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PROPERTIES OF CHEMOLITHOTROPHIC BACTERIA NEW STRAINS ISOLATED FROM INDUSTRIAL SUBSTRATES

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The purpose of the research was determination of strains *Acidithiobacillus ferrooxidans* MFLv37 and *Acidithiobacillus ferrooxidans* MFLad27, isolated from aboriginal consortium of coal beneficiation dumps and fly ash from coal combustion, resistance to heavy metals, forming part of these waste, as well as adaption ability of the strains to new substrates. New strains increased resistance to heavy metal ions as compared to *A. ferrooxidans* standard and collection strains is found; minimal inhibitory concentrations of heavy and toxic metals are determined; a number of metals that have negative impact on growth of isolated cultures are identified. It is shown that the minimal metals concentrations, at which strains growth still happens, are several times higher than their concentrations in technogenic waste. It has been found that isolated strains differed in their ability to adapt, as well as in growth rate and substrates oxidation. This is due to the specific conditions of microbiocenoses formation in making and further storage of rock dumps and fly ash, whereof the appropriate strains are isolated. The investigations indicate the necessity in directional selection of strains that are resistant to the toxic compounds and are able to oxidize various mineral substrates, as well as in their adaptation to new substrates for the extraction of heavy metals.

Key words: fly ash, rock dumps, aboriginal bacterial community, acidophilic chemolithotrophic bacteria, strains, leaching activity, ions of heavy metals.

The members of the genera *Acidithiobacillus* and *Sulfobacillus* have the greatest activity regarding leaching of metals from raw natural ores and technogenic substrates [1–3]. In previous investigations it was found that the most active group of microorganisms in the aboriginal consortium of coal beneficiation dumps and fly ash from coal combustion substrates is the group of acidophilic chemolithotrophic microorganisms as small mesophilic and most numerous moderately thermophilic ones — the members of the genera *Acidithiobacillus* and *Sulfobacillus* [4, 5]. The study of the properties of the most active pure cultures of the microorganisms isolated from the substrates aboriginal association has allowed to classify them as the representatives of *Acidithiobacillus ferrooxidans* and assign them the strain numbers MFLv37 and MFLad27 in a view of their habitats — Lviv-Volyn Coal Basin coal beneficiation dumps and fly ash from the coal combustion at Ladyzhynskaya thermal power station, respectively.

Acidithiobacillus ferrooxidans group is the most studied among acidophilic chemolithotrophic bacteria receiving energy from the oxidation of ferrous iron, elemental sulfur and its reduced compounds and sulfide minerals; it has a high level of resistance to heavy metals. Resistance is an important property of microorganisms of different taxonomic groups, thanks to which resistant forms of microorganisms in the environment with a high content of heavy metals appear. So, in [6] it is shown that the differences between the strains in substrate oxidation activity are caused by environment and conditions of microbiocenosis formation whereof the strains were isolated. *A. ferrooxidans* strain, isolated from a complex mineralogical composition substrate, is characterized by a higher growth rate and oxidation degree of various pyrite types than the strain *A. ferrooxidans* from poorer composition ore dump. In addition, in environmental objects with a high content of heavy metals, steadier to heavy metals (resistant) forms of microorganisms of different taxonomic groups appear and formed. There are data about the study of *A. ferrooxidans* strains, isolated from the natural environment, adaptation to various sulfide ores [7–9]. However, the issue of technogenic mineral raw materials usage for energy obtaining by representatives of *A. ferrooxidans* has been insufficiently studied. Therefore, the study of properties of new resistant highly active strains capable of adaptation and growth on the environmental objects with a high content of heavy metals is an urgent task of biotechnology.

The objective was to study the properties of the new isolated from technogenic waste strains of chemolithotrophic bacteria *Acidithiobacillus ferrooxidans* MFLv37 and MFLad27 — as resistance to heavy metals, and the ability to adapt to new substrates compared with highly active collection (*A. ferrooxidans* VKM468) and standard (*A. ferrooxidans* ATCC23270) strains.

Materials and Methods

Isolated strains *Acidithiobacillus ferrooxidans* MFLv37 and MFLad27, collection *Acidithiobacillus ferrooxidans* VKM468 and standard *Acidithiobacillus ferrooxidans* ATCC23270 strains are stored in the Museum of Microbiology, Virology and Biotechnology Department of Mechnykov Odesa National University, which is a branch of the National Collection of Microorganisms. Strains *A. ferrooxidans* ATCC23270 and *A. ferrooxidans* VKM468 are used in biohydrometallurgical processes for the bacterial leaching of metals from sulphide ores [10]. In addition, *A. ferrooxidans* ATCC23270 is capable of solubilizing the sulfur from the coal surface [11].

New strains *A. ferrooxidans* MFLv37 and MFLad27 are characterized by the following features. There are Gram-negative rods, 0.2–0.4 microns in size, mobile. They are obligate anaerobes, chemolithoautotrophs, strictly acidophilic, mesophilic (optimum temperature for growth is 35.0 ± 2.0 °C). They are cultivated in media with iron, sulfur, thiosulfate, use thiourea as the single energy source, capable of growth in mixotrophic conditions. They are stored on an agar culture medium in the fridge with passages on fresh medium every 1–2 months. To cultivate the strains for biomass accumulation and metal ions leaching from substrates the medium 9K is used (g/dm^3): $(NH_4)_2SO_4$ — 3,0; KCl — 0,1; $MgSO_4$ — 0,5; K_2HPO_4 — 0,5; $Ca(NO_3)_2$ — 0,01. The only source of energy is $FeSO_4 \cdot 7H_2O$ in concentrations: 44.5 g/dm^3 for maximum strains biomass accumulation and 12.0 g/dm^3 for the most effective leaching of metals from the studied substrates. The original titer of all tested strains is the same and is equal to $(4.1 \pm 0.3)\times10^3$ cells/ml.

The objects of the research were the substrates of: rock dumps resulting from the coal beneficiation of mines of Lviv-Volyn Coal Basin by gravity and flotation methods at the Central Concentrating Factory (CCF) "Chervonogradska" of the joint stock company (JSC) "Lviv Coal Company", and fly ash from high temperature combustion of mixture of domestic fossil coals at Ladyzhynskaya thermal power station.

Rock dumps substrate has the following characteristics. This is clay earth material, crystallized with the overwhelming majority of fairly large (5–7 mm) particles, represented by argillites, kaolinite, quartz sandstone-type mineral, pyrite, containing coal up to 17.0%, sulfur — up to 1.5% and organic mass — up to 2.0%. Chemical composition (%): Fe — 4.46; Al — 1.39; Si — 15.90; Ti — 0.42; Ca — 1.72; Cu — $6.22 \cdot 10^{-3}$; Zn — $1.13 \cdot 10^{-2}$; Mn — $3.18 \cdot 10^{-2}$; Pb — $0.42 \cdot 10^{-2}$; Ni — $1.34 \cdot 10^{-2}$; Cd — $2.82 \cdot 10^{-4}$; Sn — $3.52 \cdot 10^{-2}$; Cr — $0.99 \cdot 10^{-2}$; V — $1.50 \cdot 10^{-2}$; Co — $1.16 \cdot 10^{-2}$; Sr — $2.11 \cdot 10^{-2}$; Ba — $5.19 \cdot 10^{-2}$; Zr — $1.73 \cdot 10^{-2}$; Rb — $1.41 \cdot 10^{-2}$; Nb — $1.40 \cdot 10^{-3}$; La — $4.80 \cdot 10^{-3}$; Ce — $6.90 \cdot 10^{-3}$; Ga –$1.21 \cdot 10^{-3}$; Ge — $2.60 \cdot 10^{-3}$. Storage life in dumps is 1 year. The number of aboriginal acidophilus chemolithotrophic bacteria is: of mesophilic ones $(6.4 \pm 0,6)\times10^4$ and of moderately thermophilic ones $(7.4 \pm 0,3)\times10^8$ cells/ml [12].

Fly ash substrate is an amorphous poorly crystallized pulverized product with an overwhelming majority of quartz, carbonate and silicate phases, oxides of iron and aluminum. Chemical composition (%): Fe — 5.93; Al — 3.89; Si — 12.10; Ti — 4.16; Ca — 0.20; Cu — $6.82 \cdot 10^{-3}$; Zn — $3.27 \cdot 10^{-2}$; Mn — $5.73 \cdot 10^{-2}$; Pb — $1.09 \cdot 10^{-2}$; Ni — $1.77 \cdot 10^{-2}$; Cd — $5.31 \cdot 10^{-4}$; Sn — $2.07 \cdot 10^{-2}$; Cr — $2.18 \cdot 10^{-2}$; V — $2.15 \cdot 10^{-2}$; Co — $3.05 \cdot 10^{-2}$; Sr — $6.56 \cdot 10^{-2}$; Ba — $6.34 \cdot 10^{-2}$; Zr — $2.37 \cdot 10^{-2}$; Rb — $1.16 \cdot 10^{-2}$; Nb — $1.90 \cdot 10^{-3}$; La — $4.20 \cdot 10^{-3}$; Ce — $7.40 \cdot 10^{-3}$; Ga –$1.02 \cdot 10^{-3}$; Ge — $2.80 \cdot 10^{-3}$; S — 1.24; C (incomplete combustion) — 9.98.

Storage life in dumps is 1 year. The number of native acidophilus chemolithotrophic bacteria is: of mesophilic ones $(5.9 \pm 0.6) \cdot 10^4$ and of moderately thermophilic ones $(6.4 \pm 0.5) \cdot 10^7$ cells/ml.

The increase in biomass was determined by the number of cells counted in Goryaev chamber with the light microscope Primo Star PC (Germany). Analysis of solutions for metal content was performed using atomic absorption spectroscopy with devices AAS-1 (Germany) and C-115PK Selmi (Ukraine). The reliability of the results was evaluated by Student's t test with a probability of $P < 0.05$. The ratio (%) of the amount of metal that passed into the solution as a result of contact a nutrient medium with a substrate in microorganisms presence to the initial amount of metal in the original solid substrate is referred to as "degree of metal extraction". Indicator 100% corresponds to complete metal transition from the substrate into the solution.

Results and Discussion

In the initial phase the resistance of new strains *A. ferrooxidans* MFLv37 and *A. ferrooxidans* MFLad27 to heavy metals, which are the part of technogenic waste, has been studied.

Determination of copper, iron, zinc, lead, cadmium, manganese and germanium ions minimal concentration, at which *A. ferrooxidans* MFLv37 and *A. ferrooxidans* MFLad27 cells growth still happens, carried out in the course of their cultivation on an agar medium 9K with iron, adding sulfates of these metals in ever increasing concentrations. The minimal inhibitory concentration (MIC), at which the growth of the test strains may be still recorded, was determined. The term of cultivation was 7 days at 30.0 ± 2.0 °C. The results are shown in the table.

The findings indicate a high resistance level to heavy metals of strains isolated from technogenic waste of coal and energy industry. It is found that the minimal metal concentrations, at which strains growth still happens, are several times greater than their content in waste itself. The both new strains equally reacted to impact of manganese, zinc, copper and germanium. *A. ferrooxidans* MFLv37 and *A. ferrooxidans* MFLad27 strains are the most sensitive to cadmium; the most stable — to iron. By the negative influence on the growth of new isolated strains the following row is made:

$$Fe < Zn < Mn < Cu < Pb < Ge < Cd.$$

When comparing the stability of new strains with standard and collection ones it has been found that the latest were more sensitive to metals, forming part of technogenic waste. This could be explained by the fact that new strains are isolated from the aboriginal community, formed during the production and storage of technogenic objects with complex mineralogical composition and high concentrations of heavy metals. The high level of thiobacteria stability to metals is associated with the fact that the genetic basis of the resistance is extrachromosomal stability factors presence in bacteria — plasmids and transposons [13], and also with poor permeability of the cell wall for the heavy metal ions, formation of a large amount of slime, which binds and inactivates metals, et al. [14].

The next step is studying the adaptation of isolated strains *A. ferrooxidans* MFLv37 and

Minimal inhibitory metal concentrations (mg/dm^3) for *A. ferrooxidans* strains

Element	MIC, mg/dm^3				Determined concentration in substrates, mg/kg	
	A. ferrooxidans MFLv37	*A. ferrooxidans* MFLad27	*A. ferrooxidans* VKM 468	*A. ferrooxidans* ATCC23270	Rock dump	Fly ash
Cu	1200.0	1500.0	7.0	5.0	62.18	68.18
Zn	4500.0	4500.0	5.0	5.0	112.52	327.33
Mn	1500.0	1650.0	7.0	5.0	317.72	572.60
Pb	600.0	850.0	2.0	1.0	42.20	108.74
Cd	25.0	25.0	2.0	2.0	2.82	5.31
Fe	$150.0 \cdot 10^3$	$150.0 \cdot 10^3$	$10.0 \cdot 10^3$	$10.0 \cdot 10^3$	$44.57 \cdot 10^3$	$59.31 \cdot 10^3$
Ge	150.0	150.0	12.0	15.0	26.00	28.00

A. ferrooxidans MFLad27 to new technogenic waste as a new source of energy. Experiments were performed using the standard 9K medium and technogenic substrates samples as a sole energy source. Ratio of solid and liquid phases (S:L) in the experiments was the following: 1:2, 1:3 and 1:10; temperature of 30.0 ± 2.0 °C, pH ≤ 3. Successive reinoculations with growing volumes of new substrates were performed when the strains reached maximum quantity of biomass in each passage. Control was an indicator of strains biomass growth in the medium 9K with 12.0 g/dm^3 of $FeSO_4 \cdot 7H_2O$ (as the sole source of energy).

Fig. 1 and 2 show the results of strains *A. ferrooxidans* MFLv37 and *A. ferrooxidans* MFLad27 biomass growth with various amounts of substrates usage as power sources. Obviously, the both strains, which were grown for a long time using Fe^{2+}, require lengthy adaptation as to substrate from which they were isolated, and to a new substrate.

As follows from Fig. 1, when switching from ferrous iron oxidation to old substrate (dumps) and new one (ash) oxidation there is a decrease in *A. ferrooxidans* MFLv37 bacteria cell number in the first 6 hours of cultivation. Subsequently, the active biomass growth occurs on both substrates with pronounced inversely proportional dependence on its quantity: less substrate with respect to the medium, more active biomass increasing on it. In the case of the test strain cultivation at S:L = 1: 2 the time to reach the stationary phase of growth for *A. ferrooxidans* MFLv37 was 36 and 42 hours, respectively, for the new (ash) and old (dumps) substrates. Upon that, the quantity of biomass was respectively 4.0–6.0 times less than in the control experiment. With a decrease in the amount of the solid substrate to the S:L = 1: 3 the time to reach the stationary phase of growth for *A. ferrooxidans* MFLv37 was 48 hours for the new (ash) and 66 hours for the old (dumps) substrates. In addition, the quantity of biomass was respectively 1.7–2.0 times less than in the control. During the further test strain cultivation the cell number did not increase, but they remain viable. With a decrease in technogenic waste number to S:L = 1:10 *A. ferrooxidans* MFLv37 growth was observed over the entire period of cultivation, the number of bacteria was even higher than data in the control group. The sharp decrease in the number of bacteria in the early adaptation period and a minor amount of bacteria with an excess of the substrate (S:L = 1: 2, and S:L = 1: 3) may be explained by attaching a significant number of formed cells to the surface of substrate particles [15].

Similar results were obtained while studying the biomass growth of *A. ferrooxidans* MFLad27 strain on the old and the new substrates (Fig. 2) with expressed adaptation period in the first 6–12 hours, and with an excess of a substrate — up to 36 hours of cultivation. In the process of *A. ferrooxidans* MFLad27 adaptation to the growing volume of rock dumps the same pattern as that for *A. ferrooxidans* MFLv37 to fly ash are marked, with the only difference being that the strain *A. ferrooxidans* MFLad27 is much worse adapted to other technogenic substrate as an energy source.

In the next stage of the work the oxidative capacity of adapted to new substrates strains has been studied compared with maladapted strains, standard *A. ferrooxidans* ATCC23270 and collection *A. ferrooxidans* VKM468. The leaching process was performed at a ratio of

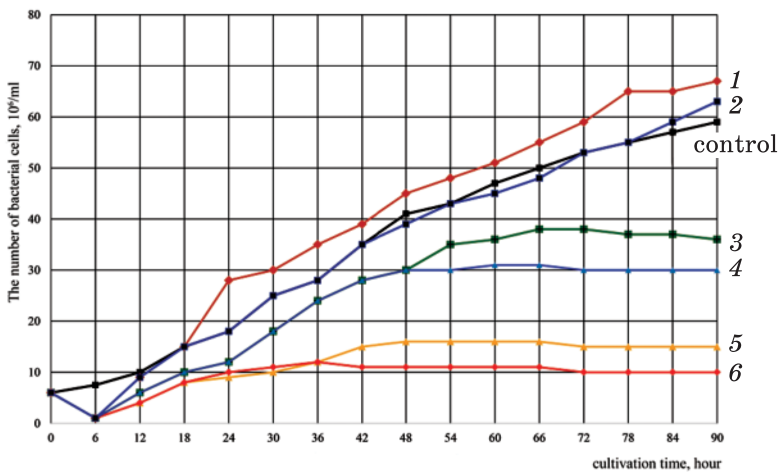

Fig. 1. **The amount of *A. ferrooxidans* MFLv37 biomass with growth on:**
1 — dumps S:L = 1:10; *2* — ash S:L = 1:10; *3* — dumps S:L = 1: 3; *4* — ash S:L = 1: 3;
5 — dumps S: L = 1: 2; *6* — ash S:L = 1: 2

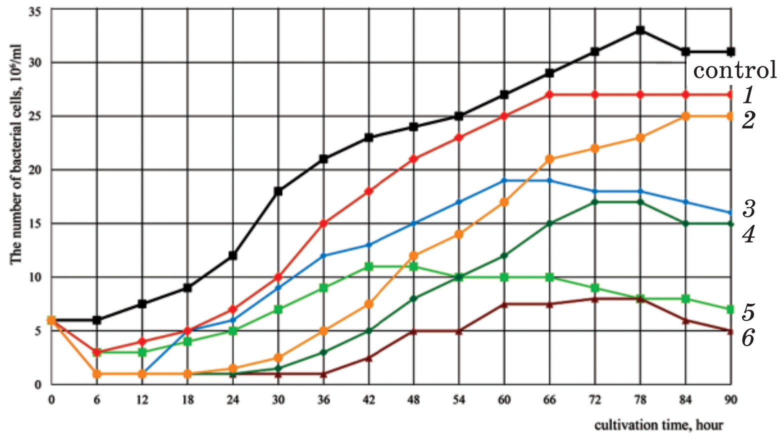

Fig. 2. **The amount of *A.ferrooxidans* MFLad27 biomass with growth on:**
1 — ash S:L = 1:10; *2* — dumps S:L = 1:10; *3* — ash S:L = 1: 3; *4* — dumps S:L = 1: 3;
5 — ash S:L = 1: 2; *6* — dumps S:L = 1: 2

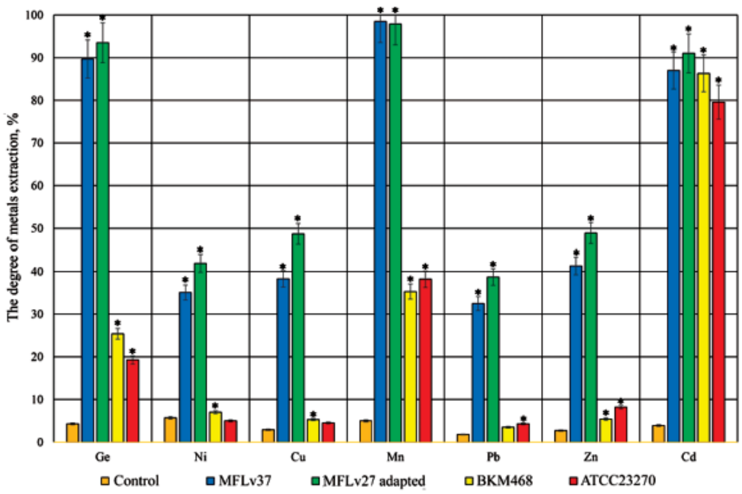

Fig. 3. **Metals recovery (%) from fly ash by *A. ferrooxidans* strains**
Hereinafter: * — $P < 0.05$ compared with the control

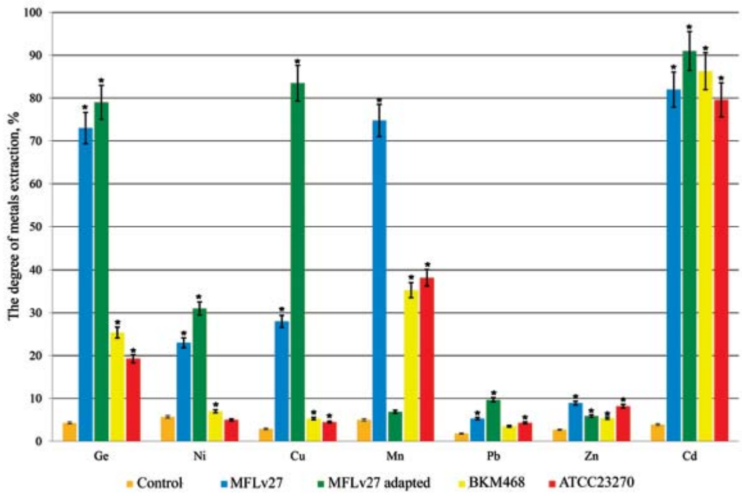

Fig. 4. **Metals recovery (%) from rock dumps by *A. ferrooxidans* strains**

S:L = 1:10, at temperature of 30.0 ± 2.0 °C, pH ≤ 2, for 7 days. The control was the results of metals leaching from substrates by nutrient medium under sterile conditions in the absence of microorganism strains. Fig. 3 and 4 show the results of these experiments.

Analysis of the results indicates that the both new isolated strains had a much higher oxidizing ability compared to the standard and the collection strains. Furthermore, adapted strains in most cases leached metals into solution more active compared to maladapted strains.

Comparison of the two strains showed that *A. ferrooxidans* MFLad27, isolated from fly ash obtained from coal combustion, exhibits less adaptability relative to growth rate (Fig. 2) and oxidation degree (Fig. 4). This indicates that *A. ferrooxidans* MFLv37 has more pronounced regulatory mechanisms, when reinoculating from one medium ($FeSO_4 \cdot 7H_2O$) to another (dump or ash substrate) an enzyme systems activation occurs that associated with the oxidation of sulfur compounds, and consequently the growth and oxidation degree increase [6, 9]. This is due to the specific conditions of rock dumps and fly ash microbiocenoses formation, whereof appropriate strains are obtained.

The results indicate that the main factors affecting the diversity of *A. ferrooxidans* strains are their formation and habitation in different ecological niches, in the substrates of different origin, as well as physico-chemical properties specificity. Search and directed selection of strains with high resistance and oxidative capacity, adaptation to the new natural and technogenic substrates are urgent tasks.

REFERENCES

1. *Ivanov M. V., Karavayko G. I.* Geological microbiology. *Mikrobiologiya*. 2004, 5 (73), 581–597. (In Russian).
2. *Norris P. R., Burton N. P., Foulis N. A. M.* Acidophiles in bioreactor mineral processing. *Extremophiles*. 2000, V. 4, P. 71–76.
3. *Blayda I. A.* Extraction of valuable metals from industrial waste biotechnological methods (Review). *Energotekhnologii i resursosberezhenie*. 2010, V. 6, P. 39–45. (In Russian).
4. *Blayda I. A., Vasileva T. V., Slyusarenko L. I., Barba I. N., Ivanitca V. A.* Composition and leaching activity of energy industrial waste microbiocenosis. *Problemy ekolohichnoi biotekhnolohii*. 2013, V. 1. Available at: http://jrnl.nau.edu.ua /index.php/ecobiotech/article/view/4592. (In Russian).
5. *Blayda I. A.* Composition and activity of bacterial community of coal tailing. *Biotechnol. acta*. 2014, 7 (5), 94–100. doi: 10.15407/biotech7.05.094.
6. *Tupikina O. V., Kondrateva T. F., Samorukova V. D., Rassulov V. A., Karavayko G. I.* Dependence of the strains *Acidithiobacillus ferrooxidans* phenotypic characteristics from the physical, chemical and electrical properties of pyrites. *Mikrobiologiya*. 2005, 74 (5), 596–603. (In Russian).
7. *Karavayko G. I., Kuznetsov S. I., Golomzik Ye. I.* The Role of Microorganisms in the leaching of metals from ores. *Moskva: Nauka*. 1972, 248 p. (In Russian).
8. *Silver M.* Metabolic mechanisms of iron-oxidizing thiobacilli. *Metallurgical applications of bacterial laeching and related microbiological phenomena*. Murr L. E., Torma A. E., Brierley J. A. (Eds). *New York: Academic Press*. 1978, P. 3–17.
9. *Grudev S. N.* The differences between the strains of *Thiobacillus ferrooxidans* on the ability to oxidize sulfide minerals. *Biogeotehnologija metallov*. Karavayko G. I., Grudev S. N. (Red.). *Moskva: Centr mezhdunarodnykh proektov GKNT*. 1985, P. 85–99. (In Russian).
10. *Tuovinen O. H., Fry I. J.* Bioleaching and mineral biotechnology. *Curr. Opin. Biotechnol*. 1993, V. 4, P. 344–355.
11. *Juszczak A., Domka F., Kozlowski M., Wachowska H.* Microbial desulfurization of coal with *Thiobacillus ferrooxidans* bacteria. *Fuel*. 1994, 74 (5), 725–728.
12. *Vasileva T. V.* Diversity of the microorganisms community in the technogenic ecosystems of Ukraine fuel-and-energy complex. *Problemy ekolohichnoi biotekhnolohii*. 2014, V. 1. Available at: http://ecobio.nau.edu.ua / index.php/ecobiotech/article/view /6660. (In Ukrainian).
13. *Khmelev I. A.* Plasmids and evolution of microorganisms. *Uspekhi sovremennoy biologii*. 1985, 99 (3), 323–337. (In Russian).
14. *Kenji Iwahorl, Fumiaki Takeuchi, Kazuo Kamimura, Tsuyoshi Sugio*. Ferrorous Iron-Dependent Volatilization of Mercuri by the Plasma Membrane of *Thiobacillus ferrooxidans*. *Apll. Envir. Microbiol*. 2000, P. 3823–3827.
15. *Kondrateva T. F., Ageeva S. N., Pivovarova T. A., Karavayko G. I.* Characteristics of chromosomal DNA restriction profiles strains *Acidithiobacillus ferrooxidans*, adapted to different substrates oxidation. *Mikrobiologiya*. 2002, 71 (4), 514–520. (In Russian).

ВЛАСТИВОСТІ НОВИХ ШТАМІВ ХЕМОЛІТОТРОФНИХ БАКТЕРІЙ, ЩО ЇХ ВИДІЛЕНО З ТЕХНОГЕННИХ СУБСТРАТІВ

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Метою роботи було встановлення резистентності виділених з аборигенного консорціуму породних відвалів вуглезбагачення і золи-виносу від спалювання вугілля штамів *A. ferrooxidans* МФLv37 і *A. ferrooxidans* МФLad27 до важких металів, що входять до складу цих відходів, а також здатності штамів до адаптації до нових субстратів. Встановлено підвищену стійкість нових штамів до іонів важких металів порівняно з типовим і колекційним штамами *A. ferrooxidans*, визначено мінімальні інгібуючі концентрації важких металів, виявлено низку металів, що негативно впливають на ріст ізольованих культур. Показано, що мінімальні концентрації металів, за яких ще відбувається ріст штамів, у декілька разів перевищують їх вміст у техногенних відходах. Встановлено, що виділені штами відрізнялися за здатністю до адаптації, а також за швидкістю росту і окиснення субстратів. Це зумовлено специфічними умовами формування мікробіоценозів у процесі утворення і подальшого зберігання породних відвалів та золи-виносу, з яких виділено відповідні штами. Проведені дослідження свідчать про необхідність спрямованої селекції культур, стійких до токсичних сполук і здатних окиснювати різні мінеральні субстрати, а також їх адаптації до нових субстратів для вилучення важких металів.

Ключові слова: зола-винос, породні відвали, аборигенне бактеріальне угруповання, ацидофільні хемолітотрофні бактерії, штами, активність вилуговування, іони важких металів.

СВОЙСТВА НОВЫХ ШТАММОВ ХЕМОЛИТОТРОФНЫХ БАКТЕРИЙ, ВЫДЕЛЕННЫХ ИЗ ТЕХНОГЕННЫХ СУБСТРАТОВ

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Целью работы было установление резистентности выделенных из аборигенного консорциума породных отвалов углеобогащения и золы-уноса от сжигания угля штаммов *A. ferrooxidans* МФLv37 и *A. ferrooxidans* МФLad27 к тяжелым металлам, входящим в состав этих отходов, а также способности штаммов к адаптации к новым субстратам. Установлена повышенная устойчивость новых штаммов к ионам тяжелых металлов по сравнению с типовым и коллекционным штаммами *A. ferrooxidans*, определены минимальные ингибирующие концентрации тяжелых металлов, выявлен ряд металлов, оказывающих негативное влияние на рост изолированных культур. Показано, что минимальные концентрации металлов, при которых еще происходит рост штаммов, в несколько раз превышают их содержание в техногенных отходах. Установлено, что выделенные штаммы отличались по способности к адаптации, а также скорости роста и окисления субстратов. Это обусловлено специфическими условиями формирования микробиоценозов в процессе образования и дальнейшего хранения породных отвалов и золы-уноса, из которых выделены соответствующие штаммы. Проведенные исследования свидетельствуют о необходимости направленной селекции культур, устойчивых к токсичным соединениям и способных окислять различные минеральные субстраты, а также их адаптации к новым субстратам для извлечения тяжелых металлов.

Ключевые слова: зола-унос, породные отвалы, аборигенное бактериальное сообщество, ацидофильные хемолитотрофные бактерии, штаммы, активность выщелачивания, ионы тяжелых металлов.