

## CHROMOSOME NUMBER AND RELATIONSHIP IN THE MAGNOLIALES

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*With plate 80 and four text figures*

BECAUSE OF THEIR important phylogenetic position, the Magnoliales offer a suggestive group for cytological investigation. It is the purpose of the present paper to report the results of a cytological survey of several members of the Magnoliales with a discussion of the bearing of these results on taxonomic and phylogenetic relationships.<sup>1</sup>

The Magnoliales, according to Hutchinson (1921), may be separated into the following families: Magnoliaceae, Winteraceae, Schisandraceae, Himantandraceae, Lactoridaceae, Trochodendraceae, and Cercidiphyllaceae. In the collections of the Arnold Arboretum members of the Magnoliaceae, Schisandraceae, Trochodendraceae, and Cercidiphyllaceae are available for study. Through the coöperation of the Royal Botanic Garden, Kew, and the John Innes Horticultural Institution, Merton, England, it has been possible to extend the range of the investigation to include all the families in the order, with the exception of representatives of the Himantandraceae and Lactoridaceae.

Table I gives the chromosome number of the species studied. The observations on the meiotic divisions were made from aceto-carminic and permanent smear preparations of pollen mother cells. The aceto-carminic preparations were found to be superior for the study of chromosome number and chiasma frequency. The counts of *Illicium anisatum*, *I. floridanum* and *Drimys Winteri* were secured from root tip material by the use of the smear-maceration method followed by staining with aceto-carminic. The remaining somatic counts were secured from root tips by use of the usual permanent preparations of sectioned material.

<sup>1</sup>The work reported in the present paper has been furthered through the coöperation of a number of investigators at this and other institutions. Thanks are due to the officials of the Royal Botanic Garden, Kew, and of the John Innes Horticultural Institution, especially to W. J. C. Lawrence, Curator, and Mr. L. La Cour of the latter institution, who provided the material of *Trochodendron aralioides*, *Illicium religiosum*, *Tetracentron sinense*, and *Kadsura japonica*. Professor I. W. Bailey of the Arnold Arboretum has kindly furnished the data regarding the anatomy of the group studied. Professor Karl Sax of the Arnold Arboretum suggested the investigation and has very generously provided the drawings and information regarding *Cercidiphyllum* and *Euptelea*.

TABLE I  
CHROMOSOME NUMBERS IN THE MAGNOLIALES

	Chromosome No.	
	(n)	(2n)
MAGNOLIACEAE		
<i>Magnolia virginiana</i> L. ....	19	
<i>Magnolia tripetala</i> L. ....	19	
<i>Magnolia Fraseri</i> Walt. ....	19	
<i>Magnolia acuminata</i> L. ....	38	
<i>Magnolia acuminata</i> var. <i>cordata</i> Sarg. ....	38	
<i>Magnolia liliflora</i> Desrouss ....	38	
<i>Magnolia Soulangiana</i> Soul. ....	38	
<i>Magnolia Soulangiana</i> var. <i>Brozzonii</i> ....	38	
<i>Magnolia Soulangiana</i> var. <i>Candolleana</i> ....	38	
<i>Liriodendron Tulipifera</i> L. ....	19	
WINTERACEAE		
<i>Illicium anisatum</i> L. ....		28
<i>Illicium religiosum</i> Sieb. & Zucc. ....		28
<i>Illicium floridanum</i> Ellis ....		28
<i>Drimys Winteri</i> Forst. ....		±76
		(4 × 19)
SCHISANDRACEAE		
<i>Schisandra sphenanthera</i> Rehd. & Wils. ....	14	
<i>Kadsura japonica</i> L. ....		28
TROCHODENDRACEAE		
<i>Trochodendron aralioides</i> Sieb. & Zucc. ....		38
<i>Euptelea polyandra</i> Sieb. & Zucc. ....	14	
CERCIDIPHYLLACEAE		
<i>Cercidiphyllum japonicum</i> Sieb. & Zucc. ....	19	
<i>Tetracentron sinense</i> Oliv. <sup>1</sup> ....		38

In the genus *Magnolia*, the forms examined were either diploids or tetraploids (Pl. 80, figs. 1, 2, 5, 7). Morinaga, et al. (1929) have reported the haploid number of *M. grandiflora* as being 56—57. The basic number in *Magnolia* is 19. It therefore appears likely that *M. grandiflora* is a hexaploid.

Both diploid and tetraploid species were found among the Magnolias which are indigenous to North America. *Magnolia liliflora*, the only Chinese species which has been studied, has 38 chromosomes. Ishikawa (1916) has given the number of one of the Chinese species of

<sup>1</sup>*Tetracentron* has been omitted by Hutchinson (1926) in his treatment of the Magnoliales. Since it has generally been considered as belonging to this group, it has been included in this study.



*Magnolia* (*M. kobus*) as 19. This indicates that there are diploid and tetraploid representatives of the genus in both North America and Asia.

*Magnolia Soulangeana* is a supposed hybrid resulting from the cross *M. denudata*  $\times$  *M. liliiflora* (Rehder, 1927). The three forms of this hybrid that were examined all proved to be tetraploids (see Table I & Fig. 8). Since *M. liliiflora* was found to be a tetraploid (Fig. 11), it is probable that the other parental species, *M. denudata*, is also a tetraploid. However, the chromosome count of this species was not secured. The meiotic divisions in the forms of *M. Soulangeana* that were examined show a slight amount of irregularity. There is also considerable pollen sterility, averaging about 25% in each of the three forms studied. In the parent species, *M. denudata* and *M. liliiflora*, only 7% of the pollen is sterile.

*Liriodendron Tulipifera* has 19 pairs of chromosomes (Fig. 4). It is taxonomically closely allied to the Magnolias. This position is well supported by the cytological evidence. The chromosome size, shape, and chiasma frequency are similar to those of the Magnolias.

The chromosome constitution of the different species of *Magnolia* do not exhibit any detectable variation. The chromosomes are all short rods of equal length, showing up very clearly at metaphase. The nature of the material has made possible the determination of chiasma frequency at early metaphase. This has been done for *Liriodendron Tulipifera* and a few representative species of *Magnolia*.

The results of this study brought out the following points:

1. There was no significant difference in the chiasma frequency per bivalent between any of the species of *Magnolia* examined. Two diploid species (*M. virginiana* and *M. tripetala*), two tetraploid species (*M. acuminata* var. *cordata* and *M. liliiflora*), and the species hybrid, (*M. Soulangeana* var. *Brozzonii*) were investigated. The chiasma frequency in the above species ranged from 1.06 in *M. tripetala* to 1.17 in *M. liliiflora*. A difference of this degree can scarcely be considered significant. The chiasma frequency of *Liriodendron Tulipifera* was 1.14, thus coming within the same range as that of the *Magnolia* species.

2. By far the greater proportion of the bivalents are paired as rods. There are usually 1—3 rings and, in the tetraploids, one or two multivalent associations. The chiasmata are invariably terminal at this stage (early metaphase).

It was thought that a study of the pollen grains of the diploid and tetraploid Magnolias might show size differences which could be used as a diagnostic character in predicting whether a particular species was

a diploid or a tetraploid. It was found, however, that the pollen grains of both diploid and tetraploid species are all very similar in size, shape, and wall sculpturing. The pollen grains of *Liriodendron Tulipifera* bear certain resemblances to those of the Magnolias, although the pollen of this species appears to have a much thicker wall, with deeper, heavier sculpturing.

Three species of *Illicium* have been examined, all of which proved to be diploids with 28 somatic chromosomes (Text fig. 1). The chromosomes of the three species that have been examined are remarkably

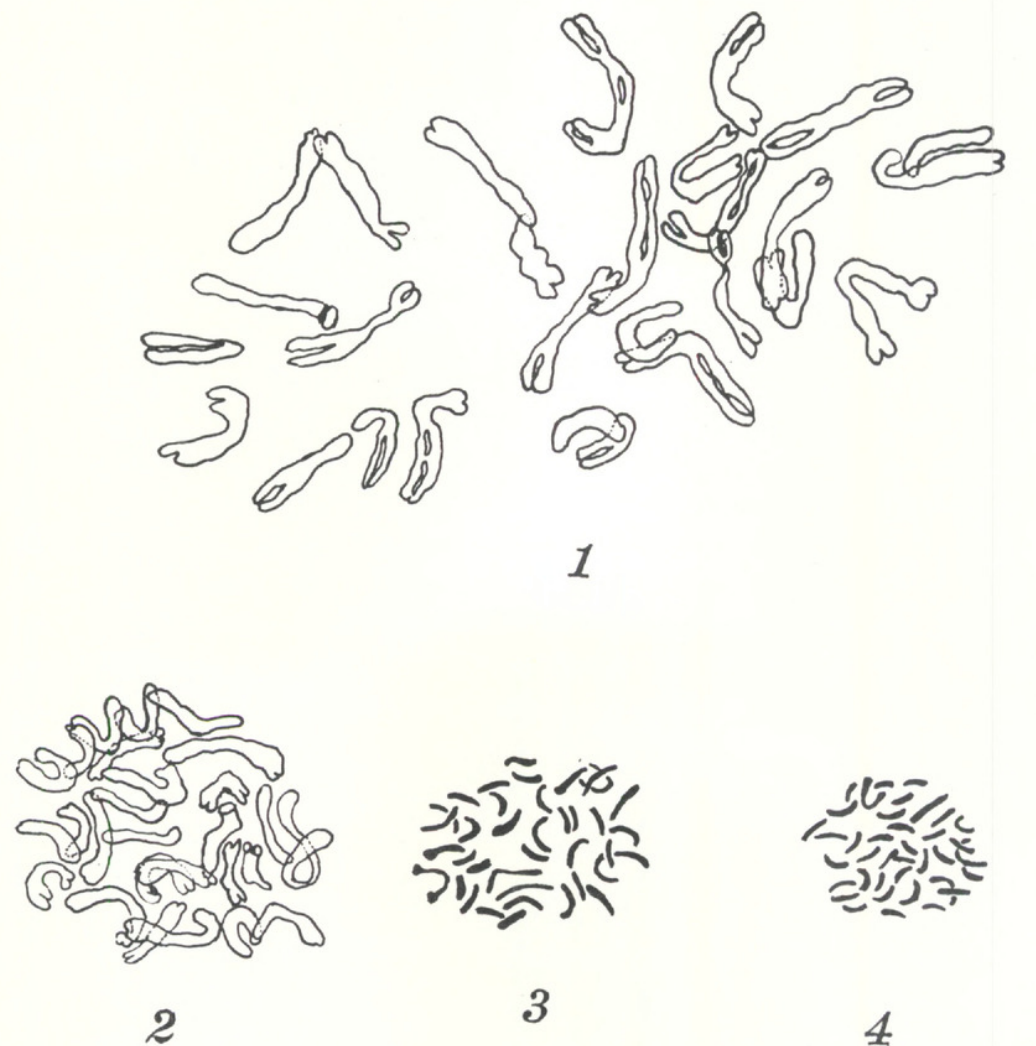


FIGURE 1. *ILlicium FLORIDANUM* .....28 chromosomes.  
FIGURE 2. *KADSURA JAPONICA* .....28 chromosomes.  
FIGURE 3. *TROCHODENDRON ARALIoidES* .....38 chromosomes.  
FIGURE 4. *TETRACENTRON SINENSE* .....38 chromosomes.

FIGURE 1. has been drawn from a root tip smear. The remainder are from permanent preparations of root tip material. × 2100.



alike and are considerably larger than those of *Drimys* and other members of the group having 19 as the basic number. Cytologically, *Illicium* is clearly distinct and bears no resemblance to either the members of the Magnoliaceae or *Drimys*, supposedly its closest allies.

Morinaga, et al. (1929) have listed the reduced chromosome number of *Illicium anisatum* as 14. The figures given by these workers, of the chromosomes during metaphase of the first division compare favorably in size and shape with those of *Schisandra sphenanthera* (Pl. 80, fig. 10).

*Drimys Winteri* has about 76 chromosomes in somatic cells. Strasburger (1905) has stated there are about 36 pairs of chromosomes in this species. Because of the large number of chromosomes and their small size, it is difficult to make absolutely certain of the number. However, it is undoubtedly between 72 and 76, with greater likelihood of the latter figure's being correct.

In *Schisandra sphenanthera* the haploid chromosome number is 14 (Pl. 80, fig. 10). The chromosome complement of this species has no affinity with that of the Magnolias or *Liriodendron*. The chromosomes are larger than those found in the Magnoliaceae, and there is a tendency toward irregularity in the time of division,—i. e., precocious division of one or two bivalents. This does not seem to affect the percentage of good pollen (94%).

*Kadsura* is a genus of climbing vines, closely allied to *Schisandra*. *Kadsura japonica*, the only species which has been examined, has 28 chromosomes (Text fig. 2). The counts were made from permanent root tip preparations. The chromosomes of this species resemble those of *Schisandra* and other representatives of the 14 chromosome group.

*Trochodendron aralioides* has a somatic chromosome number of 38 (Text fig. 3). Cytologically, its relations are with *Magnolia* and *Liriodendron* rather than with *Euptelea*, with which it has been placed by Hutchinson (1921).

*Euptelea polyandra* has 14 pairs of chromosomes (Pl. 80, fig. 9). Some secondary pairing and irregularity in the time of division, similar to that found in *Schisandra sphenanthera*, has been noted in this species.

*Cercidiphyllum japonicum*, the representative of the monotypic family Cercidiphyllaceae has 19 chromosomes (Pl. 80, fig. 3). The figure indicates that the chromosome size and shape are of the same general nature as those of *Liriodendron* and *Magnolia*. The chiasma frequency corresponds closely to that found in these two genera.



*Tetracentron sinense* has 38 chromosomes in the root tip cells (Text fig. 4). Judging by its cytological characteristics, this genus is allied to the Magnoliales. Its chromosome number definitely places it with *Magnolia*, *Liriodendron*, *Trochodendron*, etc.

## DISCUSSION

Engler and Prantl's system (Die natürlichen Pflanzenfamilien) has been criticized on the ground that the Amentiferae are regarded as a more primitive group than such polypetalous families as Magnoliaceae and Ranunculaceae. From morphological evidence, this criticism seems justified. Maneval (1914) has compiled a list of characters which are generally conceded to be more or less indicative of a primitive condition. These characters are all found in the Magnoliaceae. Sinnott (1914) has used nodal anatomy as a means of indicating phylogenetic relationships. He states: "The Magnoliaceae are perhaps more variable in nodal structure than any other family of dicotyledons." The Magnoliaceae, as used here, include, in addition to the Magnolias and *Liriodendron*, the Schisandraceae and Winteraceae of Hutchinson (1926). This variability in nodal structure is precisely what one would expect and what is actually found among primitive types.

If one accepts the customary thesis that the Magnoliales are a primitive group of dicotyledons, one would expect to find, among the families of this order, several lines of cytological development. Thus far the evidence from chromosome numbers and other cytological features indicates that there are at least two such lines. The first comprises *Magnolia*, *Liriodendron*, *Drimys*, *Trochodendron*, *Tetracentron*, and *Cercidiphyllum*. In this group the chromosomes are characteristically small, short rods with a basic number of 19 pairs. In the second group occur *Euptelea*, *Schisandra*, *Kadsura* and *Illicium*, with 14 as the basic chromosome number. The chromosome size, shape, and configurations found in this group bear considerable resemblance to one another.

In Table II an attempt has been made to compare chromosome number, type of nodal anatomy, and several other anatomical features in the genera under investigation. Perhaps the most important point brought out by this table is the correlation existing between chromosome number and type of nodal anatomy. In Group I are found genera with two types of nodal anatomy: (a) 3 traces from 3 gaps (*Tetracentron*, *Drimys*, *Cercidiphyllum*); (b)  $\infty$  traces from  $\infty$  gaps (*Trochodendron*, *Magnolia*, *Liriodendron*). The second cate-

TABLE II  
A COMPARISON OF CHROMOSOME NUMBER, NODAL ANATOMY, ETC.  
IN GENERA OF THE MAGNOLIALES<sup>1</sup>

		genus	leaf venation	vessels present or absent	vessels scalariform or porous	tracheids
GROUP I basic chromosome number—19	Type of nodal anatomy <i>3 traces from 3 gaps</i>	<i>Tetracentron</i>	palmate	absent		scalariform
		<i>Drimys</i>	pinnate	absent		rarely scalariform
		<i>Cercidiphyllum</i>	palmate	present	scalariform	
	Type of nodal anatomy <i>∞ traces from ∞ gaps</i>	<i>Trochodendron</i>	pinnate	absent		scalariform
		<i>Magnolia</i>	pinnate	present	scalariform or porous	
		<i>Liriodendron</i>	pinnate	present	scalariform or porous	
GROUP II basic chromosome number—14	Type of nodal anatomy <i>1 trace from 1 gap</i>	<i>Illicium</i>	pinnate	present	scalariform	
		<i>Schisandra</i>	pinnate	present	scalariform or porous	
		<i>Kadsura</i>	pinnate	present	scalariform or porous	
	Type of nodal anatomy <i>3-7 traces from 1 gap</i>	<i>Euptelea</i>	pinnate	present	scalariform	

<sup>1</sup>Morphological data supplied by I. W. Bailey.



gory is essentially a multiplication of the fundamental condition of 3 traces from 3 gaps. In the 14-chromosome group two types of nodal anatomy are found; (a) 1 trace from one gap (*Illicium*, *Kadsura*, *Schisandra*); (b) 3—7 traces from 1 gap (*Euptelea*). The latter condition simply represents a multiplication in the number of traces of the basic condition of 1 trace from 1 gap.

The value of nodal anatomy as an aid in classification of angiosperms and as a means of detecting phylogenetic relationships, has been fully discussed by Sinnott (1914). It is sufficient, for the present purpose, to note that it is extremely constant within most families and is of considerable value in determining broad lines of relationship. The fact that there is a close agreement between chromosome number and nodal anatomy is reasonably strong evidence that the genera placed together in Table II form a natural grouping which should be considered in any future classification of this group.

It may be well to point out, at this juncture, the significance attached to a basic number of 19 chromosomes. This is an unusual number in the plant kingdom and is very rarely met. It has not been reported in any of the families which may be considered as related to the Magnoliales. If the chromosome number had been 12 or some other number of common occurrence, it would not carry nearly the weight which it does in the present case. It is difficult to account for the occurrence of this unusual chromosome number in Group I unless we assume that the genera composing this group are made up of closely related forms.

McLaughlin (1933) has recently made a very thorough study of the systematic anatomy of the woods of the Magnoliales. In his suggested classification of the group he has eliminated *Cercidiphyllum*, *Euptelea* and *Illicium* by placing them in the Hamamelidales rather than in the Hamamelidaceae. Cytologically, there is no evidence to justify this step. Professor Sax informs me that the Hamamelidaceae have 12 rather characteristically small, short chromosomes (unpublished data). *Illicium* and *Euptelea*, with 14 chromosomes, and *Cercidiphyllum*, with 19 chromosomes, would not fit into this proposed classification.

The placing of *Drimys* and *Illicium* in the same family, as proposed by Hutchinson (1921), can hardly be sanctioned in view of the present evidence. These two genera are very dissimilar cytologically. The same criticism can be made in regard to placing *Trochodendron* and *Euptelea* in the same family, as this author has suggested (Hutchinson, 1921).



## SUMMARY

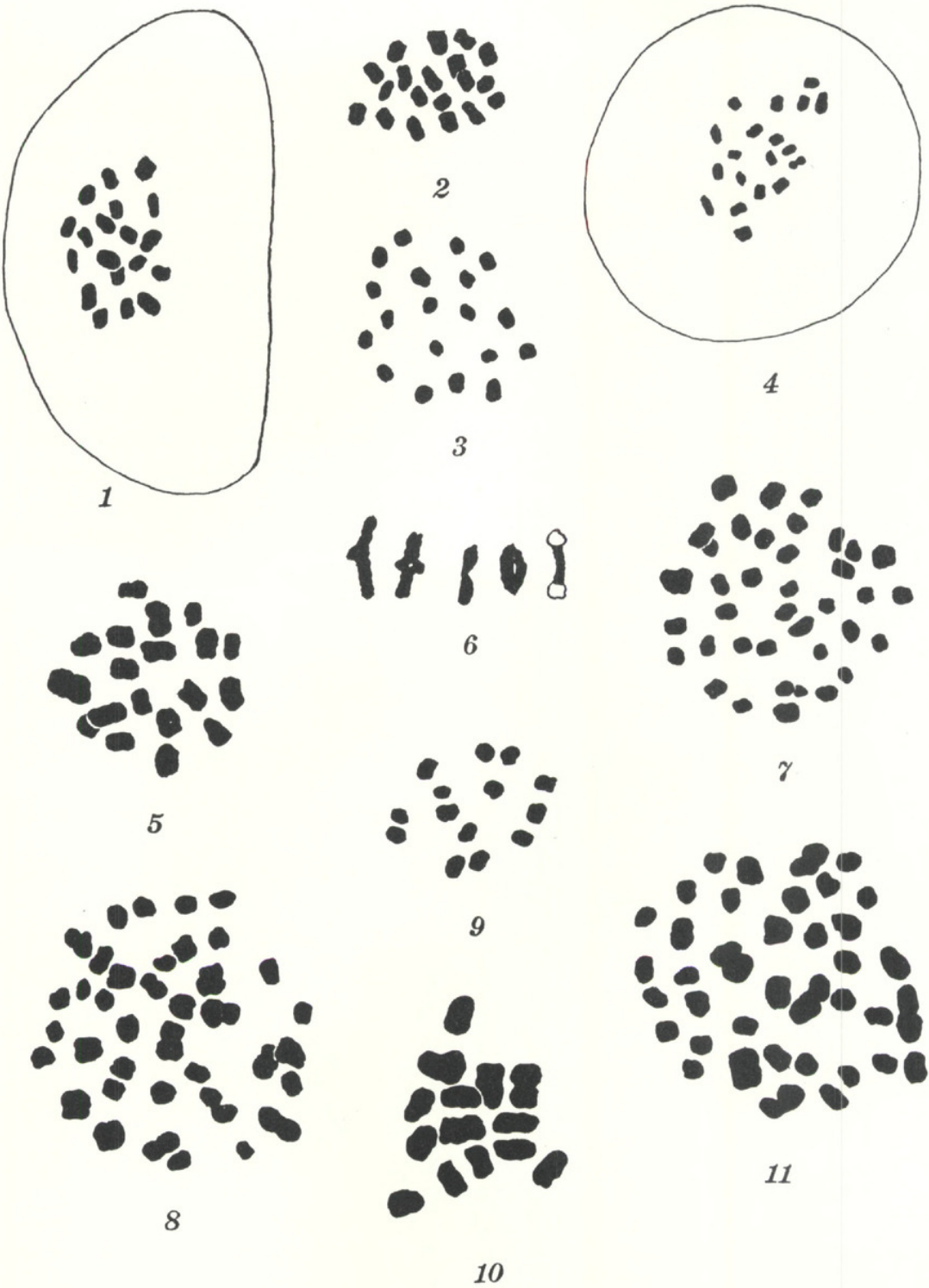
The evidence shows that in the Magnoliales thus far investigated there are two lines of cytological development. Group I has a basic chromosome number of 19. The chromosomes of this group are characteristically small, short rods. In this group the following genera occur: *Magnolia*, *Liriodendron*, *Cercidiphyllum*, *Drimys*, *Trochodendron*, and *Tetracentron*. Group II has a basic number of 14 chromosomes. In this group the chromosomes are much larger than those of Group I. The following genera occur in Group II: *Illicium*, *Schisandra*, *Kadsura*, and *Euptelea*.

There seems to be a correlation between basic chromosome number and type of nodal anatomy. Group I is characterized by the trilacunar condition and a multiplication of this condition. Group II is characterized by the unilacunar condition and a modification of this condition.

Some of the more recent systems of classification of the Magnoliales have been criticised, and it has been pointed out where certain revisions would bring about a more natural system of classification.

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CHROMOSOME NUMBER AND RELATIONSHIP IN THE MAGNOLIALES





DESCRIPTION OF PLATE 80

Chromosome numbers in the Magnoliales. The drawings are from aceto-carmin and permanent smears of pollen mother cells. Magnification about 2100.

Fig. 1.	<i>Magnolia tripetala</i> .....	II M.
Fig. 2.	<i>Magnolia virginiana</i> .....	I M.
Fig. 3.	<i>Cercidiphyllum japonicum</i> .....	I M.
Fig. 4.	<i>Liriodendron Tulipifera</i> .....	I M.
Fig. 5.	<i>Magnolia Fraseri</i> .....	I M.
Fig. 6.	Types of configurations found in <i>Liriodendron Tulipifera</i> .	
Fig. 7.	<i>Magnolia acuminata</i> .....	I M.
Fig. 8.	<i>Magnolia Soulangeana</i> var. <i>Brozzonii</i> .....	I M.
Fig. 9.	<i>Euptelea polyandra</i> .....	I M.
Fig. 10.	<i>Schisandra sphenanthera</i> .....	I M.
Fig. 11.	<i>Magnolia liliflora</i> .....	I M.

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