Stability of Ampelometric Characteristics of *Vitis vinifera* L. cv. 'Syrah' and 'Sauvignon blanc' Leaves: Impact of Within-vineyard Variability and Pruning Method/Bud Load

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Historically, grapevine (Vitis vinifera L.) leaf characterisation has been a driving force in the identification of cultivars. In this study, ampelometric (foliometric) analysis was done on leaf samples collected from hand-pruned, mechanically pruned and minimally pruned 'Sauvignon blanc' and 'Syrah' vines to estimate the impact of within-vineyard variability and a change in bud load on the stability of leaf properties. The results showed that within-vineyard variability of ampelometric characteristics was high within a cultivar, irrespective of bud load. In terms of the O.I.V. coding system, zero to four class differences were observed between minimum and maximum values of each characteristic. The value of variability of each characteristic was different between the three levels of bud load and the two cultivars. With respect to bud load, the number of shoots per vine had a significant effect on the characteristics of the leaf laminae. Single leaf area and lengths of veins changed significantly for both cultivars, irrespective of treatment, while angle between veins proved to be a stable characteristic. A large number of biometric data can be recorded on a single leaf; the data measured on several leaves, however, are not necessarily unique for a specific cultivar. The leaf characteristics analysed in this study can be divided into two groups according to the response to a change in bud load, i.e. stable (angles between the veins, depths of sinuses) and variable (length of the veins, length of the petiole, single leaf area). The variable characteristics are not recommended to be used in cultivar identification, unless the pruning method/bud load is known.

INTRODUCTION

Worldwide, up to 15 000 grapevine cultivars are grown. The characterisation of each of these cultivars has high importance for preservation and propagation. The morphological identification of *Vitis* taxa is called *ampelography*. This term was mentioned for the first time in the middle of the 17th century (Viala & Vermorel, 1910), although it dates back to ancient times (Pliny the Elder, 1855). Together with

molecular genetic and chemotaxonomic characterisation, it still is an important method in the description of cultivars today (Zulini *et al.*, 2005; Gago *et al.*, 2009). Morphological identification should be the first step in a standard protocol for the characterisation and documentation of single plants, and even for the establishment of a germplasm collection (Ortiz *et al.*, 2004), in order to avoid misnomers (Galet, 1956) and synonymy (González *et al.*, 2007). Phenotypical

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descriptions of cultivars are based on the characterisation of organs, such as roots, canes, buds, shoots, leaves, tendrils, fruits, seeds and trichomes, according to the International Organisation of Vine and Wine (O.I.V., 2009a). It should be noted that not only environmental factors, such as soil properties (Ferree & Streeter, 2004), water availability (Reynolds & Naylor, 1994) and salinity (Fisarakis *et al.*, 2001), but also the rootstock (Krstic *et al.*, 2005), nutritional status (Schreiner *et al.*, 2012) and health of the plant may have significant effects on morphological patterns (Cervera *et al.*, 2001).

The grapevine leaf is an extremely variable organ. The shape (predominantly palmate) and size of the leaf depends on the number of lobes, the length of the veins and the depth of the sinuses. Almost all cultivars have individual shapes of lamina, which present the possibility of identification (Mullins et al., 2003). Metric analyses of certain parameters of the leaf are called ampelometry. There are almost 50 biometric descriptors related to the mature leaf, e.g. size of the lamina, angles between the veins, lengths of veins, etc. The ampelometric parameters of the cultivars are available in the literature (Németh, 1966, 1967, 1970; Goussard, 2008) and "on-line", such as in the European Vitis Database (Maul et al., 2012). Ortiz et al. (2004) classified leaf characteristics into three groups, namely stable or objective characteristics, discriminant and non-discriminant characteristics. Sefc et al. (2001) reported that morphological identification could be influenced by interpretation. Digital image analysis could support ampelometric descriptions with time-efficient and objective measurements, which exclude arbitrary decisions. Software environments like Ampelocad (Alessandri et al., 1996), Digital Image Processing System MIP 1.4 (MICROM) (Martinez & Mantilla, 1995) and SuperAmpelo (Soldavini et al., 2009), as well as outline data analysis of the lamina (Kondou et al., 2002), are suitable for the digital reconstruction and characterisation of grapevine leaves. Our previous results proved that ampelometry in a raster-graphic environment is a powerful tool for the digital analysis and characterisation of grapevine leaves (Bodor et al., 2012).

The concept of within-vineyard variability is not new (Bramley & Hamilton, 2004; Hunter *et al.*, 2010). Its effect on yield and quality is well known and can be observed between and within vines (Rolley *et al.*, 2007). The description of the cultivars given by the O.I.V. also considers this type of variability of each cultivar, as in the case of 'Sauvignon blanc' leaves, where the 'Length of teeth compared with their width' (O.I.V. 078) is "short" (Nr. 3) to "medium" (Nr. 5), but other properties are also variable, such as the 'Mature leaf: degree of opening / overlapping of petiole sinus' (O.I.V. 079), and the 'Mature leaf: degree of opening / overlapping of upper lateral sinuses' (O.I.V. 082, etc.). This variability has also been observed in the case of 'Syrah' (O.I.V., 2009b).

Within ampelometric characterisation, the size of the leaf blade (*single leaf area* - SLA) is a crucial parameter, as it determines photosynthetic activity, as affected by the structure of the vine, training and trellising system, microclimate and pruning method (Clingeleffer & Krake, 1992). Pruning is an integral part of the success of any grapevine trellising system in relation to quantitative and qualitative production. The effect of pruning level on

grapevine production and physiology is well known (Intrieri et al., 2011). Detailed observations related to the change in SLA caused by bud load have been reported. For example, Clingeleffer (1984) detected no significant differences in ALA (average leaf area) and LAI (leaf area index) as a result of minimal pruning (MIN) of 'Sultana' vines compared to those subjected to conventional pruning (CP). Sommer et al. (1995) reported reduced SLA and LAI in the case of vines subjected to MIN compared to those collected from conventionally pruned 'Sultana'. Similar observations were reported by Downton and Grant (1992) on 'Riesling' and by Sommer and Clingeleffer (1993) on 'Cabernet Sauvignon'. Both reports showed almost twofold higher SLA in the case of CP compared to MIN. Later, similar results were published on 'Chardonnay' (Poni et al., 2000) and 'Sangiovese' (Intrieri et al., 2001). The lowest level of differences caused by bud load is the variance in SLA as reported by Cangi and Kilic (2011) on 'Narince'; no significant difference in MLA (mean leaf area) was found with 16 buds/vine compared to 24 buds/ vine. Except for average leaf area, the impact of viticultural practices on the deeper ampelometric description of leaves has not received any attention.

The objective of this study was to estimate withinvineyard variability and to determine the impact of viticulture practices, such as pruning method and therefore bud load/shoot number, on the stability of ampelometric characteristics of two grapevine cultivars.

MATERIALS AND METHODS

Vineyard

The experiment was carried out in 2011 on 14-year-old 'Sauvignon blanc' and 'Syrah' vines grown on the experimental farm of ARC Infruitec-Nietvoorbij, situated in the Robertson district of South Africa (Archer & Van Schalkwyk, 2007). A VSP (vertical shoot positioning) trellising system was used. The vines were spaced at 3.0 m \times 1.5 m. Both cultivars were grafted onto Richter 99 RY13 rootstock. No topping or leaf removal practices were applied. Three pruning methods were used and each treatment, carried out in a single row, comprised 44 vines per cultivar:

- hand spur pruned (HAND): 24 to 28 buds/vine
- mechanically pruned (MECH): 63 to 76 buds/vine
- minimally pruned (MIN): 97 to 128 buds/vine.

Sample collection and measurements

Sample collection and ampelometric observations were made according to the O.I.V. descriptor list (O.I.V., 2009a). Ten mature leaves were collected randomly from the middle third of 10 shoots (one sample/one plant) before harvest (at the end of March 2011). It was assumed that the collected leaves were fully developed at this time.

Characterisation of the leaves was done using 18 O.I.V. characteristics, according to the descriptor list, taking into consideration those characters recommended to be described between berry set and *véraison*: Mature leaf: size of blade (O.I.V. 065); Mature leaf: length of petiole compared to length of middle vein (O.I.V. 093); Mature leaf: depth of upper lateral sinuses (O.I.V. 094).

During the measurements, two modifications were made compared to the O.I.V. recommendation (signed with * and **):

- Mature leaf: opening/overlapping of lobes on each side of petiole sinus (O.I.V. 618*) was measured according to the distance between the tooth tip of the N₅ veins. In the case of overlapping of lobes, values are given as positive numbers, while in the case of an open sinus, values are given as negative numbers.
- Mature leaf: depths of sinuses (O.I.V. 094**) is given in numeric scale as N2-1/S1; N2-2/S2; if the value is around 1, it means the sinus was "very shallow" (O.I.V. Nr. 1), while the sinus is deeper as the value increases (O.I.V. Nr. 3-9).

Beside the 18 O.I.V. descriptors, single leaf area (SLA), length of the petiole (P) and depth of the petiole sinus (PS-1; PS-2) were also monitored. Detailed ampelometric dimensions, representing 22 biometric parameters (see Table 1) were recorded for each sample (Fig. 1), using GRA. LE.D. software according to the protocol described in Bodor *et al.* (2012).

Measured features were coded from 1 to 9, where 1 to 3 represented shorter, or low, value, and 7 to 9 coded for longer or high values according to the O.I.V. descriptor list (2009a).

Data analysis

The mean, standard deviation, minimum and maximum were calculated for each treatment and for each cultivar. Statistical analysis MANOVA (multifactorial analysis of variance) was carried out with PASW 18 (release version 18.0.0, SPSS Inc., Chicago, IL, www.spss.com) to test for significant differences between treatments and cultivars. Wilk's λ counts the proportion of variance in the combination of the dependent variables that is unaccounted for by the independent variables (fixed factors), such as treatments and cultivars. The lower the Wilk's λ values, the stronger the discriminating effects of the factors. The effect size measure (denoted by partial η^2), which gives the variance explained by a given independent factor of the variance remaining after excluding variance explained by the other independent factor, was also considered. The higher the partial η^2 values, the stronger the effect sizes (Everitt & Dunn, 1991). The power of the effect, i.e. the probability of correctly rejecting the H₀ hypothesis that assumes the homogeneity of means, is also given. High power values mean that significant results are detected when there are real differences between factor levels. Normality of residuals was checked using the D'Agostino test, with P > 0.05 (D'Agostino et al., 1990). With the aim of normalising the distribution of the data, Box-Cox transformations (Box & Cox 1964) were made in the case of some parameters, such as S-1 ($\lambda = -0.1$), N2-1/S-1 ($\lambda = -1.4$), and α ($\lambda = 0.3$). Equality of variances was tested with Levene's test. Depending on the results, Tukey's or Dunnett's post hoc test was applied. Using the main vein as central axis, the symmetry of the leaves was proved with correlation analysis of the opposite pairs (n = 60).

Graphic reconstruction

The graphic reconstructions of the leaves were made by using the datasets obtained from the 10 leaves of each of the two cultivars (Bodor *et al.*, 2012).

RESULTS

Data obtained from the ampelometric descriptions of the samples are given in Table 1. A two-way MANOVA revealed a significant multivariate main effect for both pruning method [Wilk's $\lambda = 0.04$; F (46; 64) = 5.58; p < 0.001; partial eta squared = 0.8; power to detect the effect was as high as 0.999] and cultivar [Wilk's $\lambda = 0.39$; F (23;32) = 2.2; p < 0.05, partial eta squared = 0.6; observed power was 0.95]. We thus concluded that pruning method/bud load (shoot number) had a significant effect on grapevine leaf characteristics. Foliometric characteristics changed for both the 'Syrah' and 'Sauvignon blanc' cultivars. The interaction was insignificant (p > 0.10). Given the significance of the overall test, the univariate main effects were examined. The results of the *post hoc* tests are given in Table 1.

Within-vineyard variability

The variability of characteristics within each treatment in each cultivar was analysed by coding the minimum and maximum values of measured properties according to the O.I.V. coding system from 1 to 9 (Table 1). Our observations of within-vineyard variability proved that ampelometric characteristics were highly diverse within 10 collected leaves.

In the case of 'Sauvignon blanc', the lengths of veins, sinuses and petioles showed less variability than those of 'Syrah'. Observations of angles between veins showed that 'Sauvignon blanc' had higher variability than 'Syrah'. According to the results, only length of vein N1 (O.I.V. 601) (67.72 mm to 88.8 mm = "very short") and length of vein N2-1 (O.I.V. 602) (58.49 mm to 73.96 mm = "very short") had such a low variability in MIN 'Sauvignon blanc' that it could be categorised into a class of its own. In the case of MIN 'Syrah', only the depth of the upper sinus proved to be stable.

For both cultivars, the opening of the petiole sinus (α ; PS1-PS2) proved to be the most variable characteristic. In the case of HAND 'Sauvignon blanc' it varied from -27.06° (+17.95 mm) to 33.14° (-15.59 mm) between vines, whereas in the case of HAND 'Syrah' it varied from -39.25° (+29.25 mm) to 28.99° (-15.86 mm), which means that there were variations of "open" to "overlapped" petiole sinuses in both cultivars. High variability was also observed in angle δ for both cultivars.

Effect of bud load on morphological characteristics Single leaf area

The SLA proved to have the most variable characteristic, significantly decreasing with level of bud load as induced by pruning method. Leaves on the MECH vines were 28% and those on MIN 59% smaller than those of the HAND vines (Fig. 2).

Lengths of veins

In general, the size of a leaf lamina is related to the lengths of the veins (Beslic *et al.*, 2010). Significant differences were recorded in almost all veins, but not to the same extent. Differences between the MIN and MECH datasets were much larger than those between MECH and HAND. In the case of 'Sauvignon blanc', the veins indicated with N3-1 and

N3-2 were more sensitive to pruning treatment than other veins. Furthermore, the sectors next to the petiole sinus and the N3-2 and N3-1 veins were the most variable. According to our results, 'Sauvignon blanc' leaves proved to be more sensitive to pruning system/bud load than 'Syrah' leaves (Fig. 2).

Symmetry of leaves

If the main vein is considered as a central axis, the symmetry of the leaf can be studied. Strong correlations between lengths of veins N2-1 and N2-2 ($R^2 = 0.843$), N3-1 and N3-2 ($R^2 = 0.900$), S-1 and S-2 ($R^2 = 0.821$), and PS-1 and PS-2 ($R^2 = 0.885$) were found ($R^2 = 0.885$) where found ($R^2 = 0.885$) were found ($R^2 = 0.885$) where found ($R^2 = 0.885$) were found ($R^2 = 0.885$) where found ($R^2 = 0.885$) were found ($R^2 = 0.885$) where found ($R^2 = 0.885$) were found ($R^2 = 0.885$) where found ($R^2 = 0.885$) where found ($R^2 = 0.885$) were found ($R^2 = 0.885$) where found ($R^2 = 0.885$) where found ($R^2 = 0.885$) were found ($R^2 = 0.885$) where found ($R^2 = 0.88$

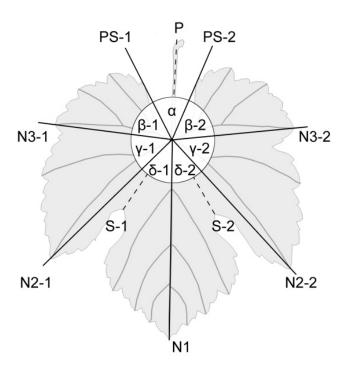


FIGURE 1
Grapevine (*Vitis vinifera* L.) leaf with indication of some ampelometric parameters.

were obtained when correlation analyses were done for both cultivars separately (data not shown). These results indicated that the change in leaf properties is symmetrical.

Depth of sinuses

No significant changes in depth of sinuses were observed, except in the case of the MIN treatment, where sinuses were 21% to 33% deeper than those of the other two treatments (p > 0,05). However, the ratio between the depth of the sinuses and the lengths of the veins (N2-1/S-1; N2-2/S-2) did not change significantly (p > 0.05).

Angles between the veins

Angles between the veins proved to be the most stable. No significant differences were observed in the case of 'Syrah' (p > 0,05). Angles between veins, viz. γ -1, γ -2 and α (opening of the petiole sinus) of 'Sauvignon blanc', were more sensitive with MIN than with HAND and MECH.

Length of the petiole and ratio to the main vein

Treatments had significant effects on the lengths of the petioles; 'Sauvignon blanc' was more sensitive to pruning method than 'Syrah'. For the MIN vines, the length of the petiole was 50% shorter than that of the HAND vines, and it was 30% shorter than that of the MECH vines. The ratio between the main vein and he petiole was almost 1:1 in the case of HAND for both cultivars. For the MIN vines, the length of the petiole decreased by almost 25% related to the length of the main vein.

DISCUSSION

The morphological description of the grapevine leaf has great importance, since other representative organs, such as bunches, can be examined only for a limited time. To obtain a reliable description of the lamina, the elasticity that may be induced by viticultural practices has to be investigated.

To estimate morphological variability caused by pruning technique, within-vineyard variability was examined first. It can be concluded that only a few characteristics in this sample set were stable. Almost all of the features "switched"

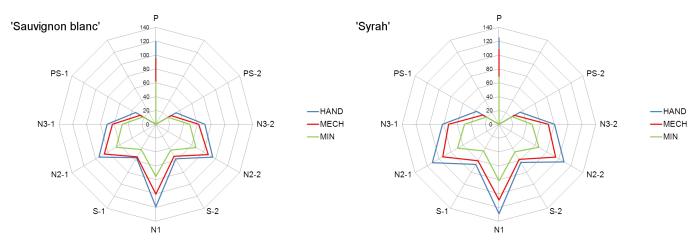


FIGURE 2

Graphic reconstruction from the average vein, petiole length and depths of sinuses data obtained from Vitis vinifera L. cv. 'Sauvignon blanc' (A) and 'Syrah' (B) vines with different bud load/shoot number.

TABLE 1 Ampelometric data for hand (HAND), mechanically (MECH) and minimally (MIN) pruned *Vitis vinifera* L. cv. 'Sauvignon blanc' and 'Syrah' vines, with the relevant O.I.V. coding.

Charac teristic	Unit/ OIV code	'Sauvignon blanc'								
		HAND				ME	СН	MIN		
		Average	St. dev.	$Min \rightarrow Max$	Average	St. dev.	$Min \rightarrow Max$	Average	St. dev.	$Min \rightarrow Max$
SLA	cm ²	184.19 _c	± 20.65	$151.36 \rightarrow 214.26$	131.91 _b	± 19.55	$114.79 \rightarrow 176.56$	75.14 _a	± 13.42	$56.98 \rightarrow 96.82$
OIV No.	-	-		-	-		-	-		-
N1	mm	118.94 _c	± 9.08	$106.02 \rightarrow 130.74$	100.98_{b}	± 9.14	$92.56 \rightarrow 123.8$	75.32 _a	± 5.89	$67.72 \rightarrow 88.8$
OIV No.	601	3		$3 \rightarrow 5$	3		<i>3</i> → <i>5</i>	1		1
P	mm	119.48 _c	± 15.96	$101.92 \rightarrow 154.34$	95.02 _b	± 5.11	$88.25 \rightarrow 102.96$	61.26 _a	± 11.14	$47.86 \rightarrow 82.07$
OIV No.	-	-		-	-		-	-		-
P/N1	ratio	1.00_{b}	± 0.09	$0.87 \rightarrow 1.23$	0.94_{ab}	± 0.08	$0.81 \rightarrow 1.1$	0.81_{a}	± 0.13	$0.66 \rightarrow 1.08$
OIV No.	093	5		$3 \rightarrow 7$	5		$3 \rightarrow 5$	3		$1 \rightarrow 5$
N2-1	mm	94.97 _b	± 6.01	$83.34 \rightarrow 105.12$	87.55 _b	± 8.43	$80.14 \rightarrow 106.4$	66.76 _a	± 5.62	$58.49 \rightarrow 73.96$
OIV No.	602	3		$3 \rightarrow 5$	3		$3 \rightarrow 5$	1		1
N2-2	mm	94.67 _b	± 8.79	$76.59 \rightarrow 104.29$	85.88 _b	± 7.26	$77.22 \rightarrow 97.51$	66.01 _a	± 6.84	$56.37 \rightarrow 76.65$
OIV No.	602	3		$3 \rightarrow 5$	3		$3 \rightarrow 5$	1		$1 \rightarrow 3$
S-1	mm	57.33 _b	± 7.80	$40.1 \rightarrow 69.14$	53.11 _b	± 8.04	$38.1 \rightarrow 62.1$	42.95 _a	± 5.60	$35.5 \rightarrow 51.51$
OIV No.*	605	3		$3 \rightarrow 5$	3		$1 \rightarrow 5$	3		$1 \rightarrow 3$
S-2	mm	55.69 _b	± 8.16	$43.12 \rightarrow 66.25$	54.18 _b	± 4.53	$49.5 \to 65.06$	41.95 _a	± 3.19	$37.42 \rightarrow 46.77$
OIV No.*	605	3		$3 \rightarrow 5$	3		$3 \rightarrow 5$	3		$1 \rightarrow 3$
N2-1/S-1	ratio	1.68 _a	± 0.23	$1.44 \rightarrow 2.23$	1.67 _a	± 0.23	$1.38 \rightarrow 2.10$	1.57 _a	± 0.24	$1.29 \rightarrow 2.03$
OIV No.	094**	3		$3 \rightarrow 5$	3		$3 \rightarrow 5$	3		$3 \rightarrow 5$
N2-2/S-2	ratio	1.63 _a	± 0.23	$1.21 \rightarrow 1.91$	1.59 _a	± 0.17	$1.41 \rightarrow 1.86$	1.57 _a	± 0.16	$1.36 \rightarrow 1.85$
OIV No.	094**	3		$3 \rightarrow 5$	3		$3 \rightarrow 5$	3		$3 \rightarrow 5$
N3-1	mm	70.79 _c	± 5.41	$59.65 \to 78.44$	62.15 _b	± 6.66	$52.71 \rightarrow 74.3$	49.12 _a	± 7.21	$38.7 \rightarrow 59.8$
OIV No.	603	5		$3 \rightarrow 5$	3		$3 \rightarrow 5$	3		$1 \rightarrow 3$
N3-2	mm	70.53 _c	± 5.85	$60.18 \rightarrow 79.14$	62.85 _b	± 6.58	$54.33 \rightarrow 72.67$	48.51 _a	± 6.46	$36.63 \rightarrow 58.52$
OIV No.	603	5		$3 \rightarrow 5$	3		$3 \rightarrow 5$	3		$1 \rightarrow 3$
PS-1	mm	33.73 _b	± 3.99	$28.45 \rightarrow 39.12$	26.21 _b	± 4.72	$20.39 \rightarrow 36.24$	21.50 _a	± 5.93	$13.82 \rightarrow 29.51$
OIV No.	-	-		-	-		-	-		-
PS-2	mm	33.59 _c	± 4.58	$24.08 \rightarrow 38.95$	25.28 _b	± 3.34	$21.6 \rightarrow 33.94$	20.12 _a	± 3.56	$15.32 \rightarrow 26.13$
OIV No.	-	-		-	-		-	-		-
δ-1		43.54° a		$30.61 \rightarrow 58.22$	37.03° _a	± 6.48	$29.36 \rightarrow 46.89$	37.2° _a	± 5.95	$28.25 \rightarrow 47.52$
				$1 \rightarrow 7$			$1 \rightarrow 5$	3		$1 \rightarrow 5$
δ-2				$21.1 \rightarrow 59.55$						
OIV No.				$1 \rightarrow 7$	3		$1 \rightarrow 3$	3		$1 \rightarrow 5$
γ-1		-	± 6.68	$54.43 \rightarrow 72.79$				-		
OIV No.		7		$5 \rightarrow 9$	7		$3 \rightarrow 9$	5		$3 \rightarrow 7$
γ-2			± 6.32	$55.82 \rightarrow 72.03$						
OIV No.		7		$7 \rightarrow 9$			$3 \rightarrow 7$	5		$3 \rightarrow 7$
β-1		-		$64.31 \rightarrow 90.18$	-					
OIV No.		9		$7 \rightarrow 9$						$3 \rightarrow 9$
β-2				$55.63 \rightarrow 102.53$	u			u		
OIV No.				$5 \rightarrow 9$			$5 \to 9$			$3 \rightarrow 9$
α		-5.53° _a	± 19.21	$-27.06 \rightarrow 33.14$	22.61° _b	± 17.73	$0.72 \rightarrow 55.36$	48.11° _c	± 22.21	$19.52 \rightarrow 74.51$
OIV No.	618*	-		-	-		-	-		-
PS1-PS2		-		+17.95 → -15.59	-	± 7.03		-	± 6.69	
OIV No.	618*	5		$3 \rightarrow 7$	3		$3 \rightarrow 5$	3	I M (2000	$1 \rightarrow 5$

Morphological characteristics signed with *, ** were estimated with modifications compared to O.I.V. (2009a) descriptor list. Different letters indicate significant differences at the 5% level.

TABLE 1 (CONTINUED).

Ampelometric data for hand (HAND), mechanically (MECH) and minimally (MIN) pruned *Vitis vinifera* L. cv. 'Sauvignon blanc' and 'Syrah' vines, with the relevant O.I.V. coding.

Charac teristic	Unit/ OIV code	'Syrah'								
		HAND				ME	СН	MIN		
		Average	St. dev.	Min → Max	Average	St. dev.	Min → Max	Average	St. dev.	Min → Max
SLA	cm ²	231.19 _c	± 56.75	$158.52 \rightarrow 322.75$	160.84 _b	± 20.39	$118.83 \rightarrow 194.58$	88.61 _a	± 34.96	56.81 → 165.75
OIV No.	-	-		-	-		-	-		-
N1	mm	128.98 _c	± 22.52	$93.93 \rightarrow 167.61$	109.30 _b	± 9.10	$94.46 \rightarrow 121.68$	81.96 _a	± 11.49	$62.85 \rightarrow 98.94$
OIV No.	601	5		$3 \rightarrow 7$	3		$3 \rightarrow 5$	1		$1 \rightarrow 3$
P	mm	124.65 _b	$\pm\ 20.80$	$98.64 \rightarrow 165.67$	107.58 _b	± 11.65	$89.11 \rightarrow 119.48$	68.07_{a}	$\pm~15.03$	53.74 105.69
OIV No.	-	-		-	-		-	-		-
P/N1	ratio	$0.97_{\rm b}$	± 0.14	$0.65 \rightarrow 1.12$	0.98_{b}	± 0.10	$0.82 \rightarrow 1.11$	0.82_{a}	± 0.11	$0.69 \rightarrow 1.11$
OIV No.	093	5		$1 \rightarrow 5$	5		$3 \rightarrow 5$	3		$1 \rightarrow 5$
N2-1	mm	$108.76_{\rm b}$	$\pm~16.04$	$78.28 \rightarrow 130.98$	94.71 _b	± 5.59	$88.05 \rightarrow 106.85$	66.29 _a	± 9.59	$54.42 \rightarrow 84.01$
OIV No.	602	5		$3 \rightarrow 7$	3		$3 \rightarrow 5$	1		$1 \rightarrow 3$
N2-2	mm	111.02 _c	\pm 18.25	$89.09 \rightarrow 152.6$	93.68 _b	$\pm~11.40$	$75.58 \rightarrow 112.8$	68.97 _a	±16.88	$49.48 \rightarrow 102.96$
OIV No.	602	5		$3 \rightarrow 9$	3		$3 \rightarrow 5$	1		$1 \rightarrow 5$
S-1	mm	63.57 _b	± 11.46	$46.88 \rightarrow 83.41$	58.20 _b	± 7.51	$46.96 \rightarrow 68.97$	45.95 _a	± 9.20	$35.29 \rightarrow 64.85$
OIV No.*	605	5		$3 \rightarrow 7$	3		$3 \rightarrow 5$	3		$1 \rightarrow 5$
S-2	mm	66.68 _b	$\pm~17.03$	$40.4 \rightarrow 105.59$	$60.60_{\rm b}$	\pm 7.96	$50.2 \rightarrow 74.42$	44.6 _a	± 13.35	$29.14 \rightarrow 74.95$
OIV No.*	605	5		$3 \rightarrow 9$	5		$3 \rightarrow 5$	3		$1 \rightarrow 5$
N2-1/S-1	ratio	1.74 _a	$\pm~0.38$	$1.22 \rightarrow 2.46$	1.64 _a	± 0.18	$1.34 \rightarrow 1.94$	1.45 _a	± 0.13	$1.29 \rightarrow 1.66$
OIV No.	094**	3		$3 \rightarrow 5$	3		$3 \rightarrow 5$	3		3
N2-2/S-2	ratio	1.71 _a	$\pm~0.26$	$1.44 \rightarrow 2.20$	1.55 _a	± 0.13	$1.33 \rightarrow 1.75$	1.57 _a	± 0.21	$1.08 \rightarrow 1.83$
OIV No.	094**	3		$3 \rightarrow 5$	3		$3 \rightarrow 5$	3		$1 \rightarrow 5$
N3-1	mm	80.10_{b}	$\pm~8.05$	$68.75 \rightarrow 89.36$	71.35 _b	± 6.44	$57 \rightarrow 79.53$	47.83 _a	± 11.60	$32.96 \rightarrow 68.32$
OIV No.	603	5		$5 \rightarrow 7$	5		$3 \rightarrow 5$	3		$1 \rightarrow 5$
N3-2	mm	81.62 _b	± 9.10	$68.51 \rightarrow 92.29$	72.91 _b	± 9.58	$58.39 \rightarrow 91.22$	49.68 _a	$\pm~12.02$	$37.89 \rightarrow 75.3$
OIV No.	603	5		$5 \rightarrow 7$	5		$3 \rightarrow 7$	3		$1 \rightarrow 5$
PS-1	mm	37.45 _b	$\pm~8.95$	$23.72 \rightarrow 56.44$	27.12 _a	±4.46	$20.99 \rightarrow 37.37$	20.74 _a	± 5.94	$11.57 \rightarrow 33.12$
OIV No.	-	-		-	-		-	-		-
PS-2	mm	35.13 _b	± 6.78	$25.28 \rightarrow 44.75$	25.82 _a	± 7.61	$16.68 \rightarrow 37.16$	22.13 _a	± 6.63	$13.71 \rightarrow 32$
OIV No.	-	-		-	-		-	-		-
δ-1	degree	45.19° a	± 5.93	$35.48 \rightarrow 53.01$	38.78° a	$\pm~10.17$	$16.21 \rightarrow 48.67$	41.53° a	± 6.69	$29.69 \rightarrow 51.32$
	607	3		$3 \rightarrow 5$	3		$1 \rightarrow 5$	3		$1 \rightarrow 5$
δ-2	degree	45.57° a	± 9.39	$28.33 \rightarrow 62.31$	43.7° a	± 5.84	$34.76 \rightarrow 51.13$	42.27° a	\pm 8.24	29.34 →51.25
OIV No.	607	3		$1 \rightarrow 7$	3		$3 \rightarrow 5$			$1 \rightarrow 5$
γ-1	degree	60.62°	± 5.13	$54.87 \rightarrow 73.17$	56.09° a	± 7.13		57.07° a	± 8.99	$40.15 \rightarrow 72.31$
OIV No.	608	7		$5 \rightarrow 9$	7		$5 \rightarrow 7$	7		$3 \rightarrow 9$
γ-2	degree	62.11° a	± 3.79	$55.98 \rightarrow 70.45$	54.6° a	\pm 8.89	$37.97 \rightarrow 66.59$	58.89° a	± 7.02	$49.9 \rightarrow 67.58$
OIV No.	608	7		$5 \rightarrow 9$	5		$3 \rightarrow 7$	7		$5 \rightarrow 7$
β-1				$58.27 \rightarrow 94.22$	77.2° _a	± 11.51	$59.34 \rightarrow 97.14$	70.53° a	\pm 17.43	$51.19 \rightarrow 101.76$
OIV No.	610	9		$7 \rightarrow 9$	9		$7 \rightarrow 9$	9		$5 \rightarrow 9$
				48.64 - 86.68						
OIV No.	610	7		$5 \rightarrow 9$	9		$7 \rightarrow 9$	7		$3 \rightarrow 9$
α				$-39.25 \rightarrow 28.99$						
OIV No.	618*	-		-	-		-	-		-
PS1-PS2	mm	$+0.83_{a}$	$\pm~16.45$	+29.25 → - 15.86	-6.76 _b	± 4.02	$-0.76 \rightarrow -13.35$	-8.54 _b	± 5.46	$-1.73 \rightarrow -17.3$
OIV No.				<i>3</i> → <i>7</i>			$3 \rightarrow 5$	5		<i>3</i> → <i>5</i>

Morphological characteristics signed with *, ** were estimated with modifications compared to O.I.V. (2009a) descriptor list. Different letters indicate significant differences at the 5% level.

one to four categories in the O.I.V. classification, within one cultivar under the same treatment. The lowest within-vineyard variability was observed in the depths of upper sinuses in both cultivars, while the highest variability occurred in the case of angles between the veins and the opening of the petiole sinus. According to the data of the 18 O.I.V. descriptors monitored in the six sample sets, 98% of the characteristics proved to be variable and only 2% were stable.

In this study, the number of shoots correlated negatively with SLA. This can be explained by the increasing number of shoots per root system that decreased the vigour per shoot. The HAND and MECH treatments had almost the same effect on the leaves, while the MIN treatment changed the SLA and lengths of the veins significantly. According to Niinemets and Fleck (2002), not only the size of the lamina is varied by modified conditions such as radiation, but the petiole properties are also affected. In this study, petiole length was decreased significantly by the MIN treatment, both in absolute value and relative to the main vein. The results parallel the observations reported by Miller *et al.* (1997), Poni *et al.* (2000) and Intrieri *et al.* (2001), namely that the SLA is sensitive to bud load and microclimate.

On the basis of change in bud load, the leaf characteristics of 'Syrah' and 'Sauvignon blanc' could be divided into two groups, namely variable and stable. Ortiz *et al.* (2004) grouped (leaf) morphological characteristics into three groups in their sample set. Symbols are used to compare those characteristics with the O.I.V. (2009a) descriptor list and our present study (#: OIV; ##: present study). The following characteristics were grouped:

- non-discriminant characteristics with the same expression in all samples (not listed);
- discriminant characteristics: W × H; L2/L (N3/N1#); L3/L (N4/N1#), L4/L (N5/N1#); d (opening of the petiole sinus, PS1-PS2); ^a + ^b (O.I.V. 607 + O.I.V. 608#; δ+γ##); ^a + ^b + ^c (O.I.V. 607 + O.I.V. 608 + O.I.V. 609#); ^a + ^b + ^t (O.I.V. 607 + O.I.V. 608 + O.I.V. 610#; δ+γ+β##); ^a + ^t (O.I.V. 607 + O.I.V. 610#; δ+β##);
- stable or objective characteristics appropriate for clear identification: shape of blade (O.I.V. 067), number of lobes (O.I.V. 068), anthocyanin coloration of the main veins on the upper side of the blade (O.I.V. 070), presence of teeth in the petiole sinus (O.I.V. 081-1), naked petiole sinus (O.I.V. 081-2), shape of upper leaf sinuses (O.I.V. 082), shape of the base of the upper leaf sinuses (O.I.V. 083-1), presence of teeth at the base of the upper leaf sinuses (O.I.V. 083-2) and density of erect hairs on the petiole (O.I.V. 091).

According to the classification of the morphological characteristics reported by Ortiz *et al.* (2004), ampelographic patterns used in our present study could be classified into discriminant characteristics with two sub-groups: stable and variable. It has to be mentioned that the grouping of variables as "stable" or "variable" depends on the sample set, the environmental circumstances and the treatment applied.

The results proved that stable characteristics (angles between the veins, depth of sinuses) could serve as

descriptors, whereas variable properties (SLA and length of veins and petioles) may depend on cultivation practices. This was supported by metrical and statistical analyses, thus providing objective conclusions on the usefulness of leaf features in cultivar identification and on the impact of cultivation.

This study provided a metrical description of the grapevine leaf and its variability, as affected by cultivation and ecological parameters. We have shown that the morphological identification of the grapevine according to the leaf should be clarified with a consideration and clear indication of at least the pruning method and bud load. It is proposed that cultivation practices should be detailed thoroughly whenever a morphological identification of the grapevine is attempted.

CONCLUSIONS

'Sauvignon blanc' and 'Syrah' leaves collected from minimally pruned, mechanically pruned and hand-pruned vines were compared using 22 ampelometric features. According to the analysis of within-vineyard variability, it can be concluded that foliometric characteristics of both of the cultivars were highly variable within treatments. Further experimentation on a large scale is required to clarify the impact of within-vineyard variability on foliometric characteristics. In the case of the ampelometric variability caused by pruning level/bud load, it can be concluded that the morphological characteristics of the leaves can be divided into two groups, viz. stable (angles between the veins, depths of sinuses) and variable (single leaf area and lengths of veins and petioles). Stable characteristics could serve as descriptors of varietal identification, whereas variable properties may depend on cultivation practices. Ampelometric (foliometric) analyses are objective and highly valuable in the assessment of genetically determined morphological characteristics influenced by environmental factors and cultivation practices.

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