

Identification by SNP Analysis of a Major Role for Cayetana Blanca in the Genetic Network of Iberian Peninsula Grapevine Varieties

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Abstract: Cayetana Blanca is a grapevine variety widely distributed in the Iberian Peninsula (Spain and Portugal). Its numerous synonyms and its presence in northern Africa point to a long history of cultivation. The goal here was to identify the genetic relationships of Cayetana Blanca with other Iberian and Mediterranean cultivars using a panel of SNP markers. A total of 243 single nucleotide polymorphisms (SNPs) and four chloroplast microsatellite loci allowed for identifying synonyms and trio (parents and offspring) and duo (parent-offspring) relationships. Six trios including Cayetana Blanca as a parent were found and supported by very high LODs. Five of the trios had the variety Alfocheiro Preto as the other parent and gave rise to cultivars used in Portugal: Cornifesto, Camarate, Mouratón, Malvasia Preta, and Periquita. The Spanish cultivar Jaén Tinto is confirmed to be an offspring of Cayetana Blanca and Listán Prieto. Additionally, synonyms and duos were detected that, in some cases, were not anticipated. The parents of Cayetana Blanca remain unknown, while the geographical origin of Cayetana Blanca could be a border region between Portugal and Spain. Results show that this cultivar has had a relevant impact in the viticulture of that area. As in other European regions, viticulture in the Iberian Peninsula is also marked by the impact of certain varieties that have worked as progenitors of many present cultivars. The results also show the interconnection between Portuguese and Spanish viticulture. Finally, the study demonstrates that SNP is a powerful tool for parentage inference in grapevine.

Key words: genetic relationships, historical genetics, parentage analysis, single nucleotide polymorphism, *Vitis vinifera*

Cayetana Blanca is the prime name of a well-known variety, cultivated in the Iberian Peninsula and the Maghreb region. In the *Vitis* International Variety Catalogue (VIVC, www.vivc.de) the cultivar appears with 65 synonyms, including Jaén Blanco, the most extended name in Spain, where this variety

is traditionally considered to have a local origin. Jaén was first cited in the 16th century (Herrera 1513) and described as a grape cultivar with large and compact bunches, large berries, and tender skins. The cultivar was widespread, giving rise to great confusion because of many synonyms and homonyms, as recorded in 1807: “El primero que dé á conocer con exâctitud todos los vidueños que se llaman Jaenes en España hará á su patria un servicio real” [“The first person to clarify exactly all cultivars that are called Jaén in Spain will make their country a royal service”] (Rojas Clemente y Rubio 1807). Present synonyms in Spain include Baladí, Baladí Verdejo, Balay (in Cordoba province), Blanca Cayetana, Pardina (Badajoz), Cagazal (La Rioja), Calagraño (La Rioja), Cayetana (Cáceres), Cirial (Jaén), Jainas (La Rioja), Maizancho (Ciudad Real), and Robal (Zaragoza). The cultivated surface for Cayetana Blanca has diminished in Spain from 71,709 ha in 1990 to 45,580 ha in 2007 (Inventario del Potencial Vitícola Campaña 2008/2009. MARM). However, it is still ranked as the sixth most planted cultivar, primarily in the central and south-southwestern regions of the Iberian Peninsula (Extremadura, Andalucía, Castilla-La Mancha, and Madrid).

The existence of synonyms and homonyms for a given cultivar makes it difficult to determine its correct identity and parentage. DNA markers and morphological markers help to identify synonyms and homonyms and to determine the genetic relationships between cultivars (This et al. 2006). Microsatellite or SSR markers have been used in pedigree studies of grape cultivars. Parentage analysis using 30 microsatellites allowed the identification of Cabernet franc and

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Sauvignon blanc as the parents of Cabernet Sauvignon (Bowers and Meredith 1997). Based on 32 loci SSR, Bowers et al. (1999) demonstrated that crosses between Pinot and Gouais Blanc gave rise to Chardonnay and Gamay as well as many other French grapevine cultivars. More recently, 20 polymorphic nuclear and three chloroplast microsatellite loci were used to identify the kinship group of Merlot (Boursiquot et al. 2009). In table grapes, analyses based on 25 nuclear and five chloroplast SSR loci were used to determine the parents of 93 table-grape cultivars (Ibáñez et al. 2009, Vargas et al. 2009).

Cayetana Blanca has been identified as an offspring of a cross between Antão Vaz and Rabo de Ovelha using data obtained for 28 microsatellite loci (Lopes et al. 2006). Although they detected incompatibilities at three loci, the authors argued that the possible existence of null alleles would explain those incompatibilities. The full compatibility at 25 loci, added to the fact that the three varieties are cultivated in a close geographical region (Alentejo in Portugal and Badajoz in Spain), supported the reliability of the suggested cross.

In this study, single nucleotide polymorphisms (SNPs) have been used to establish first-degree genetic relationships among grapevine cultivars. Compared to conventional nuclear SSR, SNPs have several features appropriate for large-scale genetic studies: they are abundant in most genomes surveyed (Brumfield et al. 2003) and they have relatively low costs and the data are very easily scored, allowing a more feasible exchange of data between laboratories. SNPs have been used recently in grapevine for individual identification, integration of genetic and physical maps, and paternity inference (Cabezas et al. 2011, Lijavetzky et al. 2007) and in the study of genetic structure and domestication history of grape (Myles et al. 2011). The power of SNPs for parentage inference has been reported previously (Anderson and Garza 2006), but almost exclusively in animal studies.

Here we present new insights regarding the genetic relationships of Cayetana Blanca through the study of synonyms and several genetically related varieties from the Iberian Peninsula (Spain and Portugal). Altogether, the results identify this cultivar as a major node in the genetic network relating grapevine cultivars in the central region between the two countries.

Materials and Methods

Plant materials. All accessions studied that are Cayetana Blanca or were found to be closely genetically related to this variety are listed in Table 1. Portuguese and Spanish accessions were sampled from the collection of grapevine varieties of El Encín (IMIDRA, Madrid). The Djinani accession was sampled from the SODEA germplasm bank in Meknes, Morocco. Síria, Sarigo, and Tinta Gorda were obtained from the Coleção Ampelográfica Nacional (CAN) of Instituto Nacional de Investigação Agrária (INIA) in Dois Portos, Portugal. When possible, varieties have been named following the prime names proposed by the *Vitis* International Variety Catalogue (VIVC, www.vivc.de).

DNA analysis. DNA was isolated from young frozen leaves using the DNeasy Plant Mini Kit (Qiagen, Valencia, CA). Previously identified SNPs (Lijavetzky et al. 2007) were genotyped at the CEGEN (Spanish National Genotyping Centre; www.cegen.org) using SNplex (Applied Biosystem, Foster City, CA), an application of high-throughput genotyping using a set of universal preoptimized reagents. This system is plex-based and each plex contains 48 markers. Each variety was genotyped using seven plexes (336 SNPs minus four, which were repeated: 332 SNPs), as described in Cabezas et al. (2011). Twenty nuclear microsatellites and four chloroplast microsatellites were also analyzed in some samples through the use of multiplex PCR and run in an Applied Biosystems 310 Genetic Analyzer (Ibáñez et al. 2009).

Table 1 Grapevine accessions included in this study.

Accession	Country ^a	VIVC prime name (number) ^b	Synonyms and sports studied
Água Santa	P	Água Santa (123)	
Alfrocheiro	P	Alfrocheiro Preto (277)	Baboso Negro
Antão Vaz	P	Antao Vaz (493)	
Camarate	P	Camarate Tinto (2018)	
Castellana Blanca	S	Castellana Blanca (40016)	
Castillo de Arcos	S	Castillo de Arcos (2328)	
Cigüente	P,S	Dona Blanca (15673)	Valenciana, Síria
Cornifesto	P	Cornifesto (2846)	
Criolla	S	Mission (7873)	Listán Prieto, Mission
Garrido Macho	S	Garrido Macho (4471)	
Jaén	M,P,S	Cayetana Blanca (5648)	Djinani, Morisca, Jaén Rosado, Sarigo
Jaén Tinto	S	Jaen Negro (5652)	
Moreto do Dao	P	Malvasia Preta (15647)	
Mouraton	P,S	Mouraton (8082)	Tinta Gorda
Periquita	P	Periquita (9152)	
Plateado	S	Doradilla (3654)	Doradilla
Rabo de Ovelha	P	Rabo de Ovelha (16956)	
Rocía	S	Rocía (40057)	
Puerto Alto	S	Puerto Alto (9619)	

^aM: Morocco, P: Portugal, S: Spain.

^bPrime name and variety number, *Vitis* International Variety Catalogue, www.vivc.de.

Parentage analysis. On the basis of SNP profiles, cultivars were analyzed for possible parent-offspring groups using Cervus 3.0 software (Field Genetics, London, UK) (Kalinowski et al. 2007). Analysis was performed with 427 cultivars as candidate parents, from a study that included many Iberian and Maghreb cultivars. LOD scores were obtained taking the natural log (log to base e) of the overall likelihood ratios for the parents and offspring trios, as implemented in Cervus 3.0. The probability of paternity exclusion was estimated for all the loci combined. Whenever possible, chlorotypes were used to determine which one of the putative parents was the mother, based on maternal transmission of chloroplasts in grapevine (Arroyo-García et al. 2002, Strefeler et al. 1992).

Results and Discussion

In a wider study, 838 plants were genotyped, many of them from Spain and Portugal, for 332 SNPs (data not shown). After discarding failed SNPs and samples, nonpolymorphic SNPs, and redundant genotypes, we obtained 427 unique genotypes for 243 SNPs. The set of cultivars included several parents and offspring trios previously described using microsatellites (Bowers et al. 1999, Cabezas et al. 2003, Ibáñez et al. 2009, Vargas et al. 2009), which were used as control in the parentage analysis. These control pedigrees and the corresponding LOD scores, which ranged from 59 to 88 with an average and median of 71, are listed in Supplemental Table 1. Reliabilities of pedigrees based on microsatellites are typically based on cumulative likelihood ratios rather than LOD scores, but in some cases both are published, allowing for comparison of SSRs and SNPs. A recent publication showed eight pedigrees involving important French cultivars such as Merlot or Cot (Boursiquot et al. 2009); LOD scores based on 20 SSR ranged from 18 to 50 with an average of 37 and a median of 42, much lower than those obtained with 243 SNPs in the present study.

From all the detected first-degree relationships, we selected those which could be directly related with Cayetana Blanca: the possible trios (parents and offspring) where one of the varieties involved was Cayetana Blanca; the putative descents of these offspring cultivars; and all cultivars that share at least one allele per locus with Cayetana Blanca and, therefore, maintain a putative parent-offspring relationship (Table 1). Most are presently cultivated either in Spain or Portugal or in both countries. In total, they account for ~50,000 ha in Spain

and 40,000 ha in Portugal, or 5% and 16% of total country-wide vineyard surface, respectively. Among these, Cayetana Blanca (45,580 ha) is the most cultivated variety in Spain and Periquita (20,500 ha) and Cigüente (11,700 ha) are the most cultivated in Portugal.

Several Portuguese cultivars are siblings. A total of six trios including Cayetana Blanca as a parent were found (Table 2, Figure 1). Five of them have the same variety as the other parent, a well-known Portuguese variety, Alfrocheiro Preto, which gave place to five varieties presently cultivated in Portugal: Periquita, Cornifesto, Malvasia Preta, Mouraton, and Camarate (Figure 2). These cultivars have been described by Galet (2000) and have different names (synonyms) in different locations of Portugal and/or Spain (Supplemental Table 2).

Alfrocheiro Preto is a red grape, one of the most widely planted (220 ha) in the Dão region (Portugal), and also found in the vineyards of Alentejo, Ribatejo, and Bairrada. Periquita (its current Portuguese name is Castelão) is a red wine cultivar known in Terras do Sado and throughout southern Portugal. Wine produced from this grape is usually quite tannic and fruity. Cornifesto is a black grapevine cultivar planted in the Douro and Alto Trás-os-Montes regions in northern Portugal. Malvasia Preta is an autochthonous red winegrape from the Douro and Dão regions (Portugal) and is also known as Moreto do Dão. The cross originating this variety, under the

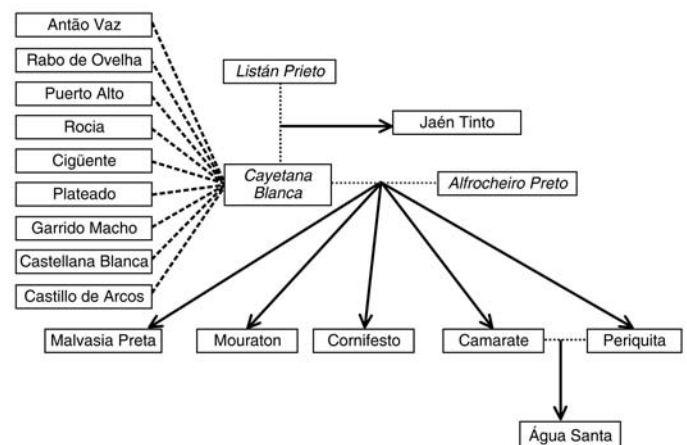


Figure 1 Genetic relationships found for Cayetana Blanca. Solid lines indicate full pedigree (trio parents and offspring), while dashed lines link cultivars sharing at least one allele per locus (possible parent-offspring relationship).

Table 2 Summary of the trios (parents and offspring) found in a parentage analysis of cultivars from Morocco and the Iberian Peninsula, where Cayetana Blanca or its descents were involved.

Offspring	H ^a	Parent 1	H ^a	Parent 2	H ^a	SNP compared (n)	LOD ^b
Jaén Tinto	D	Listán Prieto	D	Cayetana Blanca	A	220	80
Malvasia Preta	A	Cayetana Blanca	A	Alfrocheiro Preto	A	235	78
Mouraton	A	Cayetana Blanca	A	Alfrocheiro Preto	A	233	76.1
Cornifesto	A	Cayetana Blanca	A	Alfrocheiro Preto	A	232	62.9
Camarate	A	Cayetana Blanca	A	Alfrocheiro Preto	A	232	69.2
Periquita	A	Cayetana Blanca	A	Alfrocheiro Preto	A	234	75.3
Água Santa	A	Camarate	A	Periquita	A	232	81.3

^aH: Haplotype for parent and progeny.

^bLOD: LOD score of candidate parent, the most likely candidate parent is the one with the highest LOD score.

name of Mureto, has been described (Lacombe et al. 2007). This variety has a rose mutation. Mouraton is a black Spanish grapevine mainly cultivated in the border between Spain and northeastern Portugal (Arribes del Duero). Tinta Gorda is a Portuguese variety cultivated in Dão and Douro. In this study, a full match has been obtained between the SNP genotypes of this variety and Mouraton, which has allowed for discovering that these two varieties are synonyms. Camarate, a black cultivar widely cultivated in Portugal, was largely planted during the 18th century and presently covers 947 ha (Galet 2000). This variety is characterized by small to medium bunches and is very susceptible to oidium disease. Água Santa is a black winegrape cultivar authorized in the Beira Litoral, Beira Interior, Ribatejo, Madeira, and Açores regions in Portugal. This variety was obtained by Leão Ferreira de Almeida in 1948, after crossing Mortágua (syn. Camarate) and João Santarém (syn. Castelão) (Ghira et al. 1982). SNP data confirmed that this variety originated from a cross between Periquita (syn. Castelão) and Camarate, and thus is a “double grandchild” of Cayetana Blanca and Alfrocheiro Preto (Figure 1).

Jaén Tinto is a descent of Jaén Blanco and Listán Prieto. A cross between the cultivars Cayetana Blanca and Listán Prieto gave rise to Jaén Tinto (Table 2, Figure 1). This cross had been previously described (This et al. 2006) based on the genotypes for 20 microsatellite loci, with a very high LOD score of 61. Using SNP, this trio is supported with an even higher LOD score of 80. Jaén Tinto is an old Spanish cultivar (Valcarcel 1765, Rojas Clemente 1807, Abela 1885, García de los Salmones 1914) that is now cultivated in southern Spain (Galet 2000), with 1.145 ha in the Granada, Jaén, Almería, and Badajoz provinces (Figure 2).

There is another red variety called Jaen that originates from the Bierzo region, Castilla-León (Spain) (Martins et al. 1997). This Jaen is a homonym for Jaén Tinto, which corresponds to a synonym of Mencia, the typical red variety of that region.

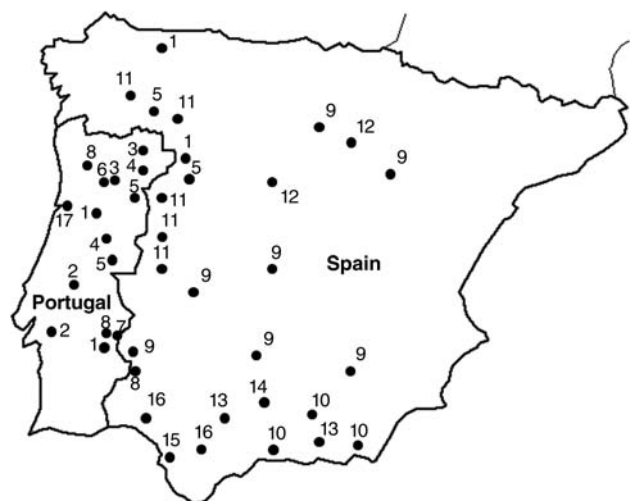


Figure 2 Map of the Iberian Peninsula showing the different locations of the offspring and other cultivars related to Cayetana Blanca: (1) Alfrocheiro Preto, (2) Periquita, (3) Cornifesto, (4) Malvasia Preta, (5) Mouraton, (6) Camarate, (7) Antão Vaz, (8) Rabo de Ovelha, (9) Cayetana Blanca, (10) Jaén Tinto, (11) Cigüente, (12) Castellana Blanca, (13) Plateado, (14) Puerto Alto, (15) Rocia, (16) Castillo de Arcos, (17) Água Santa.

The relationship between Cayetana Blanca and Jaén Tinto was also expected, based on several morphological traits. The shape of the mature leaves is highly similar: pentagonal, with rectilinear teeth, and often a tooth in the margin of the petiole sinus. The clusters of the two cultivars are similar only in shape and density: Jaén Tinto clusters are smaller than those of Cayetana Blanca. Morphological similarities between Jaén Tinto and Cayetana Blanca are exemplified by the fact that a well-known Spanish synonym for Cayetana Blanca is Jaén Blanco. Conversely, Listán Prieto and Jaén Tinto are morphologically quite dissimilar.

There are doubts about the origin of Listán Prieto, and the fact that it is the parent of Jaén Tinto could provide information. Listán Prieto is presently cultivated in several of the Canary Islands, with ~1700 ha in Tenerife, Lanzarote, and Gran Canaria, but its cultivation in the Iberian Peninsula almost disappeared after phylloxera in the 19th century. This variety is also cultivated in the Americas from California to Chile, where it is known under many names, including País, Criolla, and Mission (Milla-Tapia et al. 2007, This et al. 2006). Some bibliographic sources consider Listán Prieto as originally from the Americas (VIVC), with the suggestion that it could have been developed in South America from grapevine seeds introduced by Spanish missionaries in the 16th century (Galet 1957). Nevertheless, given that Listán Prieto is a parent of Jaén Tinto, a variety mentioned by Valcarcel (1765), indicates its existence as a cultivar earlier in the Iberian Peninsula. Thus, it would be very difficult to consider that Listán Prieto was developed in America from a seed, was multiplied and distributed there, and later brought to the Iberian Peninsula and crossed with Cayetana Blanca to produce Jaén Tinto. In addition, Herrera (1513) cited a variety called Palomino Negro that, according to his description, could possibly correspond to Listán Prieto.

Based on the maternal transmissions of chloroplasts in the *Vitis* genus (Arroyo-García et al. 2002, Strefeler et al. 1992), the analysis of the chloroplast microsatellite loci leads us to propose Listán Prieto as the mother and Cayetana Blanca as the father of Jaén Tinto. For the other crosses, the coincidence of the parental haplotypes precludes the identification of maternal and paternal parents (Table 2).

Cayetana Blanca is the putative parent of several other Iberian varieties. Cayetana Blanca is related to several Spanish varieties—Puerto Alto, Castellana Blanca, Cigüente, Plateado, Rocia, Garrido Macho, and Castillo de Arcos—sharing at least one allele per locus at the 243 SNPs studied (Figure 1). These results point to possible parent-offspring relationships between Cayetana Blanca and each of the mentioned varieties. In some cases this relationship is in agreement with some degree of morphological similarity between the related cultivars, while in others there are no conspicuous similarities.

Puerto Alto is a minor white winegrape cultivar grown in the Andalusian region (southern Spain) (García de los Salmones 1914). Castellana Blanca originated from the Rueda region (Spain) and is also known as Tolociriana and Temprana Agostea (Yuste et al. 2006). Cigüente is considered a

Spanish autochthonous cultivar and was first cited by Herrera (1513). This variety spread and is still cultivated along the Spanish-Portuguese border, following the Roman pathway known as *Vía de la Plata* (“silver way”) (Muñoz-Organero et al. 2006), where it is known by various names (Supplemental Table 2). In Portugal, its official name is *Siria*. *Plateado* is a white wine grapevine cultivated in Andalusia and first described by Candela (1971). Morphologically it is very similar to *Cayetana Blanca*. *Rocía* is a black wine cultivar from Spain also found in southern Spain (Martín et al. 2003). *Garrido Macho* is a white grapevine, long cultivated in Huelva (a southwestern Spanish region that borders Portugal), from where it spread to Cadiz and neighboring regions (Galet 2000). It is also known as *Garrido Macho*, *Parron Garrido*, and *Parron Garrido* (www.vivc.de). *Castillo de Arcos* is a white grape cultivated in Jerez, southwest Spain (www.vivc.de).

Origin and synonyms of Cayetana Blanca. *Alentejo-Badajoz*, a region in the southwest Iberian Peninsula, has been suggested as the probable place of origin of *Cayetana Blanca* (Lopes et al. 2006). In Morocco, the name for *Cayetana Blanca* is *Djinani* and is also considered a local variety. Unfortunately, there were no written records on viticulture in Morocco until the 1950s (Vidal 1951). Galet (2000) mentioned *Djinani* as a Moroccan variety considered by Truel as a synonym of *Jaén Blanco*. The accession used here was from the SODEA germplasm bank, a collection created in 1952 in Meknes by Vidal, who collected local vines from different regions, particularly from the Rif (northern Morocco). A comparison of the genetic profile of *Djinani* and *Jaén* using 20 SSR recently confirmed that they are synonyms (El Oualkadi et al. 2009). This study confirmed the identity of these synonyms and two color sports (rose) (Table 1) for the SNP markers used. Their local names and distribution are shown in Supplemental Table 2.

In Portugal, *Cayetana Blanca* is currently known as *Sari-go* and is cultivated in Douro. It is also known as *Mourisco Branco* (*Alentejo*), *Boal Carrasquenho*, and *Dona Branca* (*Estremadura*, Portugal).

The numerous Iberian cultivars identified here as progenies of *Cayetana Blanca* and the number of synonyms in Spain—*Calagraño*, *Baladí*, *Blanca Cayetana*, *Cirial*, *Baladí Verdejo*, *Jaén Empinadillo*, *Maizancho*, *Robal*, *Balay*, and *Cayetana* (Asensio 2000, Manso de Zúñiga 1905)—strongly suggest that *Cayetana Blanca* originated in the Iberian Peninsula, probably close to the present border between Portugal and Spain, where many relatives of *Cayetana Blanca* have originated and have been long cultivated (Figure 2). The probable exchange of grapevine cultivars between northern Africa and the Iberian Peninsula during centuries of close interaction would be responsible for its spread in Morocco.

The parents of Cayetana Blanca remain unknown. Twenty-eight SSR markers were used to provide evidence that *Cayetana Blanca* was the progeny of a cross between *Antão Vaz* and *Rabo de Ovelha* (Lopes et al. 2006). Although *Cayetana Blanca* displayed only one of the parental alleles at three loci, the authors posited the presence of null alleles which would still make the cross feasible. Geographical and histori-

cal reasons also supported the parentage hypothesis: *Antão Vaz* is the most important wine white cultivar cultivated in the *Alentejo* region, around *Virgueira*, Portugal (Almadanim et al. 2007, Lopes et al. 2006). *Rabo de Ovelha* is a white grapevine which at present is found throughout Portugal (Lopes et al. 2006), but it probably originated from the *Alentejo* region (Almadanim et al. 2007). *Cayetana Blanca* is mainly cultivated in the *Extremadura* region, close to the *Alentejo* region, but on the Spanish side of the Iberian Peninsula.

The analysis of 243 SNP loci in *Cayetana Blanca*, *Antão Vaz*, and *Rabo de Ovelha* allowed for discarding the suggested trio because there were incompatibilities at 10 SNPs (Supplemental Table 3). Mutation rates are in general considerably lower for SNPs than for microsatellites (Helyar et al. 2011), and thus it is expected to find fewer incompatibilities due to mutations in parentage analyses using SNP markers than microsatellite markers. In order to definitively corroborate or discard this putative parentage, we genotyped the three cultivars involved at 20 SSR loci. The trio was noncompatible at three SSR loci (Supplemental Table 4): *VMC1B11*, *VVIQ52*, and *VVMD32*. Furthermore, since *Cayetana Blanca* is heterozygous at these three mismatching loci, noncompatibilities cannot be caused by null alleles.

We also tested the possibility of pair-wise relationships of parent-offspring type in the three varieties. *Antão Vaz* and *Rabo de Ovelha* are not compatible at four SNPs. A similar result was previously found using 28 SSR markers where the two varieties share only 36% of alleles and then cannot have a parent-offspring relationship (Lopes et al. 2006). Nevertheless, *Antão Vaz* with *Cayetana Blanca* and *Rabo de Ovelha* with *Cayetana Blanca* share one allele per locus in all markers analyzed in this study. Thus, we conclude that *Antão Vaz* x *Rabo de Ovelha* is not the cross that gave rise to *Cayetana Blanca*; however, one of them could be a parent and the other an offspring, or perhaps both are offspring of *Cayetana Blanca*. This result stresses the difficulties of rejecting false trios when very closely related cultivars are involved, even using a high number of markers.

As described above, we found that *Cayetana Blanca* is related to other old and minor cultivars from Spain and Portugal, sharing one allele per locus at 243 SNPs (Figure 1, Figure 2). Any of them could be one of the parents of *Cayetana Blanca*, but, given the known history of the varieties included, we suggest that *Cigüente* or *Castellana Blanca* would be more probable.

Conclusions

This parentage analysis is one of the first in grapevine based on SNP marker data. SNPs will soon become the marker of choice for parentage analyses, as they are well suited to the high-throughput genotyping required for large studies, genotyping error rates are low, and LOD scores obtained are high, which provides strong support to the pedigrees found. This study sheds light on the genetic relationships among cultivars from Spain and Portugal related to the important cultivar *Cayetana Blanca*. The number of varieties directly related to *Cayetana Blanca* highlights its significance as a genetic node

in the network of pedigrees originating Iberian grapevine varieties. The full parentages of Jaén Tinto, Cornifesto, Camarate, Mouraton, Malvasia Preta, Periquita (Castelão), and Água Santa have been discovered or confirmed, as well as other nine varieties with putative parent-offspring relationship regarding Cayetana Blanca. Genetic and historical data point to the origin of Cayetana Blanca in the southwest Iberian Peninsula, while its parents remain unknown.

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