SCLERIFICATION IN THE BARK TISSUES OF COMMON FIR (*ABIES ALBA* MILL.)

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ABSTRACT

The sclerification process in bark tissues of common fir (*Abies alba* Mill.) has been described. The sclerification begins in 3 years old stems. Sclereids differentiate from cortical parenchyma cells and from secondary phloem parenchyma cells that do not contain phenolic deposits. The first single sclereids are formed at the interface of the cortex and nonfunctional phloem. Hereafter, a continuous layer of them is formed. Later, new sclereid layers are formed successively in nonfunctional secondary phloem and cortex. The consecutive layers are separated tangentially by phloem parenchyma cells, that accumulate large amounts of phenolic substances, and by compressed phloem cells. Laterally they are separated by phloem rays that except of some dislocations are continuous. Structural net of the cortical phloem ray cells and phloem parenchyma delineates the areas where the formations of sclereid layers occurs in nonfunctional secondary phloem. Older cortex contains more sclereid layers and the time period of their formation extends continuously.

KEY WORDS: *Abies alba*, bark, sclereid differentiation, secondary phloem.

INTRODUCTION

In trees all the tissues positioned outside cambium are called the bark. The first descriptions of the bark tissues of the conifers come from the second half of the 19th century (Hartig 1837; Sanio 1873; Strasburger 1891). At the beginning of the 20th century the anatomy of the bark tissues of the coniferous trees very extensively examined (Esau 1950, 1954, 1965; Abbe and Crafts 1939; Isenberg 1943; Chang 1954; Grillos and Smith 1959). The main effort was to recognize the structure of the secondary phloem. Available data on bark tissues of fir are scarce and they are usually provided by reviews and general descriptions of the bark tissues of the gymnosperms. The most detailed descriptions were given by Golinowski (1968, 1971a, b).

The bark of common fir (*Abies alba*) is structurally very heterogeneous. It is composed of primary and secondary tissues (Figs 1-3). Primary phloem, cortical parenchyma, secretory tissue, and epidermis belong to the primary tissues forming the bark (Fig. 1) while cambium, secondary phloem, sclereids, secretory tissue and peryderm are of secondary origin (Figs 2 and 3).

In the primary growth area the largest is the cortex. It is composed mostly of parenchymatic cells with large intercellular spaces. The cortex contains also numerous resin ducts and secretory cells filled with mucilage. During the stage of the volumetric growth the cortical cells are stretched and compressed.

All the changes occurring in the cortex are caused by the activity of the two lateral meristematic tissues: cambium and phellogen. Continuous formation of new cells, stretching and compression of already existing nonfunctional cells is reflected in the thickness of separate layers of the cortical tis-

sues and in the thickness of the bark itself. The bark of common fir remains functional for many years. This is because the first layer of phellogen remains functional for a very long time. Therefore the history of all processes occurring during the bark development may be followed. The bark is a dynamic component of a tree where qualitative and quantitative developmental changes occur.

Sclerification is a basic process associated with the volumetric growth. It causes the largest modifications in the bark (Den Outer 1967; Whitmore 1962). Sclerification takes place in the cortex and nonfunctional secondary phloem. The formation of the sclereids occurs in cycles. The length of the each cycle increases with the age of the tree (Golinowski 1971a, b). The available literature with data on the formation of the sclereids in the bark of common fir provides only the anatomical characteristic of this process. So far no developmental cytological description has been done.

The aim of this work is to describe events occurring during sclerification in common fir at the cellular level.

MATERIALS AND METHODS

The research was conducted on 13 randomly selected trees of *Abies alba*, being 35 till 135 years-old. The trees originated from the natural forest in Gromnik District in Podkarpackie (southern Poland). The samples were collected in October 1996, June 1997 and January 1998 (Table 1).

Selected trees were felled and then the samples were collected from different heights of the stem (1.5, 4.0, 8.0, 12.0, 16.0, 20.0, 24.0, 28.0, 32.0 m, and from the increments of the last 5 years). Each collected sample was a 10 cm long transverse section of a stem. They enabled estimation of the age of

Figs 1-3. Bark tissues of the 37 years-old tree (January 1998).

Fig. 1. Transverse section of 1 year-old stem. $\times 115$

Fig. 2. Transverse section of 3 years-old stem. First sclereids (S) formed in cortex (CO) and between secondary nonfunctional phloem (NFP) and cortex. $\times 115$

Fig. 3. Transverse section of 20 years-old stem. Four layers of sclereids (S) are visible in the bark. $\times 115$

C – cambial zone, CO – cortex, E – epidermis, FP – functional phloem, IS – intercellular space, NFP – nonfunctional phloem, PD – periderm, R – vascular ray, RC – resin canal, S – sclereid, X – xylem, arrows – parenchyma cells accumulating phenolic substances, asterisk – secretory cell filled with mucilage.

TABLE 1. Characteristics of trees from which the samples were collected for microscopic examinations.

Collection time	No. of tree	Age (years)	Total height (m)
October 1996			
	I	116	32
	II	97	28
	III	82	24.5
	IV	64	18
	V	45	12.5
June 1997			
	I	135	30.5
	II	111	28
	III	49	17.5
	IV	56	14.5
	V	43	5.5
January 1998			
	I	67	19.5
	II	35	15
	III	135	30.5

the stem at the sectioned level. From the south-oriented part of the stem small samples of bark were collected for microscopic examinations. Then they were fixed and embedded in epoxy resin according to procedures described in Robinson et al. (1987).

The fixation took place in a mixture of 5% glutaraldehyde and 4% paraformaldehyde in 0.1 M cacodylic buffer (pH 7.2-7.3) at the room temperature for 24 h. Then the samples were washed in several changes of the same buffer and post-fixed in 2% OsO₄ for 2 h at 4°C. They were dehydrated in increasing graded series of ethanol, that was eventually substituted with propylene oxide. They were embedded in synthetic Epon resin. The semi-thin sections (3 µm thick) were taken on the microtome 2065 Supercut (Leica/Reichert-Jung), dried at 70°C and stained with 2% Methylen Blue in 2% borax and with 1% Azur B. The slides were mounted in DePeX (SERVA). The light microscopic examinations were conducted on OLYMPUS AX 70 "PROVIS" with photographic

device OLYMPUS SC 35 and video camera SONY DXC 950P PowerHAD. Via this video camera the microscope was connected with the digital video image analysis system AnalISIS-Pro. All measurements, figure preparation and prints were made using the AnalISIS-Pro programme. All measurement were statistically tested with the SPSS/PC+ programme.

Ultrathin sections (60-80 nm thick) were taken on ultramicrotome Ultracut E (Reichert). They were collected on uncoated and formvar coated slot copper grids. They were stained with uranyl acetate followed by lead citrate and examined in JEOL JEM 100C and JEOL 1220 transmission electron microscopes.

Probes for roentgen spectroanalysis were fixed as described above but the post-fixation step with OsO₄ was omitted. Sections for examinations (120-240 nm thick) were cut on ultramicrotome Ultracut E and carbon shadowed. The sections were examined using electron microscope JOEL JEM 1220 Ex equipped with roentgen spectrometric device EDS LINK AN 10000.

RESULTS

The anatomy of the bark in the trees of different age is shown in Figs 1-3. In the first year of growth (Fig. 1) the bark tissues make about 85% of the total area of the stem transverse section. Epidermis mono layer covers the bark. The epidermis remains functional only during the first year of vegetation and in the next year is substituted by periderm (Figs 1 and 2). When the first periderm is formed the remnants of the epidermal cells are still present (Fig. 1). In the primary growth the volumetrically largest part of it represents the cortex. The cortex is composed of loosely arranged parenchymatic cells with large intercellular spaces, resin ducts and mucilage containing cells. The latter cells spread within the intercellular spaces and reach dimension several times larger than parenchymatic cells. A primary phloem is the most adaxial primary tissue of the bark. It remains functional during the entire phase of the primary growth. A secondary phloem develops at the end of the first year of shoot growth and replaces the primary phloem that ultimately degenerates. The secondary tissues cause large increase in the

TABLE 2. Thickness of tissues in the bark of *Abies alba* expressed as mean values (in µm) and standard deviations (given in square brackets).

	Age of tree (years)												
	1	2	3	4	5	6	7	10	15	20	25	30	40
Number of observations	10	11	10	12	11	11	10	9	12	5	11	5	10
Nonconducting phloem	0	192 [33]	231 [80]	612 [263]	678 [82]	868 [202]	1064 [131]	1114 [95]	1497 [204]	2657 [165]	1777 [134]	2517 [66]	2191 [137]
Conducting phloem	200 [22]	181 [34]	138 [27]	251 [54]	388 [42]	250 [41]	283 [61]	268 [38]	307 [45]	243 [54]	374 [24]	341 [29]	246 [32]
Total bark thickness	1468 [87]	2076 229	1957 [136]	2277 [514]	2590 [107]	2676 [286]	2805 [93]	2669 [100]	2940 [326]	3054 [255]	3991 [156]	3649 [101]	3244 [134]
Total thickness of sclereid layers	0	0	0	398 [124]	702 [231]	528 [84]	811 [140]	541 [80]	1144 [280]	1591 [214]	1378 [164]	1715 [260]	1747 [113]
Rate of phloem increment (µm/year)	201 [22]	188 [18]	123 [34]	215 [72]	213 [13]	186 [34]	192 [14]	118 [11]	120 [16]	151 [6]	81 [6]	95 [2]	66 [3]
Rate of increment of sclereid layers (µm/year)	0	0	0	99 [31]	140 [46]	88 [14]	116 [20]	54 [8]	76 [19]	80 [8]	55 [9]	57 [9]	47 [3]

Figs 4-6. Secondary phloem parenchyma cells from a 5 years-old stem (June 1997).

Fig. 4. Transverse section of nonfunctional secondary phloem and vascular ray cells (R). Note the large intercellular spaces (IS) aside of the ray cells caused by compression of the sieve cells (SC). PP – phloem parenchyma cells, arrows – phenolic substances. $\times 850$

Fig. 5. Early phase of accumulation of phenolic substances (arrows) in the vacuole (V) next to the tonoplast in phloem parenchyma cell. IS – intercellular space, n – nucleus. $\times 3400$

Fig. 6. Phloem parenchyma cell with vacuole filled with osmiophilic phenolic substances. IS – intercellular space. $\times 3400$

stem volume during the second year of shoot growth. However, the general pattern of the stem anatomy is not changed noticeably. In the third year (Fig. 2) the typical processes (cell compressions and stretching) associated with the stem radial and circumferential growth become more obvious. They cause slight decrease in the bark thickness in comparison to the second year annual increment (Table 2). Also the resin ducts are gradually stretched in the tangential direction. The cortical parenchyma tissue dominates on transverse sections of 3 years-old stems. These cells transport nutrient in the radial direction, store ergastic and phenolic substances as well as resins. They later differentiate either into sclereids or phellogen cells. Phenolic substances accumulate in vacuoles of some fully developed cortical and phloem parenchyma cells only (Figs 1-6). They are rarely found also in phellodermal and dead phellem cells. They appear first in the vacuole next to tonoplast (Fig. 5). Their amount gradually increases until they fill up the entire vacuole (Fig. 6). Numerous electron translucent vesicles of different shapes found in the vacuoles filled up with phenolic substances are artefacts caused by their dissolution in ethanol during sample preparation (Fig. 6). Addition of the coffein to the fixative strongly decreases the number and extent of the electron translucent vesicles in the cells accumulating phenolic substances.

The sclerification of some cortical cells begins in the third year of shoot growth. The first single sclereids were present in the intercellular spaces at the interface of the cortex and nonfunctional primary phloem and in cortical parenchyma (Fig. 2). Numerous intercellular spaces were formed close to the vascular rays where many nonconducting sieve cells occurred (Fig. 4). The development of sclereids is a gradual process. It begins from the divisions of cortical or phloem parenchyma cells (Fig. 7). The newly developed cells enlarge strongly and reach dimensions several times larger than normal cells. They acquire lobbed irregular shapes and invade intercellular spaces (Fig. 8). Their abnormal shapes are caused by their intrusive growth occurring on all edges of the cell walls facing intercellular spaces. Finally the shape of future sclereid fits the shape of the intercellular space filled by this cell (Figs 8-10). The next phase in the sclereids formation is a thickening of the cell wall (Fig. 10). Numerous small vesicles with electron translucent content accumulate close to the plasmalemma and then fuse with the plasma membrane. Their content is released and incorporated into the cell wall (not shown). Another type of vesicles involved in cell wall thickening are large vesicles with homogenous osmiophilic content (Figs 11-13). The staining pattern of material present in these large vesicles is different from the previously described. The extensive thickening of the cell wall causes folding on of its inner surface (Fig. 12). In addition to different types of vesicles also rough endoplasmic reticulum, numerous free ribosomes and elongated mitochondria are present in a parietal cytoplasm of differentiating sclereid (Fig. 11). Plastids with few small starch grains and well-developed thylakoid system filling almost the entire stroma are present in the protoplast of differentiating sclereid. These cells contain also strongly osmiophilic phenolic substances. The cells developing into sclereids possess always numerous pit fields-especially at the interface with the parenchymatic cells (Fig. 11). The cell wall thickening and maturation is the final step in the sclereid development. It leads to closure of the cell lumen and lignification of secondary cell wall. Mature sclereids are dead cells. The thick secondary walls have a striated pattern (Fig. 14). In some sclereids where part of the cell lumen has been retained phenolic substances were noticed inside.

In the next phase of sclerification new sclereids differentiate around already present single mature sclereid. They form initially groups (or nests) of sclereids. This process leads in the end to formation of almost continuous sclereid layer. This layer is separated tangentially by live phloem ray cells lined up in radial direction (Figs 3 and 9). The formation of single sclereid layer lasts for 5 or even 10 years. It lasts longer in older parts of the stem. Depending on the age of the secondary phloem, the number of sclereid layers present is different. The consecutive sclereid layers are separated one from another along the radius by the phloem parenchyma cells and compressed sieve cells (Fig. 3).

The phloem rays are sometimes curved or slightly dislocated close to developing sclereid layers (Fig. 3). However, the connection with other parenchymatic cells and the nutrient transport system in the cortex is maintained. Development of the sclereids causes compression of the cortical tissues thus total annual increments of cortex are very small (0.1-0.7 mm) (Table 2).

Light microscope analysis of the serial transverse sections showed that some phloem mother cells do not follow the phenolic substances accumulation pathway. Instead they remain meristematic for several years. Frequently these cells are found abutting to unicellular phloem rays. In this undifferentiated neotenic state they enter the phase of nonfunctional phloem development when the process of the formation of intercellular spaces begins. This is caused by compression of sieve cells. At that time these meristematic cells divide. Their divisions are asymmetric. Smaller cells usually grow intensively in different directions. Their intrusive growth leads to filling up of the intercellular spaces created in the process of sieve cells compression and stretching. Parenchymatic cells developing into sclereids retain symplastic connections with sister cells.

Large intercellular spaces are present among cortical cells in young stems. Their volume decreases with the age of the shoots and enlargement of the sclereids. The sclereids differentiate also in the cortex in the way described for sclereid formation in nonfunctional phloem. However, usually only one layer of sclereids is formed in the cortex (Figs 2 and 3).

The roentgen microanalysis method (EDX) was introduced to follow in more details the process of cell wall thickening (Lott and Spitzer 1980; Wada and Lott 1997). Using this method the elemental composition of the homogenous vesicle content, cell walls of developing and mature sclereids were compared. The comparison of the obtained spectra shows high similarity of elemental composition of structures examined. The same elements are present in all analysed structures but their proportions are different. For instance then the roentgen spectra of the mature sclereid and tracheid cell wall are compared it appears that both indicate the presence of the same elements. Additionally, the height of the picks is the same which means similar amounts of the same elements in both cell types.

The measurements of the tissues thickness measured along the stem radius was conducted using computer video image analysis system AnalISIS. They were made on different trees on part of the stems being of different age. The medium values and standard deviations of the measured samples are given in Table 2.

The one-way analysis of variance was introduced to compare the thickness of the examined tissues in relation to the age. This method was introduced because the increment of the tissue thickness is not a continuous parameter. The Leven test (not shown) showed that variance of the tissues thickness

Figs 7-10. Sclerification in secondary phloem (June 1997).

Fig. 7. Transverse section through secondary phloem of 10 years-old stem. The youngest sclereid layer (S) with a group of parenchymatic cells (PP) with irregular shapes (arrows). IS – intercellular space, PPT – parenchymatic cell accumulating phenolic substances, R – vascular ray. $\times 40$

Fig. 8. Transverse section through secondary phloem of 4 years-old stem. Irregular phloem parenchyma cells (PP) without phenolic substances differentiate into sclereids. PPT – parenchyma cell accumulating phenolic substances, R – vascular ray. $\times 45$

Fig. 9. Transverse section of the nonfunctional phloem in 102 years-old stem. Large irregularly shaped cells with thickened cell walls differentiate into sclereids (S). IS – intercellular space, PPT – parenchymatic cell accumulating phenolic substances, R – vascular ray. $\times 80$

Fig. 10. Longitudinal tangential section through nonfunctional phloem in 102 year-old stem. Irregularly shaped cell differentiates into sclereid (S). Arrows point to thickened cell walls covered by a thin layer of cytoplasm with nucleus (n). R – vascular ray, SC – sieve cell. $\times 190$

Figs 11-14. Sclereid differentiation.

Fig. 11. Phloem parenchyma cells in 28 year-old stem (October 1996). Differentiating sclereid (S) with thick secondary cell wall (CW). Arrow heads point to pit fields occupying the entire cell wall between differentiating sclereid and phloem parenchyma cell (PP). IS – intercellular space, pl – plastid, arrows – phenolic substances. $\times 3400$

Fig. 12. Phloem parenchyma cells (PP) between sclereids (S) in 19 year-old stem (October 1996). Vesicles with homogenous content (arrows) gather near the thickened cell wall. IS – intercellular space, V – vacuole. $\times 2200$

Fig. 13. Thickening of sclereid cell wall (CW) differentiating in 124 years-old stem (January 1998). Vesicles with homogenous content (arrows) fuse with plasmalemma. V – vacuole. $\times 20500$

Fig. 14. Mature sclereid (S) in 4 years-old stem (October 1996). Lignified and thickened cell wall is marked with (arrows). No cell lumen is visible. IS – intercellular space, SC – sieve cell. $\times 2700$

TABLE 3. Results of one-way analysis of variance of different tissue thickness.

Type of tissue	DF	Square deviation	F
Nonfunctional phloem	12	6251144.09	287.72***
Total thickness of sclereid layers	12	3688890.76	148.71***
Rate of phloem increment	12	27809.85	33.37***
Rate of increment of sclereid layers	12	20575.95	53.91***

*** at $p < 0.001$

in the tested years may be accepted as homogenous at the significance level 0.05 (Sokal and Rolph 1969). One-way analysis of variance indicates important differences in the thickness of all tissues examined (Table 3).

Changes in the secondary phloem are shown on Fig. 15. The thickness of the functional and nonfunctional phloem changes with time. The curves showing the continuous increase of the thickness of functional and nonfunctional phloem are similar. However, the values for the latter are several times higher. Although, the both curves are positioned on different heights they show similar fluctuations. The youngest stems (1, 2 and 4 years-old) have the thinnest nonfunctional phloem layer. This layer is especially thin in the 3rd year of growth. The functional phloem reaches the highest annual increment in 5 years-old stems.

The increment of the nonfunctional phloem is accompanied by increase of sclereid thickness. The sclereids fill up intercellular spaces formed by the compression of sieve cells. Changes of the sclereid layer thickness from the beginning of the sclereid formation till 40th year of growth are shown in Fig. 16. The large fluctuations in the thickness are very obvious in the examined time period. In older stem the sclereid layers reveal the highest relative thickness increment and they form the thickest layer in the bark. The thickness of the sclereids layer is related to the thickness of cambium layer. This relation fits to linear regression function with a high determination parameter $R^2 = 0.717$. It indicates that measured values fit to the estimated regression function at the level higher than 70% (Fig. 17).

Taking into consideration the annual sclereid layer increment (Fig. 19) it appears that the sclereids develop most ex-

Fig. 15. Mean values and standard deviations of functional and non-functional phloem thickness during the 40 years of growth.

Fig. 16. Mean values with 95% confidence interval of bark sclereids thickness during the 40 years of growth.

Fig. 17. Linear regression showing relation between the thickness of the cambial zone and the thickness of the sclereid layers. Linear regression function (y) and determination parameter (R^2).

Fig. 18. Mean values with 95% confidence limit of the annual secondary phloem increments in stems of different ages.

tensively in young 3-7 years-old stems with the maximum in 5 years-old stems. Since the 7th year of growth the annual increments become gradually smaller and reach two minima in the 10th and 40th year of growth. The secondary phloem (both functional and nonfunctional parts) reveals also similar trend of decreasing annual increments (Fig. 18). The highest annual increments of the phloem thickness occur during the first 7 years of growth with the lowest increment in 3 years-old stems.

Fig. 19. Mean values with 95% confidence limit of the annual sclereid increments in stems of different ages.

DISCUSSION

The bark comprises all the tissues located between phellogen and cambium. All newly formed bark cells originate from these two lateral meristems. Their development influences the position and functions of already existing cells of primary and secondary tissues. The bark of fir is a dynamic system that functions during the entire tree life.

In perennial plants of the moderate climate there is a clear separation between the time of growth (spring and summer) and the time of rest caused by low temperatures (late autumn and winter) (Abbe and Crafts 1939; Evert 1960; Golinowski 1971a). This successive periods of activity and rest influence all live cells in the plants and need some specific adaptations of the protoplasts, e.g.: the increase of the amount of lipids and free soluble sugars protects against low temperatures.

The observations of fusiform cambial initials indicate that they differentiate in the area of a secondary phloem into sieve and phloem parenchyma cells, mother sclereid cells, mother vascular ray cells and albumin cells. These diverse cells remain as one symplastic domain. This domain creates a kind of net formed by phloem parenchyma cells (in tangential direction) and phloem ray cells (in radial direction). The "mesh" of this net is formed by sieve cells. Parenchymatic cells are connected with one another via plasmodesmata in longitudinal and radial direction. They have also well-developed plasmodesmatal connections to parenchymatic cells of phloem rays in radial direction. The symplasts of the sieve cells ("mesh") and phloem ray cells ("net") are connected via albumin cells.

Except of differentiated parenchymatic cells in the secondary phloem there are also discrete "neotenic" undifferentiated cells. These cells are dispersed as single elements or in small groups among sieve cells close to the phloem rays. In this paper it has been shown that the structure of their protoplast is very similar to meristematic cells. They may remain as such for 3-4 years; the time that is necessary for them to become a part of nonfunctional phloem. Their ultrastructural features as: dense protoplast, divisions in different planes, irregular shape and their localization in the bark indicate that these cells are sclereid mother cells.

Parenchymatic cells arranged in regular tangential and radial layers or forming irregular groups (depending on the species) play an important role in the secondary phloem (Den Outer 1967). The parenchymatic cells in the bark play different functions depending on their position. These are: nutrients storage and transport; production and accumulation of phenolic substances, mucilages, resins and crystals; de-dif-

ferentiation into meristematic cells (phellogen) and formation of sclereids (Chattaway 1955; Whitmore 1962; Srivastava 1964; Zigler 1964; Den Outer 1967; Souter 1967; Zobel 1978).

Sieve cells in the nonfunctional secondary phloem zone are dead and gradually compressed. The first indication of their compression is a separation of their cell walls from the phloem ray cells. Then the cell walls containing many pit fields (usually radial walls) become progressively bent causing approaching of the tangential walls until the cell lumen is fully closed. The compression of the sieve cells precede sclerification and decreases the thickness of the secondary phloem and the bark itself.

Sclereids appear for the first time in 3 years-old stems of *Abies alba*. Foster (1955) reports that the development of sclereids is induced by low auxin concentration. This hypothesis may explain why in fir the first sclereids develop as late as in the 3rd year of growth. We hypothesize, that there must be some distance from the apex for the auxin concentration decrease to the level enabling sclereids differentiation. Sclerification starts in the middle of the May simultaneously with the spring xylem development (Golinowski 1971a). The beginning of the sclereid formation process influences also the development of functional phloem; decrease of its total thickness and its smallest annual increment. The total thickness of the bark decrease also in the 3rd year of growth.

Sclerification occurs via de-differentiation of parenchymatic cells or straight from neotenic sclereid mother cells. It occurs usually in nonfunctional secondary phloem and in the cortex. Differentiating sclereid may enlarge its volume several hundreds times while the neighbouring parenchymatic cells enlarge their volume only 2 for 3 times. The growth of the sclereids is a conjunction of the symplastic growth during early developmental stage with the intrusive growth in later developmental phases when the sclereid invades the intercellular spaces.

First sclereids appear as single elements at the interface of the cortex and nonfunctional phloem. Later new additional sclereids differentiate in the vicinity so the groups or nests of sclereids are formed. After several years of growth almost a continuous layer of sclereids is formed. This process occurs in cycles and repeats every 5-10 years depending on the age of the stem. Mature sclereids are dead cells with irregular shapes and very thick multilayered secondary cell walls. The cell lumen is frequently closed. Roentgenographic examinations revealed that the quantitative elemental composition of the sclereid walls is the same as the composition of the tracheal cell walls.

Developing sclereids do not destroy the system of the parenchymatic tissues. The continuity of these tissues is maintained and only some cells are dislocated. The sclereid layers create the main part of the bark. The thickness of the sclereid layer increases with the age of the stem (Hodlheid 1951; Chang 1954; Srivastawa 1963; Golinowski 1971a, b). The frequency of the formation of new sclereids decreases with the increase of the number and thickness of sclereid layers. This process is a response on the volumetric increment of the stem and also on compression and stretching of the secondary nonfunctional phloem and cortical parenchyma.

Sclereids create layers separated by tangentially positioned phloem parenchyma cells frequently accumulating phenolic substances. The presence of the parenchymatic cells between sclereid layers enable continuous growth of already existing sclereid layers (Den Outer 1967; Whitmore 1962). In the radial direction sclereid layers are separated by the phloem

rays. The direction of the rays in the nonfunctional secondary phloem reflects processes associated with volumetric growth of stem, e.g.: cell compression and stretching. This direction in some parts of the stem may be oblique or even tangential.

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SKLERYFIKACJA W TKANKACH KOROWYCH JODŁY POSPOLITEJ *ABIES ALBA* MILL.

STRESZCZENIE

Opisano proces skleryfikacji tkanek korowych jodły pospolitej *Abies alba*. Proces ten rozpoczyna się w 3 letnich pędach. Sklereidy różnicują się na terenie kory pierwotnej z komórek miękiszowych kory pierwotnej, a na terenie łyka wtórnego z komórek miękiszowych łyka, które nie wytworzyły substancji fenolowych i z komórek macierzystych sklereidów. Pierwsze pojedyncze sklereidy powstają na granicy kory pierwotnej z łykiem nieprzewodzącym i zapoczątkowują tworzenie się warstwy sklereidów. Kolejne warstwy sklereidów powstają w nieprzewodzącym łyku wtórnym i w korze pierwotnej. Im starsza jest kora, tym więcej warstw sklereidów wchodzi w jej skład i wydłuża się czas ich tworzenia.

Poszczególne warstwy sklereidów oddzielone są od siebie w płaszczyźnie stycznej komórkami miękiszowymi łyka, gromadzącymi duże ilości substancji fenolowych oraz zgniecionymi komórkami sitowymi. Natomiast w kierunku promieniowym warstwy sklereidów poprzecinane są promieniami łykowymi, które pomimo pewnych bocznych przesunięć zachowują ciągłość i stanowią ważne ogniwo w symplastycznej łączności pomiędzy łykiem przewodzącym a miękiszem kory pierwotnej. Przestrzenna sieć miękiszu promieni łykowych i miękiszu łykowego wyznacza obszary na terenie nieprzewodzącego łyka wtórnego, w których następuje rozbudowa warstw sklereidów.

SŁOWA KLUCZOWE: *Abies alba*, kora, różnicowanie się sklereidów, łyko wtórne.