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Morphology and anatomy of the root system of new potato cultivars Part II. Root anatomy*

Morfologia i anatomia systemu korzeniowego nowych odmian ziemniaka Część II. Anatomia korzeni

The comparison of primary adventitious roots structure was done in a survey study of 17 *Solanum tuberosum* cultivars differently tolerant to drought by means of microscopic methods. Cell wall autofluorescence combined with aniline blue treatment of handmade sections was shown to be a convenient method for fast examination of large amount of plant material. Secondary structure limited to the formation of vascular cambium and its derivative tissues was found in all the cultivars examined. Anatomical differences were found between cultivars, individual plants and individual roots, and they were most evident in regard to the extent of secondary structure formation and number of primary vascular tissues strands. By means of image-analysis method, cross-section areas of the root and xylem were measured in 16 cultivars, and by means of statistical analysis significant differences were found between cultivars in regard to these traits.

Key words: endodermis, exodermis, root anatomy, secondary xylem.

W pilotażowych obserwacjach wykonano przy pomocy metod mikroskopowych porównanie budowy pierwszorzędowych korzeni przybyszowych u 17 odmian *Solanum tuberosum* o zróżnicowanej tolerancji stresu suszy. Wykazano przydatność obserwacji, na skrawkach ręcznych, autofluorescencji ścian komórkowych w zestawieniu z traktowaniem błękitem aniliny do szybkiego badania dużej ilości materiału roślinnego. U wszystkich badanych odmian stwierdzono występowanie budowy wtórnej ograniczonej do wytworzenia kambium waskularnego i tkanek pochodnych. Różnice w budowie anatomicznej stwierdzono w odniesieniu do odmian, poszczególnych roślin i poszczególnych korzeni, przy czym dotyczyły one głównie stopnia zaawansowania budowy wtórnej i liczby pasm drewna pierwotnego. Przy pomocy metody analizy obrazu zmierzono pola przekroju

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poprzecznego korzenia i drewna, a analiza statystyczna wykonana dla 16 odmian wykazała, że różniły się one istotnie pod względem wymienionych cech.

Słowa kluczowe: anatomia korzenia, egzoderma, endoderma, ksylem wtórny

INTRODUCTION

Potato (*Solanum tuberosum* L.) is a species native to frost-free temperate climate, and it is sensitive to temperatures exceeding 20°C (Struik et al., 1989 a; 1989 b; Van Dam et al., 1996), which result in inhibited tuberization and reduction of photoassimilate partitioning to tubers (Ewing, 1981; Krauss and Marschner, 1984; Haynes et al., 1989; Lafta and Lorenzen, 1995; Van Dam et al., 1996; Rykaczewska, 2013, 2015 a). In Poland, due to the increasing occurrence of periods of high temperatures and drought resulting in potato production losses, studies of potato tolerance to these abiotic stresses were undertaken in the Department of Potato Agronomy of the Plant Breeding and Acclimatization Institute — NRI. The studies focused, among others, on assessments of the morpho-anatomical variability of the potato root system and seeking of a possible connection with abiotic stress tolerance. The rather shallow fibrous root system of potato plants grown from tubers is built from branching adventitious roots formed on sprouts (Cutter, 1992; Levy and Coleman, 2015). Currently, limited ability of potato root system to efficiently transport water is considered a major cause of drought sensitivity in this crop species (Chaves et al., 2003). Therefore, in the search for or development of potato cultivars better-adjusted to drought and drought-associated high temperatures, superior root architecture, root growth rate, morphology and anatomy are of interest.

The present work was intended as an exploratory study of several potato cultivars in regard to their root anatomical features that may be correlated with stress tolerance. Since statistical analysis of some anatomical traits was planned, another goal of the study was to design a simple microscopy protocol to make possible fast examination of a large number of sections, providing still robust anatomical data and digital images of quality suitable for measurements.

MATERIALS AND METHODS

Plant material

For the anatomical study the following 17 cultivars were taken: Denar, Lord, Justa, Miłek (very early), Aruba, Bila, Etola, Gwiazda, Hubal, Michalina (early), Etiuda, Finezja, Gandawa, Kuba, Oberon, Stasia and Tetyda (medium early). The cultivars, as well as growth terms, growth conditions and culture termination were the same as in Part I. of this work (Rykaczewska, 2015 b).

Among these cultivars, Aruba, Etola, Finezja and Tetyda were studied earlier. Cultivar Tetyda was proven as the most tolerant to high temperature-drought stress acting on the plants during the growing season, which manifested in a) relatively small decrease in the total yield or tuber size, b) low level of tuber deformations, and c) lack of tendency for

sprouting in the soil before harvest (Rykaczewska, 2015 a). In the same earlier experiment, similar features were found for Finezja, while Aruba was most susceptible.

Microscopy — preparation of sections

Plant material for microscopy was collected at the day of culture completion. From every cultivar, three plants were taken, and from every plant, 2 cm-long fragments were cut out of the basal part (i.e. the oldest one) of two of the primary (1st range) adventitious roots, 5 cm at most from the root base. The following fixation and staining procedures were done according to Broda (1971). Root samples were immediately placed in FAA fixative (38% formalin: 50% ethanol: glacial acetic acid, in 0.5:9.0:0.5 volume ratio) for several days at the ambient temperature. Next, the samples were rinsed in 70% ethanol until the smell of acetic acid was not perceivable, and stored in the refrigerator until sectioning.

Cross-sections were cut by hand, using razor blade, from proximal and distal end of every root sample placed between halves of *Sambucus* pith. Sections were immediately immersed in a drop of aniline blue (0.5% solution in 0.1M K_3PO_4, pH >8.5), distal and proximal sections on separate slides, and examined using a light microscope (below). Next, cover glass was carefully removed, aniline blue was dried with tissue paper, and the sections were covered with a drop of Sudan III saturated solution in isopropanol, a lipophilic orange stain for suberin detection.

Microscopy — examination of sections

Sections were examined using bright-field optics, polarization optics (for identification of crystal sand idioblasts) or UV-induced fluorescence of Provis AX (Olympus) microscope. For fluorescence, NU filter set (BP360-370 nm excitation filter, BA420 nm barrier filter, DM400 nm dichroic mirror) was used. Fluorescence technique revealed the autofluorescence of the cell walls (esp. suberized or lignified) and the fluorescence of aniline blue bound to callose. The best cross-sections (i.e. suitably thin and non-oblique) were selected and saved using a dedicated digital camera DP50 (Olympus). Digital images of 2776 × 2074 pixel resolution were saved as tiff files and processed using Photoshop CS6 (Adobe) software by means of non-destructive tools. All adjustments were done on the whole area of the image. For every magnification used, image of linear micrometric scale was saved identically as described, to provide a reference images for magnification scale bars.

Image analysis

The area measurements were done for a) cross-sections of the whole root and b) xylem. Images of root cross-section saved at 4x objective were selected from the whole image by means of tool sequence "Magic wand"/ "Reverse" in Photoshop CS6 (Adobe) software and painted uniformly black using "Pencil". The background noise was removed by the tool sequence: "Magic wand" (marking of the root)/ "Reverse"/ "Delete". The resulting image was copied, pasted into an image of standard 4000 × 4000 pixel size, saved as jpg file and used for measurements.

Images of black dots printed on a micrometric slide were processed identically. The dots had a known diameter, 0.6 mm (area 0.2827 mm^2) or 0.15 mm (area 0.0177 mm^2) for

objective 4x or 10x, respectively, and they were later used as a control of the measurement accuracy.

Images of autofluorescing xylem cross-section saved at 10x objective were selected from the whole image using the following tool sequence: "Magic wand" at the outer limit of xylem to mark dark tissues/ "delete"/ "Magic wand" at the empty area/ "Reverse"/ "Pencil" to paint xylem uniformly black/ "Magic wand" to mark xylem/ "Reverse"/ "delete" to remove any non-xylem part of the original image. The resulting image was copied, pasted into an image of standard 4000 x 4000 pixel size, saved as jpg file and used for measurements.

For software scale setting, two images of a bar 2000µm or 600µm long were prepared using images of linear micrometric scale mentioned above.

For measurements, the Fiji (Open Source, http://fiji.sc/Fiji) software was used. Images of cross-sections of a cultivar roots, as well as the respective bar and dot images were stacked, and on the bar image the scale was marked using "Straight" tool and set using "Set Scale" tool. Next, images were turned to binary with "background" set as "light" and automatic threshold calculation for every image of the stack. Using "Analyze Particles"/ "Size" tool the cross-section areas of the images were calculated for objects within a set size range (above 4µm^2) and the results were displayed as a MSExcel (Microsoft) file (from the results, the area of the bar was deleted).

Statistical analysis

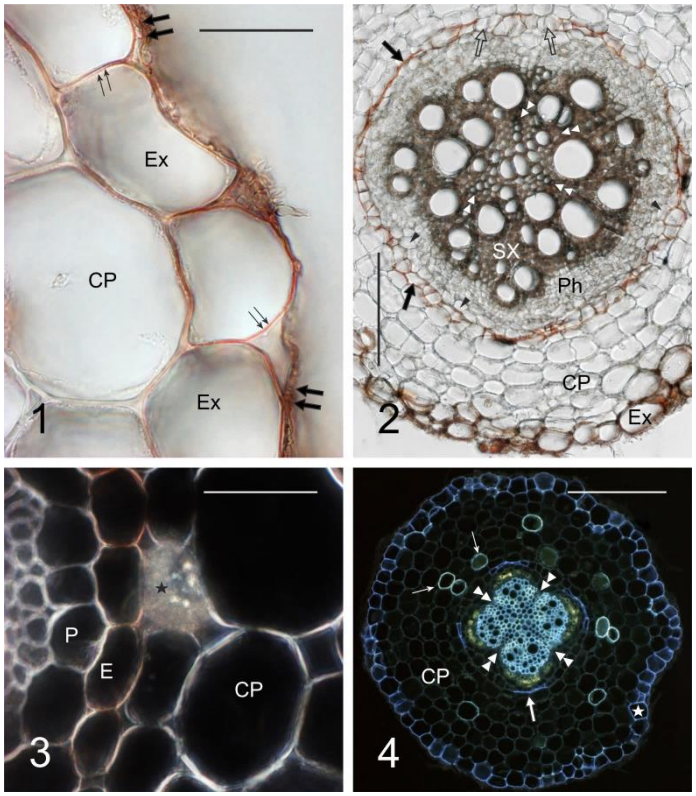
Cultivar Miłek was excluded from statistical analysis due to missing data (owing to rot, some roots disintegrated during sectioning). Because of nested observations within a plant and a root within plant, the influence of cultivar on root (or xylem) cross-section area was analyzed with a linear mixed-effect model (Pinheiro and Bates, 2000). The fitted model's diagnosis was done by graphical methods (Quinn and Keough, 2002). Since the cultivars differed in the root (or xylem) cross-section area ($P<0.001$), multiple comparisons were made by analyzing Tukey's all-pair comparisons (Hothorn et al. 2008). The analysis was conducted in *R* (R Team 2015), with the help of *nlme* (Pinheiro et al., 2013) and *multcomp* (Hothorn et al. 2008) packages.

RESULTS

Analysis of potato root structure by means of microscopy

In the basal part, adventitious roots were covered with the remnants of collapsed and degraded rhizodermis, which had no autofluorescence (Fig. 1). Within this layer, numerous bacterial and fungal hyphae were always present (not shown).

The protective function fulfilled the outer cells of primary cortex - the single layer of tightly adjoining exodermal cells (Fig. 1). Their cell walls were suberized, which was visualized by Sudan staining and whitish autofluorescence following UV excitation. In numerous roots, suberization extended into anticlinal walls of adjoining cells of primary cortex parenchyma. The latter tissue was composed of several layers of vacuolated cells (Fig. 2) that exhibited very low autofluorescence.



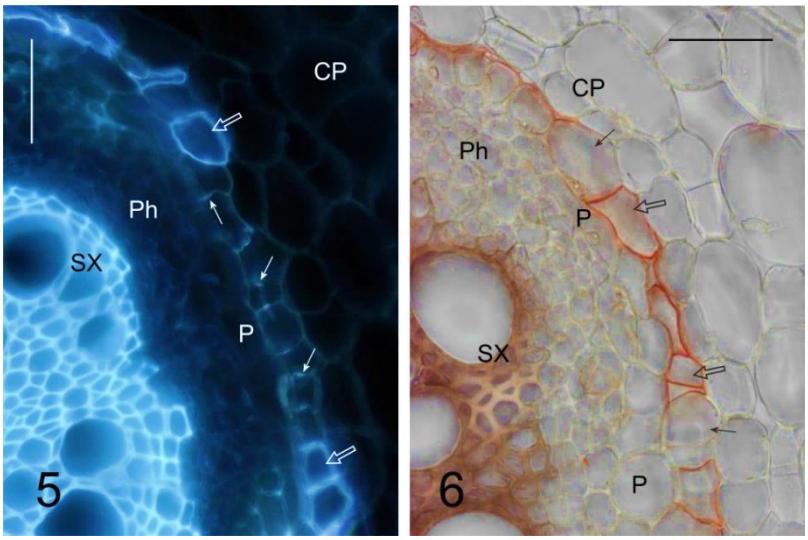
Explanations: CP - primary cortex parenchyma, E - endodermis, Ex - exodermis, P - pericycle, Ph - phloem, double arrows - degraded rhizodermis, double slim arrows - Sudan-stained suberin layer, open arrows - endodermal cells in the 1st developmental stage, arrows - endodermal cells in the 2nd developmental stage, asterisk - crystal sand in the idioblast, white asterisk - exodermis, arrowheads - pericycle, double arrowheads - protoxylem strands, white slim arrows - autofluorescence of lignified walls of cortical idioblasts

Fig. 1-4. Cross-sections of primary adventitious roots of *Solanum tuberosum* cultivars - primary cortex structure. Fig. 1. Exodermis and remnants of degraded rhizodermis, cultivar Etola, Sudan staining. Bar 0.04 mm. Fig. 2. Non-synchronous differentiation of endodermis in a tetrarch root of cultivar Tetyda, Sudan staining. Bar 0.14 mm. Fig. 3. Crystal sand idioblast, cultivar Aruba, polarization contrast. Bar 0.04 mm. Fig. 4. Autofluorescent lignified cortical idioblasts of unknown function in a tetrarch root of cultivar Aruba, UV-excited autofluorescence and aniline blue fluorescence. Bar 0.33 mm

Objaśnienia: CP - miękisz kory pierwotnej, E - endoderma, Ex - egzoderma, P - perycykl, Ph - floem, podwójne strzałki - zdegradowana ryzoderma, podwójne wąskie strzałki - warstwa suberynowa zabarwiona Sudanem, puste strzałki - komórki endodermy w 1. stadium rozwojowym, strzałki - komórki endodermy w 2. stadium rozwojowym, gwiazdka - piasek krystaliczny w idioblaście, gwiazdka biała - egzoderma, groty strzałek - perycykl, groty strzałek podwójne - pasma protoksylemu, strzałki smukłe białe - autofluorescencja zdrewniałych ścian idioblastów korowych

Rys. 1-4. Przekrój poprzeczny pierwszorzędowych korzeni przybyszowych u odmian *Solanum tuberosum* - budowa kory pierwotnej. Fot. 1. Egzoderma i pozostałości zdegradowanej ryzodermy, odmiana Etola, barwienie Sudanem. Odcinek skalujący 0,04 mm. Fot. 2. Niesynchroniczne różnicowane endodermy w korzeniu tetrarchicznym, odmiana Tetyda, barwienie Sudanem. Odcinek skalujący 0,14 mm. Fot. 3. Idioblast z piaskiem krystalicznym, odmiana Aruba, kontrast polaryzacyjny. Odcinek skalujący 0,04mm. Fot. 4. Autofluoryzujące zdrewniałe idioblasty korowe o nieznanej funkcji w korzeniu tetrarchicznym u odmiany Aruba, autofluorescencja wzbudzona promieniowaniem UV i fluorescencja błękitu aniliny. Odcinek skalujący 0,33 mm

In roots with considerable secondary growth within stele, cortical parenchyma cells were divided anticlinally, thus compensating the circumferential growth of the root. Idioblasts filled with crystal sand occurred in the cortical parenchyma of all cultivars (Fig. 3). In roots of cultivars Justa, Miłek and Aruba, cortical parenchyma contained additional idioblasts of unknown function, which were characterized with strongly autofluorescent (but not Sudan-positive) and uniformly thickened cell walls (Fig. 4). In some roots, inner parenchyma layers were arranged in radial files in cross sections, and the files were consistent with the location of endodermal cells (not shown). Generally, such arrangement results from common origin: the parenchyma file and the adjacent endodermal cell originate from the same initial within the root apical meristem. Surprisingly, this feature was inconsistent for individual plants and cultivars, suggesting variability in functioning of potato root apical meristem.



Explanations: CP - primary cortex parenchyma, P - pericycle, Ph - phloem, SX - secondary xylem, arrows - endodermal cells in the 1st developmental stage (arrows point to Casparian strip in Fig. 5), open arrows - endodermal cells in the 2nd developmental stage.

Fig. 5-6. Cross-sections of primary adventitious roots of *Solanum tuberosum* cultivars - non-synchronous differentiation of endodermis. Fig. 5. The 1st stage endodermal cells with autofluorescent Casparian strips and 2nd stage cells with autofluorescent suberin lamellae, cultivar Lord, UV-excited autofluorescence. Bar 0.03mm. Fig. 6. The 1st stage endodermal cells with non-stained walls and 2nd stage cells with suberin lamellae stained with Sudan, cultivar Tetyda. Bar 0.03 mm.

Objaśnienia: CP - miękisz kory pierwotnej, P - perycykl, Ph - floem, SX - ksylem wtórny, strzałki - komórki endodermy w 1. stadium rozwojowym (na Fot. 5. strzałki wskazują pasemko Caspary'ego), strzałki puste - komórki endodermy w 2. stadium rozwojowym.

Fot. 5-6. Przekrój poprzeczny pierwszorzędowych korzeni przybyszowych u odmian *Solanum tuberosum* - niesynchroniczne różnicowanie endodermy. Fot. 5. Komórki endodermy w 1. stadium różnicowania z autofluorescencją pasemka Caspary'ego i komórki w 2. stadium z autofluoryzującą lamellą suberynową, odmiana Lord, autofluorescencja wzbudzona promieniowaniem UV. Odcinek skalujący 0,03mm. Fot. 6. Komórki endodermy w 1. stadium różnicowania ze ścianami niezabarwionymi i komórki w 2. stadium z lamellą suberynową zabarwioną Sudanem, odmiana Tetyda. Odcinek skalujący 0,03 mm.

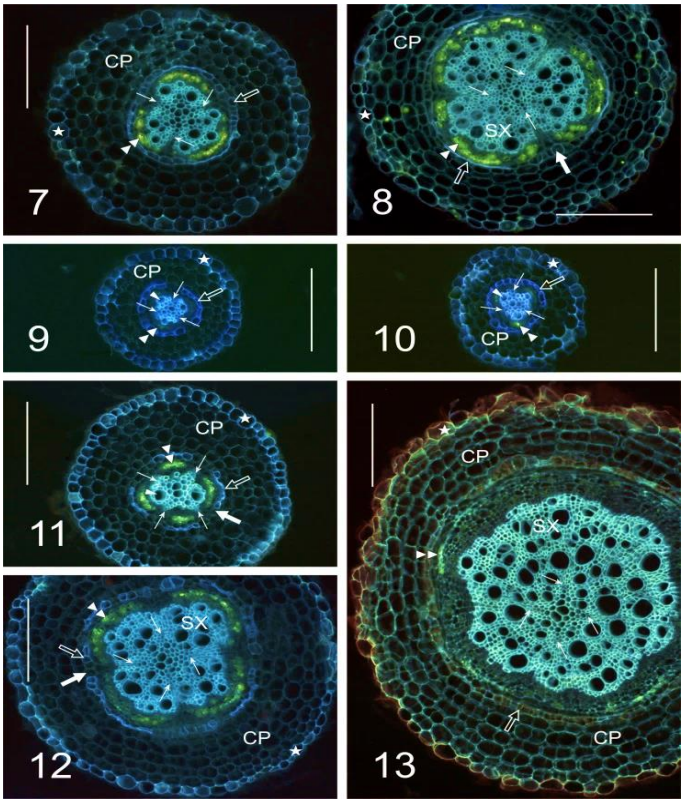
Endodermis, the innermost tissue of primary cortex, was monolayered (Fig. 2), as typical for roots. Endodermal cell walls were strongly autofluorescent (Fig. 5) and easily stainable with Sudan III (Fig. 2, 6), similarly to exodermal ones. In contrast to the latter, the fluorescence and orange tinge of endodermal cells were circumferentially diverse, and they were most intense in endodermal cells opposite primary phloem. Such image exemplified the asynchronous differentiation of endodermis. The differentiation stage reached by endodermis varied between cultivars. In most cultivars, even after secondary vascular tissues were formed, cells opposite primary xylem poles had just Casparian strips (primary stage), while the suberin lamella (secondary stage) was already deposited in cells facing phloem poles (Fig. 5). In some sections, secondary stage cells formed a closed ring, but in these roots the cell wall autofluorescence was more intense opposite phloem (not shown).

Within the stele, secondary structure differentiated limited to the formation, to the extent variable between cultivars, of cambium and secondary vascular tissues. Since no phloem fibers occurred in both primary and secondary phloem, these tissues showed scarcely any autofluorescence. Therefore, location of primary phloem was determined by the bright green fluorescence of callose-bound aniline blue, and it was very helpful for determining the number and location of primary xylem strands that were often poorly discernible from the secondary xylem due to the total lignification of both regions, as well as the abundance of xylem fibers (live in the roots examined) and absence of xylem parenchyma both within primary and secondary xylem.

Generally, potato primary adventitious roots examined in this work were triarch (Fig. 7-10), tetrarch (Fig. 11-13), pentarch (Fig. 14-15) or sextarch (Fig. 16). Most roots examined were tetrarch, and the thickest roots were found among the tetrarch ones (Fig. 13, Cv. Denar). The sextarch roots were rare. Often, together with the primary roots the lateral ones were unintentionally sectioned, and these were usually diarch, occasionally triarch, with very thin cortical parenchyma (Fig. 17).

The extent of secondary structure varied between roots of different cultivars (compare Fig. 7, 8 vs Fig. 9, 10), of the same cultivar (compare Fig. 7, 8 vs Fig. 11, 12) and the same plant (not shown), as well as the number of primary xylem (or phloem) strands. In the center of stele, a single-to-several metaxylem vessel(-s) differentiated (e.g. Fig. 11, 12), and stelar parenchyma was never observed in this location.

The cultivars differed mainly in the thickness of secondary xylem. In the thinnest roots (Fig. 9, 10, Cv. Tetyda), primary xylem was still in contact with pericycle and the very first vessels of the secondary xylem were differentiated.

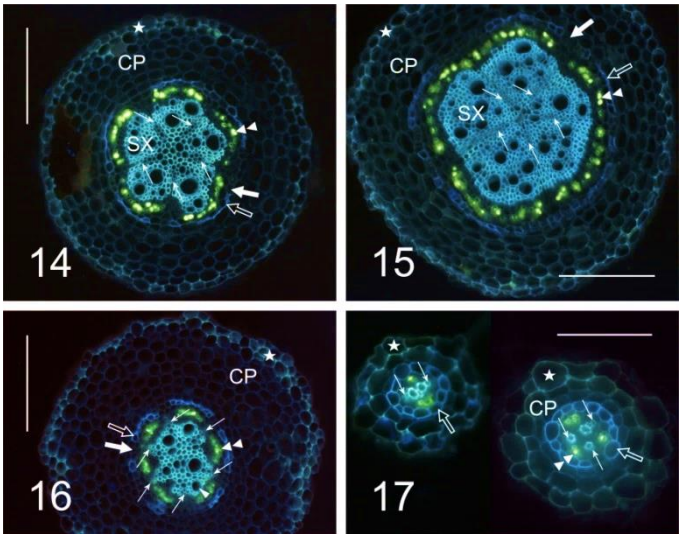


Explanations: CP - primary cortex parenchyma, SX - secondary xylem, asterisks - exodermis, arrows - endodermal cells in the 1st developmental stage, open arrows - endodermal cells in the 2nd developmental stage, slim arrows - protoxylem strands, double arrowheads - fluorescence of callose-bound aniline blue in phloem, arrowheads - the first vessels in the youngest secondary xylem

Fig. 7-13. Cross-sections of primary adventitious roots in *Solanum tuberosum* cultivars - anatomical differences in root thickness, number of primary xylem strands and secondary structure extent. Fig. 7 and 8. Triarch roots of small or moderate thickness and secondary structure, cultivar Aruba. Fig. 9 and 10. Triarch roots of the least thickness and secondary structure among all examined cultivars, cultivar Tetyda. Fig. 11 and 12. Roots of cultivar Bila - exclusively tetrarch in all Bila's sections examined. Fig. 13. Tetrarch roots of cultivar Denar - the widest thickness and secondary structure of all cultivars examined. UV-excited autofluorescence and aniline blue fluorescence. Bars 0.33 mm

Objaśnienia: CP - miękisz kory pierwotnej, SX - ksylem wtórny, gwiazdki - egzoderma, strzałki - komórki endodermy w 1. stadium rozwojowym, strzałki puste - komórki endodermy w 2. stadium rozwojowym, strzałki wąskie - pasma protoksylemu, groty strzałek podwójne - fluorescencja błękitu aniliny związanego z kalozą we floemie, groty strzałek - pierwsze naczynia w najmłodszym ksylemie wtórnym

Fot. 7-13. Przekrój poprzeczny pierwszorzędowych korzeni przybyszowych u odmian *Solanum tuberosum* - różnice anatomiczne dotyczące grubości korzenia, liczby pasm ksylemu pierwotnego i zaawansowania przyrostu wtórnego. Fot. 7 i 8. Korzenie triarchiczne o małej lub średniej grubości i przyroście wtórnym, odmiana Aruba. Fot. 9 i 10. Korzenie triarchiczne o najmniejszej grubości i przyroście wtórnym spośród wszystkich odmian badanych, odmiana Tetyda. Fot. 11 i 12. Korzenie u odmiany Bila - wyłącznie tetrarchiczne we wszystkich skrawkach z korzeni tej odmiany. Fot. 13. Korzenie tetrarchiczne u odmiany Denar - największa grubość i przyrost wtórny spośród wszystkich odmian badanych. Autofluorescencja wzbudzona promieniowaniem UV i fluorescencja błękitu aniliny. Odcinki skalujące 0,33 mm



Explanations: CP - primary cortex parenchyma, SX - secondary xylem, asterisks - exodermis, arrows - endodermal cells in the 1st developmental stage, open arrows - endodermal cells in the 2nd developmental stage, slim arrows - protoxylem strands, double arrowheads - fluorescence of callose-bound aniline blue in phloem

Fig. 14-17. Cross-sections of primary adventitious roots in *Solanum tuberosum* cultivars in comparison to lateral roots - anatomical differences in root thickness, number of primary xylem strands and secondary structure extent. Fig. 14 and 15. Pentarch roots of small or moderate thickness and secondary structure, cultivar Oberon. Fig. 16. Sextarch root, cultivar Hubal. Fig. 17. Di- and triarch lateral roots with single or double only layer of cortical parenchyma cells and no secondary growth, cultivar Hubal. UV-excited autofluorescence and aniline blue fluorescence. Bars 0.33 mm or 0.12 mm in Fig. 14-16 or 17, respectively

Objaśnienia: CP - miękisz kory pierwotnej, SX - ksylem wtórny, gwiazdki - egzoderma, strzałki - komórki endodermy w 1. stadium rozwojowym, strzałki puste - komórki endodermy w 2. stadium rozwojowym, strzałki wąskie - pasma protoksylemu, groty strzałek podwójne - fluorescencja błękitu aniliny związanego z kalozą we floemie

Fot. 14-17. Przekrój poprzeczny pierwszorzędowych korzeni przybyszowych u odmian *Solanum tuberosum* w porównaniu do korzeni bocznych - różnice anatomiczne dotyczące grubości korzenia, liczby pasm ksylemu pierwotnego i zaawansowania przyrostu wtórnego. Fot. 14 i 15. Korzenie pentarchiczne o małej lub średniej grubości i przyroście wtórnym, odmiana Oberon. Fot. 16. Korzeń sekstarchiczny, odmiana Hubal. Fot. 17. Di- i triarchiczne korzenie boczne z tylko pojedynczą lub podwójną warstwą komórek miękiszu kory pierwotnej i bez przyrostu wtórnego, odmiana Hubal. Autofluorescencja wzbudzona promieniowaniem UV i fluorescencja błękitu aniliny. Odcinki skalujące 0,33 mm lub 0,12 mm, odpowiednio na rys. 14-16 lub 17

Quantitative analysis of potato root structure

Significant differences were found between cultivars regarding the area of primary adventitious root cross-section (Table 1) or xylem cross-section (Table 2). The thinnest roots, as measured by root cross-section, were found in plants of cultivars Etola and Tetyda (early and medium early cultivars, respectively), and they were nearly 4-fold thinner than the widest ones in Denar (very early). For Etola and Denar, root cross-section area was seemingly correlated with cross-section of xylem, as the same cultivars produced the smallest and widest xylem, respectively (at this stage of research correlations were not analyzed statistically due to the low number of individuals).

Table 1

Cross-section area of the primary adventitious root in potato. Average areas marked with different letters are significantly different at P<0.001
Pole przekroju poprzecznego korzenia przybyszowego I rzędu u ziemniaka. Średnie zaznaczone innymi literami różnią się istotnie przy P<0,001

Cultivar Odmiana	Cultivar earliness group Grupa wczesności odmian	Average area of root cross-section mm × 1000^{-1} Średnie pole przekroju poprzecznego korzenia	Group Grupa
Etola	early	425	a
Tetyda	medium early	444	a
Finezja	medium early	548	ab
Gwiazda	early	676	ac
Hubal	early	714	ac
Etiuda	medium early	749	ac
Kuba	medium early	764	ac
Bila	early	771	ac
Aruba	early	815	ac
Lord	very early	890	bc
Gandawa	medium early	905	bc
Stasia	medium early	909	bc
Justa	very early	951	bc
Oberon	medium early	998	c
Michalina	early	1066	c
Denar	very early	1698	d

Table 2

Xylem cross-section area in the primary adventitious roots of potato. Average areas marked with different letters are significantly different at P<0.001
Pole przekroju poprzecznego ksylemu w korzeniach przybyszowych I rzędu u ziemniaka. Średnie zaznaczone innymi literami różnią się istotnie przy P<0,001

Cultivar Odmiana	Cultivar earliness group Grupa wczesności odmian	Average area of xylem cross-section mm × 1000^{-1} Średnie pole przekroju poprzecznego ksylemu	Group Grupa
Etola	early	58	a
Hubal	early	66	ab
Tetyda	medium early	67	ab
Finezja	medium early	70	ab
Aruba	early	81	ab
Bila	early	84	ab
Gwiazda	early	86	ab
Kuba	medium early	121	ab
Justa	very early	122	ab
Gandawa	medium early	123	ab
Stasia	medium early	126	ab
Lord	very early	133	ab
Oberon	medium early	137	ab
Etiuda	medium early	154	ab
Michalina	early	175	b
Denar	very early	307	c

Cultivars within the same earliness group usually differed in both analyzed features. It was especially evident in regard to root cross-section in medium early cultivars (Table 1). The variability between individual plants was very high within the cultivars (not shown),

the differences being even 3-fold. In some plants, also the individual roots had strikingly different cross-section areas.

DISCUSSION

In search of crops more tolerant of abiotic stress resulting from drought and/or increased temperatures, cultivars are sought that have, among other features, root systems exhibiting higher efficiency in extracting soil water (Levy and Coleman, 2015; Iwama, 2008). Such root systems should possess increased water transport efficiency, which may result from root growth rate and depth, morphology or anatomical structure.

Considering the results of this work in view of heat and/or drought stress susceptibility of potato, it is worth mentioning that the basal part of primary adventitious roots examined here are especially vulnerable, as the loss of these roots due to desiccation would deplete plant of water until it formed new adventitious roots. Therefore, it is important what protection against desiccation develop these roots. As we have shown, despite formation of cambium-derived secondary vascular tissues, root cortex was maintained with only monolayered exodermis as the sole protective tissue, and to the authors' best knowledge no *Solanum tuberosum* genotypes form periderm on roots. The blue autofluorescence of exodermal cell walls was indicative of the suberin presence. In future, it would be reasonable to extend this study to investigations of root exodermis suberization, e.g. the precise determination of chemical constituents and suberin layer ultrastructure, since its thickness or striation would influence cell wall hydrophobicity.

Generally, the anatomical features of potato primary adventitious roots examined in this work did not differ from these described earlier briefly in handbooks by Hayward (1936) and Cutter (1992). The variability of stele structure was higher than 4-6-arch mentioned by Cutter (1992), as tri-, tetra-, pent- or sextarch roots were found, with most roots tetrarch. Also, Cutter (1992) reported the 1nd stage endodermis only (Casparian strip) while in roots observed in this work, phloem-facing endodermis was always developmentally more advanced.

Regarding the results of cross-sections measurements, the presented work has to be considered as a preliminary study only. One of the reasons is that due to the low number of individuals, the variability within cultivar was very high, with frequent 3-fold differences between plants. Despite such high variability, significant differences were found between cultivars. Therefore, the experiment will be repeated in future, with higher number of individuals that would yield more reliable results.

CONCLUSION

The study demonstrates usefulness of a simple microscopy protocol and digital images analysis for examination of morphology and anatomy of the potato root system.

The results of the quantitative analysis of potato root structure presented in this paper made it possible to demonstrate the high variability of tested cultivars in terms of cross-section area and xylem cross-section area in the primary adventitious roots. The

demonstration of a relationship between tolerance of cultivars to heat and drought stresses and the anatomy of the root system will require further research.

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