

# Identification, Genome Sizes, and Ploidy of *Deutzia*

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**ABSTRACT.** The genus *Deutzia*, in the Hydrangeaceae family, includes  $\approx 60$  species that range in ploidy from diploid ( $2x$ ) to tetradecaploid ( $14x$ ). There have been extensive breeding efforts for *Deutzia*, but this has been limited to a few parental species. Although there have been numerous studies of the cytogenetics of some species of *Deutzia*, the ploidy level of many species remains unknown, and there are few cytogenetic data available for *Deutzia* hybrids and cultivars. The purpose of this study was to validate the identification and determine the genome sizes and ploidy of a diverse collection of *Deutzia* species and hybrids using cytology and flow cytometry. Accessions were identified using the most current taxonomic key and voucher specimens were deposited for each at the North Carolina State University herbarium. Corrected and updated species names are provided for all cultivars and accessions studied. Traditional cytology was performed for roots of representative taxa to calibrate the genome size with the ploidy level. The genome size and estimated ploidy were determined for 43 accessions using flow cytometry. Ploidy levels were reported for the first time for three species of *Deutzia* including *D. calycosa* ( $2n = 4x = 52$ ), *D. paniculata* ( $2n = 4x = 52$ ), and *D. glauca* ( $2n = 12x = 156$ ). The base and monoploid genome size ( $1Cx$ ) were somewhat variable and ranged from 1.20 to 2.05 pg. No anisoploid hybrids were documented, suggesting the presence of an interploid block. The information produced from this study are beneficial to future curation, research, development, and improvement of this genus with corrected nomenclature and clone-specific data regarding cytogenetics.

*Deutzia* species are a valuable group of temperate landscape plants grown primarily for their profusions of showy white to pink flowers produced in late spring. Several species have been widely cultivated in Europe since the first *Deutzia* were imported from Japan in the early 18th century. *Deutzia* gained added global popularity in the early 19th century due to the introduction of additional Asian species to cultivation (Styer and Stern, 1979). With access to this diverse germplasm, many additional *Deutzia* hybrids and cultivars were developed through the breeding and selection efforts of Victor Lemoine, his family, and the Lemoine Nursery staff in the 19th and early–

mid 20th centuries in Nancy, France (Wyman, 1971). The development of new hybrids and cultivars has continued since then.

The taxonomic history of *Deutzia* has seen it placed within both the Saxifragaceae and the Philadelphaceae families, although it is currently accepted as a member of Philadelphaceae within Hydrangeaceae (Soltis et al., 1995; Stevens, 2001). *Deutzia* is most closely allied with *Kirengoshoma*, a small genus of rhizomatous perennials that share several morphological traits with *Deutzia* (Hufford et al., 2001). *Deutzia* represents a disjunct genus with species occurring in eastern Asia and Central America. The genus has traditionally been divided into three sections based on morphological differences, with the Asian sections *Deutzia* and *Mesodeutzia* differing in the aestivation of the petals (valvate/induplicate in *Deutzia* and imbricate in *Mesodeutzia*). The central American *Neodeutzia*, rarely treated as a separate genus, differs from its Asian relatives in the number of stamens, with 12 to 15 in *Neodeutzia* compared with 10 in *Deutzia* and *Mesodeutzia* (Hwang, 1993; Styer and Stern, 1979; Zaikonnikova, 1975). Kim et al. (2015) constructed a phylogeny of the genus and suggested that polyphyletic sections *Deutzia* and *Mesodeutzia* should be merged into a single monophyletic section (*Deutzia/Mesodeutzia*), thus reducing the number of sections to two.

Polyploidy has had an important role in the evolution and divergence of angiosperms (Soltis et al., 2015; Wendel, 2015). Repeated cycles of whole genome duplication (sometimes coupled with hybridization) can lead to reproductive isolation, genomic rearrangements, enzymatic multiplicity, epigenetic changes, and novel phenotypes that contribute to biodiversity and speciation (Adams and Wendel, 2005; Chen and Ni, 2006;

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Chen and Yu, 2013; Hegarty and Hiscock, 2008; Laport and Ng, 2017; Madlung, 2013). For plant breeders, knowledge of ploidy is particularly important because it influences reproductive compatibility, fertility of hybrids, and gene expression (Ranney, 2006). There have been numerous cytological studies of *Deutzia*, beginning with Sax (1931) at the Arnold Arboretum (Boston, MA). Compared with other genera within Philadelphae, *Deutzia* exhibits extreme variability in ploidy, with  $2n = 2x = 26$  to  $2n = 14x = 182$  (Table 1), possibly with ploidy variations within species. The other speciose genus in this tribe, *Philadelphus*, has a similar number of species ( $\approx 65$ ) as *Deutzia* (Dirr, 2009). However, unlike *Deutzia*, polyploidy is not found in *Philadelphus*. The greater ploidy variation in *Deutzia* may contribute to its higher degree of morphological variation when compared with other genera in this tribe (Sax, 1931). Both *Deutzia* and *Philadelphus*, as well as *Fendlerella*, also of Philadelphae, have a base chromosome number of  $x = 13$  (Sax, 1931; Ward, 1984).

Genome size (DNA content) can reflect biodiversity, genome evolution, and taxonomic relationships (Laport and Ng, 2017; Ranney et al., 2018; Rounsaville and Ranney, 2010; Soltis et al., 2015). Genome size data can also be used to estimate ploidy in closely related taxa when properly calibrated

with known cytological standards (Jones et al., 2007; Lattier et al., 2014; Parris et al., 2010; Rounsaville and Ranney, 2010; Shearer and Ranney, 2013). A previous report of the genome size of *Deutzia* was the first report of genome size of Hydrangeaceae. Using Feulgen densitometry, Hanson et al. (2001) determined that the 1Cx genome size of *D. prunifolia* is 1.9 pg. The more recent development of flow cytometry has provided a more accurate and efficient method of determining genome size (Doležel et al., 2007). We are not aware of other reports of genome size of *Deutzia* using flow cytometry.

Despite the extensive use and wide cultivation of *Deutzia*, correct identification of the species is challenging and problematic, and differentiation between species is often subtle (Dirr, 2009). With more than 60 species of *Deutzia*, it is possible that many species and cultivars have been traded under incorrect names for years. However, an excellent key for *Deutzia* that provides clear distinctions among taxa was developed by Zaikonnikova (1975).

The broad genetic diversity and array of desirable commercial traits of *Deutzia* provide a valuable base for further breeding and development of future cultivars. However, confusion regarding the proper identity and lack of information regarding cytogenetics of particular accessions and cultivars

Table 1. Previous cytological reports for *Deutzia* species.

Taxon	Chromosome no.	Reference
<i>D. baroniana</i>	$2n = 4x = 52$	Fedorov, 1974; Hanson et al., 2001
<i>D. bungoensis</i>	$2n = 4x = 52$	Niu and Ohba, 2000
	$2n = 6x = 78$	Niu and Ohba, 2003
<i>D. compacta</i>	$2n = 2x = 26$	Fedorov, 1974
<i>D. corymbosa</i>	$2n = 2x = 26$	Sandhu and Mann, 1989
	$2n = 14x = 182$	Chatterjee et al., 1989
	$2n = 6x = 78$	Funamoto and Nakamura, 1994; Niu and Ohba 2000
<i>D. crenata</i>	$2n = 6x = 78$	Darlington and Wylie, 1955; Fedorov, 1974; Funamoto and Nakamura, 1994;
	$2n = 10x = 130$	Niu and Ohba, 2000; Terasaka and Tanaka, 1974
<i>D. discolor</i>	$2n = 8x = 104$	Darlington and Wylie, 1955; Fedorov, 1974
<i>D. floribunda</i>	$2n = 6x = 78$	Niu and Ohba, 2000
<i>D. gracilis</i>	$2n = 2x = 26$	Darlington and Wylie, 1955; Fedorov, 1974; Funamoto and Nakamura, 1992
<i>D. gracilis</i> var. <i>gracilis</i>	$2n = 2x = 26$	Niu and Ohba, 2000
<i>D. gracilis</i> var. <i>microcarpa</i>	$2n = 2x = 26$	Niu and Ohba, 2000
<i>D. gracilis</i> var. <i>zentraroana</i>	$2n = 4x = 52$	Niu and Ohba, 2000
<i>D. hypoglauca</i>	$2n = 2x = 26$	Darlington and Wylie, 1955
<i>D. longifolia</i>	$2n = 8x = 104$	Cave, 1959
<i>D. maximowicziana</i>	$2n = 2x = 26$	Funamoto and Nakamura, 1992; Niu and Ohba, 2000
<i>D. mollis</i>	$2n = 6x = 78$	Darlington and Wylie, 1955; Fedorov, 1974
<i>D. naseana</i>	$2n = 4x = 52$	Funamoto and Nakamura, 1992; Ohba and Akiyama, 1992
<i>D. naseana</i> var. <i>amanoi</i>	$2n = 4x = 52$	Ohba and Akiyama, 1992
<i>D. ogatai</i>	$2n = 10x = 130$	Niu and Ohba, 2003
<i>D. parviflora</i>	$2n = 2x = 26$	Darlington and Wylie, 1955; Fedorov, 1974
<i>D. pulchra</i>	$2n = 8x = 104$	Cave, 1964
<i>D. purpurascens</i>	$2n = 2x = 26$	Darlington and Wylie, 1955
<i>D. scabra</i>	$2n = 2x = 26$	Funamoto and Nakamura, 1992; Niu and Ohba, 2000
	$2n = 10x = 130$	Singhal et al., 1980
<i>D. scabra</i> var. <i>sieboldiana</i>	$2n = 2x = 26$	Niu and Ohba, 2000
<i>D. schneideriana</i>	$2n = 10x = 130$	Darlington and Wylie, 1955
<i>D. staminea</i>	$2n = 2x = 26$	Cave, 1963; Sandhu and Mann, 1989
<i>D. uniflora</i>	$2n = 2x = 26$	Funamoto and Nakamura, 1992; Niu and Ohba, 2000
	$2n = 6x = 78$	Cave, 1959; Fedorov, 1974
<i>D. yaeyamensis</i>	$2n = 2x = 26$	Niu and Ohba, 2000; Ohba and Akiyama, 1992
<i>D. ×candelabrum</i> ( <i>gracilis</i> × <i>scabra</i> )	$2n = 2x = 26$	Fedorov, 1974

constrain the development of informed breeding strategies. The objectives of this study were to validate the identification and determine genome sizes and estimated ploidy of an extensive collection of *Deutzia* species, hybrids, and cultivars.

## Materials and Methods

**PLANT MATERIAL.** Plant material was collected from arboreta and botanical gardens in the United States in June 2014. Plants were propagated by stem cuttings by treating with 5000 mg·L<sup>-1</sup> potassium salt of indolebutyric acid (KIBA) basal dip for 5 s and maintained under intermittent mist until rooted. Plants were then potted in 2.8-L containers with media consisting of 100% ground pine bark supplemented with 1.04 kg·m<sup>-3</sup> dolomitic lime and 0.74 kg·m<sup>-3</sup> granular micronutrients (Micromax; The Scotts Co., Marysville, OH), and grown outdoors on a gravel pad. Each container was topdressed with ≈12 g of 5- to 6-month 15N–3.9P–10K slow-release fertilizer (Osmocote plus 15–9–12; The Scotts Co.). Accessions were field-planted in Fall 2014 at the Mountain Horticulture Crops Research and Extension Center in Mills River, NC. Flowering and vegetative material of mature plants from each accession were collected and pressed in May 2018. For root tip collections, selected taxa were propagated and container-grown as described previously. All accessions were identified according to Zaikonnikova (1975) by Hembree. Herbarium specimens for each accession were prepared and deposited in the North Carolina State University Herbarium in Raleigh.

**GENOME SIZE/PLOIDY DETERMINATION.** Flow cytometry and cytology were used together to determine genome size and ploidy. For flow cytometry, samples were prepared from fully expanded leaf tissue. Approximately 0.5 cm<sup>2</sup> of leaf tissue was placed in a petri dish with 500 µL nuclei extraction buffer (CyStain PI Absolute P Nuclei Extraction Buffer; Sysmex Partec, Görlitz, Germany) and finely chopped with a razor-blade. The resulting solution was filtered through a 50-µm nylon-mesh filter (CellTrics; SysmexPartec) into a 3.5-mL polystyrene tube. Following filtration, 2 mL of nuclei staining buffer (CyStain PI Absolute P; Sysmex Partec), 6 µL RNase A (Sysmex Partec), and 12 µL propidium iodide (Sysmex Partec) were added to the mixture. Then, the solution was incubated at 4 °C in a refrigerator for at least 30 min. The incubated samples were analyzed using a flow cytometer (PA II; Sysmex Partec). A minimum of 3000 nuclei were recorded with a mean coefficient of variation of 6.3%. Two subsamples were prepared from two randomly selected leaves from each accession. Mean fluorescence for each sample was compared with an internal standard, either *Pisum sativum* ‘Ctirad’ (2C = 8.76 pg) or *Magnolia virginiana* ‘Jim Wilson’ (2C = 3.92 pg) (Doležel et al., 1998; Parris et al., 2010), depending on the genome size of the sample. Genome size (2C) was calculated as follows: [DNA content of the standard × (mean fluorescence of the sample / mean fluorescence of the standard)]. The 1Cx monoploid genome size was calculated as (2C genome size / ploidy).

To calibrate the genome size with ploidy levels, chromosome counts were performed for root tips from selected accessions. Root tips from actively growing roots were collected from the root system after removing the container. Root tips were excised and placed in a pre-fixative solution of 2 mM 8-hydroxyquinoline + 0.24 mM cycloheximide to arrest mitosis. Roots were left for 3 h at room temperature (20 °C) in the dark before being moved to a refrigerator for incubation at 4 °C for

an additional 3 h. After the cold incubation period, the roots were triple-washed in cold, distilled water before being transferred to a 1:3 fixative solution of propionic acid and 95% ethanol and stored at room temperature overnight. After a minimum of 14 h, roots were transferred to a storage solution of 70% ethanol. Fixed roots were hydrolyzed in a 1:3 solution of 12 M hydrochloric acid and 95% ethanol for 90 s. The roots were moved to a slide where the tips were excised before being transferred to a new slide with a drop of modified carbol fuchsin (Kao, 1975). After several minutes, a cover slip was placed over the root tips. Then, they were squashed and viewed with a light microscope at 1000× magnification. Highly resolved cells were observed and photographed for each counted accession.

## Results and Discussion

Forty-three accessions were included in this study representing 13 species and 7 interspecific hybrids including 24 named cultivars (Tables 2 and 3). Of the taxa studied, species designations of 13 were inconsistent with the key created by Zaikonnikova (1975) (Table 2). The names of the Zaikonnikova key were compared with the current taxonomy of *Deutzia* and updated when appropriate (The Plant List, 2019). Species names were also corrected when appropriate (Table 2). There were many possible factors that could have contributed to these discrepancies. As Zaikonnikova (1975) stated, *Deutzia* comprise “a difficult group” that present many “problems of identification.” It is difficult to know how many of our germplasm sources have been revisited and re-examined since the date of their initial accessions, and it is likely that the concept and scope of the genus *Deutzia* and species designations have changed considerably since then. Often, particularly in commerce, species names are accepted without question, especially when there are no resident experts at the receiving institution. Therefore, plants may be widely shared and distributed, and they may come to be known by a name that is taxonomically incorrect.

Chromosome counts were completed for *D. gracilis* ‘Nikko Dawn’ (2n = 2x = 26), *D. ×kalmiiiflora* (2n = 2x = 26), *D. paniculata* ‘Dippon’ (2n = 4x = 52), *D. ogatai* (2n = 4x = 52), *D. ×magnifica* ‘Nancy’ (2n = 6x = 78), and *D. discolor* (2n = 8x = 104) (Fig. 1). These taxa served as references for calibrating estimated ploidy with genome sizes.

Estimated ploidy of *Deutzia* included in this study ranged from 2x to 12x. The first known ploidy estimates of *D. calycosa* (2n = 4x = 52), *D. paniculata* (2n = 4x = 52), and *D. glauca* (2x = 12x = 156) were also determined. Ploidy of *D. ogatai*, *D. parviflora*, *D. naseana*, and *D. longifolia* were inconsistent with previous literature (Table 2). It is likely that previous cytological studies of *Deutzia* were challenged by the same taxonomic issues and difficulty with correctly identifying species that were encountered in this study. As such, it is difficult to know for certain which taxa specifically were being used in these studies and if the species designations were correct because herbarium vouchers generally do not exist for these studies. The difficulties surrounding the identification of members of this group are omnipresent and influence the results of virtually all studies thereof. Without the ability to confirm which species have been studied previously, it is sometimes difficult to validate the results of prior studies of *Deutzia*.

Several species of *Deutzia* have been reported as having a ploidy series, including *D. bungeensis*, *D. corymbosa*, *D.*

Table 2. Genome sizes and estimated ploidy levels of *Deutzia* species and cultivars.

Taxon	Received as	Accession no./NCSC voucher no. <sup>z</sup>	Source/ID no. <sup>z</sup>	2C Genome size [mean ± SE (pg)]	1Cx Genome size (pg)	Estimated ploidy (x)
<i>D. gracilis</i>						
'Nikko'	<i>D. gracilis</i>	2014-068/Hembree 2	MCIL	2.87 ± 0.01	1.43	2
'Nikko Dawn' <sup>y</sup>	<i>D. gracilis</i>	2014-057/Hembree 48	JCRA 100404	2.84 ± 0.01	1.42	2
'Pink Minor' <sup>x</sup>	<i>D. scabra</i>	2014-100/Hembree 4	Cornell BG	2.92 ± 0.03	1.46	2
<i>D. hypoglauca</i> <sup>x</sup>	<i>D. rubens</i>	2014-134/Hembree 6	USNA 67798	2.85 ± 0.04	1.42	2
<i>D. calycosa</i> <sup>x</sup>	<i>D. monbeigii</i>	2014-074/Hembree 13	USNA 59699	6.76 ± 0.12	1.68	4
<i>D. calycosa</i> <sup>x</sup>	<i>D. ningpoensis</i>	2014-067/Hembree 11	USNA 59620	6.75 ± 0.01	1.68	4
<i>D. ogatai</i> <sup>y,w</sup>	<i>D. gracilis</i> var. <i>ogatai</i>	2018-060/Hembree 49	JCRA 130638	5.45 ± 0.11	1.36	4
<i>D. paniculata</i>						
'Dippon' <sup>y,x</sup>	<i>D. gracilis</i>	2014-098/Hembree 12	Chicago Botanic Gardens	8.20 ± 0.01	2.05	4
<i>D. parviflora</i> <sup>x,w</sup>	<i>Deutzia</i> sp.	2014-065/Hembree 14	USNA 64514	7.48 ± 0.02	1.87	4
<i>D. discolor</i> <sup>y</sup>	<i>D. vilmorinae</i>	2014-107/Hembree 19	Arnold Arboretum 296-2000A	10.27 ± 0.03	1.28	8
<i>D. discolor</i> <sup>y</sup>	<i>D. discolor</i>	2014-125/Hembree 17	USNA 67633	10.71 ± 0.01	1.34	8
<i>D. discolor</i> <sup>y</sup>	<i>D. globosa</i>	2014-105/Hembree 38	Arnold Arboretum 957-86-A	11.80 ± 0.16	1.48	8
<i>D. pulchra</i> <sup>x</sup>	<i>D. taiwanensis</i>	2014-133/Hembree 21	UGA	11.08 ± 0.15	1.39	8
<i>D. crenata</i> <sup>x</sup>	<i>D. coreana</i>	2014-119/Hembree 24	Arnold Arboretum 460-73-A	11.97 ± 0.13	1.20	10
<i>D. crenata</i>		2014-066/Hembree 39	USNA 72020	12.85 ± 0.02	1.29	10
'Candidissima' <sup>x</sup>	<i>D. scabra</i>	2014-112/Hembree 29	Arnold Arboretum 923-81-A	12.69 ± 0.21	1.27	10
'Codsall Pink' <sup>x</sup>	<i>D. scabra</i>	2014-108/Hembree 26	MCIL	12.47 ± 0.02	1.25	10
'Summer Snow'	<i>D. crenata</i>	2014-058/Hembree 27	JCRA xx0228	12.51 ± 0.21	1.25	10
'Variegata' <sup>x</sup>	<i>D. gracilis</i>	2014-056/Hembree 25	JCRA xx0591	12.56 ± 0.04	1.26	10
'White Splashed'	<i>D. crenata</i>	2014-099/Hembree 32	Cornell BG	12.15 ± 0.10	1.22	10
<i>D. longifolia</i>						
'Elegans' <sup>w</sup>	<i>D. longifolia</i>	2014-106/Hembree 33	Arnold Arboretum 850-80-A	12.68 ± 0.21	1.27	10
<i>D. naseana</i> <sup>x,w</sup>	<i>D. parviflora</i>	2014-073/Hembree 40	JCRA 001389	12.88 ± 0.07	1.29	10
<i>D. schneideriana</i> <sup>a</sup>	<i>D. schneideriana</i> var. <i>laxiflora</i>	2014-102/Hembree 41	Morris Arboretum 1943-020* A	12.71 ± 0.08	1.27	10
<i>D. glauca</i> <sup>x</sup>	<i>D. glabrata</i>	2014-069/Hembree 42	MCIL	16.16 ± 0.20	1.35	≈12
<i>D. schneideriana</i> <sup>w</sup>	<i>D. schneideriana</i>	2014-122/Hembree 23	Arnold Arboretum 196-96-A	17.07 ± 0.18	1.43	≈12

<sup>z</sup>Arnold Arboretum, Boston, MA; Chicago Botanic Garden, Glencoe, IL; Cornell Botanic Gardens (Cornell BG), Ithaca, NY; William G. Hembree (Hembree); JC Raulston Arboretum (JCRA), Raleigh, NC; Morris Arboretum, Philadelphia, PA; Mountain Crop Improvement Lab (MCIL), Mills River, NC; North Carolina State University Herbarium (NCSC), Raleigh; University of Georgia (UGA), Athens; U.S. National Arboretum (USNA), Washington, DC.

<sup>y</sup>Ploidy confirmed with chromosome counts.

<sup>x</sup>Indicates keyed to new species.

<sup>w</sup>Ploidy inconsistent with the literature.

<sup>y</sup>Indicates keyed to same species as received, but taxon has been reclassified as noted here.

Table 3. Genome size and estimated ploidy levels of *Deutzia* hybrids.

Taxon	Accession no./NCSC voucher no. <sup>z</sup>	Source/ID no. <sup>z</sup>	2C genome size [mean ± SE (pg)]	1Cx genome size (pg)	Estimated ploidy (x)
<i>D. xelegantisima</i> ( <i>D. purpurascens</i> × <i>D. scabra</i> var. <i>steboldiana</i> ) 'Rosalind'	2014-109/Hembree 9	MCIL	2.67 ± 0.04	1.33	2
<i>D.</i> 'NCDX1' ( <i>D. xrosea</i> × <i>D. gracilis</i> )	H2007-190-001/Hembree 50	MCIL	2.82 ± 0.01	1.41	2
<i>D.</i> 'NCDX2' ( <i>D. xrosea</i> × <i>D. gracilis</i> ) <sup>y</sup>	H2010-310-034/Hembree 51	MCIL	2.76 ± 0.03	1.38	2
<i>D. xrosea</i> ( <i>D. gracilis</i> × <i>D. purpurascens</i> ) 'Carminea'	2014-114/Hembree 5	Longwood Gardens 1985-0281	2.92 ± 0.03	1.46	2
	2014-096/Hembree 7	Cornell BG	2.87 ± 0.05	1.43	2
	2014-063/Hembree 3	USNA 74356	2.89 ± 0.05	1.44	2
<i>D. xkalmiflora</i> ( <i>D. parviflora</i> × <i>D. purpurascens</i> ) <sup>y</sup>	2014-064/Hembree 10	USNA 59578	2.88 ± 0.04	1.44	2
<i>D. xmaghnifica</i> ( <i>D. scabra</i> × <i>D. discolor</i> ) 'Nancy' <sup>y</sup>	2014-121/Hembree 34	Arnold Arboretum 920-81-A	11.64 ± 0.04	1.46	8
	2014-101/Hembree 15	Cornell BG	9.13 ± 0.14	1.52	6
	2014-075/Hembree 16	USNA 59622	10.96 ± 0.02	1.37	8
	2014-110/Hembree 30	Arnold Arboretum 841-80-C	11.59 ± 0.06	1.44	8
'Supetba'	2014-120/Hembree 31	Holden Arboretum 69-25-85 via Morton Arboretum	11.59 ± 0.05	1.44	8
'Ebumea'					
'Formosa'	2014-113/Hembree 35	Arnold Arboretum 922-81-A	11.75 ± 0.15	1.47	8
<i>D. xhybrida</i> ( <i>D. longifolia</i> × <i>D. discolor</i> ) 'Strawberry Fields'	2014-131/Hembree 20	JCRA 001170	10.89 ± 0.08	1.36	8
	2014-070/Hembree 18	JCRA 020130	11.22 ± 0.19	1.40	8
'Tourbillon Rouge'	2014-124/Hembree 22	Holden Arboretum 99-249	11.27 ± 0.03	1.41	8
'Magicien'	2014-097/Hembree 36	Chicago Botanic Gardens	11.56 ± 0.04	1.44	8
'Pink Pompom'	2014-116/Hembree 37	Arnold Arboretum 9-87-A	11.72 ± 0.13	1.47	8
<i>D. xmyriantha</i> [ <i>D. parviflora</i> × <i>D. setchuensis</i> (Hillier and Lancaster, 2014) or <i>D. gracilis</i> × <i>D. purpurascens</i> (Sargent, 1924)]					

<sup>z</sup>Arnold Arboretum, Boston, MA; Chicago Botanic Garden, Glencoe, IL; Cornell Botanic Gardens (Cornell BG), Ithaca, NY; William G. Hembree (Hembree); Holden Arboretum, Kirtland, OH; JC Raulston Arboretum (JCRA), Raleigh, NC; Longwood Gardens, Kennett Square, PA; Morton Arboretum, Lisle, IL; Mountain Crop Improvement Lab (MCIL), Mills River, NC; North Carolina State University Herbarium (NCSC), Raleigh; U.S. National Arboretum (USNA), Washington, DC.

<sup>y</sup>Ploidy confirmed with chromosome counts.

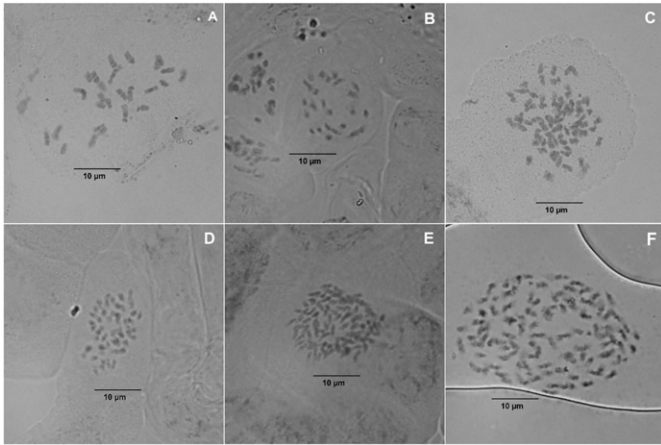


Fig. 1. Photomicrographs of condensed chromosomes of *Deutzia* taxa: (A) *D. gracilis* 'Nikko Dawn',  $2n = 2x = 26$ ; (B) *D. xkalmiiflora*,  $2n = 2x = 26$ ; (C) *D. paniculata* 'Dippon',  $2n = 4x = 52$ ; (D) *D. ogatai*,  $2n = 4x = 52$ ; (E) *D. xmaghnifica* 'Nancy',  $2n = 6x = 78$ ; (F) *D. discolor*,  $2n = 8x = 104$ .

*scabra*, *D. uniflora*, and *D. crenata*. In our study, a ploidy series was not found for *D. crenata*, but it was found for *Deutzia schneideriana*, which has not been previously reported as having a ploidy series. If the accessions in this study were not keyed and verified, then our results would have been quite different. For example, *D. gracilis* 'Pink Minor' ( $2n = 2x = 26$ ), *D. crenata* 'Codsall Pink' ( $2n = 10x = 130$ ), and *D. crenata* 'Candidissima' ( $2n = 10x = 130$ ) were received as *D. scabra* before being keyed out. *Deutzia scabra* and *D. crenata* have shared a close and confusing taxonomic and horticultural history. The common name Pride of Rochester, which is sometimes used as a cultivar name, is variously ascribed to both species (Clapham, 1959; Clarke, 2007). Additionally, *D. crenata* has, at times, been considered a synonym of *D. scabra* (Hillier Nurseries, 1974), and the Flora of China (Huang et al., 2001) recognizes *Deutzia crenata* as being synonymous with *D. scabra* var. *crenata*. Kim et al. (2015) showed that the two are very closely related as sister species in a phylogenetic study of the genus, thus adding to the confusion surrounding the circumscription of the putative taxa.

The accession received as *D. parviflora* 2014-073 and keyed to *D. scabra* var. *latifolia*, which is a synonym of the currently accepted *D. naseana*, has been reported as tetraploid (Funamoto and Nakamura, 1992; Ohba and Akiyama, 1992). This accession was found to be  $2n = 10x = 130$ , which has been reported for *D. scabra* (Singhal et al., 1980), but not for *D. naseana*.

Most of the accessions received as *D. scabra* were cultivars, including 'Candidissima' and 'Codsall Pink', two decaploids, and 'Pink Minor', a diploid. 'Candidissima' and 'Codsall Pink' were keyed to *D. crenata*. Both *D. crenata* and *D. scabra* have been reported to include decaploids (Niu and Ohba, 2000; Singhal et al., 1980). 'Pink Minor' was keyed to be *D. gracilis*, consistent with all other diploid *D. gracilis* included in this study.

Two of the three accessions that were identified as *D. discolor* were received under the names *D. vilmorinae* and *D. globosa*. The ploidy of *D. vilmorinae* and of *D. globosa* have not been reported in previous literature. Flow cytometry data indicated both to be octoploid, which is consistent with

prior reports of *D. discolor* (Darlington and Wylie, 1955; Fedorov, 1974).

*Deutzia parviflora* has been reported as diploid by Darlington and Wylie (1955) and Fedorov (1974), but our accession had a genome size consistent with other tetraploids. The accession of *D. ogatai*, which has been reported as decaploid (Niu and Ohba, 2003), was found to be tetraploid through flow cytometry and chromosome counts.

The one accession of *D. longifolia* had a genome size equivalent to other decaploids, even though it is reported as an octoploid (Cave, 1959). For *D. schneideriana*, which has been reported as a decaploid (Darlington and Wylie, 1955), there was a considerable difference in the genome sizes between our two accessions. *D. schneideriana* 2014-102, received as *D. schneideriana* var. *laxiflora*, had a genome size consistent with that of other decaploids. However, *D. schneideriana* 2014-122 had a genome size in the range expected for a dodecaploid, which has not been reported for *Deutzia*. *Deutzia glauca* had a large genome size similar to that of *D. schneideriana* 2014-122, but its ploidy has not been reported in the literature. Despite considerable efforts, we were unable to confirm the chromosome numbers of these putative dodecaploids with cytology.

The ploidy of hybrids varied from diploid to octoploid and were all isoploid (Table 3). Hybrids sometimes had inconsistent ploidies when compared to their reported parents, although it is difficult to confirm without knowing the ploidy of the exact parents. The lack of any anisoploid hybrids points to the probability of incompatibility of interploid crosses. Some efforts have been made to create interploid hybrids [e.g., *D. xhybrida*  $\times$  *D. gracilis* (T.G. Ranney, personal observation)], but no success has further reinforced the likelihood of an interploid block in *Deutzia*.

The proportions of plants used in this study that were determined to be misidentified underscore the challenges of *Deutzia* taxonomy. The correct identification and determination of genome sizes and ploidy of a wide range of *Deutzia* species and hybrids will provide a valuable resource for breeders and curators. The most current comprehensive key (Zaikonnikova, 1975) was completed in 1966 and is now older than half a century. There have been numerous taxonomic changes in the genus; therefore, an updated and revised key with referenced voucher specimens would be valuable for the continued understanding of *Deutzia*. In addition to the phylogeny of *Deutzia* (Kim et al., 2015), these data will help inform decisions regarding potential interspecific crosses with greater potential for success.

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