

A view into American grapevine history: *Vitis vinifera* cv. 'Sémillon' is an ancestor of 'Catawba' and 'Concord'

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Summary

The *Vitis vinifera* background of 'Catawba' and 'Concord' was investigated by using SSR analysis: 'Sémillon' was shown to be an ancestor of 'Catawba', while the wild parent remains unknown. 'Concord' was confirmed to be an offspring of 'Catawba' and another unknown wild parent. Since these two important American varieties most likely resulted from random natural crosses and successful selection, the original, wild growing wild donors remain unknown.

Key words: *Vitis aestivalis*; *Vitis labrusca*; genetic fingerprinting; multiplex PCR; microsatellite; SSR; parentage.

Introduction

According to MUNSON (1909) the initial stock of the later called variety 'Catawba' was discovered in a forest near the Catawba River in North Carolina in 1801. Due to its appealingly dark red berries, very juicy pulp and extraordinary aroma composition, it soon developed to the first well-known American cultivar having commercial importance (ROBINSON *et al.* 2012). Nowadays, 'Catawba' is still widespread around New York State and mainly used for table grape and juice production. In 2006, 'Catawba' was grown on an area of 522 ha in this region (ROBINSON *et al.* 2012). The winegrower John Adlum ("Father of American Viticulture") introduced 'Catawba' in the District of Columbia in the year 1823 where it became the first prominent variety planted expansively to produce table grapes, juice and wines (HEDRICK 1908). Another commercially important American cultivar, introduced after 'Catawba', is 'Concord'. Believing the historical written records, 'Concord' is a descendant of 'Catawba': Ephraim Bull from Massachusetts dug up a wild *V. labrusca* next to his fence and planted it on his lot next to other grapevine cultivars, including 'Catawba' (TUKEY 1966). E. Bull planted the seed from this *V. labrusca* accession in 1843 and 'Concord' was selected from among the seedlings. Some characteristics such as the hermaphrodite flowers of 'Concord' and the oval berries in some 'Concord' offspring gave a hint about its *V. vinifera* portion (TUKEY 1966). Both 'Catawba' and 'Concord' wines feature the typical, strong

wild foxy flavor (NELSON *et al.* 1977, RAPP *et al.* 1980, RAPP *et al.* 1993).

While the historical origin of 'Catawba' can be retraced rather easily, the actual parents were discussed extensively in the last two centuries (PRINCE 1830, TUKEY 1966, GALET 2000, PINNEY 2007). In any case, the close relatedness to a wild species and the fact that 'Catawba' arose by chance can be stated (ROBINSON *et al.* 2012). Two main origin scenarios were discussed on the basis of the ampelography: 'Catawba' to be (1) a true wild *V. labrusca* variety or (2) an interspecific cross of the wild grapes *V. aestivalis* or *V. labrusca* with an unknown *V. vinifera* cultivar introduced to North America at this time (HEDRICK 1908). *V. labrusca* is a native wild grapevine species in North America and was firstly described by LINNÉ in 1763 (AMBROSI 2011). Microsatellite analysis of the genetic resources at the JKI Geilweilerhof resulted in first indications that the European cultivar 'Sémillon' could be the *V. vinifera* parent.

Material and Methods

'Sémillon' (GALET 2000, LACOMBE *et al.* 2013), 'Catawba' and 'Concord' (HEDRICK 1908) were confirmed as ampelographically true to type accessions within the grapevine collection at the Julius Kühn-Institut (JKI), Institute for Grapevine Breeding Geilweilerhof. Analyzing the fingerprints of 600 genotypes with the statistical software FaMoz (GERBER *et al.* 2003), the one from 'Sémillon' was the only one with a good match with 'Catawba'. Young leaf material from these accessions was lyophilized (Lyocube, Christ, Germany) and DNA was extracted with the aid of a kit (NucleoSpin® 96 Plant II, Macherey-Nagel, Germany). DNA of reference accessions from the germplasm collection in Geneva were kindly provided by T. Chao (U. S. Department of Agriculture, USDA). The analysis didn't include any *V. labrusca* or *V. aestivalis* accession.

The multiplex PCR was conducted with the KAPA2G Fast Multiplex PCR Kit (KAPABIOSYSTEMS, USA) comprising up to 10 primer pairs with fluorescent labels (forward primer coupled with HEX, ROX/PET, TAMRA or FAM). PCR program: 95 °C for 3 min (initial denaturation), 95 °C for 15 s (denaturation), 60 or 58 °C for 30 s (primer annealing), 72 °C for 30 or 50 s (elongation) and 72 °C for 3 min

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(final elongation) with 30 cycles of denaturation, annealing and elongation. The 137 oligonucleotides used were VMC from the *Vitis* Microsatellite Consortium in France, UDV (DI GASPERO *et al.* 2005), VVI (MERDINOGLU *et al.* 2005)VVMD (BOWERS *et al.* 1999), VCHR (CIPRIANI *et al.* 2008), VtZAG (SEFC *et al.* 1999), in-house GF (ZHANG *et al.* 2009, FECHTER *et al.* 2012, Schwander *et al.* 2012, REX *et al.* 2014) and unpublished GF from JKI Geilweilerhof. The fragment length analysis was done on a 3130xl Genetic Analyzer (Applied Biosystems, Germany) and the corresponding Genemapper 4.0. software. To confirm the berry color locus alleles, 'Sémillon', 'Catawba' and 'Concord' were investigated by the in-house SSR marker GF02-55 for the berry color locus on chromosome 2 (AZUMA *et al.* 2011). The GF02-55 forward primer is AAAATTGAAGGACAGGAGGAGG, reverse primer is GCAAGGCTGGTCTACTCAGAAA.

Results and Discussion

Systematic SSR analysis of genotypes of the grapevine repository provided first evidence of a parent-child relation of 'Catawba' and 'Sémillon'. As a follow-up study a detailed genotyping included the putative parent of 'Catawba' ('Sémillon') and the presumed offspring 'Concord'. The 'Catawba' and 'Concord' accessions of JKI Geilweilerhof showed the identical genetic fingerprint to the USDA references, confirming their identity (LACOMBE *et al.* 2013, *VIVC Vitis* International Variety Catalogue). Tab. 1 shows 38 SSR markers as an example of in total 166 SSR markers, reasonably equally distributed throughout the genome which were applied in a mapping study. In larger parentage analysis studies was shown that 20 SSRs can be sufficient to confirm parent-child relationships (LACOMBE *et al.* 2013).

Table 1

Subset of SSR markers informative for the relationship of 'Sémillon', 'Catawba' and 'Concord', on the 19 grape chromosomes (Chr). Fragment lengths in [bp] are given for the two alleles of each cultivar. Consistent fragment lengths for 'Sémillon' and 'Catawba' are bold. Same fragment lengths for 'Catawba' and 'Concord' are in italic

Chr	SSR Marker	Sémillon alleles [bp]		Catawba alleles [bp]		Concord alleles [bp]	
1	UDV-035	148	162	<i>136</i>	162	132	<i>136</i>
	VMC9D3	199	205	205	217	<i>205</i>	221
2	GF02-11	276	276	276	281	<i>276</i>	283
	GF02-17	327	327	327	333	<i>327</i>	331
3	VMC2E7	156	160	154	160	154	<i>160</i>
	GF03-01	118	122	108	122	108	<i>122</i>
4	UDV-034	178	180	178	<i>202</i>	200	<i>202</i>
	GF04-16	316	316	286	316	280	<i>316</i>
5	VRZAG79	247	251	247	251	<i>247</i>	259
	GF05-11	304	304	<i>295</i>	304	<i>295</i>	295
6	GF06-16	257	265	<i>239</i>	265	<i>239</i>	241
	GF06-08	140	162	150	162	140	<i>162</i>
7	GF07-05	157	157	157	<i>163</i>	<i>163</i>	163
	GF07-14	209	221	<i>211</i>	221	209	<i>211</i>
8	VCHR08A	200	200	200	0	163	0
	UDV-026	127	133	127	0	157	0
9	GF09-14	356	356	356	<i>370</i>	<i>370</i>	370
	VMC6E4	141	141	<i>121</i>	141	<i>121</i>	121
10	VVIH01	244	250	<i>240</i>	244	<i>240</i>	250
	GF10-09	298	298	<i>284</i>	298	<i>284</i>	284
11	GF11-03	122	132	122	130	<i>122</i>	130
	UDV-028	140	146	140	150	<i>140</i>	146
12	GF12-05	167	171	<i>163</i>	171	<i>163</i>	169
	VMC4F3	164	170	164	<i>204</i>	168	<i>204</i>
13	VCHR13A	139	149	139	139	134	<i>139</i>
	GF13-08	348	358	348	<i>350</i>	<i>350</i>	358
14	GF14-02	221	227	221	<i>245</i>	221	<i>245</i>
	VMC6C10	110	138	122	138	126	<i>138</i>
15	GF15-06	166	178	<i>170</i>	178	<i>170</i>	170
	GF15-02	132	140	140	<i>144</i>	142	<i>144</i>
16	GF16-31	226	230	220	230	228	<i>230</i>
	GF16-25	320	320	315	320	312	<i>320</i>
17	VCHR17A	184	184	156	184	164	<i>184</i>
	GF17-07	109	109	<i>90</i>	109	<i>90</i>	<i>109</i>
18	SCU10	205	211	205	211	<i>205</i>	214
	VMC8F4.2	94	108	94	106	<i>94</i>	102
19	UDV-023	179	201	201	<i>227</i>	<i>223</i>	<i>227</i>
	GF19-10	153	155	155	<i>159</i>	<i>159</i>	159

137 markers proofed to be heterozygous for 'Catawba'. For these markers, 'Catawba' shared one allele with 'Sémillon' in any case. 'Concord' inherited always one 'Catawba' allele, either the 'Sémillon' allele or the allele of the wild ancestor confirming their parent-child relatedness (Tab. 1).

A consideration about the Mendelian heredity of the berry color supports our result on a different level: In grapes, the black berry color is dominant over red and the red color is dominant over white (BARRITT and EINSET 1969). The putative 'Catawba' ancestor 'Sémillon' is a white berry cultivar. Consequently it carries two recessive alleles for white color at the berry color locus. As 'Catawba' has red grapes, it could have either two alleles for red color or one white allele plus one dominant red allele. To support 'Sémillon' as 'Catawba's ancestor, the offspring must have inherited the white allele and thus being heterozygous at the berry color locus. The JKI Geilweilerhof in Siebeldingen maintains a population, which derived from a cross of 'Blaufraenkisch' and 'Catawba'. The black berried 'Blaufraenkisch' has the allele combinations black/white receiving the white allele from its white berried ancestor 'Heunisch Weiss' (LACOMBE *et al.* 2013). The 'Blaufraenkisch' x 'Catawba' population segregates concerning the berry color in the following way: in year 2014 out of 112 descendants, 59 were black, 8 red and 25 white. According to this, the ratio of the berry color (black : red : white) was approximately 2 : 1 : 1. As a conclusion, 'Catawba' must be heterozygous at the berry color locus carrying the allele combination red/white which is in accordance with the given 'Sémillon' descent. In case of 'Concord' having black colored berries, it could have the alleles black/white, black/red or black/black. The identified parent-child relationship between 'Catawba' and 'Concord' is also in agreement with the results that 'Concord' has inherited either its recessive red or its recessive white allele besides the black one. The SSR analysis indicated one black allele for 'Concord' and one white allele of 'Catawba', originated from 'Sémillon' (Tab. 2, Figure).

To deduce something more about the ancestry of 'Concord' regarding the wild type portion, the SSR marker information around the flower sex locus was used. This is, like the berry color locus, located on chromosome 2 (FECHTER *et al.* 2012), thus the heredity of the alleles for hermaphrodite, male and female flowers can be traced. 'Sémillon', 'Catawba' and 'Concord' exhibit hermaphrodite flower sex having one allele for hermaphroditism and one for female flowers. According to FECHTER *et al.* (2012), 'Sémillon' and 'Concord' differ in their female allele: 'Concord' has the "wild

female allele" (HFw) in contrast to 'Sémillon' having the allele typical for elite European cultivars (HFk). 'Catawba' was analyzed with the diagnostic InDel marker for the adenine-phosphoribosyl transferase (APT) and was found out also to have a recessive "wild female allele" (HFw) found in wild species. According to this, 'Catawba' inherited the female flower sex allele from the wild parent and the hermaphrodite allele from 'Sémillon' probably being more yield stable than female plants. 'Concord' inherited the 'Sémillon' part of chromosome 2 completely, as indicated in Tab. 1. This was verified by seven SSR markers equally distributed over chromosome 2 for all three cultivars (data not shown). According to this, 'Concord' inherited the hermaphrodite flower sex allele (H) from 'Sémillon' through 'Catawba' and the "wild Fw allele" came from the wild parent. Wild *Vitis* species are usually dioecious (TUKEY 1966) and the male allele is dominant over the female allele (FECHTER *et al.* 2012). If the other ancestor of 'Concord' would have been a pure wild species, it would show the female flower sex. Since 'Concord' is a product of coincidence, the wild parent is unknown. But a principal components analysis for an ancestry study from SAWLER *et al.* (2013) issued a wild portion of 49 % for 'Catawba' and 69 % in 'Concord'. Considering this

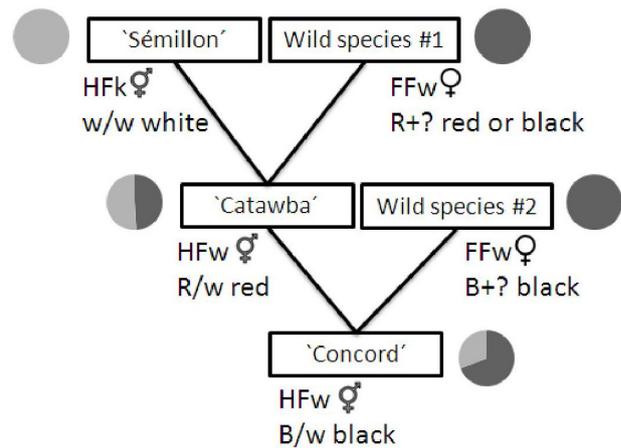


Figure: The Pedigree of 'Concord' could be retraced by using phenotypic data combined with genotypic data received from SSR markers (berry color locus) and an APT InDel marker (flower sex locus) as well as former genome-wide genetic analysis from SAWLER *et al.* (2013) concerning the wild portions (dark grey) and *V. vinifera* portions (light grey). B = black berry color allele, R = red berry color allele, w = white berry color allele. H = hermaphrodite allele, Fk = female allele, Fw = "wild female allele". ♀ = female flowers, ♂ = hermaphrodite flowers.

Table 2

Results of the analysis using the SSR marker GF02-55 with the product lengths of the cultivars 'Sémillon', 'Catawba' and 'Concord'. This marker is used for investigation of the berry color locus of grapes located on chromosome 2

Cultivar name	GF02-55 products	Allele # 1 [bp]	Allele # 2 [bp]	Genotype	Berry color
Sémillon		215	217	white/white	White
Catawba		168	215	red/white	Red
Concord		172	215	black/white	Black

information combined with our results, it can be deduced that the unknown ancestors were two different, pure wild species that inherited their "wild female alleles" respectively. The summary of the results are depicted in the Figure.

Besides the validation of the genetic background, the marker information reveals genomic regions with wild species content in the 'Catawba' offspring. The accordingly informative SSR markers can be used for background selection in future backcross breeding programs as proposed by HERZOG *et al.* (2013).

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References

- AMBROSI, H.; 2011: Farbatlas Rebsorten. Stuttgart, Germany, Ulmer.
- AZUMA, A.; UDO, Y.; SATO, A.; MITANI, N.; KONO, A.; BAN, Y.; YAKUSHIJI, H.; KOSHITA, Y.; KOBAYASHI, S.; 2011: Haplotype composition at the color locus is a major genetic determinant of skin color variation in *Vitis × labruscana* grapes. *Theor. Appl. Genet.* **122**, 1427-1438.
- BARRITT, B. H.; EINSET, J.; 1969: The inheritance of three major fruit colors in grape. *J. Am. Soc. Hortic. Sci.* **94**, 87-89.
- BOWERS, J. E.; DANGL, G. S.; MEREDITH, C. P.; 1999: Development and characterization of additional microsatellite DNA markers for grape. *Am. J. Enol. Vitic.* **50**, 243-246.
- CIPRIANI, G.; MARRAZZO, M. T.; DI GASPERO, G.; PFEIFFER, A.; MORGANTE, M.; TESTOLIN, R.; 2008: A set of microsatellite markers with long core repeat optimized for grape (*Vitis* spp.) genotyping. *BMC Plant Biol.* **8**, 1-13.
- DI GASPERO, G.; CIPRIANI, G.; MARRAZZO, M. T.; ANDREETTA, D.; PRADO CASTRO, M. J.; PETERLUNGER, E.; TESTOLIN, R.; 2005: Isolation of (AC)n-microsatellites in *Vitis vinifera* L. and analysis of genetic background in grapevines under marker assisted selection. *Mol. Breed.* **15**, 11-20.
- FECHTER, I.; HAUSMANN, L.; DAUM, M.; SØRENSEN, T. R.; VIEHÖVER, P.; WEISSHAAR, B.; TÖPFER, R.; 2012: Candidate genes within a 143 kb region of the flower sex locus in *Vitis*. *Mol. Genet. Genom.* **287**, 247-259.
- GALET, P.; 2000: Dictionnaire Encyclopédique des Cépages. Hachette, Paris, France.
- GERBER, S.; CHABRIER, P.; KREMER, A.; 2003: FaMoz: a software for parentage analysis using dominant, codominant and uniparentally inherited markers. *Mol. Ecol. Notes* **3**, 479-481.
- HEDRICK, U. P.; 1908: The Grapes of New York. Albany, NY.
- HERZOG, E.; TÖPFER, R.; HAUSMANN, L.; EIBACH, R.; FRISCH, M.; 2013: Selection strategies for marker-assisted background selection with chromosome-wise SSR multiplexes in pseudo-backcross programs for grapevine breeding. *Vitis* **52**, 193-196.
- LACOMBE, T.; BOURSQUOT, J. M.; LAUCOU, V.; DI VECCHI-STARAZ, M.; PEROS, J. P.; THIS, P.; 2013: Large-scale parentage analysis in an extended set of grapevine cultivars (*Vitis vinifera* L.). *Theor. Appl. Genet.* **126**, 401-414.
- MERDINOGLU, D.; BUTTERLIN, G.; BEVILACQUA, L.; CHIQUET, V.; ADAM-BLONDON, A. F.; DECROOÇQ, S.; 2005: Development and characterization of a large set of microsatellite markers in grapevine (*Vitis vinifera* L.) suitable for multiplex PCR. *Mol. Breed.* **15**, 349-366.
- MUNSON, T. V.; (1909). Foundations of American Grape Culture. Orange Judd Comp., New York, USA
- NELSON, R. R.; ACREE, T. E.; LEE, C. Y.; BUTTS, R. M.; 1977: Methyl anthranilate as an aroma constituent of American wine. *J. Food Sci.* **42**, 57-59.
- PINNEY, T.; 2007: A History of Wine in America from the Beginnings to Prohibition. University of California Press, Berkeley, USA.
- PRINCE, W.; 1830: Treatise on the Vine - Embracing its History from the Earliest Ages to the Present Day with Descriptions of Above Two Hundred Foreign, and Eighty American Varieties. J. Dobson Publ., New York, USA.
- RAPP, A.; KNIPSER, W.; ENGEL, L.; ULLEMMEYER, H.; HEIMANN, W.; 1980: Fremdkomponenten im Aroma von Trauben und Weinen interspezifischer Rebsorten. I. Die Erdbeernote. *Vitis* **19**, 13-23.
- RAPP, A.; VERSINI, G.; ULLEMMEYER, H.; 1993: 2-Aminoacetophenon: Verursachende Komponente der untypischen Alterungsnote (Naphthalin-ton,Hybridton) bei Wein. *Vitis* **32**, 61-62.
- REX, F.; FECHTER, I.; HAUSMANN, L.; TÖPFER, R.; 2014: QTL mapping of black rot (*Guignardia bidwellii*) resistance in the grapevine rootstock 'Börner' (*V. riparia* Gm183 × *V. cinerea* Arnold). *Theor. Appl. Genet.* **127**, 1667-1677.
- ROBINSON, J.; HARDING, J.; VOUILLAMOZ, J.; 2012: Wine grapes. A Complete Guide to 1.368 Vine Varieties, Including Their Origins and Flavours. Allen Lane, Penguin Books Ltd., London, UK.
- SAWLER, J.; REISCH, B.; ARADHYA, M. K.; PRINS, B.; ZHONG, G. Y.; SCHWANINGER, H.; SIMON, C.; BUCKLER, E.; MYLES, S.; 2013: Genomics assisted ancestry deconvolution in grape. *PLoS ONE* **8**, e80791.
- SCHWANDER, F.; EIBACH, R.; FECHTER, I.; HAUSMANN, L.; ZYPRIAN, E.; TÖPFER, R.; 2012: Rpv10: a new locus from the Asian *Vitis* gene pool for pyramiding downy mildew resistance loci in grapevine. *Theor. Appl. Genet.* **124**, 163-176.
- SEFC, K. M.; REGNER, F.; TURETSCHKE, E.; GLÖSSL, J.; STEINKELLNER, H.; 1999: Identification of microsatellite sequences in *Vitis riparia* and their applicability for genotyping of different *Vitis* species. *Genome* **42**, 367-373.
- TUKEY, H. B.; 1966: The story of the Concord grape. *J. Fruit Var. Hortic. Digest.* **20**, 54-55.
- VIVC; 2015: *Vitis* International Variety Catalogue (<http://www.vivc.de/>).
- ZHANG, J.; HAUSMANN, L.; EIBACH, R.; WELTER, L. J.; TÖPFER, R.; ZYPRIAN, E. M.; 2009: A framework map from grapevine V3125 (*Vitis vinifera* 'Schiava grossa' × 'Riesling') × rootstock cultivar 'Börner' (*Vitis riparia* × *Vitis cinerea*) to localize genetic determinants of phylloxera root resistance. *Theor. Appl. Genet.* **119**, 1039-1051.

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