#### Abstract

# of the dissertation work of **Akhmet Ainagul** for the degree of Doctor of Philosophy (PhD) in the specialty 6D070100 –Biotechnology on the topic: "**Technology of the valuable components bioleaching from phosphoruscontaining wastes of the Southern Kazakhstan**"

**General characteristics of the research.** The dissertation work is aimed at developing the technology of bio-leaching of valuable components from phosphorus-containing wastes of Southern Kazakhstan by clarifying their basic physico-chemical and biological characteristics, followed by the compilation of three compositions from isolated active strains of microorganisms used under varying conditions of waste processing to extract a complex of valuable metals.

**Relevance of the research topic**. One of the serious environmental problems of Kazakhstan is the presence of significant volumes of industrial and man-made waste. Currently, about 31.6 billion tons of industrial waste are stored on the territory of the state, including in Southern Kazakhstan more than 150.8 million tons of polymetallic, 2.8 million tons of lead-zinc and more than 500 thousand tons of phosphorus-containing waste. Active processes of urbanization have led to the fact that the places of storage of man-made waste gradually turned out to be within the city. As a result of water-wind erosion processes, man-made waste has become a source of danger to the environment and public health. Thus, medical studies have found that as a result of the influence of phosphorus-containing waste from Shymkent, the indicators of reticulocytosis and hypochromic anemia are increased in the body of people living near the storage area of toxicants.

There are many technologies for the disposal of industrial man-made waste, among them biotechnological methods are the most promising. Currently, the most widespread application in the processing of man-made waste has acquired the technology of bio-leaching, which is used in the mining industry of China, France, Pakistan, South America, Russia, Spain and Kazakhstan. Bioleaching of phosphorus-containing waste located on the territory of Shymkent in Southern Kazakhstan will not only prevent environmental impact on the environment, but will also allow additional valuable components to be extracted from them.

**The purpose of the study:** To develop a technology for bio-leaching of valuable components from phosphorus-containing wastes of Southern Kazakhstan.

#### **Research tasks**:

- Study of physico-chemical, mineralogical and biological properties of phosphorus-containing waste in Shymkent;

- Study of the peculiarities of the spread of microorganisms in phosphoruscontaining waste located in Shymkent;

- Isolation and selection of new strains of microorganisms promising for bioleaching of valuable components from phosphorus-containing waste;

- Development of technology for bioleaching of valuable components from phosphorus-containing waste in Shymkent.

**Object of research**: phosphorus-containing technogenic waste stored on the territory of Shymkent in Southern Kazakhstan.

## Scientific novelty of the work:

- It has been established that the mineralogical composition of phosphoruscontaining slag and sludge stored on the territory of Shymkent is represented by pseudovollastonite, cuspidine, melilite, akermanite, rankinite, fluorapatite, fluorite, silicocarnotite, calcite, quartz, ferrophosphore. It was revealed that the content of  $1.0 \pm 0.1$  vol.% of slag in the substrate has a stimulating effect on test objects, and the concentration of  $10.0 \pm 0.9$  vol.% is lethal to all organisms. Sensitive species of hydrobionts: *Scenedesmus guadricauda, S. protuberans, Synedra ulna, Rotatoria sallidina, Aeolosoma sp., Nematoda sp., Gammarus lacustris, Daphnia magna* can be used for bioindication purposes.

- It was found that the microbial population of phosphorus-containing waste located in Shymkent consists of heterotrophic, nitrogen-fixing, denitrifying, sulfur-oxidizing, nitrifying bacteria, micromycetes and actinomycetes, with a predominance of the proportion of micromycetes. It was revealed that the largest number of microorganisms  $10^7$ - $10^8$  cell/g is concentrated at a depth of 10-30 cm, which is explained by the presence of optimal parameters of the gas-air regime, humidity and biogenic elements.

- As a result of selection and screening work, new strains of microorganisms have been isolated from phosphorus-containing waste, promising for use in bioleaching and taxonomically defined as *Aspergillus niger ASIA*, *A. tubingensis ASPN*, *A. terreus JOM*, *A. flavus AsZ*, *A. flavus ASF*, *Sulfobacillus thermosulfidooxidans ST*, *Galionella capsiferriformans TS*, *Pseudomonas* stutzeri NJA, *Methyloversatilis thermotolerans MSO*, *Ralstonia pickettii ASA*, *R. pickettii TS2*, *Zoogloea resiniphila NS1; Acinetobacter sp. NAO*.

- On the basis of new strains of microorganisms, microbial consortia have been developed and capable of selectively extracting valuable components from waste: the TIAI consortium from strains of iron-oxidizing bacteria *Acidithiobacillus ferrooxidans* ThIO1, *A. ferrooxidans* ThIO2 extracts Zn, Al, Mn, Rb; the ANAT consortium consisting of strains of micromycetes *Aspergillus niger ASIA* and *A. tubingensis ASPN* extracts Cu, Ag, V, Ru, Mo, Ba, Zr; NEMfos consortium - from nitrifiers *Nitrosomonas europeae Nit1* and *M. thermotolerans MSO* extracts Ce and La.

- A three-stage technology of bio-leaching of phosphorus-containing waste located in Shymkent has been developed using microbial consortia TIAI, ANAT and NEMfos, which can be used separately or sequentially, depending on the initial physico-chemical characteristics of waste, with the extraction of up to  $85.2 \pm 7,8\%$  of valuable metals from waste.

## The degree of validity and reliability of the dissertation work.

The reliability and accuracy of the scientific results were determined using proven methods for assessing the physico-chemical and biological characteristics of waste and using a large sample size. The results of the obtained data are proved on the basis of a comparative assessment with the results of similar studies. The results presented in the dissertation work were obtained during many years of research conducted at least three times and statistically processed using statistical processing methods - variance and correlation-regression analysis, where the level of accuracy of experiments was determined with a significant probability of the sample under study and the reliability of the results obtained. In addition, the research results were processed using the Excel computer program to formalize them in the form of graphical data. In order to carry out the planned research and biotechnological practices, special certified methods, standards of the Russian Federation and the Republic of Kazakhstan have been applied. The equipment and materials used in the course of the study comply with the requirements of regulatory and technical documents.

# The main provisions submitted for protection:

• The refined mineralogical composition of phosphorus-containing wastes is represented by pseudovollastonite, cuspidine, melilite, akermanite, rankinite, fluorapatite, fluorite, silicocarnotite, calcite.

•  $1.0 \pm 0.1$  vol.% content of phosphorus-containing waste in the substrate has a stimulating effect on test plants with an increase in morphometric indicators by  $23.1 \pm 2.0\%$ , an increase in concentration of wastes to  $10.0 \pm 0.9$  vol. % has a lethal effect on all organisms, while phosphorus-containing sludge is more toxic than slag.

• The microflora of phosphorus-containing waste consists of 46,7% of micromycetes, 40,8% of bacteria, including heterotrophic, nitrogen-fixing, denitrifying, sulfur-oxidizing, nitrifying bacteria and 8,8% of actinomycetes.

• 12 new strains of microorganisms promising for the purposes of biogeotechnology have been identified, the taxonomic affiliation of which is determined by PCR analysis.

• On the basis of new strains of microorganisms, consortia TIAI, ANAT, NEMfos have been developed, capable of selectively leaching valuable components from phosphorus-containing waste under optimal cultivation conditions.

• The application of the developed three-stage technology of bio-leaching of phosphorus-containing waste using microbial consortia separately or in combination, depending on the variation of the physico-chemical characteristics of waste, contributes to the extraction of up to  $85,2 \pm 7,8\%$  of valuable metals from them.

# Theoretical and practical significance of the research results.

- The theoretical significance of the research results lies in the data on the reaction of test plants and hydrobiont organisms to phosphorous-containing toxicants; on the peculiarities of the spread of microorganisms in waste storage sites; the revealed structure of the microbial population of phosphorus-containing slags and slimes; isolated and described new strains of microorganisms and their taxonomic characteristics identified by PCR analysis.

- The practical significance of the research results lies in new strains of microorganisms promising for biogeotechnology; the developed microbial consortia TIAI, ANAT, NEMfos separately or together can be used to obtain concentrates of valuable components from various technogenic or mineral wastes; three-stage technology of bio-leaching can be recommended for use in the processing of various phosphorus-containing waste in Kazakhstan. The

effectiveness of the developed technology of three-stage bio-leaching is confirmed by the test certificate at the production enterprise of Kainar LLP (Appendix A).

**Approbation of the results of the dissertation**. The main results of the research work were discussed at the following international conferences: "Fundamental and applied scientific research: current issues, achievements and innovations" (Mat. XXIII Int. Sci. and Pract. Conf., Penza, 2019), "Auezov Readings - 17: new impulses of science and spirituality in the world space" (Mat. Int. Sci. and Pract. Conf., Nur-Sultan, 2019), "Scientific community of students: Interdisciplinary research" (Mat. XCIX Int. student. Sci.-pract. conf., Novosibirsk, 2020).

**Publications on the research topic.** On the topic of the dissertation, 13 scientific papers have been published at international and republican scientific - practical conferences, 1 article in the international journal from Scopus database, 3 articles in journals publications recommended by the committee for control in the field of education of the Ministry of Science and Higher Education of the Republic of Kazakhstan.

**Personal contribution of the dissertation.** The author performed a search and analysis of literary data, conducted laboratory studies and extensive tests in industrial conditions, statistically processed the obtained primary experimental data, prepared publications on the topic of the dissertation, prepared and executed the dissertation work.

**Connection with the plan of the main scientific works.** The dissertation work was carried out in accordance with the priority direction of the development of science of the Republic of Kazakhstan, aimed at the development of the waste processing sector with the production of products from secondary raw materials. Dissertation research on bioleaching of rare-earth elements from phosphorus-containing waste was carried out within the framework of the grant of the Ministry of Education and Science of the Republic of Kazakhstan No. 1969/GF4 "Development of a method for biological leaching of lanthanum, cerium and neodymium from polymetallic, phosphorus-containing and lead-zinc wastes of the South of Kazakhstan" (2015-2018) and the initiative theme of the M. Auezov SKU Research Institute of Ecology and Biotechnology of the M. Auezov SKU "Rational resource-saving technologies".

# Structure and scope of the dissertation

The dissertation work consists of an introduction; an analytical review; a chapter describing the objects and methods of research; a chapter with an analysis of the results of the study; conclusions; 5 appendices; a list of references presented by 212 titles. The dissertation consists of 147 pages, contains 60 figures and 13 tables.