ABSTRACT

of the PhD dissertation titled: "Study of the current state and genetic diversity of rare species *Juniperus seravschanica* Kom. populations in Kazakhstan" by Yermagambetova Moldir submitted for the degree of Doctor of Philosophy (PhD) on the educational program "8D05108 - Geobotany"

The relevance of research. In recent decades, in the context of global climate change and increasing anthropogenic impact on the natural ecosystem, preserving biodiversity and studying plants' genetic diversity have become highly relevant topics. Juniper (*Juniperus* L.), common in the mountainous regions of Central Asia, including Kazakhstan, plays an important role in the formation of landscapes and ecosystems. That is why one of the most pressing issues is the assessment and conservation of endemic, rare and endangered species in each region of the country at the population level.

Kazakhstan is the ninth largest country in the world by the territory and it has a unique and inimitable flora. The high mountain ranges of Altai, Northern, and Western Tien Shan surround the eastern, southeastern, and southern regions of the country. Juniper is one of the important components of pine forests in the above-mentioned mountain ranges, as well as in Northern Kazakhstan. There are about 75 species of this genus in the world, and according to various sources, from 7 to 10 *Juniperus* species grow in Kazakhstan - *Juniperus communis, J. media, J. pseudosabina, J. sabina, J. semiglobosa, J. sibirica, J. turkestanica, J. talassica, J. davurica* and *J. seravschanica*.

The object of the study is a rare species, *J. seravschanica* (zeravshan juniper), which is endemic in Central Asian endemic, listed in the Red Book of Kazakhstan (category III) due to a decreasing of its population. The study of the current state and genetic diversity of the zeravshan juniper is important because it plays an important ecological role in the formation of shrub-forest areas in the mountainous regions of the country, as well as in protection against floods and fixing the soils of mountain ranges. The leaves and cones of zeravshan juniper contain a lot of essential oils, which are sources of raw materials for pharmaceuticals. In this regard, it is an urgent task to conduct comprehensive studies on the current state of the species and its populations, its morphological and genetic diversity, and phytochemical characteristics.

Objects of study: Populations of the species *Juniperus seravschanica* Kom. (zeravshan juniper).

Subject of study. Morphological and anatomical description, floristic composition of plant communities with the participation of *J. seravschanica*, phytochemical composition of cones and leaves, molecular genetic analysis of *J. seravschanica* populations, and molecular taxonomy of the *Juniperus* genus.

Goal of the research. A comprehensive study of the current state of populations of the rare Red Book species *Juniperus seravschanica* Kom. using botanical and molecular genetic methods.

Research tasks.

1. To analyze herbarium material of species of the genus *Juniperus* in domestic and foreign herbarium funds and to collect plant material of *J. seravschanica* populations;

2. Description of *J. seravschanica* populations' characteristics;

3. Assessment of morphological and anatomical features of the *J. seravschanica* population;

4. To study the floristic composition of plant communities that include rare species *J. seravschanica*;

5. Determination of the phytochemical composition of leaves and cones of *Juniperus* species from Kazakhstan;

6. Phylogenetic analysis of juniper species using DNA barcoding technology and nucleotide sequences of chloroplast genomes;

7. Assessment of genetic diversity and population structure analysis of *J. seravschanica* populations using microsatellite DNA markers.

Research methods. Modern botanical, phytochemical, molecular genetics and statistical methods were used in the study. In 2023, the PhD student completed a scientific internship on statistical research methods at the John Innes Center, Norwich, UK.

Scientific novelty. For the first time, comprehensive studies of natural populations of the rare species *J. seravschanica* included in the Red Book have been carried out using botany, phytochemistry, and population genetics methods, including next-generation sequencing (NGS) technology. The floristic composition of plant communities with the participation of *J. seravschanica*, the number and density of populations of *J. seravschanica*, morphological features of populations, anatomical features of leaves, phytochemical composition of juniper cones and leaves were determined. For the first time, a phylogenetic analysis of Kazakhstani juniper species was carried out using nucleotide sequences of chloroplast genomes and a nuclear genome marker (ITS). For the first time, a comparative analysis of genetic diversity using highly polymorphic microsatellite DNA markers was carried out and the population structure of natural populations of *J. seravschanica* collected in Central Asia was evaluated.

Theoretical and practical significance.

The current state of populations of *J. seravschanica* has been studied: places of growth have been established, current distribution areas have been surveyed, and the floristic composition of plant communities with the participation of *J. seravschanica* has been determined.

For the first time, herbarium specimens of species of the genus *Juniperus* from the main repositories and herbarium collections, including the herbarium collection of the Institute of Botany and Phytointroduction (AA) of the Ministry of Ecology and Natural Resources of the Republic of Kazakhstan were analyzed.

Two herbarium sheets belonging to populations of *J. seravschanica* were documented and transferred to the herbarium fund of the Institute of Botany and Phytointroduction (AA) and herbarium sheets of the *J. seravschanica* and *J. sibirica* species to the herbarium collection of the Faculty of Biology and Biotechnology, Department of Biodiversity and Bioresources of the al-Farabi Kazakh National University.

The morphoanatomical and phytochemical features of the leaves and cones of juniper species were studied. The highest content of myrcene (19.7 % and 19.5 %) and

cedrol (13.1 % and 16.8 %) compared to other studied juniper species (*J. sabina*, *J. pseudosabina*, *J. turkestanica* and *J. sibirica*) was found in the leaves and cones of *J. seravschanica*.

ITS sequences of Kazakhstan's populations of *J. seravschanica* were entered into the international database of the National Center for Biotechnology Information (NCBI) under registration numbers OK325616, OK325617, OK325618 and OK325619. Also, complete nucleotide sequences of chloroplast genomes of 7 juniper species with identification numbers OL684343 (*J. seravschanica*), OQ644240 (*J. communis*), OQ644239 (*J. sibirica*), OQ644236 (*J. davurica*), OQ644238 (*J. pseudosabina*), OQ644237 (*J. semiglobosa*) and OL467323 (*J. sabina*) have been deposited in the international NCBI database. Botanical and genetic passports of junipers are included in the database "Biodiversity of the Flora of Kazakhstan (kazflora.kz). The results make a major contribution to the study of the molecular taxonomy of the genus *Juniperus*. Results of studies of chloroplast genomes with next-generation sequencing technology are a valuable source for molecular identification of plants and assessment of phylogenetic relationships at the genus level and beyond.

The main provisions of the dissertation submitted for defense:

1. The size and weight of plant cones in populations of zeravshan juniper growing in Kazakhstan correlates with the growth condition "altitude above sea level";

2. Individuals with the average generative age state represented 41.2 % of all plants in the studied populations of *J. seravschanica*;

3. The floristic composition of plant communities with the participation of *J. seravschanica*, growing in Kazakhstan, is represented by 254 species, 175 genera from 51 families;

4. Leaves and cones of *J. seravschanica* are characterized by the highest content of myrcene (19.7 % and 19.5 %) and cedrol (13.1 % and 16.8 %) compared to the other 4 studied juniper species (*J. sabina*, *J. pseudosabina*, *J. turkestanica* and *J. sibirica*);

5. Nuclear (*ITS*) and chloroplast (*accD*, *clpP-infA-matK*, *ycf1*, *ycf2* and *ycf3*) genome markers are informative tools for studying phylogenetic relationships and molecular taxonomy of the *Juniperus* genus.

6. Populations 4 (Uzbekistan), 6 (Kyrgyzstan), and 12 (Kazakhstan) of *J. seravschanica* are characterized by relatively high genetic diversity according to SSR analysis.

Relations with the plan of main scientific works. The dissertation work was carried out in the laboratory of molecular genetics at the "Institute of Plant Biology and Biotechnology" within the project AP09259027 "Study of the genetic diversity of species of the genus *Juniperus* L. growing in Kazakhstan" (2021-2023) and AP05131621 "Information system on molecular genetic and botanical documentation of wild flora of Kazakhstan" (2018-2020) funded by the Science Committee of the Ministry of Science and Higher Education of the Republic of Kazakhstan.

Personal contribution of a PhD student. The author of the dissertation made a personal contribution to the collection of literary review data on the research topic, conducting theoretical and experimental research, field trips for the collection of plant material, processing the results, as well as preparing articles for publication, and writing

and preparing the dissertation manuscript.

Approbation of research work. The results and main provisions of the dissertation were published and presented at the international scientific and practical conferences "The 5th Symposium on EuroAsian Biodiversity (SEAB-2021)" (in a hybrid format, Mugla, Turkey and Almaty, Kazakhstan, 2021), in the materials of the International Scientific-practical conference "Independence of Kazakhstan: Aspects of Biodiversity Conservation" dedicated to the 80th anniversary of Doctor of Biological Sciences, Professor N.M. Mukhitdinov (Almaty, Kazakhstan, 2021) and International Conference on Veterinary, Agriculture and Life Sciences (ICVALS) (Antalya, Turkey, 2022-2023). The main results of the dissertation were reported annually at meetings of the scientific and technical council of al-Farabi KazNU, Faculty of Biology and Biotechnology, Department of Biodiversity and Bioresources, scientific seminars of the Institute of Plant Biology and Biotechnology (IPBB), Laboratory of Molecular Genetics. The results of the dissertation were included in the annual reports of the project AP09259027 (2021-2023).

Publications. The main content of the dissertation was reported in 13 scientific publications including: 5 articles in peer-reviewed international journals included in Web of Science and Scopus databases, 2 articles in republican scientific journals recommended by the Committee for Quality Assurance in the Field of Science and Higher Education of the Ministry of Science and Higher Education of the Republic of Kazakhstan, 6 theses in collections of materials of international scientific conferences.

Structure of the dissertation. The dissertation is presented on 155 pages and consists of definitions, designations and abbreviations, introduction, literature review, research materials and methods, results and discussion, conclusion and the list of literature sources including 273 titles; the dissertation contains 28 tables, 31 figures and 6 supplementary files.