Grape Breeding for the Prairies: Inheritance of Resveratrol production in hybrid grapes

2008/09 Annual Project Summary for
The Alberta Horticultural Growers Congress and Foundational Society

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Breeding Overview

Over the last three seasons, I was able to perform multiple controlled crosses utilizing parent vines of diverse genetic backgrounds. Research started in my last year of undergraduate studies has continued through my summer employment with the Fruit Program and now as a graduate student. In 2007, with the help of summer staff, 300 vines from controlled crosses were planted in the U of S hort field plots. Of those vines two promising genotypes have been selected based on winter hardiness (and quality traits), C-16 which will be described in detail and a red-fruited (Valiant x Kay Gray) seedling which produced fruit for the first time this year. This genotype was the hardiest of all the first generation crosses to Valiant and proved to be hardier than Valiant itself having flowered above the snow line following the 2008/2009 'test winter'. This genotype may prove useful in further breeding.

Figure 1. Promising red-fruited (Valiant x Kay Gray) seedling showing greater hardiness than its parent Valiant. Fruit quality is similar to its paternal parent 'Kay Gray'. Cluster size is small because the plants are very young. Currently, there are no red grape varieties for the Prairies.

Due to the low level of hardiness observed in the Valiant-cross, first generation seedlings, I decided to return back to crosses with pure *Vitis riparia* to maintain greater hardiness in progeny. The first step was to select local *V. riparias* with much better fruit characteristics; many genotypes with

little to no ‘herbaceousness’ (a common off type flavour in wild grapes) were selected, cloned and used in controlled crosses. I have also utilized superior male *V. riparias* in breeding; the pollen parent of C-16 is a male Manitoba *V. riparia* that I found growing in an above-ground planter outside the university library, I reasoned that if it could survive -45°C with its roots above ground it must be very hardy! Instead of using inferior ‘quality’ parents like Concord or Fredonia-types, I decided on the best, low acid and neutral flavoured Swenson and French hybrids available, preferably those that already have some *V. riparia* in their backgrounds. I have also used pure *Vitis vinifera* (Pinot Noir, Merlot, Riesling, Cabernet) in some crosses and those will remain in pots in the greenhouse for future breeding. Not all crosses have been directed to wine grapes, I am also looking at producing hardy juice and table grapes; I have bred a promising seedless (Valiant x Petite Jewel) seedling that will be used in further table grape breeding.

In 2008, 3,000 experimental grape vines were planted in Block 4 of the hort field plots and this year (2009) another 4,000 were added. Next year, I hope to plant another 3,000 vines, this time with promising second-generation material based on ‘C-16’, a promising female vine. I have crossed C-16 with some of the best interspecific grape hybrids available, including some recent releases from the University of Minnesota’s grape breeding program in an attempt to combine *extreme* hardiness with high fruit quality. It is in second generation crosses like these that segregating populations begin to throw out the ‘good stuff’. I look forward to evaluating these promising crosses in the future.

Figure 2. Block 4 grape seedling nursery planted in 2008 & 2009 (Top); newly broken soil in. Block 4 ready for 2010’s seedlings (Bottom).

Details on some exciting selections

I was able to identify the superior grape seedling – accession ‘C-16’ which arose from a cross I performed between *Vitis* ‘Severnji’ and a *Vitis riparia* male pollen parent. This breeding was done as part of my undergraduate thesis project (also funded by the Alberta Hort Congress) and is a continuation of the effort to breed high quality grapes for the Canadian Prairies.

'C-16' has proven to be a superior Vitis riparia x F1 in that it not only possesses excellent fruit quality characteristics such as neutral flavours (low herbaceousness), moderate acidity and high sugar (average 23° brix in 2008 & 2009) but the vine itself has many desirable agronomic characteristics. C-16 has survived un-injured in Saskatoon, Saskatchewan through three winters including last winter which was considered particularly harsh; Saskatoon experienced 24 consecutive days where temperatures were no 'warmer' than -25°C! The majority of this cold-snap, saw temps in the -30's with an absolute minimum of -42°C. This past 'test' winter was a welcome event in that it enabled the identification the truly hardy grape genotypes in the field plots. Aside from pure indigenous *Vitis riparia*, genotype C-16 was the only seedling to suffer little to no die-back and flowered from the highest buds on canes trained up a 7' stake. For comparison, the standard Prairie-hardy genotype 'Valiant' died back to the snow line under this period of extended cold, evidently lacking true 'field' hardiness.

Figure 3. Vitis 'Valiant' with dead canes above the snow. Vine is trained on a 7' bamboo stake shown centre, picture taken Aug. 20, 2009.

Accession C-16 flowered for the second season this year (2009) and despite the below-average temperatures this summer, ripened ahead of both 'Valiant' and Manitoba *V. riparia*. Figure 4 illustrates just how early-ripening this genotype is. In a 'normal' year with adequate heat unit accumulation, C-16 may be capable of full ripeness by the last week of August; this year it was estimated to be 2 weeks earlier than Valiant. It was also observed that the fruit of C-16 ripens evenly from top to bottom (within and between clusters) achieving uniform ripeness throughout. Valiant by comparison began to ripen its bottom clusters first where heat had accumulated near the bare soil.

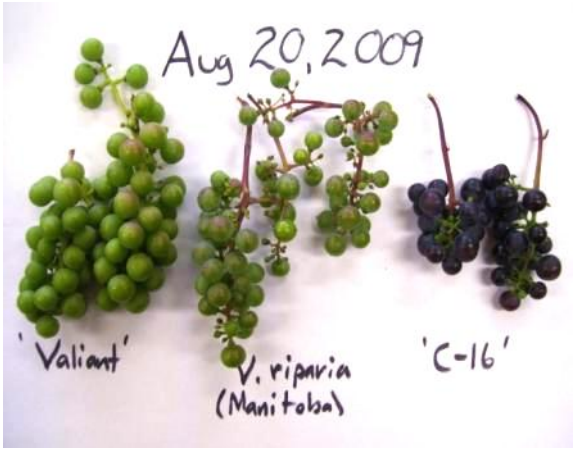

Figure 4. Vitis genotype 'C-16' fruit ripening in comparison with hardy standards 'Valiant' and Manitoba *V. riparia.* Note: C-16 has larger clusters on mature vines (Figure 7).

Figure 5. C-16 with ripe fruit at the top canes.

Figure 6. C-16 with even ripening fruit from top to bottom (Aug. 20, 2009); hardy well above the snow line as well.

Figure 7. Unripe bunch of Vitis 'Frontenac' on top comparable to ripe 'C-16' (bottom) on greenhouse-grown vines. To get large clusters like these in the field, vines must be fully mature and trained properly. Pictures taken on the same day- Sept. 20, 2009.

The pedigree of 'Frontenac', the standard wine grape cultivar in Minnesota is aprox. 25% *Vitis vinifera*, 3% *V. labrusca*, 50% *V. riparia* with lesser amounts of other *Vitis spp.* like *V. rupestris, V. aestivalis, V. berlandieri and V. cinerea.* This type of genetic background maximizes the adapted alleles from the North American species (especially *Vitis riparia*) which provides hardiness and disease resistance and minimizes the negative influence of *V. labrusca* on flavour quality. 'Frontenac' is said to possess 'Vinifera-like' flavour quality with only a theoretical quarter of its genome comprised of *Vitis vinifera* alleles.

Likewise, accession 'C-16' is comprised of aprox. 25% *Vitis vinifera*, and 50% *V. riparia* but contains no *Vitis labrusca* in its background, therefore has no potential to carry off-flavours associated with this species. The rest of C-16's pedigree is filled by *V. amurensis* another super hardy grape species from Asia. In fact it may be favourable, complementary allelic combinations from both *Vitis riparia* and *Vitis amurensis* that makes C-16 better adapted to the mid to high latitudes of the Canadian Prairies than the most Northern *Vitis riparia* ecotypes. In addition, C-16 has 'Precose de Malingre' in its background which is a super-early ripening *V. vinifera*, this vine is also the ancestor of the most notably early-ripening Viniferas like Madelaine Angevine, Sigerrebe, Ortega, Perle de Csaba and Agria. The ability to ripen earlier than Valiant and Manitoba *V. riparia* makes C-16, a potentially valuable breeding parent for the Canadian Prairies. In Saskatoon over the last three growing seasons it has been observed that C-16 not only ripens its fruit first but its also one of the first vines to ripen its canes and drops its leaves; last year (2008) being fully defoliated and dormant by Oct. 10 while Valiant still possessed green leaves and shoots.

Planned research for 2009/2010

The purpose of my graduate research project is in part to determine if C-16 is a superior breeding parent (combining abilities) compared to other standard *Vitis riparia* x F1's like Valiant, Beta, ES 8-2-24 and Frontenac. I will also look at some of the heritabilities of fruit quality traits like pH, sugar content (°Brix), acidity (TA) and the 'functional' trait, 'resveratrol production potential'. Breeding for this nutraceutical compound will give prairie grapes a commercial advantage filling a valuable niche market.

Resveratrol is an 'inducible' trait that needs some sort of stimulus to elicit its production in grape skins. The chosen elicitor for this project is UV-C radiation. This past winter with the help of your organization's funding and a departmental grant, two 60watt UV-C lamps were purchased for this purpose and assembled in a proper work station to conduct further experiments. Analytical (HPLC) equipment was also purchased so that the resveratrol content of harvested fruit can be analyzed in the second phase of my project.

Figure 8. Ultraviolet lamps used in resveratrol induction experiments

Cultivar Development

The experimental design of my thesis project utilizes a half-diallel mating scheme. This past winter I was able to complete all the crosses with an excess amount of seed resulting from the C-16 x Frontenac Gris cross (extra seedlings will be planted directly in the field plots next spring). As mentioned earlier, both parents have desirable traits and I believe that their offspring could be a good step toward cultivar development. Frontenac Gris is a grey mutation (sport) of Frontenac so it actually breeds like a white grape meaning that its offspring will carry recessive alleles for white fruit colour. I am optimistic that the above cross could produce an even hardier version of Frontenac with better characteristics as the early ripening trait of C-16 could help in producing progeny of much lower fruit acidity and like Frontenac, C-16 is very neutral-flavoured. In the next year this breeding strategy and hypothesis will be tested.

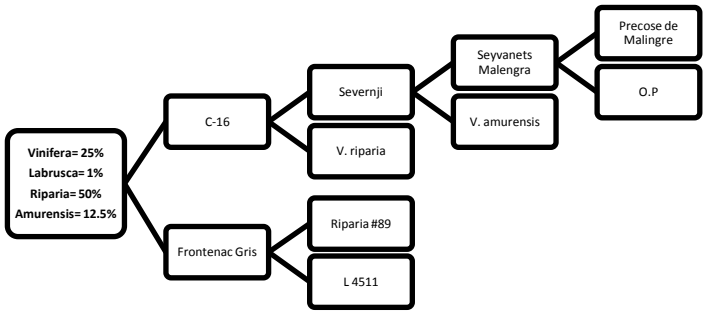

Figure 7. Genetic background of (C-16 x Frontenac Gris) progeny; maintaining a similar pedigree to Frontenac.

Summary of Grape crosses:

2007: 14 types of crosses using the following parents:

Boughen's White
Riparia
Eona
Foch
Himrod
Kay Gray
Minnesota #78
Petite Jewel
Severnji
Somerset Seedless
Suffolk Red
V. riparia
Valiant

2008: 8 types of crosses using the following parents:

Aris
DM 8521-1
ES 8-2-43
Frontenac
Himrod
Louise Swenson
Sabrevois
Selfed Eona
Selfed Valiant

V. riparia
Valiant

2009:31 types of crosses using the following parents:

#5
C-16
DG Riparia
DM 8521-1
ES 10-18-28
ES 2-8-1
ES 8-2-24
ES 8-2-43
Foch
Kandiyohi
Louise Swenson
Petite Jewel
Riparia A
Riparia I
Riparia J
Riparia K
Riparia L
Selfed ES 8-2-24
Somerset
St. Croix
Steuben
Valiant

Planned for 2010: 21 types of crosses using the following parents:

Aris
Beta
C-16
(C-16 x ES 8-2-24)
(C-16 x Frontenac Gris)
DM 8521-1
ES 10-18-30
ES 2-8-1
ES 8-2-24
Foch
Frontenac Gris
Kay Gray
LaCrescent
(Riparia K x Pinot Noir)
St. Croix
Valiant
Vandal Cliché
New York Muscat

The parents used in the breeding program represent a wide range of hardy and semi- hardy varieties useful for wine, juice and table grapes. Some crosses may result in hardy offspring but others will likely require another generation or 2 before hardiness for Prairie conditions can be restored.

Previous to this project, grapes have not been bred at the University of Saskatchewan. Thus, these crosses represent the start of an entirely new breeding program here!

Acknowledgement

I wish to thank the The Alberta Horticultural Growers Congress and Foundational Society and the U of SK's Department of Plant Sciences each of which is providing 45% of funding for my stipend and research expenses. Also, 'Garden's Alive' (a US company from Ohio) is providing 10% of the funding.

I am planning to graduate in Fall or Winter of 2010, at which time I will send a completed copy of my thesis to those who have generously provided me with support.