



E-ISSN: 2278-4136
P-ISSN: 2349-8234
JPP 2019; SP1: 439-446

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(Special Issue- 1)
2nd International Conference
**“Food Security, Nutrition and Sustainable Agriculture -
Emerging Technologies”**
(February 14-16, 2019)

**Variability in Grapes (*Vitis vinifera*) Genotypes
Using Morphological Traits**

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Abstract

Knowledge about the extent of genetic diversity/relatedness in grapes germplasm is vital for developing coherent strategies for future gains in productivity and quality. Genetic diversity of twenty four grape accessions maintained at new orchard of Punjab Agricultural University (PAU) were studied using morphological descriptors given by International Plant Genetic Resources Institute (IPGRI) for grapes. Adequate genetic variation was found among the genotypes by using young shoot, mature leaf and mature shoot descriptors. Dendrogram of twenty four genotypes was constructed based on nine morphological markers by using UPGMA which divided 24 grapes genotypes into three main groups and seven subgroups. Genetic similarity values between genotypes ranged from 0.46 to 0.90.

Keywords: Grapes. Genetic variability. Morpho-physiological traits. Cluster analysis

Introduction

The table, wine, juice and other varieties of grapes suitable for processing and the rootstocks constitute a wide range of germplasm. 12 genera and about 600 species of grapes are being cultivated in temperate, tropical and subtropical regions of the world. The genotypes of grape varieties are highly heterozygous and nearly all modern cultivated varieties are hermaphroditic, self-fertile and out-crossing (This *et al.* 2006) [7].

The variability of the grapevine can be observed in terms of both morphology and quality (Alleweldt and Possingham 1988) [2]. Ampelography studies are useful in the identification of grape cultivars (Fatahi *et al.* 2004, Rubio and Yuste 2004, Pavék *et al.* 2003) [3, 9, 6]. The genomic resources that are available to the grapevine research community have increased enormously during the past years with a renewed interest in grapevine (*Vitis vinifera* L.) germplasm resources and analysis of genetic diversity in grapes. Genetic variation, either natural or induced, is valuable for crop improvement. In the present study, genetic diversity in grape genotypes was investigated using morphological markers.

Materials and methods

Twenty four grape accessions (Table 1.) were accessed for different morphological characters. The observations on young shoot characters, mature leaf characters and mature shoot characters were recorded using the grape descriptors given by International Plant Genetic Resources Institute (IPGRI). Descriptors used for characterizing grape accessions are given in Table 2. Young shoot characters were observed at the time of bud burst while mature leaf characters were observed in August- September when leaf was fully expanded. A leaf from the middle portion of the shoot was taken. Mature shoot characters were taken in August-September, when shoot was fully developed. Statistical analysis for the data was conducted using the software programme Numerical Taxonomic and Multivariate Analysis System (NTSYS-pc) version 2.02e (Rohlf 1998) [8]. Similarity was estimated using SIMQUAL function of NTSYS, which computes a variety of similarity coefficient for qualitative data (nominal data).

Similarity matrix value based on Nei and Li (1979) ^[5] coefficient of similarity (D_{ij}) were calculated. The similarity matrix was thus generated and dendrogram was constructed

using UPGMA (Unweighted Pair Group Method using Arithmetic Averages) available in NTSYS.

Table 1: List of twenty four grape accessions accessed for different morphological traits.

Variety/rootstock	Parentage/Specie	Source of collection
Table grape		
Loose perlette	Perlette mutant	NRC Grapes Pune ^a
Seeded hybrid		
Beauty Seedless	(<i>V. Vinefera</i>)	NRC Grapes Pune ^a
Cardinal	'Tokay' x 'Riber' (<i>V. Vinefera</i>)	NRC Grapes Pune ^a
Himrod	'Ontario' x, Sultana' (<i>V. Vinefera</i>)	NRC Grapes Pune ^a
H 144	'Cheema Sahebi' x 'Catawba' (<i>V. Vinefera</i> x <i>Labrusca</i>)	ARI, Pune ^b
Muscat hamburg	'Muscat of Alexandra, x 'Trollinger Schaiava Grossa' (<i>V. Vinefera</i>)	NRC Grapes Pune ^e
Perlette	'Scolokertek Hiralyonje 26' x 'Sultanina Marble' (<i>V. Vinefera</i>)	UC, Riverside
Flame seedless	(Cardinal x Thomson seedless) x (Red Malaga x Tifafihi Ahmer) x (Muscat of Alexandria x Thomson seedless) (<i>V. Vinefera</i>)	INRA France ^d
Wine grape		
Chardonnay	'Pinot Noir' x 'Gouais' (<i>V. Vinefera</i>)	NRC Grapes Pune ^a
Chasan B	'Listan' x 'Pinot' (<i>V. Vinefera</i>)	INRA France ^d
Madeleine Angevine	'Malingre Precoce' x 'Madeleine Royale' (<i>V. Vinefera</i>)	INRA France ^d
Portan	'Grenache Noir' x 'Blauer Portugieser' (<i>V. Vinefera</i>)	INRA France ^d
Juice grape		
H 27	'Diamond jubilee' x 'Rubi Red' (<i>V. Vinefera</i>)	ARI, Pune ^b
Pusa Navrang	'Angevine x 'Rubi Red' (<i>V. Vinefera</i>)	IARI New Dehli ^c
Rootstocks		
deGrasset	<i>V. Champani</i> Planch	NRC Grapes Pune ^a
Dogirde	<i>V. Champani</i>	NRC Grapes Pune ^a
H-324	'Gulabi' x 'James' (<i>V. Vinefera</i> x <i>V. rotundifolia</i>)	ARI, Pune ^b
<i>Vitis Paviflora</i>	<i>Vitis Paviflora</i>	NRC Grapes Pune ^a
1103P	<i>V. berlandieri</i> x <i>v. riparia</i>	NRC Grapes Pune ^a
1616 C	<i>V. solonis</i> x <i>v. riparia</i>	NRC Grapes Pune ^a
Others		
A-21-2		
A-39-1		
A-46-2		

Table 2: Descriptors used for characterizing grape accessions.

S. No	Accession	Character
Young shoot		
1	Form of tip	Closed
		Half open
		Fully open
2	Anthocyanin coloration of tip	Present
		Absent
3	Shoot habit	Erect
		Horizontal
		Drooping
4	Shoot length	Shoot length was measured with the help of measuring scale and shoot from the middle of the raceme was selected.
5	No. of tendrils	Up to two
		More than two
6	Length of tendril	Very short (<11cm)
		Short (12-16cm)
		Medium (17-21cm)
		Long (22-26cm)
		Very long (>30cm)
7.	Colour of upper surface	Colour of upper surface of young shoot of 30 genotypes was noted using the horticultural colour chart.
Mature leaf		
1	Size of blade	Small
		Medium
		Larger
2	Shape of blade	Chordate
		Wedge
		Pentagonal
		Circular

3	No of lobes	Entire leaf
		Three
		Five
		Seven
4	Anthocyanin colouration on veins.	Absent
		Present
5	Profile-cross section at middle of leaf blade	Flat
		V shaped
		Undulate
6	Shape of petiole sinus	Very wide open
		Wide open
		Half open
		Closed
		Lobes overlapping
7	Tooth at petiole sinus	Absent
		Present
8	Leaf area	Mature leaves of grapes genotypes were scanned by computer scanner and leaf area was calculated using leaf area meter.
9	Colour	Colour of mature leaf of grape genotypes were compared with horticultural colour chart.
10	Number of stomata	Calculated per cm ² under microscope.
Mature Shoot		
1	Surface	Smooth
		Angular
		Ribbed
2	Thickness	Thickness of mature shoot was measured with the help of Vernier's Caliper's (cm).

Results and Discussion

Young shoot characters

As presented in Table 3 all the varieties have closed tip except Degraet and Chardonnay which have half open tip. Accessions Chardonnay, H-324, Himrod, Loose Perlette, and Perlette do not have anthocyanin colouration on the tip of young shoots, while in all other accessions it was present. Degraet and H-324 have horizontal type of shoot growth habit, while all other accessions have erect type of shoot habit. Shoot length for 24 genotypes was measured with the help of measuring scale and shoot from the middle of the raceme was selected. Longest shoot length 79.50 cm was noted in Seeded Hybrid while the shortest 7.5 cm was noted in Muscat Hamburg. Number of tendrils for each genotype was counted in young shoot selected from middle of the vine and was found that all the accessions have the three or more than three number of tendrils. There was great variability in the length of the tendrils among different grape accessions. H-27, Loose Perlette, Muscat Hamburg, and 1616 C have short length of tendril. Degraet, Flame Seedless and Perlette were having medium length tendrils, where as rest of accessions have very short tendrils.

Mature leaf characters

Size of blade of eighth leaf from start of third shoot from middle of vine was measured by measuring scale. Size of blade was found maximum in H-27 (280.85) and small in Muscat Hamburg (37.05). Shape of mature leaf blade of all the 24 accessions were compared with the shapes given by IPGRI descriptors.

Accessions Chardonnay, H-324 and Muscat Hamburg were having circular, Porton, Madeline Angevine, A-21-2 and A-46-2 has pentagonal, cordate was found in Loose Perlette where as the rest of accessions were having wedge shaped leaves. No lobe i.e entire leaf was found in Chardonnay and *Vitis parviflora* accessions. Five lobes in mature leaf were noted in, Chasan, H-144, Perlette, Portan, Pusa Navrang, Madeline Angevine and Seeded Hybrid accessions. While rest of the accessions have three lobes in mature leaf. Fig 1. Anthocyanin colouration on main veins was checked for its

presence or absence and ranked as 1 and 0, respectively. Greater variability among grape accessions were found for anthocyanin colouration of main veins on upper side of the leaf. Fig 2. Profile crosssection was found undulated in Flame Seedless, Degraet and 1616 C. Rest of the accessions were having V shaped profile cross section Great variation was found among grapes accessions in shape of petiole sinus. *Vitis parviflora* was having very wide petiole sinus Accessions Beauty Seedless, Cardinal, 1103-P and were having wide open shape of petiole sinus. However, H-27 was having overlapping type of petiole sinus Fig. 3 Presence or absence of tooth at petiole sinus was observed and ranked 0 if tooth was absent and 1 if it was present. Mature leaves of grape genotypes were scanned by computer scanner and then leaf area was calculated using leaf area meter and it was found that H-27 was having largest leaf area (177.36 cm²) while, Muscat Hamburg was having the smallest (34.51 cm²) leaf area among the grape accession under study. Leaf tip of all the 24 accessions was compared with different leaf shapes. In all the leaves type of leaf tip was found to be acuminate. Number of stomata calculated for all grape accessions under investigation are given in Table 5. Among all the accessions, H-27 (155/mm²) had the maximum number of stomata, while Dogridge was having the minimum number of stomata (77 mm²).

Surfaces of mature shoot were compared by images given in IPGRI descriptors viz., smooth, angular and ribbed. All the grape accession had smooth surface.

Thickness in mature shoot was measured with the help of Vernier's Caliper's. Maximum thickness among the accessions was noted in H-27 (1.8 cm); whereas it was minimum in Portan (0.46 cm).

Cluster analysis based on morphological data.

The dendrogram of 24 genotypes was constructed based on ten morphological markers (form of tip, anthocyanin coloration of tip, shoot habit, length of tendril, shape of blade, number of lobes, anthocyanin colouration of main veins on upper side of leaf, profile cross section, shape of petiole sinus and tooth at petiole sinus) by using UPGMA method in order

to examine the genetic diversity (Fig 4 & 5). Genetic similarity values between genotypes ranged from 0.46 to 0.90. The 24 grapes genotypes were divided into three Groups and further into seven sub groups i.e Sub group I, II, III, IV, V, VI and VII as shown in Fig 4. Subgroup 1 consisted of five accessions (Beauty Seedless, Seeded Hybrid, H-144, Cardinal, and Loose Perlette). Sub group II consisted Degraset and Flame Seedless, while sub group III consisted of 1616 C and H-27. Sub group IV consisted of A-21-2, A-46-2, while sub group V consist of five Accessions viz. A-39-1,

Chasan, Pusa Navrang, Porton, Dogridge. Sub group VI consists Muscat Hamburg and Perlette, while in Sub group VI the accessions were Chardonny, H-324, Himrod, Madeline Angevine and *Vitis parviflora*. Grape cultivars were identified by comparison of morphological characters, a method based on the visual evaluation of leaf, fruit, shoot tip and other organs Traditionally, grapevine identification has relied on the skills of ampelography. Ampelography searches for differences among varieties and ensures classification.

Table 3: Young shoot characters of grapes accessions.

S. No.	Accession	Form of tip (ā)	Anthocyanin coloration of tip (b)	Shoot habit (c)	Shoot length (cm)	No. of tendrils (ë)	Length of tendril (f)	Colour of upper surface (ğ)
1	1103-P	1	1	1	48.6	2	1	1
2	1616-C	1	1	1	44.3	2	3	1
3	A-21-2	1	0	1	47.5	2	1	1
4	A-39-1	1	1	1	46	2	1	1
5	A-46-2	1	1	1	75.7	2	1	1
6	Beauty Seedless	1	1	1	50.3	2	1	1
7	Cardinal	1	1	1	50.3	2	5	1
8	Chardonny	3	0	1	48.7	2	1	1
9	Chasan	1	1	1	66.1	2	1	1
10	De graset	3	1	5	76.5	2	5	1
11	Dogridge	1	1	1	44.7	2	1	1
12	Flame Seedless	1	1	1	60.5	2	5	1
13	H-144	1	1	1	52	2	5	1
14	H-27	1	1	1	55.7	2	3	1
15	H-324	1	0	5	47.9	2	1	1
16	Himrod	1	0	1	43.7	2	1	1
17	Loose Perlette	1	0	1	157	2	3	1
18	Medaline Angevine	1	1	1	43.3	2	1	1
19	Muscat Hamburg	1	1	1	27.5	2	3	1
20	Perlette	1	0	1	45.5	2	5	1
21	Porton	1	1	1	53.3	2	1	1
22	Pusa Navrang	1	1	1	31.4	2	1	1
23	Seeded Hybrid	1	1	1	79.5	2	1	1
24	<i>Vitis parviflora</i>	1	1	1	45	2	1	1

ā: 1- closed type; 3-half open and 5-fully open

b: 0- absent and 1; present

c: 1- erect; 5- horizontal and 9- drooping habit

ë: 1- upto two; 2- three or more than three

f: 1- very short length (<11cm); 5- medium length (19-21 cm); 7- long length (24-26 cm); 9-very long length (>30 cm)

ğ: 1- green and 0- other

Table 4: Mature leaf characters of grape accessions.

No.	Accession	Size of blade (cm ²)	Shape of blade (b)	No. of lobes (c)	Anthocyanin colouration of main veins on upper side of leaf (d)	Profile cross section (ë)
1	1103-P	145.20	2	1	1	2
2	1616-C	151.20	2	2	0	5
3	A-21-2	64.97	3	2	0	2
4	A-39-1	110.00	3	3	0	2
5	A-46-2	61.41	3	2	0	2
6	Beauty Seedless	107.50	2	2	1	2
7	Cardinal	108.30	2	2	0	2
8	Chardonny	64.75	4	1	1	2
9	Chasan	151.61	2	3	0	2
10	De graset	155.25	2	2	1	5
11	Dogridge	159.50	2	2	0	2
12	Flame Seedless	178.45	2	2	1	5
13	H-144	163.60	2	3	1	2
14	H-27	280.85	2	2	0	2
15	H-324	90.37	4	2	1	2
16	Himrod	110.60	2	2	1	2
17	Loose Perlette	84.46	2	2	1	2
18	Medaline Angevine	87.63	1	2	1	2

19	Muscat Hamburg	37.05	3	3	0	2
20	Perlette	198.45	4	2	0	2
21	Porton	70.56	2	3	0	2
22	Pusa Navrang	86.40	2	3	0	2
23	Seeded hybrid	48.00	4	3	1	2
24	<i>Vitis parviflora</i>	65.76	5	1	1	2

b: 1- cordate; 2- wedge shaped; 3- pentagonal; 4- circular

c: 1- entire leaf; 2- three; 3- five; 4- seven

ø: 0- absent; 1- present

ë: 1- flat; 2- V-shaped; 5- undulate

Table 5: Mature leaf characters of grape accession.

No.	Accession	Shape of petiole sinus (f)	Tooth at petiole sinus (ğ)	Leaf Area (cm ²)	Colour	Stomata/mm ²
1	1103-P	2	0	61.6	138A	137
2	1616-C	3	1	135	147A	108
3	A-21-2	3	0	38.7	146A	135
4	A-39-1	3	0	56.7	146B	115
5	A-46-2	3	1	23.4	147A	145
6	Beautyseedless	2	0	65.4	139B	139
7	Cardinal	2	0	63.1	147A	117
8	Chardonny	4	1	34.5	143A	145
9	Chasan	3	0	75.6	147 A	99
10	De graset	2	0	59.9	146A	154
11	Dogridge	3	0	58.3	147A	77
12	Flame Seedless	3	0	99.7	137A	135
13	H-144	2	0	57.1	139A	126
14	H-27	4	1	177.3	144B	155
15	H-324	2	1	103.6	148A	140
16	Himrod	3	1	64.9	137A	154
17	Loose Perlette	2	0	35.5	147A	103
18	MedalineAngevine	3	1	59.1	143A	108
19	Muscat Hamburg	3	0	34.7	147A	135
20	Perlette	3	0	114	139C	147
21	Porton	3	1	35.3	139A	96
22	Pusa Navrang	3	0	56.2	146B	145
23	Seeded hybrid	2	0	28.7	147A	129
24	<i>Vitis parviflora</i>	1	1	29.2	139A	139

f: 1-very wide open; 2- wide open; 3- half open; 5- closed; 6- overlapping

ğ: 0- absent; 1- present

Table 5: Mature shoot characters of grape accession.

No.	Accession	Surface	Thickness (cm)
1	1103-P	Smooth	0.54
2	1616-C	Smooth	0.52
3	A-21-2	Smooth	0.87
4	A-39-1	Smooth	0.86
5	A-46-2	Smooth	0.54
6	Beauty seedless	Smooth	0.97
7	Cardinal	Smooth	0.86
8	Chardonny	Smooth	0.88
9	Chasan	Smooth	1.5
10	Degraset	Smooth	1.3
11	Dogridge	Smooth	0.76
12	Flame Seedless	Smooth	0.99
13	H-144	Smooth	0.77
14	H-27	Smooth	1.8
15	H-324	Smooth	0.47
16	Himrod	Smooth	1.1
17	EarlyPerlette	Smooth	0.73
18	Madeline Angevine	Smooth	0.76
19	Muscat Hamburg	Smooth	0.56
20	Perlette	Smooth	0.59
21	Porton	Smooth	0.46
22	Pusa Navrang	Smooth	0.54
23	Seeded Hybrid	Smooth	0.88
24	<i>Vitis parviflora</i>	Smooth	0.77

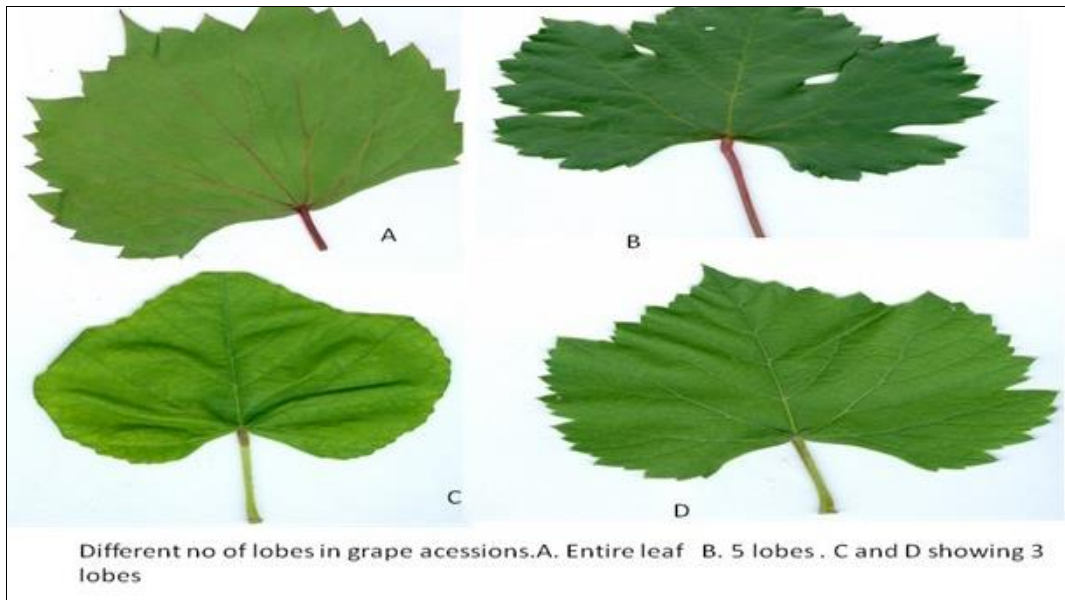


Fig 1: Diversity in number of lobes in leaves of grape accessions

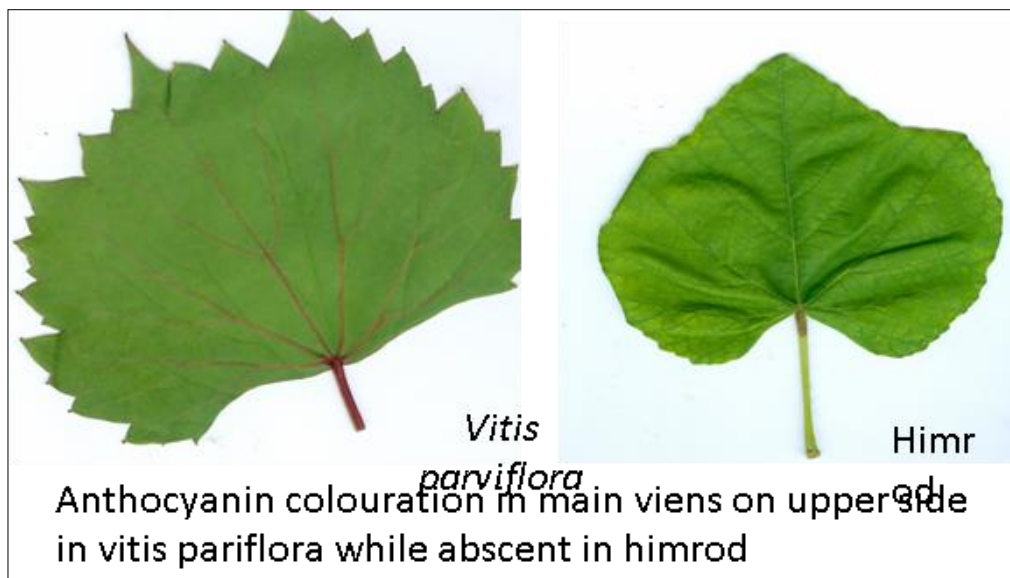


Fig 2: Anthocyanin colouration in main veins on upper side

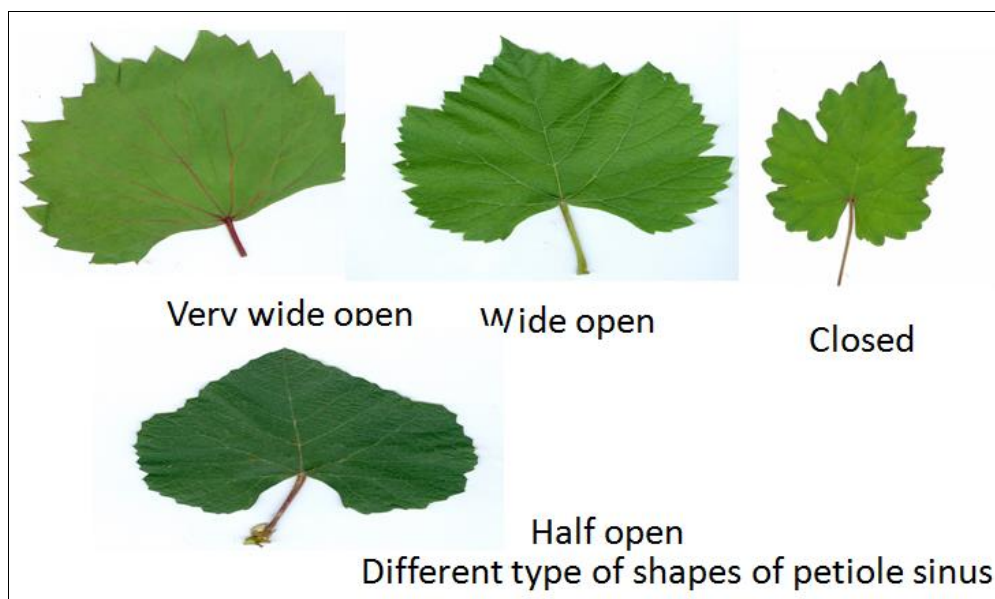


Fig 3: Different types of shapes of petiole sinus

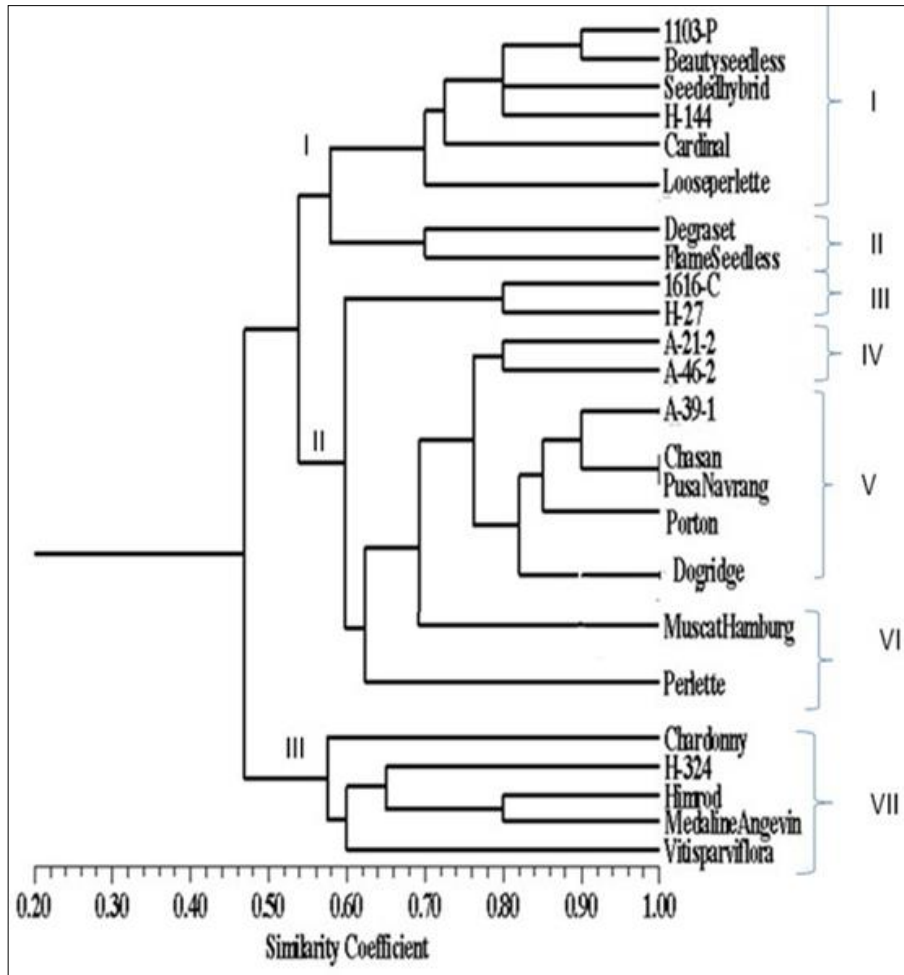


Fig 4: The dendrogram of 24 genotypes was constructed based on ten morphological markers using UPGMA method

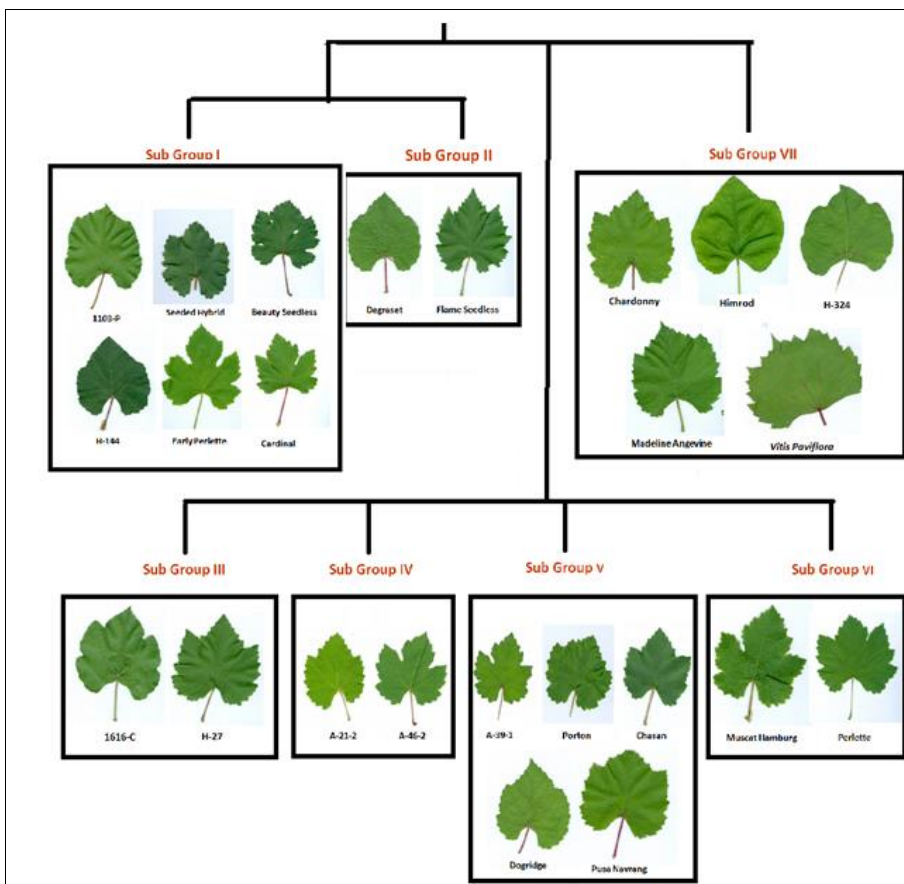


Fig 5: Grouping of 24 genotypes was constructed based on ten morphological markers using UPGMA method.

References

1. Anderson JA, Churchill GA, Autrique JE, Tanksley SD, Sorrells ME. Optimizing parental selection for genetic linkage maps Genome. 1993; 36:181-6.
2. Alleweldt G, Possingham JV. Progress in grapevine breeding. Theor. Appl. Genet. 1988; 75:669-673.
3. Fatahi R, Ebadi A, Vezvaei A, Zamani Z, Ghanadha MR. Relationship among quantitative and qualitative characters in 90 grapevine (*Vitis vinifera*) cultivars Acta Hort. 2004; 640:275-282.
4. Isci B, Yildirim HK, Altindisli A. A review of the authentication of wine origin by molecular markers. J Inst. Brew. 2009; 115(3):259-264.
5. Nei Masatoshi, Li Wen-Hsiung. Mathematical model for studying genetical variation in terms of restriction endonucleases. Proceedings of the National Academy of Sciences of the United States of America. 1979; 76(10):5269-5273.
6. Pavék DS, Lamboy WF, Garvey EJ. Selecting in situ conservation sites for grape genetic resources in the USA Genetic Resources and Crop Evolution. Biomedical Life Sci. 2004; 50(2):165-173.
7. This P, Lacombe T, Thomas MR. Historical origins and genetic diversity of wine grapes. Trends in Genetics. 2006; 22(9):511-519.
8. Rohlf FJ. NTSYS-pc. Numerical taxonomy and multivariate analysis system. (*Applied Biostatistics*, New York), 1998.
9. Rubio JA, Yuste J. Ampelographic differentiation of 'Tempranillo' clones from different area of origin, according to their synonym. Acta. Hort. 2004; 652:73-79.