Scientia Horticulturae 219 (2017) 125–130

Contents lists available at ScienceDirect

Scientia Horticulturae

journal homepage: www.elsevier.com/locate/scihorti

# Pedigree reconstruction of wine and table grape crossbreeds created in Italy by Giovanni Dalmasso

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ARTICLE INFO

*Article history:*
Received 27 October 2016
Received in revised form 24 January 2017
Accepted 28 February 2017

*Keywords:*
Microsatellites
*Vitis vinifera* L.
Genotyping
Parentage analysis
SSR

ABSTRACT

Starting in 1931, Giovanni Dalmasso carried out intense grapevine breeding activity, creating more than 100 wine and table grape crosses. He sought to contribute to the economic development of Italian viticulture, but also left considerable genetic material for further breeding programs.

Owing to the current strong interest in the genetic improvement of grapevines, the pedigrees of varieties released by breeders must be determined. The aim of this study was to genetically characterize and verify the disclosed pedigrees of Dalmasso's crosses (IDs). Nuclear microsatellite profiles (n-SSR) of 42 ID accessions and 22 genotypes declared as parents were obtained at 22 loci. By cross-validation of allele size, declared parentages were verified. When one or both disclosed parents were found to be incorrect due to inconsistent genetic data, putative parent(s) were sought via SSR profile comparison within the grapevine molecular database of CNR-Institute for Sustainable Plant Protection, and the probability estimated via IDENTITY v. 4.0 software.

Through microsatellite analysis, three duplicated genotypes were discovered and twenty ID parentages out of 39 were confirmed. In 13 IDs, one parent was incorrect, in 2 IDs, both parents were inconsistent with microsatellite profiles, and in 4 IDs the pedigree could not be verified since the pollen donor was not available. Apart from invalidated crosses likely due to pollen contamination, 5 accessions were mislabelled, either when both parents were invalidated or when two specimens of the same offspring were differently labelled. Verification of breeder's declared parentages revealed 43% of invalidated pedigrees within the investigated wine and table cross-breeds. The results provide additional insight into grapevine available diversity, and may aid the development of further breeding programs.



## 1. Introduction

Vegetative propagation of *Vitis* spp. varieties has been used for centuries, preserving the characters of the founder plants. Except perhaps in the early stages of domestication, or in cases of seed transportation by migrating populations, reproduction from seed has tended to be avoided, because of the uncertain traits exhibited by descendants. The grapevine is a highly heterozygous cross-pollinating species (This et al., 2006). However, cross-breeding is very advantageous for developing varieties with favourable traits, especially when pollination control and offspring's strict selection are applied. As far as is known, the first successes in grapevine genetic improvement by deliberate crossing date to the early nineteenth century, when Louis Bouschet obtained the two flesh-coloured varieties 'Petit Bouschet' and 'Gros Bouschet' (later used to develop other flesh-coloured wine grape varieties) (Alabouvette, 1936), and when other breeders, such as Foster in the UK and Vibert in France, released improved table grape cultivars.

After these pioneering successes, intense breeding activity, starting from the mid-nineteenth century, was chiefly directed at obtaining hybrids (mainly from North American species and *Vitis vinifera* L.) resistant to fungal diseases and phylloxera. Apart from breeding for resistance, during the last 150 years many Italian geneticists (Bruni, Cosmo, Dalmasso, Manzoni, Pirovano, Prosperi, Rigotti, Terzi, and others) as well as those from other countries (Branas, Gargiulo, Mathiasz, Müller, Olmo, Thomson, Truel, Vidal, to name a few) concentrated their cross-breeding programs on *Vitis vinifera* L., aiming to obtain new table grapes with interest-

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http://dx.doi.org/10.1016/j.scienta.2017.02.044
0304-4238/

ing features, and new wine grape cultivars from which to produce appealing marketable wines.

Among the scientists mentioned, Giovanni Dalmasso started a cross-breeding program on wine and table grapes in 1931 at the Conegliano Viticulture Experimental Station (Veneto, North-Eastern Italy). The program was continued at the University of Turin (Piedmont, North-Western Italy). The program concerning table grapes aimed: a) to obtain improved genotypes from traditional varieties; b) to merge the grape quality characteristics from two or more genotypes (berry shape, color and flavour) with resilience and adaptability, c) to create new genotypes suited also to the cooler climate of Northern Italy. The wine grape program was chiefly concentrated on developing new crossbreeds combining positive technological features. According to Prof. Dalmasso's original notes, during twenty years of activity more than 100 wine and table grape crosses were selected, leaving a large body of grape diversity and considerable genetic material for further breeding.

Among these new cultivars, 14 wine grape varieties and 6 table grape genotypes are included in the Italian National Grapevine Variety Catalogue. Some of them, like the wine grape 'Albarossa' producing remarkable wines, currently enjoy the favour of growers and are greatly appreciated by the market. Others have never emerged from collections or experimental trials.

Among the most commonly used molecular markers for genetic analyses in grapevines, microsatellite alleles are inherited in Mendelian co-dominant segregation, confirming their suitability for genome mapping and to investigate hereditability and cultivar parentage (Thomas et al., 1994; Sefc et al., 2009). They can thus be used to confirm or invalidate breeder's information on the pedigree of a breed. Before the use of molecular markers, the putative parentage of new grape cultivars was retrieved from breeders' notes, which could be incomplete or inaccurate. Several studies have used microsatellite markers to clarify the parentage relationships between grape cultivars, either traditional or modern (Thomas et al., 1994; Cipriani et al., 2010; Lacombe et al., 2013). In grape-breeding programs, the female parent is certain, while the putative pollen donor might be mistaken because of contamination during pollination. This opens the possibility of a male parent not corresponding to the reported pedigree (Dettweiler et al., 2000; Ibàñez et al., 2009; Vargas et al., 2009; Lacombe et al., 2013). In some cases, due to mistaken varietal identity, the female parent may also be incorrect (Akkak et al., 2007). In connection with Dalmasso's crosses involving 'Nebbiolo' as parent, Torello Marinoni et al. (2009) discovered that, instead of the true 'Nebbiolo', Dalmasso accidentally used 'Nebbiolo di Dronero', a synonym for 'Chatus' that differs considerably from 'Nebbiolo'.

Because of the above errors, this study aimed to clarify the pedigrees of breeds obtained by Prof. Dalmasso that are still maintained in collections or commercially exploited. The secondary aim was to genetically characterize these materials, offering reference genetic fingerprints for the use of breeders, grape growers, and the wine industry. Due to the economic relevance of table grape breeding, there is worldwide interest in knowing varieties' correct pedigrees, because this information helps breeders to plan their cross-breeding programs.

As to the grape varieties discussed, many of them, especially the table grapes, are suitable for use in further breeding programs, because they exhibit improved traits in comparison to their parents (Carlomagno et al., 2014). At the same time, new varieties are more likely to be accepted and developed in the more dynamic table grape market. In recent years, much research activity is ongoing at the Department of Agriculture, Forest and Food Sciences (University of Turin, Italy), aimed to characterize the agronomic and qualitative behaviour of Dalmasso's table grape crosses in various environments, in the attempt to encourage their use at the local and national level.

Apart from their genetic value as a source of diversity, new wine grape cultivars could play a role in the more traditionalist wine market, through favourable features such as adaptability to climate change, or by offering an original, appealing flavour.

## 2. Materials and methods

### 2.1. Plant material

The SSR profiles of 42 Dalmasso interbreeds (IDs) and of 22 cultivars declared as parents were obtained. Samples from both IDs and declared parents were taken from field collections. In one case ('Pirovano 62') it was impossible to retrieve samples of the desired accession, because it was lacking in known collections.

The varieties (IDs) under investigation were described in terms of vine morphological features during the AGER Project, 2010–2104, "An Italian Vitis database with multidisciplinary approach, for exploitation and valorisation of the regional genotypes", employing the major OIV descriptors selected from the European *Vitis* Database (http://www.eu-vitis.de), and compared with published references.

Most of the plant material was taken from two collections maintained by the University of Turin, located at Chieri (Turin Province; planted in 1975) and at Alba (Cuneo Province; planted in 1984); 22 accessions were from the CNR-IPSP collection located at Grinzane Cavour (Cuneo Province; planted in 1992); two samples were kindly provided by CREA-VIT (Conegliano, Italy). After checking the uniformity of all the vines by morphology, for each accession one vine was chosen, from which to take a sample of young leaves, and the vine was labelled for further controls. Samples consisting of 3–4 young leaves were then stored at −80 °C until extraction; DNA extraction was done by the Thomas and Scott protocol (1993), slightly modified. Table 1 lists the 42 IDs analysed with their declared parents.

### 2.2. SSR analyses

Extracted DNA samples were amplified by Polymerase Chain Reaction (PCR) using primers labelled with four different fluorescent dyes (Fam, Ned, Pet, Hex). The PCR reactions were run in a BIO-RAD T100 thermal cycler, with the following thermal profile: one cycle at 95 °C for 3 min, followed by 28 cycles at 95 °C for 30 s each, 52 °C for 45 s, 72 °C for 90 s, and a final step of 30 min at 72 °C.

Amplification products were analysed on a 3130 Genetic Analyzer capillary sequencer (Applied Biosystems, Foster City, CA, USA). The internal GeneScan size standard 500LIZ (Applied Biosystems) was included in each run. Allele sizes in the output were determined using GeneMapper v. 4.0 software (Applied Biosystems), and alleles were designated by their size in base pairs (bp).

With regard to the markers used, at the first step, all IDs under study and their putative parents were analysed by the 9 SSR markers selected by the international scientific community for common use (This et al., 2004; Grapegen06: http://www1.montpellier.inra.fr/grapegen06/technical/index.html): VVS2 (Thomas and Scott, 1993), VvMD5, VvMD7 (Bowers et al., 1996), VvMD25, VvMD27, VvMD28, VvMD32 (Bowers et al., 1999), VrZag62 and VrZag79 (Sefc et al., 1999).

Declared parentages were verified by cross-validation of allele size. When one or both disclosed parents were found to be incorrect, due to inconsistent genetic data, the putative parent(s) were sought via SSR profile comparison within the grapevine molecular database of CNR-Institute for Sustainable Plant Protection (CNR-IPSP), which contains 850 unique genotypes of European cultivars (unpublished). At the second stage, all investigated IDs, their parents validated by the first 9 markers used, and the putative parents

**Table 1**
The 42 investigated cultivars issued from Dalmasso's crosses.

| Cultivar code | Cultivar name | Utilization | Berry color[a] | Parents according to breeder notations (variety names according to VIVC[b]) |
|---|---|---|---|---|
| **II-26** | **Vega** | Wine | B | Furmint × Malvasia Cosolo (Malvasia istriana) |
| **II-32** | **Fubiano** | Wine | B | Furmint × Trebbiano toscano |
| **III-3** | | Table | B | Moscato d'Amburgo (Muscat Hamburg) × Regina bianca (Afus Ali) |
| **III-32** | | Table | B | Moscato d'Amburgo (Muscat Hamburg) × Regina bianca (Afus Ali) |
| **III-34** | **Franca** | Table | N | Moscato d'Amburgo (Muscat Hamburg) × Regina bianca (Afus Ali) |
| **IV-6** | | Table | N | Chasselas rosa (Chasselas rose) × Perla di Csaba (Csaba Gyoengye) |
| **IV-28** | **Cornarea** | Wine | N | Barbera × Nebbiolo |
| **IV-31** | **Soperga** | Wine | N | Nebbiolo × Barbera |
| **V-1** | | Table | N | Chasselas rosa (Chasselas rose) × Perla di Csaba (Csaba Gyoengye) |
| **VI-3** | **Emilia** | Table | B | Bicane × Regina bianca (Afus ali) |
| **VI-6** | | Table | B | Bicane × Regina bianca (Afus ali) |
| **VI-9** | | Table | B | Bicane × Regina bianca (Afus ali) |
| **VI-12** | | Table | B | Bicane × Regina bianca (Afus ali) |
| **VI-24** | | Table | B | Bicane × Moscato di Terracina |
| **VII-21** | **S. Martino** | Wine | N | Nebbiolo × Dolcetto |
| **VIII-1** | | Table | B | Bicane × Moscato d'Amburgo (Muscat Hamburg) |
| **VIII-5** | **Bionda** | Table | B | Bicane × Moscato d'Amburgo (Muscat Hamburg) |
| **VIII-10** | | Table | N | Bicane × Moscato d'Amburgo (Muscat Hamburg) |
| **IX-2** | | Table | B | Schiavone (Schiava grossa) × Zibibbo (Muscat of Alexandria) |
| **X-4** | | Wine | B | Verdiso × Maddalena reale (Madeleine royale) |
| **X-10** | | Wine | B | Verdiso × Maddalena reale (Madeleine royale) |
| **X-12** | **Sirio** | Wine | B | Verdiso × Maddalena reale (Madeleine royale) |
| **XI-2** | | Table | B | Moscato d'Amburgo (Muscat Hamburg) × Regina bianca (Afus Ali) |
| **XI-5** | | Table | Rs | Moscato d'Amburgo (Muscat Hamburg) × Regina bianca (Afus Ali) |
| **XI-6** | | Table | B | Moscato d'Amburgo (Muscat Hamburg) × Regina bianca (Afus Ali) |
| **XI-20** | | Table | B | Moscato d'Amburgo (Muscat Hamburg) × Regina bianca (Afus Ali) |
| **XII-26** | | Wine | B | Furmint × Malvasia trevigiana (Malvasia bianca lunga) |
| **XII-30** | | Wine | B | Furmint × Trebbiano toscano |
| **XII-37** | **Bussanello** | Wine | B | Riesling italico (Welschriesling) × Furmint |
| **XII-40** | | Wine | B | Riesling italico (Welschriesling) × Riesling renano (Riesling weiss) |
| **XIII-11** | **Covè** | Wine | B | Hàrslevelü × Malvasia trevigiana (Malvasia bianca lunga) |
| **XIV-4** | | Wine | N | Grignolino × Sangiovese grosso (Sangiovese) |
| **XIV-15** | | Wine | N | Grignolino × Sangiovese grosso (Sangiovese) |
| **XV-29** | **Nebbiera** | Wine | N | Nebbiolo × Barbera |
| **XV-31** | **Albarossa** | Wine | N | Nebbiolo × Barbera |
| **XV-34** | **S. Michele** | Wine | N | Nebbiolo × Barbera |
| **XVI-8** | **Valentino** | Wine | Rg | Nebbiolo × Dolcetto |
| **XVII-25** | **Passau** | Wine | N | Dolcetto × Nebbiolo |
| **XVIII-3** | **Viola** | Table | N | Moscato d'Amburgo (Muscat Hamburg) × I.P. 62 (Pirovano 62) |
| **XVIII-12** | **Liana** | Table | N | Moscato d'Amburgo (Muscat Hamburg) × I.P. 62 (Pirovano 62) |
| **XVIII-21** | **Giovanna** | Table | N | Moscato d'Amburgo (Muscat Hamburg) × I.P. 62 (Pirovano 62) |
| **XVIII-24** | **Teresita** | Table | B | Moscato d'Amburgo (Muscat Hamburg) × I.P. 62 (Pirovano 62) |

[a] B = white, Rg = red, N = black, Rs = rose.
[b] *Vitis* International Variety Catalogue: http://www.vivc.de/index.php.

retrieved from the CNR-IPSP internal SSR database, were analysed by a further 13 microsatellite loci. These loci (belonging to various linkage groups, different from those of the previous 9 SSR) were: VvMD21, VvMD24, VvMD26 (Bowers et al., 1999); VrZag64 (Sefc et al., 1999); VVIb01, VVIn73, VVIp31, VVIq52 (Merdinoglu et al., 2005); VMC2h4, VMC7f2, VMC5g8, VMC1e8, and VMC3d12 (Vitis Microsatellite Consortium managed by Agrogene SA, Moissy Cramayel, France). All 19 grapevine chromosomes were covered by the 22 utilised markers. Parentage consistency was evaluated statistically on 290 genetic profiles obtained at 22 SSR loci from: a) the investigated cross-breeds (42 accessions), b) the declared and putative parents (21), and c) a further 227 *V. vinifera* cultivars. Parentage probability and the main genetic parameters were estimated via IDENTITY v. 4.0 software (developed by Wagner and Sefc, 1999). IDENTITY computes the likelihood ratios (both cumulative and with an upper confidence limit of 95%) on the allele frequencies of a proposed parentage versus other possibilities. PIC (Polymorphism Information Content) was calculated via POWER MARKER version 3.25 software (http://www.powermarker.net).

## 3. Results and discussion

The unique SSR profiles of the investigated IDs and their parents are reported in the Supplementary Material. Table 2 gives the

**Table 2**
Genetic parameters of a 290-sample set genotyped with 22 SSR *loci*.

| LOCUS | $N_a$ | $H_e$ | $H_o$ | PI | PIC |
|---|---|---|---|---|---|
| **VvMD5** | 9 | 0.822 | 0.855 | 0.054 | 0.800 |
| **VvMD7** | 13 | 0.798 | 0.821 | 0.066 | 0.773 |
| **VvMD25** | 11 | 0.766 | 0.772 | 0.093 | 0.727 |
| **VvMD27** | 8 | 0.799 | 0.769 | 0.070 | 0.770 |
| **VvMD28** | 15 | 0.876 | 0.907 | 0.028 | 0.864 |
| **VvMD32** | 13 | 0.797 | 0.841 | 0.065 | 0.774 |
| **VvS2** | 12 | 0.836 | 0.855 | 0.045 | 0.819 |
| **VrZAG62** | 10 | 0.845 | 0.890 | 0.043 | 0.826 |
| **VrZAG79** | 11 | 0.837 | 0.865 | 0.043 | 0.821 |
| **VvMD21** | 5 | 0.549 | 0.572 | 0.250 | 0.502 |
| **VvMD24** | 6 | 0.684 | 0.724 | 0.156 | 0.627 |
| **VvMD26** | 8 | 0.673 | 0.755 | 0.160 | 0.619 |
| **VrZAG64** | 15 | 0.839 | 0.879 | 0.045 | 0.820 |
| **VMC1e8** | 12 | 0.839 | 0.827 | 0.046 | 0.819 |
| **VMC2h4** | 16 | 0.787 | 0.759 | 0.068 | 0.764 |
| **VMC3d12** | 10 | 0.699 | 0.710 | 0.126 | 0.664 |
| **VMC5g8** | 8 | 0.710 | 0.762 | 0.127 | 0.667 |
| **VMC7f2** | 6 | 0.600 | 0.654 | 0.195 | 0.565 |
| **VVIb01** | 5 | 0.686 | 0.731 | 0.156 | 0.628 |
| **VVIn73** | 6 | 0.289 | 0.259 | 0.517 | 0.277 |
| **VVIp31** | 12 | 0.884 | 0.921 | 0.025 | 0.873 |
| **VVIq52** | 7 | 0.673 | 0.717 | 0.168 | 0.611 |
| **Cumulative** | | | | $3.23 \times 10^{-24}$ | 0.710 |

$N_a$ = number of alleles, $H_e$ = expected heterozygosity, $H_o$ = observed heterozygosity, PI = probability of identity, PIC = polymorphism information content.

main genetic parameters of a set of 290 *V. vinifera* cultivars including the genotypes investigated: number of alleles, expected (He) and observed (Ho) heterozygosity, probability of identity (PI), and polymorphism information content (PIC). The total number of alleles was 218, and ranged from 5 to 16 for each locus, with an average of 9.9; expected heterozygosity varied between 28.9% and 88.4%, while observed heterozygosity was between 25.9% and 92.1%. The probability of finding different genotypes with the same profile at 22 SSR loci appeared to be very low ($PI = 3.23 \times 10^{-24}$): therefore identical genotypes (i.e. synonyms) correspond to identical profiles. The high discriminating power among genotypes of the set of markers used is also supported by the high value of cumulative PIC (0.710).

Genetic analysis revealed that 'VI-12' and 'VIII-5' ('Bionda') are the same genotype, as are 'VII-21' ('S. Martino') and 'VIII-10', and 'XII-26' and 'XII-30'. This finding was confirmed by the morphological characterization. Because of these duplicates, the unique genotypes studied were 39. The three cases of mistaken labelling could have occurred either during material propagation or during planting/duplicating of collections, rather than in seedling selection. Unfortunately, the breeder's notes on the morphology of the selected offspring were too concise to afford true variety identification. The question arises about the correct labelling of these duplicates, i.e. whether, for example, the genotype labelled both 'VI-12' and 'VIII-5' actually corresponds to the true 'VI-12' or to the true 'VIII-5' ('Bionda'). This problem was solved by checking the consistency of declared parentages (Table 1) with genetic data analysis (SSR profiles in Supplementary Materials and Table 3). In the example of the contenders 'VI-12' and 'VIII-5', due to the high likelihood ratio in favour of the cross 'Bicane' x 'Moscato d'Amburgo', it can be postulated that the true cross refers to 'VIII-5', namely 'Bionda'. Based on the same reasoning, the genotype consistent with being the progeny of 'Chatus' and 'Dolcetto' is the ID 'VII-21', i.e. 'S. Martino', while the true descendent of 'Furmint' and 'Malvasia bianca lunga' is 'XII-26' (Table 3), ID 'XII-30' being inconsistent with this parentage (SSR profiles in Supplementary Materials).

For the other putative parentages, the likelihood ratios (LR) calculated for proposed parenthoods in relation to other possible combinations were all quite high, and > the minimum value of 8.39E + 1 for 'II-32' ('Fubiano') (Table 3). This indicates that the proposed parents of 'Fubiano' are about 80 times more likely than other alternative combinations.

The pedigrees of 20 crosses of 39 obtained by Prof. Dalmasso were validated (Table 3). In 13 IDs, one parent (female or male) was incorrect, while for two IDs both parents were inconsistent with microsatellite profiles. In four IDs the pedigree could not be verified for both parents because, as mentioned above, one of them, namely 'Pirovano 62', was missing. However, molecular data verified the likely parenthood of the other genitor, 'Muscat Hamburg'.

In all the wine grape varieties examined, 'Nebbiolo' was invalidated as parent, 'Chatus' being the true genotype, entering the cross either as male or female partner, as has been reported (Torello Marinoni et al., 2009). Since one of the many Italian synonyms for the French 'Chatus' is 'Nebbiolo' (popular around the village of Dronero, hence 'Nebbiolo di Dronero'), this cultivar must have been utilised in the crosses instead of the true 'Nebbiolo'. Cipriani et al. (2010) indicated 'Nebbiolo' as the authentic parent of the two IDs 'Cornarea' and 'Nebbiera'; however, this demonstrates that the accession of 'Nebbiolo' they analysed (maintained at the CREA-VIT of Conegliano collection and probably the same used by Dalmasso) was mistaken, clearly actually corresponding to 'Chatus'.

The proposed parentages (Table 1) were not confirmed for ID 'XIII-11' ('Cové') nor for 'XIV-4', and nor were other possible genitors (consistent with the descendent's SSR profiles) found in the CNR-IPSP grape genetic database. These two crosses are in fact the result of self-pollination of the plant chosen as female parent

**Table 3**
Declared and validated/invalidated parentages of the 39 unique Dalmasso's interbreeds of wine and table grapes. Likelihood ratios of the proposed families are compared to other possibilities. Probabilities are based on allelic frequencies of 22 *n-SSR loci* from each genotype and (in brackets) with a 95% upper confidence threshold. X and Y: random cultivar; rel(1): close relative of Parent (1); rel(2): close relative of Parent (2).

| Accession code | Cultivar name | Declared parents (names according to VIVC) | Parents resulting from genetic analyses | | | | | | Pedigree remarks |
|---|---|---|---|---|---|---|---|---|---|
| | | | Parent (1) | Parent (2) | X x Y | (1) x X | rel(2) x (1) | (2) x X | rel(1) x (2) | |
| **II-26** | **Vega** | Furmint × Malvasia istriana | Furmint | Malvasia istriana | 1.29E + 23 (2.29E + 19) | 1.23E + 13 (1.52E + 11) | 5.86E + 3 (2.94E + 3) | 4.91E + 11 (7.47E + 9) | 2.41E + 3 (1.09E + 3) | Validated |
| **II-32** | **Fubiano** | Furmint × Trebbiano toscano | Furmint | Trebbiano toscano | 3.88E + 12 (8.94E + 9) | 7.84E + 6 (5.11E + 5) | 2.36E + 2 (1.04E + 2) | 8.82E + 7 (3.09E + 6) | 1.88E + 2 (8.39E + 1) | Validated |
| **III-3** | | Muscat Hamburg × Afus Ali | Muscat Hamburg | Afus Ali | 2.75E + 16 (4.08E + 13) | 8.02E + 7 (5.03E + 6) | 6.25E + 2 (2.84E + 2) | 3.18E + 11 (7.12E + 9) | 2.93E + 3 (1.39E + 3) | Validated |
| **III-32** | | Muscat Hamburg × Afus Ali | Muscat Hamburg | Afus Ali | 2.46E + 15 (3.89E + 12) | 6.56E + 6 (4.37E + 5) | 4.16E + 2 (1.75E + 2) | 1.08E + 11 (2.46E + 9) | 1.50E + 3 (7.21E + 2) | Validated |
| **III-34** | **Franca** | Muscat Hamburg × Afus Ali | Muscat Hamburg | Afus Ali | 1.40E + 14 (4.83E + 11) | 8.80E + 6 (6.85E + 5) | 4.51E + 2 (2.02E + 2) | 5.61E + 9 (2.60E + 8) | 1.51E + 3 (7.37E + 2) | Validated |
| **IV-6** | | Chasselas rose × Csaba Gyoengye | Muscat Hamburg | Schiava grossa | 1.73E + 18 (2.51E + 15) | 6.40E + 8 (3.42E + 7) | 1.42E + 3 (6.55E + 2) | 8.92E + 9 (2.84E + 8) | 1.76E + 3 (7.83E + 2) | Both parents invalidated |
| **IV-28** | **Cornarea** | Barbera × Nebbiolo | Barbera | Chatus | 1.11E + 14 (2.39E + 11) | 9.09E + 7 (4.90E + 6) | 5.35E + 2 (2.29E + 2) | 2.46E + 9 (1.02E + 8) | 1.35E + 3 (6.08E + 2) | Male parent invalidated [a] |
| **IV-31** | **Soperga** | Nebbiolo × Barbera | Barbera | Chatus | 1.86E + 14 (3.76E + 11) | 8.17E + 7 (4.50E + 6) | 3.83E + 2 (1.74E + 2) | 4.94E + 9 (2.03E + 8) | 1.74E + 3 (7.89E + 2) | Female parent invalidated [a] |
| **V-1** | | Chasselas rose × Csaba Gyoengye | Schiava gentile | Muscat of Alexandria | 2.99E + 15 (4.86E + 12) | 1.36E + 11 (3.80E + 9) | 2.49E + 3 (1.17E + 3) | 1.74E + 8 (9.87E + 6) | 5.52E + 2 (2.63E + 2) | Both parents invalidated |

Table 3 (*Continued*)

| Accession code | Cultivar name | Declared parents (names according to VIVC) | Parents resulting from genetic analyses | | | | | | | Pedigree remarks |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | Parent (1) | Parent (2) | X x Y | (1) x X | rel(2) x (1) | (2) x X | rel(1) x (2) | |
| **VI-3** | **Emilia** | Bicane × Afus Ali | Bicane | Afus Ali | 6.88E+14 (1.46E+12) | 1.44E+7 (1.02E+6) | 3.33E+2 (1.50E+2) | 2.22E+10 (6.88E+8) | 1.68E+3 (7.96E+2) | Validated |
| **VI-6** | | Bicane × Afus Ali | Bicane | Afus Ali | 1.38E+16 (1.82E+13) | 3.24E+8 (1.33E+7) | 4.76E+2 (2.12E+2) | 2.08E+10 (6.20E+8) | 2.21E+3 (9.95E+2) | Validated |
| **VI-9** | | Bicane × Afus Ali | Bicane | Afus Ali | 6.64E+13 (1.67E+11) | 7.00E+6 (4.73E+5) | 3.63E+2 (1.53E+2) | 5.92E+9 (2.22E+8) | 1.27E+3 (6.08E+2) | Validated |
| **VI-24** | | Bicane × Moscato di Terracina | Bicane | Chasselas | 3.16E+14 (6.80E+11) | 2.26E+8 (1.08E+7) | 6.48E+2 (2.82E+2) | 2.03E+8 (8.78E+6) | 6.75E+2 (2.91E+2) | Male parent invalidated |
| **VII-21** | **S. Martino** | Nebbiolo × Dolcetto | Chatus | Dolcetto | 4.28E+16 (6.07E+13) | 2.07E+10 (8.71E+8) | 2.39E+3 (1.16E+3) | 4.15E+10 (1.44E+9) | 2.75E+3 (1.29E+3) | Female parent invalidated [a] |
| **VIII-1** | | Bicane × Muscat Hamburg | Bicane | Muscat Hamburg | 7.64E+16 (1.18E+14) | 1.82E+9 (7.82E+7) | 9.99E+2 (4.71E+2) | 3.64E+10 (1.31E+9) | 3.16E+3 (1.50E+3) | Validated |
| **VIII-5** | **Bionda** | Bicane × Muscat Hamburg | Bicane | Muscat Hamburg | 1.07E+17 (1.23E+14) | 4.02E+9 (1.42E+8) | 1.40E+3 (6.26E+2) | 1.02E+10 (3.49E+8) | 1.51E+3 (7.17E+2) | Validated |
| **IX-2** | | Schiava grossa × Muscat of Alexandria | Schiava gentile | Muscat of Alexandria | 1.30E+19 (5.64E+15) | 9.91E+13 (1.07E+12) | 7.98E+3 (3.97E+3) | 1.43E+9 (5.84E+7) | 8.97E+2 (4.27E+2) | Female parent invalidated |
| **X-4** | | Verdiso × Madeleine royale | Madeleine Royale | Verdiso | 6.58E+16 (8.41E+13) | 9.50E+10 (2.83E+9) | 3.34E+3 (1.55E+3) | 7.11E+8 (3.02E+7) | 1.17E+3 (5.03E+2) | Validated |
| **X-10** | | Verdiso × Madeleine royale | Madeleine Royale | Verdiso | 7.58E+14 (1.66E+12) | 5.63E+9 (1.91E+8) | 1.01E+3 (4.71E+2) | 8.85E+6 (6.82E+5) | 4.79E+2 (2.10E+2) | Validated |
| **X-12** | **Sirio** | Verdiso × Madeleine royale | Madeleine Royale | Verdiso | 2.32E+16 (3.05E+13) | 4.57E+8 (2.08E+7) | 6.09E+2 (2.74E+2) | 3.61E+10 (1.14E+9) | 2.81E+3 (1.30E+3) | Validated |
| **XI-2** | | Muscat Hamburg × Afus Ali | Muscat Hamburg | Afus Ali | 2.03E+16 (3.64E+13) | 3.29E+7 (2.03E+6) | 5.33E+2 (2.33E+2) | 1.20E+11 (3.54E+9) | 3.52E+3 (1.67E+3) | Validated |
| **XI-5** | | Muscat Hamburg × Afus Ali | Muscat Hamburg | Afus Ali | 1.04E+17 (1.32E+14) | 4.42E+7 (2.81E+6) | 5.13E+2 (2.30E+2) | 7.84E+12 (1.43E+11) | 6.84E+3 (3.41E+3) | Validated |
| **XI-6** | | Muscat Hamburg × Afus Ali | Muscat Hamburg | Afus Ali | 8.11E+17 (7.03E+14) | 3.06E+8 (1.28E+7) | 6.98E+2 (3.10E+2) | 1.10E+12 (2.21E+10) | 4.04E+3 (1.94E+3) | Validated |
| **XI-20** | | Muscat Hamburg × Afus Ali | Muscat Hamburg | Afus Ali | 2.87E+16 (4.89E+13) | 1.42E+8 (8.89E+6) | 9.32E+2 (4.23E+2) | 2.95E+11 (8.44E+9) | 4.34E+3 (2.07E+3) | Validated |
| **XII-26** | | Furmint × Malvasia bianca lunga | Furmint | Malvasia bianca lunga | 2.02E+24 (1.24E+20) | 7.41E+14 (4.61E+12) | 6.06E+3 (2.96E+3) | 1.52E+13 (1.42E+11) | 5.37E+3 (2.42E+3) | Validated |
| **XII-37** | **Bussanello** | Welschriesling × Furmint | Welschriesling | Furmint | 2.11E+20 (3.38E+16) | 1.13E+15 (6.61E+12) | 1.11E+4 (5.26E+3) | 1.51E+10 (3.73E+8) | 9.61E+2 (4.32E+2) | Validated |
| **XII-40** | | Welschriesling × Riesling weiss | Welschriesling | Riesling weiss | 1.75E+18 (9.41E+14) | 3.73E+7 (1.76E+6) | 5.33E+2 (2.12E+2) | 2.38E+11 (3.50E+9) | 2.20E+3 (1.04E+3) | Validated |
| **XIII-11** | **Covè** | Harslevelue × Malvasia bianca lunga | Harslevelue | Harslevelue | – [b] | – | – | – | – | Male parent invalidated (self-pollination) [a] |
| **XIV-4** | | Grignolino × Sangiovese | Grignolino | Grignolino | – | – | – | – | – | Male parent invalidated (self-pollination) [a] |
| **XIV-15** | | Grignolino × Sangiovese | Grignolino | Moradella | 2.12E+15 (2.95E+12) | 5.88E+8 (2.72E+7) | 6.35E+2 (2.86E+2) | 5.32E+10 (1.48E+9) | 3.18E+3 (1.42E+3) | Male parent invalidated |
| **XV-29** | **Nebbiera** | Nebbiolo × Barbera | Chatus | Barbera | 6.29E+15 (7.89E+12) | 1.22E+10 (4.02E+8) | 1.60E+3 (7.25E+2) | 1.01E+9 (4.02E+7) | 7.41E+2 (3.25E+2) | Female parent invalidated [a] |
| **XV-31** | **Albarossa** | Nebbiolo × Barbera | Chatus | Barbera | 2.35E+17 (1.83E+14) | 4.36E+8 (1.87E+7) | 7.41E+2 (3.19E+2) | 1.57E+12 (2.97E+10) | 4.94E+3 (2.34E+3) | Female parent invalidated [a] |
| **XV-34** | **S.Michele** | Nebbiolo × Barbera | Chatus | Barbera | 3.54E+12 (1.13E+10) | 1.56E+8 (7.79E+6) | 4.96E+2 (2.22E+2) | 1.24E+7 (7.99E+5) | 2.41E+2 (1.09E+2) | Female parent invalidated [a] |
| **XVI-8** | **Valentino** | Nebbiolo × Dolcetto | Chatus | Dolcetto | 5.46E+17 (4.49E+14) | 3.41E+9 (1.38E+8) | 1.29E+3 (5.85E+2) | 4.06E+12 (7.74E+10) | 6.19E+3 (2.97E+3) | Female parent invalidated [a] |
| **XVII-25** | **Passau** | Dolcetto × Nebbiolo | Dolcetto | Chatus | 2.31E+18 (1.31E+15) | 3.00E+13 (3.60E+11) | 7.94E+3 (3.71E+3) | 2.63E+9 (1.14E+8) | 1.23E+3 (5.59E+2) | Male parent invalidated [a] |
| **XVIII-3** | **Viola** | Muscat Hamburg × Pirovano 62 | Muscat Hamburg | – | | | | | | unverifiable |
| **XVIII-12** | **Liana** | Muscat Hamburg × Pirovano 62 | Muscat Hamburg | – | | | | | | unverifiable |
| **XVIII-21** | **Giovanna** | Muscat Hamburg × Pirovano 62 | Muscat Hamburg | – | | | | | | unverifiable |
| **XVIII-24** | **Teresita** | Muscat Hamburg × Pirovano 62 | Muscat Hamburg | – | | | | | | unverifiable |

[a] Confirmation of prior publication.

[b] Likelihood ratios of the proposed families for selfing descents are not calculated.

(as reported by Cipriani et al., 2010 for 'Cové'), likely because of a mistake in the emasculation process. In both crosses, but particularly in the second one, the plant's vigour was depressed, and leaf morphological anomalies arose at times.

In 'VI-24' and 'XIV-15', the pollen donor was found to be a different cultivar from that expected, likely because of pollen contamination. As in the case of 'Chatus' being mistaken for 'Nebbiolo', in one cross ('IX-2') the true parent was 'Schiava gentile' (alias 'Urban blau'), a less widespread cultivar than the declared genitor 'Schiava grossa', known as 'Trollinger' and 'Frankenthal' in other central European countries, and named 'Schiavone' in Dalmasso's notes.

For two table grapevines ('IV-6' and 'V-1'), both parents were invalidated: this might be due to the mistaken identity of both parents or, more likely, to a mistake in seedling labelling, or even in material propagation from mother plants during field collection establishment or duplication. The lack of a detailed morphological description from the breeder prevents true-to-type verification of seedlings.

As already mentioned, the descent of the four offspring of 'Pirovano 62′ could not being verified, since 'Pirovano 62' appears to have disappeared from European collections. In the large INRA collection at Domaine de Vassal (France), where 'Pirovano 62' should be maintained according to the *Vitis* International Variety Catalogue (www.vivc.de), the conserved accession probably corresponds instead to 'Pirovano 61' (a sibling of 'Pirovano 62'), as INRA scientists suspect (J.-M. Boursiquot, personal communication).

Verification of parentages declared by breeders, through the inheritance of SSR markers, revealed 43% of invalidated pedigrees in the investigated cross-breeds (15 out of 35 verified pedigrees). This percentage is high compared to those reported in other studies: Lacombe et al. (2013) analysed 381 modern cultivars, and found breeder's data to be invalid in 126 genotypes (33%). In the present study, the use of 'Chatus' instead of 'Nebbiolo' in a substantial number of IDs had a multiplier effect on the inaccuracy of breeder's data. Vargas et al. (2009) also found a similar percentage of incorrect pedigrees (42%, 13 incorrect pedigrees versus 18 correct, as reported in the literature).

## 4. Conclusions

Alongside invalidated crosses, likely due to pollen contamination (when the male parent was incorrect), several cases of cultivar mislabelling (either when both parents were invalidated or when the same genotype was labelled differently) were found in the varieties investigated. This indicates that further mislabelling during vegetative propagation to establish/duplicate collections may have been added to possible cross-breeding errors, making varietal identification essential for newly-bred varieties.

The economic exploitation of new breeds by grape farmers and the wine industry may be considerably affected by their correct identification and parentage. As to breeder's activity, in particular related to table grapes, the correct information provided on the genetic basis of the crosses obtained may contribute to the development of further breeding programs. Lastly, the parentage verification and genetic characterization of 39 grapevine cross-breeds obtained by Prof. Dalmasso has provided additional insight into the available grapevine diversity.

## Funding

This research was funded by the Agricultural Research Foundation (AGER, Italy) [grant number 2010-2104].

## Acknowledgments

The authors thank the Agricultural Research Foundation (AGER, Italy) for financial support and Giovanni Dalmasso Foundation for patronage. We are grateful to Massimo Gardiman of the Consiglio per la Ricerca in Agricoltura (CREA-VIT), Conegliano (Italy) for providing two samples from their collections. We gratefully acknowledge the assistance given by J-M. Boursiquot (INRA, Montpellier, and Domaine de Vassal, France) who provided useful information on 'Pirovano 61' and 'Pirovano 62'.

## Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.scienta.2017.02.044.

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