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(*To be continued*)

A STUDY OF CHROMOSOME NUMBER IN TWO GENERA OF BERBERIDACEAE: MAHONIA AND BERBERIS

HAIG DERMEN

THE STUDY of the chromosome situation in both *Mahonia* and *Berberis* was undertaken to determine the cytological relationships between these two genera and among species in each genus. There are supposed to be some 50 species of *Mahonia* and 175 species of *Berberis*. Rehder (1927) describes 6 *Mahonia* and 48 *Berberis* species hardy in temperate North America. He also gives some interspecies hybrid forms, and one intergeneric form which has never been known to bloom. In the Arnold Arboretum there are 2 species of *Mahonia*, some 50 species of *Berberis* and the intergeneric hybrid just mentioned.

THE GEOLOGICAL HISTORY AND THE PRESENT DISTRIBUTION OF MAHONIA AND BERBERIS. Some five fossil species have been described that were unearthed from tertiary formation in the south of France, northern Italy and Switzerland which showed considerable resemblance to present forms, especially to *M. Aquifolium* and to other species similar to it (Engler & Prantl, 1891). It is a curious fact, that at present not a single species of *Mahonia* and one only of *Berberis*, namely *B. vulgaris*, is found in Europe. *Mahonia* species are found in North and Central America and Eastern and Southern Asia, and *Berberis* species in Eastern and Central Asia, in South America, a few in North America and North Africa and one in Europe (Rehder, 1927).

CYTOLOGICAL STUDIES. Two species of *Mahonia* and 42 species of *Berberis* were studied. Young anthers were smeared on slides in aceto-carmine solution and the chromosomes of pollen mother cells were stained and counted. The time of development of buds for study began in the early part of April and lasted until the end of May. Buds from *B. aggregata* were not ready until June 19.

As the meiotic chromosomes in *Mahoberberis Neuberti*, the intergeneric hybrid, could not be studied, since this plant does not develop buds, it was necessary to make the study from somatic chromosomes. We tried to get root-tips from cuttings of this and of its parent species but were unsuccessful. Later roots were dug up from the arboretum specimens and root-tips were obtained and fixed in killing solution. Root-tips were fixed in a mixture of 1 part 0.5% chromic acid and 1 part 5% commercial formalin (as stock solutions these are kept separate) in small vials for 1 hour or longer depending on the size of the root-tips and were directly transferred into absolute alcohol for 1 hour or longer. This alcohol is replaced with fresh alcohol for the same length of time, then xylol for 1 hour, placed in the oven in equal parts of xylol and soft paraffin until the paraffin melts, put through two changes of melted hard paraffin, and is then embedded and sectioned. For staining, crystal violet-iodine was used. If the material is fixed in the morning, by evening it can be ready for study. This technique has given excellent results both for chromosome counts and for the study of chromosome structure without causing plasmolysis or shrinkage of cells.

CHROMOSOME COUNTS. In the 44 species studied all but one had 14 pairs of chromosomes; one had 28 pairs, namely *B. turcomanica integerrima*, this being a tetraploid varietal form. The following are the species studied by the author: 2 *Mahonia* species: *Aquifolium*, *repens;* 42 *Berberis* species: *aemulans*, *aggregata*, *amurensis*, *amurensis japonica*, *brachypoda*, *canadensis*, *candidula*, *chinensis*, *circumserrata*, *dasystachya*, *diaphana*, *dictyophylla*, *Dielsiana*, *Fendleri*, *Gagnepainii*, *Gilgiana*, *Henryana*, *heteropoda*, *Julianae*, *koreana*, *laxiflora oblanceolata*, *Mouillacana*, *notabilis*, *ottawensis*, *Poiretii*, *polyantha*, *provincialis* var., *Purdomii*, *Rehderiana*, *Sargentiana*, *Sieboldii*, *thibetica*, *Thunbergii*, *Thunbergii Maximowiczii*, *Thunbergii minor*, *Tischleri*, *turcomanica integerrima* (28 pairs), *Vernae*, *verruculosa*, *vulgaris*, *vulgaris atropurpurea*, *yunnanensis*.

The following 10 species were studied by Tischler (1931) all but one with 14 pairs of chromosomes. One was a tetraploid species: *Mahonia Aquifolium*, *M. japonica*, *M. repens*, *Berberis Darwinii*, *B. empetrifolia*, *B. integerrima*, *B. Thunbergii*, *B. Veitchii*, *B. vulgaris*, *B. buxifolia* (28 pairs).

No exact measurements were taken but microscopic observation showed all to have apparently the same size of chromosomes. This point was readily proven when the somatic chromosomes of *M. Aquifolium*, *B. vulgaris*, and *Mahoberberis Neubertii* (the generic hybrid between the two) were studied and drawn under the same magnification.

Size of Pollen Grain

The measurements of pollen grains of a group of plants (Table I) showed some differences but were not considered very striking. The shape of pollen grains of all species of both genera were the same and in no particular detail were they found different. There was no correlation between chromosome number and pollen grain size; therefore it is considered impractical to try to determine by

TABLE I—POLLEN GRAIN MEASUREMENTS AND PERCENTAGE OF STERILITY

Name of species	Size	Sterility %
M. Aquifolium	52.8	65
M. repens	48.4	50
B. diaphana	52.8	4
B. turcomanica integerrima	52.8	10
B. circumserrata	55	nil
B. Gagnepainii	59.4	30
B. verruculosa	57.2	1
B. Vernae	41.8	5
B. brachypoda	50.6	5
B. laxiflora var.	48.4	40
B. vulgaris	45.1	20
B. heteropoda	50.6	12
B. notabilis	47.5	1
B. provincialis serrata	44	30
B. Tischleri (flowers in three)	55	20
" " (flowers in cluster)	46.2	15
B. Julianae	50.6	7
B. Sargentiana	48.4	7
B. Sieboldii	52.8	1
B. dasystachya	46.2	3
B. Fendleri	48.4	5

pollen grain measurement which are tetraploid and which are diploid forms. Some plants showed high percentage of sterility. *B. notabilis*, a hybrid form from a cross between *B. vulgaris* and *B. heteropoda*, has practically no sterile pollen grains and its parent species show quite a high percentage of sterility. Pollen grains of *B. notabilis* measured 47.5 μ in diameter, while pollen grains of *B. vulgaris* measured 45.1 μ and *B. heteropoda* 50.6 μ, the hybrid having pollen grains intermediate in size.

Species and Generic Hybrids

In the Arboretum there are two or three plants of *Mahoberberis Neubertii* that vary from each other somewhat and they all are considered to be hybrids between *M. Aquifolium* and *B. vulgaris.* These hybrids have never been known to develop flowers.

All the above evidence indicates convincingly that these two genera are very closely related. It would not be surprising that if some artificial intergeneric crosses were tried between *Mahonia* and species of *Berberis* other than *B. vulgaris*, it might result in some successful hybrids that would bloom. Some such crosses were made but only one developed fruits with seeds (Table IIa). The failure in others (Table IIb) may have been due to rain that followed pollination. Some of the crosses will be repeated next year with the hope of getting some successful intergeneric hybrids.

The genus *Berberis* has been divided into 15 groups (Rehder 1927) based on group characteristics. In Table III are listed some of the hybrid forms both of *Mahonia* and *Berberis*. The parent species are given with their climatic zone number, the name and region of countries of their native habitat, and the group number to which they belong according to their group characteristics.

This list indicates that in some cases quite divergent forms have been hybridized with apparent success. Mr. Rehder told me that most of these are chance hybrids. It is especially remarkable that some hybrids have been obtained from crosses between evergreen and deciduous species. Mr. Judd, Propagator at the Arnold Arboretum, informed me that *B. verruculosa* and *B. Gagnepainii* cross freely and give rise to many varied hybrids.

Table IIa gives the list of crosses made by the author that produced fruits; however, nothing further can be said about these till the seeds are germinated and plants grown to maturity. Table IIb contains the list of crosses that apparently were unsuccessful. As can be seen crosses were made between very divergent species, especially between evergreen and deciduous forms, as well as bebetween *Mahonia* and *Berberis* species.

Discussion

Although *Berberis* and *Mahonia* are very old genera they have the same chromosome number, and the numerous and widespread species of *Berberis* show no important differences in either chromosome number or chromosome size. The fact that the two genera can be crossed, and that species hybrids are frequently found in *Berberis*, shows that no fundamental change has occurred in the chromosomes of these genera and species. Even the species of *Berberis* from different parts of the world can often be crossed when brought together, even though they must have been separated for very long periods of time.

Since *Mahonia* and *Berberis* have the same number and size of chromosomes and can be crossed, there is some justification for including both of these forms in the same genus.

TABLE IIa—SUCCESSFUL CROSSES

Zone No.	Group No.	Habitat	Species Name ♀	Species Name ♂	Zone No.	Group No.	Habitat
VI ?	5	W. China	B. Tischleri	× B. Henryana	V	12	C. China
V	5	N. W. China	B. circumserrata	× B. notabilis			
V	12	Japan	B. Sieboldii	× B. diaphana	V	5	W. China
V	10	Turkest.	B. heteropoda	× M. Aquifolium	V		B. C. & Ore.
V	10	Turkest.	B. heteropoda	× B. Julianae	VI	4	C. China
			B. notabilis	× B. Thunbergii	V	12	Japan

TABLE IIb—UNSUCCESSFUL CROSSES

V		B. C. to Ore.	M. Aquifolium	× B. Sargentiana	VI ?	4	C. China
V		"	"	× B. Julianae	VI	4	C. China
VI ?	4	C. China	B. Sargentiana[1]	× M. Aquifolium	V		B. C. & Ore.
VI	4	C. China	B. Julianae	× "	"		"
V		B. C. to N. Mex. and Calif.	M. repens	× "	"		"
VI ?	5	W. China	B. Tischleri	× B. Julianae	VI	4	C. China
V	5	N. W. China	B. circumserrata	× B. Vernae	V	9	N. W. China
V	5	W. China	B. diaphana	× B. Sieboldii	V	12	Japan
V	5	W. China	B. Tischleri	× B. Vernae	V	9	N. W. China
"	"	"	"	× B. brachypoda	V	14	C. & N. W. China
V	5	N. W. China	B. circumserrata	× "	"	"	"

[1] *B. Sargentiana* started to form fruits but very soon after shed its fruit, both crossed and non-crossed.

TABLE III—THE LIST OF HYBRIDS (FROM REHDER 1927)

Zone No.	Group No.	Habitat	Parent species	Zone No.	Group No.	Habitat	Hybrid name
V	—	B. C. to Ore.	M. Aquifolium × M. Fortunei	?	—	China	= M. heterophylla
V	—	B. C. to Ore.	M. Aquifolium × B. vulgaris	III	15	Eu.	= Mahoberberis Neuberti
VII or VIII	—	Calif., New Mex. & Mex.	M. pinnata × M. Aquifolium	V	—	B. C. to Ore.	= M. Wagneri
VIII ?	3	Chile	B. Darwinii × B. empetrifolia	VI	1	S. Amer.	= B. stenophylla
VII ?	4	S. W. China	B. pruinosa × B. diaphana	V	5	W. China	= B. Vilmorinii
VI to VII	4	C. China	B. Veitchii × B. vulgaris	III	15	Eur.	= B. Vanfleetii
IV ?	5	Siberia	B. sibirica × B. vulgaris	III	15	Eur.	= B. emarginata
VI	6	W. China	B. Wilsonae × B. aggregata	V	6	W. China	= B. rubrostilla
V	7	N. W. Himal.	B. aristata × B. vulgaris	III	15	Eur.	= B. macracantha
V	10	Turkest.	B. heteropoda × B. vulgaris	III	15	Eur.	= B. notabilis
V	12	Japan	B. Thunbergii × B. vulgaris	III	15	Eur.	= B. ottawensis
V	12	N. China, Amurl.	B. ? Poireti[1] × B. canadensis	V	12	Va. to Ga. and Mo.	= B. durobrivensis
V	12	Caucas.	B. ? chinensis × B. vulgaris	III	15	Eur.	= B. laxiflora
V	12	Caucas.	B. ? chinensis × B. amurensis	II	15	N. E. Asia	= B. Meehanii
V	12	Va. to Ga. and Mo.	B. ? canadensis × B. vulgaris	III	15	Eur.	= B. declinata
V	12	Va. to Ga. and Mo.	B. ? canadensis × B. Fendleri	VI	12	Colo. to New Mex.	= B. Rehderiana

[1] Species with question mark are the species of which identity cannot be certain.

Species differentiation in *Berberis* is not due to changes in chromosome number or to any fundamental change in chromosome structure or genetic constitution. Most of the differences between species are those which might be attributed to mutation associated with geographic isolation. It is possible, of course, that hybridization between closely related forms has played an important part in causing variation in this genus, but the production of polyploid types or fundamental changes in the chromosome complex, produced by wide species hybridization, has evidently not played an important part in the formation of species in *Berberis*.

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CHROMOSOMES OF SOME HEVEA SPECIES

W. N. BANGHAM

IN THE SPRING of 1929 the writer, while a student at Bussey Institution, Forest Hills, Massachusetts, conducted an investigation of the cytology of some tropical plants. Thirty-four somatic chromosomes were found in the root tips of *Hevea brasiliensis* Muell. Arg. from the New York Botanic Garden. Heusser (1919) had reported 16 chromosomes in the vegetative and 8 in the generative cells.

It is possible that the tree which had furnished the above material was possibly abnormally polyploid in make-up. The investigation has been continued in Sumatra in the laboratory of the Plant Research Department of the Goodyear Rubber Plantations Co. and was extended to three other species of *Hevea* which were obtained from the Algemeen Proef Station der AVROS through the courtesy of Dr. Heusser. These other species, *H. Collina* Huber, *H. guianensis* Aubl., and *H. Spruceana* (Benth.) Muell. Arg. were budded on to *H. brasiliensis* stock with very good success. The only meristematic material available, therefore, was the growing point, which in every case gave good division figures. The species *H. guianensis* and *H. collina* were placed by Huber (2) under the subfamily *Euhevea* as they have only one ring of anthers in the staminate column, and the species *H. brasiliensis* and *H. Spruceana* he placed