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Physicochemical Characteristics of the Maltese Grapevine Varieties – Ġellewża and Girgentina

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Abstract

Two indigenous Maltese grape vine varieties (cv. Ġellewża and Girgentina) juice extracts were studied for their physicochemical properties at three different locations on the Island of Malta. The mean acidity for Ġellewża and Girgentina was 2.729±0.088 and 3.971±0.179 g/L, pH was 4.026±0.039 and 3.704±0.042 and a %Brix was 17.913±0.364 and 17.531±0.189, indicating similarities between the variety-location combinations. Spectroscopic analysis revealed significant difference between the two varieties. The Ġellewża variety exhibited a high colour index (3.055-10.774) while the Girgentina variety showed a high tonality ratio (2.656-3.111). Although, the total polyphenolic content of the two varieties was not significant differently in most cases (754.771-2643.552 mg/kg), the red grape Ġellewża had significantly higher anthocyanin content (708.236±68.451 mg/kg) compared to the white grape Girgentina (14.412±1.119 mg/kg). Principal component analysis confirmed the differences between the varieties and also exhibited distinctive location differences, based on their physicochemical characteristics.

Keywords: *Vitis vinifera* varieties, Ġellewża, Girgentina, physicochemical characteristics

1. Introduction

The dark-skinned indigenous Ġellewża grape variety has been utilised by traditional wine makers to produce a red wine. It has been also transformed into still and semi-sparkling rosé wines. Wine-makers blend this variety with Syrah, softening the spiciness of the latter wine and at the same time adding a cherry flavour. A typical *passito* is produced by drying the berries in the sun, to intensify the sugars, prior to vinification. The resultant wine is spicier with a more pungent cherry flavour and earthy undertones. On the other hand, the white indigenous Girgentina grape variety produces a crispy and fruity wine. It is usually blended with Chardonnay, to produce a smooth wine with a fruity aroma and buttery undertones (Marsovin, 2008; Delicata, 2013).

The berries of international varieties have been distinctively studied for their physicochemical characteristics. Such characteristics include proteins (Sarry et al., 2004), proanthocyanidins (Czochanska et al., 1979; Souquet et al., 1996; Sun et al., 1998), anthocyanins (Revilla et al., 2001), organic acids (Conde et al., 2007) and sugars (Liu et al., 2006). Attention has also been drawn towards the health benefits derived from the consumption of grapes and their products. These phytochemicals contribute to the antioxidant (Tamura & Yamagami, 1994; Wang et al., 1997; Orak, 2007), cardioprotective (Sato et al., 1999; Xia et al., 2010), and anticancer (Ye et al., 1999; Singletary & Meline, 2001) activities amongst others.

In this study, we attempted to determine the quality of the Maltese grapevine varieties Ġellewża and Girgentina, grown in different regions of Malta, through physicochemical characteristics, including the total acids, sugar content, pH and polyphenolic parameters.

2. Materials and Methods

2.1 Materials

The berries of two Maltese indigenous varieties, Ġellewża and Girgentina, were collected between 30th August and 21st September 2011 every two days, from different vineyards. Ġellewża grape samples were collected from Mġarr (two sites) and Burmarrad while Girgentina grape samples were collected from Siġġiewi and Mġarr (two sites).

2.2 Extraction of Grape Samples

For the total acidity, Brix and pH analyses, the fresh grape samples were crushed in a large beaker, and the filtered juice was then analysed. For the polyphenolic analyses, approximately 5 g fresh grape aliquots were extracted with 35 ml methanol:HCl (1M) (95:5). The samples were placed in an ultrasonicator for 15 minutes followed by 15 minutes centrifugation at 3000rpm. The samples (supernatants) were ready for analysis. All samples were collected in triplicates and all tests were conducted in triplicates, for a total replicate number of nine.

2.3 Titratable Acidity, Brix and pH Determination

The titratable acidity was measured by using NaOH 0.1 N and bromothymol blue as indicator. The volume of titrant was then multiplied by 0.75 and then divided by two to determine the amount of tartaric acid in g/L as titratable acidity. The pH was measured using a Thermo Scientific Orion 4-Star pH meter (USA), while the %Brix content was determined using a Hanna Instruments HI 96814 wine refractometer (USA).

2.4 UV-Vis Analysis and Folin-Ciocalteu Colorimetric Method

Each grape extract was analysed with a Lightwave II – WPA UV-Vis spectrophotometer (UK), between 200 and 800nm (wave scans) and specifically at 280 nm, 420 nm, 520 nm and 620 nm. The Ġellewża and Girgentina extracts were generally in a 1:9 and 1:4 ratio with solvent, respectively. The colour intensity, % tint, tonality ratio (Glories, 1984) and anthocyanin content were determined according to the following equations

$$\text{Colour intensity} = (A_{420}.\text{DF}) + (A_{520}.\text{DF}) + (A_{620}.\text{DF}) \quad (1)$$

$$\text{Percentage Tint} = \frac{A_{420}}{A_{520}} \times 100 \quad (2)$$

$$\text{Tonality Ratio} = \frac{A_{420}}{A_{520}} \quad (3)$$

$$\text{Anthocyanin content(mg/kg)} = \frac{1000.V_S.\text{DF}.A_{520}}{\epsilon} \quad (4)$$

Where, V_S = volume of extracted sample per gramme of grape; DF = dilution factor; ε = extinction coefficient [58.3 ml (mg.cm)]; A420, A520, A620 = absorbance values at 420, 520 and 620 nm.

The test for polyphenols, using the Folin-Ciocalteu reagent, was conducted according to Attard (2013). Briefly, to 5 µl of each grape extract, 5 µl of distilled water, 100 µl of Folin-Ciocalteu reagent (Sigma-Aldrich, pre-diluted with distilled water (1:9)) and 80 µl of sodium carbonate Na_2CO_3 (Sigma-Aldrich, 1M) were added. The absorbance was read after 20 min at room temperature at 630 nm using a micro-plate reader (BioTek ELx800, Winooski, VT, USA). Tannic acid (Sigma-Aldrich) was used as standard (60-960 mg/ml; $r^2 = 0.9940$) to prepare the calibration curve in order to determine the total amount of phenols in each grape extract (mgTAE/kg of grape extract).

2.5 Data Analysis

The results for the samples, collected over the three-week period, were averaged. The titratable acidity, pH, %Brix, colour intensity (CI), tonality ratio, anthocyanin content, polyphenolic content, anthocyanin to polyphenolic ratio were investigated with multivariate analysis for all berry samples. The correlation matrix was calculated, giving the correlation coefficients between each pair of variables tested. To identify variability and to reduce the dimensions of the data set, principal component analysis (PCA) was performed, using the XLSTAT Version 2011.5.01 software (Addinsoft, USA).

3. Results and Discussion

3.1 Titratable Acidity, pH and %Brix

In previous studies, it has been observed that different physicochemical parameters are affected by genetic variability and environmental factors (Serrano-Megias et al., 2006; Makris et al., 2006). In this study we report the differences observed for the physicochemical characteritics for the two grapevine varieties wth different environmental conditions. The results for the titratable acidity, pH and %Brix are exhibited in Table 1. In particular, acidity values for Girgentina were superior to those of Ġellewża and while the opposite was observed for the pH values. Furthermore, samples collected from Mġarr have shown that grapes from the old vines have a higher acidity compared to the grapes of the new vines ($p < 0.05$). The total acidity results of the Ġellewża and Girgentina also showed to be much lower compared to Cabernet Sauvignon, Malbec, Bonarda, Merlot and Tempranillo grape varieties (Fanzone et al., 2012), i.e. less than 4.63 g/L as opposed to values greater than 5.3 g/L, respectively. Results also showed that the new Ġellewża and Girgentina grapevines in Mġarr have a higher pH than the old grapevines. Comparing the pH of Cabernet Franc, Merlot, Sangiovese and Syrah grape varieties (Gris et al., 2010) to the Ġellewża and Girgentina, it was observed that the pH of the local varieties is quite higher. In fact the pH of the international varieties were lower than 3.58 while those for the Maltese varieties the values ranged between 3.507 and 4.205. The Ġellewża grapevines located at Burmarrad exhibited the lowest sugar content (15.181±0.274%), while the new Ġellewża and Girgentina grapevines in Mġarr have shown the highest sugar content (19.458±0.347 and 18.801±0.607%, respectively). The old Ġellewża grapevines in Mġarr showed a higher sugar content than the old Girgentina grapevine (18.526±0.468 and 17.151±0.372%, respectively), while the Girgentina grapevines at Siġġiewi (17.033±0.345%) showed a similar sugar content to the old Girgentina grapevine at Mġarr. Compared to other grape varieties, including Alicante, Cabernet Franc, Cabernet Sauvignon, Carignan, Cinsault, Grenache, Merlot, Mourvedre, Sangiovese and Syrah (Gris et al., 2010; Jensen et al., 2008) both the Ġellewża and Girgentina varieties have a much lower sugar content. This was always considered as an important issue with local grapevine varieties.

Table 1. Total acidity, pH and %Brix for the six variety-location combinations

		Titratable Acidity (g/L)	pH	%Brix
Ġellewża	Burmarrad	2.721±0.166	3.941±0.073	15.181±0.274
	Mġarr-New	2.536±0.187	4.205±0.108	19.458±0.347*
	Mġarr-Old	2.874±0.329	3.949±0.100	18.526±0.468*
Girgentina	Siġġiewi	3.700±0.351	3.716±0.104*	17.033±0.345
	Mġarr-New	3.445±0.280	3.956±0.091	18.801±0.607*
	Mġarr-Old	4.650±0.732*	3.507±0.084*	17.151±0.372

Note. Data are means±S.E.M. of three independent determinations. Means within an asterisk show a significant difference between variety-location for the individual parameters, at $P < 0.05$ by Student's *t*-test.

3.2 UV-Vis Analysis and Absorbance Values

Colour analysis was carried out by spectrophotometric methods (Gris et al., 2010). Although these parameters are usually studied in red grape varieties, similar to Ġellewża, these were also considered for Girgentina as a means to show distinctive differences between the two varieties. Percentage tint is the percentage ratio of the yellow components (proanthocyanidins) at 420 nm and the red components (anthocyanins) at 520 nm. The results for CI and Tonality are illustrated in Table 2. Results showed that the percentage tint is lower in Ġellewża grapes than in Girgentina. It is clearly indicated that although the anthocyanin content of Girgentina white grapes is low, the proanthocyanidin content is high. Consequently, the proanthocyanidin to anthocyanin percentage ratio of Ġellewża red grapes is lower than the Girgentina type. Another parameter related to percentage tint is the tonality. Tonality ratio is the classical ratio between the two wavelengths. As a result, Girgentina samples resulted in a higher tonality ratio compared to the Ġellewża samples. Colour intensity depends on pH and the content of anthocyanins. This reflects the content and structure of the anthocyanins present in both grapes and wine. In fact, Colour intensity featured distinctively with Ġellewża samples. Colour intensity is obtained by the addition of the absorbances at three wavelengths, i.e. 420, 520 and 620 nm (Glories, 1984). When comparing red grape varieties or wines, the more acidic the grape juice or wine, the brighter red the colour is (Mollah, 2011). Although Ġellewża samples showed higher colour intensities than Girgentina samples, the important parameter, in this study, was the content of anthocyanins. This is because the Ġellewża samples exihibited higher pH values than Girgentina samples, even though their colour intensities were higher. This may be due to the adaptability of the local varieties to the highly alkaline and calcareous Maltese soil.

In the measurement of the anthocyanin content, the absorbance value at 520nm is taken into consideration. Since anthocyanins have a direct influence on the red colour of the grapes, total anthocyanin content in Ġellewża samples were higher than in Girgentina (Figure 1). The Mġarr Ġellewża anthocyanin content compares well with the Syrah anthocyanin content (808-1112 mg/kg) obtained by Barbagallo et al. (2011). Ġellewża grapes compared to other varieties such as Merlot, Cabernet Sauvignon and Vranec (Ivanova et al., 2010) possess a lower tint but a higher colour intensity, while on the other hand, Girgentina grapes showed a lower colour intensity with a higher tint.

Table 2. Colour Intensity and Tonality for the six variety-location combinations

		Colour Intensity(Abs)	Percentage Tint (%)	Tonality (ratio)
Ġellewża	Burmarrad	3.055±0.689*	38.7±4.5	0.387±0.045
	Mġarr-New	9.488±0.670*	26.2±0.8	0.262±0.008
	Mġarr-Old	10.774±1.298*	26.2±0.8	0.262±0.008
Girgentina	Siġġiewi	0.447±0.088	311.1±35.5*	3.111±0.355*
	Mġarr-New	0.467±0.056	304.5±24.3*	3.045±0.243*
	Mġarr-Old	0.663±0.141	265.6±25.1*	2.656±0.251*

Note. Data are means±S.E.M. of three independent determinations. Means within an asterisk show a significant difference between variety-location for the individual parameters, at $P < 0.05$ by Student's *t*-test.

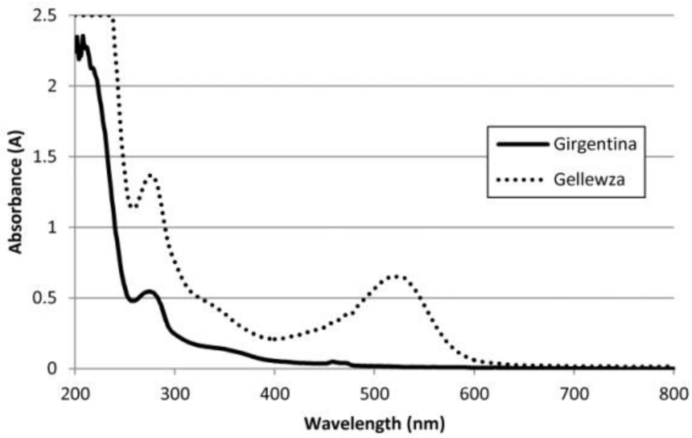

Figure 1. The UV-Vis profile for the Ġellewża and Girgentina grape varieties between 200 and 800 nm

3.3 Folin-Ciocalteu Test for Polyphenols

The determination of the polyphenolic content of Ġellewża and Girgentina grapes was carried out by using the Folin-Ciocalteu test, a commonly used spectrophotometric method in viticulture and oenology. The results for anthocyanins, polyphenols and their ratio are exhibited in Table 3. These have showed that Ġellewża and Girgentina samples collected from the old grapevines in Mġarr had the highest content of polyphenols. Compared to the other samples, grapes collected from Burmarrad possessed the lowest amount of polyphenols. Samples collected from the new grapevines in Mġarr showed that Ġellewża grapes contained a higher amount of polyphenolic compounds compared to Girgentina samples. The results also showed Girgentina from Siġġiewi to have less polyphenols than the other Girgentina grapes sampled from Mġarr, though the difference was not statistically significant.

Table 3. Anthocyanins, Polyphenols and their Ratio for the six variety-location combinations

		Anthocyanin content (mg/kg)	Polyphenolic content (mg/kg)	Anthocyanin:Polyphenols ratio
Ġellewża	Burmarrad	216.612±46.882*	754.771±207.066	0.287*
	Mġarr-New	803.092±78.727*	1853.426±178.299	0.433*
	Mġarr-Old	976.487±100.111*	2643.552±193.592*	0.369*
Girgentina	Siġġiewi	11.662±3.335	1021.562±276.026	0.011
	Mġarr-New	12.185±1.841	1447.374±286.619	0.008
	Mġarr-Old	19.032±3.468	1818.667±304.349	0.010

Note. Data are means±S.E.M. of three independent determinations. Means within an asterisk show a significant difference between variety-location for the individual parameters, at $P < 0.05$ by Student's *t*-test.

The amount of polyphenols found in Ġellewża samples collected from Burmarrad was compared to the Cinsault variety with a value of 876 mg/kg. The polyphenolic content of both the Ġellewża and Girgentina collected from the new grapevines in Mġarr, and the Ġellewża collected from the old grapevines resulted in a much higher concentration compared to the popular varieties Cabernet Sauvignon, Grenache, Merlot and Syrah. Girgetina sampled from Siġġiewi had similar polyphenolic content to the Carignan variety with a value of 1210 mg/kg (Jensen et al., 2008).

It was observed from the scree plot (data not shown) that the first three principal components accounted for 98.68 % of the total variance. This indicates that little numerical noise and/or experimental error is observed. However, the parameters studied fall within the first two principal components, which in fact contributed significantly to the total variance. The loadings plot provides the direction of each original variable, and the scores plot, indicates the position of each variety/location combination. The first factor is loaded heavily on titratable acidity, pH, colour intensity, tonality, anthocyanin content and anthocyanin/polyphenolic ratio, representing a mix of physical and chemical parameters. The second factor is heavily loaded on %brix and polyphenlic content, representing the main categories of phytochemicals in grapes. The titratable acidity and tonality showed an inverse correlation with anthocyanin/polyphenol ratio and pH. There is no direct relationship between titratable acidity and pH for grape extracts, although generally the pH decreases as the acid increases and vice-versa. The latter two parameters did not show any strong correlation with the other parameters (Figure 2A).

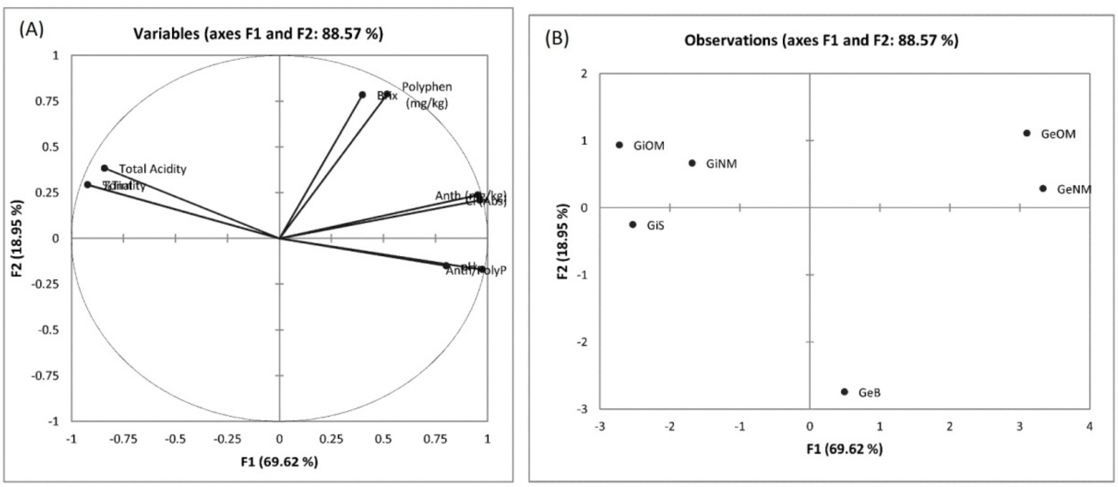

Figure 2. Graph of loading plot (A) of physicochemical parameters and scores plot (B) of six variety-location combinations: *Ġellewża* Burmarrad, Mġarr-New, Mġarr-Old (GeB, GeNM and GeOM, respectively) and *Girgentina* Siġġiewi, Mġarr-New, Mġarr-Old (GiS, GiNM and GiOM, respectively

The score plot reported in Figure 2B shows the physicochemical parameters of grape extracts in the space of the two new variables F1 and F2. Moving along F1 from left to right in the graph, we observed that the Girgentina grape extracts exhibited siginificantly different physicochemical characteristics from the Ġellewża grape extracts. Moving along F2 from top to bottom, the Mġarr-grown Ġellewża and Girgentina vines exhibited superior sugar and polyphenolic contents, as compared to those grown at Burmarrad and Siġġiewi.

4. Conclusion

These results show that the two Maltese grapevine varieties, Ġellewża and Girgentina, are distinct from other international grapevine varieties, based on their physicochemical characteristics. There were also remarkable differences between the Ġellewża and Girgentina, which also are reflected in the locality, hence environmental differences. The high polyphenolic content in both grape varieties, indicates that the grape products may be valued for their nutritional properties, apart from the production of wine.

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