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TECHNICAL NOTE

AMINO ACIDS PROFILE OF TWO GALICIAN WHITE GRAPEVINE CULTIVARS (GODELLO AND TREIXADURA)

NOTA TÉCNICA

PERFIS DE AMINOÁCIDOS DE DUAS VARIEDADES GALEGAS BRANCAS DE VIDEIRA (GODELLO AND TREIXADURA)

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SUMMARY

Amino acids constitute a source for yeast metabolism and, thus are of paramount importance to wine flavour and aroma. Their concentrations in must depend greatly on grape variety. The aim of this work was to describe the amino acids profile of two white grapevine varieties: 'Godello' and 'Treixadura', produced in Galicia (NW Spain). The study was carried out over two consecutive vintages (2012 and 2013). Amino acids profiles of the musts were determined using HPLC. Although the musts from the two varieties presented common major and minor amino acids, the greatest contents were observed for 'Treixadura'. 'Godello' and 'Treixadura' varieties were arginine-accumulators, since this was the amino acid found at the highest level in the musts from the two cultivars. Amino acids contents were higher in 2012 than in 2013, likely due to climate differences between years.

RESUMO

Os aminoácidos constituem uma fonte para o metabolismo das leveduras e são de suma importância para o sabor e o aroma do vinho. As suas concentrações no mosto dependem grandemente da variedade de videira. O objetivo deste trabalho foi descrever o perfil de aminoácidos de duas variedades brancas de videira: 'Godello' e 'Treixadura', produzidas na Galiza (Noroeste de Espanha). O estudo foi realizado em duas vindimas consecutivas (2012 e 2013). Os perfis de aminoácidos dos mostos foram determinados utilizando HPLC. Os mostos das duas variedades apresentaram maiores e menores aminoácidos comuns, sendo observadas maiores concentrações na 'Treixadura'. No mosto das variedades 'Godello' e 'Treixadura' o aminoácido predominante foi a arginina. As concentrações de aminoácidos foram maiores em 2012 do que em 2013, provavelmente devido a diferenças climáticas entre anos.

Key words: amino acids, Godello, Treixadura, vintage.
Palavras-chave: aminoácidos, Godello, Treixadura, colheita.

INTRODUCTION

Amino acids present in musts act as nutrients required for growth and development of yeasts during alcoholic fermentation. Some of them are precursors of many compounds that contribute to wine aroma: higher alcohols, aldehydes, ketones and esters (Swiegers *et al.*, 2005; Moreno-Arribas and Polo, 2009; Styger *et al.*, 2011).

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Many factors affect the amino acid composition of musts, mainly the grape cultivar used, weather conditions and viticultural practices (Hernández-Orte *et al.*, 1999; Lee and Schreiner, 2010; Garde-Cerdán *et al.*, 2014; Ortega-Heras *et al.*, 2014). Grape variety is the most important determinant of the type and concentrations of amino acids that accumulate in the grape berry (Kliewer, 1970; Stines *et al.*, 2000). When other factors are held comparatively constant, climate is the dominant factor that influences berry and wine quality (Storchmann, 2005; Makra *et al.*, 2009). Of the climate variables, temperature has been recognised as a primary driver of vine growth and berry/wine composition (Winkler, 1974; Soar *et al.*, 2008). In hot and dry climates, grape maturation is rapid and the concentrations of different compounds in the berries is greater than in more humid and cool climates. Moreover, a dilution effect occurs in rainy years, affecting the organoleptic and sensory properties of the musts since the compounds accumulated in the grapes have been diluted (Barmuud *et al.*, 2014).

Despite the wide range of factors affecting the amino acid composition in grapes and wines, some studies have successfully used this amino acid pattern to differentiate wines according to variety and vintage (Soufleros *et al.*, 2003; Herbert *et al.*, 2005; Martínez-Pinilla *et al.*, 2013). Therefore, the importance of using free amino acids profiles to characterize musts from minority varieties in different regions would allow to characterize wines with their own personality and different from the rest that exist in the international market.

In Galicia (NW Spain), white grapevine (*Vitis vinifera* L.) cultivars are mainly grown. Among them, 'Godello' and 'Treixadura' represent a relevant area of cultivation. 'Godello' provides intense taste to wines, but it possesses a lower aromatic potential than 'Albariño' (Versini *et al.*, 1994). 'Treixadura' is one of the most important white grapevine cultivars in Galicia (Blanco *et al.*, 2012). It is aromatic, giving fruity, floral and balsamic notes to the wines.

Some important components of wine aroma depend on the amino acids composition of the grapes and this has never been described for these cultivars. Therefore, the aim of the present study was to assess the amino acids profiles of the musts from two white grapevine (*Vitis vinifera* L.) cultivars which are traditionally cultivated in Galicia: 'Godello' and 'Treixadura'. This is the first study that determines the amino acid concentrations of musts from these cultivars.

MATERIAL AND METHODS

Samples

The grape samples were collected in two consecutive vintages (2012 and 2013) at the experimental farm of the Estación de Viticultura e Enoloxía de Galicia (EVEGA) in Leiro (Ourense, NW Spain) within the Ribeiro Designation of Origin (42º 21.62' N, 8º 7.02 W, elevation 110 m). Soil is sandy-textured with a low water storage capacity.

The studied vineyards (0.2 ha) were planted with 'Godello' and 'Treixadura' grapevine cultivars, native from Galicia. Plants were planted in 1998, grafted onto rootstock 196-17C and vertically shoot-positioned in a single cordon with 10-12 buds per plant. Spacing was 2.4 m x 1.25 m. In both vintages, grapes were harvested at their optimum degree of maturity, which was assessed by their soluble solids content.

Climate data were collected at a weather station located on-site. Yearly average temperatures were 13.08 ºC and 13.47 ºC in 2012 and 2013, respectively. Total yearly rainfall amounts were 841 mm and 1282 mm for 2012 and 2013, respectively. The average temperatures for the growing season (April to September) were 17.2 ºC and 17.9 ºC for 2012 and 2013, respectively. Total rainfall amount for the growing season was 352.8 mm in 2012 and 269 mm in 2013. The two years studied (2012 and 2013) showed different distribution in rainfall; in 2012, a very low amount of precipitation fell from January to March (51 mm), whereas in 2013 more than 500 mm fell during those three months. The growing season was more rainy in 2012; nevertheless, in August and September (when most metabolites are being concentrated in grapes) less rain fell in 2012 (63 mm) than in 2013 (109 mm). Moreover, higher monthly average temperatures were recorded in 2013; however, the differences in temperatures between both years were mainly observed in wintertime.

Grapes were harvested when they attained their optimum maturation, implying different dates for each cultivar. Approximately, three lots of 60 kg each were harvested for each variety, each corresponding to one field replication (Trigo-Córdoba *et al.*, 2015). Grapes were destemmed, crushed and pressed with a pneumatic press. Basic attributes of the must (pH, degrees Brix, titratable acidity, malic and tartaric acid contents) were determined following the official methodology (OIV, 2009). Must samples were processed following standard protocols for white winemaking (further details can be found in Trigo-Córdoba *et al.*, 2014) and wines were analyzed using official methods (OIV, 2009).

Chemical reagents

A Milli-Q equipment (Millipore, Bedford, MA, USA) was used for generating ultra-pure water. Super-gradient HPLC grade acetonitrile and methanol were purchased from Scharlau (Sentmenat, Spain). Ammonium chloride was from Merck (Germany). L-aspartic acid, L-glutamic acid, L-asparagine, L-serine, L-glutamine, L-histidine, glycine, L-threonine, L-arginine, L-α-alanine, γ-aminobutyric acid (GABA), L-proline, L-tyrosine, L-valine, L-methionine, cysteine, L-isoleucine, L-tryptophan, L-leucine, L-phenylalanine, L-ornithine hydrochloride, L-lysine, and diethylethoxymethylenemalonate (DEEMM) were from Acros Organics (New Jersey, USA). Solutions of amino acids were prepared with HCl 0.1 N.

Analytical methods

The determination of the amino acids present in the grapes was performed following the method proposed by Gómez-Alonso *et al.* (2007) and Garde-Cerdán *et al.* (2009), with slight modifications.

The derivatization of amino acids and ammonium was carried out by reaction of 1.75 mL of borate buffer 1 M (pH = 9), 0.75 mL of methanol, 1 mL of sample without any pre-treatment, 20 μL of internal standard (L-2-aminoadipic acid, 1 g/L), and 30 μL of DEEMM (diethylethoxymethylenemalonate) in a screw-cap tube over 30 minutes in an ultrasound bath. Then, the sample was heated at 70 ºC for 2 hours in order to allow for the complete degradation of the excess of DEEMM and reagent by-products. Determinations were carried out in triplicate for the three different samples (one from each field replication).

The HPLC analysis was performed using Agilent 1100 series equipment (Agilent Technologies, Palo Alto, CA, USA). Chromatographic separation was carried out in a Zorbax Eclipse AAA column (C18), particle size 5 μm (150 mm x 4.6 mm) thermostated at 22 ºC. A pre-column was also used (Zorbax Eclipse AAA, 12.5 mm x 4.6 mm).

Amino acids were eluted under the following conditions: 0.8 mL/min flow rate, 10% B during 20 min, then elution with linear gradients from 10% to 17% B in 10 min, from 17% to 19% B in 0.01 min, maintained during 0.99 min, from 19% to 19.5% B in 0.01 min, from 19.5% to 23% B in 8.5 min, from 23% to 29.4% B in 10.49 min, from 29.4% to 72% B in 8 min, from 72% to 82% B in 5 min, from 82% to 100% B in 4 min, maintained during 3 min, followed by washing and reconditioning the column. Phase A was 25 mM acetate buffer (pH 5.8) with 0.4 g of sodium azide; phase B was 80:20 (v/v) mixture of acetonitrile and methanol.

The injected volume was 50 μL. For detection, a photodiode array detector monitored at 280 and 269 nm was used for amino acids and ammonium, respectively. In these conditions, 22 amino acids and the ammonium ion (Figure 1) were separated and identified according to the retention times and UV-vis spectral characteristics of the derivatives of the corresponding standards, and were quantified using the internal standard method. Quantification of amino acids was carried out immediately after the derivatization reaction in order to obtain reliable results of the amino acids concentrations in the must, since Gómez-Alonso *et al.* (2007) reported that proline concentrations were not stable over the time.

Statistical analysis

A two-way ANOVA mixed effects model was used for assessing the effects of grape cultivar (fixed factor) and vintage (random factor) for the general attributes of the musts and for the amino acids concentrations. A principal component analysis (PCA) was carried out using the seven amino acids found at the highest concentrations in the musts from the two cultivars studied, because they represented more than 70% of the total free amino acids contained in the must. Statistical analysis was carried out using R software version 2.15 (R Core Team, 2012).

RESULTS AND DISCUSSION

General attributes of the must

Differences among cultivars are reflected in must attributes such as titratable acidity, pH and tartaric acid content, being musts from 'Treixadura' the least acidic ones (Table I).

Vintage exerted a significant effect on soluble solids content and titratable acidity of the must. However, there were no significant interactions between cultivar and vintage for the must attributes considered (Table I).

In addition, lower values of titratable acidity were observed in 2013 compared to 2012 for both cultivars. These differences were caused by a reduction in tartaric acid content in the case of 'Godello' (Table I).

Amino acids profiles

'Treixadura' presented higher contents in glutamic acid and glutamine than 'Godello'. In fact, all the amino acids determined in this experiment were significantly different between cultivars (Table II).

Quantitatively, 'Treixadura' musts presented higher total amino acids contents in both vintages (Table II). Vintage exerted a significant influence on the concentrations of all the amino acids except for aspartic acid, glycine, proline and ornithine. The interaction between cultivar and vintage was significant for aspartic acid, asparagine, glutamine, glycine, arginine, GABA, tyrosine and cysteine (Table II).

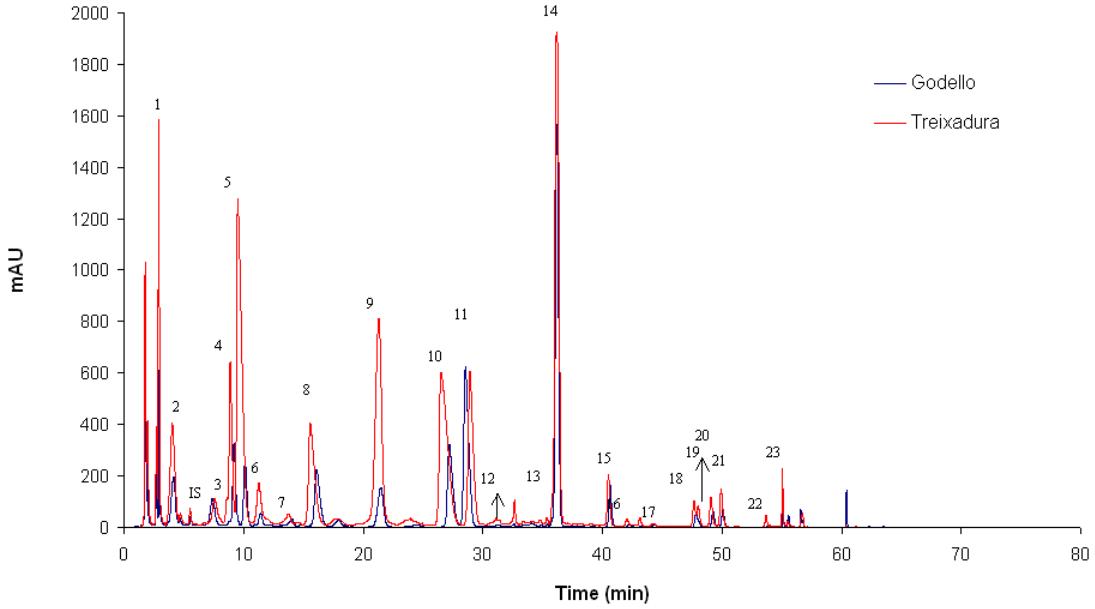

Figure 1. Chromatogram of the amino acids profiles of 'Godello' and 'Treixadura' must samples. Peak numbers correspond to the order of the compounds shown in Table II (IS indicates internal standard).

Cromatograma dos perfís de aminoácidos dos mostos de 'Godello' e 'Treixadura'. Os números nos picos correspondem à ordem dos compostos apresentados no Quadro II (IS indica padrão interno).

Table I

General attributes of the musts from the two studied cultivars and the two vintages surveyed

Atributos gerais dos mostos das duas variedades e das duas vindimas estudadas

Cultivar	Vintage	Attribute				
		Soluble solids (º Brix)	Titratable acidity (g/L tartaric acid)	pH	Malic acid (g/L)	Tartaric acid (g/L)
Godello	**2012**	22.3 a	7.7 b	3.2 a	2.5 a	8.7 a
	2013	24.0 b	6.3 a	3.3 a	2.3 a	8.0 a
Treixadura	**2012**	22.1 a	5.6 a	3.4 a	2.8 a	5.5 a
	2013	25.0 b	5.1 a	3.6 b	2.5 a	6.3 a
Cultivar		ns	**	**	ns	***
Vintage		*	*	ns	ns	ns
Cultivar x Vintage		ns	ns	ns	ns	ns

Different letters next to an attribute indicate significant differences between years for a given variety ($p < 0.05$).
For the factorial analysis: ns = non-significant; * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$.

Table II

Amino acids and ammonium contents (mg/L) in the musts from two Galician white grapevine cultivars and the two vintages surveyed

Concentração de aminoácidos e ião amónio (mg/L) nos mostos das duas variedades galegas brancas de videira e das duas vindimas estudadas

Compound	Godello		Treixadura		Cultivar	Vintage	Cultivar x Vintage
	2012	2013	2012	2013			
Aspartic acid	24.24 a	39.72 b	55.46 b	36.22 a	***	ns	***
Glutamic acid	46.43 a	84.48 b	54.92 a	103.27 b	*	***	ns
Asparagine	4.91 b	1.92 a	12.80 b	5.74 a	***	***	*
Serine	33.65 b	24.50 a	49.75 b	40.82 a	***	***	ns
Glutamine	57.42 b	34.76 a	201.03 b	113.32 a	***	***	**
Histidine	17.58 b	8.06 a	31.04 b	15.24 a	***	***	ns
Glycine	4.45 b	2.42 a	3.99 a	5.22 b	***	ns	***
Threonine	58.99 b	34.59 a	77.82 b	61.15 a	***	***	ns
Arginine	175.72 b	62.11 a	252.39 a	222.11 a	***	***	*
Alanine	67.50 b	34.07 a	127.40 b	92.66 a	***	***	ns
γ-Aminobutyric acid (GABA)	92.29 b	29.54 a	68.12 b	38.54 a	**	***	***
Proline	1.74 a	3.55 b	5.07 a	4.70 a	***	ns	*
Tyrosine	4.13 b	2.66 a	12.89 b	4.80 a	***	***	***
Ammonium chloride	88.60 a	134.08 b	141.52 a	216.89 b	***	***	***
Valine	16.89 b	13.58 a	20.29 a	19.17 a	***	ns	ns
Methionine	1.89 a	1.48 a	3.72 b	2.08 a	***	**	ns
Cysteine	2.27 a	2.13 a	2.14 a	2.75 b	***	**	***
Isoleucine	7.77 b	5.55 a	14.43 a	10.19 a	***	**	ns
Tryptophan	7.36 b	5.47 a	18.29 b	13.46 a	***	***	ns
Leucine	10.29 b	5.87 a	13.87 b	10.06 a	***	***	ns
Phenylalanine	8.98 a	7.81 a	26.54 a	23.63 a	***	*	ns
Ornithine	0.62 b	0.44 a	1.94 a	1.65 a	***	ns	ns
Lysine	3.74 b	2.07 a	5.90 a	4.68 a	***	***	ns
Sum of amino acids	648.86 b	406.76 a	1059.79 b	831.46 a	***	***	ns

Different letters next to the amino acid concentration value indicate significant differences between years for a given variety ($p < 0.05$).
For the factorial analysis: ns = non-significant; * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$.

Six amino acids represented 69 - 76% of the total amino acids contents for the musts of the two considered cultivars: arginine, alanine, threonine, GABA, glutamine and glutamic acid. The most abundant one was arginine, representing approximately 25% of the total amino acids content in the musts (Figure 2). However, the percentage of these amino acids varied from year to year and glutamic acid was the most abundant amino acid in 'Godello' musts from 2013 instead of arginine (Figure 2), whereas the percentage of glutamine was greater in 2012 for this cultivar.

Vintage exerted a significant effect on most of the amino acids considered in this study (Table II). 'Godello' musts presented significant differences in amino acids contents between years except for three amino acids, whereas 'Treixadura' musts did not show differences between years for seven amino acids (Table II). The contents in ammonium ion were significantly higher in 2013 for both varieties.

'Godello' musts presented three amino acids with a similar content in 2012 and 2013 (methionine, cysteine and phenylalanine). Three amino acids appeared in greater contents in musts from 2013 (aspartic acid, glutamic acid and proline). The remaining 16 amino acids presented higher contents in musts from 2012 (Table II).

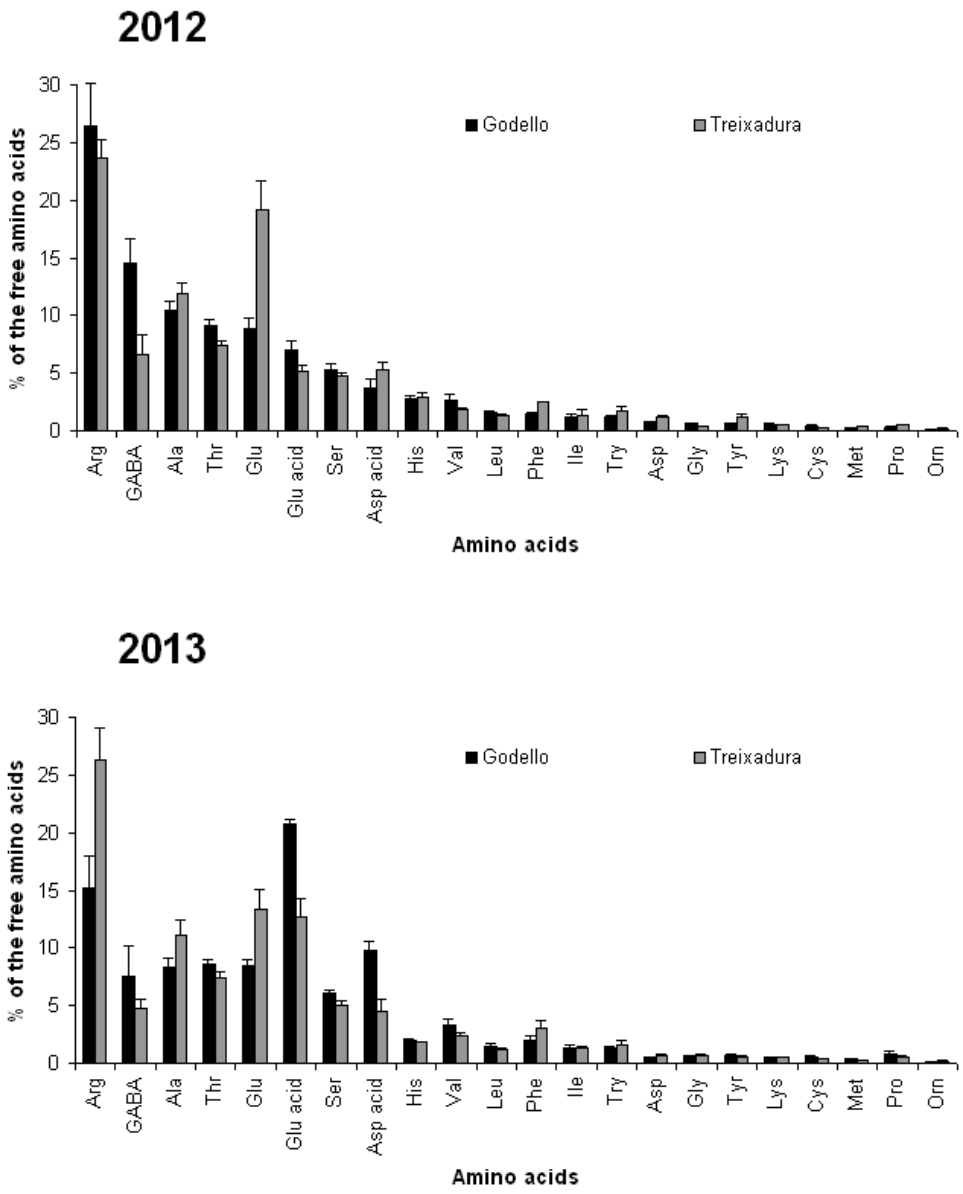

Figure 2. Percentage of each amino acid over the total amino acids in 'Godello' and 'Treixadura' musts for the 2012 and 2013 vintages. Abbreviations: Arg (arginine), GABA (γ-aminobutyric acid), Thr (threonine), Ala (alanine), Ser (serine), Glu (glutamine), Glu acid (glutamic acid), Val (valine), Phe (phenylalanine), Leu (leucine), His (histidine), Asp acid (aspartic acid), Ile (isoleucine), Try (tryptophan), Tyr (tyrosine), Asp (asparagine), Lys (lysine), Cys (cysteine), Pro (proline), Gly (glycine), Orn (ornithine), Met (methionine).

Percentagem de cada aminoácido nos mostos de 'Godello' e 'Treixadura' nas vindimas de 2012 e 2013. Abreviaturas: Arg (arginina), GABA (ácido γ-aminobutírico), Thr (treonina), Ala (alanina), Ser (serina), Glu (glutamina), Glu acid (ácido glutâmico), Val (valina), Phe (fenilalanina), Leu (leucina), His (histidina), Asp acid (ácido aspártico), Ile (isoleucina), Try (triptofano), Tyr (tirosina), Asp (asparagina), Lys (lisina), Cys (cisteina), Pro (prolina), Gly (glicina), Orn (ornitina), Met (metionina).

'Treixadura' musts presented seven amino acids contents similar between the two considered vintages: arginine, proline, valine, isoleucine, phenylalanine, ornithine and lysine. Three amino acids appeared at greater concentrations in musts from 2013 than in those from 2012: glutamic acid, glycine and cysteine.

The remaining 12 amino acids appeared at higher concentrations in musts from 2012 (Table II).

PCA was able to discern between varieties and years according to the seven major amino acids found in the must (Figure 3), proving the importance of variety and vintage on the amino acids concentration of musts. Principal components (PC) 1 and 2 explained 95.7% of the total variance in the dataset, 72.8% for PC1 and 22.9% for PC2. In the construction of PC1, the contents of arginine, alanine, histidine, threonine and glutamine had a major influence; whereas in the construction of PC2 the contents in glutamic acid, GABA and glutamine were more important. 'Treixadura' musts were related to higher concentrations of arginine, alanine, glutamine, histidine and threonine (Figure 3). In contrast, 'Godello' from 2012 was related to high concentrations of GABA.

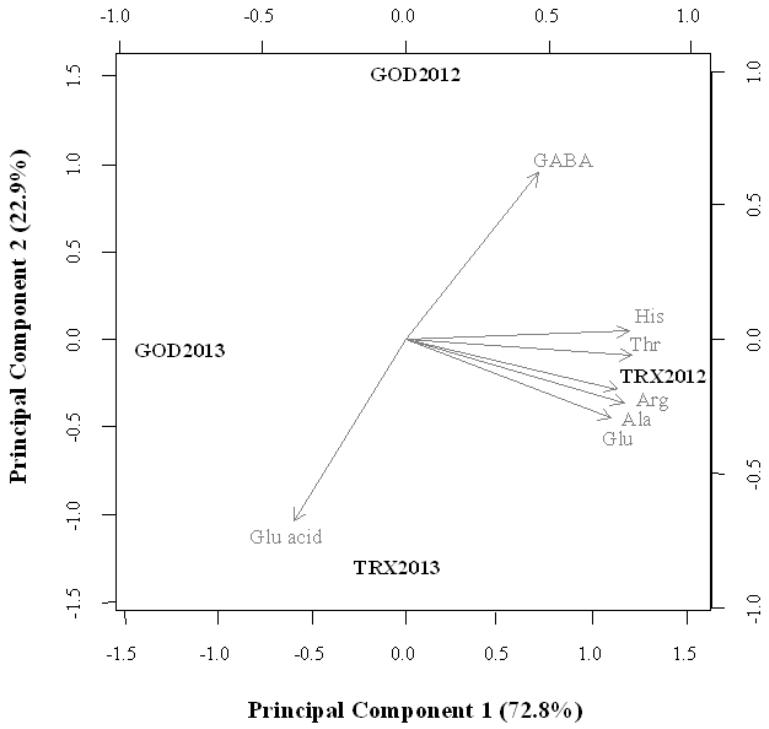

Figure 3. Principal component analysis (PCA) of 'Godello' and 'Treixadura' musts amino acids contents (seven major amino acids). Biplot for the first two components, including the projections of the variables and cases on the factor plane. GOD = Godello, TRX = Treixadura.

Análise em componentes principais (ACP) das concentrações de aminoácidos (sete aminoácidos maioritários) dos mostos de 'Godello' e 'Treixadura'. Biplot das duas primeiras componentes, incluindo as projeções das variáveis e dos casos. GOD = Godello, TRX = Treixadura.

The concentrations of some amino acids differed between the two cultivars studied. 'Treixadura' showed higher values for 14 amino acids in both vintages and for seven amino acids in some year. It is noticeable the highest value for glutamine in 'Treixadura' musts when compared with that of 'Godello'. These results clearly reflected the effect of grape variety on the amino acids concentrations in the musts.

In general, amino acids contents were greater in 2012 than in 2013, which might be caused by the different climate conditions occurring in both years. In fact, rainfall was lower in 2012 (841 mm) than in 2013 (1282 mm); while evapotranspiration was very similar between years (914 mm and 955 mm for 2012 and 2013, respectively), indicating that 2012 could be considered a drier year. These results are in accordance with those presented for another Spanish

white grapevine cultivar, 'Verdejo', in three consecutive years (Ortega-Heras *et al.*, 2014), who observed greater amino acids concentrations in the warmer and drier year.

In the two vintages studied, the major amino acids in the grapes from the studied cultivars were arginine, glutamine, glutamic acid and alanine. These amino acids represented, respectively, 15.1 – 26.4%, 8.5 – 19.1%, 5.1 – 20.7% and 8.3 – 11.9% of the total amino acids present in grapes, depending on the cultivar and vintage. Arginine has been pointed out as the most abundant amino acids in grapes (Bell and Henschke, 2005), in accordance with our findings.

All amino acids were in the standard range of concentrations found for these compounds (Bell and Henschke, 2005) except for proline, which was under the reported concentrations. This may have been caused by the fact that proline accumulation occurs late in ripening (Stines *et al.*, 2000), whereas arginine accumulation begins before veraison. The proline/arginine ratio is used to classify grape cultivars according to their ability to accumulate either one or the other of these two amino acids (Garde-Cerdán *et al.*, 2009). This ratio is a function of the cultivar studied. In our study 'Godello' and 'Treixadura' musts presented proline/arginine ratios lesser than 1, being in 2012 between 0.01 in 'Godello' and 0.02 in 'Treixadura' and in 2013 between 0.02 in 'Treixadura' and 0.06 in 'Godello', thus these varieties are arginine accumulators. These ratios are similar to those reported for 'Syrah' by Garde-Cerdán *et al.* (2009), who observed that proline concentrations in musts from 'Monastrell', 'Syrah', 'Merlot' and 'Petit Verdot' were lower than 5% of the total free amino acids content at harvest time.

The proline concentrations found in the samples from 'Godello' and 'Treixadura' were very low (Table II) in disagreement with many studies (Stines *et al.*, 2000; van Heeswijck *et al.*, 2001). However, the precautions for proline quantification following in the HPLC method (Gómez-Alonso *et al.*, 2007) used in the current study were carefully taken into account. We do not have an explanation for these results; however, other authors reported low amounts of proline for other varieties such as 'Syrah' and 'Merlot' (Garde-Cerdán *et al.*, 2009). 'Verdejo' also showed lower amounts of proline when compared to arginine (Ortega-Heras *et al.*, 2014), although proline concentrations increased when grapes were over-matured. In fact, the accumulation of proline is not uniform throughout berry development but confined to the later stages of ripening, as observed by Stines *et al.* (2000) in 'Gewurztraminer', 'Chardonnay', 'Cabernet Sauvignon' and 'Muscat Gordo'. This may indicate that 'Godello' and 'Treixadura' samples collected in the current study were not over-matured and proline biosynthesis mechanisms were still not finished. Furthermore, proline and arginine metabolism may be linked and the final concentration of one is influenced by the other, with arginine acting as a precursor for at least some of the proline accumulated (Kliewer, 1968).

General attributes of the wines

Significant differences between cultivars were detected for titratable acidity, pH and tartaric acid content (Table III). Vintage exerted a significant influence on alcoholic grade and tartaric acid content; whereas the interaction between cultivar and vintage did not influence any of the attributes considered.

In the case of 'Godello', significant differences in tartaric acid content were observed between vintages. 'Treixadura' wines presented a significantly higher alcoholic grade in 2013 than in 2012 (Table III).

Direct precursors of higher alcohols and volatile fatty acids (Styger *et al.*, 2011), such as the three nonpolar branched-chain amino acids (valine, leucine and isoleucine), appeared under 5% in the studied samples. In fact, several authors noticed a significant relationship between must amino acids and wine aromatic composition (Guitart *et al.*, 1999; Hernández-Orte *et al.*, 2002). Falqué *et al.* (2001) observed a higher concentration of aromatic compounds in 'Albariño' than in 'Treixadura' wines; several of those are directly related to the amino acids of the musts, such as 2-phenyl ethanol that can be glycosylated as aroma precursor in grape berries. Other compounds allowed Falqué *et al.* (2001) to classify the wines according to grape variety, among these compounds methionol is dependent on the concentration of methionine in the musts.

Threonine influences wine aroma composition (Hernández-Orte *et al.*, 2002), since it is strongly related to odorants from the fatty acid synthesis. In this study, 'Treixadura' presented greater contents in this amino acid than 'Godello' musts, pointing out a greater aromatic potential of the former variety.

Furthermore, the higher the must content in phenylalanine, the higher the wine relative contents of β-phenylethanol and isobutanol, and the lesser the wine relative contents of isoamyl alcohol (Hernández-Orte *et al.*, 2002). In the current study, 'Treixadura' musts presented greater phenylalanine contents than 'Godello' musts.

Table III

General attributes of the wines from the two studied cultivars and the two vintages surveyed

Atributos gerais dos vinhos das duas variedades e das duas vindimas estudadas

Cultivar	Vintage	Attribute				
		Alcoholic grade (% Vol.)	Titratable acidity (g/L tartaric acid)	pH	Malic acid (g/L)	Tartaric acid (g/L)
Godello	**2012**	13.5 a	7.1 a	3.09 a	1.8 a	3.3 b
	2013	14.1 a	6.6 a	3.14 a	2.2 a	2.1 a
Treixadura	**2012**	13.5 a	5.2 a	3.45 a	1.5 a	2.0 a
	2013	14.7 b	6.0 a	3.40 a	2.5 a	1.4 a
Cultivar		ns	*	*	ns	*
Vintage		*	ns	ns	ns	*
Cultivar x Vintage		ns	ns	ns	ns	ns

Different letters next to an attribute indicate significant differences between years for a given variety ($p < 0.05$). For the factorial analysis: ns = non-significant; * $p < 0.05$.

The two cultivars studied ('Godello' and 'Treixadura') presented similarities in the sense that the major and minor amino acids were common. This might indicate an adaptation of these varieties to the region, as previously reported by Garde-Cerdán *et al.* (2009) for 'Monastrell', 'Syrah', 'Merlot' and 'Petit Verdot' in South East Spain.

CONCLUSIONS

'Godello' and 'Treixadura' musts differed on the concentrations of their amino acids profiles. 'Treixadura' presented the highest contents in amino acids. This fact points out the important effect of variety on the amino acids profile of musts. However, arginine was the major amino acid for the two varieties and the minor amino acids (methionine, ornithine, and glycine) were common for them.

A strong effect of vintage on the amino acids content in grapes was observed. This effect may have been caused by the different degree of berry maturation due to the particular weather conditions observed between years.

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