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DETERMINATION OF ALKALOIDS IN "COLCHIKUM KESSELRINGII" PLANT BY CHROMATOGRAPHIC METHOD

Abstract: In this study, experiments were conducted on the chromatographic determination of alkaloids in the "Colchikum Kesselringii" plant, and the results of the experiments were given in the article. Alkaloids in the "Colchikum Kesselringii" plant were determined using paper chromatography. The acetic acid solution was cooled, made alkaline with ammonia solution and extracted with chloroform. As a result, a collection of alkaloids of basic and phenolic nature was isolated.

Keywords: "Colchikum Kesselringii" plant, alkaloid, chromatography, paper chromatography, extraction.

Introduction: Alkaloids are widely distributed in the plant world. Plants containing alkaloids have been used by mankind since ancient times as a healing agent. Alkaloids have a strong physiological effect on the body, a small dose of them is therapeutic, and a large amount is toxic. Since alkaloids are valuable medicinal substances, interest in their study has increased.

In the 20th century, the discovery of chromatography and spectroscopy was the impetus for the development of alkaloid chemistry.

All types of chromatographic methods (adsorption-ion exchange) are widely used in the analysis of alkaloids contained in alkaloid-containing plants. These methods are used to determine how many and what compounds are present in the alkaloid extract of plant raw materials, to isolate some of the alkaloids and to determine their amount.

The chromatographic methods of distribution on paper and thin layer are very useful for determining how many alkaloids are present in plants and their approximate identification.

Experimental part

Determination of alkaloids in "Colchikum Kesselringii" plant using paper chromatography. 25 ml of 1% hydrochloric acid is added to 1 g of dried and crushed product in a 100 ml flask and shaken for 1 hour or heated in a boiling water bath for 5 minutes. The mixture was cooled, filtered through a funnel into a 100 mL flask, and ammonia was added until alkaline. The liquid mixture is then shaken through chloroform in a separatory funnel and the alkaloids are isolated. The separation is dripped onto the starting line of the chromatography paper using a capillary, and the chromatography paper is placed in a chamber containing n-butanol-acetic acid-water (5:1:4) system. The chromatogram is dried and sprayed using a pulverizer with a solution prepared by the modified Dragendorf reagent. As a result, an orange stain of alkaloid is formed. After drying the chromatogram, the Rf of the substance found is determined. To do this, the length of the distance traveled by the solvent is divided by the length of the system traveled.

7.2 grams of the fraction of neutral alkaloids isolated from the rhizomes of white saffron (S kesselringii Rgl) collected during the fruiting period was passed through a chromatographic column containing 40 g of aluminum oxide. Compounds were eluted in the following sequence (only fractions containing individual compounds were reported).

1. Eluate I - ether-chloroform, I:I, 40 mg of 2-demethyl - b - lumicolchicine, Rf - 0.79 (system X), (t_c=236-237°C)

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2. Eluate 2 - ether-chloroform, I:2, I:4, 0.32 g of 3-demethyl - b - lumicolchicine, Rf - 0.40 (t_c=198-200°C)

3. Eluate 3 - ether-chloroform, I:4 i chloroform, 4.03 g 3 - demethylcolchicine, Rf - 0.36 (t_c=179-182°C)

4. Eluate - chloroform, 20 mg of 3-demethyl-g-lumicolchicine, Rf - 0.31 (t_c=287-288°C)

All substances were crystallized in acetone.

Isolation of alkaloids from the flower of white saffron (S kesselringii Rgl).

1.0 kg of flowers of White savrinjon (S kesselringii Rgl) collected in mountainous areas of Surkhandarya region were extracted with 5 liters of 3% acetic acid solution. The acid extraction was continued for one day and approximately 3.6 L of extract was isolated. Then this work was carried out for four days with 4 liters (total 16 l) of 3% acetic acid solution. As a result, a total of 23 l of extract was obtained.

The medium of the obtained extract was brought to rN=1 with dilute sulfuric acid (1:1) solution (a total of 0.4 l of sulfuric acid was used). Then it was washed with ether three times, 4 liters each. The strongly acidic aqueous extract was washed four times with 4 liters of chloroform, and a pool of neutral and phenolic alkaloids was isolated (Extract A).

The acetic acid solution was cooled, made alkaline with 25% ammonia solution (up to rN=8) and extracted 5 times with 3 liters of chloroform each. A collection of alkaloids of basic and phenolic nature was isolated (extract B).

The chloroform extracts A and B were concentrated by evaporation of the solvents until 400 ml remained. Alkaloids of the phenolic-acidic fraction were separated into phenolic and acidic alkaloids for the first time. For this purpose, the alkaline and aqueous extracts obtained from extract A with chloroform were treated first with dilute sulfuric acid solution (up to rN=1), then with ammonia (up to rN=8.5) while cooling, and extracted four times with 140 ml of chloroform. In this, a fraction of phenolic alkaloids was formed. After bringing the remaining aqueous solution to an acidic environment, a collection of alkaloids with an acidic nature was isolated in the following amounts.

The discussion of the results

Table 1

Alkaloids isolated from the flowers of white saffron (S kesselringii Rgl).

Alkaloid nature	Amount of alkaloid, g	Alkaloid content, %
Alkaloids of a neutral nature	0,20	0,06
Phenolic alkaloids	0,72	0,2
Alkaloids of acidic nature	0,84	0,23
Alkali-insoluble compounds	0,08	0,02
Phenolic-based alkaloids	0,02	0,01
A collection of alkaloids	1,86	0,52

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Alkaloids extracted from plant flowers were determined by chromatographic analysis. According to the analysis results, the collection of alkaloids extracted from the flowers of the plant does not differ in quality from the collection of alkaloids extracted from its leaves. The results of chromatographic analysis are presented in the table:

Table 2

Chromatographic analysis of alkaloids isolated from the flowers of white saffron (S kesselringii Rgl).

Fraction	Alkaloids and their R_f	Chromatographic system
Alkaloids of a neutral nature	0.28 – colchicine 0.42 – β-lumicolchicine 0.96 – not determined	7
Alkaloids of phenolic nature	0.28 – 2-demethyl β-lumicolchicine 0.06 – 3-demethylcolchicine 0.42 – not determined 0.78 – not determined	7
Alkaloids of acidic nature	0.30 – 3-demethylcolchizein 0.66 – not determined	7
Alkali insoluble bases	0.16 – not determined 0.56 – not determined 0.75 – not determined	1
Phenolic alkaloids	0.67 – Lutein 0.57 – not determined 0.43 – Lutein 0.26 – isoregelinone 0.15 – isoregecholin	1 2

Conclusions: The alkaloids contained in the “Colchikum Kesselringii” plant were determined by chromatographic method. The alkaloids contained in the “Colchikum Kesselringii” plant were determined using paper chromatography. The acetic acid solution was cooled, made alkaline with ammonia solution and extracted with chloroform. As a result, a collection of alkaloids of basic and phenolic nature was isolated. It was found that the total alkaloids extracted from the flowers of the plant do not differ in quality from the total alkaloids extracted from its leaves.

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