Phenolic Compound Profiles of Cold Climate Wine Grape Cultivars

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Background and Rationale:

The physical and chemical composition of wine grapes at harvest is a key factor that determines the fruit quality characteristics, and ultimately, the quality of the wine produced. These characteristics of grape vine leaves, canes and roots also reflect attributes such as disease resistance or other stress tolerance traits. These chemical profiles have been well characterized for *Vitis vinifera* cultivars, yet little is known about the chemical composition of cold climate wine grape cultivars. While knowledge of the chemical profiles of berries is important for selecting optimal harvest times to make quality wines, understanding the chemical profiles of vine tissues is also important for breeding the next generation of disease resistant and stress tolerant cultivars. A workflow has been established for chemical profiling of berry composition including phenolic compounds during fruit ripening. A strategy is described here for profiling stilbenoids, which are a large group of phenolic compounds in *Vitis* (including resveratrol) that are associated with human health benefits and fungal disease resistance.

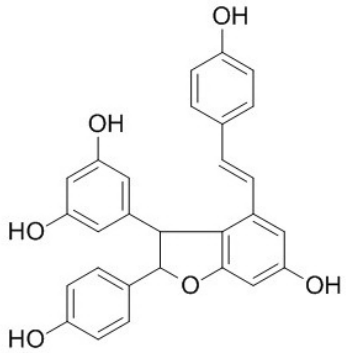

Figure 1: The antifungal stilbenoid ε-viniferin was first identified in *Vitis vinifera.* It and other potentially novel phenolic compounds are also likely to be present in cold climate cultivars. Stilbenoids such as ε-viniferin exist in both grape berries and vines in different chemical forms including monomers (such as resveratrol), dimers (such as ε-viniferin), trimers, tetramers and higher order multimers.

Experimental Design:

The favored hypothesis is that stilbene multimers (dimers, trimmers, tetramers etc.) are assembled under genetic control by diverse sets of enzymes that vary with pedigree across cold climate grapevine cultivars. An alternative hypothesis is that oligomerization occurs spontaneously by oxidation or during lignification and is not under genetic control. To test these hypotheses, stilbenoids were profiled from a broad sampling of cold climate grapevine cultivars to assess overall stilbenoid compound diversity. The analysis was carried out on 17 cultivars including one *Vitis riparia*, one *V. amurensis,* and 15 cold climate wine grape or table grape hybrid cultivars (listed in Figure 2).

Methods:

Dormant cane tissue was collected from the 17 cultivars in March of 2013 and pulverized in a burr grinder. The powdered cane tissue was extracted with 60% acetone/40% water (v/v) prior to liquid chromatography-mass spectrometric (LC-MS) analysis. Stilbenoid compound profiles were measured separately by LC-MS using a hybrid quadruple Orbitrap Q Exactive mass spectrometer equipped with a Dionex Ultimate 3000 HPLC. Chromatographic separation was accomplished using a Waters C_{18} reversed phase, HSS T3 column (2.1 × 100 mm, 1.8 µm) eluted with a gradient of 0.1% formic acid in water to 0.1% formic acid in acetonitrile.

Results:

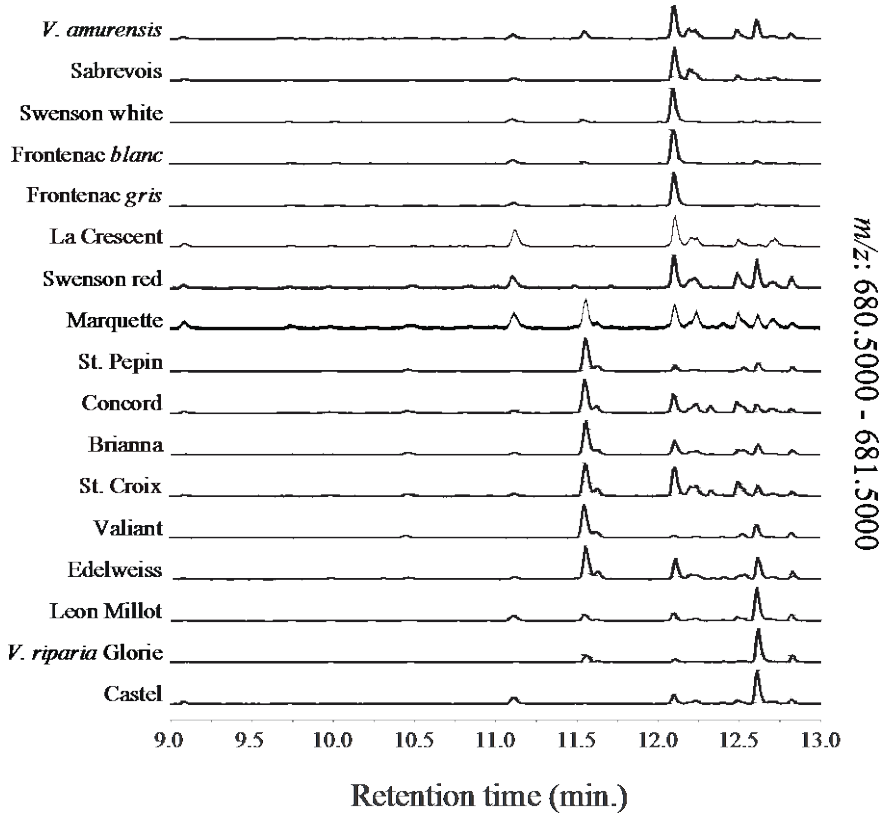

Figure 2: LC-MS stilbene trimer profiles. This figure shows extracts of cane tissues from the cultivars listed along the left side of the figure. Only chemicals with masses consistent with one class of trimeric stibenoids will appear as peaks in the traces next to each cultivar name. The peaks appearing at different retention times (bottom axis) indicate chemically distinct trimeric stilbenoid of the same mass. The intensities of different trimeric stibenoids change from cultivar to cultivar.

What the results mean:

- The high variability and apparent influence of pedigree on stilbenoid profile patterns are consistent with genetic control of oligomerization rather than spontaneous oligomerization
- Stilbenoid profiles are consistent with a high degree of genetic variation in stilbenoid biosynthesis within existing cold climate cultivars
- These findings show that there is potential for correlating disease resistance phenotypes with stilbenoid profiles, which may be further linked with biosynthetic genes for use in marker assisted selection for future breeding efforts.