentration of ETS was conducted by densitometration of chromatographic plates (Laser densitometer LKB Ultrascan XL, Sweden).

Determination of endogenous respiration of yeast was performed by using polarographic method (Polarograph LP 60) with open electrode of Clarke type at 37 °C. Respiration rate per unit of time for 1 mg of protein fractions was calculated [15].

The investigation of influence of ethylthiosulfanilate on permeability of the cytoplasmic membranes of yeast was determined photometrically by the release of low-molecular compounds having a maximum absorption at λ-260 nm [16].

All experiments were conducted at least three times using appropriate controls. Statistical analysis was performed using the software package of Microsoft Excel, statistical significance was evaluated by Student's *t*-test. The difference was significant when $P < 0.05$.

Results and Discussion

Previous studies revealed that ETS is a compound with broad spectrum of action and wide intervals of minimal inhibitory concentration (MIC) toward different groups of microorganisms (MIC towards micromycetes was within 3.6-500 μg/ml, gram-negative bacteria – 30-250 μg/ml, gram-positive bacteria – 31.2-125.0 μg/ml, and yeast of genus *Candida* – 30-500 μg/ml [10, 12]).

Yeast-like fungus *C. tropicalis* was used as a culture test for determination of ETS effect. Fungi of the genus *Candida* are one of the prime of fungal diseases of humans and animals. In this work subfungicidal concentration of ETS for *C. tropicalis* – 125 μg/ml was used.

Nowadays a number of antimicrobial agents is known, the mechanism of their action is associated with dysfunction of nucleic acids or enzymes that are involved in their synthesis. In this regard, the study of the impact of ETS on *C. tropicali*s nucleic acids was carried out.

As shown in Table 1, ETS (125 μg/ml) inhibits the synthesis of DNA and RNA. The residual concentration of DNA in the cells was 10.68 ± 0.01 and 49.74 ± 0.05 μg/ml for RNA, that correspond to 27.52 and 39.13% of control, respectively. Namely, concentration of nucleic acids in *C. tropicalis* cells under the influence of ETS decreased 3.6-fold for DNA and 2.56-fold for RNA. Simultaneous inhibition of both types of nucleic acids – RNA and DNA usually occurs when the chemotherapeutics act on the integrity of the cell membranes [16].

A relatively small number of yeast species are characterized by stable respiratory system. In most yeast, growth of yeast under condition of periodic cultivation is accompanied by changes in the structure of the respiratory chain, especially at the initial and terminal parts. This explains more complex functions of respiratory chain and remarkable adaptive properties of yeast organisms [17].

The inhibition of endogenous respiration of yeast under the action of ETS was studied by polarographic method. According to the obtained polarograms the consumption of oxygen in the process of respiration of *C. tropicalis* yeast cells in the presence of ETS was calculated. The calculation results are presented in Table 2.

ETS inhibits respiration of yeast cells by 87%. The obtained data indicates a significant inhibition of endogenous cellular respiration by ETS. More detailed mechanism of ETS action on respiration of cells is possible to establish only as the result of the study of its impact on key catabolic processes and enzymes of the respiratory chain. It may be a subject for further in-depth study.

Table 1. Changing of the nucleic acids concentrations in C. tropicalis cells under the influence of subfungicidal concentration of ETS

Sample	DNA		RNA	
	μg/ml	% from control	μg/ml	% from control
Control	38.81 ± 0.02	100.00	127.11 ± 0.75	100.00
ETS, 125 μg/ml	10.68 ± 0.01	27.52	49.74 ± 0.05	39.13

Table 2. Values of endogenous respiration of yeast C. tropicalis under the influence of ETS subfungistatic concentrations

Sample	Concentration of oxygen nanoatoms, O_2/min·mg^{-1} protein	Respiratory depression, % from control
C. tropicalis cells	264.00 ± 5.84	0
ETS, 125 μg/ml	34.3 ± 7.8	87

Chemotherapy drugs of different nature can cause the delay of ontogenetic development of crops, leading to the increase of the polar fraction of lipids and unsaturated fatty acids in cells [16]. Results of investigations of content of total lipids in *C. tropicalis* cells under the influence of ETS subfungicidal concentration are shown in Table 3.

In the yeast cells that were grown in the presence of ETS the qualitative changes of lipids composition were not detected. In all samples the same lipid components: phospholipids, monoglycerides, diglycerides, triglycerides, sterols, free fatty acids, methyl esters of fatty acids and squalene were found.

But quantitative content of individual fractions varied significantly. A significant accumulation of squalene and methyl esters of fatty acids up to 269.9% of the control was detected under ETS action. Along with the accumulation of squalene accumulation of diglycerides to 242.6%, cholesterol – to 155.4% and slightly increase of triglyceride concentrations – to 116.2% in comparison with control occured. At the same time a decrease in the phospholipids concentration – 47.4%, monoglycerides – 13% and a slight decrease in free fatty acids – 73.4% were observed.

Redistribution of total lipids under the ethylthiosulfanilate action may be of great biological significance. The increase in the concentrations of triglycerides can reflect the growth of the part of lipids, which are the reserve energy substances. It is also know that triglycerides, because of the high exchange rate of membrane lipids, can be used as diacylated intermediates for the biosynthesis of phospholipids [18], the concentration of which decreased under the ETS action.

It is noteworthy that the action mechanism of many antifungal compounds is related to sterols. Ergosterol in fungi cells is required for cell proliferation and its absence could cause the stop in growth of crops. Moreover, ergosterol in cytoplasmic membrane of fungi similar to cholesterol in the membrane of mammalian cells influences the flow and membrane integrity as well as its functions [18]. Thus, considering the adaptive role of membrane lipids it can be assumed that the redistribution of total lipids regulation promotes structural and functional state of membranes, resulting in changing cell sensitivity to chemotherapy drugs.

Phospholipids, which represent a larger part of membrane lipids, play a decisive role in the structure and function of membrane. Specific set of phospholipids induces necessary conformation of enzymes localized in membrane and ensures the activity of their receptor areas. The composition of phospholipids affects structural lability and permeability of cytoplasmic membranes [19].

Considering the importance of phospholipid in the membrane structural organization the study of phospholipid content in *C. tropicalis* cells were carried out.The data (Table 4) revealed that subfungicidal concentration of ETS leads to a decrease in almost all classes of phospholipids relative to control cells: glycophospholipids – to 68.3%, phosphatidylinosite – to 39.9%, phosphatidylethanolamine – to

Table 3. Fractional composition of total lipids of C. tropicalis under the influence of ETS subfungicidal concentration (125 μg/ml)

Class of lipids	Control		Ethylthiosulfanilate		
	μg/g of yeast	% of total lipidpool	μg/g of yeast	% of total lipidpool	% of control
Phospholipids	72.5 ± 8.1	22.00	34.4 ± 10.9	10.97	47.44
Monoglycerides	123.0 ± 6.1	37.32	16.0 ± 5.4	5.10	13.00
Diglycerides	6.1 ± 1.5	1.85	14.8 ± 5.4	4.72	242.62
Sterols	33.2 ± 8.6	10.09	51.6 ± 9.9	16.46	155.42
Triglycerides	14.8 ± 4.1	4.49	17.2 ± 5.8	5.49	116.21
Free fatty acids	18.4 ± 4.9	5.58	13.5 ± 4.8	4.30	73.36
Methyl esters of fatty acids and squalene	61.5 ± 34.4	18.67	166.0 ± 13.8	52.96	269.91
Total	329.5 ± 6.81	100.0	313.5 ± 8.0	100	

Table 4. Fractional composition of phospholipids of C. tropicalis under the influence of ETS subfungicidal concentration (125 μg/ml)

Class of lipids	Control		ETS		
	μg/g of yeast	% of total pool of phospholipids	μg/g of yeast	% of total pool of phospholipids	% of control
Glycophospholipids	51.4 ± 3.0	23.1	35.1 ± 3.0	20.3	68.3
Lysophosphatidylcholine	21.6 ± 3.0	9.7	35.1 ± 3.0	20.3	162.5
Phosphatidylinosite	54.1 ± 3.0	24.3	21.6 ± 0.3	12.5	39.9
Phosphatidylcholine	24.3 ± 3.0	11.0	27.0 ± 3.0	15.7	111.1
Phosphatidylethanolamine	21.6 ± 0.3	9.7	13.5 ± 3.0	7.8	62.5
Neutral lipids	49.5 ± 3.0	22.2	40.5 ± 3.0	23.4	81.8
Total	222.5	100.0	172.8 ± 2.2	100.0	

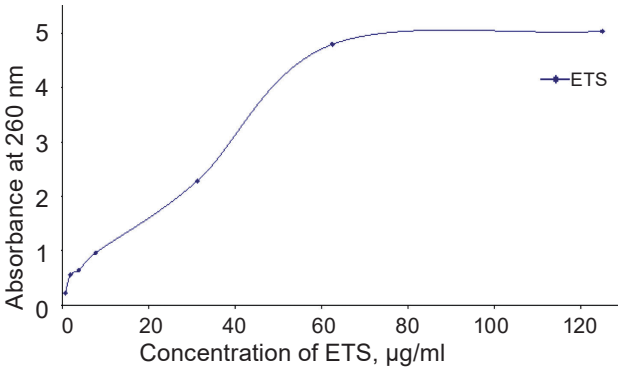
62.5%, neutral lipids – to 81.8%. Also, an increase of lysophosphatidylcholine – to 162.5% and phosphatidylcholine – to 111.1% was observed.

Redistribution of phospholipids under ETS action may occur, probably, due to its effect on the enzymes that catalyzed transformation of one subtypes of phospholipids into another. Changes of membrane lipids can lead to destabilization of cytoplasmic membrane, changing its permeability and as a result – decrease in cell viability.

It is known that most antifungal drugs act on the cytoplasmic membranes level. A characteristic feature of these compounds is the ability to bind with membranes of microorganism cells, leading to leakage of cell vital metabolites, potassium ions, inorganic phosphorus, sugars, amino acids, purine and pyrimidine bases. Based on the data of changes in the chemical composition of membrane lipids, we assumed that interaction with cytoplasmic membrane of yeast may be involved in the ETS mode of action.

Functional state of cell membranes of *C. tropicalis* was evaluated by the level of release of low molecular components of nucleotide nature – pyrimidine and purine bases under the action of different concentrations of ETS. Detection of the substances (λ 260 nm) is a sensitive test that characterizes the state of barrier permeability of membrane and can be used to study the process kinetics.

The measurement of the release of compounds (λ 260 nm) from *C. tropicalis* cells has shown that this process starts immediately after the ETS addition into the incubation medium (*C. tropicalis* cells concentration – 10^6 cells/ml, Fig.). Changing of ETS concentrations from 0 to 62.5 μg/ml led to 4.8-fold increase in the output of purine- and pyrimidine



The permeability of the cell membrane C. tropicalis under the influence of different concentrations of ETS

containing compounds from cells compared with the reference cells. In the concentration range 62.5-125 μg/ml ETS caused almost complete loss of a pool of compounds, that illustrates the plateau in Figure. The obtained results indicate that the ETS has membranotropic effect at subfungicidal and fungistatic concentrations.

There are findings about correlation of permeability barrier with membrane lipids composition [19]. ETS, interacting with surface structures of cells, including cytoplasmic membrane, probably initiates profound structural adjustment, the result of which is the increased permeability and, possibly, inhibition of membranes' physiological functions. Thus, the data suggest that the action mechanism of ETS can be related to disruption of the cytoplasmic membrane, that leads to significant defects in the flow of nutrients into the cells and removing from them vital metabolites.

It was proved that the ETS in subfungicidal concentration (125 mg/ml) significantly influence the metabolism of yeast-like fungi *C. tropicalis*. ETS decreased endogenic respiration by 87% and caused a decrease in nucleic acids pool in pathogenic cells to 27.52% for DNA and to 39.13% for RNA from referent cells level. Significant differences in adipogenesis of *C. tropicalis* under ETS action were shown. Subfungicidal concentration led to a decrease in the content of almost all classes of phospholipids, but provided an increase in the concentration of lysophosphatidylcholine – by 16.25% and phosphatidylcholine – by 11.11%. ETS at fungistatic and subfungicidal concentrations has membranotropic effect and provide a high degree of cooperativity of structural transitions of cell membranes.

ВПЛИВ ЕТИЛТІОСУЛЬФАНІЛАТУ НА *Candida tropicalis*

Л. Б. Орябінська[1], С. О. Старовойтова[2], С. В. Василюк[3], В. П. Новіков[3], В. І. Лубенець[3]

[1]Національний технічний університет України «Київський політехнічний інститут імені Ігоря Сікорського»;
[2]Національний університет харчових технологій, Київ, Україна;
[3]Національний університет «Львівська політехніка», Україна;
e-mail: vlubenets@gmail.com

Проблема резистентності мікроорганізмів до антибіотиків стимулює пошук нових хіміотерапевтичних препаратів. На основі солей тіосульфокислот синтезовано етилтіосульфанілат (ЕТС, S-етил-4-амінобензенсульфонотіоат, S-етиловий естер 4-амінобензентіосульфокислоти) – структурний аналог низки фітонцидів. У роботі вивчали вплив ЕТС на умовно патогенні дріжджоподібні гриби *Candida tropicalis*. Показано, що ЕТС у субфунгіцидній концентрації (125 мкг/мл) впливав на окремі ланки як конструктивного, так і енергетичного метаболізму грибів: пригнічував ендогенне дихання і знижував пул нуклеїнових кислот. На додаток було виявлено значні зміни в ліпогенезі *C. tropicalis*. Встановлено, що ЕТС у фунгістатичних та субфунгіцидних концентраціях виявляв мембранотропний ефект та забезпечував високу ступінь кооперативності структурних переходів мембран досліджуваного штаму.

Ключові слова: *Candida tropicalis*, антигрибкова дія, вплив етилтіосульфанілату.

ВЛИЯНИЕ ЭТИЛТИОСУЛЬФАНИЛАТА НА *Candida tropicalis*

Л. Б. Орябинская[1], С. А. Старовойтова[2], С. В. Васылюк[3], В. П. Новиков[3], В. И. Лубенец[3]

[1]Национальный технический университет Украины «Киевский политехнический институт имени Игоря Сикорского»;
[2]Национальный университет пищевых технологий, Киев, Украина;
[3]Национальный университет «Львовская политехника», Украина;
e-mail: vlubenets@gmail.com

Проблема резистентности микроорганизмов к антибиотикам стимулирует поиск новых химиотерапевтических препаратов. На основе солей тиосульфокислот синтезирован этилтиосульфанилат (ЭТС, S-этил-4-аминобензолсульфонотиоат, S-этиловый эфир 4-аминобензолтиосульфокислоты) – структурный аналог ряда фитонцидов. В работе изучали влияние ЭТС на условно патогенные дрожжеподобные грибы *Candida tropicalis*. Показано, что ЭТС в субфунгицидной концентрации (125 мкг/мл) влиял на отдельные составляющие как конструктивного, так и энергетического метаболизма грибов: подавлял эндогенное дыхание и снижал пул нуклеиновых кислот. Дополнительно были выявлены значительные изменения в липогенезе *C. tropicalis*. Установлено, что ЭТС при фунгистатических и субфунгицидных концентрациях проявлял мембранотропный эффект и обеспечивал высокую степень кооперативности структурных переходов мембран исследуемого штамма.

Ключевые слова: *Candida tropicalis*, противогрибковое действие, влияние этилтиосульфанилата.

References

1. Nakamura Y, Matsuo T, Shimoi K, Nakamura Y, Tomita I. S-methyl methane thiosulfonate, a new antimutagenic compound isolated from *Brassica oleracea* L. var. botrytis. *Biol Pharm Bull.* 1993; 16(2): 207-209.
2. Peinado MJ, Ruiz R, Echávarri A, Rubio LA. Garlic derivative propyl propane thiosulfonate is effective against broiler enteropathogens *in vivo*. *Poult Sci.* 2012; 91(9): 2148-2157.
3. Block E, Thiruvazhi M, Toscano PJ, Bayer T, Grisoni S, Zhao SH. Allium chemistry: structure, synthesis, natural occurrence in onion (*Allium cepa*), and reactions of 2,3-dimethyl-5,6-dithiabicyclo[2.1.1]hexane S-oxidens. *J Am Chem Soc.* 1996; 118(12): 2790-2798.
4. Takada N, Watanabe M, Suenaga K, Yamada K, Kita M, Uemura D. Isolation and structures of hedathiosulfonisc asids A end B, novel thiosulfonic acids from the deep-sea urchin *Echinocardium cordatum*. *Tetrahedron Lett.* 2001; 42(37): 6557-6560.
5. Lubenets V. I. Thiosulphonic acids derivatives: synthesis and properties. *Ukr Khimich Zh.* 2003; 69(3): 109-117. (In Ukrainian).
6. Dos Santos Edos A, Gonçalves FM, Prado PC, Sasaki DY, de Lima DP, Macedo ML. Synthesis method for thiosulfonate and report of its insecticidal activity in *Anagasta kuehniella* (Lepidoptera: Pyralidae). *Int J Mol Sci.* 2012; 13(11): 15241-15251.
7. Halenova TI, Nikolaeva IV, Nakonechna AV, Bolibrukh KB, Monka NY, Lubenets VI, Savchuk OM, Novikov VP, Ostapchenko LI. The search of compounds with antiaggregation activity among S-esters of thiosulfonic acids. *Ukr Biochem J.* 2015; 87(5): 83-92.
8. Nawrot U, Zaczyńska E, Czarny A, Lubenets V, Karpenko E. Antifungal activity of synthetic derivatives of allicin – continued research. *Mikologia Lekarska.* 2012; 19(4): 143-146.
9. Lubenets V, Karpenko O, Ponomarenko M, Zahoriy G, Krychkovska A, Novikov V. Development of new antimicrobial compositions of thiosulfonate structure. *Chem Chem Technol.* 2013; 7(2): 119-124.
10. Pershin GN, Kukushkina TS, Guskova TA. Esulan. *Khim Farmatsevt Zhurn.* 1976; 10(11): 146-150. (In Russian).
11. Banya AR, Karpenko OY, Lubenets VI, Baranov VI, Novikov VP, Karpenko OV. Influence of surface-active rhamnolipid biocomplex and ethylthiosulfanilate on growth and biochemical parameters of plants in oil contaminated soil. *Biotechnol Acta.* 2015; 8(5): 71-77.
12. Starovoitova SO, Oriabinska LB, Lubenets VI. Spectrum antimicrobial action of the original antifungal drug Esulanum. *Res Bull Nat Techn Univ Ukraine KPI.* 2015; 3: 68-76. (In Ukrainian).
13. Chemistry and biochemistry of nucleic acids / Edited by Zbarsky YB, Debov SS. L.: Medytsyna, 1968; 429 p. (In Russian).
14. Biological membranes. Methods / Edited by J. Findle. M.: Mir, 1990; 423 p. (In Russian).
15. Paranko SI, Gavryliuk VG, Golodok LP, Vinnikov AI. The study of respiration processes in clinical antibiotic-resistant strains of staphylococci. *Visnyk Dniprop Univ. BIOLOGY Series Biol Ecol.* 2006; 1(14): 140-144. (In Russian).
16. Eschenfeldt WH, Zhang Y, Samaha H, Stols L, Eirich LD, Wilson CR, Donnelly MI. Transformation of fatty acids catalyzed by cytochrome P450 monooxygenase enzymes of *Candida tropicalis*. *Appl Environ Microbiol.* 2003; 69(10): 5992-5999.
17. Zvyagilskaya RA. Yeast mitochondria: distinguishing features, and contribution to solving the general problems of bioenergetics. (Review). *Prikl Biokhim Mikrobiol.* 1995; 31(1): 50-59. (In Russian).
18. Mysiakina IS, Funtikova NS. Study of the composition of the lipid fungus Mucor strain of INMI under conditions of growth retardation with Nystatin. *Mikrobiologiia.* 1991; 60(4): 645-651. (In Russian).
19. Konev SV. Structural lability of biological membranes and regulatory processes. Minsk: Nauka i tehnika, 1987; 240 p. (In Russian).

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