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The Muscats: A molecular analysis of synonyms, homonyms and genetic relationships within a large family of grapevine cultivars

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Summary

The Muscats are a large, wide-spread family of grapevines, having in common besides the name the typical Muscat flavour. A huge number of synonyms and homonyms makes it difficult to identify them. Sixty-four accessions were analysed in the present work; they are representatives of the huge variability of this family as far as morphological aspects, berry colour and size, time of ripening and aptitude for wine and/or table grape production are considered. An analysis was performed at two isozymes and 25 microsatellite loci. The 64 accessions were reduced to 20, which were easily distinguishable from each other at the molecular level by as few as two microsatellite loci. The remaining 44 were found to be synonyms. Three mutants with red and pink coloured berries were identified in the Moscato bianco group. Moscato nero encompasses at least two, Moscato rosa three different varieties. It seems that only two of the analysed Muscats are the main progenitors of the Muscat family: Moscato bianco and Muscat of Alexandria, which in turn are joined by a direct parent-offspring link. We were unable to discriminate biotypes belonging to the same cultivar by microsatellite markers.

K e y w o r d s : Muscat grapevines, synonyms, homonyms, discrimination, pedigree, microsatellites.

Introduction

The large family of Muscat vines is extensively spread all over the world; they have in common the characteristic Muscat flavour, from which their name is derived. The centre of origin has been assumed to be Greece, but DALMASSO *et al.* (1964) referred more prudently and generically to the Eastern Mediterranean basin. Almost all ampelographers agree that Muscat grapevines are known and appreciated since ancient times (SCIENZA *et al*. 1989). The Muscat germplasm has been used extensively by grape breeders; numerous crosses can be found in botanical gardens and ampelographic collections while others became popular either for winemaking, such as Muscat Ottonel, or for table grape production, *e.g.* Italia. Nowadays ampelography uses new tools to describe and identify varieties, among them are molecular markers. In recent years there has been an increasing development of microsatellite markers for grapevine. They were first isolated by THOMAS and SCOTT (1993), then by BOWERS *et al*. (1996) and by REGNER and coll. (pers. comm.). In 1999 the search for new microsatellites in grape was pushed forward by the "Vitis Microsatellite Consortium", established with the aim to isolate a large number of these markers in the frame of an international cooperation.

Microsatellite markers are very polymorphic; they give information at a single locus level and have a codominant Mendelian inheritance. Therefore they are very powerful for parentage analyses: THOMAS and SCOTT (1993) reported a pedigree analysis for Marroo Seedless, BOWERS and MEREDITH (1997) identified the parents of Cabernet Sauvignon and MEREDITH *et al*. (1999) those of Petit Sirah. SEFC *et al.* (1997 a) suggested a pedigree for 9 European varieties, REGNER *et al.* (2000) proposed a pedigree for Pinots.

In the present study a molecular analysis based on microsatellite markers was carried out for 64 Muscat accessions with different aims: firstly to clarify homonyms and synonyms; secondly, to investigate the genetic relationship among these accessions and to establish a hypothesis about their origin and finally to test the discrimination power of microsatellite markers among clones/biotypes belonging to the same variety. For this last goal 24 accessions of Moscato bianco collected from different sites were typed.

In addition the 64 Muscat accessions were analysed by two isoenzymatic systems: GPI (Glucose Phosphate Isomerase, EC 5.3.1.9) and PGM (Phosphogluco Mutase, EC 5.4.2.2) (CALÒ *et al.* 1989).

Material and Methods

P l a n t m a t e r i a l : Leaf samples were collected from 64 Muscat accessions (see Tab. 1) cultivated in the field collection of the Istituto Sperimentale per la Viticoltura of Conegliano. A few accessions were kindly supplied by the Istituto Agrario di S. Michele all'Adige (Trento, Italy) or were obtained from private vineyards. Leaf samples of 5 varieties chosen as outgroup were also collected to complete the sample set.

I s o z y m e a n a l y s i s : This analysis was performed with freshly collected leaves for GPI and PGM systems, following the method previously described by CRESPAN *et al.* (1998). The leaf extract was electrophoresed in a potato starch gel and stained by dipping the gel slices into solutions containing, besides other reagents, the appropriate substrate for isozyme reaction; the patterns were recorded as allele combinations, referring to the codes published by CALÒ *et al.* (1989).

M i c r o s a t e l l i t e D N A a n a l y s i s : The DNA was extracted according to the method described by CRESPAN *et al.* (1999). Twenty-five loci were analysed: VVS1 and VVS2

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Table 1

List of the 64 analysed accessions of Muscats, grouped by synonyms and in alphabetical order

The representative variety of a group is written in bold. At the bottom: the list of the outgroup varieties

DNA-No	Muscat accessions	Col*.	Source	Collection
42	**Moscadoul**	B	Dr. Bruni	ISV Susegana
54	**Moscatellone bianco**	W	Ist. Colt. Arb. Univ. Bari	ISV Susegana
49	**Moscatello nero**	B	Ist. Colt. Arb. Univ. Firenze	ISV Susegana
56	Moscatello nero	B	Crispiano (Taranto)	ISV Spresiano
79	Moscatello nero	B	Ist. Colt. Arb. Univ. Bologna	ISV Susegana
60	**Moscatello selvatico**	W	Az. Agr. Jatta, Ruvo di Puglia (Bari)	ISV Spresiano
46	Moscato selvatico	W	Ist. Colt. Arb. Univ. Bari	ISV Susegana
11	**Moscatel ruso**	W	Jerez de la Fronteira (Spain)	ISV Susegana
53	**Moscato**	B	Provincia di Padova	ISV Susegana
123	**Moscato bianco**	W	CNR Torino	
32	Moscatel fino	W	Jerez de la Fronteira (Spain)	ISV Susegana
120	Moscatello	W	Az. Ricci (Toscana)	Dr. Storchi
121	Moscatello	W	Az. Banfi (Toscana)	Dr. Storchi
122	Moscatello	W	Az. Poggione (Toscana)	Dr. Storchi
48	Moscatello bianco	W	Ist. Colt. Arb. Univ. Firenze	ISV Susegana
66	Moscatello bianco	W	Az. Bello, Crispiano (Taranto)	ISV Spresiano
50	Moscatello bianco	W	Ist. Colt. Arb. Univ. Bologna	ISV Susegana
64	Moscatello bianco di Basilicata	W	Az. Gaudiano, Potenza	ISV Spresiano
119	Moscato bianco	W	Toscana	
114	Moscato bianco	W	Parenzo (Croatia)	
65	Moscato bianco	W	Università degli studi di Firenze	ISV Spresiano
61	Moscato bianco	W	C.N.R. Centro Migl. Gen.Torino	ISV Spresiano
62	Moscato bianco di Basilicata	W	Az. Gaudiano, Potenza	ISV Spresiano
33	Moscato Chambave	W	Raccolta Di Rovasenda, Alba (Piemonte)	ISV Susegana
39	Moscato dei Colli Euganei	W	Conte Emo, Arquà (Padova)	ISV Susegana
115	Moscato di Momiano	W	Momiano (Croatia)	
16	Moscato di Montalcino	W	**	ISV Susegana
55	Moscato di Montalcino	W	Università degli studi di Firenze	ISV Spresiano
22	Moscato di Tempio	W	**	ISV Susegana
51	Moscato reale	W	Ist. Colt. Arb. Univ. Bari	ISV Susegana
38	Moscato rosa	Rs	Università di Belgrado	ISV Susegana
25	Moscato rosso	B	Raccolta Di Rovasenda Alba (Piemonte)	ISV Susegana
45	Muscat blanc à petites grains	W	E.N.S.A.M. Montpellier (France)	ISV Susegana
12	Muscat d'Alsace blanc	W	Colmar (France)	ISV Susegana
9	Muscat d'Alsace rouge	B	Colmar (France)	ISV Susegana
43	Muscat de Frontignan	W	Maclet Botton Villefranche (France)	ISV Susegana
24	**Moscato bianco casalese**	W	Prof. Zarattaro, Casale Monferrato (Piemonte)	ISV Susegana
63	**Moscato di Scanzo**	B	Ist. Colt. Arb. Univ. Milano	ISV Spresiano
19	**Moscato giallo**	W	Raccolta Di Rovasenda, Alba (Piemonte)	ISV Susegana
37	Moscato armeno	W	Prof. Musci (Bari)	ISV Susegana
20	Moscato fior d'arancio	W	Padova	ISV Susegana
128	Moscato giallo	W	Istituto Agrario di S.Michele all'Adige, Trento	
47	Moscato saraceno	W	Ist. Colt. Arb. Univ. Bari	ISV Susegana
21	Moscato sirio	W	Conte Emo, Arquà (Padova)	ISV Susegana
127	**Moscato fior d'arancio**	W	Istituto Agrario di S.Michele all'Adige, Trento	
23	Moscato bianco grosso	W	Università di Belgrado	ISV Susegana
29	Moscato Jesus	W	Prof. Musci (Bari)	ISV Susegana
116	**Moscato nero**	B	Breganze (Vicenza)	
27	Moscato nero	B	Raccolta Di Rovasenda, Alba (Piemonte)	ISV Susegana
75	**Moscato rosa**	Rs	Breganze (Vicenza)	
124	**Moscato rosa**	B	Istituto Agrario di S.Michele all'Adige, Trento	

Tab. 1 (continued)

DNA-No	Muscat accessions	Col*.	Source	Collection
26	**Moscato violetto**	B	Staz. Sperim. Agr. e For. S. Michele all'Adige, Trento	ISV Susegana
41	**Muscat of Alexandria**	W	**	ISV Susegana
35	Moscatel blanco	W	Jerez de la Fronteira (Spain)	ISV Susegana
34	Moscatel de Malaga	W	Jerez de la Fronteira (Spain)	ISV Susegana
67	Moscato francese	W	Vecchie varietà di Velletri (Lazio)	ISV Susegana
13	**Muscat of Hamburg**	B	ISV S.O.P. Bari	ISV Susegana
40	Moscatellone	B	**	ISV Susegana
14	Moscatel negro	B	Jerez de la Fronteira (Spain)	ISV Susegana
7	Muscat Madresfield Court	B	**	ISV Susegana
8	Muscat noir de H. Marsel	B	Jerez de la Fronteira (Spain)	ISV Susegana
10	**Muscat Ottonel**	W	Hungary	ISV Susegana
31	Muscat Ottonel	W	Trier (Germany)	ISV Susegana
44	**Thomuscat**	W	Davis (California)	ISV Susegana
Outgroup varieties				
129	Corvina veronese	B		ISV Susegana
80	Sangiovese	B		ISV Susegana
68	Pinot nero	B		ISV Susegana
18	Riesling renano	W		ISV Susegana
98	Sultanina	W		ISV Susegana

ISV = Istituto Sperimentale per la Viticoltura.
Col* = colour: W = white, B = black, Rs = pink.
** In some cases it was not possible to go back to the source of the material present in the collection for a very long time.

(Thomas and Scott 1993), VVS29 (Thomas, pers. comm.; CSIRO Plant Industry, Adelaide, Australia), VVMD5, VVMD7, VVMD8 (Bowers *et al.* 1996), VVMD14, VVMD17, VVMD21, VVMD24, VVMD25, VVMD26, VVMD28, VVMD31, VVMD32, VVMD34, VVMD36 (Bowers *et al.* 1999), VRZAG21, VRZAG47, VRZAG62, VRZAG64, VRZAG79 (Regner, pers. comm.), IVS2, ISV3 and ISV4 isolated in our institute which is a member of the VMC.

The PCR reaction mixture (25 µl final volume) contained: 10 ng total DNA, 0.5 U Taq DNA polymerase (Polymed, Firenze) and 1x relative buffer (Polymed, Firenze), 1.5 mM $MgCl_2$, 200 µM of each dNTP, 20 pmol of each primer. The PCR was performed in a PTC-100 thermal cycler (MJ Research, Massachussetts, USA) with a two-step protocol (Sefc *et al.* 1997 b): 5 min at 95 °C, 10 cycles of 50 °C for 15 s, 94 °C for 15 s, followed by 23 cycles of 50 °C for 15 s and 89 °C for 15 s, and a final step of 8 °C for at least 10 min to stop the reaction.

5 µl of the PCR product were tested on a 2 % agarose gel. On the basis of the signal intensity, 1-2 µl of amplified DNA was used for electrophoresis. Samples were denatured at 94 °C for 3 min in a buffer containing formamide and loaded onto a sequencing gel (5 % polyacrylamide, TBE 1 x, urea 7 M). Amplification products of cultivars carrying alleles of known molecular size were used as a reference for allele sizing. Bands on the gel were revealed by silver staining (Bassam *et al*. 1991, Tixier *et al.* 1997) immediately after run; the glass plate with the gel was dipped in different solutions as follows: 20 min in 10 % acetic acid; 4 rinses of 5 min each with distilled water; 30 min in 0.2 % $AgNO_3$ and 0.05 % formaldehyde; a brief rinse of 10 s with distilled water and finally 5-10 min in 3 % NaOH and 0.05 % formaldehyde, until bands appeared. The gels were manually scored at least twice and the images were recorded by a scanning apparatus (Epson GT-6500, Seiko Epson Corporation, Japan).

Data management: Two indices, δ_t and PD, were calculated to evaluate the allelic variability and the informative power of microsatellite loci: δ_t is a measure of H (expected heterozygosity), corrected on the sample size (Morgante *et al.* 1994), whereas PD estimates the discrimination power of loci as a function of genotype frequencies (Testolin *et al.* 2000). The formula to calculate δ_t is:

$$\delta_t = (1 - \Sigma p_i^2)\, N/(N-1)$$

where p_i is the frequence of the i^{th} allele, $1 - \Sigma p_i^2 = H$ and N the sample size. The formula to calculate PD is:

$$PD = 1 - \Sigma G_i^2$$

where G_i is the frequency of the i^{th} genotype.
A matrix of genetic similarity was generated by calculating the Dice coefficient (Dice 1945) for all pairs of cultivars. Then a similarity dendrogram was obtained by applying the UPGA (Unweighed Pair-Group Analysis) procedure, using the STATISTICA software (StatSoft Inc. 1993). All pairs of cultivars were finally compared with each other at all loci to find the parent-offspring putative relationships.

Results

As a result of the analysis carried out at 25 microsatellite and two isozyme loci, only 20 different molecular fingerprints were found, which are therefore referable only to 20 different Muscat varieties; they are listed in Tab. 2. The same profiles of these 20 varieties were found in the other 44 Muscat accessions at all examined loci: varieties with the same pat-

Table 2

Isozyme and molecular data of the 20 identified Muscat genotypes and of the 5 varieties chosen as outgroup, listed in alphabetical order (the alleles are in bp)

DNA-No	Muscat varieties	Col.	GPI*	PGM*	VVS1		VVS2		VVS29		VVMD5		VVMD7		VVMD8		VVMD14		VVMD17		VVMD21		VVMD24		VVMD25	
42	Moscadoul	B	ab	aa	190	181	143	135	179	175	236	232	247	237	157	-	232	222	222	220	256	235	214	210	259	245
54	Moscatellone bianco	W	ab	aa	190	181	133	133	171	171	246	232	249	239	157	141	232	228	222	221	266	249	219	210	245	243
79	Moscatello nero	B	bb	aa	181	181	135	133	171	171	228	226	249	239	143	141	241	227	221	220	266	249	214	210	267	253
60	Moscatello selvatico	W	ab	aa	181	181	151	149	171	171	232	232	253	249	141	-	241	235	221	220	266	249	214	210	267	253
11	Moscatel ruso	W	ab	aa	187	181	155	133	179	171	240	232	253	251	141	-	232	227	220	220	266	249	214	214	253	249
53	Moscato	B	ab	aa	181	181	135	133	171	171	236	228	249	245	143	141	241	241	222	212	256	249	214	214	259	245
123	Moscato bianco	W	ab	aa	181	181	133	133	171	171	236	228	249	233	141	-	241	222	222	220	266	249	219	214	253	245
24	Moscato bianco casalese	W	aa	aa	190	181	133	133	181	171	228	226	239	233	143	-	241	222	222	220	249	249	219	210	253	245
63	Moscato di Scanzo	B	aa	aa	183	181	135	133	171	171	240	236	249	239	143	141	222	222	222	221	266	249	219	218	259	245
19	Moscato giallo	W	aa	aa	187	181	143	133	171	171	240	228	249	239	143	141	259	241	222	212	266	256	219	214	259	245
127	Moscato fior d'arancio	W	aa	aa	190	181	133	133	179	171	236	228	249	247	143	141	227	222	222	212	266	249	214	210	253	245
116	Moscato nero	B	ab	aa	181	181	133	133	179	171	228	226	251	247	143	141	241	222	221	220	266	256	219	214	253	245
27	Moscato nero	B	aa	aa	181	180	143	135	179	171	240	236	253	233	147	141	241	241	222	220	266	249	219	210	259	259
75	Moscato rosa	Rs	ab	aa	183	180	133	133	181	179	232	228	249	239	143	-	232	227	222	220	266	266	214	214	253	245
124	Moscato rosa	B	ab	aa	181	181	135	133	171	171	240	236	249	239	141	-	m.d.		222	222	266	243	219	210	259	245
26	Moscato violetto	B	aa	aa	181	181	133	133	181	171	228	226	249	247	143	141	230	222	222	222	266	256	219	210	253	245
41	Muscat of Alexandria	W	aa	ad	181	180	149	133	179	171	232	228	251	249	141	-	241	232	220	220	266	256	214	214	253	253
13	Muscat of Hamburg	B	aa	ad	190	181	149	135	179	171	238	232	249	247	157	141	241	232	222	220	256	249	214	214	259	253
10	Muscat Ottonel	W	aa	aa	190	181	143	133	179	171	228	226	243	239	143	141	235	227	222	212	266	266	214	214	259	253
44	Thomuscat	W	ab	ad	188	181	151	149	171	171	234	232	253	249	145	-	238	232	222	220	266	256	219	214	253	243
Outgroup varieties																										
129	Corvina veronese	B	ad	ac	188	162	155	151	181	179	240	232	239	239	167	141	241	230	224	212	249	249	218	210	267	245
68	Pinot nero	B	bc	ab	190	183	151	137	179	171	238	228	243	239	143	141	241	222	220	212	249	249	218	216	253	243
18	Riesling renano	W	cc	ab	190	190	151	143	179	171	234	226	257	249	147	143	234	232	221	220	249	249	218	210	259	253
80	Sangiovese	B	ab	aa	181	181	133	133	171	171	236	226	263	239	147	143	235	232	221	21	249	243	216	210	245	245
98	Sultanina	W	ab	ad	188	181	151	145	179	171	234	234	253	239	157	145	238	228	222	222	256	249	219	210	253	243

*The letters correspond to the alleles described in CALÒ *et al.* (1989) as follows: a = 1, b = 3, c = 4, d = 5.
m.d. = missing data
Col.: see Tab. 1

Tab. 2 (continued)

DNA-No	VVMD26		VVMD28		VVMD31		VVMD32		VVMD34		VVMD36		VRZAG21		VRZAG47		VRZAG62		VRZAG64		VRZAG79		ISV2		ISV3		ISV4	
42	251	251	239	237	214	212	273	257	240	240	295	240	206	200	159	157	191	179	191	137	254	254	143	141	139	139	197	187
54	251	249	261	251	212	212	273	257	242	240	270	254	206	206	169	163	199	195	159	143	250	246	165	141	139	133	177	169
79	251	251	251	247	216	213	273	265	240	240	266	254	206	190	172	157	195	185	159	139	254	248	143	143	139	131	169	169
60	251	251	251	247	216	213	273	265	240	240	266	254	206	190	172	157	199	185	141	137	254	250	165	143	139	139	195	191
11	249	249	247	237	216	214	265	257	248	240	288	254	206	200	157	157	187	185	141	139	254	246	143	141	145	139	177	169
53	263	251	261	249	216	212	273	265	248	240	266	244	206	206	163	157	201	185	141	137	254	238	143	143	139	133	187	177
123	251	251	271	249	216	212	273	265	240	240	264	244	206	206	172	157	195	185	159	141	254	250	143	141	139	133	187	169
24	251	249	261	247	212	212	265	253	242	240	264	254	214	206	159	157	195	187	159	159	258	254	169	151	139	133	169	169
63	255	251	249	237	216	216	271	241	240	240	295	244	206	190	172	157	193	185	163	159	254	250	159	141	139	133	187	169
19	251	249	249	239	212	210	273	259	240	240	295	264	206	190	157	157	187	185	159	141	254	248	167	141	139	133	191	169
127	251	249	271	249	216	212	273	241	240	240	264	244	206	200	163	157	203	185	159	137	254	250	165	143	139	133	169	169
116	251	249	271	271	224	196	265	263	240	240	264	264	190	190	163	157	203	191	143	139	254	250	143	141	139	133	177	169
27	251	249	261	231	224	212	273	241	242	240	254	252	206	190	172	157	199	195	157	141	254	250	151	141	133	131	197	169
75	249	249	271	247	224	216	265	241	240	240	264	254	206	200	167	157	193	185	139	139	254	248	141	141	139	133	195	169
124	251	251	271	251	216	212	273	265	248	240	264	254	206	200	172	157	187	185	159	141	254	248	165	143	133	133	187	187
26	251	249	261	249	216	216	273	251	240	240	270	244	206	190	161	157	203	185	159	141	254	244	161	141	133	133	177	169
41	251	249	271	247	224	216	273	265	240	240	264	254	206	190	172	157	203	185	141	139	254	246	143	141	139	133	195	169
13	251	249	247	239	216	212	273	273	240	240	295	254	206	190	163	157	191	185	191	139	254	238	161	141	139	139	187	169
10	251	249	271	261	216	212	265	241	240	240	276	264	206	206	167	157	193	187	159	137	258	254	143	141	139	133	177	169
44	249	249	247	211	224	212	273	251	240	240	254	250	202	190	172	157	187	185	143	139	258	254	143	143	139	133	195	191
Outgroup varieties																												
129	251	251	261	261	216	212	273	241	248	240	276	254	206	190	167	157	193	187	143	137	250	250	165	151	139	133	187	169
68	255	249	239	221	216	216	273	241	240	240	254	254	206	200	167	163	193	187	163	139	244	238	165	151	145	133	177	169
18	251	251	237	231	214	204	273	253	240	240	264	254	206	202	167	159	203	193	159	137	244	242	151	143	145	139	197	169
80	249	249	247	237	212	212	257	253	240	240	264	264	204	202	163	157	195	193	139	137	258	242	165	143	139	139	197	177
98	251	249	247	221	212	212	251	251	248	240	268	250	202	190	172	159	187	187	159	143	258	246	143	143	139	133	193	191

terns were considered as synonyms and were grouped together in Tab. 1; the best known or the most ancient representative of that group is given in **bold**. Most of these groups are small, whereas the Moscato bianco group is very large, including 27 accessions.

Microsatellite markers showed a high polymorphism, the number of alleles found in the 20 unique genotypes of Muscats ranging from three at the locus VVMD34 to 11 at VVMD36, with an average of 6.58 (Tab. 2).

Both indices of genetic diversity, δ_t and PD, were high and followed the same trend, with an average of 0.719 and 0.793 respectively; VVMD14, VVMD28 and VVMD36 were the most discriminating loci (Fig. 1).

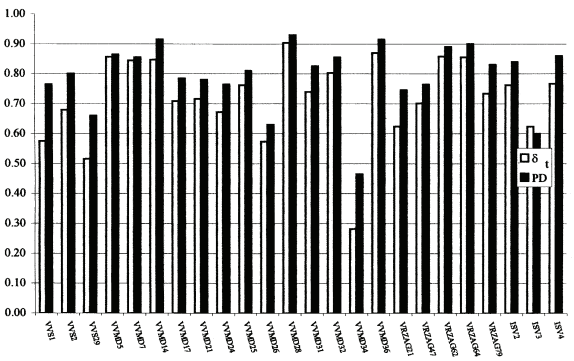

Fig. 1: δ_t and PD values calculated for each locus from the analysis of the 20 Muscat genotypes.

In many cases the polymorphism at a single locus was able to discriminate among most genotypes (Tab. 2): we frequently found unique profiles for single varieties due to the presence of an allele or a combination of alleles absent in all other varieties; *e.g.*, the locus VVMD36 gave 14 different patterns, VVMD28 gave 12 and VVMD14 gave 10. All 20 Muscat genotypes were easily separated by simultaneous comparison of their profiles at just two microsatellite loci (VVMD36 and VVS1).

The similarity tree, constructed by only one representative of each group, led to a further grouping (Fig. 2). Two main groups, which appear in the centre and at the right-hand side of the tree, showed the closest relationship among the accessions grouped at any distance: the most ancient

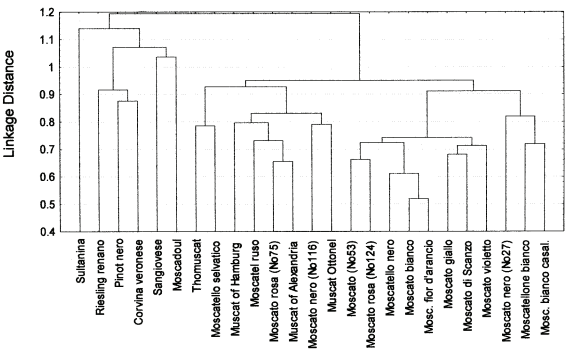

Fig. 2: Dendrogram produced by UPGA using the DICE's coefficient for the 20 Muscat genotypes and further 5 grape cultivars.

cultivar in each group was identified as Muscat of Alexandria in the first group and Moscato bianco in the second.

These groupings agree with the pedigree analysis, carried out by checking the compatibility as parent-offspring for each pair of accessions, independently of the parentage direction, which cannot be deduced by marker data (Fig. 3).

Discussion

The fingerprinting of 64 cultivars of the Muscat family allowed to identify several accessions showing identical banding pattern. These accessions were considered as synonyms and grouped together (Tab. 1). The main groups are discussed below.

Homonyms and synonyms in the Muscat family: The **Moscato bianco** is cited in literature as a very old variety known to have many different names representing small variants of the name Moscato (Muscat, Muskat, Moscatello, Muskateller) or which add the name of the region where the variety had been introduced to the basic name (*e.g.* d'Alsace, de Frontignan, di Montalcino, di Tempio). We checked 27 different accessions (Tab. 1) of which 18 were named differently; the remaining had their names in common with other accessions but originated from a different repository.

The group included also variants for berry colour, *e.g.* the Moscato rosa of the University of Belgrade, the Muscat d'Alsace rouge and the Moscato rosso. All ampelographers

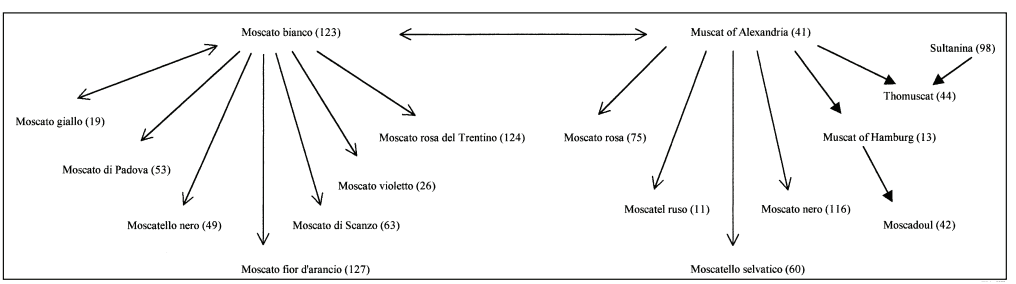

Fig. 3: Putative genetic relationships parent-offspring among 16 of the 20 identified Muscats. In brackets the accession numbers referred to Tab. 1. Double arrows: the descent direction is highly uncertain; single arrows: probable direction of crosses; thick arrows: safe direction.

agree that Moscato rosso resembles perfectly Moscato bianco; the intensity of colour may change due to the cultivation site, which explains names such as Muscat rose and Muscat gris (Molon 1906). This is not surprising, since mutation of skin colour is a rather common event in grape and in other fruit. A mutation of skin colour often occurs as a sport mutation and this makes it difficult to find differences in the marker profile between the new genotype and the original one, if a small amount of the genome is screened; moreover, a random intense genome screening, for example with AFLP markers, may bring about disappointing results (Sefc *et al*. 1998).

Muscat of Alexandria is also a very ancient cultivar with many different names. Molon (*l.c.*) reports about the history of some of them: *e.g.* the name Muscat of Alexandria seems to be relatively recent, since it was found for the first time in a Catalogue of Certosini frères in Paris, printed in 1713; the oldest wide-spread name of the variety is Zibibbo bianco, which reminds on the name of a cape at the African coast. It is likely that the name Muscat of Alexandria is derived from Alexandria in Egypt, where it was extensively grown.

Muscat of Alexandria has also mutants for berry colour, *e.g.* Muscat de Alexandria rouge, with red berries (not analysed here, see Galet 1964).

As far as the **Moscato giallo** group is concerned, Cosmo and Calò (1964) noted some analogies with Moscato sirio; our data confirm that the two varieties are synonyms. The group includes also Moscato armeno, Moscato saraceno and Moscato fior d'arancio grown in the province of Padova. The latter is genetically quite different from the true Moscato fior d'arancio grown in the area of Trento.

Two new varieties have entered the list of varieties related to **Muscat of Hamburg**: the Muscat noir de H. Marsel and the Muscat Madresfield Court. Thanks to this fortuitous comparison, it was possible to discover the origin of Muscat of Hamburg, since Muscat Madresfield Court is a known cross between Muscat of Alexandria and Black Morocco.

Moscato fior d'arancio was shown to be different from Moscato giallo, with which it has been confused sometimes; it includes two further accessions, Muscat Jesus and Moscato bianco grosso, confirming the observations of Di Rovasenda (1877). Also Viala and Vermorel (1901), report that Moscato fior d'arancio is a synonym of Muscat Jesus.

Finally a few cases of homonymy have to be described. The two accessions called Moscato nero showed a different marker profile: the Moscato nero from the Di Rovasenda's collection and the Moscato nero found in the province of Vicenza are different, each one showing a unique profile. The three accessions of Moscatello nero analysed here resulted, in another comparison, to be identical to three accessions of Aleatico nero at 12 microsatellite loci and for both GPI and PGM isozymes (data not shown). Effectively, the names Moscatello nero and Moscato nero are used in some Italian areas to indicate the Aleatico nero (Di Rovasenda *l.c.*).

The two Moscato fior d'arancio analysed in the present work are two different varieties: the one grown in the area of Padova (No 20) belongs to the Moscato giallo group of synonyms, whereas the second one, grown in the area of Trento, has been grouped together with Moscato Jesus and Moscato bianco grosso and is the true Moscato fior d'arancio cited by ampelographers.

We analysed three accessions of Moscato rosa: one (No 38) is a mutant of the Moscato bianco for the berry colour; another (No 124), cultivated in Trentino, is well known for its rose smelling and has black berries (Scienza *et al*. *l.c.*); the third (No 75) was found in the province of Vicenza. All three were found to be different varieties.

Genetic similarity and pedigree analysis: The data of the VVMD8 locus were not used in these comparisons, since the fact of null alleles has already been reported (Crespan *et al*. 1999).

Molecular data were elaborated in two ways: by drawing a dendrogram with Dice coefficients (Fig. 2) and by simply comparing the data locus by locus for pairs of varieties (Fig. 3).

For 16 of the unique Muscat genotypes it was possible to establish a direct parent-offspring link, since they shared at least one allele per locus, isoenzymatic alleles included (Fig. 3). This link is indicated by arrows in Fig. 3 (thick arrows: known or easily understandable cross directions; thin arrows: the direction may only be guessed; double points: no hypotheses). So far, two families were identified: one referred to Moscato bianco and the other to the Muscat of Alexandria. Moscato bianco is directly linked (parenthood relationship) to Moscato from Padova (No 53), Moscatello nero, Moscato fior d'arancio, Moscato di Scanzo, Moscato violetto, Moscato rosa of Trentino (No 124) and Moscato giallo; the latter was joined with a double pointed arrow, since it originates from the Middle East, the supposed origin of *Vitis vinifera* L. Muscat of Alexandria is linked to the Moscato rosa of Vicenza (No 75), Moscatel ruso, Moscatello selvatico, Moscato nero (No 116), Muscat of Hamburg and Thomuscat.

Moscato nero (No 27), Moscato bianco casalese (No 24), Moscatellone bianco (No 54) and Muscat Ottonel (No 10) showed no close parentage. Nevertheless, from Fig. 2 we may gather some information on a probable indirect parentage: in fact, these 4 varieties are part of a huge family of Muscats and are well separated from the outgroup varieties, which forms a lateral branch at the left side. Moreover, Moscato nero (No 27), Moscato bianco casalese (No 24) and Moscatellone bianco (No 54) resemble much more to Moscato bianco while Muscat Ottonel (No 10) belongs to the family of Muscat of Alexandria. An indirect descent from Moscato bianco and Muscat of Alexandria, respectively, through subsequent crosses may be assumed.

The Muscat lineage figured out in this paper, with Moscato bianco and Muscat of Alexandria as the main progenitors of the great Muscat family, is supported by historical evidence since both Moscato bianco and Moscato of Alexandria are described in ampelographic manuals as very ancient grapevine cultivars.

Interestingly, Moscato bianco and Muscat of Alexandria share at least one allele per locus, so that a direct parent-offspring relationship can be postulated for these two varieties. From our molecular data it is not possible to infer

the direction of the cross and therefore we can not establish which is older.

In addition, we can confirm reports in literature that the Californian Thomuscat, obtained in 1949 by Henderson (EYNARD *et al*. 1981) is a crossing of Muscat of Alexandria and Sultanina bianca.

The Moscadoul is the only Muscat grouped in the dendrogram of Fig. 2 with the outgroup varieties. This classification may be due to the fact that it is a hybrid of the Galimbert-Coulondre collection, obtained by crossing of 12375 and Muscat of Hamburg (GALET 1956). According to our molecular data Muscat of Hamburg has confirmed to be a parent.

None of the 20 primary Muscat varieties identified in this work derived from self-pollination of any other variety of the study.

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Erratum

In the paper

The Muscats: A molecular analysis of synonyms, homonyms and genetic relationships within a large family of grapevine cultivars

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Vitis **40** (1), 23-30 (2001)

some numbers of alleles were misprinted in Tab. 2.
Part of Tab. 2 is given below with corrected numbers in bold.

DNA-No/Muscat varieties	Col.	GPI*	PGM*	VVMD17	VVMD21	VVMD25	VVMD26	VVMD28	VVMD31	VVMD36	VRZAG21
42 Moscadoul	B	ab	aa	222 220	256 235	259 245	251 251	239 237	214 212	295 240	206 200
54 Moscatellone bianco	W	ab	aa	222 221	266 249	245 243	251 249	261 251	212 212	270 254	206 206
79 Moscatello nero	B	bb	aa	221 220	266 **243**	**259** 253	251 **249**	**249 239**	216 **212**	**254 244**	**206 206**
60 Moscatello selvatico	W	ab	aa	221 220	266 249	267 253	251 251	251 247	216 213	266 254	206 190
11 Moscatel ruso	W	ab	aa	220 220	266 249	253 249	249 249	247 237	216 214	288 254	206 200
53 Moscato	B	ab	aa	222 212	256 249	259 245	263 251	261 249	216 212	266 244	206 206
123 Moscato bianco	W	ab	aa	222 220	266 249	253 245	251 251	271 249	216 212	264 244	206 206
24 Moscato bianco casalese	W	aa	aa	222 220	249 249	253 245	251 249	261 247	212 212	264 254	214 206
63 Moscato di Scanzo	B	aa	aa	222 221	266 249	259 245	255 251	249 237	216 216	295 244	206 190
19 Moscato giallo	W	aa	aa	222 212	266 256	259 245	251 249	249 239	212 210	295 264	206 190
127 Moscato fior d'arancio	W	aa	aa	222 212	266 249	253 245	251 249	271 249	216 212	264 244	206 200
116 Moscato nero	B	ab	aa	221 220	266 256	253 245	251 249	271 271	224 196	264 264	190 190
27 Moscato nero	B	aa	aa	222 220	266 249	259 259	251 249	261 231	224 212	254 252	206 190
75 Moscato rosa	Rs	ab	aa	222 220	266 266	253 245	249 249	271 247	224 216	264 254	206 200
124 Moscato rosa	B	ab	aa	222 222	266 243	259 245	251 251	271 251	216 212	264 254	206 200
26 Moscato violetto	B	aa	aa	222 222	266 256	253 245	251 249	261 249	216 216	270 244	206 190
41 Muscat of Alexandria	W	aa	ad	220 220	266 256	253 253	251 249	271 247	224 216	264 254	206 190
13 Muscat of Hamburg	B	aa	ad	222 220	256 249	259 253	251 249	247 239	216 212	295 254	206 190
10 Muscat Ottonel	W	aa	aa	222 212	266 266	259 253	251 249	271 261	216 212	276 264	206 206
44 Thomuscat	W	ab	ad	222 220	266 256	253 243	249 249	247 **221**	224 212	254 250	202 190
Outgroup varieties											
129 Corvina veronese	B	ad	ac	224 212	249 249	267 245	251 251	261 261	216 212	276 254	206 190
68 Pinot nero	B	bc	ab	220 212	249 249	253 243	255 249	239 221	216 216	254 254	206 200
18 Riesling renano	W	cc	ab	221 220	249 249	259 253	251 251	237 231	214 204	264 254	206 202
80 Sangiovese	B	ab	aa	221 **212**	249 243	245 245	249 249	247 237	212 212	264 264	204 202
98 Sultanina	W	ab	ad	222 222	256 249	253 243	251 249	247 221	212 212	268 250	202 190