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Determination of the quantitative content of procyanidins in hawthorn fruits

Aim. To determine the quantitative content of procyanidins in hawthorn fruits.

Results and discussion. It was found that the quantitative content of procianidins in *Crataegus succulentha* Sarg. fruits was 1.45 % ± 0.02; *C. prunifolia* Sarg. – 1.24 % ± 0.05; *C. pseudokyrtostila* Klok. – 1.15 % ± 0.03; *C. leiomonogyna* Klok. – 1.28 % ± 0.04; *C. arnoldii* Sarg. – 1.38 %± 0.02; *C. submollis* Sarg. – 1.37 % ± 0.03; *C. mollis* Sarg. 1.48 % ± 0.01.

Experimental part. The content of procyanidins was determined by the spectrophotometric method calculated with the reference to cyanidin chloride. The optical density of the test solution was measured at a wavelength of 545 nm. The study was performed on a Thermo Fisher Scientific model EVOVUTION 60S spectrophotometer.

Conclusions. Procyanidins of fruits of 7 nonofficinal species of the genus *Crataegus* L. have been studied. It has been determined that all samples under research meet the requirements of the SPhU and EuPh by the indicator "Assay for procyanidins", which should be at least 1 %.

Key words: hawthorn; fruits; procyanidins

Н. В. Сидора, А. М. Ковальова, І. А. Данилова

Визначення кількісного вмісту проціанідинів плодів глоду

Мета. Встановлення кількісного вмісту проціанідинів плодів глоду.

Результати та їх обговорення. Встановлено, що кількісний вміст проціанідинів у плодах *Crataegus succulentha* Sarg. склав 1,45 % ± 0,02; *C. prunifolia* Sarg. – 1,24 % ± 0,05; *C. pseudokyrtostila* Klok. – 1,15 % ± 0,03; *C. leiomonogyna* Klok. – 1,28 % ± 0,04; *C. arnoldii* Sarg. – 1,38 % ± 0,02; *C. submollis* Sarg. – 1,37 % ± 0,03; *C. mollis* Sarg. 1,48 % ± 0,01.

Експериментальна частина. Вміст проціанідинів у сировині визначали спектрофотометричним методом у перерахунку на ціанідину хлорид. Оптичну густину випробовуваного розчину вимірювали при довжині хвилі 545 нм. Дослідження проводили на спектрофотометрі Thermo Fisher Scientific model EVOVUTION 60S.

Висновки. Досліджені проціанідини плодів 7 неофіційних видів роду *Crataegus* L. Встановлено, що усі досліджені зразки відповідають вимогам ДФУ та EuPh за показником «Кількісне визначення проціанідинів», який повинен бути не менше 1 %.

Ключові слова: глід; плоди; проціанідини

Н. В. Сидора, А. М. Ковалева, И. А. Данилова

Определение количественного содержания процианидинов плодов боярышника

Цель. Установление количественного содержания процианидинов плодов боярышника.

Результаты и их обсуждение. Установлено, что количественное содержание процианидинов в плодах *Crataegus succulentha* Sarg. составило 1,45 % ± 0,02; *C. prunifolia* Sarg. – 1,24 % ± 0,05; *C. pseudokyrtostila* Klok. – 1,15 % ± 0,03; *C. leiomonogyna* Klok. – 1,28 % ± 0,04; *C. arnoldii* Sarg. – 1,38 %± 0,02; *C. submollis* Sarg. – 1,37 % ± 0,03; *C. mollis* Sarg. 1,48 % ± 0,01.

Экспериментальная часть. Содержание процианидинов в сырье определяли спектрофотометрическим методом в пересчете на цианидина хлорид. Оптическую плотность измеряли при длине волны 545 нм. Исследования проводили на спектрофотометре Thermo Fisher Scientific model EVOVUTION 60S.

Выводы. Исследованы процианидины плодов 7 неофицинальных видов рода *Crataegus* L. Установлено, что все исследованные образцы отвечают требованиям ГФУ и EuPh по показателю «Количественное определение процианидинов», который должен быть не меньше 1 %.

Ключевые слова: боярышник; плоды; процианидины

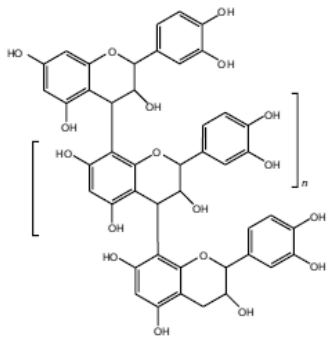
As a result of the phytochemical studies conducted it has been found that hawthorns fruits contain a wide range of phenolic compounds, namely flavonoids, anthocyanins, hydroxycinnamic acids, tannins [1, 2, 3, 4]. The State Pharmacopoeia of Ukraine (SPhU) and the European Pharmacopoeia (EuPh) recommend standardization of the medicinal plant raw material (MPRM) "*Crataegi fructus*" by the content of procyanidins [5].

Procyanedins are biologically active compounds; they belong to the class of flavonoids and are polymers that include the catechin chains by their chemical structure (Scheme 1).

It is known that procyanidins have the antioxidant, antihypertensive, antibacterial, anti-inflammatory properties and inhibit lipid peroxidation [6, 7, 8].

In order to standardize the fruits of nonofficinal hawthorn species we believe that the scientific interest is to determine the content of procyanidins in this raw material.

The aim of the work is to determine the quantitative content of procyanidins in hawthorn fruits.



Scheme 1. The chemical structure of procyanidins

Materials and methods

The objects of our study were fruits of *C. succulentha* Sarg., *C. prunifolia* Sarg., *C. pseudokyrtostilla* Klok., *C. leiomonogyna* Klok., *C. arnoldii* Sarg., *C. submollis* Sarg., *C. mollis* Sarg. All samples of the raw material were collected on the territory of Ukraine.

Determination of procyanidins in hawthorn fruits

To 2.5 g of the powdered raw material add 30 ml of *ethanol R (70 % V/V)*, heat under a reflux condenser for 30 min and filter. Wash the residue with 10.0 ml of *ethanol R (70 % V/V)*. Add 15.0 ml of hydrochloric acid *R1* and 10.0 ml of water *R* to the filtrate. Heat under a reflux condenser for 80 min. Allow to cool, filter and wash the residue with *ethanol R (70 % V/V)* until the filtrate is colorless. Dilute the filtrate to 250.0 ml with *ethanol R (70 % V/V)* [9]. Evaporate 50.0 ml of this solution in a round-bottomed flask to approximately 3 ml and transfer to a separating funnel. Rinse the round-bottomed flask sequentially with 10 ml and 5 ml of water *R* and transfer to the separating funnel. Shake the combined solution with 3 quantities of *butanol R*, each of 15 ml. Combine the organic layers and dilute to 100.0 ml with *butanol R*. Measure the absorbance of the solution at 545 nm [10].

When adding *hydrochloric acid R* to the filtrate acid cleavage of procyanidins occurs (Scheme 2).

As can be seen from Scheme 2, as a result of the reaction cyanidin and catechol are formed.

Results and discussion

The content of procyanidins (%) calculated with the reference to cyanidin chloride was determined using the following expression:

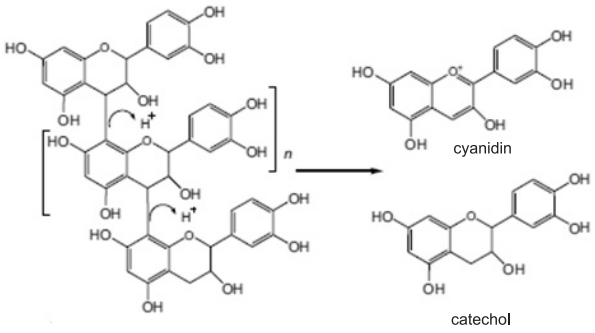
$$X = \frac{A \times 500}{75 \times m},$$

where: A – is the absorbance of the test solution at 545 nm; m – is the sample weight of the raw material studied, g.

For determination the specific absorbance of cyanidin chloride equaled 75 was used.

The absorption spectra of procyanidins of hawthorn fruits are shown in Fig. 1-7. The results of determination of the quantitative content of procyanidins are given in Table.

As can be seen from Table, the quantitative content of procyanidins in the MPRM studied (%) ranges from 1.15 ± 0.03 to 1.48 ± 0.01. The highest content is determined for *C. mollis* Sarg fruits.



Scheme 2. The mechanism of acid cleavage of procyanidin

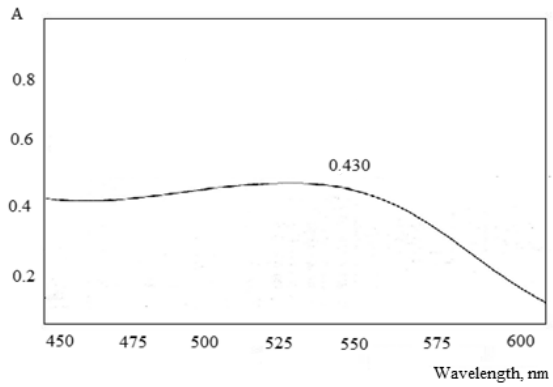


Fig. 1. The absorption spectrum of procyanidins of *C. pseudokyrtostilla* fruits

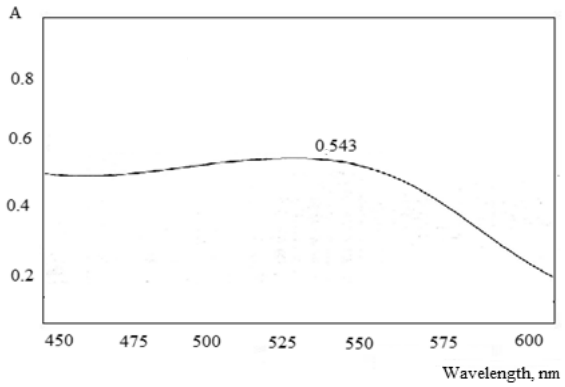


Fig. 2. The absorption spectrum of procyanidins of *C. succulentha* fruits

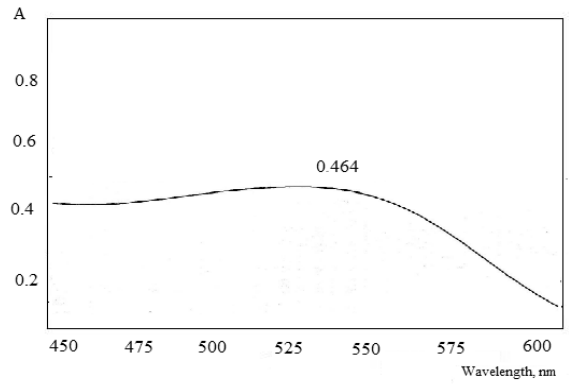


Fig. 3. The absorption spectrum of procyanidins of *C. prunifolia* fruits

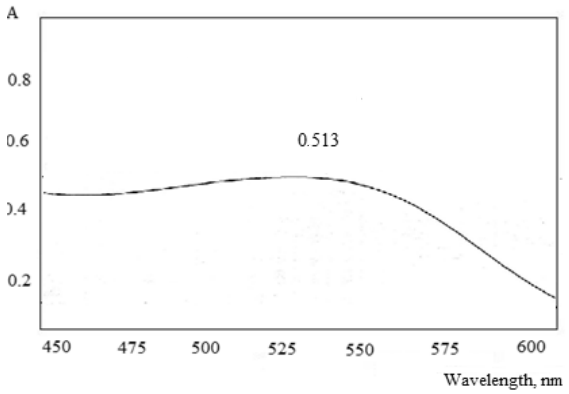


Fig. 6. The absorption spectrum of procyanidins of *C. submollis* fruits

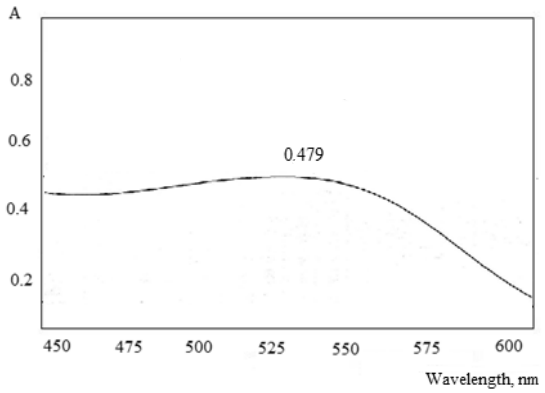


Fig. 4. The absorption spectrum of procyanidins of *C. leiomonogyna* fruits

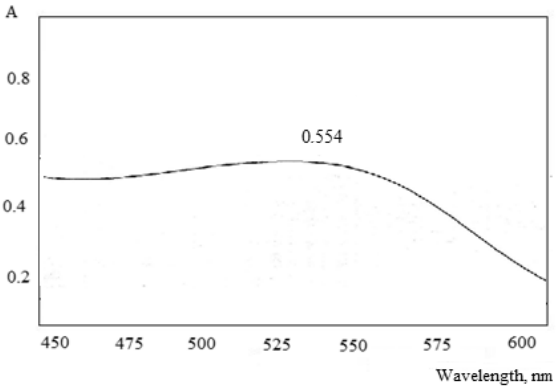


Fig. 7. The absorption spectrum of procyanidins of *C. mollis* fruits

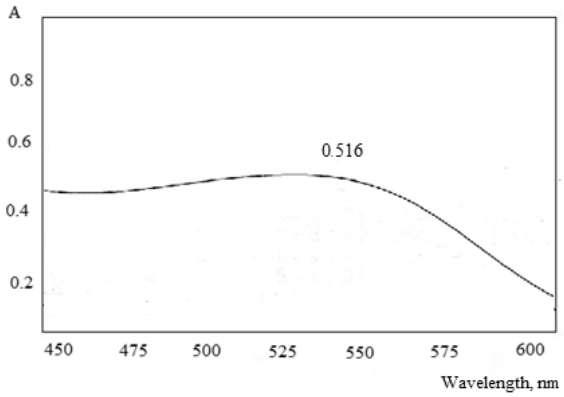


Fig. 5. The absorption spectrum of procyanidins of *C. arnoldii* fruits

Table

The content of prjcyanidins in hawthorn fruits

The raw material	Content, $\overline{X} \pm \Delta\overline{X}$ (%)
C. succulentha Sarg. fruits	1.45 ± 0.02
C. prunifolia Sarg. fruits	1.24 ± 0.05
C. pseudokyrtostilla Klok. fruits	1.15 ± 0.03
C. leiomonogyna Klok. fruits	1.28 ± 0.04
C. arnoldii Sarg. fruits	1.38 ± 0.02
C. submollis Sarg. fruits	1.37 ± 0.03
C. mollis Sarg. fruits	1.48 ± 0.01

Conclusions

For the first time procyanidins of fruits *C. succulentha* Sarg., *C. prunifolia* Sarg., *C. pseudokyrtostilla* Klok., *C. leiomonogyna* Klok., *C. arnoldii* Sarg., *C. submollis* Sarg. and *C. mollis* Sarg. have been identified. By the procyanidin content all test samples correspond to the requirements of the SPhU and EuPh, according to which this indicator for hawthorn fruits should be at least 1 %.

Conflict of interests: authors have no conflict of interests to declare.

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