

ISO-CHROMOSOMES AND THE ORIGIN OF TRIPLOIDY IN HYBRIDS BETWEEN OLD AND NEW WORLD SPECIES OF *PHILADELPHUS*

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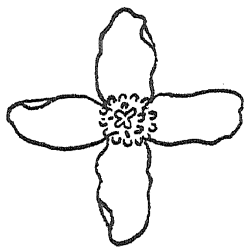
THE genus *Philadelphus* comprising about 40 species of deciduous shrubs is found in isolated regions in North America, East Europe, Caucasus and East Asia to Himalayas. The fossil history of *Philadelphus* goes as far back as Eocene times when the genus had a continuous distribution in the Northern Hemisphere. With the rise of the Rocky Mountains, the distribution of *Philadelphus* in the New World became discontinuous and the result has been the evolution of distinct eastern and western geographical races in North America. The West American species, which are more closely related to the fossil types, have inflorescences which are racemose panicles. The Ice Age in America saw the migration of *Philadelphus* southwards as far as Mexico, and in these regions have evolved, desert forms like the *Microphylli*, which have small leaves and scented flowers which are usually solitary. At least four distinct geographical races have evolved in America as a result of isolation (Rehder, 1940).

In Asia a north-south differentiation of *Philadelphus* occurred as a result of the Himalayan uplift. To the North Asian group, the *Coronarii* series of Rehder, also belong the single European species *P. coronarius* and the related *P. caucasicus* which survived the last Ice Age by finding shelter in the mountains of South Europe and Caucasus respectively. The Indian species of *Philadelphus*, *P. tomentosus* and *P. nepalensis* of the Himalayas, are closely related to these, and link the *Coronarii* of Europe with their Siberian, Manchurian and Japanese counterparts. The second group of Asian *Philadelphus*, the *Sericanthe*, are found in Central and West China. They resemble *Deutzia*, the genus most closely related to *Philadelphus* and have the same basic number of 13 chromosomes. While high polyploidy occurs in *Deutzia* (Sax, 1931), *Philadelphus* has remained diploid ($2n = 26$) nature since Eocene time (Bangham, 1929).

In an earlier paper (E. K. Janaki Ammal, 1951) I have traced the history of *Philadelphus* in cultivation and reported on the occurrence of triploids

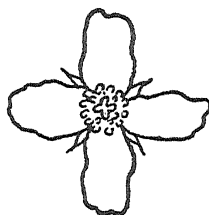
P. coronarius

$2n = 26$



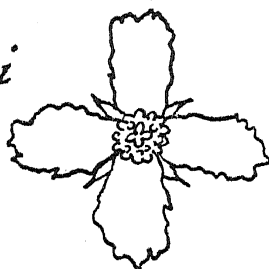
P. microphyllus

$2n = 26$



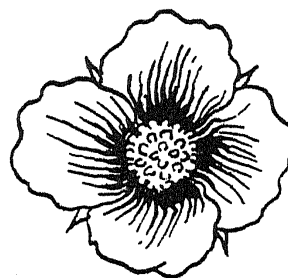
P. Lemoinei

$2n = 26$



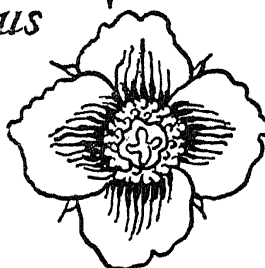
P. Coulteri

$2n = 26$



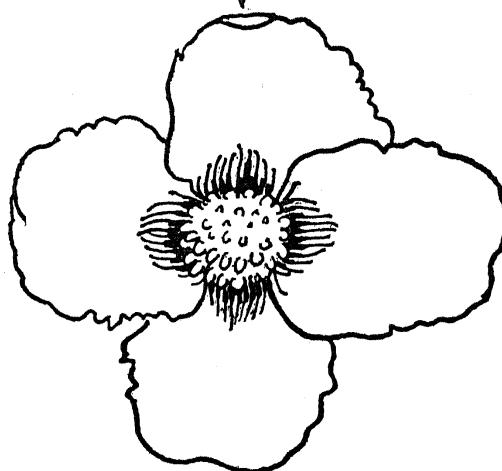
P. purpureo-maculatus

$2n = 26$



P. "Belle Etoile"

$2n = 39$



Diagrammatic representation of the parentage of the triploid "Belle Etoile".

amongst the progeny of garden hybrids between species widely separated in nature.

These triploids ($2n = 39$) which had much larger flowers than diploids were first found in the progeny of the tri-specific hybrid *P. purpureo-maculatus*. The grandparents of this hybrid were the European species *P. coronarius* and the fragrant Arizonian species *P. microphyllus*. The resulting hybrid *P. lemoinei* was crossed with the Mexican species *P. coulteri*. This gave the sweet scented and purple tinted *P. purpureo-maculatus*. Both fragrance and size have been enhanced as a result of triploidy in the progeny of this garden favourite. The best known of them is "Belle Etoile" (see diagrammatic sketch).

The production of unreduced gametes which is the main cause of aberrant triploid seedlings in cultivated plants has been shown to be due to errors in meiosis. Triploids are not only of immediate value to horticulture and agriculture as being more decorative or vegetatively productive plants, but they also become the potentials of hexaploids by chromosome doubling. Hexaploids are increasingly found in regions where the distribution of diploid species overlap. This is a common occurrence in the case of *Magnolia*, *Camellia* and *Rhododendron* (E. K. J., 1950, 1952 *a* and 1952 *b*). The chromosome mechanism that is responsible for the production of triploids in garden hybrids between species widely separated in nature as in *Philadelphus* is of interest for understanding what happens in nature when diploid populations overlap as a result of migration or due to changes in the physiography of land masses.

THE CHROMOSOME MORPHOLOGY IN *Philadelphus*

For the study of chromosomes, flower-buds were fixed in acetic alcohol 1:3 and stained with acetocarmine. The chromosome complement of *Philadelphus* was fairly uniform in all the 29 species I examined. It was found to be composed of 13 pairs of chromosomes. The chromosome morphology is best studied during mitosis in pollen grains (Fig. 1). Of the 13 chromosomes, composing the haploid complement, 4 are small, 8 medium and 1 long. The single long chromosome and 2 of the small chromosomes have medium constrictions while in the remaining 10 it is sub-terminal. The haploid formulæ may be represented as:

$$1 L_m + 8 M_{st} + 2 S_{st} + 2 S_m.$$

The large L chromosome stands out from the rest by being more heterochromatic than the others.

CHROMOSOME BEHAVIOUR IN *Philadelphus*

Thirteen bivalents are formed regularly during meiosis in all the species studied (Figs. 2 and 3). Of these five have 2 chiasmata and eight have 1 chiasma only, the chiasmata frequency being 1.38.

In spite of regular pairing pollen sterility was found to be fairly high in some of the species studied (see Table I).

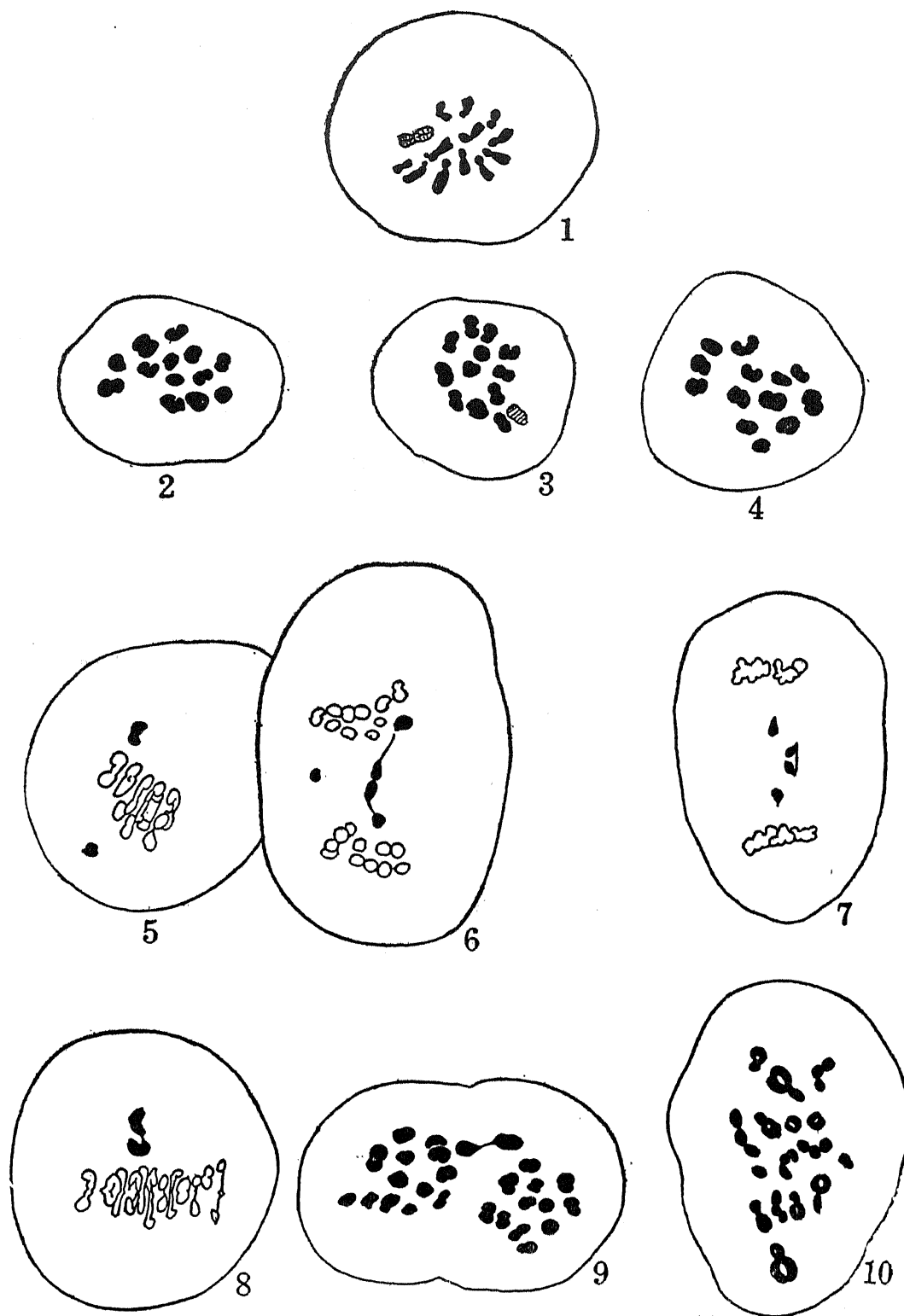
An examination of pollen mother cells in the parent species showed that the behaviour of the single large pair of chromosomes differed from the rest of the complement at meiosis. It did not congress on the plate simultaneously and behaved as if it had a defective chromosome. This behaviour was accentuated in the hybrids. Figs. 5-8 show the behaviour of the L chromosome in two hybrids *P. zeyheri* and *P. lemoinei*. In *P. lemoinei* the two large chromosomes failed to pair quite often and the formation of chiasma between the two arms of the univalent chromosomes was often noted (Fig. 5) showing that this chromosome is an *iso-chromosome*. Misdivision was fairly common and clearly seen during anaphases (Figs. 6-7).

The behaviour of the iso-chromosome differed in the different species and could be correlated with the pollen sterility. Thus in some of the species like *P. grandiflorus* of America and *P. tomentosa* of the Himalayas, the pollen sterility was almost as great as in interspecific hybrids. In general, the iso-chromosomes of the *Coronarii* and *Speciosii* groups were found to be more inert than in the other races.

The percentage of pollen fertility of 9 species and 3 hybrids is given in Table I.

TABLE I

<i>P. microphyllus</i>	% 95
<i>P. californicus</i>	94
<i>P. argyrocalyx</i>	93.6
<i>P. coulteri</i>	92.4
<i>P. Sakamuri</i>	80
<i>P. coronarius</i>	62
<i>P. mexicana</i>	62
<i>P. tomentosa</i>	54
<i>P. grandiflorus</i>	49
<i>Hybrids</i>	
<i>P. zeyheri</i> (<i>P. coronarius</i> × <i>P. grandiflorus</i>)	50
<i>P. lemoinei</i> (<i>P. coronarius</i> × <i>P. microphyllus</i>)	45
<i>P. purpureo-maculatus</i> (<i>P. lemoinei</i> × <i>P. coulteri</i>)	41



FIGS. 1-10.

In the *Tri-specific hybrid Philadelphus purpureo-maculatus* (*P. lemoinei* \times *P. coulteri*), the chiasma frequency was reduced to 1.0. Out of 192 pollen grains of *P. purpureo-maculatus* examined, 20 were found to be binucleate and 1 was four-nucleate. Pollen fertility was only 41%. Fig. 9 shows the formation of binucleate pollen grains by the non-separation of 2 iso-chromosomes during anaphase. When these function the result is a triploid.

CHROMOSOME BEHAVIOUR IN TRIPLOID *Philadelphus*

Upto 11 trivalents were found in pollen mother cells of the triploid "Belle Etoile" (Fig. 10); Of the remaining chromosomes either one or two are bivalents, the rest being univalents. Iso-chromosomes were generally found at the periphery of the cell forming a trivalent, or associated as a bivalent and univalent. The pollen fertility in the triploid was reduced to 24%.

While geographical isolation in *Philadelphus* has not appreciably affected pairing between the 12 pairs of chromosomes in hybrids, it has a distinct effect on the pairing of iso-chromosomes. It seems from these studies that heterozygosity is more pronounced in the iso-chromosome than in the ordinary chromosome.

The persistence of an iso-chromosome in the chromosome complex of the genus *Philadelphus* in which the species are so many and so widely separated in nature is interesting when compared to the function of iso-chromosome in other plants. Permanent iso-chromosomes have been noted in other genera of plants, notably in the *Solanaceae* (E. K. J., unpublished). In *Nicandra* which is a monotypic genus and probably a relic species, Darlington and E. K. J. (1945) found that plants with single iso-chromosome showed delayed germination. The evolutionary bearing of iso-chromosome of *Philadelphus* could only be studied by tracing its behaviour and persistence in successive generation of diploids. This work had to be postponed for geographical reasons. However, Sampson (1955) has studied the progeny of one of the triploids—Bicolore, and according to his findings the chromosome numbers of the progeny are nearer diploids than triploids. He also finds that the distribution of extra chromosome is not at random.

SUMMARY

1. All *Philadelphus* species in nature are diploids $2n = 26$.

2. The chromosome complement of species examined was found to be fairly uniform in morphology and could be represented by the haploid formulæ.

$$1 L_m + 8 M_{st} + 2 S_{st} + 2 S_{sm}.$$

3. The L chromosome in *Philadelphus* was found to be an iso-chromosome with a weak centromere.

4. While geographical isolation in *Philadelphus* has not appreciably affected pairing between the 12 pairs of chromosomes in hybrids, it has a distinct effect on the pairing of iso-chromosomes.

5. Pollen fertility of species and hybrids is related to the behaviour of iso-chromosome.

6. Triploidy in interspecific hybrids is due to the formation of un-reduced gametes by non-separation of iso-chromosomes at meiosis.

ACKNOWLEDGEMENT

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EXPLANATION OF TEXT-FIGURES

FIG. 1. Somatic metaphase in pollen grain division of *P. microphyllus* showing 13 chromosomes with iso-chromosome shaded.

FIGS. 2-4. Metaphase I in pollen mother cells of *P. coronarius*, *P. lemoinei* and *P. microphyllus* showing 13 bivalents.

FIG. 5. Metaphase I of *P. lemoinei* showing 2 iso-chromosomes outside the plate, one paired on itself.

FIG. 6. Anaphase I of *P. lemoinei* showing misdivision of paired iso-chromosomes.

FIG. 7. Anaphase of *P. lemoinei* showing misdivision in unpaired iso-chromosome.

FIG. 8. Metaphase I in *P. zeyheri* showing non-congression of paired iso-chromosome.

FIG. 9. The formation of binucleate pollen grain in *P. purpureo-maculatus* by non-disjunction of iso-chromosome.

FIG. 10. Metaphase I in "Belle Etoile," the triploid hybrid.

All the figures were drawn at the magnification $\times 1,800$.