ANNOTATION

of dissertation for the degree of Doctor of Philosophy (PhD) in the specialty 6D060600 – «Chemistry»

MUKAZHANOVA ZHAZIRA

Investigation of the chemical composition and biological activity of some species of plants of family *Scrophulariaceae*

General characteristics of the work. The dissertation work is aimed atstudying the chemical composition of plants of the genus *Verbascum orientale L*, *Verbascum densiflorum L.* and *Verbascum phoeniceum L.* of the *Scrophulariaceae* family growing in East Kazakhstan; extracting new individual compounds and studying their biological activity; developing optimal flowcharts for the isolation of biologically active compounds and establishing their structure.

Relevance of the research topic. In recent decades, interest in herbal medicine has been steadily growing. Minimum side effects and the possibility of long-term use of herbal products are some of their main advantages. Currently, about 3,000 medicinal products are used for treatment in Kazakhstan, the range of which is systematically expanding. One-third of these medicines are produced from medicinal plants and herbs. This indicates the prospects for the development of phytochemical research and makes it possible to increase the number of new medicines released in pure form.

The natural flora of Kazakhstan has more than 6000 plant species, among them exists a significant number of raw resources for biologically active substances that are used in domestic pharmaceutical production. The creation of new production facilities based on the latest technologies enable a more complete use of the unique potential of the plant resources of our republic. One of the priority tasks set by the Government of the Republic of Kazakhstan in the field of drug provision is the development of the pharmaceutical industry to meet the needs of the republic in medicines, including the production of phytopreparations based on raw materials from the domestic plants. Kazakhstan pharmaceutical enterprises produce 15% of their own medicines, of which herbal based drugs account for approximately 40%. Therefore, the creation of highly effective medicines of domestic production, the proposal of new methods for the isolation of biologically active complexes is considered an urgent task.

East Kazakhstan is characterized by a wide variety of vegetation cover. The *Scrophulariaceae* family is represented by 137 species and 20 genuses, which is approximately 4.6% of the species and 9.5% of the generic compositions in the world volume. However, not all plant species of the genus *Verbascum* have been subjected to systematic research, in this regard, the study of chemical composition, the development of methods for isolating phytopreparations and biologically active substances, the study of biological activity is particularly relevant.These will

contribute to the creation of more effective domestic phytopreparations, which makes it possible to expand the range of medical products of the Republic of Kazakhstan.

Purpose and objectives of the study. The main purpose of the research work is to develop a method for obtaining new biologically active compounds and to establish the chemical composition of plants of the genus *Verbascum orientale L.*, *Verbascum densiflorum L*. and *Verbascum phoeniceum L.* of the *Scrophulariaceae* family growing in East Kazakhstan.

In order to achieve this goal , the following tasks were carried out:

1. Investigation of the qualitative composition and quantitative content of the main groups of biologically active substances (BAS) in the studied plant species (*Verbascum orientale L.*, *Verbascum densiflorum L.* and *Verbascum phoeniceum L.*) and a comparative analysis was done;

2. Development and establishment of optimal technological parameters for the production and separation of biologically active compounds (flavonoids, phenylpropanoids, iridoids);

3. Isolation of individual compounds and establishment of their structure using by chemical and physico-chemical analysis methods;

4. Study of the biological activity of isolated phytopreparations and individual compounds for their use in medicine.

Objects of research – the aerial parts of *Verbascum orientale L.*, *Verbascum densiflorum L.* and *Verbascum phoeniceum L.*plants of the *Scrophulariaceae* family growing in East Kazakhstan were harvested in three vegetation phases (budding - in June, flowering - in July and fruiting - in August-September) in 2018-2019 from East Kazakhstan.

The main provisions submitted for protection:

- The largest amount of biologically active substances is contained in plants of genus *Verbascum orientale L*.

- The optimal parameter for the isolation of biologically active substances from the aerial part of the *Verbascum orientale L.* plant is the ratio of raw materials and solvent of 1:9 respectively.

- A plant of the genus *Verbascum orientale L.* contains luteolin 7-O-β-D-glucopyranosyl-3-O-(3-hydroxy-4-methoxy)-cinnamate, not previously described in the literature.

- Ethyl acetate, butanol extracts and compound luteolin 7-O-β-D-glucopyranosyl-3-O-(3-hydroxy-4-methoxy)-cinnamate isolated from *Verbascum orientale L.* plants have immunomodulatory activity at the level of ibuprofen.

Scientific novelty

1. Within the framework of the state program, studies of the chemical composition of three plant species were carried out: *Verbascum orientale L.*, *Verbascum densiflorum L* and *Verbascum phoeniceum L.* growing in Eastern Kazakhstan. A comparative phytochemical analysis of the studied plant samples was determined.

2. For the first time in the work, in order to isolate and separate biologically active substances, a basic block scheme and technology for obtaining polyphenolic compounds are proposed. To obtain biologically active compounds from plants and

optimize the technology, the methods of classical maceration and circulating extraction in the Soxlet apparatus were used. To isolate a phenylpropanoid complex from plants of the *Verbascum* genus with an immunomodulatory effect, an effective sorbent MCI gel CHP-20P was proposed and individual compounds were obtained using preparative recycling high-performance liquid chromatography.

3. 13 biologically active compounds were isolated from the studied samples of plants of the Verbascum genus, one of which, 7-O-β-D-glucopyranosyl-3-O-(3-hydroxy-4-methoxy) - luteolin cinnamate, is not previously described in the literature, and 87 lipophilic substances were also identified from the hexane extract of plants. The structures of the compounds have been analyzed by chemical (acid, alkaline hydrolysis) and physico-chemical methods such as 1H and 13C NMR, HMBC, HSQC, COSY, NOESY, UV, IR spectroscopy and mass spectrometry (EI-MS, ESI-MS, FAB-MS).

4.The biological activity of the developed 12 phytopreparations and 1 compound isolated from plants of the genus *Verbascum* growing in East Kazakhstan was studied. Cytotoxic, immunomodulatory, antioxidant and antibacterial activity of the obtained phytopreparations were revealed.

Practical significance of the work. New sources of biologically active substances from plants of the genus *Verbascum* have been identified and proposed.

The results of biological screening indicate that biologically active substances, as well as crude extracts obtained from *Verbascum* have cytotoxic, immunomodulatory, antioxidant and antibacterial effects, individual compounds (7-O-β-D-glucopyranosyl-3-O-(3- hydroxy-4-methoxy) cinnamate of luteolin) exhibit high immunomodulatory activity. Thus, the data obtained can be applied in the agro-industry and in the development of effective domestic medicines with a certain activity in pharmaceuticals. The results of the research are introduced into the educational process in the discipline: «Chemistry of natural compounds", «Chemistry and technology of natural compounds».

Recommendations for the specific use of the obtained results and initial data

As a result of biological screening, phytopreparations obtained from plants of the genus *Verbascum orientale L., Verbascum densiflorum L.* and *Verbascum phoeniceum L* .of the *Scrophulariaceae* family growing in East Kazakhstan have cytotoxic, immunomodulatory, antioxidant and antibacterial activities. The results can be recommended for use in bioorganic chemistry, pharmacy and agriculture. Methods of separation and isolation of flavonoid and phenylpropanoid compounds are proposed for introduction into the practice of the educational process. In addition, the results of this dissertation work can be used in lectures of the following disciplines: «Chemistry of natural compounds", «Chemistry and technology of natural compounds». The results of the research work are protected by the patent of the Ministry of Justice of the Republic of Kazakhstan for a utility model «Method for obtaining a complex with an immunomodulatory effect» (№6334, 20.04.2021, bull. №33. 2021/0358.2 from 10.04.2021). An act of introduction into the educational process in the discipline «Chemistry of natural compounds» 6B01504 –Chemistry

and 6B01507-Chemistry-Biology and «Chemistry and technology of natural compounds» for EP 7M05302 – Chemistry (№1, 26.10. 2021) has been drawn up.

Technical and economic efficiency and scientific level in comparison with the best achievements in this field

– In this dissertation work, an optimal sheme for the separation and isolation of new biologically active substances from plants of the genus *Verbascum* of the *Scrophulariaceae* family has been developed. The structure of the isolated individual compounds is characterized with modern spectral analysis methods (IR, UV, NMR: ^{1}H, ^{13}C, 2D NMR: HMBC, HMQC, COSY, NOESY, EI-MS, ESI-MS, FAB-MS).

– For one of the new compound (7-O-β-D-glucopyranosyl-3-O-(3 - hydroxy-4-methoxy) cinnamate of luteolin) not described in the literature, immunomodulatory activity was observed.

– The results of the biological screening study allow us to offer these compounds and biologically active complexes as cytotoxic, immunomodulatory, antioxidant and antibacterial agents, which could in future expand the range of new highly effective domestic drugs, as well as contribute to agriculture and the educational process.

The author's personal contribution consists in choosing the direction of research, setting goals and objectives, searching and analyzing literary data on the topic of the dissertation, harvesting the plants (raw materials), performing the theoretical and experimental part of the dissertation work, processing materials, interpreting and formatting the research results and discussing them, as well as preparing scientific articles for publication in republican and international publications.

Connection of the topic with the research plan and various government programs.

The dissertation work was carried out in accordance with the plan of research work carried out at the Department of Chemistry of the S.Amanzholov East Kazakhstan University and jointly within the framework of the Scientific and Technical Program of the Ministry of Education and Science of the Republic of Kazakhstan AP05131716 «Development of scientific foundations for the isolation of new domestic preparations from plant raw materials for medicine and agriculture» (state registration no. 0118RK00459), deadlines 2018-2020.

Degree of reliabilityand approbation of the work. The main provisions and results of the dissertation work have been tested in the following international and national conferences:

1. International scientific and practical conference: «Uvaliev readings - 2018. Trends in the development of Modern science and education» (November 2018, Ust-Kamenogorsk, Kazakhstan);

2. XLII International Scientific and Practical Conference: Results of research activities 2018: inventions, methods, innovations (December 2018, Moscow, Russia);

3. Materials of the V International Scientific-Practical Conference «Integration of the Scientific Community to the Global Challenges of Our Time» (February 12-14 2020, Tokyo, Japan);

4. VII International Scientific and Practical Conference «Europe and the Turkic World: Science, Technology and Technology » (4-6 May, 2022, Mersin, Turkey);

5. International scientific and practical online conference «Uvaliev Readings - 2020» on the topic «Topical issues of science and education development» (November 2020, Ust-Kamenogorsk, Kazakhstan);

6. Materials of the VI International Scientific-Practical Conference «Integration of the Scientific Community to the Global Challenges of Our Time» (February 10-12, 2021, Yokohama, Japan).

Publications. The main research results on the topic of the dissertation are presented in 11 published works, of which: 1 article in an international peer-reviewed journal with a non-zero impact factor, which is included in the Scopus and Web of science (Q4) databases, 3 articles in publications recommended by the Committee for Quality Assurance in Education and Science of the Ministry of Education and Science of the Republic of Kazakhstan; 1 a patent for a utility model is protected by «A method for obtaining a complex with an immunomodulatory effect»: «RSE for PCB «Sarsen Amanzholov East Kazakhstan State University» of the Ministry of Education and Science of the Republic of Kazakhstan (KZ), RSE «National Institute of Intellectual Property» of the Ministry of Justice of the Republic of Kazakhstan; 6 articles of reports at international conferences.