

ANNOTATION

dissertation work of Daurov D.L. on the topic «Assessment potential of sweetpotatoes (*Ipomoea batatas* L.) for phytoremediation of plumbum in contaminated areas of southern Kazakhstan», submitted for the degree of Doctor of Philosophy (PhD) in the educational program 8D05204 – Ecology

Relevance of the research topic

Currently, due to the intensive pace of industrial production, the content of anthropogenic pollution in the environment is growing. Very dangerous pollutants are heavy metals: arsenic (As), cadmium (Cd), mercury (Hg), selenium (Se), lead (Pb), zinc (Zn), nickel (Ni), molybdenum (Mo), copper (Cu), antimony (Sb), etc. It is known that excessive content of heavy metals can lead to serious environmental consequences. Heavy metals have become the main pollution problem in the industrial regions of Kazakhstan and cannot be destroyed by degradation. In our Republic, the emission of powerful man-made pollution occurs in the South Kazakhstan region. The cities of Shymkent and Kentau are included in the zone of increased soil pollution with heavy metals. As is known, the main concern in the city of Shymkent is the former Shymkent lead plant, which actively produced lead and zinc in Soviet times.

The scientific novelty lies in identifying sweetpotato (*Ipomoea batatas* L.) varieties tolerant to lead capable of remediating contaminated soils, as well as determining the gene expression spectrum in tolerant and sensitive sweetpotato varieties in response to elevated lead concentrations. Sweetpotato has a well-developed biomass of the upper part of the plant, which consists of vines up to 5 meters long and a well-developed root system, which makes this crop a good candidate for remediating heavy metals from contaminated soils. Previously, work on the development of biotechnology for obtaining virus-free planting material of sweetpotato showed that this crop grows well in our climatic conditions. Previously, cleaning of areas contaminated with heavy metals by sweetpotato was not carried out. Exposure to heavy metals activates a large number of genes and proteins, linking signaling pathways that provide tolerance to various heavy metals. Sweetpotatoes are known to be rich in antioxidants such as beta-carotene and anthocyanin. There is a lot of information about the antioxidant systems of cells and their activation under stress. As a rule, under the influence of unfavorable conditions, oxidative stress processes develop in the cells of living organisms, caused by the generation of reactive oxygen species (ROS). These systems include both low-molecular non-protein antioxidants (carotenoids, proline, ascorbate, glutathione, flavonoids, etc.) and specific antioxidant enzymes (SOD, CAT, peroxidases, etc.) and SH proteins. Studying the features of the functioning of antioxidant systems is important for understanding how plants adapt to anthropogenically changed environmental conditions.

Purpose of the research: Selection, testing and study of sweetpotato varieties tolerant to elevated lead concentrations in soil, as well as those capable of

accumulating lead in tissues for phytoremediation of contaminated areas in southern Kazakhstan.

To achieve this goal, the following research objectives have been set:

1. Determination of the level of soil pollution with lead and other heavy metals by the Shymkent Lead Plant.
2. Conduct experimental cultivation of sweetpotatoes in lead-contaminated areas of southern Kazakhstan.
3. Optimization of solid nutrient medium for sweetpotato cultivation.
4. Optimization of nutrient medium for *in vitro* cloning and transplantation of sweetpotato plants.
5. Determination of maximum permissible concentrations of heavy metals in the nutrient medium.
6. *In vitro* screening (at least 50 varieties) and selection (at least 2 varieties) of sweetpotatoes for lead tolerance.
7. Test for growth and development of sweetpotato under controlled conditions in lead contaminated soil.
8. Biochemical analysis of sweetpotatoes for enzyme activity.

Description of the main results of the study

As a result of determining the level of soil pollution with lead and other heavy metals by the Shymkent Lead Plant, high concentrations of lead (Pb), zinc (Zn) and cadmium (Cd) were detected. Soil analysis from three local residents' sites revealed exceedances of maximum permissible concentrations (MPC).

As a result of *in vitro* screening of sweetpotato plants at various concentrations of heavy metals, 32 varieties of sweetpotatoes out of 57 were taken for work. Then, to select from 32 varieties of tolerant and sensitive plants, a phenotypic assessment was carried out for leaf damage on a scale from 0 to 10. Thus, two tolerant varieties KO-12 and KO-16 and two sensitive varieties KO-7 and KO-5 to lead were selected.

High tolerance of the sweetpotato variety KO-12 compared to rapeseed and sunflower when exposed to high concentrations of heavy metals was confirmed. Phenotypic evaluation as well as physiological and biochemical analysis confirmed the tolerance of KO-12 and KO-16 varieties to heavy metals, while KO-7 and KO-5 varieties were sensitive.

The difference in the gene expression spectrum of tolerant and sensitive sweetpotato varieties was determined by comparative transcriptome analysis. To understand the mechanism of tolerance to lead, zinc and cadmium at the molecular level, we investigated the expression of 4 genes: Metallothionein (IbMT1), ATPase 3 (HMA3) homolog gene, PODs (swpb3 and swpa4). From the obtained results, it was revealed that lead, zinc and cadmium treatment showed different changes in gene expression between the tolerant and sensitive variety.

Experimental cultivation of sweetpotato in areas contaminated with heavy metals was carried out. The results of atomic absorption spectroscopy analysis showed that sweetpotatoes adsorb high concentrations of lead and zinc. The bioconcentration and translocation coefficient revealed that the ability of sweetpotatoes to accumulate metals was distributed as follows: Pb>Zn>Cd. The

index for lead and zinc was >1 , which suggests that sweetpotatoes have great potential as a phytoremediant.

Sweetpotatoes were processed for starch and analyzed for heavy metal content. The results of atomic absorption spectroscopy analysis revealed low levels of heavy metals in starch that did not exceed the MAC.

Based on the results obtained, it can be confidently assumed that sweetpotatoes have good adsorption of heavy metals by roots and leaves, as well as the ability to remediate lead.

Scientific novelty. The ability of sweetpotato (*Ipomoea batatas* L.) to remediate contaminated soils was studied for the first time in Kazakhstan, and the spectrum of gene expression in tolerant and sensitive sweetpotato varieties in response to elevated lead concentration was determined.

Theoretical and practical significance. The results of the research with sweetpotatoes will allow us to solve many biotechnological issues in the future, including environmental restoration using phytoremediation methods, increasing productivity by creating varieties with enhanced antioxidant properties, which will significantly increase their stress tolerance and reduce crop losses under unfavorable environmental conditions.

The main provisions submitted for defense:

1. The level of soil contamination with lead and other heavy metals by the Shymkent Lead Plant has been determined.

2. Experimental cultivation of sweetpotatoes was carried out in the lead-contaminated territory of southern Kazakhstan. A method for obtaining virus-free sweetpotato material was developed.

3. Optimized solid nutrient medium for sweetpotato cultivation.

4. Optimized nutrient medium for cloning and transplanting sweetpotato plants *in vitro*.

5. The maximum permissible concentrations of heavy metals in the nutrient medium have been determined.

6. *In vitro* screening (at least 50 varieties) and selection (at least 2 varieties) of sweetpotatoes for tolerance to lead were carried out.

7. Test for growth and development of sweetpotato under controlled conditions in lead contaminated soil was carried out.

8. Biochemical analysis of sweetpotatoes for enzyme activity was carried out.

Degree of reliability and validation of research results. The main studies were carried out in accordance with the thematic plans of research and development, the results of which are annually reported and presented at the meetings of the Academic Council of the RSE on the Right of Economic Management "Institute of Plant Biology and Biotechnology" of the SC MNVO RK (IPB), in the materials of the international scientific and practical conference "Ecological Genetics and Population Health: Achievements and Prospects" dedicated to the 80th anniversary of Doctor of Biological Sciences, Professor, Academician of the National Academy of Sciences of the Higher School of the Republic of Kazakhstan and IAIN Bigaliyev Aitkhazhi Bigaliyevich (Almaty, January 18, 2023), the International Scientific Forum "Omarov Readings: Biology and Biotechnology of the XXI Century"

(Almaty, April 14, 2023), the International Conference "International Conference on Plant Biology and Biotechnology (ICPBB 2024)" (Almaty, June 3-6, 2024). The reliability and validity of the research results are ensured by the clarity of the methodological positions applied in the experimental work, corresponding to the object, subject, goals and objectives of the dissertation.

The results of this dissertation will improve the level of research work in the field of creating heavy metal accumulator plants. Knowledge in the field of ecology, plant biotechnology and molecular biology will increase significantly due to the acquisition of new theoretical and practical knowledge. In addition, the results of the study will lay the foundation for phytoremediation of locally contaminated heavy metals areas of Kazakhstan, which will have a positive impact on improving the environmental situation and public health.

Relationship with other research works. This dissertation work was completed within the framework of the GF project AP09259945 «Potential of sweetpotatoes (*Ipomoea batatas* L.) for phytoremediation of plumbum in contaminated areas of southern Kazakhstan», 2021-2023.

Publication of dissertation results. The main results of the dissertation were published in 11 scientific articles in journals and conference proceedings, including 5 articles in the journal included in the Scopus database with a percentile of 59% (Q2), 69% (Q2), 79% (Q1), 86% (Q1) and 87% (Q1), 2 scientific articles in journals recommended by the Committee for Quality Assurance in Science and Higher Education of the Ministry of Science and Higher Education of the Republic of Kazakhstan, 4 publications in the proceedings of an international scientific conference.

Personal contribution of the author. The author personally formulated the purpose and objectives of the research, conducted field studies and laboratory analyses to select tolerant and sensitive varieties of sweetpotatoes to lead, and determined the difference in the spectrum of gene expression in tolerant and sensitive varieties of sweetpotatoes using comparative transcriptome analysis, processed the obtained data and interpreted them, and performed statistical processing of the results.

Structure of the dissertation. The total volume of the dissertation is 115 pages. The list of references includes 361 titles, the dissertation contains 29 figures, and 12 tables.