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PHYTOCHEMICAL INVESTIGATION OF WILD COLCHICUM SZOVITSII FISCH. GROWING IN ARMENIA

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The chemical compound of plant Colchicum Szovitsii growing in Aragatsotn province has been investigated. Seven alkaloids including colchicin, flavanoid-apigenin and tannins were discovered and isolated.

Colchicum Szovitsii – alkaloid – colchicin – shovicin – shovicidin

Հետազոտվել է Հայաստանի Արագածոտնի մարզում աձող Շովիցի շնդեղի քիմիական կազմը։ Հայտածվել և անջատվել են 7 ալկալոիդներ (այդ թվում` կոլխիցին), ապիգենին ֆլավոնոիդ և դաբաղանյութեր` տաննիդներ։

Շովիցի շնդեղ – ալկալոիդներ – կոլխիցին – շովիցին – շովիցիդին

Исследовался химический состав безвременника Шовица, произрастающего в Арагацотнской области Армении. Проявлены и выделены 7 алкалоидов, в том числе колхицин, флавоноид апигенин и дубильные вещества-танниды.

Безвременник Шовица – алкалоиды – колхицин – шовицин – шовицидин

Colchicum is found in Mediterranean countries of Europe, in South Africa, in Caucasus and elsewhere. There are about 225 colchicum species. They are member of the family Colchiceae and are included in alkaloid containing plants.

The alkaloids in colchicum are among alkaloids that contain nitrogen in the side chain, which were studied by some authors [1,4-6]. Climatic conditions have certain influence on alkaloid compound of plants [3]. Tropical plants are very rich of alkaloids. Even one of the most poisonous plants Aconitum which is found in Sweden, Central Asia and Armenia, is no longer considered to be poisonous in Sweden. The height from sea level also has influence on the accumulation of alkaloids. The plants, which grow in subalpine and alpine zones, contain more alkaloids. Soil composition, water regime, stages of plant growth and development also influence the accumulation of alkaloids. When alkaloids accumulate in the seeds, they promote seed germination and when they accumulate in the roots, they regulate metabolism. Alkaloids increase the sensitivity of cell and tissue towards the spectrum of the sun rays which promotes more effective use of the sun energy. Some alkaloids also take part in physiological processes of the plant, as breathing. Alkaloids are mostly in the form of bases and salts.

Colchicum Szovitsii, which is typical to Armenian flora, blossoms in early spring. It is found in high mountain zones of Aragatsotn province. It has some qualitative and quantitative phytochemical peculiarities depending on climatic conditions. According to Yusupov [4,5] Colchicum Szovitsii contains colchicine and its derivatives, which are cellular poisons, stop mitosis, change chromosome character and have some anti-tumor effect. Colchicine is also used agriculture polyploid in for getting forms of plants [3].

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Colchicine is greatly used in medicine for treating periodic disease [8]. "Colchicine" medicine is now registered by the Ministry of health of RA. Taking into account the unique properties and great demand for Colchicum, the aim of the work is to carry out phytochemical investigation of wild Colchicum Szovitsii.

During 2007-2008 Colchicum Szovitsii was introduced into hydroponics culture [2]. At present the plants collected from the alpine zone of Mount Aragats have been introduced into soilless cultivation. Investigations on the following issues were carried out: mineral nutrition optimization, nutrition frequency and planting density. Before being introduced into hydroponics and soil culture (control) some part of the wild colchicum was undergone to phytochemical research, which can be the basis of comparative studies of plants and biosynthesis of important secondary metabolites (alkaloids, flavanoids, tannins).

Materials and methods. The plants were collected from the subalpine zone of Mount Aragats (2000 m above sea level) during the blossoming period, in April, 2011 (fig. 1).



Fig. 1. Colchicum Szovitsii in subalpine zone of Aragats (blossoming stage, 04/26/2011)

Overground (the flower with stem) and underground (bulbs) parts of the dry air plants were separated for research. Standard tins Silufol UV-254 were used for analytic and preparative thin layer chromatography (TLC) in the following systems of solvents: chloroform-methanol 9:1 (1) and chloroform-methanol-benzol 10:3:3 (2). 5% water solution of FeCI3, ultraviolet rays (UV) were used for discovering flavanoids. The alkaloids were discovered with Drangendorf reactive, iodine steam and UV-lamp. The amount of tannins was determined by 1% gelatin (in 10%NaCl solution) [7]. The spectrums were taken with Specord UV-Vis equipment and Specol 11 for UV spectrophotometry was used for analysis.

The alkaloids isolated by standard colchicin, relevant Rf-s (2) in solvent system and UV- spectroscope. According to Yusupov's method [6] sum amount of neutral and basic alkaloids was isolated [picture 2].

Individual alkaloids were isolated from the amount of neutral and basic alkaloids and their quantity was determined by preparative TLC (2), in solvent system.

The experiments were carried out twice.

Results and Discussion. The results of research had shown (tab.1) that both over-ground and underground parts of Colchicum Szovitsii had the same chemical composition but different in quantity. Neutral alkaloids, which isolated according to the scheme of picture 2 [6] (colchicine, 3-dimethyl- β -lumicolchicin, 2-dimethylcolchicein, shovicamin) in the over-ground part of the plant exceeded their percentage content 1.3 times in bulbs. In the over-ground part the content of basic alkaloids (O-methyl-kreyzigin, shovicin, shovicidin) was more than in bulbs.

Promotion of biosynthesis of tannins was observed in bulbs which exceeded 1,3 times its content in the overground part [7].





Physicochemical indices of the above-mentioned alkaloids are presented in Tab. 2. As we can see the least polar alkaloid is O-methylcreyzegin, the Rf of which is 0.76 in (2) solvent system, and the most polar alkaloid is shovicidin the Rf of which is 0.15 in (2) solvent system.

Over-ground part	Bulbs
0.066 ± 0.002	0.052 ± 0.001
0.080 ± 0.002	0.060 ± 0.004
0.080 ± 0.005	0.053 ± 0.008
0.022 ± 0.001	0.023 ± 0.004
0.041 ± 0.001	0.040 ± 0.005
0.082 ± 0.002	0.062 ± 0.005
0.042 ± 0.002	0.050 ± 0.005
2.00 ± 0.200	2.50 ± 0.200
	$\begin{array}{c} Over-ground part \\ 0.066 \pm 0.002 \\ 0.080 \pm 0.002 \\ 0.080 \pm 0.005 \\ 0.022 \pm 0.001 \\ 0.041 \pm 0.001 \\ 0.082 \pm 0.002 \\ 0.042 \pm 0.002 \\ 2.00 \pm 0.200 \end{array}$

Table 1. Percentage content of biologically active substances isolated
from Colchicum Szovitsii *

* The percentage content of the above mentioned alkaloids and tannins are presented by dry weight of the overground and underground parts of the plants [table 1]. The experiments were repeated twice and \pm is the standard deviation of the experiments.

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Alkaloids	Indices <u>Rf*(</u> 2)solvent
	$\lambda^{**}_{max}(nm)$
Colchicin	<u>0.71</u>
	248;350
3 dimothyl B	0.60
Jumicelebiein	228:264
lumicolement	
2-dimethylcolchicein	<u>0.54</u>
	226;290
Shovicamin	0.21
	258;287
O-methyl-kreyzigin	0.76
	258 ;290
Shovicin	<u>0.35</u>
	260;290
Shovicidin	0.15
	280

 Table 2. Physico-chemical indices of alkaloids isolated from Colchicum Szovitsii

*Rf- The distance travelled by a given component divided by the distance travelled by the solvent front

** λ max-m maximum length of wave

The results of research have shown that both over-ground and underground parts of wild Colchicum Szovitsii contain the same alkaloids. The amount of colchicin in the overground part exceeds its content in the bulbs about 1.2 times. The diagram of alkaloid separation of wild Colchicum Szovitsii can be used for research of hydroponics crops. The isolated alkaloids can serve as standards for qualitative and quantitative research.

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