ABSTRACT

of the dissertation on the topic **"Pharmacognostic study of Karatau eryngium (Eryngium karatavicum Iljin) and obtaining a phytosubstance on its base"** for the degree of Doctor of Philosophy (PhD) in specialty 6D110400 – «Pharmacy»

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Relevance of the research topic: According to the World Health Organization (WHO), products of natural origin are important sources of various biologically active substances, since chemical substances of plant origin contain various compounds, determining their belonging to which group, studying physical and chemical properties, conducting qualitative and quantitative analysis, consideration of production methods and selection of the optimal one is one of the most pressing problems in the development of the pharmaceutical industry. It is known that in our Republic pharmacy is of great importance from the social, economic and strategic points of view of the development of science.
Implementation of the national drug policy, i.e. reducing dependence on foreign pharmaceutical products by providing the population with high-quality, effective, safe and inexpensive drugs, developing the domestic market, increasing the financial sustainability of the healthcare system, searching for new medicinal compounds, developing and introducing into practice domestic drugs drugs of natural origin is very important. In this regard, the most in-depth study of herbal medicines produced in our country has high scientific and technical significance.

The flora of the Republic of Kazakhstan is very rich in medicinal plants. More than 600 endemic plants are registered in our country. They contain a large amount of medicinal raw materials, which have not been fully studied.

In accordance with section 4 of the concept of healthcare development of the Republic of Kazakhstan until 2026 (Resolution of the Government of the Republic of Kazakhstan dated November 24, 2022 945), domestic pharmaceutical production and the national sanitary and epidemiological service for innovative medicines ensure internal needs, independence from the global pharmaceutical market and biosafety of the population.

In pursuance of the Comprehensive Plan for the Development of the Pharmaceutical Industry of the Country, aimed at 2020-2025, the issue of organizing the production of medicinal products based on medicinal plants on the territory of the Republic of Kazakhstan with different chemical compositions and broad pharmacological effects has a significant status for our state.

Pharmacognostic study of plant raw materials and obtaining a drug based on it with low toxicity and broad pharmacological action is considered an important issue.

The purpose of the research work:

Pharmacognostic study of the Karatau eryngium, preparation and standardization of a phytosubstance based on it.

Research objectives:

- Determination of morphological, anatomical and diagnostic characteristics of Eryngium karatavicum Iljin.

- Study of the chemical composition of medicinal plant materials and identification of the main groups of biologically active compounds of Eryngium karatavicum Iljin

- Standardization of raw materials Eryngium karatavicum Iljin in accordance with pharmacopoeial requirements.

- Preparation and standardization of Eryngium karatavicum Iljin extracts with the selection of optimal technologies.

- Determination of the stability of the extract from raw materials Eryngium karatavicum Iljin

- Study on the safety of extract from raw materials Eryngium karatavicum Iljin

- Determination of pharmacological activity of Eryngium karatavicum Iljin extract.

- Objects of research: the aboveground part of Eryngium karatavicum Iljin and an extract obtained from the aboveground part of the plant.

Scientific novelty of the research:

For the first time in Kazakhstan, a pharmacognostic analysis of medicinal plant raw materials Eryngium karatavicum Iljin was carried out. Macro- and microscopic analysis, commodity analysis, and phytochemical analysis were carried out. For a comparative study of the chemical composition of medicinal plant raw materials Eryngium karatavicum Iljin, thick extracts were obtained by fractional maceration percolation methods with exposure to ultrasound, their chemical composition was determined by gas chromatography using a mass spectrometric detector. As a result, the maceration method using ultrasound was chosen as the optimal technology for obtaining an extract from plant raw materials Eryngium karatavicum Iljin; during the study of its phytochemical composition, over 50 chemical compounds were identified. Antimicrobial activity was determined against test strains S. aureus, E. coli, P. aeruginosa, C. albicans, Str. Pneumonia, K. pneumonia, as well as antioxidant and cytotoxic effectiveness.

The scientific novelty of the research is confirmed by a patent under registration number No. 8783 (Method of obtaining Eryngium karatavicum Iljin extract with antimicrobial action).

The main positions of the dissertation research submitted for defense:

- Results of studying the distribution area, development of technology for procurement of raw materials, determination of pharmacognostic features and pharmaceutical-technological parameters of plant raw materials Eryngium karatavicum Iljin and its standardization.

- Results of studies on the selection of the optimal technology for obtaining an extract from raw materials Eryngium karatavicum Iljin, determining its component composition and standardization, assessing its safety and pharmacological activity.

Practical significance of the obtained results

Drafts of regulatory documents for plant raw materials Eryngium karatavicum Iljin and extracts based on it have been developed, analysis methods have been tested and implemented:

- The technology for collecting and preparing plant raw materials Eryngium karatavicum Iljin is presented. Identification of medicinal plant raw materials was confirmed by the State Institution of the Republic of Kazakhstan “Institute of Botany and Phytointroduction”. Registration certificate number No. 01-08/200

- On the practical application of the technology for collecting and harvesting plant raw materials Eryngium karatavicum Iljin, an Act of Implementation in the Syrdarya-Turkestan State Regional Natural Park was presented;

- The results of work were included in the educational process of the School of Pharmacy of NCJSC Karaganda Medical University in the section "Pharmacognosy research of plants containing phenolic compounds" of the discipline Pharmacognosy for students of educational programs 6B110103 Pharmacy and 6B07201 Technology of pharmaceutical production "

- An act of implementation of the technological process for obtaining an extract by percolation method from plant raw materials Eryngium karatavicum Iljin is presented at the Department of Pharmaceutical Technology of NCJSC Kazakh National Medical University named after S.D. Asfendiyarov

- An act of implementation of the technological process for obtaining an extract by maceration from plant raw materials Eryngium karatavicum Iljin is presented at the Department of Pharmaceutical Technology of NCJSC Kazakh National Medical University named after. S.D. Asfendiyarov.

- Completed scientific internship at KazNU named after. Al-Farabi as part of his doctoral dissertation

Personal contribution of the doctoral candidate

The doctoral candidate independently reviewed and analyzed information from domestic and foreign sources on the topic of the dissertation, and completed all experimental work on the assigned research tasks. All research results obtained are confirmed by the use of modern analysis methods and equipment in research centers and laboratories.

The reliability and validity of the research results is confirmed by the fact that the work performed is intended to solve a current problem, carried out in leading modern world-class research centers and by regulatory documents.

Approbation of the dissertation results:

The main results of the dissertation work are presented and published in the proceedings of international conferences:

To the 130th anniversary of S. Zh. Asfendiyarov "University Days 2019: Pediatrics of the 21st century. International scientific and practical conference "Modern problems and trends" (Almaty, Kazakhstan, 2019).

International scientific and practical conference dedicated to the memory of Doctor of Pharmaceutical Sciences, Professor Rakhimzhan Dilbarkhanuly, organized within the framework of "Formation and development prospects of the scientific school of pharmacy: Continuity of generations" (Almaty, Kazakhstan, 2019).

"Scientific discussion: Current issues, achievements and innovations in medicine" material of the XIV International Scientific and Practical Conference of Young Scientists and Students, dedicated to the "Year of Rural Development, Tourism and Folk Crafts (2019-2021)" (Dushanbe, Tajikistan, April 19 , 2019).

Materials of the VI International Scientific Conference "Farabi Alemi" for students and young scientists (Almaty, Kazakhstan, 2019).

Publications

The results of the dissertation research were published in 9 scientific papers, including: 1 publication in an international journal included in the Web of Science Core Collection and Scopus database, 3 publications in journals recommended by the Committee for Quality Assurance in the Sphere of Education of the Ministry of Education of the Republic of Kazakhstan, 4 publications in materials of International scientific and practical conferences, 1 patent for invention.

Conclusion

1. Plant raw material of Eryngium karatavicum Iljin was collected during the flowering phase in summer in accordance with proper requirements. The herb was dried in a well-ventilated area at room temperature 25±5°C. It was checked that the collected raw materials did not contain solid particles of soil, dirt, dust, or insects.

The raw materials were placed in kraft paper bags indicating the name of the raw material, place of procurement, collection time and net weight.

The study of the anatomical and morphological features of the raw material Eryngium karatavicum Iljin revealed the following diagnostic signs: Diagnostic signs at the microscopic level can be determined the following:

-shape of cells of the main epidermis of the leaf, presence of diacite-type stomata;

-isolateral type of leaf with multilayered epidermis with lower and upper

sides of the sheet;

-presence of calcium oxalate drusen visible from the leaf surface;

-the presence of small containers with essential oil on the cross section of the leaf and stem.

Identification of samples of plant raw materials Eryngium karatavicum Iljin was carried out according to the following parameters: macro- and microscopic features of raw materials, qualitative and quantitative composition of BAS. Qualitative and quantitative research revealed the presence of flavonoids, amino acids, alkaloids, polysaccharides, hydrolyzed and condensed tannins, essential oils (terpenes), phenolic acids, coumarins and saponins.

According to the order of the Minister of Health of the Republic of Kazakhstan No. KR DSM-20 dated February 16, 2021, quality indicators of plant raw materials Eryngium karatavicum Iljin were determined and a quality specification was developed.

According to the order dated October 28, 2020, the results obtained during a long period of research on Eryngium karatavicum Iljin raw materials allow you to set a temperature of 25 ± 2 ° C, a relative humidity of 60 ± 5%, and a shelf life of 2 years.

The technological parameters of Eryngium karatavicum Iljin plant raw materials were determined: specific, bulk and bulk weight, porosity, porosity, free volume of the raw material layer, extractant absorption coefficient, extractive substances.

Extracts of traditional and modern extraction methods were obtained on the basis of plant raw materials Eryngium karatavicum Iljin. The classical method is percolation; the modern method is fractional maceration using ultrasound. The extract obtained by maceration using ultrasound was selected as the optimal extract and the parameters of its extraction were determined.

The quality indicators of the extract based on plant raw materials Eryngium karatavicum Iljin were determined according to the order of the Minister of Health of the Republic of Kazakhstan No. KR DSM-20 dated February 16, 2021: description, identification, dry residue, weight loss during drying, heavy metals,

microbiological purity, quantification, packaging, labeling, transportation, storage, shelf life, the main pharmacological action.

The data obtained on the long-term testing of an extract based on plant raw materials Eryngium karatavicum Iljin according to the order of the Minister of Health of the Republic of Kazakhstan No. KR DSM -165/2020 dated October 28, 2020 showed that at a temperature of 25 ± 2 ° C, relative humidity 60 ± 5%, there were no significant changes in the results of determining quality indicators.

Referring to the data from the literature published so far, substances with biologically active properties belonging to various classes of natural organic compounds have been found in plants. The chemical composition of many Eryngium plants consists mainly of polyacetylene compounds, three-terpene saponins, terpenoids, phenolic compounds, tannins, vitamins and other biologically active substances.

The development of pharmaceutical production in the Republic of Kazakhstan is crucial for the development of the country's economy and the potential of the population. Due to its numerous benefits, drugs derived from medicinal plants are known to be very widely used in medicine.

2. To develop the efficiency of the process of extracting extracts from the Karatau dogwood herb (Eryngium karatavicum Iljin), the following technological parameters were studied and determined: grinding of plant raw materials - 3-5 mm; specific gravity 1.34 g/cm3; mass mass - 0.25 g/cm3; volumetric mass - 0.47 g/cm3; porosity - 0.63 g/cm3; divisibility - 0.57 g/cm3; free volume of the layer - 0.84 g/cm3; extractant absorption coefficient: water - 5.1 ml/g; 30% ethanol - 4.12 ml/g; 50% ethanol - 3.92 ml/g; 70% ethanol-3.53 ml/g; 90% ethanol-2.91 ml/g. the yield of extractives showed relatively the highest value in the extract from 90% ethyl alcohol. The data obtained will be used in the technological process for the production of extraction preparations based on the herbs of Eugene karatau.

A significant amount of microelements iron (Fe), zinc (Zn), manganese (Mn) and macroelements sodium (Na), magnesium (Mg), calcium (Ca) in the raw material of the above-ground part of the plant. The content of heavy metals meets the requirements for medicinal plant raw materials and food products.

In the studied sample of Eryngium karatavicum Iljin raw materials, there are 18 types of amino acids, 8 of which are essential: valine, leucine, isoleucine, threonine, methionine, phenylalanine, lysine, tryptophan, 8 replaceable: glycine, serine, cysteine, proline, aspartic acid, glutamic acid, Alanine, tyrosine and conditionally substituted histidine, arginine were found to be amino acids.

In the course of the research carried out on the topic of the dissertation, the content of fatty acids was determined using a gas chromatography flame ionization detector,

among which there are saturated, monounsaturated and polyunsaturated fatty acids: monounsaturated fatty acids - palmitolein (0.0028%), eruc (0.1961%), nervon (0.0154%), hamolene (0.008%), myristolein (0.0037%), polyunsaturated fatty acids - linol (0.1803%), arachidone (0.1368%), linolene (0.0078%), linoleaidin (0.046%).

Of the saturated fatty acids - fatty (butanoic) (2.35%), caproic (95.62%), capric (decane) (0.063%), myristic (0.257%), isoheptadecanic (0.085%), heptadecanic (0.016%) , stearic (1.042%) acid. Fatty acids, including unsaturated fatty acids, are involved in various biochemical processes in the human body, so it is considered important that their amount is normal.

According to the regulatory documentation, the mandatory standardization of medicinal plant raw materials identified 4 main potentially toxic elements (cadmium, mercury, lead and arsenic), as a result of which cadmium, mercury and arsenic were not detected. In addition to heavy metals, studies were carried out on the content of radionuclides, pesticides, mycotoxins, the purity of microorganisms and compliance with standards was established. Based on these indicators, a specification for the quality of plant raw materials has been compiled.

As a result of long-term stability tests of raw materials at a temperature of (25 ± 2) °C and relative humidity (60 ± 5)%, the shelf life is set at 24 months.

Pharmacognosic study of raw materials, a comprehensive phytochemical study of the plant Eryngium karatavicum Iljin allows us to identify raw materials, establish quality indicators and draw up a regulatory document (OD) for the studied raw materials.

Based on the results of studies of acute and subacute toxicity, Eryngium karatavicum Iljin was classified as toxicity class V, that is, practically non-toxic substances, in accordance with the modified classification of the Organization for Economic Assistance and Development (OECD). Eryngium karatavicum Iljin ethanol extract was found to be active against gram-positive, gram-negative bacteria and two reference yeast strains (Candida albicans and Aspergilus). MIC values were similar for all reference microorganisms tested, and these values were 10 mg/ml for yeasts and 20 mg/ml for bacteria. E. coli and S. for all bacteria tested except epidermidis, the MBC to MIC ratio is 1, which means that although the test compound shows that it has bactericidal properties, the MFC to MIC ratio was between 1 and 2, indicating on the fungicidal activity of the extract.

Assessing the completeness of the assigned tasks

While maintaining the internal unity of the dissertation , research was carried out in full on the assigned tasks to determine the distribution area of plant raw materials Eryngium karatavicum Iljin, the development of appropriate technology for the procurement of raw materials, pharmacognostic features and the results of

determining pharmaceutical-technological parameters and standardization; on choosing the optimal technology for obtaining an extract from Eryngium karatavicum Iljin, determining its component composition and standardization, safety and biological activity assessment.

Recommendations and background data for specific use of the results

Determination of the distribution area of plant raw materials Eryngium karatavicum Iljin, development of an appropriate technology for harvesting raw materials, pharmacognostic features and results of determining pharmaceutical and technological parameters and standardization; the choice of an effective technology for obtaining various types of extract from Eryngium karatavicum Iljin, the determination of its component composition and standardization, the determination of safety and antioxidant, antimicrobial and anti-inflammatory properties allow it to be recommended to domestic pharmaceutical industries as a pharmaceutical substance.

A draft of regulatory documents has been developed for the plant raw materials Eryngium karatavicum Iljin, and an extract based on it.

Assessment of the scientific level of the work performed in comparison with the best achievements in this field

The results obtained on the completed dissertation work are confirmed by 1 patent for an invention, 3 articles in journals recommended by the Committee for Quality Assurance in the Sphere of Education of the Ministry of Education of the Republic of Kazakhstan; 1 publication in an international journal included in the Scopus database and the Web of Science Core Collection. In addition, the main research results were reported at International scientific and practical conferences (Kazakhstan, Tajikistan).

The scientific and methodological level of the dissertation meets the modern requirements for this category of work.