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ABSTRACT BOOK


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CORRELATION BETWEEN HISTOCHEMICAL EXPRESS METHOD AND INSTRUMENTAL METHODS OF ANALYSIS IN PHYTOCHEMISTRY

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Macrochemistry and microchemistry of plants is designed to detect an insignificant content of substances in organs, tissues, and cells and determine their distribution and localization. The use of a microscope greatly increases the sensitivity of the reaction and makes it possible to trace the reaction where it remains hidden to the naked eye. The amount of substances involved in microscopic reactions during the study is expressed in micrograms.

According to Pellicciari C. (2010), a wide development of histochemical research will especially be possible in the field of plant biology [1]. The author says that the number of plant histochemical articles published has been relatively small since the '50s of the last century, but about 200 have been published in 2001-2010 years. The highest amount of articles about plant histochemistry has been published in 2011 according to the search in the National Library of Medicine. It is suggesting that the histochemical approach is becoming increasingly attractive for the plant chemistry.

As well as any other methods of studying the nature around us, microscopic chemistry has its advantages and disadvantages: sensitivity, rapidity of work, simplicity of the technique, locality of the reaction, the complexity of the composition of the cellular content, cell death, the reliability of the reaction, the effect of sample preparation [2,3].

The aim of our work was to study polysaccharides, flavonoids, hydroxycinnamic acids by histochemical methods and by instrumental methods (spectrophotometry, HPLC) in medicinal plant material of different plants.

The study of presence of polysaccharides in Akebia quinata leaves, Primula veris leaves and flowers, Rosa damascena buds and petals has been done under the microscope using the methylene blue reagent, phloroglucinol solution and potassium permanganate solution. For the histochemical determination of flavonoids in Agrimonia eupatoria and Akebia quinata plant material the 10% sodium hydroxide solution has been used. Hydroxycinnamic acids have been identified in plant material of Agrimonia eupatoria, Akebia quinate, and Actinidia arguta by complex reagent: Arnov's reagent (sodium molybdate and sodium nitrate), sodium hydroxide solution, hydrochloric acid solution [2,3]. The quantitative content of polysaccharides and mucilage has been studied by mucilage index and gravimetry. The study of the qualitative composition and quantitative content of flavonoids, hydroxycinnamic acids has been performed by spectrophotometry and HPLC methods.

The obtained results of quantitative determination confirm the assumptions about the presence of certain classes of biological active substances in the studied samples of plant materials during histochemical reactions. The color intensity in the places of localization of biological active substances in the tissues of plant raw materials correlates with the obtained results of quantitative content.

Literature:

[1] C. Pellicciari. European Journal of Histochemistry. 2010, 54:e51, 242-248.
[2] F. A. Badria, W. S. Aboelmaaty. Acta scientific pharmaceutical sciences. 2019, 3(7), 88-100.
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(PP-49)

DETERMINATION OF CARBOXYLIC ACIDS IN THE FRUITS OF *ACTINIDIA* Lindl. BY TLC AND HPLC METHOD

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The qualitative composition and quantitative content of aliphatic and aromatic carboxylic acids in the fruits of *Actinidia* Lindl has been studied. The ripe fruits of *A. chinensis* and *A. arguta* were selected. Fruits were collected in early September at the research areas of the M.M. Grishko National Botanical Garden of National Academy of Sciences of Ukraine in 2019.

The study of the qualitative composition and quantitative content of aliphatic and aromatic acids was performed by TLC and HPLC methods. TLC studies were performed in a solvent system of *n*-butanol-formic acid-water (10:1:2), ethyl acetate-formic acid-water (3:1:1), *n*-butanol-formic acid-water (75:15:10) and *n*-butanol – formic acid – water (4:1:5) compared to standard samples. The chromatograms were treated with 0.05% alcohol solution of bromothymol blue and 0.1% alcohol solution of sodium 2,6-dichlorophenolindophenolate.

HPLC studies were performed on a liquid chromatograph Agilent Technologies 1200. Separation was performed on a chromatographic column Zorbax SB-Aq (4.6 mm, 150 mm, 3.5 μm) (Agilent Technologies, USA). Detection was performed using a diode-matrix detector [1,2]. An aqueous solution of phosphoric acid was used to extract aliphatic carboxylic acids. Elution was performed in isocratic mode. The acetonitrile (A) and the 0.1% solution of phosphoric acid in water (B) (1:99) were used as the mobile phase [1]. Aromatic acids were extracted by methanol. The methanol (A) and the 0.1% formic acid solution in water (B) were used as the mobile phase. Elution was performed in a gradient mode: 0 min -A (25%): B (75%); 25 min - A (75%): B (25%); 27 min - A (100%): B (0%); 35 min - A (100%): B (0%) [2]. The identification and quantitative analysis were performed using standard solutions of carboxylic acids.

The presence and quantitative content of isocitric, tartaric, malic, fumaric, ascorbic acid in the fruits of *A. chinensis* has been found. An isocitric acid predominates ($1.39 \cdot 10^4$ μg/g). The tartaric, malic, ascorbic and fumaric acids have been identified in *A. arguta* fruits. A tartaric acid predominates ($1.69 \cdot 10^3$ μg/g). The sum of content of aliphatic carboxylic acids is dominated in the fruits of *A. chinensis* ($1.75 \cdot 10^4$ μg/g) compared to *A. arguta* ($2.3 \cdot 10^3$ μg/g). It confirms the taste of the fruits.

The research of aromatic carboxylic acids in the fruits of selected species of *Actinidia* revealed the presence of 5 compounds. The caffeic, syringic, *p*-coumaric, sinapic, *trans*-cinnamic acids have been identified in *A. chinensis* fruits. In the fruits of *A. arguta*. - gallic, *trans*-ferulic, sinapic, *trans*-cinnamic, salicylic acids. A quinic acid was founed in both studied raw materials: *A. chinensis* - $6.55 \cdot 10^2$ μg/g, *A. arguta* - $5.06 \cdot 10^2$ μg/g.

Literature:

[1] C. Agius, S. von Tucher, B. D. C. Poppenberger , W. Rozhon. *MethodsX*. **2018**, *5*, 537-550.
[2] B. R. Sumere. *Ultrasonics sonochemistry*. **2018**, *48*, 151-162.