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Paperbark Maple (*Acer griseum*) Conservation Project

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INTRODUCTION

Despite being a well-known highly ornamental and popular garden plant, paperbark maple, *Acer griseum*, is listed as endangered in its native habitat in central China (Fig. 1).

Figure 1. *Acer griseum* form, fall color, and bark.

As far as can be determined, there have been a limited number of introductions from the wild, with five into the United States, one into the United Kingdom, and one into Finland. The first collection was made by British plant collector and explorer Ernest H. "Chinese" Wilson (E.H. Wilson) for James Veitch and Sons Nurseries in 1901. One other collection of two seedlings was collected by Wilson in 1907 for the Arnold Arboretum. Remarkably, there was good germination from the seed collected in 1901, something that is highly unusual for this species. By 1912, Veitch and Sons Nurseries was offering *A. griseum* for sale in its catalogue. These two collections became the basis for cultivated material in the West.

Besides the two known introductions of *A. griseum* by Wilson, seed was received in the early 1990s from China by Heritage Seedlings, (Salem, Oregon) but it is not known if the seed actually germinated, seedlings collected by the North American-China Plant Exploration Consortium (NACPEC) on its 1994 expedition to Hubei Province, and seed collected in China for Arboretum Mustila (Elimaki, Finland) in 2010. It is likely that plants grown from seed from the Wilson collections and Heritage Seedlings is the source of all plants currently in cultivation. With this in mind, the Morris, Morton, and Arnold Arboreta, and the Beijing Botanical Garden initiated the *Acer griseum* Conservation Project to determine whether the diversity of cultivated plants in the U.S.A. and U.K. accurately reflects the genetic diversity of plants in the wild, or if further efforts are needed to conserve this species.

MATERIALS AND METHODS

This project was divided into the following phases:

- In the summer and fall of 2013 and fall of 2014, Aiello, Bachtell, and Dosmann sampled trees from throughout the U.S.A.
- In July 2014, Aiello and Bachtell visited mature trees throughout the U.K., visiting a total of nine locations and sampling 24 *A. griseum*.
- In September 2015, Aiello, Bachtell, Dosmann, and Wang visited and sampled native populations of *A. griseum* in five provinces throughout its native range in central China.
- Additional collections from China were made by Dosmann in Hubei Province (2014), by staff from the Hunan Forest Botanical Garden in Hunan Province (2015), and from Hangzhou Botanical Garden (2016).
- Samples were sent from Arboretum Mutila, Elimaki, Finland (2016).
- In the fall of 2015 and summer 2016, Aiello sampled trees from other sites on the East Coast of the U.S.A.
- In the summer of 2016, Aiello sampled trees at three locations in the Pacific Northwest.

As far as can be determined, no seed was collected in China by Western botanists for most of the 20th century. Since the 1990s Heritage Seedlings has been the largest source of *A. griseum* seedlings sold throughout the U.S.A., using a seed orchard that was grown from seed collected from a tree at North Willamette Research & Extension Center, Aurora, Oregon, from the Highland Park trees, and possibly from the Henan collection.

Subsequently, the next wild collection occurred on the 1994 North America-China Plant Exploration Consortium (NACPEC) expedition. On the expedition, 16 seedlings were collected, and 10 of these are still alive: four at the Morris, three at the Arnold, and three at the U.S. National Arboretum in Washington, D.C. One other tree of wild origin was included in this study — in the garden of plant explorer Dan Hinkley (Seattle, Washington) — although in

the summer of 2015 unfortunately, this tree suddenly died.

In July 2014 Aiello and Bachtell visited venerable *A. griseum* specimens throughout the U.K. (Fig. 2) with a goal of visiting as many trees known or purported to have been collected by E.H. Wilson in 1901.

Figure 2. Example of an old specimen Aiello and Bachtell visited of *A. griseum* in the U.K.

We visited a total of nine locations and sampled 24 *A. griseum* — some of these were known to be grown from the Wilson collection, others were suspected to be from Wilson, and others originated are from subsequent generations of seedlings. The gardens that were visited were as widespread as the Royal Botanic Garden Edinburgh (Scotland), Newby Hall and Gardens (North Yorkshire, England), Dyffryn Gardens (Vale of Glamorgan, Wales) and Highdown Gardens (West Sussex, England). Many of these collections include original introductions from China, and together they provide an intriguing insight into the world of collecting 100 years ago.

In September of 2015, we completed the third phase of this project, when a NACPEC expedition sampled wild populations of *A. griseum* across its native range in central China. Aiello, Bachtell, Dosmann, and Wang travelled within an approximately 500-mile radius of capital city of Xi'an in Shaaxni Province. The locations stretched from Gansu south to Sichuan and Chongqing, into Shaanxi, Hubei and Hunan, and north into Henan and Shanxi.

Even more helpful was recent work on the distribution, conservation status, and genetic diversity of natural populations of *A. griseum* that had been conducted by a group of researchers at the Chinese Academy of Forestry, Beijing (Chen et al. 2013; Sun et al. 2014). Among these provinces the populations are disjunct from each other, and plants are often scarce within a given area. Despite this, on this expedition we visited nine locations of *A. griseum* in five provinces, resulting in 66 trees sampled, along with two seeds collections.

In late 2015 and then in the summer of 2016, samples were collected from various locations in the eastern U.S.A. A final phase of the project was in late July and early August 2016 when Aiello travelled to Oregon and Washington to sample trees at Heritage Seedlings, North Willamette Research & Extension Center, and Washington Park Arboretum.

In total, the U.S.A. trees that were sampled included:

- Twenty American trees of wild-origin (nine form E.H. Wilson, ten from NACPEC, and one from Hinkley) — 2013 and 2014.
- Six plants of nursery origin from the Morton Arboretum, Lisle, Illinois — 2013.
- One tree at the Glenn Dale, Maryland research station — 2015.

- Two trees at the New York Botanical Garden, Bronx, New York — 2015.
- Two trees from Mount Auburn Cemetery, Cambridge, Massachusetts — 2016.
- Five trees of cultivated origin at the former site of Princeton Nurseries, Allentown, New Jersey — 2016.
- Five trees from Washington Park Arboretum, Seattle, Washington — 2016.
- Two trees at the North Willamette Research & Extension Center, Aurora, Oregon — 2016.
- Forty trees from Heritage Seedlings, Salem, Oregon — 2016.

Leaf samples from all of the sites were collected for RAD-sequence analysis to help answer the question of the degree of genetic diversity represented in cultivation compared to that in the wild. Dr. Andrew Hipp, Systematist and Director of the Herbarium at The Morton Arboretum conducted the analysis of the DNA of the wild and cultivated trees to answer our question of how much genetic diversity is represented in cultivation. The results support the idea that Wilson's 1901 collection provides the basis for all trees in the U.K. and most of what had been in the U.S.A. in the 20th century (Fig. 3).

PROPAGATING TREES FROM TWO UNPROTECTED POPULATIONS

In September 2015 during the third phase of the project, most of the populations that we sampled were located in areas with some level of forest preserve and protection status. But we encountered two populations of paperbark maple that were growing in remote hillsides managed by local farmers. These two populations were located in Chengkou, Chonqing Municipality and near Xixia, Henan Province. The population in Chengkou was growing in an area that is periodically clear cut for fire and construction wood. These trees ranged from 4 to 15 m in height, but some had large basal diameters (up to 45 cm; 17.7 inches) which demonstrated that they had been coppiced several times over a period of many years. The population in Henan was more scattered, and we observed six trees scattered over an area of a few miles.

Wang exchanged contact information with the farmers in both locations so that they could collect dormant scion wood from these trees at a later date for us to use for propagation. The farmers were each paid a stipend, and instructed not to cut down the trees and to cut away vegetation near the trees in order to open up the nearby canopy in the hope to initiate stronger annual growth, and thus better scions for grafting in the future.

In February 2018 we conducted a grafting trial to determine the best understock to use and to hone our grafting skills with *A. griseum*. Two different bare-root rootstocks, sycamore maple (*A. pseudoplatanus*) and sugar maple (*A. saccharum*) were used in the trial. The scions for this trial were collected from trees growing at the Morris Arboretum and Arnold Arboretum from plants of the 1994 NACPEC expedition. Depending on the diameter of the scions either a side veneer or cleft grafting technique was used. Following graftage, the graft wound was wrapped with 0.004 mil 1-in. wide × 2¾ in. long pre-cut pieces of clear poly grafting tape.

Then the grafts were placed in a hot pipe calluser tube in a heavily shaded and vented greenhouse (Fig. 4). The tube's temperature was set to a temperature between 20°C – 22°C (70°F – 74°F) and the grafts were kept in place for 21 days. The grafts were rotated 180 degrees after 10 days to allow for more even heat exposure to both sides of the graft union. After this, the grafts were placed in a sealed box and held in a cooler at 4.4°C (40°F) for 14 days before being potted up. In total 99 grafts were made (49 using *A. pseudoplatanus* rootstock and 50 using *A. saccharum* rootstock).

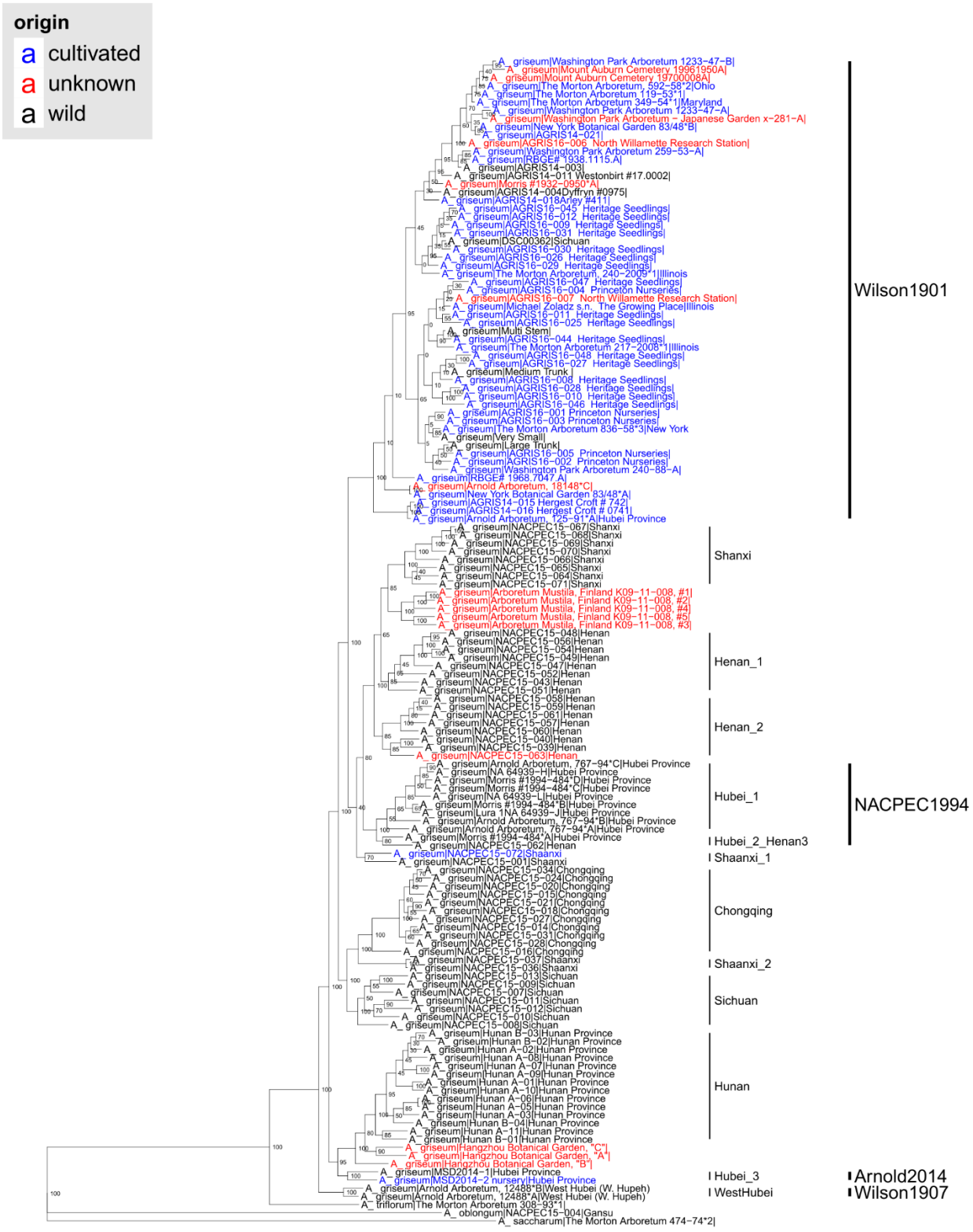

Figure 3. Phylogenetic relationship between different *Acer griseum* accessions.

A total of 71.4% of the grafts were successfully using the *A. pseudoplatanus* rootstock — this species has the remarkable trait of being a "universal rootstock" for many maples, including *A. griseum*. None of the grafts using the *A. saccharum* rootstock were successful. Due to the lack of visual evidence of callus formation when the grafts were in the hot-pipe calluser the grafts were allowed to remain there for an additional 10 days, but to no avail. A noticeable flow of sap arose from the rootstock (and eventually a rather foul odor). It appeared that the sap flow from the rootstock somehow impeded callus formation on both the scion and rootstock.

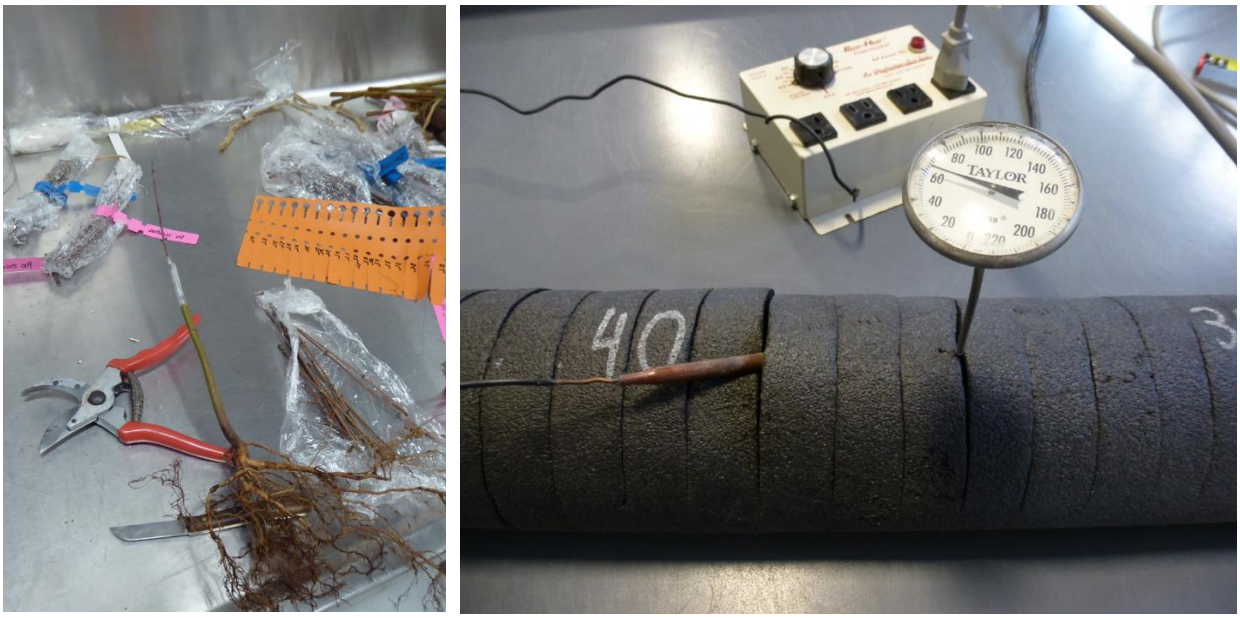

Figure 4. Example of grafted scion (left) and hot pipe calluser tube (right).

Scions from the two unprotected populations from China (Henan and Chongqing) were collected in February 2019. Due to various bureaucratic delays, it was only in late March of 2019 they reached the USDA National Plant Germplasm Plant Quarantine Center located in Beltsville, Maryland. Grafting was performed in early April 2019. Based on the success experienced in the 2018 grafting trial only *A. pseudoplatanus* rootstock was used. As in the earlier propagation trial two graft cuttage techniques were used, either side veneer or cleft, and this was determined largely by the diameter of the scions. Many of the scions were exceedingly thin likely due to the non-vigorous growth of the parent trees. This made the graftage difficult to perform.

Due to the shipping delay and the typically warm ambient temperature during April in Beltsville, Maryland the hot pipe calluser was actually placed in a walk-in cooler that was maintained at 3.3°C (38°F) so the buds on the scions would remain dormant. Following graftage the grafts were treated similarly as the graft in the earlier trial. After 28 days in the heat calluser the grafts were potted up and placed in a shaded greenhouse. After another 45 days the grafts were inspected for success. The percentage of success was lower than the earlier trails and this was believed to be due to the thin diameter of the scions and the substantial delay in time involved with their collection and transport to the U.S.A. from China. There

were 54 total grafts attempted with 21 being judged as successful: a 38.9% success rate.

These plants are required to remaining under quarantine until they are released which is currently scheduled for 2022. Once released they will be distributed to members of NACPEC for further observation, cultivation, and propagation. An additional set of grafts was taken successfully in China. Both groups of young plants will provide the basis for ex situ conservation of the two unprotected populations. It will be interesting to observe their growth performance and to see if this new genetic material improves the viability of the seed set by other *A. griseum* already well established in these NACPEC member gardens public gardens.

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