The anatomical structure of the common fir (*Abies alba* Mill.) bark

I. Development of bark tissues

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Abstract:

Qualitative and quantitative changes in bark tissues of the fir (*Abies alba*) connected with the long-term cambium activity were investigated: 1) the development of secondary phloem elements forming the annual ring in different-aged parts of the stem was studied in detail; 2) the terms of formation of the phloem elements were established as well as the duration of the periods of their differentiation and functioning; 3) observations were made of the changes occurring in nonconducting phloem and primary cortex tissues; 4) the course of development of the periderm was studied.

INTRODUCTION

The term bark denotes in the present paper the shoot tissues situated outside the cambium, both in the primary and secondary stem structure.

The anatomical structure of the bark tissues of coniferous trees has been described in the literature of the 19th century.

T. Hartig (1851) and H. Mohl (1855) were the first to give a characteristic of the elements of secondary phloem in gymnosperms.

The problem is exhaustively elaborated in the papers of: Bertrand (1874), Moeller (1882), Russow (1882), Strasburger (1891), Holdheide (1951) and Chang (1954).

The structure of secondary phloem in some species of the *Pinaceae* family has been described by numerous investigators: Abbe and Crafts (1939), Huber (1939), Grillos and Smith (1959), Srivastava (1963), Huber and Liese (1963), Evert and Alfieri (1965) Srivastava and O'Brien (1966 a, b), Wooding (1965), Den Outer (1967), Alfieri and Evert (1968) and others.

As seen from this brief review, the literature dealing with the anatomical structure of bark tissues of conifers is extensive, nevertheless only few works deal with the development of bark in specific ontogenesis. Information on the structure of fir bark is only fragmentary.

The present study was undertaken for investigating the qualitative and quantitative changes in the bark tissues due to cambial activity of many years duration. Particular attention was devoted in these investigations to the following problems:

- 1) the characteristic of the elements of the secondary phloem forming the annual rings in different-aged shoots,
- 2) determination of the time of phloem formation and differentiation and of the period of their functionality;
- 3) the changes occurring in phloem and in primary cortex tissues;
- 4) the characteristic of the peridermis.

The species *Abies alba* Mill. was chosen for the investigations, because the bark of this fir does not practically desquamate, thus, it is possible to follow the changes in the bark tissues in the course of individual growth of trees even a hundred years old.

Most of the terms used in the present paper have been defined by K. Esau (1939, 1950, 1965). Use is also made of the terminology suggested by Wilson, Wodzicki and Zahner (1966) for describing the development stages of secondary xylem and phloem.

MATERIAL AND METHODS

The material for investigations on bark development was taken from natural fir stands (average tree age 30 years) in the years 1966 and 1967 at 15-day intervals during the vegetation period and at 30-day intervals in the period of dormancy. For microscopic examination sections of the stem aged 1, 2, 3, 4, 5, 10, 15 and 20 years were taken, each time at mid length of the annual elongation (Diagram 1). The material was collected in the state forest district Gromnik, forestry Kańska Górna, section 109a. Each time the tree was felled and the stem length, the particular annual increment in length and the age of the tree were determined and segments were cut at mid length of the annual length increments and the N—S direction was marked on them. The basic investigations on development, the results of which are reported here, were performed on material collected in 1967.

Anatomical-histological examination was carried out on living or fixed in Cr-A-F (0.5—0.5—20) material. For staining live tissues iodine-potassium-iodine, anilin blue and erythrosine were used. Observations on living tissues were made on cross, longitudinal-radial and longitudinal-tangential sections prepared by means of a razor or gilette blade. Parti-

*Schematic drawing
of tree stem, sites
of sampling marked,
Scale 1:30*

*Scheme of bark
structure $\times 30$*

*Scheme of annual
secondary phloem
increment structure*

Diagram 1. Schemes of tree stem, bark structure and annual secondary phloem

cular elements of the differentiating secondary phloem were also prepared out. For this purpose pieces of bark 4×8 cm in size were separated from the wood, stained in an aqueous erythrosine or anilin blue solution, and then with a sectioning needle the newly differentiated elements of secondary phloem were prepared out. Good results were also obtained by making incisions and tearing off particular layers of secondary phloem. By the application of these methods it was possible to observe several intact sieve cells (Plates III and IV), an achievement difficult to obtain when using the microtome, in view of the considerable length of these cells.

The fixed material was embedded in paraffin (soft tissues) and celloidin (hard tissues). Cross, longitudinal-radial and -tangential sections $10-50 \mu$ thick, prepared by means of a rotary or sliding Reihert microtomes, as well as free-hand sections were examined. The fixed material was stained after Cheadle, Gifford and Esau (1953). Potassium hydroxide and sudan III were used for staining suberised walls, floroglucine with hydrochloric acid served for staining lignified walls, and carmi-

Diagram 2. Percent of cross section surface area occupied by cell lumen in various types of functional sieve cells.

S₁ — sieve cells overwintering in a partially differentiated state; S₂ — early phloem sieve cells; S₃ — sieve cells of transitional zone; S₄ — late phloem sieve cells

ne for staining cellulose walls. Starch distribution in bark tissues was investigated by means of Lugol's solution, the presence of tannins was revealed by iron chloride, resinous substances were stained with copper acetate. Schultze's reagent was used for maceration.

The microphotographs were made in an N.F.p.K. — Plan Zeiss microscope with an M.F. Zeiss attachment. Microscopic measurements were performed in the same microscope.

To obtain information on the relation of wall thickness in sieve cells to their size, the dimensions of these cells were measured as well as their lumen diameter. Preliminary investigations demonstrated differences in the size of the sieve cells and in the thickness of their walls on cross sections of different-aged shoots. In connection with this, measurements were performed on shoots aged 1, 2, 3, 4, 5, 10, 15 and 20 years,

taken from trees at the following dates: Feb. 15, Apr. 1, May 1, June 1, July 1, Aug. 7, Sept. 7, Oct. 5, Nov. 28. The cell diameter was measured from middle lamella to middle lamella, and of the cell lumen from cell wall to cell wall. The difference between the cell diameter and the cell lumen divided by two gives the thickness of the cell wall. In each group ten cells were measured in radial and tangential direction, and the arithmetic mean was calculated. The radial and tangential diameter of the cell and of its lumen being known, the surface area of the cross section occupied by the cell lumen could be calculated as well as the surface area of the cross section of the whole sieve cell. Then the percent of the cross section surface area occupied by the cell lumen and the cell wall was calculated. The method of measurement was similar as that described by Esau and Cheadle (1958).

CAMBIUM

Introduction. In order to understand the ontogenesis and interrelations of the secondary phloem elements, a knowledge of the structure of the cambium and of the way of division of its cells are necessary. The meaning of the term cambium has been discussed in more detail by Bannan (1955). It denotes not only the layer of initial cells, but also the zone of undifferentiated or dividing mother cells of the xylem and phloem (Bannan, 1955; Esau, 1965).

Cambium consists of two types of cells: elongated fusiform cells with tapering ends and isodiametric ray initials (Bannan, 1955, 1962; Srivastava, 1963a and b; Esau, 1965).

The radial row of cambium-derived cells arising by way of periclinal division both on the xylem and phloem sides is called the radial tier (Esau and Cheadle, 1955; Cheadle and Esau, 1958).

The changes occurring in the cambium in connection with growth in girth are described in detail by Bailey (1923). According to this author the increase in the circumference of the cambium in gymnosperms occurs by an increase in length, in the tangential diameter, and in the number of cambial fusiform initials and as well as an increase in the tangential diameter and the number of ray initials. These changes occur owing to intrusive growth, anticlinal division and the loss of cambial fusiform initials (Klinken, 1914; Bailey, 1923; Whalley, 1950; Bannan, 1951, 1953, 1956, 1957, 1965; Berghoorn, 1940, 1941; Evert, 1960; Hejnowicz, 1961, Srivastava, 1963 and others).

Bailey (1923), Whalley (1950) and Bannan (1957) claim that the anticlinal divisions are of pseudotransversal character. According to Bannan (1956), Hejnowicz (1961) and Whalley (1950) intensive intrusive growth occurs after anticlinal division. Bannan

Plate I

Bark tissues of 4-year-old stem

- A — cross section. Material collected on July 1. $\times 50$
 - B — longitudinal radial section of secondary phloem. Material collected on May 1. $\times 100$
 - C — tangential view of sieve cells with vertically oriented strands. Material collected on July 1. $\times 250$
 - D — phloem parenchyma cells from conducting phloem zone, longitudinal tangential section. Material collected on June 1. $\times 250$
 - E — longitudinal tangential section through nonconducting phloem. Material collected on June 1. $\times 100$
- C — cambial zone, CO — cortex, CP — crushed nonfunctional phloem, FP — functional phloem, IS — intercellular spaces, PD — periderm, PS — parenchyma strands, R — ray, RC — resin canal, RS — resin space, S — sieve cell, Scl — sclereids, SP — secondary phloem, st — strands, W — secondary wall

Plate II

Development of secondary phloem in 2-year-old stem

- A — cross section. Material collected on May 1. $\times 200$
- B — cross section. Material collected on June 1. $\times 200$
- C — cross section. Material collected on June 1. $\times 585$
- D — longitudinal radial section. Material collected on June 1. $\times 585$
- E — cross section. Material collected on July 1. $\times 200$
- F — cross section. Material collected on Aug. 7. $\times 200$
- G — cross section. Material collected on Sept. 7. $\times 180$
- H — longitudinal radial section. Material collected on May 1. $\times 180$
- I — Longitudinal radial section. Material collected on May 1. $\times 250$.

Ar — Albuminous cells of radial system, Av — albuminous cells of axial system, C — cambial zone, IS — intercellular spaces, PS — parenchyma strands, R — ray, S_1 — sieve cells overwintering in partly differentiated state, S_2 — early phloem sieve cells, S_3 — sieve cells of transitional zone, S_4 — late phloem sieve cells, SA — sieve area, X — xylem

Plate III

A, B, C, D, E — Fragments of prepared out sieve cells differentiating from the cells overwintered in partly differentiated state. Five-year-old stem. Material intravitaly stained in aqueous erythrosine solution. Material collected on Apr. 17. Microphotograph A enlarged $\times 300$. Microphotographs B, C, D, E enlarged $\times 760$.

F, G, H, I — Fragments of prepared out early phloem sieve cells. Five year-old stem. Material intravitaly stained in aqueous erythrosine solution. Material collected on May 1. $\times 300$

N — nucleus, *n* — nucleolus, *SA* — sieve area, *SB* — slime bodies

Plate IV

Differentiating early phloem sieve cells

Ten-year-old stem. Material intravitaly stained in aqueous erythrosine. Material collected on June 1

A — cross section through secondary phloem. $\times 250$

B, C, D, E — fragments of prepared out differentiating sieve cells.

Microphotographs B and C enlarged $\times 120$, microphotographs D and E enlarged $\times 300$

C — cambial zone, Cp — nonfunctional phloem, Fp — functional phloem

(1956) and Hejnowicz (1961) report that, after the loss of the fusiform initials of the cambium, a rapid intrusive growth and frequent anticlinal divisions occur in the remaining fusiform cells of the cambium.

The cambial fusiform initials formed in the pseudotransversal division in the course of intrusive growth undergo symmetric or asymmetric periclinal divisions. The row of cells formed in symmetric periclinal divisions is called the regular tier (Srivastava 1963 a, b). The cambial fusiform cells undergoing asymmetric periclinal division give rise to daughter cells of unequal lengths. The longer cells differentiate as elements of phloem and xylem, and the shorter ones remain in the cambial zone and serve as initials in subsequent divisions (Bannan 1953; Srivastava, 1963 a). As the result of further asymmetric periclinal divisions, the original length of the initials is reduced. This process leads either to a complete loss of the fusiform initials in the cambial zone or to the formation from their part of new ray initials (Whalley 1950; Bannan 1956, 1960; Bannan and Bayly 1956). The rows of cells formed in the process of asymmetric periclinal division are called declining tiers (Srivastava 1963).

Own observations. In studies on development, three stages may be distinguished in the cambium activity:

- 1) a period of winter dormancy (it is in this period that division does not occur in the cambial zone);
- 2) a period of radial growth of the cells of the cambial zone (preparatory phase for future division in the cambial zone);
- 3) period of division of initials in the cambium.

The division of cambium initials occurs earlier in the apical part of the tree and latest in the zone of the root neck. Investigations on material collected on April 15, 1967 showed divisions of cambial cells in the apical parts of the stem (age 2, 3, 4, 5 and 10 years), whereas in the lower parts of the trunk they did not yet occur (age 15 and 20 years). Owing to division of the cambial initials, elements of early phloem (Apr. 7 to May 1), of early xylem (Apr. 20 to June 1) of late phloem (May 15 to July 1) and of late xylem (June 15 to Aug. 30) are successively formed.

The period of radial growth of the cells of the cambial zone is preceded by a period of cambium initials division about 15—30 days earlier. It occurs owing to a considerable increase in the radial diameter of the cells of the cambial zone. Investigations demonstrated that, simultaneously with the radial growth of the cambial initials, there occurs a process of differentiation of sieve cells from the cells overwintering at the border of the cambial zone. In 1967 the period of radial growth of the cells of the cambial zone fell to the last days of March and the first half of April.

Division in the cambial ends first in the upper part of the stem (around mid August), and much later in the remaining parts of the trunk

(to the end of August). In the period of dormancy the width of the cambial zone varies from 4 to 6 cells, and the cell walls, particularly their radial walls, thicken greatly.

CONDUCTING PHLOEM

1. Ontogenesis of sieve cells overwintering in a partly differentiated state

Conducting phloem in *Abies alba* consists of the following elements: sieve cells, phloem-parenchyma cells, parenchyma of pith rays, albuminous cells and crystal cells.

In the increment of secondary phloem, early and late phloem can be distinguished. The first elements differentiating in the vegetation period are the sieve cells which arise from cells overwintering in partly differentiated state at the border of the cambial zone. In the material investigated, mostly two layers of partly differentiated sieve cells were found, forming the border between the cambial zone and secondary phloem. On the cross section the cells of the inner layer are smaller than those of the outer one, their size exceeds, however, at least two times the size of the cells of the cambial zone. They also differ from the cambial zone cells by their thick tangential walls; they exhibit dense cytoplasm in their apical parts. The nucleus is distinctly shifted to one end of the cell and elongated, much larger as compared with the nuclei of cambial zone cells, it also stains more intensively and has a homogeneous structure (nucleoli cannot be distinguished). The processes of differentiation of the outer layer cells in late autumn are more advanced, nevertheless the walls of these cells are about two times thinner as compared with those of the sieve cells of the late phloem. As compared with the latter cells they contain less slime bodies they are completely deprived of callose and have no developed sieve areas. In the second half of October they are partly crushed along the radial axis. In two-year shoots crushed cells were observed as early as the end of September. In the winter period the processes of differentiation of these cells are completely inhibited. They are reactivated simultaneously with radial growth of the cells of the cambial zone. In the spring process of their differentiation noteworthy is their intrusive growth. In the growing apical part of the cell a large amount of cytoplasm is present with distinctly delimited dark granular agglomerations (Plate III, A, B). At a certain distance from the apical part there is a greatly elongated nucleus surrounded by large quantities of slime substances (Plate III, A, C). After the end of intrusive growth, the amount of slime bodies decreases, the plasma adheres to the walls, and inside the cell there appear thin internal strands running often parallel to the cell length. In the course of differentiation the shape of the nucleus changes (Plate III C, D), the nucleoli become visible again. In

Diagram 3. Scheme of formation of annual ring of secondary phloem in the successive annual length increments of the stem of a 30-year-old fir

Diagram 4. Certain processes occurring in the bark of *Abies alba* within one year (1967).

S_1 — sieve cells overwintering in partly differentiated state; S_2 — early phloem sieve cells; S_3 — transitional zone sieve cells; S_4 — late phloem sieve cells; X_1 — early xylem; X_2 — late xylem

the end stage of differentiation the nucleus disintegrates (Plate III E), sieve areas form in the radial walls with pores lined with a thin callose layer. Towards the end of functioning of these cells the amount of callose slightly increases. In nonconducting phloem definitive callose was not found in overwintering crushed cells. The period of functioning of these cells varies from 4 to 6 weeks (Diagrams 3 and 4).

2. Early phloem

The sieve cells of early phloem form as the result of the first initial cells divisions in the cambium. In 2-, 3- and 4-year-old shoots the early phloem elements were produced before Apr. 15. The same elements in 5-, 10- and 20-year shoots formed in the period Apr. 15 to May 1. The first divisions in the cambial zone were observed in 5—10 year shoots in the material collected on Apr. 15.

Observations concerning the ontogenesis of sieve cells of the early phloem confirm in general the results of Evert and Alfieri (1965)

Plate V

Characteristic of secondary phloem in 5-year stem

Material collected on May 1

- A — cross section through secondary phloem. $\times 60$
 Microphotographs B, C, D, E, F, G and H show the successive fragments of the tissues presented in microphotograph A beginning with cambium (photo B) up to the first sclerification (photo H). $\times 585$
- B — early phloem formed up to May 1, 1967 and late phloem formed in 1966
- C — late phloem formed in 1966
- D — early phloem formed in 1966 and fragments of late phloem formed in 1965
- E — fragment of late phloem and early phloem formed in 1965
- F — late phloem and fragment of early phloem formed in 1964
- G — late phloem and early phloem formed in 1963
- H — sclereids within secondary phloem formed in 1962
- Av — Albuminous cells of axial system, IS — intercellular spaces, PS — parenchyma strand, R — ray, Scl — sclereids

Plate VI

Sclerification of secondary phloem

- A, B — three-year shoot. Material collected on Aug. 7. Cross section through secondary phloem. At border between secondary phloem and primary cortex, the first sclereid layer is forming (Sc1₁). $\times 200$
- C — five-year stem. Material collected on Nov. 28. Cross section through secondary phloem and fragment of primary cortex. $\times 60$
- D — fifteen-year stem. Material collected on Nov. 28. Cross section through secondary phloem. Within it three layers of sclereids (Sc1₁, Sc1₂, Sc1₃). $\times 60$
- E — fifteen-year stem. Material collected on Nov. 28. Fragment of secondary phloem on cross section. Resin spaces visible (RS) formed between 2nd (Sc1₂) and 3rd (Sc1₃) sclereid layer. $\times 200$
- F — fifteen-year stem. Material collected on June 1. Fragment of secondary non-conducting phloem on longitudinal radial section. The two outer layers of sclereids (Sc1₁ and Sc1₂) are visible. $\times 60$
- C — twenty-year stem. Material collected on Feb. 15. Fragment of secondary phloem on cross section, 4th (Sc1₄) and 3rd (Sc1₃) sclereid layer are visible. $\times 60$
- H — twenty-year stem. Material collected on Feb. 15. Fragment of 4th sclereid layer on cross section. $\times 585$
- I — five-year stem. Material collected on Nov. 28. Sclereids prepared out of 1st layer. $\times 60$
- J, K — ten-year stem. Material collected on Nov. 28. Sclereids prepared out of 2nd layer. $\times 150$.
- L, M, N, — fifteen-year stem. Material collected on Nov. 28. Sclereids prepared out of 3rd layer. $\times 300$

as regards other species of *Pinaceae*. The first stage in the ontogenesis of the sieve cell is a considerable increase in the cell volume. In the cytoplasm contiguous to the wall of the differentiating sieve cell of early phloem there appear: the cell nucleus, plastides, mitochondria and other minute grains described in detail by Abbe and Crafts (1939). It should be stressed that in the course of maturation of the sieve cell, its walls thicken greatly, particularly the tangential ones (Plate III *F, G, H, I*; Plate IV). In the end stage of differentiation the plasmatic internal strands appear most distinctly within the surface of the sieve area in which plastides, mitochondria, and other granular formations are present. Minute starch grains staining violet with Lugol's solution were observed in the plastides. Towards the end of functioning of the sieve cell the amount of callose in it increases up to the formation of definitive callose.

The period of functioning of sieve cells in early phloem is 30—45 days (Diagrams 3 and 4). It should be mentioned that sieve cells differentiated earlier are functional for a shorter time (ca. 30 days) as compared with those differentiating later. The period of functioning of the sieve cells was observed to be dependent on the increments of early phloem. In 2- and 3-year-old shoots the early phloem increment constitutes about 8 sieve cells in radial file, functioning for 3—4 weeks. In 15- and 20-year-old shoots the radial row of early phloem consists on the average of 4 sieve cells, and the time of their functioning is 5—6 weeks. These observations point to a dependence between the duration of the differentiation process of the sieve cell and the time of its later functioning.

The early increment of secondary phloem is built of 4—8 layers of sieve cells and one of phloem parenchyma cells (Plates II and V). The layer of phloem parenchyma cells separates the sieve cells of early phloem from the those of late phloem. The greatest growth increments of early phloem are noted in 2- and 3-years-old shoots. Early phloem is built of regular tiers. In 15- and 20-year-old shoots, declining tiers formed of albuminous cells of the axial system occur sporadically.

Measurements of the length of early phloem sieve cells showed a wide variation in the dimensions of these cells within the tree material studied (collected on June 1). In the 2-year shoots the length of the sieve cells is on the average 900—1000 μ , in 5-year-old shoots 1300—1400 μ and in 15- and 20-year shoots it reaches 2000—2100 μ .

Measurement of the cross section and lumen of early phloem sieve cells, and the percent of the surface area of the cross section occupied by the cell lumen and wall, calculated from these measurements lead to the following conclusions (see diagram 2): 1) in the apical part (1-, 2-, 3- and 4-year shoots) the surface area of the cross section increases successively (from 98—520 μ^2); 2) in 5-, 10-, 15- and 20-year shoots the surface area of the cross section shows no tendency to increase, oscillating within 260—550 μ^2 ; 3) the percent of the cross section area occupied by the cell

wall is highest in the apical part of the tree and gradually diminishes in the subsequent lower situated parts of the stem; 4) in the period May 1 to June 1, the sieve cell walls in early phloem greatly thicken (as shown by measurements performed on material collected on May 1 and June 1); 5) in the period July 1 to Aug. 7, the wall thickness slightly decreases in the early phloem sieve cells; 6) a slight reduction of the cell wall thickness, and particularly of the tangential walls, is noted in the nonconducting sieve cells of early phloem.

3. Late phloem

In the late phloem of shoots up to 5 years old a zone of transitional phloem is distinguished (Plate II). This could be done on the basis of the structure of the sieve cells (wall thickness, lumen diameter), the date of formation and the period of their functioning (Diagrams 2, 3, 4). The sieve cells of the transitional zone function only in one vegetation season, whereas those of late phloem overwinter alive and function for a certain time in the next season.

The process of late phloem elements differentiation is similar to that of the sieve cells of early phloem. In the cytoplasm of the differentiating cells, a large number of mitochondria was found; the sieve cells of late phloem contain, moreover, a large quantity of slime bodies. Only in few preparations were transcellular internal strands observed.

The transition zone of late phloem consists of 2—4 cell layers. It is separated from the late phloem by a layer of parenchyma cells. These cells in late phloem, unlike those arising in early phloem, do not form a regular tangential strand. In the phloem, of the transitional zone declining tiers formed of albuminous cells of the axial system and of crystal cells are visible. The sieve cells of the transitional zone form in the period May 15 to June 1. The process of their differentiation last on the average 2—4 weeks, and the period of their functioning exceeds 2 months.

Late phloem sieve cells are produced in the apical part of the tree stem (1-, 2- and 3-year shoots) in the period July 1 to Aug. 1, in 4- and 5-year shoots between July 1 and July 15, and in 10-, 15- and 20-year shoots between May 15 and June 1. The process of differentiation of late phloem sieve cells lasts about 4—6 weeks, the first cells differentiating within about 4 weeks, and the later ones taking longer (about 6 weeks). In the last days of September and the beginning of October the sieve cells of late phloem accumulate large quantities of callose. During radial growth of the cambial zone cells (early spring) the amount of callose in these cells decreases. In the apical part (2-, 3-, 4- and 5-year shoots) the overwintering sieve cells of late phloem function up to May 1, in 10- and 15-year shoots up to May 15, and in 20-year shoots up to June 15. It

results from the foregoing that the late phloem sieve cells in the lower part of the tree stem function longest, for a period of about one year.

Late phloem consists on the average of 4 layers of sieve cells and one layer of phloem parenchyma cells. The latter do not form a regular tangential strand. Radial rows of late phloem sieve cells not including phloem parenchyma cells are sporadically observed. The declining tiers of albuminous cells of the axial system and crystal cells are numerous. Partly differentiated sieve cells overwintering at the border of the cambial zone, form in the apical part (1- and 2-year shoots) in the first half of August, and in the remaining part of the shaft in its second half. The process of differentiation of these cells is inhibited in the first half of October.

Measurements of the surface area of the cross section of late phloem sieve cells and of their lumen, and the percent of this surface occupied by the lumen and cell wall calculated therefrom lead to the following conclusions (see diagram 2): 1) the cross section surface area of late phloem sieve cells varies within the limits 180—640 μ^2 , the cells in the apical part exhibiting mostly a smaller cross section; 2) the percent of the cross section surface area occupied by the cell wall is highest in the one-year shoot and gradually diminishes in lower parts of the stem; 3) the thickness of the cell walls is maximal in the first half of August and does not change till mid September, then they become somewhat thinner and remain so throughout the winter period; 4) in spring the thickness of the overwintering late phloem sieve cell walls slightly increases; 5) the walls of the nonconducting sieve cells of late phloem and particularly those of the tangential ones also decrease in thickness.

Staining of cell walls of conducting phloem sieve cells confirms the results of Srivastava (1963) who claimed that the secondary wall of the sieve cells in *Pinaceae* stains less intensively as compared with the middle lamella.

4. Development of conducting phloem in the vegetation season

Diagram 3 shows the picture of the dynamics of development of conducting phloem in the course of the vegetation season in the particular mid parts of annual length increments of a 30-year-old fir. On the axis of ordinates, sieve cells formed successively in the vegetation season are laid off. The axis of abscissae presents the development of secondary phloem in the examined parts of the trunk. The bars denote the number of sieve cells formed in various periods of the vegetation season (the conducting cells are denoted by smaller black bars and the overwintering late phloem sieve cells by black-and-white bars). The diagram, moreover, shows the changes in the girth of the secondary phloem in the

vegetation season in various parts of the shaft. It may be generally said that, the zone of conducting functional phloem is widest in May, June and up to mid July, and from Aug. 15 to Sept. 15.

5. Characteristics of the parenchymal elements

Cells of phloem parenchyma in *Abies alba* occur in the vertical strands of the axial system (Plates I, II, V and VI). They arise from transversal division of the phloem mother cells. On the longitudinal section of a phloem parenchyma cell they are rectangular. On the cross section in the zone of conducting phloem their shape is close to rectangular, but in nonconducting phloem their volume increases gradually and they assume an oval shape (Plate V). The walls of these cells are not lignified, they have pitted areas joining the parenchyma cells with one another and with the ray parenchyma. The phloem parenchyma cells contain starch grains, tanniferous substances, resins and lipids. They form on the cross section more or less regular tangential cell strands. The number of phloem parenchyma strands formed in the course of a vegetation season varies within the stem at different levels (Diagram 1).

Albuminous cells occur both in the axial and in the radial system. The albuminous cells of the axial system may occur in the form of single vertical cell strands similarly as do the phloem parenchyma cells (Plate II H). Most frequently, however, they form radial plates of cells (Plate II I). Analysis of the radial tiers of conducting phloem and of the cambial zone indicates the following possibilities of formation of albuminous cells of the axial system: 1) as the result of transverse divisions of phloem mother cells (single strands form); 2) owing to further periclinal divisions of the transversely divided phloem mother cells. These processes lead to a decline of the radial tiers, the formation of radial plates of cells being only the initial phase of this process.

In *Abies alba* the albuminous cells of the radial system (Plate II C, D) occur most frequently on both sides of the pith rays. These cells are an element of the rays and arise in the process of their formation. This process has been extensively discussed by Chrysler (1913), Barghoorn (1940); Bannan (1950, 1951, 1953, 1965); Srivastava (1963a, b) and Den Outer (1967).

The albuminous cells of the axial and radial systems, in contrast to phloem parenchyma cells, communicate with the sieve cells through the sieve areas. These areas connecting sieve cells with radial albuminous cells have much larger pores as compared with those of the axial system. Moreover, in the sieve areas connecting sieve cells with the albuminous cells of the radial system, callose was found on both sides, whereas on the sieve areas connecting albuminous cells of the axial system with the

sieve cells, either no callose was observed on both sides, or it occurred only on the side of the sieve cell.

In *Abies* one-file pith rays occur. Crystal cells are present both in the axial and in the radial system.

NONCONDUCTING PHLOEM

The changes in nonconducting phloem consist in crushing of the sieve cells, an increase in volume of the phloem parenchyma cells, formation of intercellular spaces, formation of resin spaces and sclerification of secondary phloem (Plate V). In the material studied, formation of the first sclereid layer was observed in a 3-year-old shoot (Diagram 1; Plate VI, A, B). Cells of the phloem parenchyma formed in the first annual ring of secondary phloem undergo sclerification. Within the first annual ring of the 3-year shoot, numerous intercellular spaces are observed formed as the result of tearing off of the crushed sieve cells from the ray parenchyma cells.

In the ontogenesis of the phloem parenchyma cells undergoing sclerification, the following successive processes can be distinguished: 1) increase in volume of the phloem parenchyma cells; 2) accumulation of storage (starch, lipids and tanniferous) substances; 3) enzymatic decomposition of starch grains — the sugars are dissolved in the cell sap; 4) appearance in the cytoplasm of large amounts of plastide-like bodies as well as of mitochondria; 5) intensive growth of the parenchymal cells (they usually intrude into the intercellular space due to the crushing of sieve cells); 6) enzymatic breakdown of the walls of the crushed sieve cells adjacent to the enlarging parenchyma cells; 7) lignification of the parenchyma cell walls; 8) continuation of intrusive growth of the sclereid arms. Very often the sclereid mother cells adhere to the radial cells. The pith rays passing between the extending sclereids were never crushed (Plate VI).

The process of multidirectional growth of the phloem parenchyma cells of the first annual ring, from which sclereids will form, occurs in the period between May 15 and June 15. It consists in the first place in intrusion of these cells into the intercellular spaces. The process of lignification of the cell walls of the forming sclereids is most intensive in the period June 15 to July 15. Intrusive growth of the sclereid arms may be resumed in the next vegetation season. In the period of the 4, 5, 6 and 7th year further filling of the sclereid layer occurs. The phloem parenchyma cells of the second annual ring undergo sclerification.

In the 6-, 7-, 8- and 9-year shoots, under the sclereid layer, intercellular spaces form in which resin spaces grow making the process of continuous thickening of the sclereid layer impossible. In 7- and 8-year shoots, a second sclereid layer forms under the resin spaces. The processes con-

Plate VII

Characteristic of primary cortex

- A — one-year stem. Material collected on Aug. 7. Cross section. $\times 60$
- B — one-year stem. Material collected on Sept. 7. Cross section. Arrows point to crushed parenchyma cells. $\times 60$
- C, D — two-year stem. Material collected on Oct. 5. Cross section. Arrows point to divided parenchyma cells. $\times 200$
- E, F — two-year stem. Material collected on Oct. 5. Strands of divided parenchyma cells on cross section. $\times 585$
- G — three-year stem. Material collected on Aug. 7. Fragment of primary cortex tissue on cross section. Arrows point to crushed parenchyma cells. $\times 200$
- H — five-year stem. Material collected on July 1. Sclereid in primary cortex. $\times 200$
- IS — intercellular spaces, PD — periderm, RC — resin canal, RS — resin space, Sc1 — sclereids

Plate VIII

Characteristic of periderm

- A — newly formed stem. Material collected on May 15. Epidermal cells and hairs on cross section, $\times 200$
- B — one-year stem. Material collected on July 1. Periderm on cross section. $\times 250$
- C — one-year stem. Material collected on Aug. 7. Periderm on cross section. $\times 200$
- D — one-year stem. Material collected on Sept. 7. Periderm on cross section. $\times 200$
- E — two-year stem. Material collected on Sept. 7. Fragment of primary cortex tissues and periderm on cross section. $\times 120$
- F — three-year stem. Material collected on Aug. 7. Periderm $\times 250$
- G — ten-year stem. Material collected on Sept. 7. Stoma cross section. $\times 250$
E — epiderms, P — cork, Ph — phellogen

nected with the formation of this layer are of similar character as the previously described course of formation of the first layer.

The 10-year-old shoots have two sclereid layers. The process of intrusion into the second layer progresses. The sieve cells between the sclereid layers are crushed. In 15-year-old shoots there are three layers of sclereids, and the second and third layer are gradually filled. Between these layers resin spaces are found. The pith rays are wavily bent, and crushing of their parenchyma cells was not observed.

In nonconducting phloem in 20-year shoots four layers of sclereids are present. Between these layers resin spaces occur, frequently surrounded by phloem parenchyma cells.

PRIMARY CORTEX TISSUES

In the primary cortex tissues the following types of parenchyma cells can be distinguished (Plate VII):

- 1) cells capable of division,
- 2) cells containing chloroplasts,
- 3) cells containing ergastic substances (starch grains, tanniferous substances, lipids),
- 4) cells containing resinous substances (resin spaces),
- 5) epithelial cells lining resin canals,
- 6) parenchyma cells surrounding resin canals.

In shoots with primary structure (May 15), cells constituting the primary cortex have a dense cytoplasm, large nuclei and are regularly distributed. The parenchyma cells in the inner zone of primary cortex are larger as compared with those of the peripheral zone, and form a more distinct system of intercellular spaces.

The formation of the secondary structure (May 15 to June 15) is connected with essential changes in the primary cortex structure. The parenchyma cells containing tanniferous substances greatly increase in volume, particularly those above the phloem. The parenchyma cells containing chloroplasts do not increase in volume, but their walls were found to thicken somewhat. Resin spaces considerably increase in volume, but their walls remain thin. The resin canals also increase, the process being preceded by epithelial cell division.

In the period up to July 15, death of the enlarged parenchyma cells, above all over the phloem, was observed. In primary cortex tissues large spaces form, owing to stretching, crushing and tearing of the above described cells. Parenchyma cell division was observed in August and September. The newly formed tissue fills the intercellular spaces. In August and September the death of developed parenchyma cells progresses. The remaining walls of the crushed cells undergo enzymatic

breakdown. A further process of enlargement of the resin canals and resin spaces was observed. Three layers of collenchyma cells containing chloroplasts form under the periderm. In the parenchyma cells containing ergastic substances, the starch content increases. Sporadic divisions of primary cortex parenchyma cells were observed in October. In the period Nov. 1 to March 15 no division of parenchyma cells was noted. In this period an increase in the cytoplasm density occurred as well as an enlargement of nuclei in the divided parenchyma cells. An increased number of mitochondria was also found in cells of this type. These observations were confirmed on material collected in December, January, February and March of the years 1966 and 1967, both in 2-year shoots and older ones. Intensive cell division occurs in the period March 15 to May 15 in the primary cortex parenchyma. This regularity concerns also 3-, 4-, 5-, 10-, 15- and 20-years-old shoots. It should be stressed that cell division in primary cortex parenchyma was observed up to mid October, usually, however, this process is most intensive in early spring. In summer months the divided parenchyma cells increase in volume and their walls thicken. In later months, parenchyma cell division is localised in the inner primary cortex layer. The dividing cells are tangentially elongated and divide generally along radial walls. In two-year shoots, dying of enlarged parenchyma cells was also observed, the cells then were stretched, torn and crushed, and their walls underwent enzymatic breakdown. In several cases large thin-walled parenchyma cells were observed filling the spaces formed by cell tearing. Most frequently, however, these spaces are filled with parenchyma tissue, the cells of which undergo intensive division. Parenchyma cells containing chloroplasts occur in groups, they often form tangential strands and do not increase in volume. The resin canals enlarge. Pairs of divided epithelial cells were most frequently observed in August and September. The resin spaces also increase in volume, they occur above all in the peripheral zone of primary cortex. There are two or three collenchyma cell layers filled with chloroplasts under the periderm.

In 3-year stems cells containing calcium oxalate crystals were observed in the primary cortex. These cells appeared towards the end of August. In this period the formation of the first sclereids was observed within the primary cortex. Sclerification involved parenchyma cells containing ergastic substances. These cells form tangential strands in the peripheral primary cortex layer. In connection with the growth of the tree in girth, the tangential strands of these cells were torn, and generally the marginal cells of these strands underwent sclerification. These cells exhibit multidirectional intrusion growth, filling first of all the intercellular spaces. Then lignification of their walls occurs. This process has a course similar to the previously described sclerification process in secondary phloem. It should be mentioned that the primary cortex sclereids

have shapes more irregular than those formed in secondary phloem. This is due to the looser arrangement of the primary cortex tissues in connection with the continuous structural changes of the cells constituting this zone.

In older stems, all the above described processes also occur. Parenchyma cell division generally occurs in the primary cortex zone directly adjacent to the secondary phloem. In connection with the increase in volume of the shoot, the primary cortex tissues are torn, and the spaces left by them are filled by the dividing parenchyma cells. In the peripheral primary cortex layer the already existing foci of sclereids extend, and new groups of them form. The resin spaces increase in volume. They usually occur in the peripheral primary cortex zone. The walls of these cells are thin and formed of cellulose. Their shape is generally tangentially flattened. Under the periderm two or three layers of colenchyma are visible. In the colenchyma cells chloroplasts are present the year round. The parenchyma cells containing ergastic substances form usually tangential strands running in the outer zone of the primary cortex. In older shoots large resin canals are found. Around the enlarging resin canals no sclereids are seen. Frequently, a partial disappearance of sclereids formed in secondary phloem and contiguous to the extending resin canals was noted.

PERIDERM

In the newly formed (May 15) 4 cm long stem, the epidermal cells have thin cellulose walls, a dense cytoplasm and large cell nuclei. Frequent cell divisions along the anticlinal walls were observed in the epiderm. The epidermal cells produce two- or three-cell hairs (Plate VIII A). In 16—cm shoots (June 1) the epidermal cells develop thick cutinised outer walls. In the cell layer under the epiderm, periclinal division occurs. Shoots collected on June 15 have a periderm consisting of a uniseriate phellogen layer and a layer of differentiating cork cells under the epiderm. Periclinal division of phellogen cells was also observed. In shoots examined on July 1 the periderm consists of two layers of differentiated cork cells and one phellogen layer. The epidermal cells have thick walls impregnated with cutin (Plate VIII B).

From July 1 to Aug. 18 no phellogen cell division is noted. In shoots collected on September 7 the periderm consisted of five to six cork cell layers and one layer of phellogen cells (Plate VIII D). The phellogen cells have large nuclei, some of them undergo periclinal division, large quantities of tanniferous substances were found in them. In one-year shoots phellogen cell division is most intensive towards the end of August and at the beginning of September, and lasts up to mid October. In the first days of October a phelloderm cell layer forms, and in mid October the

phellogen cells undergo their last division and overwinter in this stage. Observation of material collected in the winter period seems to indicate that the process of differentiation of the inner cork cell layer is inhibited towards the end of October. The cells are much smaller than the cork cells differentiated earlier, and they contain large amounts of tanniferous substances.

In 2-year shoots two cork layers form in the period Apr. 15 to May 1. The two inner cork cell layers formed in the preceding year are crushed. In May and June numerous tears were observed in the periderm. Below the tear, intensive phellogen cell division was observed. The first layer of the formed cork cells exhibits lignified outer walls, the remaining ones are thin and impregnated with suberine. Under this layer thin-walled cork cells form. In material collected on July 1, two to three cork cell layers were observed, formed at the sites where the periderm was torn. In material from Aug. 7, four to five newly formed cork cell layers were found, and in material from Sept. 7 under the tear (from outside) the following cells were noted: one cork cell layer with lignified outer walls, two to three layers of crushed cork cells, one layer of phellogen cells and one phelloderm cell layer.

Material collected in mid October exhibited in the entire periderm two newly formed cork cell layers. The phellogen cells were divided. It would result therefrom that between September and October the activity of phellogen is high (Diagram 4).

In 3-, 4- and 5-year-old shoots cell division was observed in the entire phellogen in spring (up to June 1) and in autumn (Sept. 15 to Oct. 15). In July and August intensive division occurs under the tears. Thus, this process resembles that described above.

In the periderm of 10-, 15- and 20-year stems thin- and thick-walled cork cells and phelloid cells were found, the walls of which underwent sclerification (Plate VIII, G). In the periderm cells, large quantities of tanniferous substances are deposited. Up to June 1 there occurs in these shoots a single division of phellogen cells. In the summer months sporadic divisions of phellogen cells were observed under the tears in the periderm. Intensive division in the entire phellogen cell layer was observed between Aug. 16 and Oct. 15. In this period two to four layers of cork cells are formed.

DISCUSSION

In the vegetation period, the cambium activity in the fir (*Abies alba*) shows distinct seasonal variation, both as regards division intensity and the type of phloem and xylem elements produced. Cambium initials division occurs earlier in the apical part of the tree, and latest in the root neck zone. The end of this process has an analogous course. These obser-

vations confirm the results of Klimas (1967) who investigated cambium activity in *Pinus silvestris* in 1962, 1963 and 1964 in Poland.

At the border between the cambial zone and secondary phloem, partly differentiated sieve cells overwinter (Brown, 1915, Elliot, 1935; Abbe and Crafts, 1939; Artschwager, 1950; Bannan, 1955; Srivastava and O'Brien, 1966a). Some of the investigators did not observe the occurrence of partly differentiated sieve elements in the period of winter dormancy (Swarbrick, 1927; Gill 1932; Alfieri and Evert, 1968). In the winter period the processes of cell differentiation are inhibited. Their reactivation occurs in the phase of radial growth of the cambial zone cells. Evert (1963) has some reservations as to the use of the terms "immature" or "partly differentiated" sieve elements in defining these cells. He believes that cambium-derived cells overwintering on the phloem side may undergo in spring periclinal division like cambium initials, therefore they should be considered as a part of the cambial zone. In *Abies alba* no periclinal divisions of these cells were observed in the period of their maturation in early spring. The observations performed seem to indicate that the ontogenesis of these cells differs both from the formation of early phloem sieve cells and of those of late phloem.

The literature concerning the seasonal development of phloem and xylem brings many controversial views on the rhythmicity and terms of the beginning and ending of the formation of these tissues. Many investigators suggest that the formation of xylem precedes that of phloem (Strasburger, 1891; Cockerham, 1930). Knudson (1916) and Lodewick (1928) report that phloem and xylem formation starts and ends simultaneously. The latest investigations, however, demonstrated that, both in gymnosperms and in woody dicotyledonous plants, differentiation of new phloem precedes by several weeks the new xylem differentiation (Evert, 1960, 1963; Davis and Evert, 1965; Derr and Evert, 1967; Klimas, 1967; Alfieri and Evert, 1968). Strasburger (1891) and Cockerham (1930) were of the opinion that inhibition of development of xylem elements occurs earlier than that of phloem elements formation. Bannan (1955) considered that there is no established scheme determining the end of the processes of xylem and phloem formation in a given species. The present studies confirm fully the results obtained by Evert and his collaborators. It has been moreover, established that, as the result of division of cambium initials, the elements of early phloem, early xylem, late phloem and late xylem form successively. This points to a periodicity in the production by the cambium of phloem and xylem elements.

The annual ring of secondary phloem consists of early and late phloem (Huber, 1939, 1949; Abbe and Crafts, 1939; Holdheide, 1951; Holdheide and Huber, 1952; Alfieri and Evert, 1968;

Golinowski, 1968). The structure of the annual secondary phloem ring shows differences along the stem. They concern both the number of cells and the type of the elements constituting the annual ring. The dimensions of the sieve cells also vary within the tree stem. The thickness of the cell walls in relation to the cross section surface area gradually decreases from the apical part of the stem towards the root neck. The length of the early phloem sieve cells increases in the successively lower situated parts of the trunk. This seems to indicate that the differences in the structure of the annual ring of secondary phloem result from the nonuniform activity of cambium in the stem. In the apical part, where the annual secondary phloem rings are large, the sieve cells are short, they have thick walls and a small lumen, whereas in the middle and lower part of the tree, where the annual rings of secondary phloem are small, the sieve cells are long and have a large lumen and relatively thin walls.

Abbe and Crafts (1939) and Holdheide (1951) report that sieve cells differentiated at the beginning of the vegetation season have walls thinner and a lumen larger than those differentiated in the second part of the vegetation season. The measurements of the surface area of the sieve cells cross sections both in early and late phloem, and of their lumen, and calculation on this basis of the percent of the cross section occupied by the cell lumen and cell wall confirm these observations. It was also found that: 1) the percent of cross section occupied by the cell walls is highest in the apical part of the tree and gradually decreases in its successive lower situated parts; 2) in the period May 1 to June 1 there occurs a considerable thickening of the sieve cell walls in early phloem; 3) in late phloem the cell wall thickness reaches its maximum in the first half of August, and does not change any more till mid September. Then the walls become somewhat thinner and overwinter in this state. In the period of spring functioning of these cells their walls thicken again. The changes in cell wall thickness in the period of functioning of the early and late phloem sieve cells indicate that the substances forming the cell wall, and the structure of these walls have an essential influence on the process of translocation. Buvat (1963a and b) found that, in the sieve tubes of *Cucurbita pepo*, the endoplasmic reticulum forms numerous vesicles which are then forced into the cell wall. Buvat suggests that these vesicles are the result of pinocytosis and accumulate substances derived from the necrotic cell walls, which pass into the cells or, inversely, they translocate material indispensable for callose and cell wall production. Esau and Chandle (1958) demonstrated in dicotyledons a wide variability of the wall thickness in functional sieve tubes and they are of the opinion that this fact should be taken into consideration in the elucidation of translocation.

In most of the species studied, secondary phloem formed in the annual ring functions only over one vegetation season (Esau 1939, 1950; Hu-

ber, 1939). The latter author claims that, in some species (including the fir), mature sieve elements overwinter in live state and function until new ones are formed. These results were confirmed in the investigations of Alfieri and Evert (1968) as well as in the present study. It was found, moreover, that the period of functioning of early phloem sieve cells amounts to 30—45 days, and of those of the transitional zone exceeds two months, whereas late phloem cells function for about one year. It would result therefrom that the period of functioning of sieve cells in *Abies alba* is short. It seems that conduction of assimilation products occurs also in sieve cells in the last phase of their differentiation (see Plate IV). These observations agree with the suggestion of Kollmann and Schumacher (1964) who report that in *Metasequoia glyptostroboides* the sieve cells in intermediate stages of differentiation can conduct assimilation products. Kollmann (1965) affirms that in *Metasequoia*, assimilates labelled with ^{14}C accumulated in the region of youngest sieve cells between the cambium and the first fiber strand.

In mature sieve cells the pores on the sieve areas are lined with a thin callose layer forming a ring through which the internal strands connecting the protoplasts of various cells pass. The mature and conducting sieve elements contain relatively little callose (Esau, 1961; Ullrich, 1962). With ageing of the sieve elements, the amount of callose lining the pores increases (Abbe and Crafts, 1939; Esau 1939; Srivastava, 1963). Callose is also deposited over the entire surface areas (Esau, 1939, 1950, 1965). Accumulation of callose in large amounts indicates that the activity of the sieve cells has ceased. The callose mass at this stage of development of the sieve element was named by Lecomte (1889) definitive callose. In the process of dying of the sieve cell, first the internal strands, then the contents of the cell disappear, and after a certain time callose gradually also disintegrates (Evert and Alfieri, 1965).

Some investigators believe that the functional sieve elements do not contain callose, and that the large amount of callose in fixed material is an artefact (Zimmermann, 1960; Evert and Derr, 1964). Other scientists consider that callose occurs in functional sieve elements (Engleman, 1965; Esau and Cheadle, 1965). In the ontogenesis of sieve cells in *Abies alba* various quantities of callose were observed both on the sieve areas connecting the sieve cells with one another and those connecting the sieve cells with albuminous cells. This leads the present author to the supposition that callose occurs in functional sieve cells.

Strassburger (1891), Hill (1901), Grillos and Smith (1959) and Srivastava (1963) claim that sieve cells are connected with albuminous cells by one-sided sieve areas which develop only on the side of the sieve cell on the common wall. Evert and Alfieri

(1965) do not confirm this view. While investigating the sieve area connecting sieve cells with albuminous cells, they recorded the following cases: 1) absence of callose on both sides, 2) callose on both sides, 3) callose only on the side of the sieve cell. Srivastava and O'Brien (1965b) did not find callose in the pores forming the sieve area which joins the sieve cells with albuminous cells. It is suggested on the basis of the present observations that the albuminous cells of the axial system have a different development determination than those of the radial system.

The following differences were found between the albuminous cells of the axial and radial systems: 1) the albuminous cells of the axial system arise as the result of transversal division of phloem initials. This is the initial phase of the process leading to the decline of the radial file, and it occurs usually in late phloem. Albuminous cells of the radial system are a functional element of the pith ray. The process of formation and differentiation of these cells is closely correlated with its formation; 2) formation of sieve areas connecting the albuminous cells of the radial system with the sieve cells occurs beyond the cambial zone and is the last stage of differentiation of the conducting system of secondary phloem. The sieve area of the albuminous cells of the axial system, connecting them with sieve cells, differentiates within the cambial zone. This points to a correlation between the processes of differentiation of these two types of secondary phloem elements; 3) the radial system of albuminous cells have more numerous better developed sieve areas as compared with the cells of the axial system; 4) the albuminous cells of the radial system are connected with the sieve cells generally by two-sided sieve areas. The albuminous cells of the axial system are connected with the sieve cells by one-sided sieve areas which develop only on the side of the sieve cell on the common wall. Surface areas were also observed on which there was no callose on either side; 5) the axial system of albuminous cells seem to be very active physiologically in the period of radial growth of the cambial zone cells. This activity is connected with the reactivation of differentiation of the partially differentiated sieve cells overwintering on the border of the cambial zone, and with the beginning of division of the cambial initials.

The changes in nonconducting phloem consist in crushing of the sieve cells, enlargement of the phloem parenchyma and differentiation of some of the phloem parenchyma cells to sclereids (Shimakura 1936; Holdheide 1951; Chang 1954; Srivastava 1963; Golinowski 1968).

Shimakura (1936) claims that the secondary changes in the primary cortex of *Abies firma* consist above all in the enlargement of parenchyma cells. Holdheide (1951) calls attention to the formation of sclereids within the primary cortex. In *Abies alba* the following proces-

ses were observed in the primary cortex tissues: enlargement and death of parenchyma cells, secondary division of these cells, division of the epithelial cells of the resin canals, formation of resin spaces and of sclereids.

Investigations of the peridermis of *Abies alba* confirm in general the results of Moeller (1882), Holdheide (1951) and Chang (1954). The cork is composed of thin-walled and thick-walled layers. Sometimes the cork layer contains cells which do not undergo suberisation. This type of cork was called phelloid by Höhnelt (1878). In one-year shoots phellogen cell division is most intensive towards the end of August and at the beginning of September. In older shoots, one or two cork cell layers are formed in spring. A high phellogen activity is noted in September. The stomata in fir contain 3—10 layers of stone cells. They greatly resemble those of the pine which have been described in detail by Wutz (1955).

CONCLUSIONS

1. In the vegetation period, the cambium activity in *Abies alba* shows seasonal variation as regards intensity of division, and the type of phloem and xylem elements formed.

2. At the border of the cambium and secondary phloem zone, partly differentiated sieve cells overwinter. Reactivation of the processes of differentiation in these cells occurs in the phase of radial growth of the cambial zone cells. The present observations indicate that the ontogenesis of these cells differs both from the development of early and late phloem sieve cells.

3. Division of cambial initials occurs earliest in the apical part of the tree, and latest in the root neck zone, the end of this process runs a similar course.

4. Owing to cambial initials division, elements of early phloem, early xylem, late phloem and late xylem form successively. This points to a periodicity in the formation of phloem and xylem elements by the cambium.

5. The structure of the annual ring in secondary phloem shows differences within the stem. They concern both the number of cells, and the type of elements constituting the annual ring. The dimensions of the sieve cells also vary in the trunk. The thickness of the cell wall in relation to the surface area of the cross section gradually decreases from the apical part of the shaft towards the root neck. The length of the early phloem sieve cells increases in the successively lower situated parts of the stem. This seems to suggest that the variability in the structure of the annual ring of secondary phloem is due to nonuniform activity of the cambium in the stem. In the apical part, where the secondary phloem annual ring is large, the sieve cells are short, thick-walled with a small

lumen, whereas in the middle and lower part of the tree, where the annual secondary phloem rings are small, the sieve cells are long, they have a wide lumen and relatively thin walls.

6. The period for which the particular types of sieve cells function in the secondary phloem also differs widely. The present investigations indicate that both the structure and the period of functioning of the particular sieve cell types are determined by various physiological requirements of the tree. This problem should be further studied from the cytological and physiological point of view.

7. The results obtained suggest a different developmental determination of the albuminous cells of the axial and radial systems.

8. Changes in the nonconducting phloem lead to differentiation of the phloem parenchyma cells to sclereids. The course of this process is not equal throughout the stem. The ontogenesis of the phloem parenchyma cells requires further elucidation.

9. In primary cortex tissues there occur, in connection with the increase of the tree girth, the following secondary processes: crushing and tearing of the parenchyma cells, division of parenchyma cells, enlargement of primary resin canals and sclerification of some of the cells of the primary cortex parenchyma.

10. In the vegetation period phellogen activity exhibits seasonal differences both as regards the intensity of division and type of the cork cells produced.

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Budowa anatomiczna kory jodły pospolitej (Abies alba Mill.).

I. Rozwój tkanek korowych

Streszczenie

Celem pracy było zbadanie zmian jakościowych i ilościowych tkanek korowych w pniu jodły w związku z długoletnią działalnością miazgi.

Materiał do badań rozwojowych pobierano w naturalnym drzewostanie jodłowym (przeciętny wiek 30 lat) w latach 1966 i 1967 w odstępach 15-dniowych w okresie wegetacji oraz w odstępach 30-dniowych w okresie spoczynku. Próbkę do badań mikroskopowych stanowiły wyrzynki strzały w wieku: 1, 2, 3, 4, 5, 10, 15 i 20 lat, pobierane każdorazowo w połowie długości przyrostów rocznych (wykres 1).

Łyko przewodzące u *Abies alba* zbudowane jest z następujących elementów: komórek sitowych, miększu łykowego, miększu promieni rdzeniowych, komórek białkowych i komórek kryształonośnych.

Budowa przyrostu rocznego łyka wtórnego wykazuje zróżnicowanie w obrębie pnia (wykres 1). Różnice dotyczą zarówno liczby komórek, jak również typów elementów, stanowiących przyrost roczny. Rozmiary komórek sitowych również wykazują zróżnicowanie w obrębie pnia.

W przyroście rocznym łyka wtórnego wyróżniłem łyko wczesne i łyko późne. W pędach do 5 lat w łyku późnym wyodrębniłem łyko strefy przejściowej. Podział ten przeprowadziłem na podstawie budowy komórek sitowych (grubość ścian, wielkość światła), terminów wytwarzania oraz okresu ich funkcjonowania.

W okresie wegetacyjnym podziały komórek inicjalnych kambium najwcześniej zachodzą w wierzchołkowej części drzewa, najpóźniej w strefie szyi korzeniowej; zakończenie tego procesu ma analogiczny przebieg.

W wyniku podziału komórek inicjalnych kambium wytwarzane są kolejno: elementy łyka wczesnego, elementy drewna wczesnego, elementy łyka późnego i elementy drewna późnego.

Długość okresu różnicowania oraz funkcjonowania jest niejednakowa w poszczególnych typach komórek sitowych łyka wtórnego.

Jako pierwsze w sezonie wegetacyjnym różnicują się komórki przezimowujące na granicy strefy kambialnej i łyka wtórnego. Reaktywacja procesów różnicowania w tych komórkach następuje w fazie wzrostu radialnego komórek strefy kambialnej. Okres funkcjonowania tych komórek waha się w granicach od 4 do 6 tygodni. Moje obserwacje wskazują na to, że ontogeneza tych komórek różni się od ontogenezy komórek sitowych łyka wczesnego, jak i łyka późnego.

Komórki sitowe łyka wczesnego wytwarzane są jako rezultat pierwszych podziałów komórek inicjalnych kambium. Okres ich różnicowania wynosi przeciętnie 2 tygodnie, zaś okres funkcjonowania nie przekracza 6 tygodni.

Proces różnicowania komórek sitowych strefy przejściowej trwa około 4 tygodni, natomiast okres ich funkcjonowania przekracza 2 miesiące.

Proces różnicowania komórek sitowych łyka późnego trwa około 6 tygodni. Komórki te przezimowują w stanie żywym i funkcjonują w następnym sezonie wegetacyjnym przeciętnie do połowy czerwca.

Przeprowadzone badania komórek białkowych sugerują inną determinację rozwojową komórek białkowych systemu osiowego w porównaniu z komórkami białkowymi systemu promieniowego.

Zmiany w łyku nieprzewodzącym polegają na: zgniataniu komórek sitowych, zwiększaniu się objętości komórek miększu łykowego, tworzeniu przestworów międzykomórkowych, tworzeniu komórek śluzowych i skleryfikacji niektórych komórek miększu łyka wtórnego.

W tkankach kory pierwotnej, w związku z przyrostem drzewa na grubość, zachodzą następujące wtórne procesy: zgniatanie i rozrywanie komórek miękiszowych, podziały komórek miękiszowych, rozrastanie się pierwotnych zbiorników żywicznych, skleryfikacja niektórych komórek miękiszu kory pierwotnej.

W okresie wegetacyjnym czynność fellogenu wykazuje wyraźne różnice sezonowe, zarówno pod względem intensywności podziałów, jak również typu wytwarzanych komórek korka.

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