ANNOTATION

of the dissertation submitted for the degree of Doctor of Philosophy (PhD)
under the educational program 8D05108 – Geobotany

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"Current state of *Humulus lupulus* L. populations in the Almaty region and prospects of their use"

General characteristics of the study. The present dissertation is devoted to a comprehensive investigation of the current state of natural populations of *Humulus lupulus* L. growing in the Almaty and Zhetysu regions of the Republic of Kazakhstan, taking into account their ecological-geographical, coenotic, morphological, phytochemical, and molecular-genetic characteristics.

Relevance of the research topic.

Currently, there is a global trend toward expanding the use of natural medicinal products and biologically active compounds of plant origin in medicine. A significant proportion of pharmacological raw materials continues to be represented by wild medicinal plants, whose reserves are formed exclusively within natural ecosystems. At the same time, the demand for medicinal plant raw materials is not only maintained but is steadily increasing, whereas existing technologies for their harvesting, reproduction, and rational utilization under natural conditions remain insufficiently developed in many cases.

Plant-derived preparations are known to exert a milder, more complex, and physiologically balanced effect on the human body compared to synthetic analogues, which determines their widespread use in the treatment of chronic diseases. *Humulus lupulus* L. (common hop) is among the valuable medicinal plants characterized by pronounced pharmacological activity. Preparations based on this species are used as analgesic, sedative, and hypnotic agents, as well as for the treatment of increased nervous excitability, sleep disorders, neuralgia, vegetative-vascular dystonia, functional disorders of the cardiovascular system, tachycardia, and the early stages of hypertension.

In addition to its medicinal importance, *Humulus lupulus* L. is widely used in the brewing industry, food production, and biotechnology due to its rich content of biologically active compounds, including flavonoids, phenolic substances, essential oils, and prenylated chalcones. These compounds exhibit antioxidant, antimicrobial, anti-inflammatory, and other pharmacologically valuable properties, which significantly increase the scientific and practical interest in this species.

According to the classification of medicinal plant resources, *Humulus lupulus* L. is characterized by a wide distribution range; however, it possesses limited natural raw material reserves because it does not form extensive and dense stands under natural conditions. Despite its economic and medicinal value, information on the current status, population structure, ecological characteristics, and genetic diversity of natural populations of *Humulus lupulus* L. in Kazakhstan remains insufficient.

In recent years, studies of natural plant populations have increasingly employed an integrated approach combining ontomorphological, population-ecological, ecological-coenotic, and spatial research methods. Such an approach makes it possible to objectively assess the current status of a species within plant communities, identify patterns of its spatial organization, and predict population dynamics under changing environmental conditions. Furthermore, modern phytochemical and molecular genetic methods provide valuable information on the biological activity, genetic identity, and adaptive potential of plant populations.

In this regard, a comprehensive study of natural populations of *Humulus lupulus* L. in the Almaty and Zhetysu regions of the Republic of Kazakhstan, including analyses of their ecological

and coenotic characteristics, population structure, resource potential, phytochemical composition, antimicrobial activity, and molecular genetic features, represents an important scientific task of considerable theoretical and practical significance. An integrated assessment of the species contributes to the conservation of biodiversity, sustainable utilization of medicinal plant resources, and expansion of scientific knowledge concerning the ecology and biology of *Humulus lupulus* L. under the environmental conditions of southeastern Kazakhstan.

Therefore, a comprehensive investigation of natural populations of *Humulus lupulus* L. is highly relevant from ecological, resource-management, pharmacological, and conservation perspectives.

Objective of the study

The objective of the study was to conduct a comprehensive assessment of the current status of natural populations of *Humulus lupulus* L. in the Almaty and Zhetysu regions based on the analysis of ecological and coenotic habitat conditions, structural organization of coenopopulations, phytochemical characteristics of plant raw materials, and molecular genetic features of the species.

Research objectives:

1. To characterize the habitats and distribution range of *Humulus lupulus* L. populations.
2. To investigate the floristic composition of *Humulus lupulus* L. populations.
3. To study the structural characteristics of *Humulus lupulus* L. coenopopulations (abundance and density).
4. To conduct a phytochemical investigation of *Humulus lupulus* L.
5. To evaluate the application of molecular genetic methods in studying the genetic structure of *Humulus lupulus* L. populations.
6. To provide a comprehensive assessment of the state of the investigated natural populations of *Humulus lupulus* L.

Object of the study

The object of the study was natural populations of *Humulus lupulus* L. growing in the Almaty and Zhetysu regions of the Republic of Kazakhstan, including the territories of Almaty City, Talgar District, Enbekshikazakh District, Sarkand District, and Alakol District.

Research methods

The following research methods were employed during the dissertation study: geobotanical, soil analytical, phytochemical, microbiological, and molecular genetic methods.

Scientific novelty and significance of the dissertation

For the first time, a comprehensive ecological-geobotanical, phytochemical, antimicrobial, and molecular genetic study of natural populations of *Humulus lupulus* L. growing in the Almaty and Zhetysu regions of the Republic of Kazakhstan was carried out. The study enabled a comprehensive assessment of their current status, biological activity, ecological characteristics, and prospects for rational utilization.

For the first time, the distribution, floristic composition, coenopopulation structure, ecological features, phytochemical profile, antimicrobial activity, and molecular genetic characteristics of natural populations of *Humulus lupulus* L. in southeastern Kazakhstan were investigated in an integrated manner. New data were obtained on the population status of the species, its resource potential, and the biological activity of plant raw materials.

The theoretical significance of the research lies in the fact that the obtained results expand scientific knowledge concerning the ecological and coenotic characteristics, population organization, phytochemical composition, antimicrobial activity, and genetic structure of *Humulus lupulus* L. under the environmental conditions of southeastern Kazakhstan. The findings contribute to a better understanding of the adaptive potential, ecological stability, and biological diversity of the species within its natural habitats.

The practical significance of the study lies in the possibility of applying the obtained data to develop scientifically based recommendations for the conservation, restoration, and rational

utilization of natural hop resources, as well as in pharmacognosy, phytochemistry, biotechnology, and educational programs for training specialists in biological and medical sciences.

The obtained results may serve as a scientific basis for monitoring natural populations of *Humulus lupulus* L., assessing their resource value, developing conservation strategies, and identifying promising populations for the sustainable use of medicinal plant resources. The identified biologically active compounds and antimicrobial properties of the extracts indicate the potential application of the species in pharmaceutical, food, and biotechnological industries.

Main provisions submitted for defense:

1. Natural populations of *Humulus lupulus* L. in the Almaty and Zhetysu regions are characterized by pronounced ecological and coenotic differentiation determined by climatic and soil conditions.
2. The structure and abundance of *Humulus lupulus* L. coenopopulations vary depending on habitat conditions, which determines the stability and dynamics of the species.
3. Wild-growing hop populations of the region contain a wide range of biologically active compounds possessing antimicrobial and pharmacological potential.
4. Molecular genetic analysis using the nuclear marker *ITS* and the chloroplast markers *matK*, *rbcL*, and *psbA–trnH* confirmed the taxonomic identity of the studied samples as *Humulus lupulus* L. and enabled the assessment of their phylogenetic position.
5. The obtained ecological-geobotanical, phytochemical, and molecular genetic data can be used to develop recommendations for the conservation and sustainable use of natural populations of *Humulus lupulus* L.

The practical significance of the study lies in substantiating the possibility of using natural populations of *Humulus lupulus* L. as a promising source of medicinal plant raw materials. Populations characterized by a high content of biologically active compounds may be recommended for further application in pharmaceutical, food, and biotechnological developments. The phytochemical data obtained during the study expand the scientific basis for the utilization of *Humulus lupulus* L. as a valuable source of natural bioactive substances and may be used in the development of herbal medicines, dietary supplements, and functional products. The identified populations can serve as potential sources of plant material for cultivation, breeding programs, and resource management strategies. The established antimicrobial activity of *Humulus lupulus* L. extracts indicates the prospects for further pharmacological and toxicological investigations aimed at the development of new phytopharmaceutical products. The results also provide a basis for future studies focused on the isolation, characterization, and practical application of biologically active compounds with antimicrobial and antioxidant properties. The obtained results may be applied in the development of measures for the conservation, rational utilization, and restoration of common hop natural resources in Kazakhstan. Furthermore, the findings may be used in biodiversity conservation programs, monitoring of natural populations, and the sustainable management of medicinal plant resources. The research outcomes can also be incorporated into educational and scientific activities in the fields of geobotany, ecology, pharmacognosy, phytochemistry, and plant biotechnology.

Personal contribution of the doctoral student

The personal contribution of the doctoral student to each publication consisted of planning and conducting the full range of field, laboratory, and analytical studies related to the dissertation topic. The author carried out the collection, systematization, and analysis of primary data; conducted geobotanical, phytochemical, antimicrobial, and molecular genetic investigations; performed statistical processing and interpretation of the obtained results; prepared scientific generalizations; authored and published scientific papers; and completed the writing and formatting of the dissertation.

Research results:

1. As a result of field investigations conducted in the Almaty and Zhetysu regions, five natural populations and twelve coenopopulations of *Humulus lupulus* L. were identified and characterized. The species was found to be primarily confined to floodplain, meadow-shrub, and foothill habitats characterized by favorable soil and climatic conditions and sufficient moisture availability.
2. Floristic analysis revealed that plant communities containing *Humulus lupulus* L. comprised 99 species of vascular plants belonging to 64 genera and 31 families. The dominant families were Poaceae (21.2%) and Asteraceae (19.2%), followed by Fabaceae (8.1%) and Rosaceae (5.1%). Analysis of floristic similarity using the Jaccard index demonstrated a certain degree of similarity in species composition among individual populations.
3. Investigation of the coenopopulation structure revealed differences in plant abundance, density, and age composition. The abundance and density of five natural populations were evaluated. Population density varied from 5 to 20 individuals per m² and was determined by moisture conditions and phytocenosis structure. The highest abundance was recorded in foothill and floodplain habitats with a stable hydrological regime. Vegetative individuals predominated in all populations, ensuring clonal regeneration. The spatial structure was characterized as tussock-clonal.
4. Ecological analysis of the flora showed that mesophytic and mesoxerophytic species predominate in plant communities containing *Humulus lupulus* L., which corresponds to the association of hops with moderately moist habitats. The studied phytocenoses are characterized by differences in their ecological structure due to variations in the hydrological regime and habitat conditions.
5. Phytochemical analysis of *Humulus lupulus* L. cones performed using HPLC and GC–MS methods revealed the presence of major groups of biologically active compounds, including flavonoids, phenolic compounds, terpenoids, organic acids, and essential oil components. The quercetin content varied depending on the region of growth and amounted to 7.445 mg/L in Alakol, 4.695 mg/L in Almaty, and 4.444 mg/L in Shelek. It was established that a 50% hydroalcoholic extract of the plant exhibited antimicrobial activity against *Candida albicans* ATCC 10231 (1250 μg/mL) and *Staphylococcus aureus* ATCC 6538-P (5000 μg/mL).
6. Molecular genetic analysis of the studied *Humulus lupulus* L. samples was performed using the nuclear marker *ITS* and the chloroplast markers *matK*, *rbcL*, and *psbA–trnH*. Comparison of the obtained nucleotide sequences with reference sequences available in the GenBank (NCBI) database confirmed the taxonomic identity of the studied samples as *Humulus lupulus* L. Phylogenetic analysis made it possible to assess the position of the studied samples relative to the reference sequences of *Humulus lupulus* L. based on the nuclear and chloroplast markers used.

Publications and approbation of the research

The main results of the dissertation research have been published in 8 scientific works, including: 1 article published in a peer-reviewed scientific journal indexed in the Scopus database (Q1); 3 articles published in scientific 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; 3 conference papers published in the proceedings of international and national scientific conferences; and 1 patent of the Republic of Kazakhstan.

The principal results of the study were presented at international and national scientific and practical conferences and received positive evaluation from the scientific community.

Volume and structure of the dissertation

The dissertation consists of 177 pages and includes the following sections: abbreviations and symbols, introduction, literature review, materials and methods, results and discussion, conclusions, references, and appendices.

The reference list comprises 169 sources, including 132 publications in English. The dissertation contains 26 tables, 52 figures, and 1 appendix.