

Horticulture, hybrid cultivars and exotic plant invasion: a case study of *Wisteria* (Fabaceae)

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Exotic *Wisteria* species are highly favoured for their horticultural qualities and have been cultivated in North America since the early 1800s. This study determines the identity, genetic diversity and hybrid status of 25 Asian *Wisteria* cultivars using plastid, mitochondrial and nuclear DNA data. Fifteen (60%) hybrid cultivars were identified. All of the '*Wisteria sinensis*' cultivars sampled are hybrids with *W. floribunda*. Although *W. sinensis* and *W. floribunda* are recognized invasive species in the southeastern USA, the relationships of horticultural cultivars to naturalized plants was previously unknown. Haplotype analysis of nuclear data identifies four haplotypes shared between cultivated stock and naturalized populations in the southeastern USA. In addition, US invasive haplotypes are present in New Zealand-derived cultivars although, to date, naturalized *Wisteria* has not been documented in New Zealand. Finally, these data are used to make recommendations to horticulturalists of select species cultivars which may be less likely to invade US landscapes. © 2008 The Linnean Society of London, *Botanical Journal of the Linnean Society*, 2008, 158, 593–601.

ADDITIONAL KEYWORDS: genetic diversity – hybridization – naturalization – plant breeding – urban ecosystems – *Wisteria floribunda* – *Wisteria sinensis*.

INTRODUCTION

Ornamental plant horticulture has had a long history in the USA, beginning with the first botanical garden started by John Bartram in Philadelphia in 1728 (Hedrick, 1988). From this modest start, the nursery and horticulture business in the USA was flourishing by the early 19th century. The growth and sale of useful and attractive plant species inspired American plant collectors to travel around the world in search of novel plants. Both native and exotic plants continue to be selected and introduced into the horticultural trade for their beauty and ease of propagation (Mack,

2005). Today, nearly 60 000 species and varieties of plants are offered by North American nurseries (Ewel *et al.*, 1999).

Unfortunately, many horticultural plants are able to escape the garden and invade natural ecosystems (Reichard & White, 2001). In the eastern USA, 61–68% of plants that were deliberately introduced before 1900 have become naturalized (Mack & Erneberg, 2002). In addition, Reichard & Campbell (1996) found that over 50% of all US invasive plants were introduced for horticultural or ornamental purposes. Because of the increasing negative impacts of invasive species on natural ecosystems, it is crucial to understand the relationship between the plants that are propagated and sold and those that are degrading native habitats.

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HISTORY OF *WISTERIA* CULTIVATION

Plants in the genus *Wisteria* Nutt. are woody vines with deciduous compound leaves and showy, pendent flowers. Currently, *Wisteria* includes five recognized species (Valder, 1995), a number that may grow with taxonomic revision (Zhi, 2005). In the USA, all five species are commonly available in horticulture: the North American natives *W. frutescens* (L.) Poir. and *W. macrostachya* Nutt. ex Torr. & Gray and three exotics, *W. brachybotrys* Siebold & Zucc., *W. floribunda* (Willd.) DC. and *W. sinensis* Sweet (Valder, 1995; Isely, 1998). *Wisteria brachybotrys* and *W. floribunda* are native to Japan and *W. sinensis* is native to southeastern China.

Wisteria spp. are highly favoured for their horticultural qualities and have been cultivated in Japan and China for over 12 centuries (Valder, 1995). The first North American *Wisteria* species was reported in cultivation in England by 1724 under the name *Glycine* (Hedrick, 1988). The Asian species arrived into Western cultivation after China and Japan opened to foreign collectors in the early 19th century. In 1816 John Sims sent *Glycine (Wisteria) sinensis* to England from a garden in Canton, China; *Wisteria floribunda* was first introduced by Phillipp von Siebold to Europe in 1856. These species were introduced to the USA nearly simultaneously (Valder, 1995). Today, *Wisteria* spp. are commonly cultivated throughout the temperate regions of North America, Europe, Asia, Australia and New Zealand.

During their long history of cultivation, *Wisteria* spp. have been selected for a number of different morphological forms. Variation in flower characteristics, including white, pink, mauve, lilac and purple petal coloration and double-flowered forms are known. In addition, there are cultivars with variegated leaves or impressive golden autumn leaf colours, and even dwarf (bonsai) forms are available. Valder (1995) recognized six commercially available cultivars of *W. frutescens*, five of *W. macrostachya*, five of *W. brachybotrys*, 14 of *W. sinensis* and over 27 of *W. floribunda*. In addition, there are a number of unverified cultivar names, cultivars of unknown species identity and suspected hybrids available for purchase (Valder, 1995).

INVASIVE *WISTERIA*

In areas where *Wisteria* spp. are cultivated, these plants have had the opportunity to become naturalized into their new environment. Chinese and Japanese *Wisteria* spp. are considered invasive in 15 eastern states (Remaly, 1999; Alien Plant Working Group, 2005). Naturalized plants occur throughout the eastern USA and range from Vermont to Florida and westward to Texas and Arkansas (USDA, NRCS,

2004). Asian *Wisteria* spp. are listed as invasive by state Exotic Pest Plant Councils (EPPC) in Alabama, Georgia, Florida, Tennessee, South Carolina and Virginia (Miller, Chambliss & Barger, 2004; ALIPC, 2006). Exotic *Wisteria* are designated and monitored as suspected invasive species by the US Fish and Wildlife Service (USDA Forest Service, 2001). It is unknown how long naturalized Asian *Wisteria* plants have been in the USA, but, based on the size of escaped plants, it is likely to exceed 50 years.

HYBRID STATUS OF INVASIVE PLANTS

Preliminary research conducted in our laboratory showed that naturalized plants throughout the southeastern USA are cryptic hybrids between *W. floribunda* and *W. sinensis* (Trusty *et al.*, 2007b). It is unknown whether these naturalized hybrids were formed in urban gardens or whether they are hybrid horticultural cultivars that have escaped. Although the high genetic diversity of naturalized hybrids suggests that hybridization is an ongoing phenomenon, understanding the role of horticulture in the spread of potentially invasive cultivars or genotypes is integral to limiting the future spread of these invasive plants.

This study investigates the identity, genetic diversity and hybrid status of 25 *Wisteria* cultivars currently available in horticulture. This information is the basis for determining the relationships of horticulturally propagated *Wisteria* cultivars to invasive plants in order to identify cultivars/genotypes that are potentially invasive. Finally, these data are used to make recommendations for the horticultural propagation of select non-invasive cultivars and the continued use of *Wisteria* in US landscapes.

MATERIAL AND METHODS

Collections of named *Wisteria* cultivars were made from the living collections of Scott Lathrop (Santa Ana, CA, USA) and Peter Valder (Sydney, Australia). A reference collection of *Wisteria* species from their native ranges was made from wild-collected plants housed in botanical garden living collections. Variable mitochondria and plastid regions were discovered by amplification of previously published gene regions (Taberlet *et al.*, 1991; Zanlugo, Litvak & Jordana, 1991). Two nuclear regions were developed to work within *Wisteria* species in our laboratory through the use of sequence characterized amplified regions (Trusty *et al.*, 2007a, b). Data from 25 individuals in 10 populations of naturalized *Wisteria* are as described in Trusty *et al.* (2007b). Details of the plant material and GenBank accession numbers of the taxa sampled in this study are listed in Table 1. DNA was extracted from fresh or silica dried leaf samples using

Table 1. Genetic identity of study accessions

Genus	Species	Cultivar name	<i>trmL/F</i>	<i>cob</i>	Nuclear 997	Nuclear 824	Notes	Naturalized allele species and location	Genebank accession number
<i>Wisteria</i>	<i>floribunda</i>	'Domino'	F	F	F/F	F/F	Lilac-blue flowers		EF371949-EF371950
<i>Wisteria</i>	<i>floribunda</i>	'Hagoromo Nishiki'	F	F	S/F	F/F	Golden variegated leaves		EF371943-EF371944
<i>Wisteria</i>	<i>floribunda</i>	'Harlequin'	F	F	F/F	F/F			EF371971-EF371972
<i>Wisteria</i>	<i>floribunda</i>	'Honbeni'	F	F	F/F	F/F	Often called Rosea; pink-lavender flowers	F2/Charleston, SC, USA	EF371935-EF371936
<i>Wisteria</i>	<i>floribunda</i>	'Issai Perfect' 1	F	F	F/F	F/F	15 leaflets; light lilac-blue flowers		EF371947-EF371948
<i>Wisteria</i>	<i>floribunda</i>	'Issai Perfect' 2	F	F	F/F	F/F	15 leaflets; light lilac-blue flowers		EF371953-EF371954
<i>Wisteria</i>	<i>floribunda</i>	'Issai Perfect' 3	F	F	F/F	F/F	15 leaflets; light lilac-blue flowers		EF371973-EF371974
<i>Wisteria</i>	<i>floribunda</i>	'Jako'	S	F	F/F	F/F	White-flowered; Japanese introduction		EF371969-EF371970
<i>Wisteria</i>	<i>floribunda</i>	'Macrobotrys'	F	F	F/F	F/F	Exceptionally long racemes		EF371939-EF371940
<i>Wisteria</i>	<i>floribunda</i>	'Macrobotrys Cascade'	S	S	F/F	S/F	Ellerslie Racecourse, Auckland, New Zealand	F1/throughout southeastern USA S1/throughout southeastern USA	EF427645-EF427646
<i>Wisteria</i>	<i>floribunda</i>	'Nana Richins Purple'	F	F	S/F	F/F	Dwarf cultivar		EF371977-EF371978
<i>Wisteria</i>	<i>floribunda</i>	'Royal Purple'	F	F	S/F	F/F	Darkest-flowered cultivar		EF371937-EF371938
<i>Wisteria</i>	<i>floribunda</i>	'Shiro Noda'	F	F	F/F	F/F	Only white-flowered		EF371931-EF371932
<i>Wisteria</i>	<i>floribunda</i>	'Violaceae Plena' 1	F	F	F/F	F/F	Only double-flowered <i>floribunda</i>		EF371945-EF371946
<i>Wisteria</i>	<i>floribunda</i>	'Violaceae Plena' 2	F	F	F/F	F/F	Only double-flowered <i>Wisteria</i>	F1/throughout southeastern USA	EF371985-EF371986

Table 1. *Continued*

Genus	Species	Cultivar name	<i>trmL/F</i>	<i>cob</i>	Nuclear 997	Nuclear 824	Notes	Naturalized allele species and location	Genebank accession number
<i>Wisteria</i>	<i>sinensis</i>	'Amethyst'	S	S	S/F	F/F	New Zealand introduction		EF371951-EF371952
<i>Wisteria</i>	<i>sinensis</i>	'Augusta's Pride'	F	F	S/S	F/F	US introduction		EF427647-EF427648
<i>Wisteria</i>	<i>sinensis</i>	'Blue Sapphire'	S	S	S/S	S/F	New Zealand introduction	F1/throughout southeastern USA	EF371955-EF371956
<i>Wisteria</i>	<i>sinensis</i>	'Cooke's Special' 1	F	S	F/F	F/F	California introduction	F3/ Eufala, AL, USA	EF371959-EF371960
<i>Wisteria</i>	<i>sinensis</i>	'Cooke's Special' 2	S	S	S/S	S/F	California introduction	F1/throughout southeastern USA	EF371979-EF371980
<i>Wisteria</i>	<i>sinensis</i>	'Mercury'	S	S	S/S	S/F		S1/throughout southeastern USA	EF371957-EF371958
<i>Wisteria</i>	<i>sinensis</i>	'Southern Belle'	S	S	S/S	S/F	US introduction	F1/throughout southeastern USA	EF371981-EF371982
<i>Wisteria</i>	<i>sinensis</i>	'Texas White'	F	F	F/F	F/F	US introduction		EF371933-EF371934
<i>Wisteria</i>	<i>sinensis</i>	'C.C. Select'	F	S	F/F	F/F			EF371961-EF371962
<i>Wisteria</i>	<i>sinensis</i>	'Cannington'	F	F	F/F	F/F	UK National <i>Wisteria</i> collection site; Somerset, UK		EF371965-EF371966
<i>Wisteria</i>		'Caroline' 1	F	S	S/F	F/F	Hybrid from New Zealand	F1/throughout southeastern USA	EF371929-EF371930
<i>Wisteria</i>		'Caroline' 2	S	S	S/F	S/S	Hybrid from New Zealand		EF371963-EF371964
<i>Wisteria</i>		'Lavender Lace' 1	F	F	F/F	F/F	Hybrid from US garden		EF371941-EF371942
<i>Wisteria</i>		'Lavender Lace' 2	F	S	S/F	F/F	Hybrid from US garden		EF371983-EF371984
<i>Wisteria</i>		'Texas Purple'	S	S	S/S	S/F	Hybrid from Texas	F1/throughout southeastern USA	EF371967-EF371968
<i>Wisteria</i>		'White Delight'	F	F	F/F	F/F			EF371975-EF371976

Recommended cultivars are in bold type.
cob, cytochrome *b*; F, *Wisteria floribunda*, S, *Wisteria sinensis*.

the 2×CTAB (cetyltrimethylammonium bromide) protocol (Doyle & Doyle, 1987).

NUCLEAR DATA

Two nuclear regions, 824 and 997, were amplified separately using primers w898-824F (5′CATGTTG CATTCAATCTTGG3′), w898-824R (5′GCCTCCATA CAAGTTAGTTG3′), w843-997F (5′GAATCAACGCT GAACGTT3′) and w843-997AluR (5′GGTTCAATT TATTGATGTG3′). These primers were used to amplify all the samples in this study with the following PCR reaction conditions: 0.4 μM forward primer, 0.4 μM reverse primer, 1× Taq polymerase buffer, 0.2 μM dNTPs, 0.25 U Taq polymerase (Eppendorf) and 1 μL of DNA in a 25-μL reaction volume. Thermocycler conditions were 94 °C for 1 min; 35 cycles of 94 °C for 30 s, 52 °C for 30 s, 72 °C for 1 min; followed by 72 °C for 10 min.

Region 824

The PCR product was cloned for each species using the TOPO-TA cloning kit with plasmid vector pCR2 according to the manufacturer's protocol (Invitrogen). Colonies were screened for inserts using PCR primers and the thermocycler programme described in the cloning kit. PCR products of clones were cleaned using Microcon PCR filter units (Millipore). Clones were sequenced in two directions with the cloning primers using the dideoxy chain termination method with an ABI PRISM Dye Terminator Cycle Sequencing Ready Reaction kit. Sequences were obtained on an ABI 3100 automated sequencer following the manufacturer's instructions. Two to four clones were sequenced in two directions for each sample.

Cloned sequences of region 824 were 632 bp long and were aligned manually. Haplotypes were classified as identical only if they matched at every sequence character. Each sample had either one (homozygous) or two (heterozygous) haplotypes corresponding to a diploid chromosome number within *Wisteria*. The sequence data were imported into the program TCS 1.13 (Clement, Posada & Crandall, 2000) and a haplotype network was generated using the 90% statistical parsimony limit. The use of statistical parsimony allows for the valid estimation of genealogical relationships among haplotype (allele) copies at the population level (Clement *et al.*, 2000). This methodology assumes that ancestral haplotypes are the most frequent sequences sampled and allows for recombination between existing haplotypes to be represented in the phylogram or network (Templeton, Crandall & Sing, 1992). Population studies of haplotypes represented by traditional bifurcating trees would be erroneous.

Region 997

Amplified products of nuclear region 997 were approximately 400 bp and contained a single HpyCH4 IV restriction site at 84 bp in *W. floribunda* haplotypes. For all individuals, 5 μL of amplified product was cut for 1 h at 37 °C under the following conditions: 0.5 μL HpyCH4 IV enzyme, 2 μL of 10 × NEB (New England Biolabs) buffer 1 and 12.5 μL of water. Twenty microlitres of the reaction volume were run out on a 1.5% agarose gel containing 4 μL ethidium bromide in 1 × SB (sodium borate) buffer and visualized on an ultraviolet (UV) transilluminator. Haplotypes were scored as *W. sinensis* (single, uncut band), *W. floribunda* (two, smaller cut bands) or hybrid (all three bands).

PLASTID DATA

The plastid *trnL* intron and *trnL-trnF* intergenic spacer (*trnL/F*) of wild collected *W. floribunda* and *W. sinensis* were amplified using the c and f primers according to the protocol described in Taberlet *et al.* (1991). Products were sequenced using the amplification primers as described above. Primers WistrnLF (5′AGTTGACGACATTTCTTAC3′) and WistrnLR (5′GGAGTGAATGGTTTGATCAATG3′) were designed to amplify a 250-bp region that contains a 30-bp deletion in *W. sinensis*. Products for all samples were visualized on a 1.5% agarose gel. Bands were scored by size as *W. floribunda* (F) or *W. sinensis* (S).

MITOCHONDRIAL DATA

A 398-bp fragment of the mitochondrial cytochrome *b* (*cob*) gene that contains an *AseI* restriction site at base 125 in *W. sinensis* was amplified using primers COBRSF1 (5′CATATTGACTTTCTCTCGCC3′) and COBRSR1 (5′GAATAGGATGACTCAGCGTC3′) according to the PCR conditions described in the nuclear methodology above. Five microlitres of PCR product were cut with 0.5 μL *AseI*, 2 μL of NEB buffer 3 and 12.5 μL of distilled water for 1 h at 37 °C and visualized on a 1.5% agarose gel containing 4 μL ethidium bromide in 1 × SB buffer and on a UV transilluminator. Bands were scored by size as cut or uncut, indicating *W. floribunda* (F) or *W. sinensis* (S), respectively.

RESULTS

Table 1 contains the plastid, mitochondrial and nuclear region identification for the 31 accessions of 25 named cultivars sampled in this study. The presence of nuclear haplotypes shared with naturalized individuals is noted (Table 1). Fifteen (60%) hybrid cultivars were identified.

Plastid and mitochondrial markers are normally uniparentally inherited in plants and therefore represent a single maternal or paternal lineage without recombination. In *Wisteria*, the mitochondrial marker *cob*, used in this study, is maternally inherited, whereas the plastid marker (*trnL/F*) is paternally inherited (Hu, Zhang & Sodmergen, 2005; Trusty *et al.*, 2007a). Of the cultivars sampled, three (*W. floribunda* 'Jako', *W. floribunda* 'Lavender Lace' 2 and *W.* 'Caroline') had contrasting cytoplasmic DNA parentage indicating hybrid status.

Nuclear regions are biparentally inherited and are present in two copies in diploid *Wisteria*. If an individual is the product of a first generation cross (F1) between *W. sinensis* and *W. floribunda*, it should have a single copy from each parent for both region 824 and 997. None of the hybrid cultivars sampled was an F1 plant. Hybrid cultivars were identified as later generation (F2+ and backcrosses) hybrids. A single cultivar, *W. sinensis* 'Augusta's Pride' was homozygous for *W. floribunda* alleles for region 824 and homozygous for *W. sinensis* alleles for region 997, whereas most cultivars were hybrid for only one of the two nuclear regions (14 cultivars). The use of two nuclear regions aided in the overall identification of hybrids. Region 997 alone only identified seven (47%) of hybrid individuals whereas region 824 alone identified six (40%) different hybrid lines. In contrast, cytoplasmic markers only identified three hybrid individuals (20%). The discovery and use of additional nuclear markers may be useful in identifying more hybrids within the cultivars sampled.

Haplotype analysis of sequence data of nuclear region 824 identified two networks (Fig. 1). The top network contains the seven haplotypes related to *W. sinensis*, and the bottom network represents the relationships of the 45 *W. floribunda* haplotypes. The central box haplotype represents the inferred ancestral haplotype and is the most common haplotype in each network (Templeton *et al.*, 1992). All of the *W. sinensis* haplotypes were found in hybrid plants (black circles), with the exception of the wild-collected *W. sinensis* individual. In contrast, 58% of *W. floribunda* cultivars were true *W. floribunda* as represented by grey circles. Three out of seven (43%) putative hybrid cultivar individuals were actually true *W. floribunda* species. The hybrid and species haplotypes are intermixed, indicating that hybridization is not restricted to one or a few cultivated lineages.

When the morphological characteristics of the Asian cultivated forms of *Wisteria* are mapped onto the nuclear haplotype network, it is apparent that the white-flowered and double-flowered forms have been selected more than once. *Wisteria sinensis* 'Texas White', *W.* 'White Delight' and *W. floribunda* 'Jako' are all white-flowered cultivars that have unique nuclear

haplotypes. In addition, despite the species identification in the cultivar name, all white-flowered cultivar haplotypes belong to the *W. floribunda* evolutionary lineage. This pattern of unique nuclear haplotypes is also found in the two accessions of the double-flowered form *W. floribunda* 'Violaceae Plena'. Although there is only a single cultivar name for double-flowered *Wisteria*, the plants sampled in this study are genetically distinct and represent separate selection events.

DISCUSSION

Ornamental cultivars are often selected for a wide range of morphological characters, such as colour, scent, height, leaf shape and autumn colour. Plant collectors and breeders search out unique morphological and phenological characteristics in wild populations and from open breeding collections to enhance the original stock and to offer diverse forms for sale (Vainstein, 2002). These actions enhance genetic diversity in cultivated stock (Sakai *et al.*, 2001). The large natural ranges, long history of cultivation and wide variety of unique cultivated forms of Chinese and Japanese *Wisteria* suggest that these species represent a diverse gene pool. Our research has shown that the great majority of cultivars have unique haplotypes for nuclear region 824. These data imply that the high genetic diversity of Asian *Wisteria* may be a result of a large number of introductions from wild populations and/or a high proportion of outcrossed seeds currently maintained in cultivation. Finally, the genetic diversity of white- and double-flowered *Wisteria* cultivars corroborates the numerous selection events in the history of *Wisteria* cultivation.

Once a unique horticultural plant form is found, often it must be asexually propagated to remain 'true' to its morphological traits of interest. *Wisteria* cultivars are asexually propagated through grafting or cutting techniques (Ingram & Yeager, 1990). This process ensures that the progeny is an exact genetic replica of the parent plant. Such unique forms as double-flowered *Wisteria* (*W. floribunda* 'Violaceae Plena') and the many white-flowered cultivars must be asexually propagated to retain these morphological characters. The three accessions of *W. floribunda* 'Issai Perfect' were genetically identical and are likely to have been the result of asexual propagation.

In contrast to asexually propagated lines, most of the cultivars in the study that were represented by two or more accessions were not genetically identical. The two accessions of *W. floribunda* 'Violaceae Plena', *W. sinensis* 'Cooke's Special', *W.* 'Caroline' and *W.* 'Lavender Lace' were markedly different from one another. It may be that some of the cultivated forms breed true for their morphological traits and are

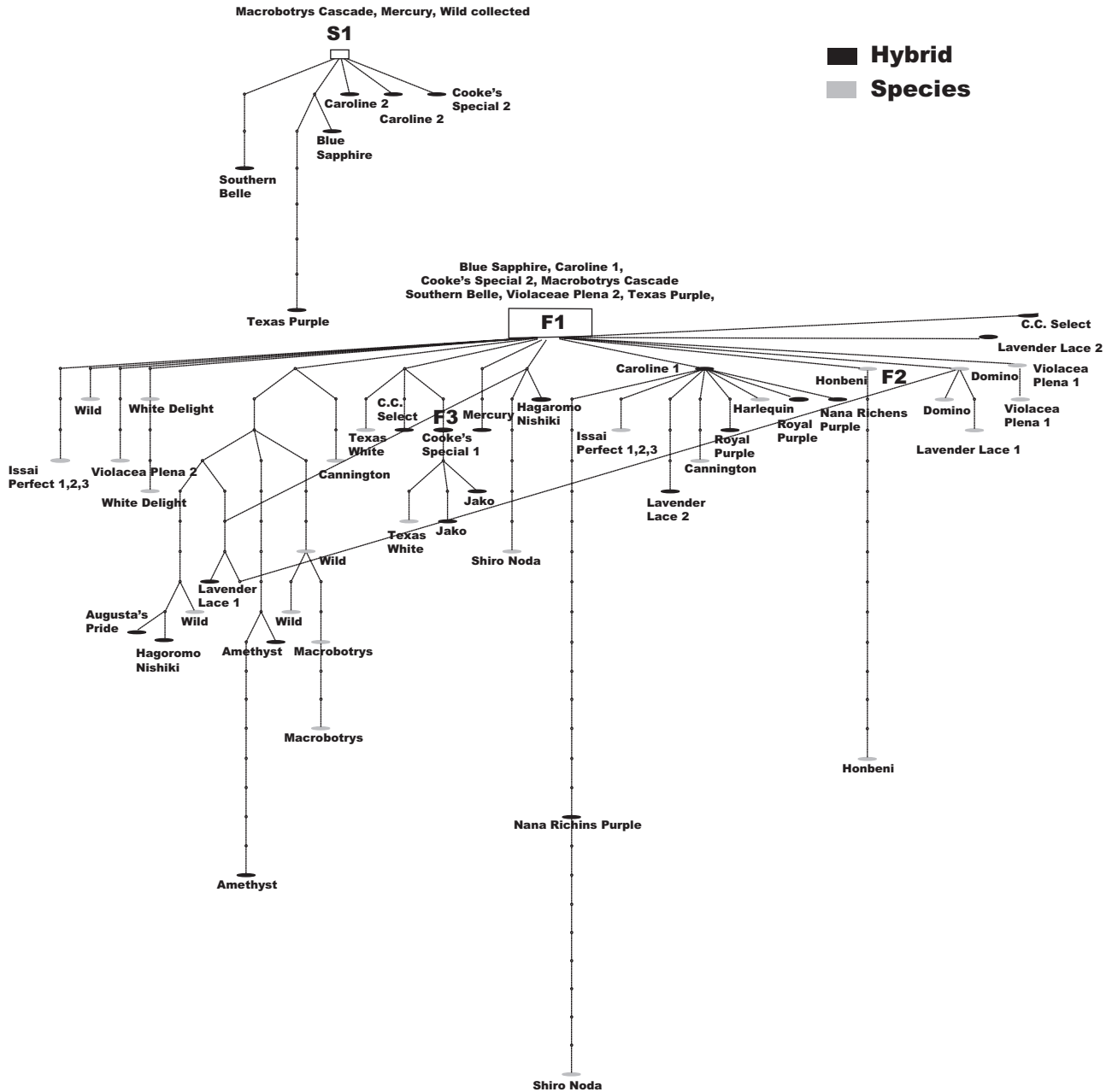


Figure 1. Haplotype networks for nuclear region 824 of cultivated *Wisteria* samples. Top network contains all *W. sinensis* haplotypes; bottom network contains all *W. floribunda* haplotypes. Each branch length implies a single mutational difference and black dots represent unsampled haplotypes. Haplotype names correspond to cultivars listed in Table 1.

being propagated by seed. Additionally, there may be a high diversity of similar morphological forms that are being combined under a single name. Finally, there is the possibility that the cultivar sampled has been mislabelled/misidentified.

Hybrids of Japanese and Chinese *Wisteria* have become common invasive plants in the southeastern USA (Trusty *et al.*, 2007a, b). This study found that hybrids are also common in horticultural cultivation.

All of the recognized *W. sinensis* species cultivars sampled in this study were hybrids with *W. floribunda*. In fact, all of the cultivated hybrids are late generation (F2 or greater), which indicates that these hybridization events were not recent. As most of the hybrid *W. sinensis* cultivars sampled in this study were developed in the USA and New Zealand, it is likely that they are the result of hybridization events outside of China. The 824 nuclear region haplotype

S1 was found in 53% of naturalized US *Wisteria*, in 14% of cultivar accessions and occurred in the Chinese wild-collected accession (Trusty *et al.*, 2007b). To date, this is the only cultivated *W. sinensis* haplotype found in US naturalized populations. Six other *W. sinensis* haplotypes occurred in cultivation, but none of these haplotypes was present in the naturalized individuals sampled in the previous study.

Although true *W. floribunda* cultivars are available in the horticulture trade, 60% of *floribunda* cultivars sampled were hybrids with *W. sinensis*. Most of these cultivars were found to be those developed in the USA and New Zealand (Valder, 1995). Although there is a long history of co-cultivation of these plants in Japan, only one Japanese introduction (*W. floribunda* 'Jako') was found to be a hybrid. Six of the *W. floribunda*-*W. sinensis* hybrid cultivars share *W. floribunda* haplotypes with naturalized plants, whereas only two 824 nuclear region haplotypes are shared between naturalized plants and *W. floribunda* species cultivars. The three haplotypes shared are F1, F2 and F3. Haplotype F1 was present in 43% of naturalized individuals and 23% of cultivated individuals, whereas haplotypes F2 and F3 were each recovered in a single cultivar (3% of the total, respectively).

Why do US- and New Zealand-bred Asian *Wisteria* cultivars share haplotypes with naturalized plants? This result can arise in two ways; US- and New Zealand-bred *W. sinensis* cultivars may be more invasive in the USA or perhaps US breeders have selected their breeding stock from open-bred or naturalized populations. The availability and abundance of naturalized hybrid *Wisteria* in the southeastern USA, and the relatively recent introduction of US cultivars to horticulture, lends weight to the latter hypothesis. These US hybrid cultivars may then have been sent to New Zealand to form the breeding/propagation stock there. No reports of naturalized *Wisteria* have been found from New Zealand, but the availability of potentially invasive haplotypes suggests that these cultivars should be monitored closely.

Once a plant invasion has reached critical numbers, the costs of control increases, while the effectiveness of control decreases (Cousens & Mortimer, 1995). Eradication of the abundant fertile hybrid *Wisteria* in natural environments throughout the southeast will be difficult because of time and cost constraints (Miller, 2003). In areas where naturalized *Wisteria* is not yet a problem, preventing the introduction of invasive genotypes may be the best strategy. This study provides information that can be used to reduce or eliminate the further introduction of potentially invasive genotypes. From our data, we recommend that all hybrid cultivars be voluntarily removed from the US nursery trade. Most of these cultivars are

known to be or are potentially invasive in the USA. In addition, the removal of these cultivars would reduce the chance of spontaneous hybridization with true *W. floribunda* cultivars. Nursery owners should focus propagation efforts on native (*W. frutescens*/*W. macrostachys*) or the non-invasive *W. floribunda* cultivars identified here (Table 1). In addition, all non-native *Wisteria* propagation should proceed via asexual techniques. It is recommended that all newly developed cultivars be genetically tested to determine their hybrid status before release to the US buying public. This study is the first to recognize the invasive potential of hybrid *Wisteria* cultivars and identify species cultivars whose propagation may not negatively impact natural ecosystems in the USA.

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