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ВИЗНАЧЕННЯ ПОЛІФЕНОЛЬНИХ СПОЛУК У МІСЦЕВИХ ЛІКАРСЬКИХ РОСЛИНАХ ТА ЗНАЧЕННЯ ЇХ ДЛЯ ЗДОРОВ'Я

Пилипенко О.О., Суховірська Л.П., Оперчук Н.І.

DETERMINATION OF POLYPHENOL COMPOUNDS IN MEDICINAL PLANTS OF LOCAL HABITAT AND THEIR IMPORTANCE FOR HEALTH

[1]PYLYPENKO O.O.,
[1]SUKHOVIRSKA L.P.,
[1,2]OPERCHUK N.I.
[1]Donetsk National Medical University, Kropyvnytskyi, Ukraine
[2]State Institution «Kirovohrad Regional Center for Disease Control and Prevention, Ministry of Health of Ukraine», Kropyvnytsky, Ukraine

Polyphenolic compounds are found in many plants, most often tannins, humic acids, melanins and flavonoids. In medicine, certain medicinal plants are used, which have a high content of polyphenolic compounds [1]. Most often, it is tannins, flavonoids. Because they have astringent, anti-inflammatory, anti-edematous, antihistamine and antimicrobial action [2]. Modern problems of constant stress and emotional overload have led scientists to search for safe plant-derived adaptogens [3]. As it turned out, polyphenolic compounds have the following properties. It is known that polyphenolic compounds are very necessary for humans and animals, as they also have pronounced antioxidant properties and others. Nevertheless, living organisms cannot synthesize this type of compounds on their own. Therefore, the main source of income is plant food [4]. All the above-described properties of polyphenolic compounds explain why the interest in their research and finding in plant raw materials does not disappear [5]. Plant extracts, which have a high content of

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[1]Пилипенко О.О., [1]Суховірська Л.П., [1, 2]Оперчук Н.І.
[1]Донецький національний медичний університет, Кропивницький, УКраїна
[2]ДУ «Кіровоградський регіональний центр з контролю та профілактики хвороб МОЗ України», Кропивницький

***Мета:** дати оцінку можливості використання деяких лікарських рослин з вмістом поліфенольних сполук (танінів та галової кислоти) у лікарській справі.*
***Матеріали та методи.** Для визначення кількості поліфенольних сполук використовується метод спектрофотометрії з реактивом Фоліна-Чокальтеу. За стандартну пробу обрано розчин галової кислоти. Об'єктами дослідження були сухі екземпляри рослин: кора дуба (quercus cortex), кореневища гірчаку зміїного (bistortae rhizomata), галли листя дуба (gallae turcicae), кореневища перстачу прямостоячого (tormentillae rhizomata). З цієї сухої сировини виготовляються експериментальні зразки у вигляді водних та спиртових екстрактів.*
***Результати.** Дослідження виявили, що всі обрані рослини мають достатній вміст танінів для підтвердження їхньої лікувальної здатності. Найбільша кількість виявилася у галах листя дуба. Спиртові екстракти вилучали поліфенольні сполуки із рослинної сировини краще, ніж водні. На точність результатів впливають якість подрібнення сировини, час витримки екстрактів, концентрація екстрагенту. Рекомендовано водні витяжки використовувати як лікувальні препарати для тих, кому протипоказані спиртові екстракти.*
***Висновки.** Досліджені сухі екземпляри рослин (кора дуба (quercus cortex), кореневища гірчаку зміїного (bistortae rhizomata), гали листя дуба (gallae turcicae), кореневища перстачу прямостоячого (tormentillae rhizomata) містять поліфенольні сполуки (танін та галову кислоту) у достатній кількості для забезпечення лікарського ефекту. Показано, що екстракція поліфенольних сполук з рослинної сировини з використанням спиртових розчинів є більш ефективною, ніж водні витяжки. Рекомендовано використовувати рослинні продукти з дубильними речовинами у лікарській практиці.*

Ключові слова: поліфенольні сполуки, спектрофотометрія, метод Фоліна-Чокальтеу, екстракція, дубильні речовини.

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polyphenolic compounds, are widely used not only as biologically active additives, but also as medicinal drugs in medicine. Therefore, studies are being conducted to detect these compounds in various plants [6].

Simple, reliable, highly sensitive and accurate calculation methods are needed to quantify these substances [7]. These include colorimetric (spectrophotometric) method using Folin-Ciocalteu reagent containing phosphormolybdenum heteropolyacids [8, 9]. Phenolic compounds in an alkaline environment reduce these acids. The reduction product is molybdenum blue or heteropolysin, which is a complex blue compound. The brightness and intensity of the color depends on the amount of polyphenolic compounds in the test samples [10].

Objective. Assess the possibility of using some medicinal plants containing polyphenolic compounds (tannins and gallic acid) in medicine.

Experimental Details. This research measures the content of polyphenolic compounds, which include both tannins and gallic acid, to compare the best-selected medicinal properties in medicinal plants. These medicinal plants are the most accessible and grow in our region.

Materials and Methods. The Folin-Ciocalteu methodwas used to determine the amount of polyphenolic compounds [11, 12]. Measurements were performed using a spectrophotometer. A solution of gallic acid was taken as a standard sample. The objects of the study were dry specimens of plants: Oak bark(quercus cortex), Rhizomes of Bistorta officinalis (bistortae rhizomata), galls of oak leaves (gallae turcicae), Rhizomes of Potentilla erecta (tormentillae rhizomata). Aqueous and alcoholic extracts were made from this dry raw material. These solutions were used as experimental samples.

For the experiment, according to the method of execution, 0.2 cm^3 of the obtained aqueous extracts and alcohol extracts were taken from the studied plant samples. This volume was transferred to a 25 cm^3 volumetric flask. A 2 cm^3 Folin-Ciocalteu reagent was added to the same flask. After a few minutes, 7.5% sodium hydrocarbonate solution was added and adjusted to the mark with water. Optical density measurements were performed after 40 minutes on a spectrophotometer using a light filter with a wavelength of 745 nm, in cuvettes with an absorbing light layer thickness of 10 mm. As a reference solution, a standard sample of gallic acid was used, which was prepared in the same way as the test solution. Calculations were performed according to formula (1) to determine the percentage of total polyphenolic compounds, based on gallic acid.

$$X = \frac{D_1 \cdot C \cdot V_{total} \cdot 100 \cdot 100}{D_0 \cdot m \cdot V \cdot (100 - W)} \quad (1)$$

D_1 – optical density of the test solution; D_0 – optical density of the gallic acid comparison solution; C – concentration of the comparison solution of gallic acid, g/ml ($0.4x10^{-3}$); m – sample of raw materials, g; V_{total} – total extract volume, ml; V – volume taken for determination, ml; W – weight loss during drying of raw materials, %.

Results and Discussion. To conduct research, a number of extractants were made from the studied raw materials. Extraction was performed with 70% alcohol solution. For extraction used dried, crushed, ground in a mortar and sifted vegetable raw materials. The samples were filled with alcohol and kept for 45 minutes at a temperature of 45^0C. Then the settled liquid was drained and centrifuged for 2 minutes at a speed of 16000 rpm. Liquid separated by centrifugation was used for the studies.

Crushed and sifted raw materials were selected for the production of water extract. The appropriate amount of raw material was transferred into a flask, filled with hot water. The flask with the mixture was then heated in a water bath under reflux for 30 minutes, stirring occasionally. The cooled water extracts were filtered, mixed, and brought to the mark with water if necessary.

Prepared samples of medicinal plants were tested for polyphenolic compounds. Their presence was determined by interaction with a solution of iron (III) ammonium sulfate. The color of different intensities from blue to green indicated the presence of this type of compounds in the samples. Solutions of Galls of oak leaves and Rhizomes of Bistorta officinalis were blue, indicating a predominance of hydrolyzed tannins of polyphenolic nature. For Oak bark and Rhizomes of Potentilla erecta, the solutions with these samples gave a green color, indicating a predominant amount of condensed tannins. After identification of polyphenolic compounds, optical density was taken from all samples. The formula calculates the percentage of total polyphenolic compounds, based on gallic acid.

According to figura 1, it can be seen that the samples extracted with water have the highest content of polyphenolic compounds in Galls of oak leaves, followed by Rhizomes of Bistorta officinalis, Penultimate place in Oak bark and the lowest percentage of Rhizomes of Potentilla erecta.

Alcohol extracts of the studied samples of medicinal plants have slightly higher rates. But the percentage has the same sequence as for aqueous extracts.Three different concentrations of et-

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[1]Donetsk National Medical University, Kropyvnytskyi, Ukraine

[2]State Institution «Kirovohrad Regional Center for Disease Control and Prevention, Ministry of Health of Ukraine», Kropyvnytskyi, Ukraine

Objective. *Assess the possibility of using some medicinal plants containing polyphenolic compounds (tannins and gallic acid) in medicine.*

Materials and methods. *The Folin-Ciocalteu reagent spectrophotometry method is used to determine the amount of polyphenolic compounds. The gallic acid solution was selected as the standard sample. The objects of the study were dry specimens of plants: Oak Bark (quercus cortex), Rhizomes of Bistorta officinalis (bistortae rhizomata), Galls of Oak leaves (gallae turcicae), Rhizomes of Potentilla erecta (tormentillae rhizomata). Experimental samples in the form of aqueous and alcoholic extracts are made from this dry raw material.*

Results. *Studies have shown that all selected plants have sufficient tannin content to confirm their healing properties. The largest number was found in the galls of oak leaves. Alcohol extracts extracted polyphenolic compounds from plant raw materials better than aqueous ones, but the difference is relatively small. Therefore, aqueous extracts can be used as therapeutic drugs for those who are contraindicated alcohol extracts.*

Conclusions. *The studied dry specimens of plants (oak bark (quercus cortex), rhizomes of snake mustard (bistortae rhizomata), galls of oak leaves (gallae turcicae), rhizomes of erect cinquefoil (tormentillae rhizomata) contain polyphenolic compounds (tannin and gallic acid) in sufficient quantities to provide medicinal effect. It has been shown that the extraction of polyphenolic compounds from plant materials using alcohol solutions is more efficient than aqueous extracts. It is recommended to use herbal products with tannins in medicinal practice.*

Keywords: polyphenolic compounds, spectrophotometry, Folin–Ciocalteu method, extraction, tannins.

hanol were used. This is necessary to determine which ethyl alcohol solution best extracts polyphenolic compounds from plants.

Extracts of medicinal plants were made from 50%, 70% and 96% ethanol solutions. Then, the optical density of all samples was measured and the percentage of polyphenols was calculated according to formula 1. Detailed information on the results is given in table.

According to the obtained data, it was found that the best extraction results have 50% and 70% ethanol solutions. Their meanings are very close. However, the best extraction is 70% ethanol solution. For the convenience of further manipulations, the average result is calculated, which is also shown in table.

Therefore, the highest content of polyphenolic compounds was found in 70% alcohol solution Galls of oak leaves (gallae turcicae).

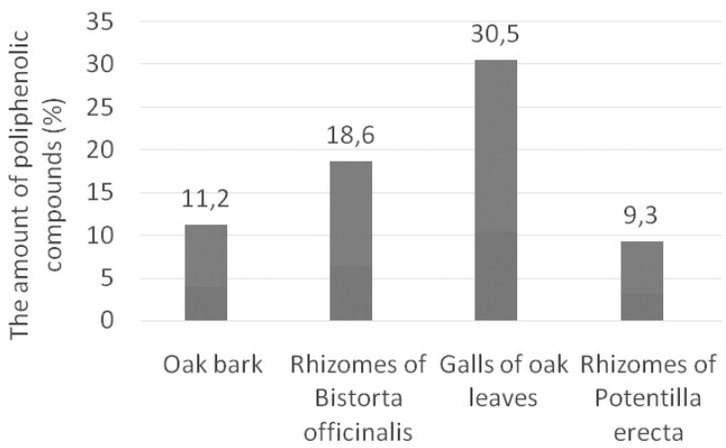
Generalized concentration values are the average value calculated for each sample. The average percentage of polyphenolic compounds in the samples is shown in Figura 2.

In order to compare the quantitative content of polyphenols in aqueous and alcoholic samples, it is appropriate to look at Figura 3, which shows in detail the difference between them.

Comparing the obtained data, it can be said that alcohol extracts extracted polyphenolic compounds from plant raw materials better than aqueous ones, but the

Figura 1

The percentage of polyphenolic compounds in the test aqueous extracts



difference is relatively small. Therefore, aqueous extracts can be used as therapeutic drugs for those who are contraindicated alcohol extracts.

Conclusion. Thus, studying four species of medicinal plants that contain polyphenolic compounds. It was found that the largest number of tannins and their derivatives have Galls of oak leaves (gallae turcicae). Aqueous solution of Galls of oak leaves (gallae turcicae) contains 30,5% of polyphenolic compounds, and alcohol extract (average value) – 37,1%. Their value is quite high compared to other studied samples. Nevertheless, all plant extracts have a sufficiently high content of polyphenolic compounds, which confirms their claimed medicinal properties. The correctness of the experiment is affected by the quality of grinding and the exact holding time of the samples for extraction. In the course of experimental studies, it was found that the concentration of the alcohol solution is also important for the completeness of the extraction of polyphenolic compounds. It was considered that 70% ethanol solution is better than 50% and 96% solutions extract tannins from plants. The lowest percentage of extraction were samples with an alcohol content of 96%. Therefore, taking into account the concentration of ethanol is a necessary condition for obtaining a solution with a high content of polyphenolic compounds. Alcohol and aqueous extracts of plants did not have a significant difference in the quantitative content of the studied compounds. Consequently, the use of both types of extracts is appropriate for application in the pharmaceutical and medical fields.

Table

The influence of alcohol extractant concentration on completeness of polyphenolic compounds extraction

C_2H_5OH (%)	Amount of polyphenolic compounds (%)			
	Oak bark (quercus cortex)	Rhizomes of Bistorta officinalis (bistortae rhizo-mata)	Galls of oak leaves (gallae turcicae)	Rhizomes of Potentilla erecta (tormentillae rhizomata)
50	14,85	19,72	36,81	11,32
70	15,27	21.94	39.38	12,68
96	13,32	18.75	55,12	9,87
Average value	14,5	20,2	37.1	11,4

Figura 2

The percentage of polyphenolic compounds in the test alcohol extracts

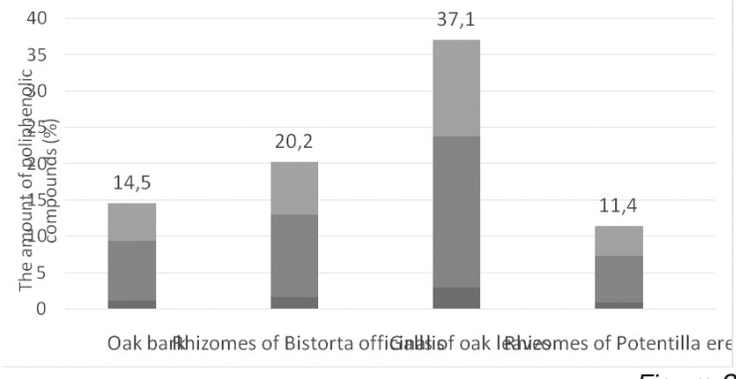
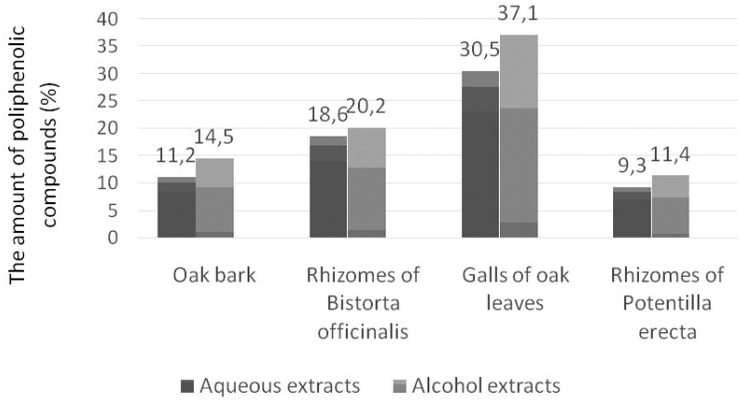


Figura 3

The influence of the nature of the extractant on the completeness of the extraction of polyphenolic compounds



ЛІТЕРАТУРА

1. Scalbert A., Williamso G. Dietary Intake and Bioavailability of Polyphenols. *J. Nutrition.* 2000. Vol. 130. P. 2073-2085.

2. Ignat I., Volf I., Popa V.I. A Critical Review of Methods for Characterization of Polyphenolic Compounds in Fruits and Vegetables. *Food Chem.* 2011. Vol. 126. P. 1821-1835.

3. Стешенко О. М., Арсеньєва Л.Ю., Ройко О.Ю., Паламарчук О.П. Дослідження фенольних сполук рослинних адаптогенів з метою внесення до рецептури функціональних харчових продуктів. *Науковий вісник Львівського національного університету ветеринарної медицини та біотехнологій імені С.З. Ґжицького. Сер. Хар-*

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чові технології. 2015. Т. 17, № 4. С. 130-136.
4. Prior R.L., Wu X., Schaich K. Standardized Methods for the Determination of Antioxidant Capacity and Phenolics in Foods and Dietary Supplements. *J. Agric. Food Chem.* 2005. Vol. 53. P. 4290-4302.
5. Khoddami A., Wilkes M.A., Roberts T.H. Techniques for Analysis of Plant Phenolic Compounds. *Molecules.* 2013. Vol. 18 (2). P. 2328-2375.
6. Georgй S., Brat P., Alter P., Amiot M.J. Rapid Determination of Polyphenols and Vitamin C in Plant-Derived Products. *J. Agric. Food Chem.* 2005. Vol. 53. P. 1370-1373.
7. Escarpa A., Gonzalez M.C. Approach to the Content of Total Extractable Phenolic Compounds from Different Food Samples by Comparison of Chromatographic and Spectrophotometric Methods. *Anal. Chim. Acta.* 2011. Vol. 427. P. 119-127.
8. Porgali E., Вьуьktuncel E. Determination Of Phenolic composition and Antioxidant Capacity of Native Red Wines by High Performance Liquid Chromatography and Spectrophotometric Method. *Food Res. Intern.* 2012. Vol. 45. P.145-154.
9. Singleton V.L., Orthofer R., Lamuela-Raventos R.M. Analysis of Total Phenols and Other Oxidations Substractes and Antioxidans by Means of Folin-Ciocalteu Reagent. *Methods in Enzymology.* 1999. Vol. 29. P. 152-178.
10. Денисенко Т.А., Вишникин А.Б., Циганок Л.П. Особенности взаимодействия 18-молибдодифосфата и реактива Фолина-Чокальтеу с фенольными соединениями. *Аналитика и контроль.* 2015. Vol. 19 (3). P. 242-251.
11. Ainsworth E.A., Gillespie K.M. Estimation of Total Phenolic Content and Other Oxidation Substrates in Plant Tissues Using Folin-Ciocalteu Reagent. *Nature Protocols.* 2007. № 2. P. 875-877.
12. Пономарьова Л., Ярощук Р., Коваленко І., Гузь О. Визначення сумарного вмісту фенольних сполук в екстракті з листя Ginkgo Biloba *L. Scientific Works.* 2019. Т. 82 (2). 68-73. https://doi.org/10.15673/swonaft.v82i2.1165

REFERENCES

1. Scalbert A. and Williamso G. Dietary Intake and Bioavailability of Polyphenols. *J. Nutrition.* 2000 ; 130 : 2073-2085.
2. Ignat I., Volf I. and Popa V.I. A Critical Review of Methods for Characterization of Polyphenolic Compounds in Fruits and Vegetables. *Food Chem.* 2011 ; 126 : 1821-1835.
3. Steshenko M., Arsenieva L., Roiko A. and Palamarchuk A. Doslidzhennia fenolnykh spoluk roslynnykh adaptoheniv z metoiu vnesennia do retseptury funktsionalnykh kharchovykh produktiv [Plant Research Phenolic Compounds Adaptogen with a View to the Recipe Functional Food]. *Scientific Messenger of LNU of Veterinary Medicine and Biotechnologies. Series «Food Technologies»*. 2015 ; 17 (4) : 130-136 (in Ukrainian).
4. Prior R.L., Wu X. and Schaich K. Standardized Methods for the Determination of Antioxidant Capacity and Phenolics in Foods and Dietary Supplements. *J. Agric. Food Chem.* 2005 ; 53 : 4290-4302.
5. Khoddami A., Wilkes M.A. and Roberts T.H. Techniques for Analysis of Plant Phenolic Compounds. *Molecules*. 2013 ; 18 (2) : 2328-2375.
6. Georgй S., Brat P., Alter P. and Amiot M.J. Rapid Determination of Polyphenols and Vitamin C in Plant-Derived Products. J. *Agric. Food Chem*. 2005 ; 53 : 1370-1373.
7. Escarpa A. and Gonzбlez M.C. Approach to the Content of Total Extractable Phenolic Compounds from Different Food Samples by Comparison of Chromatographic and Spectrophotometric Methods. *Anal. Chim. Acta*. 2011 ; 427 : 119-127.
8. Porgali E. and Buyuktuncel E. Determination of Phenolic composition and Antioxidant Capacity of Native Red Wines by High Performance Liquid Chromatography and Spectrophotometric Method. *Food Res.* Intern. 2012 ; 45 : 145-154.
9. Singleton V.L., Orthofer R., Lamuela-Raventos R.M. Analysis of Total Phenols and Other Oxidations Substractes and Antioxidans by Means of Folin-Ciocalteu Reagent. *Methods in Enzymology.* 1999 ; 29 :152-178.
10. Denisenko T.A., Vishnikin A.B. and Tsiganok L.P. Osobennosti vzaimodeistvyia 18-molibdodifosfata u reaktiva Folina-Ciocalteu s fenolnymi soedineniyami [Reaction Features of 18-Molibdodiphosphate and Folin-Ciocalteu Reagent with Phenolic Compounds]. *Analitika i kontrol*. 2015 ;19 (3) : 242-251 (in Russian).
11. Ainsworth E.A. and Gillespie K.M. Estimation of Total Phenolic Content and Other Oxidation Substrates in Plant Tissues Using Folin–Ciocalteu Reagent. *Nature Protocols.* 2007 ; 2 : 875-877.
12. Ponomarova L.N., Yaroshchuk R.A., Kovalenko I.N. and Guz O.I. Vyznachennia sumarnoho vmistu fenolnykh spoluk v ekstrakti z lystia Ginkgo Biloba L [Determination of the Total Content of Phenolic Compounds in the Extract of Leaves Ginkgo Biloba L]. *Scientific Works.* 2018 ; 82. P. 68-73 https://doi.org/10.15673/swonaft.v82i2.1165 (in Ukrainian).

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