

IRSTI 31.23.99

¹A. Kurmanbayeva, ³Zh. Shynykul, ²Yang Ye, ¹M.A. Dyusebaeva, ^{1*}J. Jenis¹Faculty of Chemistry and Chemical Technology, Al-Farabi Kazakh National University, Almaty, Kazakhstan, *e-mail: janarjenis@mail.ru²Shanghai Institute of Materia Medica, Chinese Academy of Science, Shanghai, China³Key Laboratory of Animal Models and Human Disease Mechanisms of Chinese Academy of Sciences and Yunnan Province, Kunming Institute of Zoology, Kunming, Yunnan, China.

Chemical Constituents of *Artemisia sublessingiana*

Abstract. One of the main interests of research in the field of medicinal plants is based on the results of potentially active properties of medicinal plants, one of which is current plant – *Artemisia sublessingiana* known among the locals in Kazakhstan and other Central Asian countries as having healing properties. Definitely, detailed chemical constituents of this herb have not been explored yet. In this study, we have investigated the main bioactive contents of the whole plant *A. sublessingiana* in terms of its moisture content (10.00 %), total ash (12.42 %), and extractives (22.88 %), also have analyzed the chemical constituents. Therefore, eleven macro- and microelements were identified in the ash of *A. sublessingiana* by using the method of multi-element atomic emission spectral analysis, and the main elements were K (47.310 %), Ca (41.910 %), Fe (4.984 %), Zn (0.126 %), and Mg (0.3475 %). Moreover, 8 fatty and 20 amino acids were identified from the further chemical investigation of the medicinal plant. Among of them, linoleic acid and oleic acid were recorded as the main components of fatty acids at C_{18:2} (69.1 %) and C_{18:1} (18.3 %) respectively, while glutamate, aspartate and alanine were the major contents of the amino acids at 640 mg/100 g, 1120 mg/100 g and 890 mg/100 g respectively.

Key words: *Artemisia sublessingiana*, bioactive constituents, macro-micro elements, GC-MS.

Introduction

Artemisia sublessingiana is a flowering plant of the family Compositae (Asteraceae). Within this family, *Artemisia* is included in the tribe Anthemideae and comprises over 500 species, which are mainly widespread found in Asia, Europe and North America. Asia has the greatest concentration of *Artemisia* species, with 150 accessions for China, 174 in the ex-USSR, about 50 reported for Japan, 35 species in Iran and 81 species of the genus found in Kazakhstan [1-3]. The genus *Artemisia* is known for its medicinal and aromatic properties, and used for the production of essential oils beneficial in medicine, food and cosmetics. Based on the phytochemical and pharmacological studies, *Artemisia* is a valuable medicinal source with neuroprotective, gastroprotective, anti-oxidative, anti-inflammatory, and anti-cancer effects. Farther, extensive studies on the chemical compounds composition of *Artemisia* have led to the identification of bioactive flavonoids, coumarins, sesquiterpen lactones, oxygenated monoterpenes, and sesquiterpenes [4-6].

Humans with inadequate or monotonous nutrition, poor quality of drinking water, decreased amount of mineral substances due to bleeding, disease, alcohol and some drugs that bind or cause loss of trace elements are the main causes of the lack of the biologically necessary essentials. At the same time, humans and animals can only acquire the irreplaceable essential nutrients (for example: niacin or choline) by feeding on plants or plant-based products. [7]. **Minerals, amino acids, fatty acids and vitamins** are the essential nutrients for humans.

The importance of obtaining “mineral salts” with food lies in the fact that these elements are part of the enzymes and other substances necessary for the body – participants in biochemical reactions. Consequently, in order to maintain optimal health, appropriate levels of consumption of certain chemical elements are required. Minerals are extensively divided into major minerals (macro-minerals) and trace minerals (micro-minerals). Major minerals include calcium (Ca), magnesium (Mg), potassium (K), and iron (Fe) among others [8]. As a result of the deficiency of essential amino acids in the human

body, synthesis of proteins is violated, which leads to weakening of memory and mental abilities, and a decrease in immunity (resistance of the organism to diseases) [9]. Essential fatty acids are important for the cardiovascular system: they inhibit the development of atherosclerosis, improve blood circulation, and have cardio protective and antiarrhythmic action. Polyunsaturated fatty acids reduce inflammatory processes in the body and improve the nutrition of tissues. The daily human need is estimated at 5-10 grams [10-11]. All essential amino acids, fatty acids and minerals can be found in plants and plant-based foods. A suitable combination of vegetarian or vegan products provides a person with a sufficient amount of essential nutrients [12-15].

Herbal application of species *Artemisia* as a treatment tool have been known around from immemorial time with many of them being tested through several generations because of widely distributed genus of the plant family *Asteraceae*. Nowadays, *Artemisia* species are an estimable source of sesquiterpenes, essential oils and other pharmacological active compounds which have been reported to carry various biological activities including, anti-tumor, anti-oxidant, antibacterial, anti-viral, anti-fungal, anti-cancer and to possibly solve some serious disease [16-17]. In addition, modern research in various fields of science, beside medicine, find a unique application of species *Artemisia* as a depurative, disinfectant, insecticidal anti-feedant, digestive and counteracting agent to insect poison [18-20]. However, most species of *Artemisia* have not been fully explored.

In current study has been made the investigation of chemical constituents, in intent to determine the amount of essential substances as a natural source from Kazakh medicinal plant of *A. sublessingiana* grown in Almaty region of Kazakhstan for the first time.

Materials and Methods

A. sublessingiana plants were collected in September 2017 at Kapchagay Lake of Almaty region, Kazakhstan and identified by Dr. Alibek Ydyrys. Voucher specimens (9358-S) were deposited in the Herbarium of Laboratory Plant Biomorphology, Faculty of Biology and Biotechnology, Al-Farabi Kazakh National University, Kazakhstan.

The quantitative and qualitative analysis of the biologically active constituents of the plant were carried out according to the methods reported in the State Pharmacopeia XI edition techniques.

The elemental constituents of the ash of *A. sublessingiana* were analyzed using multi-element atomic emission spectral analysis method in Shimadzu 6200 series spectrometer. These experiments were carried out in the "Center of Physico-Chemistry methods of research and analysis" of the Republican State Enterprise Kazakh National Al-Farabi University.

To determine the amino acid composition, renow of the raw material was made using GC/MS device. GC-MS analysis involves the test of the aerial part of *A. sublessingiana* by Gas Chromatograph coupled with Mass Spectrometer using polar mixture of 0.31% carbowax 20 m, 0.28% silar 5CP and 0.06% lexan in chromosorb WA-W-120-140 mesh column (400x3mm). The column temperature was programmed from 110 °C (held for 20min) at 6°C/min from 110 °C to 180 °C at 32 °C /min from 185 °C to 290 °C. When it reached 250 °C, it was left to stay constant till the end of the output of all amino acids.

To determine the fatty acid composition of the raw material, dried plant extract of *A. sublessingiana* was extracted with a chloroform-methanol mixture (2: 1) for 5 minutes, the extract was filtered through a paper filter and concentrated to dryness. Then, to taken extract add 10 ml of methanol and 2-3 drops of acetyl chloride and undergo further methylation at 60-70° C in a special system for 30 minutes. The methanol was removed by rotary evaporation and the samples were extracted with 5 ml of hexane and analyzed for 1 hour using a gas chromatograph «CARLO-ERBA-420» allocated in the Kazakh Academy of Nutrition. As a result, chromatograms of methyl esters of the fatty acids were obtained. 8 fatty acids were identified, by comparison with authentic samples by the time of exit from column.

Results and discussion

The moisture content (10.00 %), total ash (12.42 %), extractives (22.88 %) and the quantitative-qualitative contents of the biologically active constituents of the aerial parts of *A. sublessingiana* were determined (Table 1). Ash content makes it possible to judge the content of organic and mineral substances in the sample qualitatively. As a rule, the lower the content of organic substances, the higher the ash content. The content of the ash substances reveals the dependence on the properties of the test sample to certain organs and tissues of plants. Thus, the proportion of the ash component is high in tissues rich in catalytically active proteins using activator ions and ions in the cytosol composition. Mois-

ture content is mostly water, with its participation, the dissolved nutrients enter the plant through the roots and move from one cell to another, in the aqueous medium, electrolytic dissociation of these compounds occurs and the assimilation of ions containing the necessary elements of mineral nutrition by plants.

The percentage of extractives gives an idea about the content of metabolic substances in the organic extract of the plant; this is the total amount of phenolic compounds, terpenes, and polysaccharides, together with coumarins, sesquiterpen lactones, oxygenated monoterpenes, and sesquiterpenes.

Table 1 – Quantitative analysis of the main bioactive contents of *A. sublessingiana*

Contents, %					
Moisture content	Ash	Extractives	Organic acids	Flavonoids	Duplicates
10.00	12.42	22.88	0.446	0.057	0.60

In the present study, eleven macro-, micro-elements such as K (47.310 %), Ca (41.910 %), Fe (4.984 %), Zn (0.126%), Mg (0.347 %), Cu (0.081 %), Cd (0.010 %), Pb (0.060 %), Ni (0.00720 %), Mn (0.347 %), and Na (4.890) were determined in the ash of *A. sublessingiana* (Table 2). Trace elements are necessary for living organisms to ensure normal life activity. With a lack of potassium, there are disruptions in the work of the heart and skeletal musculature. Prolonged potassium deficiency can cause acute neuralgia. Prolonged deficiency of calcium and vitamin D in the diet leads to an increased risk of osteoporosis, and in infancy causes rickets. In living organisms, iron is an important microelement that catalyzes the processes of oxygen exchange (respiration). The main

intracellular depot of iron is the globular protein complex – ferritin. Lack of iron is manifested as a disease of the body: chlorosis in plants and anemia in animals. Zinc is essential for the production of sperm and male hormones, is essential for the metabolism of vitamin E, important for the normal functioning of the prostate, involved in the synthesis of various anabolic hormones in the body including insulin, testosterone and growth hormone, and necessary for the breakdown of alcohol in the body, alcohol dehydrogenase. **Magnesium is necessary** in maintaining the normal function of the nervous system and heart muscle, has a vasodilation effect, stimulates bile secretion, and increases the motor activity of the intestines, which helps to eliminate cholesterol from the body.

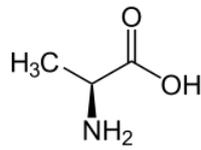
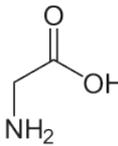
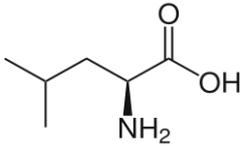
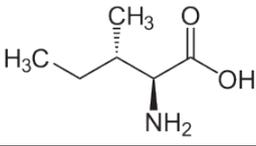
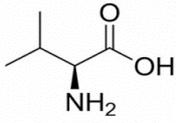
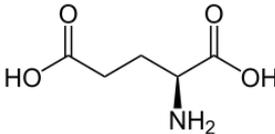
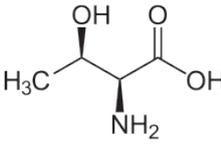
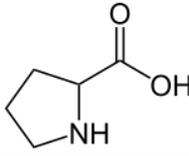
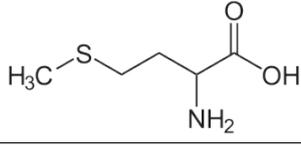
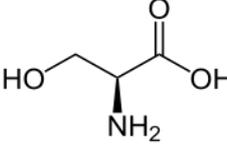
Table 2 – Composition of macro-micro elements from the ash of *A. sublessingiana*

Element	Cu	Zn	Cd	Pb	Fe	Ni	Mn	K	Na	Mg	Ca
%	0.0810	0.126	0.010	0.060	4.984	0.0072	0.347	47.31	4.89	41.91	10.82

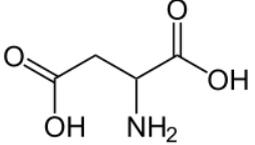
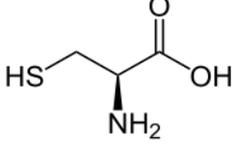
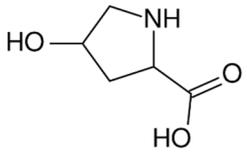
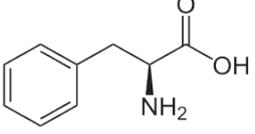
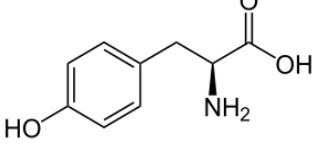
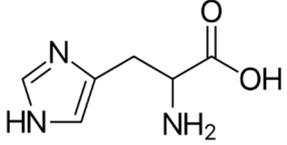
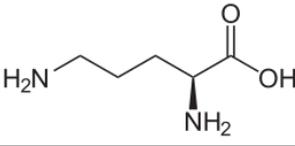
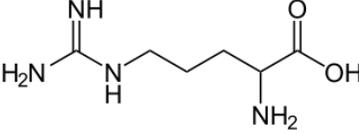
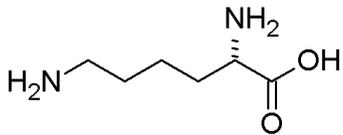
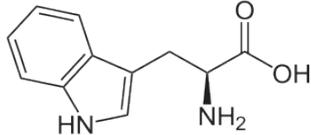
In the composition of amino acids mainly was glutamate (2640 mg/100g), aspartate (1120 mg/100g) and alanine (890 mg/100g) (Table 3). Glutamic acid is used as a chiral building block in organic synthesis, in particular, the dehydration of glutamic acid leads to its lactam-pyroglutamic acid (5-oxoproline), which is a key precursor in the synthesis of unnatural amino acids, heterocyclic compounds, biologically active compounds and so forth. Aspartate itself and

its salts are used as components of medicines. For example, the drug containing aspartate of potassium and magnesium, is used in the therapy of cardiovascular disorders. α -Alanine is part of many proteins, β -alanine is part of a number of biologically active compounds. Alanine is easily converted into the liver in glucose. This process is called the glucose-alanine cycle and is one of the main ways of gluconeogenesis in the liver.

Table 3 – Amino acid contents of *A. sublessingiana*

№	Amino acids	Molecular formula	Structure	MW	Amount in plant, mg/100g
1	Alanine	$C_3H_7NO_2$		89	890
2	Glycine	$C_2H_5NO_2$		75	452
3	Leucine	$C_6H_{13}NO_2$		131	420
4	Isoleucine	$C_6H_{13}NO_2$		131	390
5	Valine	$C_5H_{11}NO_2$		117	342
6	Glutamate	$C_5H_9NO_4$		147	2640
7	Threonine	$C_4H_9NO_3$		119	320
8	Proline	$C_5H_9NO_2$		115	760
9	Methionine	$C_5H_{11}NO_2S$		149	115
10	Serine	$C_3H_7NO_3$		105	372

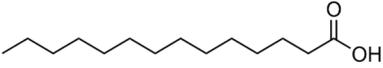
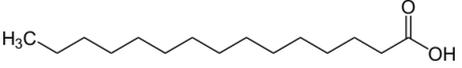
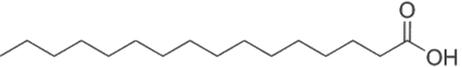
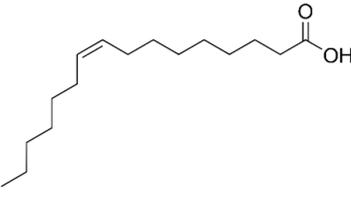
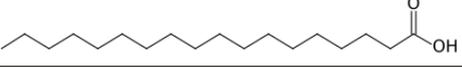
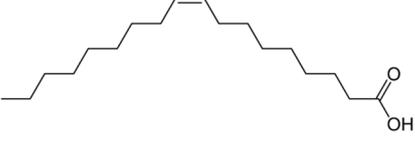
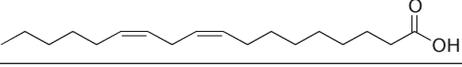
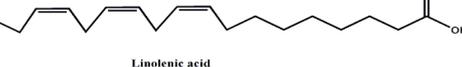
Continuation of table 3

№	Amino acids	Molecular formula	Structure	MW	Amount in plant, mg/100g
11	Aspartate	$C_4H_7NO_4$		133	1120
12	Cysteine	$C_3H_7NO_2S$		121	56
13	Oxyproline	$C_5H_9NO_3$		131	7
14	Phenylalanine	$C_9H_{11}NO_2$		165	326
15	Tyrosine	$C_9H_{11}NO_3$		181	354
16	Histidine	$C_6H_9N_3O_2$		155	270
17	Ornithine	$C_5H_{12}N_2O_2$		132	2
18	Arginine	$C_6H_{14}N_4O_2$		174	486
19	Lysine	$C_6H_{14}N_2O_2$		146	318
20	Tryptophan	$C_{11}H_{12}N_2O_2$		204	100

Results of determining the quantitative composition of the fatty acids showed that in *A. sublessingiana* from the major contents were: linoleic acid $C_{18:2}$ (69.1%) and oleic acid $C_{18:1}$ (18.3%) (Table 4). Palmitic acid is used in the production of stearin, detergents, cosmetics, lubricating oils and plasticizers; calcium palmitate is used as a component of the com-

positions for hydrophobization of tissues, skin, wood, emulsifier in cosmetic preparations, and lubrication in the production of tablets. Increased consumption of linoleic acid associated with dietary recommendations may reduce the risk of cardiovascular disease. Linoleic acid has a long-term positive effect on the prevention of type 2 diabetes mellitus.

Table 4 – Fatty acid contents of *A. sublessingiana*

№	Fatty acids	Molecular formula	Structure	MW	Amount in plant, %
1	Meristic acid $C_{14:0}$	$C_{14}H_{28}O_2$		228	0.7
2	Pentadecanoic acid $C_{15:0}$	$C_{15}H_{30}O_2$		242	1.2
3	Palmitic acid $C_{16:0}$	$C_{16}H_{32}O_2$		256	5,7
4	Palmitoleic acid $C_{16:1}$	$C_{16}H_{30}O_2$		254	0.3
5	Stearin acid $C_{18:0}$	$C_{18}H_{36}O_2$		284	4.2
6	Oleic acid $C_{18:1}$	$C_{18}H_{34}O_2$		282	18.3
7	Linoleic acid $C_{18:2}$	$C_{18}H_{32}O_2$		280	69.1
8	Linolenic acid $C_{18:3}$	$C_{18}H_{30}O_2$	 Linolenic acid	278	0.5

Conclusion

In conclusion, we investigated the main bioactive chemical contents of the whole plant *Artemisia sublessingiana* in terms of its moisture content (10.00 %), total ash (12.42 %), and extractives (22.88 %) as well as identified the composition of the macro- and microelements from its ash. Eleven macro- and

microelements in the ash of *A. sublessingiana* were analyzed using the method of multi-element atomic emission spectral analysis in the “Center of Physico-Chemistry methods of research and analysis”, and the main elements were K (47.310 %), Ca (41.910 %), Fe (4.984 %), Zn (0.126 %), Mg (0.347 %). Farther, 8 fatty and 20 amino acids were identified from the chemical investigation of the medicinal plant.

Moreover, Linoleic acid C_{18:2} (69.1%) and oleic acid C_{18:1} (18.3 %) were recorded as the main components of the fatty acids, while glutamate (2640 mg/100g), aspartate (1120 mg/100g) and alanine (890 mg/100g) were determined as the main amino acid components.

Acknowledgement

The work was supported by grants from Ministry of Education and Science of Kazakhstan (0118PK00458).

References

- Mukhitdinov N.M., Parshina G.N. (2002) Medical plants.
- Bora, K.S., Sharma, A. (2011) The genus *Artemisia*: A comprehensive review. *Pharm. Biol.* vol. 49, pp. 101–109.
- Mukhitdinov N.M., Mamurova A.T. (2013) Medical plants.
- Ahuja A., Yi Y.S., Kim M.Y., Cho J.Y. (2018) Ethnopharmacological properties of *Artemisia asiatica*: A comprehensive review. *Journal of Ethnopharmacology*, pp.117-128.
- Bayzakova G. L. (2016) Investigation of chemical compounds of *Artemisia Dracunculus*.
- Aitkazovich N. (2008) Research of chemical constituents of *Artemisia Leucodes* and *Artemisia Pontica*.
- Nosratpour M., Jafari S.M. (2018) Bioavailability of Minerals (Ca, Mg, Zn, K, Mn, Se) in Food Products. *Reference Module in Food Science*.
- Taghi Gharibzahedi S.M., MahdiJafar S. (2017) The importance of minerals in human nutrition: Bioavailability, food fortification, processing effects and nanoencapsulation. *Trends in Food Science & Technology*, vol. 62, pp.119-132
- Mc Dougall J. (2002) Plant Foods Have a Complete Amino Acid Composition. *Circulation*, vol. 105, iss. 25. pp. 197.
- Schagen Silke K., Vasiliki A. Zampeli, E. Makrantonaki and Christos C. Zouboulis. (2012) Discovering the link between nutrition and skin aging. *Dermatoendocrinol, iss. 4, no. 3*, pp. 298-307.
- Doni C, Sharma P, Saikia G, Longvah T. (2018) Fatty acid profile of edible oils and fats consumed in India, *Food Chemistry*, vol. 238, pp. 9-15.
- Baiseitova M.A., Aisa H., Jenis J. (2015) Chemical constituents of *Dracocephalum nutans*. *International Journal of Biology and Chemistry*, vol. 8, iss. 2, pp. 90-97.
- Abilova Zh.A., Baiseitova A.M., Jenis J. (2017) Investigation of chemical constituents of *Bergeria Crassifolia*. “News of the NAS RK, Series of Chemistry and Technology”, pp. 24-30.
- Muzichkina R.A., Abilov Zh.A., Korulkin D.Yu. (2010) Basics of Chemical Natural Compounds. Almaty.
- Burasheva G.Sh., Eskalieva B.K., Kipchakbayeva A.K. (2016). Medical plants of Kazakhstan. Almaty.
- Altunkaya A., Yildirim B., Ekici K., Terzioğlu O. (2014) Determining essential oil composition, antibacterial and antioxidant activity of water wormwood extracts. *The Journal of FOOD*, vol. 39, iss. 1, pp. 17-24.
- Shafi G., Hasan T.N., Syed N.A., Al-Hazzani A.A., Alshatwi A.A. (2012) *Artemisia absinthium* (AA) a novel potential complementary and alternative medicine for breast cancer. *Journal Molecular biology reports*, vol. 39, iss. 7, pp. 7373–7379.
- Gohari A.R., Kurepaz-Mahmoodabadi M., Saeidnia S. (2013) Volatile oil of *Artemisia santolina* decreased morphine withdrawal jumping in mice. *Journal Pharmacognosy Res.*, vol. 5, pp.118-120.
- Martinez-Diaz R.A., Ibañez-Escribano A., Burillo J., Heras L.D.L., Prado G.D. (2015) Trypanocidal, trichomonocidal and cytotoxic components of cultivated *Artemisia absinthium* Linnaeus (Asteraceae) essential oil. *Journal Mem Inst Oswaldo Cruz*, vol. 110, pp. 693-699. (in Brazil)
- Lee J.Y., Chang E.J., Kim H.J., Park J.H., Choi S.W. (2002) Antioxidative flavonoids from leaves of *Carthamus tinctorius*. *Arch Pharm Res*, vol. 25, pp. 313-319.