Received: 09.08.2025 | Revision: 17.08.2025 | Accepted: 17.08.2025

Agriculture. Plant growing

doi: 10.31210/spi2025.28.03.11
UDC 615.32:58+547.963
ORIGINAL ARTICLE

Scientific Progress & Innovations
https://journals.pdaa.edu.ua/visnyk

28 (3)
2025

The peculiarities of agglutinins' activity in male and female plants of Ginkgo biloba (*Ginkgo biloba* L.)

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Article info

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Citation: Samorodov, V., & Pospielov, S. (2025). The peculiarities of agglutinins' activity in male and female plants of Ginkgo biloba (*Ginkgo biloba* L.). *Scientific Progress & Innovations*, 28(3), 69–73. doi: 10.31210/spi2025.28.03.11

Lectins, or hemagglutinins, are protein compounds that are capable of selectively and reversibly binding carbohydrates without their chemical transformation, exhibiting the property of agglutinating cells and particles. Owing to their different functions, they have significant theoretical and applied importance in biology and medicine, which makes the search for new sources of these substances topical. Ginkgo biloba (*Ginkgo biloba* L.), the only modern representative of Ginkgopsida class, is known for its rich phytochemical composition, which determines the use of its raw materials in the pharmaceutical industry. The purpose of the study was to determine the pH profile of phytohemagglutinins' activity in cone berries, buds, leaves and one-year shoots of female and male ginkgo trees. The objects were the samples collected from plantings in Poltava. The study was conducted using the standard method for assessing the degree of human hemagglutination. The accumulated experimental data showed that agglutinins were present in all the studied plant organs however their activity varied significantly depending on the tissue and sex of the trees. In cone berries, the maximum activity was recorded in the pedicle and collar, while in seeds proteins were concentrated mainly in the sclerotesta. The vegetative organs also showed sexual differentiation: in female trees, the lectins' activity in the buds, shoots, and especially in the leaves was significantly higher than in male trees. At the same time, in the leaf blades of female trees, the level of hemagglutination in the pH range of 6.0–8.0 reached 12.3 points, while in male trees it did not exceed 6.5 points. The most pronounced differences were observed in the alkaline pH zone, where the activity reached the maximum values. The obtained results confirm the new fact for the science – the presence of agglutinins in *Ginkgo biloba L.* and their different localization depending on the organ and sex of the plant. The revealed conformities to the laws of nature may have practical significance as marker traits for identifying female and male plants in the pre-reproductive period of ontogenesis, which is promising for pharmacognostic and biotechnological research.

Keywords: Ginkgo biloba, *Ginkgo biloba*, lectins, proteins, agglutination, sexualization of trees.

Особливості активності аглютинінів у жіночих і чоловічих рослинах гінкго дволопатевого (*Ginkgo biloba* L.)

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Лектини, або гемаглютиніни, є білковими сполуками, що здатні вибірково та зворотно зв'язувати вуглеводи без їхнього хімічного перетворення, проявляючи властивість аглютинації клітин і часток. Завдяки різноманітним функціям вони мають значне теоретичне й прикладне значення у біології та медицині, що зумовлює актуальність пошуку нових джерел цих речовин. Гінкго дволопатеве (*Ginkgo biloba* L.), єдиний сучасний представник класу Ginkgopsida, відомий багатим фітохімічним складом, що обумовлює використання його сировини у фармацевтичній промисловості. Метою дослідження було визначення pH-профілю активності фітогемаглютинінів у шишкоягодах, бруньках, листках і однорічних пагонах жіночих та чоловічих дерев гінкго. Об'єктами були зразки, зібрані з насаджень у Полтаві. Дослідження проводили за стандартною методикою оцінки ступеня аглютинації еритроцитів людини. Накопичені експериментальні дані показали, що аглютиніни присутні в усіх досліджених органах рослини, однак їх активність значно варіювала залежно від тканини та статі дерев. У шишкоягодах максимальна активність була зафіксована в ніжці та комірці, тоді як у насінні білки зосереджувались переважно в склеротесті. Вегетативні органи також виявили статеву диференціацію: у жіночих дерев активність лектинів у бруньках, пагонах і особливо в листках була значно вищою, ніж у чоловічих. При цьому у листкових пластинках жіночих дерев рівень гемаглютинації у діапазоні pH 6,0–8,0 доходив до 12,3 бали, тоді як у чоловічих не перевищував 6,5 бали. Найбільш виражені відмінності спостерігалися у лужній зоні pH, де активність сягала максимальних значень. Отримані результати засвідчують новий для науки факт наявності аглютинінів у *Ginkgo biloba* L. та їхню різну локалізацію в залежності від органу і статі рослини. Виявлені закономірності можуть мати практичне значення як маркерні ознаки для ідентифікації жіночих і чоловічих особин у прегенеративний період онтогенезу, що є перспективним для фармакогностичних і біотехнологічних досліджень.

Ключові слова: гінкго дволопатеве, *Ginkgo biloba*, лектини, білки, аглютинація, сексуалізація дерев.

Бібліографічний опис для цитування: *Самородов В. М., Поспєлов С. В.* Особливості активності аглютинінів у жіночих і чоловічих рослинах гінкго дволопатевого (*Ginkgo biloba* L.). *Scientific Progress & Innovations*. 2025. № 28 (3). С. 69–73.

Introduction

Lectins or hemagglutinins are substances of protein nature that have the unique property of selectively and reversibly binding carbohydrates without their chemical transformation. This is the most simply manifested in the form of agglutination of particles and cells, for example, erythrocytes [1]. It has been determined that any living cell contains not one, but several lectins. Their functions are different and important, due to which lectins are increasingly used in many theoretical and applied fields of biology and medicine [1–2]. This is evidenced by the fact that the catalogs of well-known world biotechnological and chemical companies offer a large list of commercial preparations of lectins and their components, the number of which increases annually. That is why the search for new sources of these compounds is a topical task of biological science [3].

Ginkgo biloba (*Ginkgo biloba* L.) is the only representative of Ginkgopsida class, Ginkgoaceae family of Pinophyta section [5]. This tree is considered to be the oldest among the trees in the world, as it appeared on the Earth 250 million years ago. During this historical period, various climatic cataclysms and disasters occurred, but the species remained in natural biocenoses [6–8]. The trees even survived in the epicenter of the nuclear explosion in Hiroshima [8]. Such resistance to environmental factors is associated with the unique phytochemical composition of this species, which contains a large number of biologically active substances [4]. In different countries of the world, various medical preparations with pronounced cerebral protective effects, as well as bio-active dietary supplements, are produced from ginkgo raw materials [4, 6–8]. As to their popularity, they take the first places in various ratings. In addition to leaves, ginkgo cone berries and seeds are used [8, 10].

Ginkgo has been cultivated in Ukraine since 1809 and its popularity is growing [10–14]. It is worth mentioning that the first plantations of the crop have already been created, which should provide pharmaceutical production with its own raw materials. At the same time, phytochemical research in Ukraine needs to be deepened [9,15].

The aim of the study

The purpose was to investigate the pH profile of phytohemagglutinins' activity in different parts of Ginkgo biloba cone berries, buds, leaves, and one-year shoot growths of female and male trees.

Materials and methods

The object of the studies were buds, one-year shoot growths, leaves and cone berries of one-year-old Ginkgo biloba (*Ginkgo biloba* L.), trees that entered the generative phase of development. They grow in different locations in the city of Poltava: the Arboretum of Poltava State Agrarian University (No. 1); the Botanical Garden of V. G. Korolenka Poltava National Pedagogical University (No. 2); Vaviloviaria – the memorial park of M. I. Vavilova Poltava Agricultural Research Station of the Institute of Pig Breeding and Agro-Industrial Complex of the National Academy of Agrarian Sciences (No. 3) [14]. The raw materials were harvested at different times: buds in April, leaves and one-year shoot growths from male and female specimens, as well as cone berries from female plants – in October (***Figure 1***).

The activity of hemagglutinins was determined by the generally accepted method according to the level of agglutination of human erythrocytes [16, 17]. The raw materials were dried to an air-dry state, crushed and extracted with physiological solution in a ratio of 1 : 10. The intensity of agglutination was defined and evaluated in immunological plates, visually in points in a series of eight dilutions. The maximum activity in the experiments made 8 × 3 points = 24 points [17]. It was previously established that the lectins' activity depends on the pH of the medium and the appropriate method was developed according to which the intensity of the agglutination reaction was determined in the pH range from 4.0 to 8.0 [18]. For this purpose, 0.05 ml of buffer solution was added to each of the eight cells of the plate, 0.05 ml of extract was added and a series of consecutive two-fold dilutions was conducted. After that, 0.05 ml of 2% suspension of washed-out human erythrocytes was added to each cell, the plate was incubated at a temperature of +25°C for one hour. The assessment was made visually according to a five-point scale: 0 points – agglutination is absent; 3 points – maximum agglutination [17].

Results and discussion

Agglutinins' activity in the cone berries of Ginkgo biloba. It is known that the cone berry of ginkgo has three clearly expressed coatings [5, 12]. The outer one is juicy, yellow-amber colored with a waxy cover and an unpleasant odor (sarcotesta or arilus). The middle one is hard, ridged (sclerotesta); the inner one is film-like. Under the coatings, the seed is found, which contains an embryo with 2–3 cotyledons and the developed endosperm [10]. The analysis of the obtained research results allows us to state the presence of phytohemagglutinins in the above-mentioned cone berry coatings and seed elements. However, the proteins' activity in each of them differed significantly. It was the lowest in freshly harvested seeds. In all female plants, the lectins' activity equaled 0.1–0.3 points, and was determined only in the acidic zone, at pH of 4.0–4.5. It is noteworthy that practically almost all seed agglutinins are concentrated in the sclerotesta, which practitioners call the "stone". As for the sarcotesta, the activity of lectins in it is higher than in the sclerotesta and seeds. Depending on the studied location of each tree, it varies from 6.8 to 8.3 points, and the maximum values were determined at pH=7.0–8.0.

It was found that the agglutinins with the highest activity were localized in the cone berry pedicle (***Table 1***). At the same time, we practically did not observe any difference between the locations of the studied trees. It is also worth mentioning that a high activity was determined in almost the entire pH range, from 4.5 to 8.0. A general conformity was observed: in the acidic zone (pH=4.0–6.0) the activity made from 6.0–15.0 to 21.0–24.0, and in the alkaline zone (6.5–8.0) – from 21.0–24.0 to 22.2–24.0. Even at the highest dilutions of the extracts, the agglutination reached its maximum value.

The similar conformities were also revealed at determining the activity of hemagglutinins in the collar – the base of the cone berry pedicle. Therefore, in the future, all of the above mentioned makes us think about the physiological nature of the places of lectins' concentration, their role in the processes of fertilization and embryogenesis in ginkgo. This is all more interesting due to the fact that undeveloped cone berries, which fall off at different stages of their formation, have almost the same lectin activity as normally formed ones – 20.5 points.

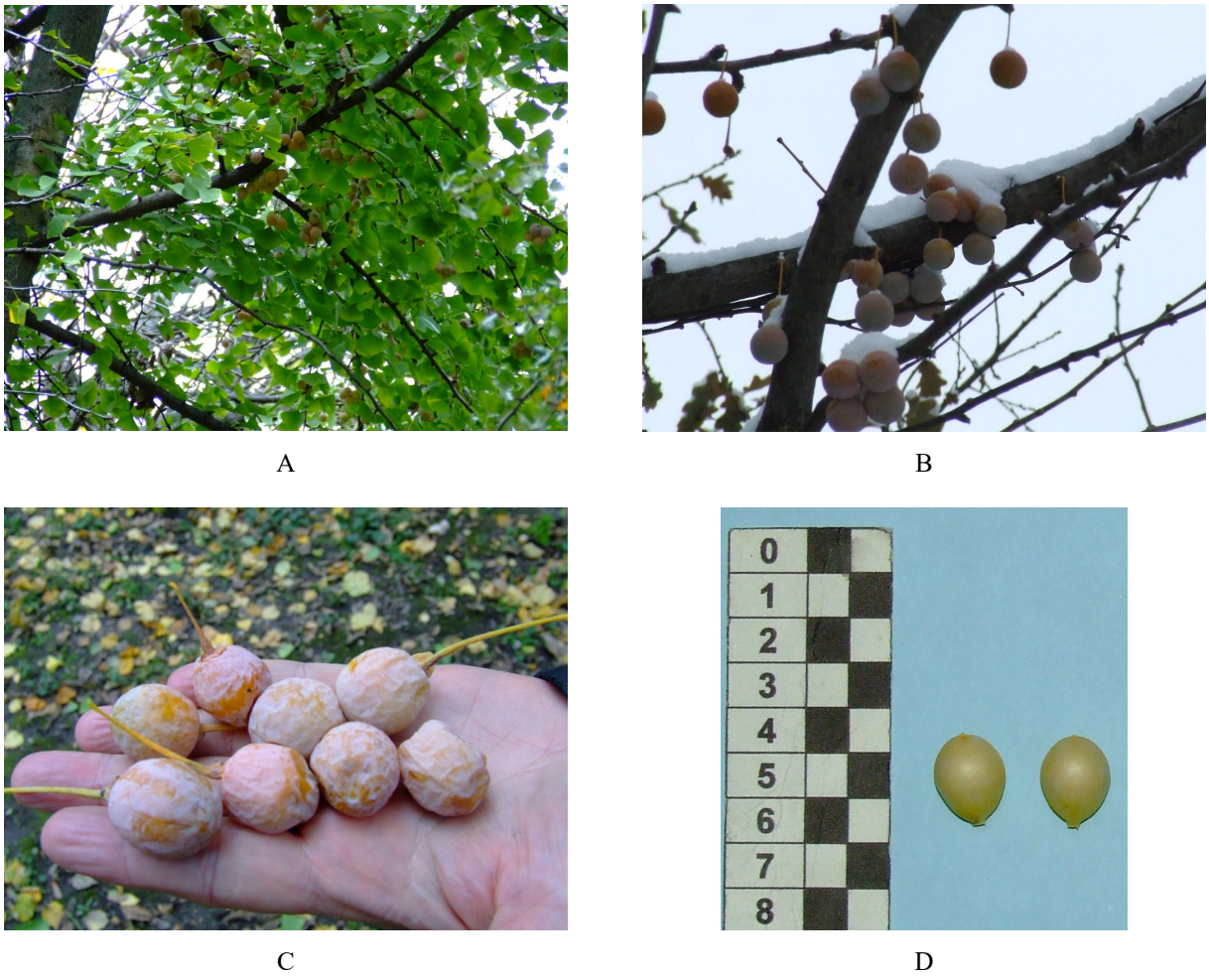

Figure 1. Cone berries (A-C) at different periods of ripening and seeds (D) of Ginkgo biloba (*photo by S. Pospielov*)

Table 1
pH profile of agglutinins' activity in extracts of Ginkgo biloba pedicle (M±m, n=4)

Tree locations	pH								
	4.0	4.5	5.0	5.5	6.0	6.5	7.0	7.5	8.0
No. 1	14.6±1.2	23.4±0.5	23.5±0.7	23.2±0.7	23.3±0.7	23.3±0.9	23.6±0.5	23.1±0.7	23.0±0.9
No. 2	8.9±0.9	21.9±0.5	22.8±1.0	23.1±0.7	23.4±0.8	22.1±0.7	23.3±1.0	23.0±0.4	22.6±1.0
No.3	5.8±0.7	11.8±0.7	19.9±1.2	20.5±0.6	20.6±0.8	20.8±0.7	20.1±0.7	22.3±1.0	22.0±1.5

Agglutinins' activity in the vegetative organs of Ginkgo biloba. Important conformities that had not been previously described in the literature available to us were discovered during the study of lectins in the buds, shoots, and leaves of male and female Ginkgo biloba trees (***Figures 2, 3***). It has been found that there is a relationship between the intensity of hemagglutination and the sexualization of trees.

The analysis of the pH profile in the buds indicates a low level of agglutination in the range of pH=4.0–5.5 (the maximum of up to 0.83 points), with an increase in pH from 6.0 to 8.0 in the buds of female plants, the activity increased to 3.67–9.83 points, and in the buds of male plants, it was significantly lower – from 1.83 to 4.33 points. In one-year-old shoots the indicated pattern was preserved, at the same time, the activity of lectins was observed almost in the entire pH range from 4.5 to 8.0. In the shoots of female plants it made 3.67–7.33 points, and in male ones – 1.67–3.83 points.

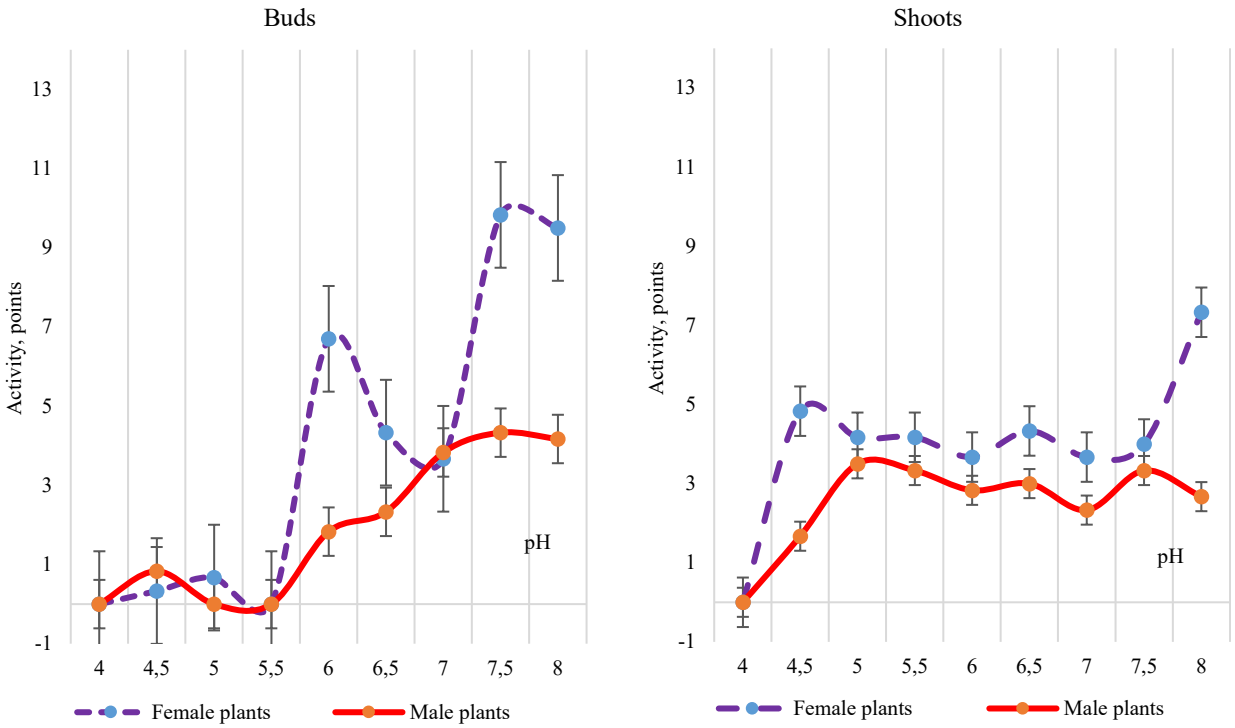

Figure 2. pH profile of agglutinins' activity in bud extracts and one-year growths of Ginkgo biloba shoots of different sexes

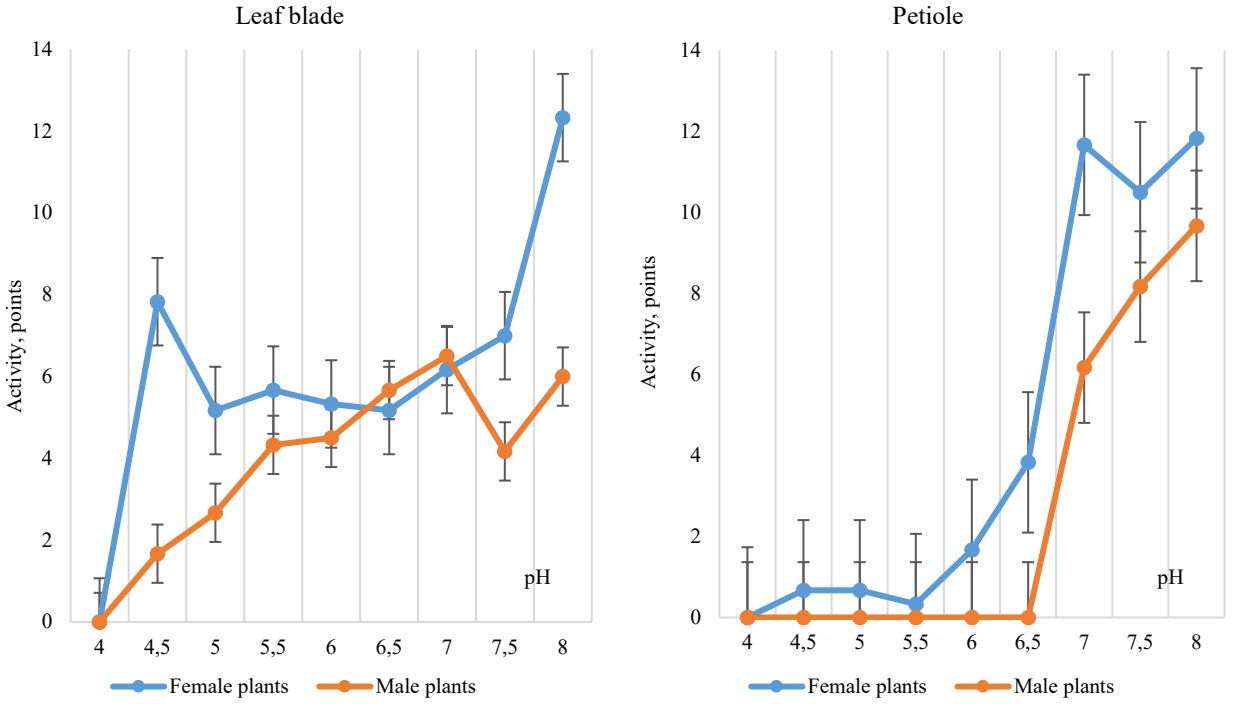

Figure 3. pH profile of agglutinins' activity in leaf extracts of Ginkgo biloba of different sexes

In the leaves, the difference in lectins' activity was even more expressed, especially in leaf blades. In the latter ones, the activity was determined in practically the entire pH range: in female plants – from 5.17 to 12.33 agglutination points, in male plants – 1.67–6.5 points. In the leaf petioles of female plants, agglutination was the minimal at pH=4.0–5.5 (up to 0.67 points) and it rapidly increased to 11.83 points in the range of pH=6.0–8.0. In the leaf petioles of male trees, agglutination was absent in the range of pH=4.0–6.5, and the maximum made 9.67 points at pH=8.0.

There have already been reports of certain differences in the chemistry of heterosexual ginkgo plants. The scientists from Yuriia Fedkovycha Chernivtsi National University have proven that the leaves and roots of cuttings from female ginkgo trees accumulate more

protein substances in comparison with male ones [19]. The researchers from O. V. Fomina Botanical Garden of Tarasa Shevchenka National University of Kyiv have found that ginkgo plants of different sexes accumulate in their leaves unequal amounts of such an important protein component as proline amino acid. Its amount was almost twice as high in the leaves of female trees as in male ones. It is interesting that the maximum difference was manifested in October during the leaf fall [20]. The similar conformities were established earlier for sea buckthorn. Ye. M. Herhel's studies have proven that female plants accumulate more lectins in their leaves in comparison with male ones [21].

The conformities to the laws of nature described above suggest a possible applied significance of the research. It is quite obvious that the assessment as to lectins' activity will help to distinguish female and male plants at the early stages of ontogenesis not only in ginkgo, but also in other types of fruit, rare, and medicinal crops, which will allow the formation of highly productive plantations.

Conclusions

The conducted studies allow us to state the previously not described fact of the presence of agglutinins in Ginkgo biloba both in cone berries and buds as well as in one-year shoot growths and leaves. It has been found that the place of maximum activity of lectins in cone berries is the pedicle and collar. In the seeds, agglutinins were localized in the sclerotesta. In the leaves of female ginkgo trees, the hemagglutinating activity of the extracts was higher than in male plants. According to the assessment of the pH activity profile, the highest values are recorded in the alkaline zone, with a pH range of 6.5–8.0. The revealed features can be marker signs for recognizing female and male Ginkgo biloba plants in the pre-generative period of ontogenesis.

Conflict of interest

The authors state that there is no conflict of interest.

References

1. Antoniuk, V. O. (2005). *Lectins and their raw material sources*. Lviv: Danylo Halytsky Lviv National Medical University.
2. Bairak, O. (2017). *My favorite tree: An autobiographical essay*. Poltava: Hovorov S. V. Publisher.
3. Becker, H. G. (2016). *Ginkgo. Weltenbaum. Wanderer zwischen den Zeiten*. Leipzig: Buch Verlag für die Frau GmbH.
4. Dobroskok, V., Dziuba, O., & Palagecha, R. (2009). Mechanism of adaptation of some relict plants during their introduction. *Bulletin of Taras Shevchenko National University of Kyiv: Introduction and Preservation of Plant Diversity*, 25-27, 60–61.
5. Felicitas, B. (2013). *Ginkgo: Der Baum des Lebens. Ein Lesebuch*. Berlin: Insel Verlag.
6. Herhel, Ye. M. (2012). Study of the lectin content in the leaves of plants of the Oleaster family (*Elaeagnaceae* Juss.). *Phytotherapy Journal*, 4, 51–54.
7. Huz, M. M., Hrechanyk, R. M., & Ostudimov, A. O. (2008). Features of *Ginkgo biloba* reproduction *in vitro*. *Scientific Bulletin of UNFU: Collection of Scientific and Technical Works*, 18 (7), 7–16.
8. Kovalov, V. M., Martynov, A. V., Krasnikova, T. O., & Stepanova, S. I. (2001). Plant lectins as biologically active substances for the development of pharmaceuticals. *Physiologically Active Substances*, 1 (31), 74–78.
9. Molotkovskyi, H. Kh., & Butnytskyi, I. M. (1977). Rooting of male and female cuttings of *Ginkgo biloba* and features of protein accumulation in them. In *Proceedings of the VI Congress of the Ukrainian Botanical Society* (pp. 55–56). Kyiv.
10. Ostudimov, A. O., & Huz, M. M. (2010). Features of seed reproduction of *Ginkgo biloba*. *Scientific Bulletin of UNFU: Collection of Scientific and Technical Works*, 20 (11), 8–15.
11. Palamarchuk, O. P., Dzhurenko, N. I., Steshenko, O. M., & Chetvernia, S. O. (2015). Modern possibilities of the complex use of medicinal plants promising for optimizing human adaptive status. In *Agrobiodiversity for improving nutrition, health and life quality* (Part II, pp. 521–526). Nitra.
12. Pospelov, S. V. (2018). Evaluation of the phytohemagglutinins activities of Echinacea species in ontogenesis. *Phytochemistry*, 421–440. https://doi.org/10.1201/9780429426155-20
13. Pospielov, S. V., & Shershova, S. V. (2012). Researches of biological activity of lectin-containing extracts of pale coneflower (*Echinacea pallida* (Nutt.) Nutt.). *Scientific Progress & Innovations*, 2, 47–51. https://doi.org/10.31210/visnyk2012.02.09
14. Pospielov, S. V., Samorodov, V. M., & Pospielova, H D. (2001). Main directions and results of lectin biology research at Poltava State Agricultural Institute. *Bulletin of Poltava State Agricultural Institute*, 4, 42–47.
15. Samorodov, V. M., & Bairak, O. M. (2016). Results of the introduction of *Ginkgo biloba* L. in the Poltava region. *Journal of Native and Alien Plant Studies*, 12, 225–228. https://doi.org/10.37555/.12.2016.173403
16. Sinitsyna, L. V. (2000). The history of the study and distribution of *Ginkgo biloba* L. in Ukraine. *Bulletin of Kyiv University. Biology*, 31, 44–47.
17. Tereshchuk, A. I. (2009). *Ginkgo – the healer*. In *Ukrainian folk traditions* (2nd ed., revised and expanded). Kyiv: Logos Ukraine.
18. Turanska, S. P., Petranovska, A. L., Turov, V. V., & Gorbyk, P. P. (2020). Lectins: obtaining, properties, application in biology and medicine. *Surface*, 12 (27), 289–326. https://doi.org/10.15407/surface.2020.12.289
19. Unseld, S. (2019). *Goethe und der Ginkgo: Ein Baum und ein Gedicht*. Berlin: Insel Verlag.
20. Yudina, Yu. V., Kryklyva, I. O., Ruban, O. A., Demianenko, V. H., & Maslii, Yu. S. (2010). Study of the qualitative and quantitative content of polyphenolic compounds in *Ginkgo biloba* leaves. *Ukrainian Journal of Clinical and Laboratory Medicine*, 5 (2), 49–52.
21. Zuzuk, B. M., Kutsyk, R. V., Tomchuk, Yu., & Darmohrai, R. E. (2001). Ginkgo biloba (*Ginkgo biloba* L.) (An analytical review). *Provisor*, 19, 34–38.

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