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ANATOMIC STRUCTURE OF ONE-YEAR-OLD SHOOTS OF *ACTINIDIA* SPECIES

Objective *— to study the anatomical and morphological structure of one-year-old shoots of the genus Actinidia L. species, which are introduced in Forest-Steppe of Ukraine.*

Material and methods. *The subject of the study were representatives of the genus Actinidia, growing in the collection of the acclimatization department of fruit plants of the M.M. Gryshko National Botanical Garden of the NAS of Ukraine. The studies were carried out in 2016-2017 with the using of generally accepted methods. Anatomical and morphological features of one-year-old shoots of the genus Actinidia species, which are introduced in the conditions of Forest-Steppe of Ukraine, were studied.*

Results. *In A. kolomikta the thick layer of periderm with flattened, suberin-saturated cells is characteristic, which indicate the better protection from abrupt temperature fluctuations, and thus the better resistance to frost and drought. The female plants had significantly thicker periderm than the male ones. A. arguta has the thickest periderm, which, however, contains a lot of air. An indirect indicator of the increased resistance to frost of these taxa is also the most intense starch accumulation in the perimedullar zone, especially in the female plant shoots. According to the structure of the integumentary tissue, the least resistant to the low temperatures has A. macrosperma.*

Conclusions. *The investigation shows that introduced the genus Actinidia species have some features of shoots anatomy, which may be used for prediction of the success of introduction. It was shown, that the high adaptive potential of A. kolomikta is characterized by much flattened suberinized cells, and so this species has the better resistance to frost and drought. The significantly thicker layer of periderm in the shoots of the A. kolomikta female plants compared to male ones is detected. Representatives of A. arguta (A. arguta, sorts Kyivska krupnoplidna and Don Huan) have the thickest layer of periderm filled with air. Their high frost resistance is supported by the most intensive accumulation of starch in the perimedullar zone (especially in the shoots of female plants). The least resistant to the low temperatures, according to anatomy, has A. macrosperma. The data obtained add to the knowledge of the anatomy of the stems of different actinidia species and may be used in the selection of cultivars with increased persistence.*

Key words: *Actinidia* L., species, anatomical and morphological structure, shoots, male and female plant, persistence, spare substances.

The augmentation of species composition of fruit-bearing plants with valuable nutritional and medicinal properties is of great strategic significance for sustainable development. Such plants include species of blueberry, cranberry, sea buckthorn, actinidia and magnolia vine, with high content of biologically active compounds in fruit and vegetative organs. A special place belongs to plants of the genus *Actinidia* Lindl. *(Actinidiaceae* Hutch.).

The genus *Actinidia* contains about 76 species, found in tropical, subtropical and temperate areas of East Asia (China, Japan, the Corean Peninsula and Russian Far East). According to the floristic regionalization, the genus is found in the Eastern Asiatic region of the Holarctic kingdom, with the center of development in South-Western China [4, 8, 9, 10]. The most widespread species of actinidia in the world is *A. deliciosa* (A.Chev.) C.F. Liang ex A.R. Ferg., known as kiwi, one of the most important industrial fruit cultures in the world. Due to low frost resistance, the plants of this species may grow only in tropical or subtropical climate.

Lately, considerable attention of scientists and practitioners of many countries of the world (the USA, Poland, Italy, France, Belgium, New Zealand) attracts other more winter-resistant species of actinidia, in particular *A. arguta* (Siebold et Zucc.) Planch. ex Miq. The first industrial plantations

of this crop are founded there, selection work with this plants is continuing, the cultivation and storage technologies of actinidia fruits are developing. Significant contribution to the introduction of certain *Actinidia* species in culture has been made in Ukraine, particularly, in M.M. Gryshko National Botanical Garden of the NAS of Ukraine (NBG), where introductory studies of actinidia are carried out more than 60 years. Today, the collection of *Actinidia* of the NBG is one of the richest in Ukraine. It is represented by six species, namely *A. kolomikta* (Rupr. et Maxim.) Maxim., *A.arguta* (Siebold et Zucc.) Planch. ex Miq., *A. purpurea* Rehd., *A. polygama* Siebold et Zucc., *A. chinensis* Planch., *A. macrosperma* C.F.Liang. As a result of the breeding work in the NBG, 19 high-yielding cultivars are produced that are promising for widespread introduction to the horticulture [12].

The success of the introduction depends greatly on the genetically determined properties of plants that determine the level of their endurance to the environment. At the same time, when plants are transferred to new conditions, the adaptive potential of species is realized through some physiological-biochemical and anatomical and morphological alterations. Therefore, the study of the anatomical structure of individual organs of plants is extremely important for assessing the impact of environmental factors and identifying the adaptation of plants to the natural environment. The anatomical and histochemical studies of plants to determine their level of resistance to the new climatic conditions are known, which indicate that the frost resistance of woody plants depends on their anatomical, morphological and physiological characteristics, in particular on the intensity of accumulation of spare substances (sugars, starches) and from the age of plants [2, 5, 7]. Known works are devoted to the investigation of anatomical structure of vegetative organs of lianes from the *Actinidiaceae* family [1, 3, 10, 11].

The value of anatomical features of one-year-old shoots as an integrated system of tissues for diagnostic is given in some works on woody plant anatomy [7]. One of the way of prediction of the efficiency of acclimatization is the investigation of the plant anatomical structure.

Objective — to study the anatomical and morphological structure of the one-year-old shoots of introduced species (*A. arguta, A. kolomikta, A. polygama* and *A. macrosperma*) regarding their frost and drought resistance.

The internal structure of vegetative organs of woody plants is of great interest not only from the practical point of view, but in the light of theoretical questions of plant physiology and some problems of systematic and phylogeny.

Material and methods

Objects of the study were four species of the genera *Actinidia*: *A. kolomikta, A. arguta, A. polygama, A. macrosperma* and two sorts selected in NBG — Kyivska krupnoplidna (female plants, F) and Don Huan (male plants, M).

For anatomical research the samples were taken in early March and in late April. We used the middle part of one-year-old shoots of female and male specimens.

The samples were fixed in FAA and sectioned into 10—15 μm thick slabs using freezing microtome, then stained by phloroglucin and I2—KI solution to detect lignified structures and starch, respectively [6]. The measurements were done using Image J program and XSP-146TR microscope.

The data were statistically processed using Statistica 8. The significance of results was determined using Student's t-criterion. The pictures were taken with digital camera Canon Power Shot A630.

Results and discussion

The vascular system of actinidia stem belongs to the transient woody type in plant evolution [2, 7]. Stems of studied species have similar structure. The primary covering of the stem, the epidermis is unicellular, covered with a thin cuticle and lacking chloroplasts. Under the epidermis is the periderm, made up by thin-walled cells stacked into dense layers. Beneath the periderm is the medullar collenchyma and the small cells of cortex parenchyma, filled with chloroplasts. The cortex parenchyma and phloem are separated by bast fibers, with cells distinguished by thickened walls. A well-de-

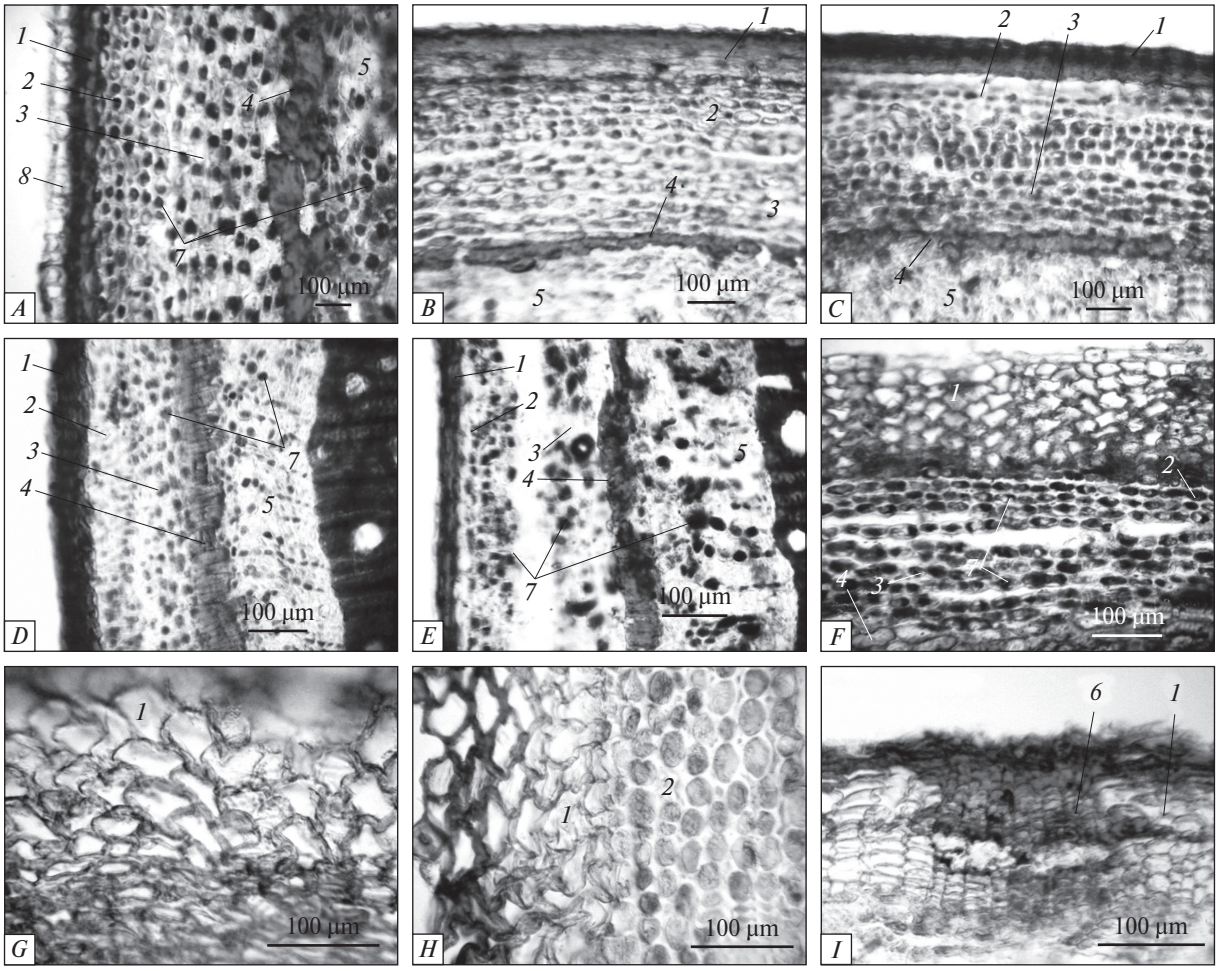

Fig. 1. Cross section of one-year-old shoots: *A — A. macrosperma* (F); *B — A. kolomikta* (F); *C — A. kolomikta* (M); *D — A. polygama* (F); *E — A. polygama* (M); *F* — Kyivska krupnoplidna (F); *G* — Don Huan (M); *H — A. arguta* (F); *I — A. arguta* (M): *1*— periderm; *2* — collenchyma; *3* — parenchyma; *4* — bast fibers; *5* — phloem; *6* — lenticels; *7* — starch; *8* — residual epidermis

veloped primary phloem is represented by a ring of woody bast fibers, the middle consists of large rounded or polygonal parenchyma cells, which on the periphery make up the perimedullar zone (Fig. 1). With age, medullar cells die off, and the shoot is hollowed out. Despite significant similarity of the internal structure of one-year-old shoots of studied species we saw some differences in each of them and in specimens of different sex.

At the beginning of vegetation, the species main difference is the degree of periderm development. The stem of *A. macrosperma* is covered by an almost whole one-layer epidermis, beneath which the periderm has only 1-2 suberinized layers (Fig. 1, *A*). The periderms of *A. polygama* and *A. kolomikta* consist respectively of 6—7 and 7—8 layers of strongly flattened, suberin-filled cells (Fig. 1, *B—E*). Female specimens of these species have somewhat thicker periderm. Thus, a shoot of 'Kyivska krupnoplidna' has 5—7 layers of deformed, air-filled cells with tortuous suberin-filled radial walls. In contrast, only the few upper layers of periderm are suberinized in male specimens ('Don Huan') (Fig. 1, *F, G*). The shoot of female specimens of *A. arguta* has a similar structure of periderm with several outer layers (5—6) suberinized and starting to lignify. In this species, the cork of males is somewhat thicker than that of females, there are

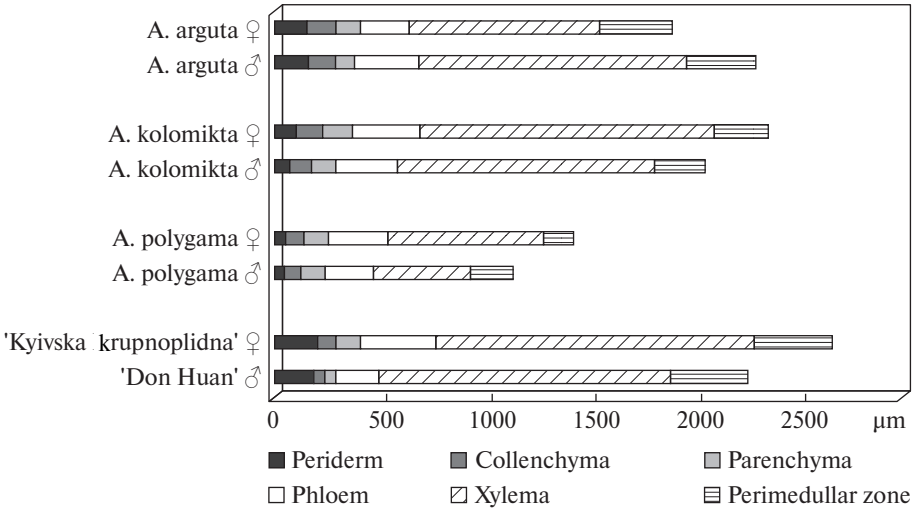

Fig. 2. Histogram of anatomical parameters of one-year-old shoots: of *Actinidia* species and sorts

up to twelve layers (Fig. 1, *H, I*). In all studied species, one-year-old shoots had a lot of lenticels.

Beneath periderm is lamellar collenchyma. It has the most layers in the shoots of *A. arguta* and in the studied sorts (7—8 in female plants, 5 in male plants), the least in *A. polygama*, 4—5 layers in both male and female specimens. There is no sharp difference between collenchyma and parenchyma. In all studied species, there are 5—6 layers of parenchyma cells with thickened walls and well-developed intercellular spaces. Notably, in early March, collenchyma and parenchyma in the male and female plants of *A. arguta* and 'Don Huan' have no starch reserves. In 'Kyivska krupnoplidna' and the female *A. macrosperma* however there are a lot of starch grains in these tissues. As for *A. polygama*, there is a lot of starch inclusions in collenchyma and parenchyma of both male and female plants.

Also, in March a lot of starch inclusions are seen in the phloem of *A. polygama,* 'Kyivska krupnoplidna' and *A. macrosperma*. In other species, no starch was found in phloem. In the medullar rays of all species except *A. kolomikta* there were starch grains. The perimedullar zone (including elements of the primary xylem) of female plants of all studied species except *A. kolomikta* had large reserves of starch. Meanwhile, in male plants the

Table. **Morphometrical parameters (μm) of one-year-old shoots of the genus *Actinidia* species and sorts**

Species / sort	Periderm	Collenchyma	Parenchyma	Phloem	Xylem	PMZ
A. arguta (F)	159 ± 17	139 ± 9	112 ± 12	235 ± 69	916 ± 37	330 ± 46
A. arguta (M)	169 ± 8	118 ± 7 *	101 ± 22	295 ± 28 *	1276 ± 30 *	328 ± 18
A. kolomikta (F)	109 ± 25	120 ± 20	139 ± 27	333 ± 13	1394 ± 45	249 ± 41
A. kolomikta (M)	71 ± 22 *	107 ± 9	107 ± 23 *	305 ± 88	1230 ± 73 *	227 ± 16
A. polygama (F)	57 ± 9	93 ± 13	115 ± 23	282 ± 12	735 ± 116	134 ± 65
A. polygama (M)	49 ± 15	93 ± 8	101 ± 24	232 ± 16 *	467 ± 69 *	194 ± 90
Kyivska krupnoplidna (F)	202 ± 13	104 ± 8	112 ± 12	353 ± 17	1509 ± 44	377 ± 25
Don Huan (M)	186 ± 20	60 ± 13 *	41 ± 9 *	213 ± 43 *	1378 ± 96 *	372 ± 17
A. macrosperma (F)	74 ± 9	96 ± 7	139 ± 14	238 ± 23	913 ± 59	235 ± 56

N o t e: PMZ is perimedullar zone; * — statistically significant ($p < 0.05$) differences compared to female plants.

starch was absent in this region only in 'Don Huan'. Interestingly, the perimedullar zone is thickest in *A. arguta* and 'Kyivska krupnoplidna', which combined with the presence of starch indicates larger starch reserves (Fig. 2). The medullar parenchyma has no starch inclusions. The maximal amounts of medullar parenchyma were seen in the stem of *A. polygama*.

As can be seen from Table, the male plants of all species except *A. arguta* either have significantly thinner periderm, collenchyma, parenchyma, phloem, xylem and perimedullar space (except *A. polygama*) or a tendency to decrease compared to female ones.

As it was in phloem and perimedullar zone where starch was found most intensively, the thickness of these zones can be one of the criteria to differentiate males and females. Male plants of *A. arguta* have better developed phloem and xylem compared to females. Sizes of perimedullar zone do not statistically significantly differ between male and female plants.

Thus, according to the structure of outer layers, the most similar were females of *A. arguta*, 'Kyivska krupnoplidna' and 'Don Huan'. Significantly thicker periderm was found in female plants of *A. kolomikta* compared to male plants indicating better frost resistance of the former. Also in *A. kolomikta* the thick periderm layer consisting of much flattened suberinized cells indicates the better defense against abrupt temperature fluctuations and less water losses, and so better frost and drought resistance. In *A. arguta* and sorts Kyivska krupnoplidna and Don Huan, the thickest periderm is found (Table), however, it is filled with air. The least resistant to the action of low temperatures, by tissue structure, was *A. macrosperma*. An indirect indicator of better frost resistance of above taxa is the large deposits of starch in the perimedullar zone, especially seen in shoots of female plants.

Conclusions

Introduced the genus *Actinidia* species have some features of anatomy of shoots that may be used for prediction of the success of introduction. It was shown, that the high adaptive potential of *A. kolomikta* is connected with flattened suberinized cells, which provide the better protection of plants from frost and drought. The significantly thicker layer of periderm in the shoots of the A. kolomikta female plants compared to male ones is detected.

Representatives of *A. arguta* (*A. arguta*, sorts Kyivska krupnoplidna and Don Huan) have the thickest layer of periderm filled with air. Their high frost resistance is supported by the most intensive accumulation of starch in the perimedullar zone (especially in shoots of female plants). The least resistant to low temperatures, according to anatomy, has *A. macrosperma*.

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АНАТОМІЧНА СТРУКТУРА ОДНОРІЧНИХ ПАГОНІВ ВИДІВ АКТИНІДІЇ

Мета — вивчити анатомо-морфологічну структуру однорічних пагонів видів роду *Actinidia* L., інтродуктованих у Лісостепу України.

Матеріал та методи. Предметом дослідження були представники роду *Actinidia* L., які зростають у колекції відділу акліматизації плодових рослин Національного ботанічного саду імені М.М. Гришка НАН України. Дослідження проведено в 2016—2017 рр. з використанням загальноприйнятих методів. Вивчено анатомо-морфологічні особливості однорічних пагонів видів роду *Actinidia*, інтродукованих у Лісостепу України.

Результати. Для *A. kolomikta* характерний товстий шар перидерми із дуже сплющених, насичених суберином клітин, що свідчить про кращий захист рослин від різких коливань температури, і, відповідно, про кращу морозо- та посухостійкість. Виявлено, що перидерма пагонів жіночих особин *A. kolomikta* порівняно з чоловічими була товщою. У представників *A. arguta* найтовща перидерма, яка насичена повітрям. Про кращу морозостійкість зазначених таксонів опосередковано може свідчити найбільш інтенсивне накопичення крохмалю у перимедулярній зоні, особливо в пагонах жіночих рослин. Найменш стійкою до дії низьких температур за будовою покривної тканини виявилася *A. macrosperma*.

Висновки. Встановлено, що інтродуковані види актинідії мають особливості анатомічної структури пагонів, які можуть бути використані для прогнозування успішності інтродукції. Показано, що анатомічна будова однорічних пагонів *A. kolomikta* вирізняється дуже сплюснутими суберинізованими клітинами, що забезпечує високу стійкість рослин до несприятливих чинників довкілля, зокрема до низьких температур і посухи. Виявлено значно товщу перидерму у пагонах жіночих рослин *A. kolomikta* порівняно з чоловічими, що корелює з їх високою морозостійкістю. Однорічні

пагони представників *A. arguta* (*A. arguta*, сорти Київська крупноплідна та Дон Жуан) мають найтовщий шар перидерми, наповнений повітрям. Їх висока морозостійкість пов'язана з найбільш інтенсивним накопиченням крохмалю в перимедулярній зоні (особливо в пагонах жіночих рослин). Найменш стійким до низьких температур є *A. macrosperma*. Отримані дані розширюють уявлення про особливості анатомічної будови пагонів видів актинідії і можуть бути використані в селекції для створення сортів з підвищеною стійкістю.

Ключові слова: *Actinidia* L., вид, анатомо-морфологічна будова, пагони, чоловічі та жіночі рослини, стійкість, запасні речовини.

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АНАТОМИЧЕСКАЯ СТРУКТУРА ОДНОЛЕТНИХ ПОБЕГОВ ВИДОВ АКТИНИДИИ

Цель — изучить анатомо-морфологическую структуру однолетних побегов видов рода *Actinidia* L., интродукцированных в Лесостепи Украины.

Материалы и методы. Предметом исследования были представители рода *Actinidia* L., произрастающие в коллекции отдела акклиматизации плодовых растений Национального ботанического сада имени Н.Н. Гришко НАН Украины. Исследования проведены в 2016—2017 гг. с использованием общепринятых методов. Изучены анатомо-морфологические характеристики однолетних побегов видов рода *Actinidia,* интродукцированных в Лесостепи Украины.

Результаты. Для *A. kolomikta* характерен толстый слой перидермы с сильно сплющенными насыщенными суберином клетками, что указывает на лучшую защиту растений от резких колебаний температуры, и, соответственно, на лучшую морозо- и засухоустойчивость. Выявлено, что перидерма побегов женских особей *A. kolomikta* по сравнению с мужскими была более толстой. У представителей *A. arguta* самая толстая перидерма, которая содержит много воздуха. Про лучшую морозостойкость упомянутых таксонов косвенно может свидетельствовать наиболее интенсивное накопление крахмала в перимедуллярной зоне, особенно в побегах женских растений. Наименее устойчивой к действию низких температур по строению покровной ткани оказалась *A. macrosperma*.

Выводы. Установлено, что интродуцированные виды актинидии имеют особенности анатомического строения побегов, которые могут быть использованы для прогнозирования успешности интродукции. Показано, что анатомическое строение однолетних побегов *A. kolomikta* характеризуется сильно сплюснутыми суберинизированными клетками, что обеспечивает высокую стойкость растений к неблагоприятным условиям окружающей среды, в частности, к низким температурам и засухе. Обнаружена значительно более толстая перидерма в побегах женских особей *A. kolomikta* по сравнению с мужскими, что указывает на их более высокую устойчивость к морозу. Однолетние побеги представителей *A. arguta* (*A. arguta*, сорта Киевская крупноплодная и Дон Жуан) имеют самый толстый слой перидермы, наполненный воздухом. Их высокая морозостойкость связана с наиболее интенсивным накоплением крахмала в перимедулярной зоне побега (особенно в побегах женских растений). Наименее устойчивым к низким температурам является *A. macrosperma*. Полученные данные расширяют представления об особенностях анатомического строения побега видов актинидии и могут быть использованы в селекции для создания сортов с повышенной устойчивостью.

Ключевые слова: *Actinidia* L., вид, анатомо-морфологическое строение, побеги, мужские и женские растения, устойчивость, запасные вещества.