

## PHENOLIC COMPOUNDS AS ONE OF THE BIOCHEMICAL INDICATORS OF FROST RESISTANCE OF THE GENUS *CYNOXYLON* L. REPRESENTATIVES IN THE CONDITIONS OF FOREST-STEPPE OF UKRAINE

*The study of phenolic compounds in shoots of the genus Cynoxylon L. representatives due to the sharp fluctuations of temperatures during winter dormancy. Analysis of the obtained experimental data showed that the species and forms of the genus Cynoxylon differ significantly in content of phenolic compounds in the period of sharp fluctuations of temperatures. But, the histogram depending of the content of phenolic compounds from average daily temperatures of all investigated species has similarities and differences. It is established that Cynoxylon japonica (Siebold & Zucc.) Nakai has a larger amplitude in the content of phenols in the temperature change unlike Cynoxylon florida Raf., where the change in the accumulation of phenols occurs more smoothly. C. japonica responds to more short-term changes in temperature contrast C. florida, where the changes of accumulation of phenolic compounds occur only during prolonged cold snaps, or warming. All this indicates a higher frost resistance C. japonica in relation to C. florida.*

**Key words:** phenolic compounds, *Cynoxylon florida*, *Cynoxylon japonica*, Forest-Steppe of Ukraine.

Frost resistance is determined by many factors, in particular the important role of secondary metabolites involved in the biochemical adaptation of plants. This group includes phenols. These compounds are formed in all organs of plants from sugars and participate in the process of cellular respiration, transferring hydrogen from oxidative molecules. Phenolic compounds exhibit a strong effect on the growth of plants, inhibiting germination of seeds, growth of stems and roots. They have strong antibacterial properties and provide plant immunity to fungal and especially to bacterial infection [2].

In adverse conditions, plants synthesize an increased amount of phenolic compounds, because in most cases they have a protective effect [5]. Often the healthy plant has no protective phenols, but they are formed in it as reaction to infection with the causative agent of a disease. Phenolic compounds have an important role in wound healing, cell division, and in protecting tissues from penetrating radiation, free radicals, mutagens and strong oxidizers.

Phenolic compounds can act as low molecular weight antioxidants, protect cells from the effects of oxidative stress developing in conditions of hypo-

thermia [8, 9, 10, 12]. Having high reaction activity due to the presence in the structure of aromatic rings and free hydroxyl groups, they can easily engage in free-radical reactions and connect the active forms of oxygen and peroxy radicals produced in the cells under stress conditions [7, 11, 13].

A characteristic feature of the phenolic compounds that distinguish them from many other secondary metabolites is their ability to interact with proteins by forming hydrogen bonds [1, 3]. In this regard, some of them may participate in the regulation of enzyme activity, affecting, therefore, on the metabolic processes in the period of low-temperature adaptation.

The aim of our study — to determine the content of phenolic compounds in sprouts of different species, forms and cultivars of the genus *Cynoxylon* L. in the winter and the dependence of these indicators of frost resistance of plants.

### Material and methods

The experience was launched on 12 December 2014 the average daily temperature  $-2^{\circ}\text{C}$  and completed 4 February 2015. In this period was very severe fluctuations in temperature from  $-10^{\circ}\text{C}$  to  $+10^{\circ}\text{C}$ . Samples were collected weekly. The number of sampling — 9. Objects of research

were *C. florida* Raf.(forms 1, 2 and var. *rosea*) and *C. japonica* (Siebold & Zucc.) Nakai (forms 1, 2, 3, ‘Milky Way’ and ‘Wolf eyes’).

For quantitative definition of phenols was used the methodology [4], based on oxidation of a reactant of Folin-Chokolta containing a tungstate of sodium and phosphomolybdate of sodium with formation of a blue complex, having absorption maximum at the wave length of 730 nm, which intensity of coloring is estimated by a photoelectrocolorimetric method was used.

For objects studied visual observations conducted general condition of the plants in the spring to determine their hardiness [6].

### Results and discussion

Analysis of the obtained experimental data showed that the types and forms of the genus *Cynoxylon* differ significantly in content of phenolic compounds in the period of sharp fluctuations of temperatures. But, the histogram depending of the content of phenolic compounds from average daily temperatures of all investigated species has similarities and differences.

It is established that *C. japonica* has a larger amplitude in the content of phenols in the temperature change unlike *C. florida*, where the change in the accumulation of phenols occurs more smoothly. *C. japonica* responds to more short-term changes in temperature contrast *C. florida*, where the changes of accumulation of phenolic compounds

#### Air temperature (°C) at a height of 2 m above ground surface (Kyiv)

Date	The minimum temperature	The maximum temperature
12.12.2014	-0.5	+1.8
18.12.2014	-1.3	+3.0
24.12.2014	+4.5	+7.5
30.12.2014	-9.3	-6.7
06.01.2015	-10.1	-9.0
14.01.2015	+2.1	+9.5
21.01.2015	0	+0.9
28.01.2015	-3.7	-0.1
04.02.2015	-0.7	+3.9

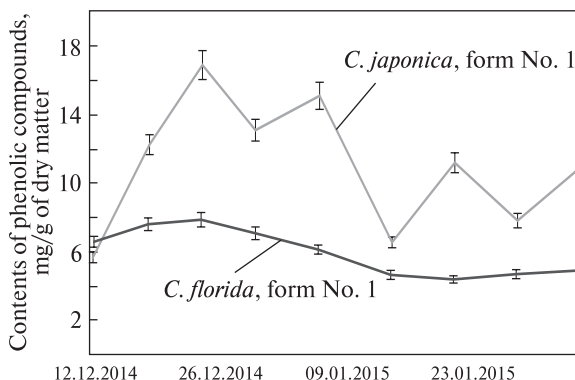


Fig. 1. The total content of phenolic compounds in sprouts of *Cynoxylon japonica*, form No. 1 and *Cynoxylon florida*, form No. 1 in the period of winter dormancy

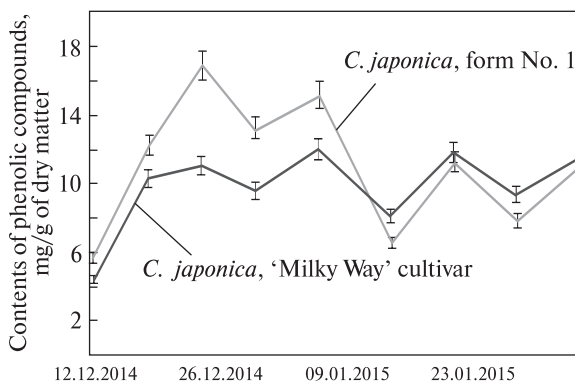


Fig. 2. The total content of phenolic compounds in sprouts of *Cynoxylon japonica*, form No. 1 and *Cynoxylon japonica*, ‘Milky Way’ cultivar in the period of winter dormancy

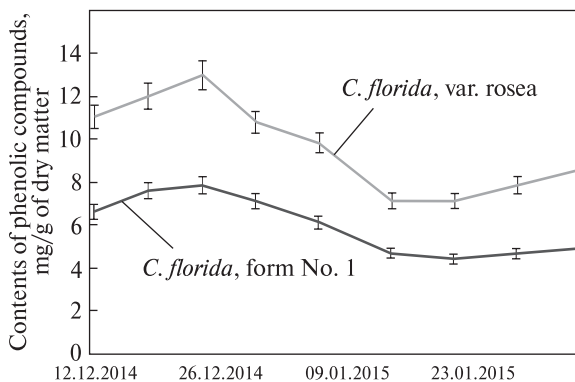


Fig. 3. The total content of phenolic compounds in sprouts of *Cynoxylon florida*, form No. 1 and *Cynoxylon florida*, var. *rosea* in the period of winter dormancy

occur only during prolonged cooling or warming. In addition, the data obtained show that *C. japonica* has less time induction of the primary stress response is 4–5 days. *C. florida* in this period is 10 days. All this indicates a higher frost resistance *C. japonica* in relation to *C. florida*.

These visual observations confirm the results of biochemical researches within a species, but no difference was observed between hardness *C. japonica* and *C. florida*.

### Conclusions

Due to the fact that the dynamics of phenolic compounds in plants of the genus *Cynoxylon* virtually unexplored, the study of this subject has been a priority task. The increase in the content of phenolic compounds in plants of the genus *Cynoxylon* in the process of low-temperature adaptation testifies to their important role in protecting cells from the action of stress factors. Because the dynamics of phenolic compounds in plants of the genus *Cynoxylon* displays its interaction with the environment, these compounds can be used as biochemical marker to evaluate the level of adaptation within species.

1. Барабой В.А. Биологическое действие растительных фенольных соединений / В.А. Барабой. — К.: Наук. думка, 1976. — 260 с.
2. Блажей А.С. Фенольные соединения растительно-го происхождения / А.С. Блажей, Л.П. Шутый. — М.: Мир, 1977. — 239 с.
3. Запрометов М.Н. Способность изолированных хлоропластов из листьев фасоли осуществлять биосинтез фенольных соединений / М.Н. Запрометов, Т.Н. Николаева // Физиология растений. — 2003. — Т. 50, № 5. — С. 699–702.
4. Ксендзова Э.Н. Прием количественного определения фенольных соединений в растительных тканях / Э.Н. Ксендзова // Бюл. ВНИИ защиты растений. — 1971. — № 20. — С. 55–58.
5. Сарану Л.П. Фенольные соединения и рост растений / Л.П. Сарану, В.И. Кефели // Фенольные соединения и их биологические функции. Материалы 1-го Всесоюз. симп. по фенольным соединениям. — М.: Наука, 1968. — С. 129–138.
6. Соколов С.Я. Современное состояние теории акклиматизации и интродукции растений / С.Я. Соколов // Тр. БИН АН СССР. Интродукция растений и зеленое строительство. — 1957. — Вып. 6. — С. 34–42.

7. Antioxidative phenolic compounds from sage (*Salvia officinalis*) / M. Wang, M. Rangarajan, Y. Shao, E. Voie, T. Huang // J. Agric. Food Chem. — 1998. — Vol. 46. — P. 4869–4873.
8. Kondo N. Enhancement of the tolerance to oxidative stress in cucumber (*Cucumis sativus* L.) seedlings by UV-B irradiation: Possible involvement of phenolic compounds and antioxidative enzymes / N. Kondo, M. Kawashima // J. Plant Res. — 2000. — Vol. 113. — P. 311–317.
9. Larson R.A. The antioxidants of higher plants / R.A. Larson // Phytochemistry. — 1988. — Vol. 27. — P. 969–978.
10. Rice-Evans C.A. Antioxidant properties of phenolic compounds / C.A. Rice-Evans, N.J. Miller, G. Paganga // Trends in Plant Science. — 1997. — Vol. 2. — P. 152–159.
11. Terao J. Protective effect of epicatechin, epicatechin gallate and quercetin on lipid peroxidation in phospholipid bilayers / J. Terao, M. Piskura, Q. Yao // Archives of Biochemistry and Biophysics. — 1994. — Vol. 308. — P. 278–284.
12. Wingsle G. Low temperature, high light stress and antioxidant defence mechanisms in higher plants / G. Wingsle, S. Karpinski, J. Hallgren // Phytan (Austria). Special issue. — 1999. — Eurosilva 4. — P. 253–268.
13. Zhao H. Protective effects of exogenous antioxidants and phenolic compounds on photosynthesis of wheat leaves under high irradiance and oxidative stress / H. Zhao, Q. Zou // Photosynthetica. — 2002. — Vol. 40(4). — P. 523–527.

### REFERENCES

1. Baraboj, V.A. (1976), Biologicheskoe dejstvie rastitelnyh fenolnyh soedinenij [The biological effect of plant phenolic compounds]. Kyiv: Naukova dumka, 260 p.
2. Blazhej, A.S. and Shutyj, L.P. (1977), Fenolnye soedinenija rastitelnogo proishozhdenija [Phenolic compounds of plant origin]. Moskva, Mir, 239 p.
3. Zaprometov, M.N. and Nikolaeva, T.N. (2003), Sposobnost izolirivannyh hloroplastov iz listiev fasoli osushchestvlyat biosintez fenolnyh soedinenij [The ability of chloroplasts isolated from leaves of bean implement the biosynthesis of phenolic compounds]. Fiziologia rastenij [Physiology of Plants], vol. 50, N 5, pp. 699–702.
4. Ksendzova, E.N. (1971). Prijom kolichestvennogo opredelenija fenolnyh soedinenij v rastitelnyh tkanjah [Reception quantification of plant's phenolic compounds in tissues]. Byulleten Vsesoyuznogo nauchno-issledovatel'skogo instituta zashchity rastenij [Bulletin All-Union Scientific Research Institute of Plant Protection], No 20, pp. 55–58.
5. Saranpuu, L.P. and Kefeli, V.I. (1968), Fenolnye soedinenija i rost rastenij [Phenolic compounds and plant growth] Materialy I Vsesoyuznogo simpoziuma po fenolnym soedinenijam [Materials of the first All-Union

- Symposium on phenolic compounds]. Moskva, Nauka, pp. 129–138.
6. Sokolov, S.J. (1957), *Sovremennoe sostojanie teorii akklimatizacii i introdukcii rastenij* [The present state of the theory of acclimatization and plant introduction] Trudy botanicheskogo instituta AN SSSR. Introdukcija rastenij i zel'noe stroitel'stvo [Works of the Botanical Institute of the USSR. Plant Introduction and Green Building], vol. 6, pp. 34–42.
  7. Wang, M., Rangarajan, M., Shao, Y., Voie, E. and Huang, T. (1998), Antioxidative phenolic compounds from sage (*Salvia officinalis*). J. Agric. Food Chem., vol. 46, pp. 4869–4873.
  8. Kondo, N. and Kawashima, M. (2000), Enhancement of the tolerance to oxidative stress in cucumber (*Cucumis sativus* L.) seedlings by UV-B irradiation: Possible involvement of phenolic compounds and antioxidative enzymes. J. Plant Res., vol. 113, pp. 311–317.
  9. Larson, R.A. (1988), The antioxidants of higher plants. Phytochemistry, vol. 27, pp. 969–978.
  10. Rice-Evans, C.A., Miller, N.J. and Paganga, G. (1997), Antioxidant properties of phenolic compounds. Trends in Plant Science, vol. 2, pp. 152–159.
  11. Terao, J., Piskura, M. and Yao, Q. (1994), Protective effect of epicatechin, epicatechin gallate and quercetin on lipid peroxidation in phospholipid bilayers. Archives of Biochemistry and Biophysics, vol. 308, pp. 278–284.
  12. Wingsle, G., Karpinski, S. and Hallgren, J. (1999), Low temperature, high light stress and antioxidant defence mechanisms in higher plants. Phytion (Austria). Special issue, Eurosilva 4, pp. 253–268.
  13. Zhao, H. and Zou, Q. (2002), Protective effects of exogenous antioxidants and phenolic compounds on photosynthesis of wheat leaves under high irradiance and oxidative stress. Photosynthetica, vol. 40(4), pp. 523–527.

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#### ФЕНОЛЬНІ СПОЛУКИ ЯК ОДИН ІЗ БІОХІМІЧНИХ ПОКАЗНИКІВ МОРОЗОСТІЙКОСТІ ПРЕДСТАВНИКІВ РОДУ *CYNOXYLON* L. В УМОВАХ ЛІСОСТЕПУ УКРАЇНИ

Проведено вивчення фенольних сполук у пагонах представників роду *Cynoxylon* L. у зв'язку з різкими коливаннями температури під час зимового спокою, щоб визначити реакцію на зниження температури.

Аналіз отриманих даних показав, що види і форми роду *Cynoxylon* суттєво відрізняються за вмістом фенольних сполук у період різких коливань температур. Гістограма залежності вмісту фенольних сполук від середньодобових температур усіх досліджених видів має спільні риси і відмінності. Встановлено, що *Cynoxylon japonica* (Siebold & Zucc.) Nakai має велику амплітуду вмісту фенолів за зміни температури на відміну від *Cynoxylon florida* Raf., у якого зміна накопичення фенолів відбувається поступово. *C. japonica* реагує на короткострокові зміни температури, тоді як у *C. florida* зміна накопичення фенольних сполук відбувається лише під час тривалих знижень температури або потепління. Це свідчить про вищу морозостійкість *C. japonica* порівняно з *C. florida*.

**Ключові слова:** фенольні сполуки, *Cynoxylon florida*, *Cynoxylon japonica*, Лісостеп України.

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#### ФЕНОЛЬНЫЕ СОЕДИНЕНИЯ КАК ОДИН ИЗ БИОХИМИЧЕСКИХ ПОКАЗАТЕЛЕЙ МОРОЗОУСТОЙЧИВОСТИ ПРЕДСТАВИТЕЛЕЙ РОДА *CYNOXYLON* L. В УСЛОВИЯХ ЛЕСОСТЕПИ УКРАИНЫ

Проведено изучение фенольных соединений в побегах представителей рода *Cynoxylon* L., чтобы определить реакцию на снижение температуры в связи с резкими колебаниями температуры во время зимнего покоя. Анализ полученных данных показал, что виды и формы рода *Cynoxylon* существенно отличаются по содержанию фенольных соединений в период резких колебаний температур. Гистограмма зависимости содержания фенольных соединений от среднесуточных температур всех исследованных видов имеет общие черты и отличия. Установлено, что для *Cynoxylon japonica* (Siebold & Zucc.) Nakai характерна большая амплитуда содержания фенолов при изменении температуры в отличие от *Cynoxylon florida* Raf., у которого изменение накопления фенолов происходит более плавно. *C. japonica* реагирует на краткосрочные изменения температуры, тогда как у *C. florida* изменение накопления фенольных соединений происходит только во время длительных похолоданий или потепления. Это свидетельствует о более высокой морозоустойчивости *C. japonica* по сравнению с *C. florida*.

**Ключевые слова:** фенольные соединения, *Cynoxylon florida*, *Cynoxylon japonica*, Лесостепь Украины.