

FARMATSEVTIKA TA'LIM VA TADQIQOT INSTITUTI
ФАРМАЦЕВТИЧЕСКИЙ ИНСТИТУТ ОБРАЗОВАНИЯ И
ИССЛЕДОВАНИЙ
INSTITUTE OF PHARMACEUTICAL EDUCATION AND RESEARCH

“ZAMONAVIY FARMATSEVTIKA SOHASINI RIVOJLANISHINING
DOLZARB MASALALARI VA TENDENSIYALARI” MAVZUSIDAGI
II XALQARO ILMIY-AMALIY ANJUMAN MATERIALLARI

МАТЕРИАЛЫ II МЕЖДУНАРОДНОЙ НАУЧНО-ПРАКТИЧЕСКОЙ
КОНФЕРЕНЦИИ «АКТУАЛЬНЫЕ ВОПРОСЫ И ТЕНДЕНЦИИ
РАЗВИТИЯ СОВРЕМЕННОЙ ФАРМАЦЕВТИЧЕСКОЙ ОТРАСЛИ»

ABSTRACT BOOK OF THE II INTERNATIONAL SCIENTIFIC AND
PRACTICAL CONFERENCE “CURRENT ISSUES AND TRENDS IN THE
DEVELOPMENT OF THE MODERN PHARMACEUTICAL INDUSTRY”

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Farmatsevtika ta'lim va tadqiqot instituti Ilmiy Kenglashining 2024 yil 27 - sentabr 2-sonli qarori bilan chop etishga tavsiya etildi.

Рекомендовано к печати решением №2 Ученого совета Фармацевтического института образования и исследований от 27 сентября 2024 года.

Recommended for publication by decision No. 2 of the Scientific Council of the Institute of Pharmaceutical Education and Research dated September 27, 2024

соединений. диссертация... доктора химических наук: 02.00.06. - Москва, 2007. - 211 с.: ил. РГБ ОД, 71:07-2/67

IDENTIFICATION OF VITAMIN K IN THE LEAVES OF PLANTAGO MEDIA L. USING THE TLC METHOD

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Introduction: A purposeful search for biologically active substances and an investigation of their pharmacological properties are the most important tasks of the pharmaceutical science. Extracts and individual compounds from medicinal plants are characterized by higher efficacy, have less manifestation of side effects providing them a lower toxicity.

The genus of Plantago contains a high amount of flavonoids and its derivatives, lipids, triterpenes, polysaccharides, vitamins and minerals that can be responsible for those bioactivities. The high content of biologically active compounds determines the wide spectrum of biological activity of Plantain [1]. Phylloquinone (vitamin K1) performs an important role in the body's vital functions, particularly in the coagulation cascade. Recent studies have proven the important role of phylloquinone for bones, neurological and vascular health. Vitamin K may also be beneficial for metabolic disorders (diabetes mellitus, excess weight, etc.), liver and kidney diseases, immune function and weight management [2, 3]. Therefore, many dietary supplements containing natural sources of phylloquinone have appeared recently. In this regard, the development of reliable methods for identifying this component in plant raw materials, and subsequently in dietary supplements, is becoming increasingly relevant [4]. The development of reliable methods for studying the phylloquinone content in plants of the Plantago L. family is required.

The methods used to study of content vitamin K include spectrophotometry, fluorimetry, chromatography. Most experiments employ high-performance liquid chromatography (HPLC) with various detection methods or mass spectrometry. Gas chromatography is used less frequently. However, thin-layer chromatography (TLC) is a simple technique for analyzing lipophilic vitamins and allows for the reliable identification of vitamin K.

Purpose of the research: Identification of vitamin K1 in Plantago media leaves using thin-layer chromatography and to compare of the effectiveness of detecting phylloquinone in different chromatographic systems.

Materials and methods: The study used Plantago media L. leaves collected during the flowering period in southern Ukraine. The raw material was dried to an air-dry state in a drying oven at 50°C.

The sample of the plant material was powdered to a particle size of 1 mm, and then it was transferred into a 15 ml flask, dissolved in 10 ml of hexane. The obtained solution was shaken on a mechanical shaker. The extract was filtered. The solvent was evaporated using a rotary evaporator until the volume of 2-3 ml. TLC separation was performed on 10×10 cm aluminum foil-backed TLC silica gel without pretreatment. Standard (10 μL) (Sigma Aldrich, $\geq 99\%$) and sample solutions (20 μL) were applied onto the plates as 8-mm bands by micropipette (CAMAG Muttenz, Switzerland). The plates were dried for 3 min. Formic acid-purified water-ethyl acetate (1:1:8) and isopropyl alcohol-benzene (1:10) systems were used as mobile phases. The chambers were previously saturated with mobile phase vapours for 20 min at room temperature. TLC was performed by the ascending method. The development distance was 115 mm. Chromatograms were dried at room temperature for 10 min. The plates were viewed under a UV cabinet (CAMAG Muttenz, Switzerland) in absorption-reflection mode at 254 nm. The substances were identified by comparing the color, fluorescence, size and retention factor of the spots obtained for the test sample and a solution of the standard substance.

Results and conclusions: Thin-layer chromatography as a method of identification of biologically active substances is one of the most accessible for wide application. Modern methods of thin-layer chromatography make it possible to identify compounds in complex compositions inherent in plant raw materials, dietary supplements, and multicomponent medicines by selecting chromatography conditions [4].

During our research, we identified vitamin K1 in plant material *Plantago media L.*, followed by viewing the chromatogram in UV light at a wavelength of 254 nm. Spots with green fluorescence were observed, which were identified as vitamin K1. Both used systems made it possible to detect vitamin K1 in the studied plant material, but better separation was achieved with the use of isopropyl alcohol-benzene (1:10) solvent system as the mobile phase.

The developed technique of thin-layer chromatography allows effective and reliable identification of vitamin K1 in the leaves of *Plantago media L.*

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ЭТИЛОЛМАЛЕИНИМИДНИ БРОМЛАШ РЕАКЦИЯЛАРИ

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Долзарблиги: N-(β-Гидроксиэтил)малеинимидни қўш боғи ҳисобига борадиган бромлаш реакцияси бир неча хил эритувчи шароитларида турлича бориши аниқланди ва тегишли бромсуксинимид олинди.

Жаҳонда кимё саноатида малеин кислотанинг амидлари ва имидларидан ноёб хоссали органик ва сополимер материаллар олиш бўйича мақсадли тадқиқотлар амалга оширилмоқда. Айтиш мумкинки, малеинимиднинг галогенли ҳосилаларини синтез қилишда мавжуд бўлган турли усуллардан фойдаланиш заруратлиги, баъзи ҳолларда эса олиниши қийин бўлиши, янги ҳосилалари синтези, назарий ва амалий аҳамиятини ўрганиш ва бу йўналишдаги ишларни ривожлантиришга, янада қулай янги изланишлар олиб боришга, малеинимид аналоглари олинишида арzonроқ реагентлардан фойдаланган ҳолда синтез усулларини ривожлантириш масалалари долзарблигини тақозо этади. Тўйинмаган дикарбон кислотанинг азот сақлаган ҳосилаларини олишда синтез вақтини қисқартириш, реакциянинг селективлигини ва маҳсулот унумини ошириш, амалий қўлланилиш соҳаларини кенгайтириш алоҳида аҳамият касб этади.

Натижа ва хulosалар: *Галогенлаш реакциялари;* Синтез асосида олинган малеинимид аналоги N-(β-гидроксиэтил)малеинимидни [1] кимёвий хоссаларини ўрганиш мақсадида, галогенлаш реакцияларидан қўш боғи ҳисобига борадиган бромлаш реакцияси олиб борилди. Бунинг учун бромнинг сувдаги, хлороформдаги, метанолдаги ва тетрахлорметандаги эритмаларидан фойдаланилди. Жараёнда 4 та бром эритмаларнинг ранги турли вақтларда ўзгариши кузатилди. Бромли сувда олиб борилган бромлаш реакцияларида бромли сувнинг ранги 30 дақиқадан кейин ўзгариши кузатилди. Хлороформдаги эритмада бромлаш реакцияси 140 дақиқаларда ранги ўзгарган бўлса, метанолдаги бромли эритмада олиб борилган реакция эса 48 соатдан кейин бром рангининг ўзгариши қўрилди. Бромли бирикма оқ чўкма ҳолида фильтрлаб олинди ва қуритилди:

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