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Yield and grape must composition in 'Cabernet Sauvignon' grape vine subjected to potassium fertilization in high altitude soil

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ABSTRACT: Potassium (K) increase in the soil can affect grape must yield and quality; and consequently, the quality of wine; however, studies in this field approaching altitude soil in Southern Brazil (used for viniculture) are scarce. The aim of the present study is to evaluate yield, critical K level in the leaves and their association with grape must composition in 'Cabernet Sauvignon' vines subjected to K application in high altitude soil in Southern Brazil. The experiment was performed in a 'Cabernet Sauvignon' vineyard subjected to the application of different K doses (0, 30 and 60 kg K_2O ha^{-1}) in three seasons. Potassium (K) availability in soil and leaves, as well as total soluble solids (TSS), pH, total titratable acidity (TA) content in grape must were evaluated. Potassium fertilizer applications increased K availability in the soil, but its concentration only increased in leaves collected at full bloom in the 2013/2014 season. Potassium applications in the soil did not affect yield, but K concentration increase in the leaves led to increased TSS and pH values in grape must, as well as to TA and tartaric acid decrease, a factor that can impair the quality of wine.

Key words: available K; leaf analysis; leaf K; *Vitis vinifera* L.

Produtividade e composição do mosto em videira 'Cabernet Sauvignon' submetidas à adubação potássica em solo de elevada altitude

RESUMO: O incremento de K no solo pode afetar a produtividade e qualidade do mosto, por consequência, do vinho. Mas, estudos desta natureza são escassos em solos de altitude do Sul do Brasil, que são incorporados a viticultura. O estudo objetivou avaliar a produtividade, nível crítico de K em folhas e a relação com a composição do mosto em uva 'Cabernet Sauvignon' submetidas à aplicação de K em solo de altitude no Sul do Brasil. O experimento foi realizado em vinhedo da 'Cabernet Sauvignon', cujas plantas foram submetidas a aplicações de doses de K (0, 30 e 60 kg K_2O ha^{-1}) durante três safras. Foram avaliados os teores de K disponível no solo e nas folhas, além dos teores de sólidos solúveis totais (SST), pH do mosto, acidez titulável (AT) e quantidade de ácido tartárico no mosto. As aplicações de fertilizantes potássicos aumentaram os teores de K disponíveis no solo, mas somente incrementaram a concentração de K nas folhas coletadas no pleno florescimento na safra 2013/2014. As aplicações de K no solo não afetaram a produtividade, mas, o incremento da concentração de K em folhas promoveu aumento dos valores de SST e no pH do mosto, e diminuição dos valores de AT e ácido tartárico o que pode prejudicar a qualidade do vinho.

Palavras-chave: K disponível; análise foliar; K folha; *Vitis vinifera* L.

Introduction

Altitude soils in Southern Brazil, including the ones cultivated with vines, are often acidic and present high organic matter contents and variation in potassium (K) availability level (Almeida et al., 2018). The highest natural exchangeable K contents are observed in soil with potassium from other sources such as minerals (Teske et al., 2013) and with high cation exchange capacity (CEC), which accounts for exchangeable K absorption - there must be K balance in the solution absorbed by plants (Poonpakdee et al., 2018), including vines. However, grapes from producing 'Cabernet Sauvignon' vineyards - including those grown in high altitude soils in Santa Catarina State (SC) - are destined to fine wine production. Potassium absorption is often observed because of K export by grape bunches and of the depletion of native exchangeable K content from soil or even of K derived from fertilization, such as pre-planting, before vineyard. Therefore, it is necessary fertilizing the soil for potassium maintenance (or production) during vines' productive period.

Fertilization with Potassium is necessary and K maintenance doses must be in compliance with the exchangeable K content in soil (extracted by Mehlich-1), as well as with total K concentration in leaves and yield expectations (CQFS-RS/SC, 2016). However, estimates about soil exchangeable K are not always related to yield parameters or to grape yield in producing vineyards, since vines' root system exploits a larger soil volume than the herein sampled soil volume (Tramontini et al, 2013; CQFS-RS / SC, 2016) and has internal nutrient reserves, including K (Pradubsuk & Davenport, 2010). Therefore, estimates of nutrient concentrations, such as estimates of K content in leaves, (assessed at full bloom or at berries-color–change time) can be closely related to vineyards' yield parameters, as well as to grape must oenological parameters such as (TST), pH, total titratable acidity (ATT), malic acid, among other parameters (Ciotta et al., 2016; Messiga et al., 2016) that can influence the quality of the wine (Walker & Blackmore, 2012). It is likely estimating K critical level in leaves (collected at full bloom) to assess oenological quality parameters in grape must, since these parameters can also apply to other wine types. Based on such estimates, producers could predict the quality of the wine and make better decisions about its storage and the target public for each wine type.

The predictability of K concentration in the leaves can be used as tool to estimate oenological quality parameters in grape must - this procedure is justified by the normal K application in the soil. Plant growth, mainly in soil presenting medium or clayey texture and high organic matter content does not affect yield (Sipiora et al., 2005; Ciotta et al., 2016). However, K accounts for 75% of the cation observed in most berries (Rogiers et al., 2017); thus, high K concentrations in vines are likely diagnosed through leaf analysis, since they are related to K concentration in berries. In this case, higher pH and ATT in grape must result from K and from the formation of organic acid complex such as K bi-tartrate, which can precipitate in must (Davies et al., 2006). This process can have negative effect on the color (Walker & Blackmore, 2012), aroma and stability of wine (Davies et al., 2006); besides, higher pH in grape must is associated with fermentation stability (Conde et al., 2007). However, knowledge on must from grape grown in altitude soil - which often presents clayey texture and high organic matter content – remains incipient. This soil type is associated with higher natural CEC values recorded in experiments conducted in more than one season, as the present one.

The aim of the present study was to evaluate the yield, critical K level in the leaves and their association with the composition of grape must from 'Cabernet Sauvignon' vines subjected to K application in high altitude soil in Southern Brazil.

Materials and Methods

The study was conducted in a vineyard located in Água Doce, Midwestern Santa Catarina State (SC), Southern Brazil (latitude 26º 42 '10 ", longitude 51º 43' 49"). Climate in the region is classified as temperate oceanic (Cfb) with mild summers, according to Köppen-Geiger's classification. The number of accumulated chilling hours (≤ 7.2°C) ranges from 642 to 778 hours per year. Mean air temperature and rainfall are shown in Figure 1. The soil in the vineyard was classified as Cambissolo Húmico (Santos et al., 2013) or Typic Humicryept (Soil Survey Staff, 2010) or Humic Cambisol (FAO et al., 1998).

'Cabernet Sauvignon' vines (*Vitis vinifera* L.) were grafted onto 'Paulsen 1103' rootstock (*Vitis berlandieri* × *Vitis rupestris*). The vines were trained in the vertical shoot position (VSP) system and spaced 2.9 m ×1.5 m from each other, thus totaling 2,299 plants per hectare. Soil chemical features in the 0-20 cm layer before experiment installation were 445 g kg^{-1} clay (pipette method) and 63 g kg^{-1} organic matter (Walkley-Black method); pH in water 6.1 (ratio 1: 1); 6.9 and 4.7 cmol$_c$ kg^{-1} of Ca and Mg, respectively (extractor KCl 1 mol L^{-1}); 7.2 and 260 mg kg^{-1} of available P and K (Mehlich 1 extractor) (Tedesco

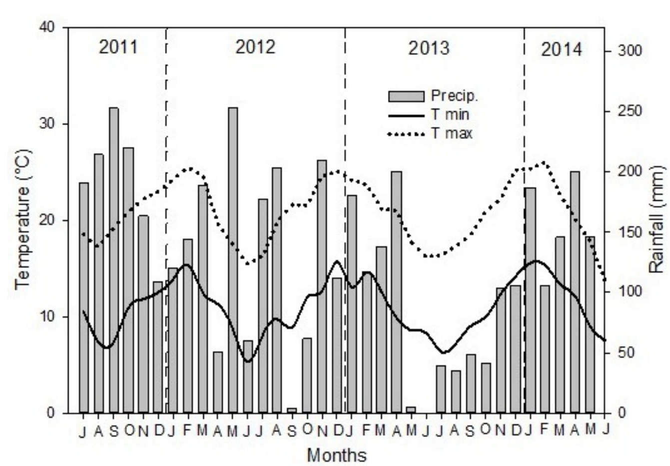


Figure 1. Rainfall (mm), maximum mean temperature (T max) and minimum mean temperature (T min) recorded from July 2011 to June 2014 in Água Doce – SC (Epagri, 2019).

et al., 1995); 13.1 and 17.5 $cmol_c$ kg^{-1} of $CEC_{effective}$ and $CEC_{pH7.0}$, respectively. Vines were subjected to applications of 0, 30 and 60 kg K_2O ha^{-1} $year^{-1}$ from the 2011/2012 to 2013/2014 season - applications were performed at the beginning of bud burst. Potassium chloride (KCl, 60% K_2O) applied on crown projection area and on soil surface (without incorporation) was the K source. The research followed the randomized blocks experimental design with four replications. Each plot comprised six vines, but only the three central plants were evaluated.

Interline vegetation mostly encompassed white clover (*Trifolium repens*), ryegrass (*Lolium multiflorum*) and Festuca *(Festuca arundinacea*). Vegetation was grazed to 10 cm tall every 50 days; plant waste was deposited on soil surface.

Soil samples were collected in the 0-20 cm layer after the berry-color-change time, in all assessed seasons. Soil was air dried, ground, sieved in 2-mm mesh and subjected to K availability analysis (Mehlich 1 extractor) through lame photometry. In total, 10 mature leaves per plant were collected in the mid-third of the branch of the year at full flowering and berry-color-change time. The leaves were dried in forced air oven at 65°C until constant weight was reached; next, they were ground and subjected to sulfur digestion (Tedesco et al., 1995). Potassium concentration in the extract was determined through flame photometry.

The number of clusters per vine was counted at harvest; all clusters were collected and weighed to determine yield. A sample comprising five clusters per vine was evaluated to assess mass (g), length (cm), and width (cm). Three samples, with 50 berries each, were randomly collected from different clusters, at different positions. Berries were crushed for grape extraction; grapes were subjected to chemical analysis in order to determine soluble solids content (ºBrix) in RTD-45 digital refractometer adjusted to temperature compensation (Instrutherm, São Paulo, Brazil); titratable acidity through titration (0.1 N NaOH) in 1% phenolphthalein (meq L^{-1}); pH measured in AD1030 pH meter (ADWA, Szeged, Hungary); and tartaric acid calculated in LC-10A high-performance liquid chromatography equipment (Shimadzu, Kyoto, Japan).

Results were subjected to analysis of variance; regression equations were adjusted through linear model testing through F test when effects were significant – results showing significance level lower than 5% ($p < 0.05$) were chosen. Regressions among K concentration in the leaves, flowering yield, total soluble solids, grape must pH, total acidity and tartaric acid in grape must were also performed. Relative yield (rr) was calculated through equation rr = (rt / rm) * 100; wherein, rt is treatment yield and rm is maximum yield. The rr results were related to K content in the leaves at full flowering in complete leaves - it was calculated to find the sufficiency level referring to 90% rr. Mathematical adjustment between variables was performed by applying the Mitscherlich model $\hat{y}=a\,(1-bx)$; where in, $\hat{y}$ represents rr, a and b are constants, and x is the nutrient content in the leaves.

Results and Discussion

Potassium availability in the 0-20 cm soil layer increased after the application of K fertilizer at the tested doses (Table 1); the recorded results were 3.6, 5.6 and 3.1 mg K kg of K_2O applied in the 2011/2012, 2012/2013 and 2013/2014 seasons, respectively. This result derives from K fertilizer solubilization in the soil, which increases K forms and the amount of K solution available in the soil (exchangeable), estimated in Mehlich-1 extractor (Poonpakdee et al., 2018). The K contents in all treatments and seasons were very high (> 240 mg K kg, for soils with $CEC_{pH7,\ 0}$ 15.1 to 30.0 $cmol_c$ kg) (CQFS-RS/SC, 2016), except for the control soil, in 2013/2014 season, which recorded high K content (121-240 mg of K kg, for soils with $CEC_{pH7,0}$ 15.1 to 30.0 $cmol_c$ kg) (CQFS-RS/SC, 2016). Very high or high K availability (exchangeable) levels in vineyard soils, without nutrient application, likely resulted from the weathering of minerals such as micas and feldspars that end up working as K source (Melo et al., 2004). These minerals can be found in acidic volcanic rocks composing the soil in the assessed region (Dortzbach et al., 2016). In addition, these forms of K (native to the soil), mainly those available in the solution, can be cyclized by native or implanted vegetation (Pérez-Álvarez et al., 2015; Oliveira et al., 2016) such as cover crop species found in vineyards – this process reduces the availability of K forms in the soil over time (Oliveira et al., 2016). On the other hand, part of K available in the control soil and in treatments, in the assessed seasons, possibly derived from pre-planting fertilization, which was carried out before vineyard installation (Schmitt et al., 2014; Dalbó et al., 2015; Ciotta et al., 2016) - this procedure is natural in experiments ran in orchards or vineyards installed during crop production (Dalbó et al., 2015; Ciotta et al., 2016).

However, in the 2012/2013 season, K levels in the soil increased due to the application of K doses (Table 1) but, in the 2013/2014 season, K availability levels decreased both in the control soil and in soils subjected to two K doses. This outcome can be related to higher K export by grape bunches, since there was greater grape yield in this season than in the 2012/2013 one. This outcome can be explained by the fact that approximately 70% of K absorbed from the soil could have been allocated to bunches (Pradubsuk & Davenport, 2010) and approximately 9 kg of K_2O could have been exported per ton of grapes (Ganeshamurthy et al., 2011). Based on these results, grape yield expectations should continue to be used as auxiliary prediction criterion to define the K dose needed by the grapevine, as already recommended by CQFS-RS/SC (2016). Potassium fertilizers can be applied even in soil recording intermediate and high K content, since the depletion of K forms available is fast, mainly in years of higher grape yield (Boonterm et al., 2013). Assumingly, the behavior observed in altitude Humic Cambissol soil in Midwestern Santa Catarina State has been incorporated to grapevine cultivation, but it may not have non-exchangeable K reserves capable of buffering exchangeable K, since there was approximate decrease by 100 mg K kg^{-1} in the soil. The observed decrease was most likely caused by grape export, since soil relief was

Table 1. Exchangeable K content in soil and total K content in the leaves at two collection times, number of bunches per plant, bunches mean mass, yield, bunches length and mean width of 'Cabernet Sauvignon' grapevines subjected to the application of increasing K_2O doses in the soil.

Variable	K_2O dose (kg ha^{-1})			Equation	R^2	CV (%)
	0	30	60			
2011/2012 season						
Exchangeable K in the soil - after grape harvest (mg kg^{-1})	233.4	277.4	448.8	y = 212.2 + 3.59 x	0.90**	17.5
Total K in the leaves - flowering (g kg^{-1})	13.6	14.1	14.6	-	ns	5.2
Total K in the leaves – berries-color-change season (g kg^{-1})	-	-	-	-	-	-
Number of bunches per vine	16	13	15	-	ns	17.2
Mean bunch mass (g)	239	202	214	-	ns	9.7
Mean bunch length (cm)	12.9	12.8	12.0	-	ns	9.6
Mean bunch width (cm)	4.6	4.6	4.6	-	ns	5.3
Yield (Mg ha^{-1})	8.5	6.1	7.1	-	ns	14.4
2012/2013 season						
Exchangeable K in the soil - after grape harvest (mg kg^{-1})	205.3	307.5	540.7	y = 183.2 + 5.60 x	0.95**	16.0
Total K in the leaves - flowering (g kg^{-1})	10.9	10.6	12.1	-	ns	9.5
Total K in the leaves – berries-color-change season (g kg^{-1})	20.2	20.6	23.5	-	ns	19.5
Number of bunches per vine	12	11	12	-	ns	13.4
Mean bunch mass (g)	69	62	62	-	ns	29.0
Mean bunch length (cm)	11.2	11.4	11.6	-	ns	8.7
Mean bunch width (cm)	5.2	4.8	5.5	-	ns	10.2
Yield (Mg ha^{-1})	1.8	1.5	1.8	-	ns	29.3
2013/2014 season						
Exchangeable K in the soil - after grape harvest (mg kg^{-1})	143.1	213.0	330.1	y = 135.1 + 3.13 x	0.98**	23.2
Total K in the leaves - flowering (g kg^{-1})	21.8	18.4	20.6	-	ns	6.8
Total K in the leaves – berries-color-change season (g kg^{-1})	14.8	17.9	19.5	y = 15.09 + 0.077 x	0.97**	7.3
Number of bunches per vine	20	22	25	-	ns	12.9
Mean bunch mass (g)	115	124	132	-	ns	10.8
Mean bunch length (cm)	13.7	12.2	14.4	-	ns	10.7
Mean bunch width (cm)	5.8	5.6	6.5	-	ns	15.7
Yield (Mg ha^{-1})	5.3	6.3	7.6	-	ns	22.7

ns = not significant; ** = significant, $p < 0.05$; *** = significant, $p < 0.01$; CV = coefficient of variation.

flat and reduced surface runoff and debris transfer from soil cover species; moreover, pruned branches remained on the soil in the vineyards.

Potassium concentration in the leaves increased due to K dose when leaves were collected in the berries-color-change time in the 2013/2014 season (Table 1). Total increment of 0.08 g K was observed for each Kg of K_2O applied to the soil. There was no K concentration increase in leaves at leaf collection and full bloom. Potassium concentrations in leaves collected at leaf-color-change time in all seasons were normal (8-16 g kg^{-1}) or excessive (above 16 g kg^{-1}) (CQFS-RS/SC, 2016). Potassium concentrations in leaves collected at flowering time in vines that were not subjected to K application or to applications of two K doses in 2013/2014 season were approximately twice as higher as those observed in the previous season. This outcome is likely related to the higher absorption of K in the soil by vines, which reinforces the depletion of the available levels in the soil, since higher grape yield and, consequently, greater K export were observed in 2013/2014 (Ciotta et al., 2016). Based on this result, flowering must be the proper leaf collection stage to diagnose nutritional status in vines (Ciotta et al., 2016). However, it is necessary defining the critical K levels in the leaves based on quantitative parameters such as yield, or on qualitative ones such as grape must or wine oenological parameters.

Yield and its components (number of bunches per vine, mean bunch mass, yield, mean bunch length and bunch width) were not affected by the tested K doses (Table 1). However, yield results observed in the three evaluated seasons showed increase in this variable after K concentration increased in leaves collected at flowering (Figure 2A; Figure 2B). Yield increased by 0.36 Mg ha^{-1} of grape, on average, for each increase in K concentration units in leaves at flowering. There was no yield increase at berries-color-change time related to K content in the leaves and also between soil content and productivity (data not shown). Lack of association between yield and K content in the leaves may have resulted from vine roots' ability to absorb K forms available in deeper soil layers - 30 and 90 cm (Sipiora et al., 2005), rather than only in the 0-20 cm layer. Soil microorganisms and plant roots may have strategies such as H^+ or organic acids release to stimulate the solubilization of minerals that have K in their composition - this process has increased the availability of K forms in the soil (Sharma et al., 2016). The closer association between K concentration in the leaves in the assessed seasons and yield may have resulted from internal K reserves in perennial organs such as roots - decrease was observed in years recording higher grape yield and, consequently, greater K export by bunches (Pradubsuk & Davenport, 2010; Ganeshamurthy et al., 2011). Thus, K concentration gradient forms inside

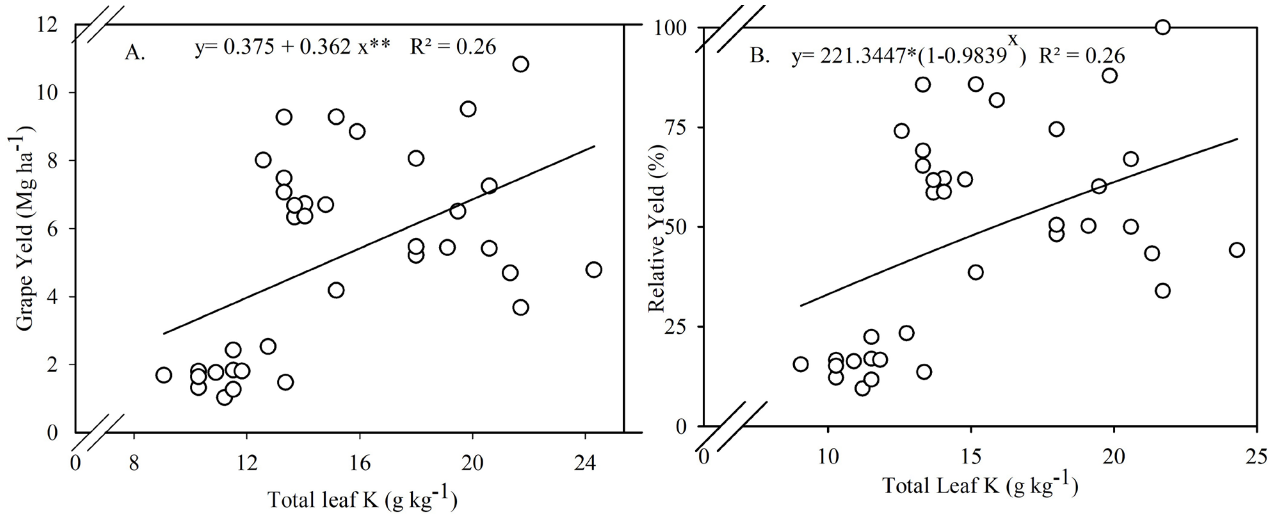


Figure 2. Linear regression between total K in leaves at flowering and the yield (A.) and relative yield (B.) of 'Cabernet Sauvignon' grapes subjected to the application of K_2O doses.

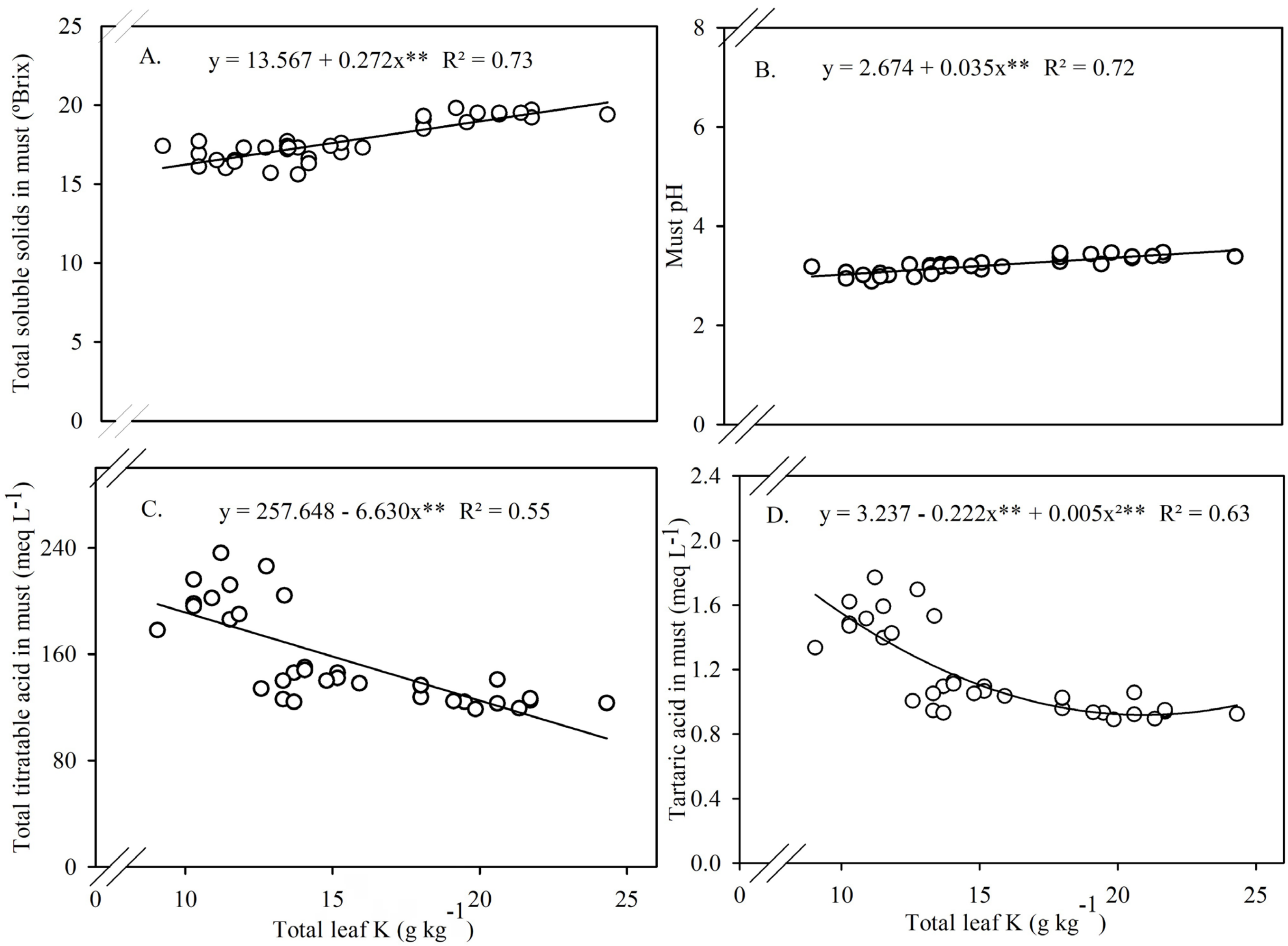
plants (more K was observed in plants subjected to nutrient addition and lesser K was observed in plants recording lower K application history); it can be diagnosed through leaf analysis, according to which, vines presenting smaller K reserves may produce smaller amounts of grapes likely because of issues related to photosynthesis, mainly to functions related to water in plants, such as growth process, cell maintenance and turgor, and water transport via phloem (Liesche, 2016; Rogiers et al., 2017).

Applications of K doses did not affect grape must oenological parameters such as SST, pH, ATT and tartaric acid (Table 2). However, increased K concentration in leaves collected at flowering led to increased SST and pH values (Figure 3A; Figure 3B) and to decreased ATT and tartaric acid values in grape must (Figure 3C; Figure 3D). Increased SST values in grape must resulted from increased production of sugars; these values can contribute to proper grape must fermentation (Conde et al., 2007). On the other hand, increased pH values and decreased values of ATT and acids, such as the tartaric acid, resulted from complex formations - including organic acids and K - to form K bi-tartrate, which has negative effects on wine stability since it shortens wine storage time (a fact not always desirable) (Davies et al., 2006). Titratable acidity recorded high values and it reflected the concentrations of free organic acids, mainly of tartaric and malic acids, in grapes. These results are likely related to low pH values in grape must, whose overall pH in the present study was optimum (> 3.0 and < 3.8), because numbers above and below the ideal pH can cause preclude grape must fermentation and change organoleptic features, color, flavor and the oxidation power of wine (Kodur, 2011). Lower SST values in grape must often consist of 95% sugars (Smart & Robinson, 1991) and result from the smaller number of clusters per plant – these clusters were noticeably smaller in length and width, which resulted in lower yield (Table 1). Thus, SST tends to be more concentrated in berries from smaller clusters (Guilpart et al., 2014).

Table 2. Composition of grape must from 'Cabernet Sauvignon' grapevines subjected to the application of increasing K_2O doses in the soil.

Variable	K_2O dose (kg ha^{-1})			Equation	R^2
	0	30	60		
		2011/2012 season			
Total Soluble Solids (ºBrix) in grape must	17.3	17.2	16.7	-	ns
Grape must pH	3.18	3.22	3.19	-	ns
Grape must total titrated acidity (meq L^{-1})	136.5	144.5	137.5	-	ns
Grape must tartaric acid (meq L^{-1})	1.02	1.08	1.03	-	ns
		2012/2013 season			
Total Soluble Solids (ºBrix) in grape must	16.9	16.7	16.5	-	ns
Grape must pH	3.04	3.00	3.00	-	ns
Grape must titrated acidity (meq L^{-1})	198.0	205.0	211.0	-	ns
Grape must tartaric acid (meq L^{-1})	1.49	1.54	1.58	-	ns
		2013/2014 season			
Total Soluble Solids (ºBrix) in grape must	19.5	19.0	19.5	-	ns
Grape must pH	3.38	3.33	3.44	-	ns
Grape must titrated acidity (meq L^{-1})	128.0	131.1	122.3	-	ns
Grape must tartaric acid (meq L^{-1})	0.96	0.98	0.92	-	ns

ns = not significant ($p \geq 0.05$).



** = significant, p < 0.01.

Figure 3. Linear regression between total K (g kg^{-1}) in the leaves at flowering and total soluble solids in grape must (°Brix) (A), grape must pH (B), total titratable acid in grape must (meq L^{-1}) (C) and Tartaric acid in grape must (meq L^{-1}) (D) from 'Cabernet Sauvignon' subjected to the application of K_2O doses.

Conclusions

The application of potassium fertilizers increased K availability levels in the soil, but only increased K concentration in leaves collected at full bloom in the 2013/2014 season.

Potassium applications in the soil did not increase grapevine yield but it increased total soluble solids, grape must pH, and decreased total titratable acid and tartaric acid in grape must, which can impair the quality of the wine.

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