

## COMPARATIVE STUDY OF THE SECONDARY PHLOEM OF SOME WOODY DICOTYLEDONS

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### SUMMARY

The anatomical features of the secondary phloem of 444 species of woody dicotyledons, belonging to 24 families, all from the Ivory Coast, West-Africa, were used to find correlations among the different types of secondary phloem characteristics as indicated by Zahur (1959). Special attention is given to companion cells since according to Zahur (1959), the type is fairly constant for natural categories. A reinvestigation of Zahur's data also revealed some correlations not noticed by him.

In the axial system of the secondary phloem of the investigated species both a primitive companion-cell type (type A) and in less extent a primitive sieve-tube type (type I) are associated and therefore correlated with an advanced axial-parenchyma type (type +) and mechanical-tissue type (type I). Also the reverse correlation is present.

A correlation between a primitive companion-cell type and a primitive sieve-area type (type a) as one could conclude from Zahur's data, is not present in our material. Also the correlation between an advanced companion-cell type (type C) and an advanced sieve-area type (type c), is difficult to ascertain or entirely absent. The above described correlation is less evident or even absent in those species in which a primitive sieve-tube type (type I) is present together with an advanced companion-cell type (type C) and in those species in which an advanced sieve-tube type (type III) is present together with a primitive companion-cell type (type A). In the secondary phloem of the concerned species from the Ivory Coast this is rather often the case.

If a storied structure is present sieve-tube type III occurs associated with companion-cell type C (and B). Such a combination of the two factors clearly indicates a primitive axial-parenchyma type (type -) and mechanical-tissue type (type 3), since both factors point in the same direction.

### 1. INTRODUCTION

ZAHUR (1959) made a comparative study of the secondary phloem of woody dicotyledons based on 423 species of 259 genera, representing 85 families from temperate and tropical regions, mainly the Hawaiian Islands. He distinguished three types of sieve tubes, based on their length and diameter and the position of the sieve plates (*table 1*). His study demonstrated that:

- a. there is a positive correlation between sieve-tube length on one side, and length of end walls or sieve plates, number of sieve areas on a sieve plate, size of pores, and frequency of secondary partitioning of sieve-tube mother cells on the other. Sieve-tube type I has long end walls which are correlated with a great number of sieve areas (*table 1*). Sieve-tube type I also has narrow pores and is derived without secondary partitioning of its mother cell;

- b. correlations between sieve-tube type and all other phloem characteristics are difficult to ascertain or entirely absent. Only a correlation with the mechanical-tissue type is present when species are considered in which secondary partitioning of the sieve-tube mother cells did not occur. In that case he found that sieve-tube type I and III are correlated with mechanical-tissue type 1 and 3, respectively;
- c. a storied structure is rarely found with sieve-tube type I, but is more commonly related with sieve-tube type III;
- d. the type of companion cell is fairly constant for natural categories of familial, and sometimes even of ordinal ranks.

It is regrettable that in Zahurs work there was not more extensive utilization of the comprehensive descriptions and detailed tables to correlate phloem characters. In our opinion some correlations were not noticed by him. So besides using the results of our own investigation, we also used Zahurs data to point out some correlations overlooked by him.

## 2. MATERIALS AND METHODS

Bark samples used are from the Versteegh and Den Outer collection, Ivory Coast, West Africa (1969). The collection is housed at the Department of Plant Cytology and Morphology, Agricultural University, Wageningen, the Netherlands. All material studied is accompanied by herbarium vouchers. Samples were collected from stems at breast height and immediately fixed in FAA. 444 Species of 247 genera of woody dicotyledons, belonging to 24 families were used to find correlations among the different types of secondary phloem characteristics. To get an impression of the anatomy of the secondary phloem of a family, only those families were taken of the Ivory Coast collection (1969), of which eight or more species were collected. These families are (between brackets number of genera and total number of species investigated): Anacardiaceae (7, 11), Annonaceae (15, 25), Apocynaceae (21, 29), Caesalpiniaceae (20, 26), Celastraceae (Hippocrateaceae included; 2, 11), Combretaceae (5, 16), Connaraceae (7, 9), Dichapetalaceae (1, 27), Ebenaceae (1, 12), Euphorbiaceae (30, 45), Flacourtiaceae (7, 8), Guttiferae or Clusiaceae (Hypericaceae included; 7, 8), Loganiaceae (3, 10), Meliaceae (7, 12), Mimosaceae (12, 17), Moraceae (5, 17), Ochnaceae (5, 8), Papilionaceae (17, 31), Rubiaceae (32, 57), Sapindaceae (10, 13), Sapotaceae (10, 11), Sterculiaceae (8, 18), Tiliaceae (7, 10), Verbenaceae (8, 13).

The investigated species are summarized in DEN OUTER (1972), those of the family Dichapetalaceae in VAN VEENENDAAL & DEN OUTER (1978)\*. As compared with ZAHUR (1959) fewer families but with each on the average more species were investigated. Of the families covered by us the Connaraceae, Dichapetalaceae and Ochnaceae were not looked at by Zahur.

Anatomical features were studied in transverse, radial and tangential sections and macerations. All sections were embedded in Kaisers gelatin-glycerin. Means

\* A list of the investigated species with their phloem characteristics is available upon request.

and ranges of the diameter and the length of sieve-tube members, parenchyma strands and ray height are based on at least twenty-five individual measurements. The sieve-tube type, sieve-area type, companion-cell type, axial-parenchyma type and mechanical-tissue type were classified according to ZAHUR (1959; see also *table 1*), ray type according to KRIBS (1935) though adopted for the rays of the secondary phloem by ZAHUR (1959), and bast type according to DEN OUTER & FUNDTER (1976).

If a positive relation (association) between two characteristics of the secondary phloem is present in 60 percent or more of the investigated cases, the two are called correlated. Correlations are thus not mathematically defined.

### 3. RESULTS

A comparison of the secondary phloem characters of the investigated species is given in four tables, viz.:

*Table 2*, relation of companion-cell type to phloem-ray type;

*Table 3*, secondary phloem characters of species with a storied structure;

*Table 4*, relation of companion-cell type to mechanical-tissue type;

*Table 5*, relation of companion-cell type to bast type.

From *table 2* it appears, among others, that when sieve-tube type I is present, the rays belong to the heterogeneous II type or in a lesser extent to the heterogeneous I type, but they are never homogeneous. The correlation here is much more distinct than that based on Zahurs data.

A storied structure does not occur together with sieve-tube type I (*table 3*). Of the 444 investigated species 45 possess a storied structure; they belong to the families Caesalpiniaceae, Malvaceae, Papilionaceae, Tiliaceae and Sterculiaceae (usually only the axial system is storied here). A storied structure is especially found when the bark possesses sieve-tube type III. In the bark with a storied structure an orderly sequence in four (bast type 4m) often occurs and this is nearly always associated with the presence of true phloem fibres (mechanical-tissue type 3). At the same time the sieve-area type is either b or c but never a; also the percentage of the companion cells lying against the radial sieve-tube walls is higher in comparison with that of species without a storied structure. Moreover the percentage of homogeneous rays is high.

All these characteristics are related to the fact that companion-cell type A does not occur. Especially companion-cell type C is correlated with the above mentioned characteristics. In contrast with type C, companion-cell type A corresponds more with mechanical-tissue type 1 (the lowest percentage true phloem fibres, the highest percentage stone cells or sclereids), than with type 3 (*table 4*). Companion-cell type A also corresponds more with bast type 2mi, s and g, than with 4m, consequently more with axial-parenchyma type + than with - (*table 5*).

Table 1. Secondary phloem characters determined by Zahur arranged in three groups.

	Primitive	Intermediate	Specialized
Fusiform cambium initial type	1 long (400 $\mu$ m or more)	m medium-long	s short (less than 350 $\mu$ m)
Sieve-tube type	I long (500 $\mu$ m or more); very oblique sieve plates with 10 or more sieve areas; sieve pores small; diameter sieve tube approx. equal to diameter of parenchyma cell	II intermediate between types I and III	III short (100–300 $\mu$ m); slightly oblique to transverse simple sieve plates; diameter sieve tube smaller than diameter parenchyma cell
Companion-cell type	A much shorter than the sieve-tube elements; occur usually single	B as long as the sieve-tube elements they accompany	C as long as the sieve-tube elements, but septated to form a strand of cells
Sieve-area type	a many, well-developed sieve areas in the side walls, equally spaced, approx. of the same size as the sieve areas on the sieve plates	b a small number, poorly developed sieve areas in the side walls, unequally spaced or diffuse	c sieve areas in the side walls entirely absent or obscure
Axial-parenchyma type	– not abundant; usually in tangential bands; some enlargement of the cells in the inactive phloem and some variation in cell size	$\pm$ intermediate between type – and type +	+ abundant, ground tissue; the cells enlarge considerably in the inactive phloem and cell size varies greatly
Mechanical-tissue type	3 true fibres, arranged in continuous or interrupted tangential bands	2 fibre-sclereids, originated in the non-functioning phloem; scattered or in short tangential bands	1 sclereids in groups or scattered or no mechanical tissue at all
Phloem rays	heterogeneous	intermediate	homogeneous
Storied structure	absent	almost absent	present
Secondary partitioning of sieve-tube mother cell	absent	intermediate	present

#### 4. DISCUSSION

Using Zahurs data we established the correlation between the type of companion cell and some other phloem characteristics. It shows that the companion-cell type A is more or less correlated with the axial-parenchyma type +, mechanical-tissue type 1 and sieve-area type a. Companion-cell type C on the other hand is more or less correlated with the axial-parenchyma type -, mechanical-tissue type 3 and sieve-area type c. Of the genera in the group with sieve-tube type I, 55% have companion-cell type C; the other 45% possess companion-cell type A and B. Also often associated with sieve-tube type I, the elements of which arose from long cambial initials and which seldom occur in storied structure, are axial-parenchyma type -, mechanical-tissue type 3 and sieve-area type c.

Of the genera in the group with sieve-tube type III, 45% have companion-cell type A. The bark which possesses sieve-tube type III, is often associated with axial-parenchyma type +, mechanical-tissue type 1, and sieve-area type a.

So, if we consider all Zahurs species together, both a primitive companion-cell type (type A) and also more or less an advanced sieve-tube type (type III) on one side, are associated with a primitive sieve-area type (type a) and advanced axial-parenchyma type (type +) and mechanical-tissue type (type 1) on the other. One can also postulate the reverse. The observed correlation for the sieve-tube type is in contrast with Zahurs own conclusion that sieve-tube type I and III are correlated with mechanical-tissue types 1 and 3, respectively. This (however) was only found when he considered species in which secondary partitioning of sieve-tube mother cells did not occur.

The Sympetalae (Gamopetalae), which are usually characterized by sieve-tube type III, have secondary partitioning of the sieve-tube mother cell much more commonly than the Choripetalae (Polypetalae), which are usually characterized by sieve-tube type I. Secondary partitioning of the sieve-tube mother cell into two or more sieve-tube members and sieve-tube type III may be considered more advanced than non-partitioning of the mother cell and sieve-tube type I (ZAHUR 1959). To consider mechanical-tissue type 3 more advanced than mechanical-tissue type 1 is a more difficult problem, because Sympetalae usually lack mechanical tissue. Furthermore within the bark phylogeny the development of two different tissues can take place in opposite directions. So an advanced sieve-tube does not necessarily implicate that all bark characteristics are advanced; see also ROTH (1981). Also according to CARLQUIST (1975) one can probably dismiss ZAHURS (1959) conclusion that sieve-tube elements are more primitive in less specialized dicotyledons as a *prima facie* conclusion. Furthermore CARLQUIST (1961) questioned Zahurs speculation that secondary partitioning indicates a phylogenetic advance because this feature occurs in families that are relatively primitive in many other features. According to ESAU (1979) the feature secondary partitioning, like nacreous wall thickening though potentially useful in systematics, cannot be mainingfully discussed with regard to their evolutionary significance.

From Zahurs data it is also evident, that:

a. when storied structure occurs, the sieve tubes usually belong to type III;  
b. neither sieve-tube nor companion-cell type are correlated with the phloem-ray type. The only conclusion could be that probably sieve-tube type III is slightly more often associated with heterogeneous phloem rays than is sieve-tube type I. This is just the opposite of what one would expect, since heterogeneous rays occur more often in species with primitive properties. But again an advanced axial system does not necessarily implicate an advanced horizontal (ray) system (Den Outer 1967).

A reinvestigation of Zahurs data, as shown above, might justify the following assertion. The length of the sieve tube determines the sieve-tube type. This type is more or less associated with certain other characteristics of the secondary phloem, and it also indicates whether or not we can expect a storied structure. The same applies for the companion-cell type. So from Zahurs data one could conclude that measurements of sieve-tube element lengths and determination of the companion-cell type would indicate what other characteristics can be expected in the secondary phloem of a concerned species.

Our results based on material from the Ivory Coast generally correspond with our findings using Zahurs material. However, the correlation between companion-cell type A and sieve-area type a, as one could conclude from Zahurs data, is not present in our material. Furthermore, the correlation between companion-cell type C and sieve-area type c, is difficult to ascertain or entirely absent. Also the not very pronounced relation between sieve-tube type I on one side, and mechanical-tissue type 1, bast type 2mi, s and g and therefore axial-parenchyma type + on the other, is in contrast with the correlation we could detect from Zahurs data, unless one takes into account those Hawaiian species in which secondary partitioning of the sieve-tube mother cells does not occur. So conclusions based on a reinvestigation of Zahurs data give the same correlations for the companion-cell type except those with the sieve-area type; for the sieve-tube type only if those species were considered in which secondary partitioning of sieve-tube mother cells did not occur. In the Ivory Coast material we did not investigate if secondary partitioning of the sieve-tube mother cells was present or not, since both according to CARLQUIST (1975) and ESAU (1979) this feature cannot be meaningfully discussed with regard to their evolutionary significance. In any case both material from the Ivory Coast and to a certain extent also that from the Hawaiian islands, show that primitive sieve-tube and companion-cell types (types I and A, respectively) are correlated with specialized other bark characteristics (mechanical-tissue type 1, and bast type 2mi, s and g which means axial-parenchyma type +) and vice versa.

However, companion-cell type C rather often occurs together with sieve-tube type I and companion-cell type A together with sieve-tube type III. So in many cases it is not immediately possible to make out or predict the other bark characteristics that one could expect using both the above mentioned characters, unless storied structure is present. Yet using both Zahurs and our own data one could say that measurements of the sieve-tube member length of a certain species to be investigated, implicates the sieve-tube type and probably nothing else; the

determination of the companion-cell type on the other hand often gives an indication about the remaining characteristics. The latter also implies that the remaining bark characteristics indicate what kind of companion-cell type one could expect. The type of companion cell is also fairly constant for natural categories as was already pointed out by Zahur.

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### LEGENDS TO TABLES 2-5

- Bast type 4u, 4mr, 4mi = orderly sequence in four, three or two uniseriate (u) or multiseriate (m) tangential layers, arranged in a regular (r) or irregular (i) way as follows:  
3u, 3mr, 3mi sequence in four: a tangential layer of fibres (crystalliferous cells), followed by a layer of axial-parenchyma cells, a layer of sieve tubes, a layer of axial-parenchyma cells, a layer of fibres (crystalliferous cells), etc.;  
2u, 2mr, 2mi sequence in three: one layer of axial-parenchyma cells is absent; sequence in two: a tangential layer of axial-parenchyma cells followed by a layer of sieve cells, a layer of axial-parenchyma cells, etc.
- s = sieve tubes scattered within an axial system composed of parenchyma cells  
g = sieve tubes in groups within an axial system composed of parenchyma cells
- place companion cell || = companion cells only against the radial walls of the sieve tube  
◇ = companion cells appear as though cut out of the corner of the sieve tube
- phloem-ray type He = heterogeneous phloem rays; procumbent and upright cells are present  
Ho = homogeneous phloem rays; only procumbent or only upright cells are present
- He/He I = uniseriate rays and multiseriate rays with long uniseriate tails  
He/He II = uniseriate rays and multiseriate rays with short uniseriate tails  
He/He III = only uniseriate rays are present

Table 2. Relation of companion-cell type to phloem-ray type (between brackets, percentage).

*Sieve-tube type I:*

comp.-cell type	no. of species studied	phloem-ray type					
		Hol	HolII	HolIII	Hel	HelI	HelII
A	2				2 (100)		
B	6				5 (83)	1 (17)	
C	47				9 (19)	36 (77)	2 (4)
Total	55				16 (29)	37 (67)	2 (4)

*Sieve-tube type II:*

A	59	4 (7)	6 (10)	1 (2)	26 (44)	20 (34)	2 (3)
B	36	1 (3)	2 (5)		15 (42)	15 (42)	3 (8)
C	81	7 (9)	28 (34)	1 (1)	20 (25)	20 (25)	5 (6)
Total	176	12 (7)	36 (20)	2 (1)	61 (35)	55 (31)	10 (6)

*Sieve-tube type III:*

A	78	4 (5)	13 (17)	2 (3)	23 (29)	29 (37)	7 (9)
B	75	2 (3)	10 (13)		19 (25)	41 (55)	3 (4)
C	60	6 (10)	16 (27)	3 (5)	17 (28)	12 (20)	6 (10)
Total	213	12 (6)	39 (18)	5 (2)	59 (28)	82 (38)	16 (8)

*Sieve-tube type I, II and III:*

A	139	8 (6)	19 (14)	3 (2)	51 (37)	49 (35)	9 (6)
B	117	3 (3)	12 (10)		39 (33)	57 (49)	6 (5)
C	188	13 (7)	44 (23)	4 (3)	46 (24)	68 (36)	13 (7)
Total	444	24 (5)	75 (17)	7 (2)	136 (31)	174 (39)	28 (6)

Table 3. Secondary phloem characters of species with a storied structure (45 out of 444 investigated species; between brackets, percentage).

Sieve-tube type I	—	Mechanical-tissue type fibre	44 (98)
Sieve-tube type II	4	Mechanical-tissue type fibre-sclereid	—
Sieve-tube type III	41	Mechanical-tissue type stone cell	27 (60)
	—	Mechanical-tissue type no mech. tissue	—
	45	Mechanical-tissue type crystal cell	45 (100)
Sieve-area type a	—	Bast type	4 mr 5 (11)
Sieve-area type b	39	Bast type	4 mi 20 (44)
Sieve-area type c	6	Bast type	3 mr 2 (4)
	—	Bast type	2 mr 3 (7)
	45	Bast type	2 mi 4 (9)
		Bast type	s 11 (25)



Table 3, continued

Companion-cell type A	—	Phloem-ray type	Ho I	3 (7)
Companion-cell type B	20	Phloem-ray type	Ho II	17 (38)
Companion-cell type C	25	Phloem-ray type	Ho III	1 (2)
	—	Phloem-ray type	He I	1 (2)
	45	Phloem-ray type	He II	23 (51)
		Phloem-ray type	He III	—
				45
Place companion cell	15			
Place companion cell ◇	30			
	45			

Table 4. Relation of companion-cell type to mechanical-tissue type (between brackets, percentage).

*Sieve-tube type I:*

comp.-cell type	no. of species studied	fibre	fibre-sclereid	stone cell	no. mech. tissue	crystal cell
A	2	1 (50)		1 (50)		1 (50)
B	6	5 (83)	1 (17)	6 (100)		6 (100)
C	47	13 (28)	3 (6)	44 (94)		46 (98)
Total	55	19 (35)	4 (7)	51 (93)		53 (96)

*Sieve-tube type II:*

A	59	11 (19)	10 (17)	56 (95)	1 (2)	54 (92)
B	36	21 (58)	4 (11)	28 (78)		35 (97)
C	81	56 (68)	11 (14)	61 (75)		79 (98)
Total	176	87 (49)	25 (14)	145 (82)	1 (—)	168 (95)

*Sieve-tube type III:*

A	78	31 (40)	9 (12)	62 (79)	1 (1)	62 (79)
B	75	46 (61)	12 (16)	45 (60)	1 (1)	66 (88)
C	60	46 (77)	5 (8)	40 (67)	2 (3)	59 (98)
Total	213	123 (58)	26 (12)	147 (69)	4 (2)	187 (88)

*Sieve-tube I, II and III:*

A	139	43 (31)	19 (14)	119 (86)	2 (1)	117 (84)
B	117	72 (62)	17 (15)	79 (68)	1 (1)	107 (91)
C	188	114 (61)	19 (10)	145 (77)	2 (1)	184 (98)
Total	444	229 (52)	55 (12)	343 (77)	5 (1)	408 (92)

Table 5. Relation of companion-cell type to bast type (between brackets, percentage).

<i>Sieve-tube type I:</i>										
comp.-cell type	no. of species studied	bast type								
		4u, 4mr	4mi	3u, 3mr	3mi	2u	2mr	2mi	s	g
A	2								1 (50)	1 (50)
B	6					1 (17)			2 (33)	3 (50)
C	47		3 (6)		1 (2)	8 (17)		3 (6)	26 (56)	6 (13)
Total	55		3 (5)		1 (2)	9 (16)		3 (6)	29 (53)	10 (18)
<i>Sieve-tube type II:</i>										
A	59	6 (10)	1 (12)			5 (8)	1 (2)	8 (13)	34 (58)	4 (7)
B	36	2 (6)	7 (19)			4 (11)		2 (6)	20 (55)	1 (3)
C	81	1 (1)	15 (19)	2 (2)	4 (5)	3 (4)	4 (5)	4 (5)	45 (55)	3 (4)
Total	176	9 (5)	23 (13)	2 (1)	4 (2)	12 (7)	5 (3)	14 (8)	99 (56)	8 (5)
<i>Sieve-tube type III:</i>										
A	78	8 (10)	8 (10)			8 (10)	1 (1)	3 (4)	38 (49)	12 (16)
B	75	5 (6)	26 (35)			5 (7)	2 (3)	4 (5)	32 (43)	1 (1)
C	60	4 (7)	15 (25)	2 (3)	3 (5)	1 (1)	4 (7)	6 (10)	21 (35)	4 (7)
Total	213	17 (8)	49 (23)	2 (1)	3 (1)	14 (7)	7 (3)	13 (6)	91 (43)	17 (8)
<i>Sieve-tube type I, II and III:</i>										
A	139	14 (10)	9 (7)			13 (9)	2 (1)	11 (8)	73 (53)	17 (12)
B	117	7 (6)	33 (28)			10 (9)	2 (2)	6 (5)	54 (46)	5 (4)
C	188	5 (3)	33 (18)	4 (2)	8 (4)	12 (6)	8 (4)	13 (7)	92 (49)	13 (7)
Total	444	26 (5)	75 (17)	4 (1)	8 (2)	35 (8)	12 (3)	30 (7)	219 (49)	35 (8)