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Genetic Resources for Table Grapes in the National Plant Germplasm System

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Table Grapes in History

Grapes, members of the genus *Vitis*, have been used by man for at least 23,000 years (15) and actively cultivated for 5000+ years (16). Many authorities agree that earliest cultivation of grapes, of the species *Vitis vinifera* L., likely occurred in the vicinity of the Trans-Caucasus region, but gradually spread throughout the Mideast, the Mediterranean area, and beyond (11) with evidence of multiple origins for cultivated types (2). Grape germplasm has long been selected for table use, as many old cultivars have markedly larger fruit, crisp flesh, larger clusters, seedlessness, and other traits very different from wild grapes (11). Many other classic table grapes were dual purpose: suitable for table use and wine (e.g. 'Chasselas', 'Cinsault', and 'Muscat of Alexandria'). However, active breeding of grape cultivars specifically for table use was initiated in Europe at the end of the 19th century (8).

Current and New Table Grapes

Seedlessness has become the first requirement of a table grape in the minds of most consumers in the US, where the vast majority of table grape production and sales are based on seedless cultivars. Interestingly, this is not true in most of the world, where seedlessness is critical only for raisins, and flavor may be of greater importance for table use. For example, 'Italia' a seeded grape with a pronounced musky aromatic flavor (muscat) is one of the leading cultivars in Italy, which is the world's number one table grape producing country (9).

Within global viticulture, grape production for wine greatly predominates, and newly developed wine cultivars are of minor importance. In California, which produced 86% of U.S. grapes (wine, table and raisin), 2007 production area was 194,000, 33,000, and 92,000 hectares respectively (10). The table grape industry has been far more receptive to new cultivars than the wine industry and the USDA/ARS release 'Flame Seedless' (14) shares top honors with 'Thompson Seedless' as the most widely planted cultivars for table use in California. Other new releases, such as 'Autumn Royal' and 'Princess', are gaining in acreage (6). 'Thompson Seedless' is the most widely planted U.S. grape cultivar, and is used for raisins, table grapes and wine. This very old cultivar has more than 50 synonyms, with 'Sultanina' and 'Kishmish' being among the most widely used around the world. It likely originated in western Asia and has been used as a source of seedlessness for breeding many of the world's seedless grape cultivars. Most seedless grapes in commerce are "stenospermocarpic", requiring pollination and initial ovule development to set fruit and varying in the degree of seed trace lignification. A few cultivars, most notably 'Black Corinth' (sometimes marketed as Champagne grape, and also used to produce the raisins known as Zante currants), are truly parthenocarpic but produce tiny fruits much smaller than other commercial grapes.

Most taxonomic assessments conclude that there are as many as 60 species in the genus *Vitis*, with about half native to North

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America (8). There is only one species native to Europe and West Asia, and this species, *Vitis vinifera*, is responsible for more than 90% of commercial grape production in the U.S. and around the world. Most grape species are interfertile (except that the subgenus *Muscadinia* does not readily cross with the bunch grapes) and this has resulted in use of interspecific hybridization in many grape breeding programs.

Development of new table grape cultivars falls into three broad categories. Most table grapes are pure *V. vinifera*, and breeding for improved cultivars within this species likely remains the dominant component of table grape breeding around the world and within the U.S. Use of interspecific hybridization largely focuses on introgression of characteristics from non- *V. vinifera* grapes such as different and interesting flavors, colors, and improved adaptation to biotic and abiotic stress, into hybrids with largely *V. vinifera*-like fruit quality and often seedlessness. Finally, there are several active programs breeding for improved cultivars within other species. In the U.S., this is best exemplified by programs focusing on improvement of the Muscadine grape (*Vitis rotundifolia* Michx.) which is native to the southeastern U.S. and is well adapted to pest pressures and environmental conditions of this region.

Grape Germplasm Resources for Plant Improvement

The U.S. National Plant Germplasm System (NPGS) is a component of the Agricultural Research Service of the United States Department of Agriculture (USDA). The National Clonal Germplasm Repository (NCGR) in Davis, California is one of 27 sites in the U.S. National Germplasm System, and is one of nine repositories which emphasize clonal materials. The NCGR houses most of the Mediterranean-adapted fruit and nut crop collections in the U.S., including grapes. The sister repository in Geneva, N.Y. includes most cold-adapted grape accessions. NPGS missions are to acquire, preserve, characterize and distribute germplasm resources of our designated crops. It should be noted that all accessions maintained are distributed without restriction, and therefore, no patented material is included.

Vitis collection of the NCGR. The Davis NCGR collection houses the world's largest grape species collection, including 46 *Vitis* species and totaling around 700 accessions. The repository also maintains a collection of over 1200 accessions of *V. vinifera*, about half of which are "wine grapes" and the other half "table grapes". In addition, the collection has over 900 designated hybrids, ranging from recognized "French hybrids" to *V. labrusca* types and breeders' selections. The collection is further complimented by ~100 *V. rotundifolia* cultivars, a large assortment of wild collected material, and breeders' selections.

The *V. vinifera* collection includes cultivars from the following geographic areas: 167 accessions from France, 159 from Greece, 139 from Italy, 65 from Afghanistan, 61 from Germany, 46 from India/Pakistan, 24 from Hungary, and 16 from North Africa. Where possible, we compare the SSR (simple sequence repeat, a DNA codominant marker very useful for genotyping) fingerprint data from our accessions with those from "type" material at other nations' national collections to verify the identity of Davis NCGR cultivars.

Table grapes in the NCGR collection. Over 1300 Davis-NCGR accessions are considered "table grapes." These include 574 named *V. vinifera* cultivars, 626 "hybrids" (203 American hybrids, 171 French hybrids and 52 *V. rotundifolia* hybrids and 113 advanced breeder selections), 23 *V. labrusca* L. cultivars and 123 *V. rotundifolia*. Among these are 94 seedless accessions and 62 tetraploids. The amazing diversity in this collection offers the opportunity for breeders to select parents providing a host of distinctive traits and also may provide interesting niche cultivars for small acreage plantings. The table grape collection includes great variabil-

ity for fruit color, bloom date, fruit ripening date, compactness of clusters, size of clusters, cold tolerance, disease tolerance/resistance, crop size, plant vigor and many other traits. Grape breeders might be interested in *vinifera* traits such the huge clusters and berries of the 'Uzbekistan Muscat', the early ripening of 'Yaghotti No 2' or 'Perlette', or the long shelf life of 'Calmeria'. Within hybrid accessions, interesting traits include the disease resistance of some cultivars bred in Florida by Fennell and Mortensen, 45 cm-long clusters of 'Koshu Sanjaku', or unusual colors found in some *V. labrusca* hybrids. A niche marketer might be interested in French heirloom 'Chasselas' table grapes with their soft translucent skins, the enormous clusters of 'Marvel of Vaucluse', the albino berries with green rachis in 'Sultana Marble' or the intense perfume flavors of 'Suavis'. They may also wish to target cultivars from specific countries for specialized ethnic markets.

Vitis collection of the PGRU. The Geneva, NY genebank is known as the Plant Germplasm Resources Unit (PGRU), and is located in the Finger Lakes region of western New York, on the research campus of Cornell University called the New York State Agricultural Experiment Station. The PGRU consists of a vegetable crops seed repository and a clonal repository which houses three collections – the apples, the tart cherries, and the cold hardy grapes. The grape collection includes roughly 1500 accessions, focusing on cold hardy material, and therefore few *V. vinifera* accessions are maintained. About 750 of the grape accessions are classified as *V. hybrid*. Most of those hybrids are of the so-called French-American type, which are crosses of *V. vinifera* and American germplasm. These crosses were generally made to combine European quality and American hardiness and disease resistance. There are 188 accessions of the species *V. riparia* Michx., the riverbank grape, that has a native range covering half of the U.S. and well into Canada.

Table grapes in the PGRU collection. Only about 100 PGRU accessions are considered table grapes, and most are interspecific hybrids. Most native grape species are not especially palatable to most people. The most notable cold-hardy U.S. native is *V. labrusca*, the so-called fox grape. *V. labrusca* is best known as the main source of the "grapey" or "foxy" flavor component that comes across so strong in purple grape juice, grape jelly and grape candies. The hybrid cultivar 'Concord' has quite a bit of *V. labrusca* in its pedigree and is widely used in juices and jellies. The PGRU has three pure *V. labrusca* cultivars and one advanced line/selection, as well as many *V. lubrusca* hybrids. Most other hybrid table grapes in the PGRU collection are derived from breeding efforts in which *V. vinifera* has been crossed with other species, for cold-hardiness, disease resistance, and other traits of interest.

Breeding Value of the Vitis Species Collections

The *Vitis* species collections of the NPGS include diverse material with resistance to many biotic and abiotic stresses. Transfer of these genes into cultivars with commercial fruit quality could greatly reduce the need for pesticides and reduce the risk of fruit or plant damage from extreme weather events. In addition, some of these species include flavor and/or appearance traits which would broaden diversity in cultivated grapes.

Unfortunately, these species also have undesirable traits which make them poorly suited to be the parents or even grandparents of commercial table grapes. Conventional breeding would likely require many generations of backcrossing to commercially suitable parents to establish the desired "wild" trait into a largely domesticated background.

Recent development of a "fast-cycling" grape may greatly accelerate this process. Tissue-culture was used to generate plants which are totally derived from the L-1 layer of 'Pinot Meunier', a periclinal chimera of 'Pinot noir'. 'Pinot Meunier' is notable for having a fuzzy surface to its leaves and a

dusty appearing skin on the grape berries, as though the fruit had been sprinkled with flour ("meunier" means "miller" in French). Interestingly, the resulting plants also had extremely short internodes and no tendrils, but instead, flowers at virtually every node (3). These plants produce flowers and fruit while vines are quite young, and many of its hybrids show the same trait. These traits are the result of a semi-dominant mutation in a gene involved in plant response to gibberellins. Indeed, application of GA is required for germination of seeds that will show the fast-cycling phenotype. This 'Pinot Meunier' variant has been developed in the U.S., by Peter Cousins and David Tricoli, and dubbed 'Pixie'. Limited quantities of budwood are now available in the Davis-NCGR.

Crosses between 'Pixie' and a wild grape expressing desired disease resistance (with a known molecular marker), would provide progeny with the 'Pixie' phenotype and resistance marker for use in further crosses within a year of the original hybridization. Repeated crossing, using a group of quality cultivated material as recurrent parents, with selection for 'Pixie' phenotype, the resistance marker, and desired fruit characteristics should yield plants with improved resistance to the target disease with few other "wild" traits within five years. In the final generation, selection for the non-'Pixie' phenotype and resistance marker coupled with more intensive quality and horticultural selection would provide quality cultivars with the desired resistance trait.

An alternative use of the species collection is through transgenic methods. Regulations regarding transgenics make it cheaper and faster to release cultivars using transgenes from closely related species.

Information on the NPGS Grape Collections

As resources permit, data are collected on the characteristics of accessions in the NPGS collections. An ongoing focus has been use of genetic fingerprinting to better understand the genetic diversity in the collections, and assist in genebank management. We collect descriptor data on traits which would be of more interest to horticulturists. Data can be accessed on the websites http://www.ars-grin.gov/dav/ and http://www.ars-grin.gov/gen/, and many are accessible on GRIN (http://www.ars-grin.gov/npgs/acc/acc_queries.html). A National Grape Registry (NGR, www.ngr.ucdavis.edu) has been developed to provide user-friendly, single-site access to information on virtually all grape material in the U.S. So far the NGR includes 713 grape cultivars with synonyms, references, and identified sources for obtaining nursery stock. Data are listed for 5 public collections and 67 commercial nurseries.

Characterization data. In the last few years we have collected detailed data on 100-200 cultivars each year, typically grouping together material originating in the same country. To facilitate comparison across years, a set of diverse standard accessions are included each year. In the last few years we have collected information on grapes of Afghan, Greek, Italian, Portuguese, and Spanish origin. Thirty-six characteristics are included in the dataset, ranging from date of budburst, to node of first cluster, cluster and berry morphology, flavor, Brix at several dates, etc. Photo-documentation is a critical component of accession characterization.

Genetic structure and differentiation in Vitis vinifera L. To evaluate genetic relatedness among numerous *V. vinifera* accessions, eight simple sequence repeat (SSR) loci [*VVS2* (13), *VVMD5, VVMD6, VVMD7, VVMD27, VVMD28, VVMD31*, and *VVMD32* (4,5)] were used to fingerprint 222 cultivated *V. vinifera* and 22 wild *V. vinifera* ssp. *sylvestris* (1). Extensive genetic polymorphism and high levels of heterozygosity were observed among the accessions. All eight loci assayed were polymorphic with the number of alleles per locus ranging from 5 for *VVMD6* to 19 for *VVMD28*, with a total of 94 alleles among the accessions assayed.

Statistical analysis revealed sixteen genet-

ic groups structured into three major clusters, which were consistent with the classical eco-geographic grouping of grape cultivars, *occidentalis, pontica*, and *orientalis* (11). French cultivars appeared to be distinct and showed close affinity to the wild progenitor, *V. vinifera* ssp. *sylvestris* from southwestern France (Pyrenees) and Tunisia, which may reflect the origin of domestication for many traditional French wine cultivars. Associations revealed in this analysis are consistent with other reports of known or presumed parent-progeny relationships (1).

Genetic diversity and classification within V. vinifera table grapes. Analysis of 40 seedless table grape cultivars using eleven highly polymorphic SSR loci confirmed several known synonyms, discovered a previously unknown synonym and disproved an alleged synonym in the literature (7). The data were consistent with known parentage, where such data were available. Two mislabeled vines in the collection were identified. Cluster analysis grouped the cultivars loosely into three groups: a group of nine mostly Middle Eastern cultivars, a group of 22 accessions mostly from Russia and Afghanistan, which were morphologically similar to 'Thompson Seedless', and a third very loose group of 11 accessions consisting mostly of eastern European wine grape cultivars.

Complete SSR fingerprinting of grape accessions. Fingerprinting has recently been completed on a large proportion of the *Vitis* collection of the NPGS. These data will be invaluable for collection management. Since the SSRs used will include the standards for international grape comparison (12), these data will be invaluable for verifying named cultivar accession identities, by matching data with type collections around the world.

Acquisitions and distribution from the NPGS Vitis collection. We are committed to acquiring additional material and are very interested in learning of opportunities, with a special interest in protecting collections which may otherwise be lost. It is NPGS policy to distribute plant material, free of charge, to research interests around the world (see websites at http://www.ars-grin.gov/dav/ and http://www.ars-grin.gov/gen/). Requesters should realize that few NCGR accessions have recent records of virus testing.

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NON-CHEMICAL CONTROL OF DOGWOOD BORER IN APPLE

Dogwood borer infestation of burrknot tissue on apple dwarfing rootstocks is an increasing problem throughout the northeastern United States. The insecticide chlorpyrifos is currently the only efficient chemical control available, but because of EPA scrutiny of pesticides and the desire to increase options available to growers, other control measures are needed. Physical barriers to dogwood borer oviposition may offer a effective control. Kain et al. tested four types of barriers: white latex paint; trunk wraps of Tyvek®; trunk wraps of self-adhesive veterinary gauze; a sprayable, nonwoven ethylene vinyl acetate (EVA). These treatments were compared with both chlorpyrifos and an untreated check. All barriers were effective in preventing dogwood borer infestation and remained intact for the first growing season. After two growing seasons, the EVA treated trees were significantly less infested than trees in the paint treatment or an untreated check. Although barriers were significantly less intact in the Tyvek and gauze treatments than in the EVA treatment in year two, borer infestations were equivalent. EVA was the least costly of the barriers and its cost may be competitive with conventional chemical control. Paraphrased from D. P. Kain et al. 2010. Journal of Entomological Science 45(1):35-43.