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Descriptions of Medicago species derivatives available to the public in Plant Introduction Programs in the United States and Australia

Edwin Bingham

Wisconsin Falcata, WISFAL-3, PI 690431, in United States Plant Introduction Program

PEDIGREE: WISFAL-3 descends from Medicago falcata PI 172980, PI 251205, PI 251830, and PI 253443 reported in Crop Sci. 15:889, 1975. These PI entries were mainly diploids, but contained occasional tetraploid plants. Pure M. falcata tetraploids with yellow flower color and M. falcata pod shape were selected for use. The tetraploids in diploid populations likely arose by union of restitution gametes. Tetraploids were identified in the field by larger plant and leaf size, and confirmed in the greenhouse by fertility in crosses with alfalfa, and by chromosome count. Initial tetraploid seed for WISFAL-3 was produced by hand intercrosses of the above PI plants in the greenhouse in 1975. This tetraploid seed, and seed from the diploid plant introductions reported in Crop Sci. 15:889, 1975, were planted in long term nurseries for natural selection for adaptation to Wisconsin, and the North Central Region. The first nursery was at Arlington, WI, from 1977-1982; next at West Madison Area Research Station (WMARS), from 1984-1989. Nurseries were planted in rows, one meter apart, with rows alternating diploid and tetraploid, to allow crossing of diploids and tetraploids, with the potential of new tetraploids by restitution gametes from the diploids. Nurseries were managed as hay fields, never irrigated, never sprayed, until the last year, when the second crop let flower, and set seed by open pollination by native bees. The process was slow recurrent selection for adaptation/naturalization.

In 1990, a sample of tetraploid seed was planted and allowed to flower, to identify plants with yellow flowers, to use for hand pollination seed increase in the winter greenhouse. Finally, this seed was further increased at Fresno, CA, in a 20X20 foot cage, in 1992. This is the WISFAL-3 seed submitted.

NARRATIVE: WISFAL-3, is the last in a series of three tetraploid populations of M. falcata developed from 16 diploid Plant Introductions. The first in the series was WISFAL (PI 560333), the second WISFAL-2 (PI 659349), and third WISFAL-3 (PI 690431). Elevation of diploid germplasm to the tetraploid level permits efficient germplasm transfer to alfalfa in tetraploid X tetraploid crosses.

WISFAL-3 plants are robust with broad crowns, branched root system, and yellow flowers. When cut for hay, WISFAL-3 produces a large first crop and less in succeeding crops. It has not been tested under grazing, but is expected to do as well as WISFAL-1 that persists well in pastures. WISFAL-3 was bred for adaptation by three cycles of crossing plants that survived for five years in three different nurseries. This resulted in WISFAL-3 being the most persistent of all lines after nine years in a nursery at WMARS (2009-2018). WISFAL-3 maintained a visual stand with no large gaps, while WISFAL-1

and several alfalfa cultivars including Vernal had some survivors, but large gaps in stand. Recurrent selection for adaptation/naturalization was effective, and resulted in the remarkable persistence of WISFAL-3.

Alfalfa X Medicago arborea, **Alborea 301, Pl 690775**, in United States Plant Introduction Program

PEDIGREE: Alborea 301 is derived from sexual hybrids of alfalfa X Medicago arborea. Both species are autotetraploids, 2n=4x=32 chromosomes. Only selected alfalfa genotypes produced hybrids when pollinated with pollen from M. arborea. Yellow flower color from M. arborea was used to identify hybrids, that were later confirmed with DNA analysis. The alfalfa parents were MBms, a male sterile plant from Magnum III X Blaser XL, and M8, a complex 3-way hybrid involving three diverse M. sativa genotypes, and three different tetraploid M. falcata genotypes (WISFAL PI 560333). The M. arborea parents were from PI 199254, 504540, 249937, and 330677. Full details are available on line in Medicago Genetic Reports, Vol. 5 onward, on line Journal: Plants MDPI 2:343-353, and Agricultural Science 28:40-46 (Journal of the Australian Inst. Ag. Sci. and Tech.-not on line). Twenty hybrids were produced since 2006, each retaining 8-15 percent M. arborea germplasm. Hybrids were intercrossed and advanced up to three generations in Alborea 301. M. arborea traits segregating in advanced generations indicate that most of the M. arborea genome is represented in Alborea 301.

NARRATIVE: Medicago arborea is a woody perennial bush, native to the Mediterranean region. M. arborea traits of interest include robust stature, large leaves, drought tolerance, large seeds, winter activity, and frost tolerance. Frost tolerance was unexpected, but M. arborea plants have been left in the field until early December, endured several frosts including minus 10C, and retained their green leaves when moved to the greenhouse. Surplus plants left in the field have been observed to grow shoots half meter tall into deep snow drifts. Thus, M. arborea plants are winter active and frost tolerant, as are some Alborea plants. The morphology of Alborea plants is variable, and ranges from resembling alfalfa to resembling M. arborea, with the majority of the plants somewhere in between. Many Alborea plants are more robust than alfalfa, with larger leaves and flowers, and larger pods and seeds. Alborea plants tend to have narrow crowns, and a more branching root system than alfalfa. About half of Alborea plants will have a range of variegated flower color due to the yellow from M. arborea and the purple from alfalfa. Most of the remainder will have either yellow or purple flowers, plus a few plants with cream flowers. Alborea seed production in the field isolation with wild bees is about 80% of alfalfa. If competing bloom of alfalfa is present, Alborea seed production is scant. Alborea crosses well with alfalfa in hand crosses in the greenhouse, thus germplasm can be easily transferred to alfalfa. Moreover, most Alborea X alfalfa progeny express positive heterosis.

Alfalfa Perennial Annual Crosses, **ALPAX, PI 691838**, involving M. arborea, intertexta, sativa, and truncatula, in United States Plant Introduction Program

PEDIGREE: Four Medicago species contributed to ALPAX: 1- M. sativa, three alfalfa clones, M8, MBms, and 6-4ms. 2- M. arborea plants, used as pollen parents. M, sativa and M. arborea are perennial and heterozygous tetraploids, 2n=4x=32 (see: Plants MDPI 2:343-353, on the web). 3- M. truncatula, and 4- M. intertexta, both annual and homozygous. The first perennial X annual cross was purple-flowered alfalfa clone 4x MBms X 4x M. truncatula in 2006. The hybrid had variegated flowers due yellow from M. truncatula. Four more hybrids were obtained in crosses of M8 X 4x M. truncatula, identified by yellow flower color from M. truncatula (Medicago Genetic Reports vol. 13, 2013; www.medicago-reports.org). Since the 2013 report, these hybrids were intercrossed, advanced to F3, and used in a number of experiments. In 2017, a selfsterile, light yellow flowered F3 plant was selected and crossed with M. intertexta. Two hybrids were obtained with dark yellow flower color and robust vigor from M. intertexta. These hybrids contained fractions of genomes from three species: M. sativa, M. truncatula, and M. intertexta, with M. sativa retaining the largest fraction. The next cross added the M. arborea species to ALPAX. This cross involved a hybrid derivative of alfalfa X M. arborea, named 'Alborea' (see: Agricultural Science 28: 40-46, 2016). A self-sterile Alborea plant from PI 690775 was used as seed parent, and pollinated with M. intertexta pollen. One robust hybrid was obtained with strong yellow flower color from M. intertexta. Over the years, these materials with four Medicago species in their pedigree, were crossed in various combinations for experiments, and studied in segregating generations for traits discussed in the following narrative. ALPAX is remnant seed of these experiments.

NARRATIVE: Alfalfa Perennial Annual Crosses, ALPAX, is an off-shoot of research on the feasibility of crossing perennial and annual species of Medicago. ALPAX in intended as a source of new variation for use in alfalfa breeding. There is new variation in the size of seed, leaves, general morphology, and fertility, including autogamy. Hence, it should be useful in restructuring alfalfa. Traits segregating in ALPAX include flower color, number of flowers per raceme, seed size and shape, leaf size and shape, size and extent of serrations, and new plant and root morphology. Also included are pod size and shape, low to high pollen production, and low to high levels of autogamous seed production. Furthermore, recombination in advanced generations will continue to produce new variation. Hence, intercrossing and advancing generations is recommended. ALPAX has not been tested for disease, insect, or stress reactions. Alfalfa Perennial Annual Crosses-Large, **ALPAX-LG, PI 692164**, in United States Plant Introduction Program

PEDIGREE: Plants with large seeds (approximately twice the size of alfalfa seeds) were selected from PI 690775 and crossed with plants selected from PI 691838, that had seeds larger than alfalfa, but smaller than seeds of PI 690775. PI 690775 was derived from hybrids of alfalfa X Medicago arborea, named Alborea 301, as described in the statement for PI 690775. PI 691838 was derived from perennial X annual interspecific crosses involving alfalfa, M. arborea, M. intertexta, and M. truncatula, named ALPAX, as described in the statement for PI 691838. Large-seeded plants from both plant introductions tended to have larger leaves and stems than alfalfa. Ten such plants selected from each PI were paired with the opposing PI and crossed by hand in the greenhouse in winter. Pairs were rotated each day until a partial diallel of PI pairs was completed. This is the ALPAX -LG seed in this release.

NARRATIVE: Alfalfa Perennial Annual Crosses-Large, ALPAX-LG, is a product of research on the feasibility of crossing perennial and annual species of Medicago, plus selection for large plant and seed size. ALPAX-LG is intended as a source of new variation for alfalfa breeding, especially large plant and seed size. Other traits segregating in ALPAX-LG include flower color, number of flowers per raceme, pod shape, leaf size and shape, size and extent of serrations, and new plant and root morphology. Moreover, variation will continue to be produced by recombination in advanced generations of ALPAX-LG. Hence, intercrossing and generation advance is recommended. ALPAX-LG has not been tested for disease, insect, or stress, reactions.

Continued on next page: **Descriptions of Medicago species derivatives placed in the South Australian Research and Development Institute SARDI in 2018 and 2019.**

Alfalfa X Medicago arborea, Alborea-101, APG 84501 in the SARDI Program 2018

PEDIGREE: Alborea-101 was developed from 15 alfalfa X M. arborea hybrids reviewed in Bingham et al. 2013. The first six hybrids were intercrossed to produce an Alborea synthetic population, and additional hybrids were added to the synthetic as they were obtained. Alborea selections from the synthetic population were use in several different breeding strategies to pyramid traits from M. arborea. Remnant seeds of crosses and breeding populations between 2007 and 2017 were bulked for Alborea-101. Most of the alfalfa contribution to Alborea was from two northern-adapted alfalfa clones that would cross with M. arborea. The M. arborea contribution was pollen collected from 10-12 M. arborea plants over the years. The genetic base of Alborea-101 and Alborea 301 (described above) is similar except that Alborea 301 is somewhat broader, and was advanced one more generation before release.

NARRATIVE: Medicago arborea is a woody perennial bush, native to the Mediterranean region. M. arborea traits of interest include robust stature, large leaves, drought tolerance, large seeds, winter activity, and frost tolerance. Frost tolerance was unexpected, but M. arborea plants have been left in the field until early December, endured several frosts including minus 10C, and retained their green leaves when moved to the greenhouse. Surplus plants left in the field have been observed to grow shoots half meter tall into deep snow drifts. Thus, M. arborea plants are winter active and frost tolerant, as are some Alborea plants. The morphology of Alborea plants is variable, and ranges from resembling alfalfa to resembling M. arborea, with the majority of the plants somewhere in between. Many Alborea plants are more robust than alfalfa, with larger leaves and flowers, and larger pods and seeds. Alborea plants tend to have narrow crowns, and a more branching root system than alfalfa. About half of Alborea plants will have a range of variegated flower color due to the yellow from M. arborea and the purple from alfalfa. Most of the remainder will have either yellow or purple flowers, plus a few plants with cream flowers. Alborea seed production in the field isolation with wild bees is about 80% of alfalfa. If competing bloom of alfalfa is present, Alborea seed production is scant. Alborea crosses well with alfalfa in hand crosses in the greenhouse, thus germplasm can be easily transferred to alfalfa. Moreover, most Alborea X alfalfa progeny express positive heterosis.

Medicago Perennial X Annual Species Crosses, **M-PAX**, sample to SARDI in 2019.

The pedigree and narrative for M-PAX are basically the same as for ALPAX above, except that seed submitted for M-PAX was from early generations of the program, and that for ALPAX was advanced one more generation. Thus, the genetic bases are similar, but differ by new variation from recombination, and random drift in the generation advance.

Wisconsin Falcata, WISFAL-3. Sample sent to SARDI in 2019.

The pedigree and narrative are above for WISFAL-3.