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Identification and characterization of grapevine genetic resources maintained in Eastern European Collections

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Summary

The Near East and the Caucasus regions are considered as gene and domestication centre for grapevine. In an earlier project "Conservation and Sustainable Use of Grapevine Genetic Resources in the Caucasus and Northern Black Sea Region" (2003-2007) it turned out that 2,654 accessions from autochthonous cultivars maintained by Armenia, Azerbaijan, Georgia, Moldova, Russian Federation and Ukraine in ten grapevine collections may belong to 1,283 cultivars. But trueness to type assessment by morphology and genetic fingerprinting still needed to be done. In COST Action FA1003 a first step in that direction was initiated. The following countries participated: Albania, Armenia, Austria, Azerbaijan, Bulgaria, Croatia, Georgia, Hungary, Latvia, Moldova, Romania, Slovakia, Slovenia and Ukraine. Mainly *Vitis vinifera* accessions (1098 samples) and 76 *Vitis sylvestris* individuals were analyzed by nine SSR-markers (VVS2, VVMD5, VVMD7, VVMD25, VVMD27, VVMD28, VVMD32, VrZag62, VrZag79). Cultivar identity confirmation/rejection was attempted for 306 genotypes/cultivars by comparison of the generated genetic profiles with international SSR-marker databases and ampelographic studies. The outcome proved unambiguously the necessity of morphologic description and photos (a) for comparison with bibliography, (b) for a clear and explicit definition of the cultivar and (c) the detection of sampling errors and misnomers. From the 1,098 analyzed accessions, 997 turned out to be indigenous to the participating countries. The remaining 101 accessions were Western European cultivars. The 997 fingerprints of indigenous accessions resulted in 658 unique profiles/cultivars. From these 353 (54 %) are only maintained in the countries of origin and 300 (46 %) unique genotypes exist only once in the Eastern European collections. For these 300 genotypes duplicate preservation needs to be initiated. In addition, the high ratio of non redundant genetic material of Eastern European origin suggests an immense unexplored diversity. Documentation of the entire information in the European *Vitis* Database will assist both

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germplasm maintenance and documentation of cultivar specific data.

Key words: biodiversity; grapevine; microsatellites; identification; documentation; germplasm preservation.

Introduction

The Near East and the Caucasus regions are considered as the origin of viticulture and the area of domestication. Already in the 1920's Negrul was the first in identifying Caucasus as the grapevine gene primary centre. His perception was based on the abundantly thriving wild wines and the enormous morphologic diversity he encountered (Alleweldt 1965). Being gene and domestication centre, grapevine genetic diversity is highly expected in that area. For investigation of that rich resource a survey of the grapevine germplasm present in Armenia, Azerbaijan, Georgia, Moldova, Russian Federation and Ukraine took place from 2003 to 2007 in the scope of the project "Conservation and Sustainable Use of Grapevine Genetic Resources in the Caucasus and Northern Black Sea Region" (Maghradze *et al.* 2009). It was funded by the government of Luxembourg and managed by Bioversity. Within the five project years an inventory was established encompassing the accessions of ten grapevine collections (Töpfer *et al.* 2009). Synonymous cultivar designations were registered under one common prime name. Prime names were assigned with a *Vitis* International Variety Catalogue (*V*IVC) variety number, a prerequisite for their uploading into *V*IVC. In addition, each accession obtained an accession number, a prerequisite for their uploading into a second database, the European *Vitis* Database. The outcome of 5 years of intensive collaboration was the preliminary conclusion that the maintained 2,654 accessions may belong to 1,283 cultivars (Töpfer *et al.* 2009). But trueness to type assessment by morphology and genetic fingerprinting still needed to be done to validate this compilation. Numerous studies had already shown that synonymy, homonymy and misnaming produced misleading results as described e.g. by Schneider *et al.* (2001), Karataş *et al.* (2007), Storchi *et al.* (2011) and Castro *et al.* (2012). These findings corroborated a previous investigation, detecting up to 10 % misnomers within grapevine collections (Dettweiler 1991). To solve such questions of cultivar identity, molecular markers proved to be highly effective for grapevines as demonstrated in almost 300 studies (http://www.vivc.de/searchBibliography/dbBibliography.php?retval=3600). Furthermore, an independent and complementary confirmation of results by ampelography remains good scientific practice. For that purpose cultivar references are needed in the form of morphological descriptions, drawings and photos. With respect to the present study an important step in that direction was made with the edition of the "Caucasus and Northern Black Sea Region Ampelography" initiated by Osvaldo Failla (Maghradze *et al.* 2012) comprising 267 autochthonous cultivars. This ampelography turned out to be a prerequisite for the objectives of COST Action FA1003 in terms of identification respectively distinction of Eastern European grape cultivars. The activities presented here and carried out by COST Action FA1003 / Working Group 1 aimed at: (a) creating a true to type inventory of germplasm existing in Eastern European collections by determining the accessions identity, (b) identifying endangered germplasm and thus initiating duplicate conservation and (c) documenting the accessions data in the European *Vitis* Database.

The following countries participated to achieve these goals: Albania, Armenia, Austria, Azerbaijan, Bulgaria, Croatia, Georgia, Hungary, Latvia, Moldova, Romania, Slovakia, Slovenia and Ukraine. They provided leaf material for SSR-marker analysis or allelic profiles from a total of 1,098 mainly *Vitis vinifera* accessions and 76 *Vitis sylvestris* individuals from Eastern European collections, respectively.

Material and Methods

For the present study participating collections were asked to provide material from cultivars which originated from their country. In total 21 collections from 14 countries were involved and 1,174 accessions were genotyped. The contribution per country in terms of accessions is given in Tab. 1. Institutes marked with an asterisk carried out DNA-analysis themselves according to the protocols indicated in Tab. 1. The remaining institutions sampled young leaves, placed them in labeled envelopes with folded blotting paper, added silica gel for drying and shipped the material to one of the following institutes for nuclear microsatellite analysis: INRA Montpellier (Laucou *et al.* 2011), CRA-VIT Conegliano (Migliaro *et al.* 2013), IASMA San Michele all'Adige (Basheer-Salimia *et al.* 2014) and JKI Geilweilerhof (Neuhaus *et al.* 2009). Nine SSR-markers (VVS2, VVMD5, VVMD7, VVMD25, VVMD27, VVMD28, VVMD32, VrZag62 and VrZag79) recommended by the European project GrapeGen06 were applied (Maul *et al.* 2012). SSR-marker data were coded for comparability of microsatellite profiles according to This *et al.* (2004). Respective SSR-marker descriptors are accessible via the European *Vitis* Database (http://www.eu-vitis.de/docs/descriptors/mcpd/OIV801_OIV806_5Juli2012.pdf). Coded fingerprints were collected, transferred into one data set and analyzed for matching allelic profiles by seven large SSR-marker databases: from Italy: CRA-VIT Conegliano, CNR Grugliasco and IASMA San Michele all'Adige; from Spain: IMIDRA Alcalá de Henares and ICVV Logroño; from France: INRA Montpellier and from Germany: JKI Geilweilerhof. The number of matching allele sizes were indicated by the SSR-marker databases. Mismatches at one or two loci were accepted to consider further studies. Probable identities were compiled. Examination of the findings took place by an expert group from CNR Grugliasco, INRA Montpellier and JKI Geilweilerhof. To confirm or reject the identities found the group consulted bibliographical references, herbarium material, photographs and further SSR-marker-data sources (http://www.vivc.de/searchBibliography/dbBibliography.php?retval=3600). Identity lists were prepared encompassing the Multi-Crop Passport Descriptors for grapevine (http://www.eu-vitis.de/docs/descrip-

Table 1

Summary of the analyzed accessions: Albania, Armenia, Austria, Azerbaijan, Bulgaria, Croatia, Georgia, Hungary, Latvia, Moldova, Romania, Slovakia, Slovenia and Ukraine

Country	Institute code	No. of accessions	No. of accessions unique or repeated 2 to 9 times									No. of unique profiles	Unique genotypes not repeated in Western Europe	
			unique	2	3	4	5	6	7	8	9		No.	%
1. Albania	ALB017	13	11	1								12	6	50
2. Armenia	ARM011	95	50	9	3	1		1		1		65	49	75
3. Austria	AUT024*	10	10									10	3	30
4. Azerbaijan	AZE007, AZE015	108	45	13	3	7						68	49	72
5. Bulgaria	BGR013*	189	173	8								181	63	35
6. Croatia	HRV014*	21	17	2								19	9	47
7. Georgia	GEO014, GEO015, GEO036, GEO037	370	129	34	22	5	5	3	5		1	204	106	52
8. Hungaria	HUN 08, HUN005*, HUN007, HUN045	47	41	3								44	28	64
9. Moldavia	MDA004	41	31	2	2							35	17	49
10. Romania	ROM 06/ROM045	57	47	3		1						51	3	6
11. Ukraine	UKR050	57	50	2	1							53	19	36
12. Slovenia	SVN018*	29	24	1	1							26	7	27
		1037	628									768	359	
13. Latvia (fungus resistant genotypes)		11	5	3								8		
14. Slovakia SVK 01* (mainly new crosses)		50	50									50		
		61												
Total:		1098												
Number of genotyped *Vitis vinifera* subsp. *sylvestris* accessions														
	ARM011	13												
	AZE015	55												
	GEO	8												
		76												

*Protocols for DNA-analysis are given in the articles behind the institute code: AUT024 (Regner *et al.* 2006); BGR013 (Dzhambazova *et al.* 2009); HRV014 (Zulj Mihaljevic *et al.* 2013); HUN005 (Galbacs *et al.* 2009); SVN018 (Stajner *et al.* 2014); SVK 01 (Dokupilová *et al.* 2014).

tors/mcpd/MCPD-for-Grapevine-10Feb12.pdf): accession name, accession number, remarks to the accession name, *V*IVC variety name, *V*IVC variety number and confirmation by bibliography. Estimation of the status of germplasm preservation in Eastern European countries was based on the following criteria: number of unique fingerprints in the country of origin of the cultivars and duplication of genotypes in the seven institutions carrying out allelic profile comparison and maintaining large grapevine germplasm repositories. The grapevine collection of the Department of Agricultural & Environmental Sciences, University of Milan, was also considered as it maintains 160 Georgian grapevine cultivars.

Results and Discussion

The search for matching profiles in the seven SSR-marker databases revealed that from the 1,098 genotyped and mainly *Vitis vinifera* accessions, 997 turned out to be indigenous to the participating countries (Tab. 2). The remaining 101 accessions were Western European cultivars (e.g. 'Luglienga bianca', 'Madeleine Angevine' and 'Pinot'), hybrids (e.g. 'Invulnerable' and 'Silva'), rootstocks (e.g. Rupestris du Lot and Selektion Oppenheim 4) and new crosses (e.g. 'Ametyst' and 'Neronet'). Only the 997 accessions representing basically traditional autochthonous cultivars were investigated in more detail. Further analyses of profiles revealed the existence of 659 unique profiles/cultivars (Tab. 2). With respect to cultivar recognition somatic mutations could only be considered if phenotypic information was available like for 'Rkatsiteli' with white berries and 'Rkatsiteli Vardisperi' with red berries.

Determination of identity: The definition of the identity of an accession requires very careful consideration and should combine the information and knowledge of two independent methods: molecular data and the morphologic description, including photos and herbaria.

Table 2

Summary of the results for the analyzed Eastern Europe grapevine accessions, indicating the number of genotypes present in collections of Eastern Europe and Western Europe

No. of repetitions	No. of accessions	No. of unique profiles	Genotypes unique in Eastern Europe	Genotypes repeated in Western Europe			
				Total	In one collection	In two collections	In more than two collections
unique	499	499	300	199	116	51	32
2x	168	84	40	44	14	18	12
3x	120	40	8	32	5	13	14
4x	56	14	2	12	2	4	6
5x	25	5	1	4	0	1	3
6x	18	3	1	2	0	1	1
7x	56	8	1	7	0	4	3
8x	16	2	0	2	0	0	2
9x	27	3	0	3	1	0	2
12x	12	1*	0	1	0	0	1
Total:	997	659	353	306	138	92	76
			54 %	46 %	21 %	14 %	11 %

*Sultanina.

Due to various reasons, in COST Action FA1003 the morphology of only a handful of the 997 genotyped accessions was described. First of all trueness to type could not be confirmed for the 300 unique accessions/cultivars lacking description and not duplicated elsewhere. However confirmation of identity was attempted for the 306 genotypes matching with cultivars maintained in Western European collections, namely the collections maintaining SSR-marker databases and Department of Agricultural & Environmental Sciences, University of Milan. Cultivar determination was also attempted for some of the 53 cultivars which were duplicated in Eastern European collections only (Tab. 3, example 6). Confirmation of identity was done combining reference material from Western European collections (like descriptions of morphology, herbarium material and photos), ampelographies, other cultivar describing documents and further SSR-marker data sources. Mainly three cases could be differentiated, for which some examples are given in Tab. 3:

- Identical or very similar accession/cultivar names and identical fingerprints: in this case trueness to type was clear, provided the genotyped material derived from distinct sources.
- Identical or very similar accession/cultivar names and different fingerprints (*i.e.* homonymy): in this case it had to be determined (a) which of the fingerprints corresponded to the true to type genotype, (b) which of the names were true homonyms, and (c) which were misnomers (Tab. 3, example 1).
- Different accession/cultivar names and identical fingerprints (i.e. synonymy): in this case the true designation needed to be figured out and synonyms respectively misnomers needed to be identified. Sampling errors or mutations were possible as well, but could not be verified without information of morphologic characteristics of the accessions (Tab. 3, examples 2 to 6).

For the time being a preliminary list displaying the findings from the 306 accessions/cultivars duplicated in Western Europe and the 53 accessions/cultivars duplicated in Eastern Europe is in preparation. Thereafter a comprehensive assessment of trueness to type is envisaged requiring the detailed knowledge of the curators in charge. A prerequisite are descriptions of the most important characteristics of the accessions and photos of shoot tip, mature leaf and bunch. These data together with the genetic fingerprint should be uploaded into the European *Vitis* Database, where they will serve as reference, useful for further identification work in the Eastern European collections. In that context the importance of the accession number needs to be stressed for traceability to the original plants and reliability of accession/cultivar specific information. In this context the accession number is a mandatory key within the European *Vitis* Database. It needs to be unique and never reused. Only Multi-Crop Passport Descriptor data encompassing accession numbers are accepted for data import. The complete accession specific information (descriptor data, photographs, SSR-marker data, virus status, etc.) is linked to that number. Besides trueness to type of accessions, management and preservation of genetic resources rely on the appropriate use of accession numbers.

As a result it can be concluded that for the first time Eastern European germplasm was systematically investigated on such a large scale with contribution of collections and molecular laboratories from 18 countries. The benefit of supporting material like ampelographies became evident as well as the voluntary agreement of researchers to use the nine GrapeGen06 SSR-markers and to publish the accessions respectively the cultivars profiles. The bibliographical data of the used articles are accessible via http://www.vivc.de/searchBibliography/dbBibliography.php?retval=3600. In particular the "Caucasus and Northern Black Sea Region Ampelography" (Maghradze *et al.* 2012) was of great

Table 3

Some examples of misnomers, synonymies and homonymies among analyzed grapevine accessions

Accession name	Accession number	Prime name	*I*VC variety number	Remark to accession name	Bibliography	VVMD5:1	VVMD5:2	VVMD25:1	VVMD25:2	VVS2:1	VVS2:2	VVMD7:1	VVMD7:2	VVMD27:1	VVMD27:2	VVMD28:1	VVMD28:2	VVMD32:1	VVMD32:2	VrZAG62:1	VrZAG62:2	VrZAG79:1	VrZAG79:2
Example 1: Misnomers. Identical accession names and distinct fingerprints: Morphology of Karmir Kakhani given by Maghradze *et al*. (2012) matched with accession's FRA139-2278Mtp1 morphological features. In addition FRA139-2278Mtp1-genetic fingerprint corresponded to the one published by Vouillamoz *et al*. (2006) for the same cultivar. Hence both Armenian Karmir Kakhani accessions were considered as misnomers.																						
Karmir Kakhani	FRA139-2278Mtp1	Karmir Kakhani	6000	true name	Maghradze *et al*. 2012, p. 56	242	242	249	267	135	151	243	253	180	195	238	244	250	272				
Karmir Kakhani	ARM011-IV-13	Karmir Kakhani 2 (non identified)	not yet registered	misnomer		230	236	245	249	153	157	243	247	186	195	246	258	250	250	196	196	249	249
Karmir Kakhani	ARM	Karmir Kakhani 3 (non identified)	not yet registered	misnomer		240	248	249	250	135	141	245	247	180	195	236	248	256	272	188	200	245	245
Example 2: Misnomers**.** Distinct accession names (distinct morphology) and matching fingerprints: Images of Shavkapito (Maghradze *et al*. 2012, p. 225) and Shavtkhila (Kartavčenko *et al*. 1954- 1966) were not the same, thus they could not be considered as synonyms. Because the cultivar occurred in two Italian collections under the name Shavtkhila , this designation was chosen as the correct cultivar name. Mchvartala was considered as misnomer because a distinct most likely true to type fingerprint for Mchvartala exists.. Another Shavkapito from GEO015 showed the same allelic profile like Adreuli Vardisperi from GEO036.																						
Shaftkhila	GEO036- III - 05	Shaftkhila	11723	true name	Kartavčenko *et al*. 1954-1966, vol. 3, p. 500	236	236	239	255	135	141	239	247	180	194	236	258	250	272	190	200	243	249
Shavkapito	GEO036- III - 08	Shaftkhila	11723	misnomer																			
Mchvartala	GEO036- I - 13	Shaftkhila	11723	misnomer																			
Sciavtchila	ITA362-3034	Shaftkhila	11724	alternative spelling																			
Shaftkhila	ITA035-125	Shaftkhila	11725	true name																			
Example 3: Unsolved trueness to type. Distinct accession names and matching fingerprints: An image of Kumsi Tetri (Maghradze *et al*. 2012) could not be compared with Tsnoris Tetri as in the Russian Ampelography (Kartavčenko *et al*. 1963-1970) no photo was given. Thus synonymous relationship between the two accessions could not be settled.																						
Kumsi Tetri	GEO036- V - 09a	Kumsi Tetri or Tsnoris Tetra	6556 or 12706	misnomer or true name?	Maghradze *et al*. 2012, p. 203 (Kumsi Tetri) or Kartavčenko *et al*. 1963-1970, vol. 3, p. 394 (Tsnoris Tetri - no photo given)	236	240	241	267	137	141	247	253	180	186	244	258	262	272	190	200	249	251
Tsnoris Tetra	GEO036- I - 14	Kumsi Tetri or Tsnoris Tetra	6557 or 12706	misnomer or true name?																			
Example 4: Synonymies. Distinct accession names and matching fingerprints. Independently Mechtka from BGR013 and Mecka from DEU098, respectively Tourchiena Boia from BGR013 and Turchiena Coia from FRA139 displayed most similar names and showed a matching allelic profile with Koiniariko. Therefore both names were considered as synonyms of prime name Koiniariko. Images of leaves and cluster from DEU098 matched with those of Del Zan *et al*. 2004.																						
Mechtka	BGR13-P15#786	Koiniariko	20739	synonym	Del Zan *et al*. 2004, p. 583	230	234	241	241	133	139	247	249	186	186	258	260	272	272	186	204	243	251
Tourchiena Boia	BGR13-P15#1340	Koiniariko	20739	synonym																			
Turchiena Coia	FRA139-0Mtp1576	Koiniariko	20739	synonym																			
Koiniariko	FRA139-0Mtp2019	Koiniariko	20739	true name																			
Mecka	DEU098-1991-186	Koiniariko	20739	synonym																			

Tab. 3, continued

Example 5: Synonymies. Distinct accession names and matching fingerprints: The allelic profiles of Bela Dinka from BGR013 matched with those of Ribolla Spizade (CIPRIANI *et al*. 2010), Bela Dinka (STAJNER 2013) and Prosecco lungo (CRESPAN *et al*. 2009). In this case morphologic description and photographs of Bela Dinka from Serbia (DRAGOSLAV, pers. communication 2014) were compared with Glera lunga (CRESPAN, pers. communication 2014), which is the official name of Prosecco lungo in the Italian Catalogue (http://catalogoviti.politicheagricole.it/result.php?codice=359).
Matching were the following important characteristics:
- High density of prostrate hairs on shoot tip
- strong intensity of anthocyanin coloration of prostrate hairs on the shoot tip
- three lobed leaves
- rectilinear tooth shape
- strong overlapping of petiole sinus
- short elliptic berry shape
- high density of prostrate hairs and very high density of erect hairs.
Bela Dinka was chosen as prime name as it is considered being an old autochthonous Serbian cultivar.

Bela Dinka	BGR13-P15#145	Bela Dinka	16848	true name	CALO *et al*. 2006, p. 638	228	248	239	255	133	143	239	247	180	195	236	244	256	272	188	194	249	259

Example 6: Misnomers. Distinct accession names and matching fingerprints AND identical accession names and distinct fingerprints: Gara Goybendam and Makhmudu are cultivars existing in Azerbaijan only. Owing to the existence of a second genetic profile from two accessions Mahmudu, it is most likely that Mahmudu from AZE015 is a misnomer and is in fact Gara Goybendam. This assumption needs to be reviewed.

Goybendam	AZE007	Gara Goybendam	21979	true name	PENAHOV and SELIMOV 2008, p. 203 (no photo)	234	240	245	255	143	155	235	249	195	195	236	258	256	270	194	200	257	259
Mahmudu	AZE015-2012-2	Gara Goybendam	21979	misnomer																			
Mahmudu	AZE007	Makhmudu	23305	true name	PENAHOV and SELIMOV 2008, p. 246 (no photo)	230	240	241	245	145	155	239	249	195	195	244	258	270	272	188	200	251	257
Mahmudu	AZE	Makhmudu	23305	true name																			

assistance. With these tools in hand a prime name was attributed to almost 50 % of the 659 unique genotypes.

Duplicated germplasm in Eastern European and Western European countries: A further aim of the present study was the identification of endangered germplasm. With regard to the investigated grapevine cultivars endangered means that genotypes are present in small numbers, e.g. preserved in only one or two collections. In these cases there is a serious risk of cultivar loss and thus genetic erosion. In such cases measures for duplication are needed.

The open question was which of the autochthonous cultivars exist in their country of origin only and which are maintained e.g. in Western Europe. To answer that question the seven collections and SSR-marker databases (see Material and Methods), as well as the Italian grapevine collection of the University of Milan in Torrazza Coste (Pavia) conserving 160 Georgian accessions were consulted. It was assumed that these collections carry the vast majority of grapevine genetic diversity while in other Western European collections Eastern European germplasm is rare. Thus, the results of profile comparison were used for determining the number of unique genotypes, duplicated once, or to find such being not in Western European grapevine collections (Tab. 1).

For each country the number of investigated accessions, the number of duplications within the collection/country and the number of unique profiles are indicated. Furthermore the number and percentage of unique genotypes are given existing in their country of origin only and in Western Europe, respectively. The percentage of genotypes per country duplicated in Western Europe ranged between 25 and 94 %. It turned out that from the Caucasian countries Armenia and Azerbaijan about one fourth is maintained in Western Europe. In contrast, Georgian cultivars are quite frequently encountered in Western Europe. However, the 48 % duplication is due to joint efforts of Georgia and Italy. In addition the figures indicate an increase of duplication by moving further West with some fluctuation between countries. This might be due to individual selection of the proposed material. For example the Romanian partner had chosen most common cultivars. This resulted in 94 % preservation in Western Europe. Bulgaria proposed rare germplasm yielding just 65 % duplication. From Albania, Croatia and Moldavia about 50 % are found in Western European collections, even more are maintained from Ukraine (64 %), Austria (70 %) and Slovenia (73 %).

The high number of accessions and unique autochthonous genotypes investigated in that study allowed an estimation of their preservation status even if not all the accessions which are maintained in repositories of Eastern European countries were investigated. Tab. 2 gives an overview of the maintenance situation summarized over all countries. The first column of Tab. 2 indicates how often an accession is duplicated. The following columns display the number of analyzed accessions, the resulting number of unique profiles, the number of unique profiles not duplicated in Western Europe and how many repositories in Western Europe maintain the material. Overall it can be stated

that from the 659 unique profiles/cultivars 54 % are maintained in the Eastern European countries only. In addition 300 (46 %) genotypes exist in just one Eastern European collection. The analysis of more accessions maintained in Eastern European countries might reduce that proportion but on the other hand could add further unique fingerprints as well. As a first step the 300 unique and not duplicated genotypes need to be fully described and respective data documented in the European *Vitis* Database. In a second step duplication needs to be initiated. Another finding is that apparently the germplasm maintained only in Eastern Europe did infrequently move between the Eastern European countries. This is proven by comparison of the total number of genotypes not duplicated in Western Europe of Tabs 1 and 2. The respective added numbers of unique genotypes over all countries in Tab. 1 resulted in 359, in Tab. 2 in 353 genotypes. These figures demonstrate that for the present study real autochthonous cultivars were selected. Furthermore, the analysis indicates that in Eastern Europe a substantial degree of unexploited genetic diversity is buried. This treasure needs to be conserved and characterized for future generations.

Conclusions

So far a first list displaying the trueness to type data from 306 accessions/cultivars duplicated in Western Europe and 53 accessions/cultivars duplicated in Eastern Europe is in preparation. With respect to cultivar identity assessment, the study revealed unambiguously the necessity of morphologic description and photos from the three most indicative organs, shoot tip, leaf and bunch, (a) for comparison with bibliography, (b) for a clear and explicit definition of the cultivars identity and (c) for the detection of sampling errors and misnomers. Intense exchange between collection curators and skilled personnel are needed to work on questionable accessions.

From the 997 accessions of Eastern European cultivars included in that study, 659 unique profiles/cultivars were found. Three hundred unique profiles/cultivars are most likely endangered as they, according to the analyzed sample, exist in only a single collection. For these 300 genotypes duplicate preservation needs to be initiated. In addition, the high ratio of non redundant genetic material of Eastern European origin suggests an immense unexplored diversity. A quite high number of the rare cultivars were phenotyped within the same COST Action FA1003 (Rustoni *et al.* 2014a, b). This was a first step to evaluate agronomic features and to rediscover valuable Eastern European germplasm for end users like grape growers, wine makers, professional associations and syndicates or national and international organizations like the Organization for Vine and Wine (O. I. V.).

Documentation of the entire information in the European *Vitis* Database will assist both germplasm maintenance and documentation of cultivar specific data. The importance of the accession number is again emphasized. To draw a real picture of the situation continuation of DNA-fingerprinting is strongly recommended.

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