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# Nitrogen fertilization of Cabernet Sauvignon grapevines: yield, total nitrogen content in the leaves and must composition

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**ABSTRACT.** Grapevines grown on sandy soils are subjected to the application of supplemental nitrogen (N); however, there is little information available regarding the impact of these applications on yield, plant nutritional state and must composition. The aim of this study was to evaluate the yield, nutritional state and must composition of grapevines subjected to N fertilization. Cabernet Sauvignon grapevines were subjected to annual applications of 0, 10, 15, 20, 40, 80 and 120 kg N ha<sup>-1</sup> in 2008, 2009 and 2010. During the 2008/09, 2009/10 and 2010/11 harvest seasons, leaves were collected during full flowering and when the berries changed color, and the total N content was analyzed. The grape yield and the enological characteristics of the must were evaluated. The response to applied N was low, and the highest Cabernet Sauvignon grape yield was obtained in response to an application of 20 kg N ha<sup>-1</sup> year<sup>-1</sup>. The application of N increased the nutrient content in the leaf collected at full flowering, but it had little effect on the total nutrient content in the must, and it did not affect the enological characteristics of the must, such as soluble solids, pH, total acidity, malic acid and tartaric acid.

Keywords: urea, leaf analysis, yield components, Vitis vinifera.

# Adubação nitrogenada de videiras Cabernet Sauvignon: produtividade, nitrogênio total em folhas e composição do mosto

**RESUMO.** As videiras cultivadas em solos arenosos são submetidas a aplicações de nitrogênio (N), porém são poucas as informações sobre o seu impacto na produção, estado nutricional e a composição do mosto. O trabalho objetivou avaliar a produção, estado nutricional e composição do mosto em videiras submetidas à aplicação de N. Videiras Cabernet Sauvignon foram submetidas à aplicação anual de 0, 10, 15, 20, 40, 80 e 120 kg N ha<sup>-1</sup>, em 2008, 2009 e 2010. Nas safras de 2008/09, 2009/10 e 2010/11 foram coletadas folhas no pleno florescimento e na mudança da cor das bagas, e analisado o teor de N total, avaliada a produção de uva e no mosto foram determinados atributos enológicos. A resposta à aplicação de N foi pouco expressiva, tanto que a maior produção de uva de Cabernet Sauvignon foi obtida com a aplicação de 20 kg de N ha<sup>-1</sup> ano<sup>-1</sup>. A aplicação de N aumentou o teor do nutriente na folha coletada no pleno florescimento, mas pouco afetou o teor de nutrientes totais no mosto e não interferiu os atributos enológicos do mosto, como os sólidos solúveis, pH, acidez total e ácido málico e tartárico.

Palavras-chaves: ureia, análise foliar, componentes de produção, Vitis vinifera.

#### Introduction

The state of Rio Grande do Sul has the largest area of grapevine cultivation in Brazil, with approximately 50,389 thousand hectares, and it is responsible for the production of more than 90% of the domestic wine. In RS, the Serra Gaúcha region, located in the northeast region of the state, is the most important wine growing region in the country. In the 1970s, vineyards for the cultivation of red *Vitis vinifera* grapes, such as Cabernet Sauvignon, were established in the natural fields of the Campanha Gaúcha region, which is located in the

southeast of Rio Grande do Sul State. The majority of these soils have a sandy surface horizon and a low or medium organic matter content, which is believed to result in a low capacity to supply mineral nitrogen (N) to plants (BRUNETTO et al., 2007). However, these soils also contain the intense growth of cover crop species in the vineyards. If the cover crop consists of leguminous plant species, they may promote the biological fixation of atmospheric N. In conjunction with gramineous plants or other species, the cover crop may take up other nutrients, including N, from deeper layers of the soil. The subsequent decomposition of the aboveground plant

tissues and senescent roots may release the N contained in the tissues to the grapevine root zones and allow i to be utilized by the plants (BRUNETTO et al., 2009; 2011). However, the N content of whole grapevine leaves, which are used as the diagnostic organ of the nutritional state, is commonly below normal (<16 g kg<sup>-1</sup>) (CQFS-RS/SC, 2004) and requires N fertilization.

Currently, in the state of RS and Santa Catarina (SC), the N dose to be applied to the soil is established based on the total nutrient content in the whole leaf or in the petiole, which is collected at veraison and in accordance the expectation regarding grape yield (CQFS-RS/SC, 2004). However, the leaves collected at veraison are not always sufficiently sensitive for an accurate diagnosis of the increase in total N within the plant. Therefore, the N availability in the soil does not always directly correlate with grape yield. This is likely due to its dilution in the organ sampled or by its redistribution to annual organs, such as grape clusters and new growing shoots, or even to perennial organs, especially to branches older than one year and to the roots (BRUNETTO et al., 2006a; 2008a; 2009). Collection during other periods, such as during full flowering, may be more appropriate for the detection of total N in the leaves and might improve the analysis of the nutritional state. In addition, the impact of N application to the soil is not sufficiently understood, and the adequate dose to be applied to improve grape yield and parameters such as the number of clusters per plant, the weight and number of berries per cluster and the 100-berry weight, is unknown (BELL; ROBSON, 1999; BRUNETTO et al., 2007; 2008b). Must composition indicators, such as the total nutrient content, are also unknown. The N content is a determining factor for must fermentation because it affects the microbial biomass. the fermentation rate and fermentation time (BELTRAN et al., 2005; JIRANEK et al., 1995). In addition, there is a lack of information on the impact of N application on potassium (K). An increase of K in the must may lead to the formation of K bitartarate in the wine, and this may increase the pH of the wine, thus accelerating its oxidation and reducing its quality over time (MPELASOKA et al., 2003). There is little information regarding the importance of soluble solids, which when in the degrees Brix scale, represent 90% of the sugars found in the must, and pH, total acidity, tartaric acid and malic acid, which represent more than 90% of the total acids in the berry and indicate the stability and longevity of the wine (SPAYD et al., 1994; BRUNETTO et al., 2008a). The effects of N fertilization on the grapevine are dependent on the chemical attributes of the soil and climatic conditions, which also determine the typical features of the wine; therefore, carrying out regional field experiments is necessary, preferentially spanning more than one crop season to increase the reliability of the data. The most appropriate dose of N to be applied can then established. The effects of nitrogen supplementation on the nutritional state of the plant and on the composition of grape also need to be determined.

The aim of this study was to evaluate the yield, nutritional state and composition of the grape must from Cabernet Sauvignon grapevines subjected to N fertilization in sandy soil and to provide data to improve recommendations for N applications to the grapevine soil.

# Material and methods

The experiment was conducted in sandy soil in a commercial vineyard located in the municipality of Rosário do Sul in the state of Rio Grande do Sul, in the Campanha Gaúcha region in the south of Brazil. The vineyard was planted in 2004 using the espalier trellis training system using the Cabernet Sauvignon variety, which was grafted onto the rootstock SO4, at a density of 3704 plants per hectare (1.0 x 2.7 m). The climate in the region is the Cfa type, with average temperatures ranging from 11.9 to 23.5°C and an annual average rainfall of 1599 mm. Other climatic data observed during the study are listed in Table 1. The soil was Sandy Typic Hapludalf (SOIL SURVEY STAFF, 1999), and prior to setting up the experiment, it exhibited the following attributes in the 0-20 cm layer: clay 70.0 g kg<sup>-1</sup>; organic matter 10.0 g kg<sup>-1</sup>; pH in water 5.5; exchangeable Al 0.0 cmol<sub>c</sub> dm<sup>-3</sup> (KCl 1 mol L<sup>-1</sup> extractor); exchangeable Ca 0.9 cmol<sub>c</sub> dm<sup>-3</sup> (KCl 1 mol L<sup>-1</sup> extractor); exchangeable Mg 0.6 cmol<sub>c</sub> dm<sup>-3</sup> (KCl 1 mol L<sup>-1</sup> extractor); available P 30.1 mg dm<sup>-3</sup> (Mehlich 1 extractor); available K 48.0 mg dm<sup>-3</sup> (Mehlich 1 extractor) and CEC<sub>pH7.0</sub> 4.7 cmol<sub>c</sub> dm<sup>-3</sup>.

Table 1. Average air temperature, rainfall and hours of sunlight in the experimental area during the 2008, 2009 and 2010 crop seasons.

	200	8		2009	1	2010 Climatological normal										
3.4 .1								Tempe	rature							
Month	Min	Med.	Ma	x.	Min.	Med.	Max.	Min.	Mean	Max.	Min.	Med.	Max.			
								°C								
July	11.0	) [	14.9	19.6	6.3	10.2	15.1	6.9	10.9	17.0	9.1	12.9	18.2			
August	9.8	1	14.1	19.5	10.3	15.2	20.8	7.3	11.6	17.7	9.3	13.6	19.2			
September	8.8	1	13.2	18.5	10.2	14.6	19.2	9.8	14.4	20.2	10.6	14.9	20.4			
October	13.1		16.8	21.2	12.0	16.7	22.6	10.7	16.2	23.2	12.3	17.0	22.8			
November	14.7	7 1	19.4	25.0	17.6	21.6	26.7	12.9	19.7	27.7	14.2	18.9	24.8			
December	15.6	5 2	20.3	26.2	17.0	21.2	26.4	16.7	23.7	31.2	16.0	20.7	26.7			
January	16.1	. 2	20.4	25.7	18.1	22.0	26.8	19.7	25.0	31.6	17.3	21.8	27.8			
February	17.8	3 2	21.7	26.6	19.1	23.0	28.4	18.0	22.8	29.3	17.3	21.7	27.5			
March	17.1	. 2	21.0	26.2	16.8	20.7	25.6	15.5	21.0	27.9	16.1	20.3	26.0			
April	13.7	7 1	18.4	24.1	13.4	17.5	22.4	13.9	18.5	25.1	13.3	17.5	22.9			
May	11.1		15.6	20.8	11.1	14.2	17.7	9.7	13.8	19.8	10.4	14.5	20.0			
June	7.5	1	11.2	15.9	9.0	13.1	18.0	7.5	11.2	16.8	8.6	12.8	17.9			
Mean	13.0	) 1	17.3	22.4	13.4	17.5	22.5	12.4	17.4	24.0	12.9	17.2	22.9			
	2008		22009 2010		Climatological normal			2008	2009	2010	Climatologic	al normal				
Month	·					Rain	fall		Insolation							
					mm				hours							
July	7	73.0	97.	.8	295.3		161.0			144.9	144.9 166.7 1		)			
August	1	98.5	257	'.9	53.3		165.0		193.5	183.0	135.9	159.0				
September	1	44.1	411	.7	182.6		185.0		185.8	134.4	183.7	162.0				
October	3	09.6	145	5.1	19.4		156.0		154.8	194.1	273.8	192.0				
November	7	70.3	359	0.5	29.0		140.0		240.8	141.1	281.1 219.0		)			
December	8	35.8	232	2.6	56.0		144.0		261.7	224.3	293.2	239.0				
January	2	69.6	296	5.4	61.6		140.0		235.7	189.2	259.1	231.0	)			
February	1-	44.5	167	'.1	67.8		139.0		174.0	205.3	219.7	199.0				
March	9	90.6	57.	.2	82.8		128.0		228.1	192.5	250.5		)			
April	2	24.2	142	2.1	103.5		114.0		243.4	180.1 212.4		173.0				
May	1.	34.7	154	.7	42.6		107.0		162.3	98.4	176.6	162.0				
June	8	32.9	129	.9	118.4		157.0		155.4	142.4	146.0	142.0	)			
Sum	16	527.8	245	2.0	1112.3		1736.0		2411.2	2029.7	2598.7	2240.	0			

The grapevines were subjected to an application of 0, 10, 15, 20, 40, 80 and 120 kg N ha<sup>-1</sup> year<sup>-1</sup>, in August 2008, 2009 and 2010, which coincided with the beginning of bud break, for a total of 0, 30, 45, 60, 120, 240 and 360 kg N ha<sup>-1</sup> over the course of three seasons. Nitrogen was applied manually in the form of urea (45% N) on the soil surface, without incorporation, and in 0.5 m wide strips in the plant rows (projection of the plant canopy). Throughout the grapevine cycle, the strip where N was applied was subjected to glyphosate herbicide applications for weed control. Over the three crop seasons, the grapevines were subjected to applications of phosphorus (P) and K, following the official recommendations for the crop (CQFS, 2004). After winter pruning, the plants had approximately eight new shoots, creating a total of 16 buds per plant. The experimental design was a randomized block with three replicates. Each plot consisted of five plants, and evaluations were conducted on the three central plants on an annual basis.

In the 2008, 2009 and 2010 crop seasons, in October, when the grapevines were in full-flower, and in January, when the berry color changed, 15 mature leaves per plant were collected from both sides of the plant row. The leaves were then dried in a forced air laboratory oven at 65°C until they

reached a constant weight. The dried leaves were ground using a Wiley mill and analyzed for total N. For total N analysis, 0.2 g of dry tissue was added to a 100 mL digestion tube with 0.7 g of a digestion solution (90.9% Na<sub>2</sub>SO<sub>4</sub> and 9.1% CuSO<sub>4</sub> 5H<sub>2</sub>O), 2 mL of concentrated H<sub>2</sub>SO<sub>4</sub> and 1 mL of H<sub>2</sub>O<sub>2</sub>. Next, the tubes were placed in a block digester at 150°C. During the digestion, the temperature was gradually raised (50°C every 30 min) to 350°C. Once the extract turned a light greenish-yellow color, the tubes remained in the block digester at a temperature of 350°C for an additional 60 min. Next, 20 mL of distilled water and 10 mL of NaOH 10 mol L<sup>-1</sup> were added to each digestion tube, and the tubes were immediately connected to a Kjeldahl semi-micro steam distiller. The distillation was carried out until 35 mL of distillate remained in 5 mL of the indicator, boric acid. Immediately, the extract was titrated with H<sub>2</sub>SO<sub>4</sub> 0.05 mol L<sup>-1</sup> to quantify the total N in each sample.

Once the berries matured in March of 2009, 2010 and 2011, the number of clusters per plant was counted and the clusters were collected. The clusters were immediately weighed using a digital balance, and five clusters per plant were separated and the number of berries per cluster was counted. Next, 100 grape berries were collected from the upper, middle and lower sections of the five clusters, and they were

weighed using a digital balance. The berries were divided into two portions. Thirty berries were ground in a blender and each sample was subjected to total N, P and K analysis, in which 5 mL of each sample was subjected to the sulfuric acid digestion described above. The total N in the berries was determined using the same method as previously described, while the total K was determined using a flame photometer, and the total P was determined according to the method of Murphy and Riley (1962). The remaining berries were macerated, and the pH values of the must were determined using a digital potentiometer. The soluble solids in the must were quantified using a portable digital refractometer with temperature control, and the total acidity was measured by titration with NaOH 0.1 mol L-1. The malic acid and tartaric acid were quantified by high performance liquid chromatography (HPLC) using a PerkinElmer liquid chromatograph operated using isocratic conditions with a diode array detector and a 20 µL Rheodyne 7125 injector. The results obtained were subjected to analysis of variance using the Sisvar statistical program (FERREIRA, 2000). When the effects were determined to be significant, regression equations were developed; the linear and quadratic models were tested using the F test, and models were chosen that had a probability of error of less than 5% (p < 0.05).

# Results and discussion

## Total nitrogen content in the whole leaf

The total N content of the whole leaves collected at full flowering of the Cabernet Sauvignon grapevines increased in a quadratic manner with the dose of N applied to the soil over the 2008/09, 2009/10 and 2010/11 crop seasons (Table 2). However, in the leaves collected during the berry color change, N application did not affect the total nutrient content in the whole leaf. In most cases, throughout the three crop seasons, the nutrient levels in the leaves collected during the berry color change were lower than those in the leaves collected at full flowering. These results indicate that

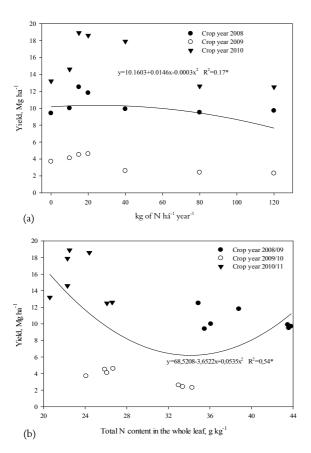
the leaves at full flowering were more sensitive than the leaves collected at berry color change for the diagnosis of N content increases within the plant resulting from the increase in the N dose applied to the soil and to the mineral N content. Similar results were obtained by Brunetto et al. (2008a) in Bordô and Couderc cultivar grapevines grown in an Inceptisol soil with an organic matter content of 11.0 g kg-1 (similar to the organic matter content of the soil of the present study) and subjected to the application of N in the Planalto (Plateau) region of RS. The lack of sensitivity of the leaves collected during berry color change for the detection of N content changes within the plant may be attributed to the decrease of the mineral N content in the soil. Decrease in the soil, especially of NO<sub>3</sub>-N, resulted from soil surface runoff and from percolation through the soil profile (LORENSINI et al., 2012). The Sandy Typic Hapludalf used in the present study has a sandy surface horizon and contained 10.0 g kg<sup>-1</sup> of organic matter, which is considered low (CQFS-RS/SC, 2004). Another reason for the lack of sensitivity of the leaves to the applied N is the dilution of the N contained in the leaf and its redistribution from the leaves to other plant organs, especially to new organs, clusters and shoots. This results in an increase in dry matter and therefore, a drain on nutrients. In addition, in organs older than one year, such as branches from the previous year, stems and roots, the N may be stored and redistributed to the organs for growth in the next cycle (BRUNETTO et al., 2006b, 2008b, 2009; YU et al., 2012). However, even without an increase in the total N content in the whole leaf in response to an increase in the applied N, the total N content in the leaves collected at the berry color change was considered to be average (16.0-24.0 g kg<sup>-1</sup>) or above average ( $> 24.0 \text{ g kg}^{-1}$ ) in all of the N doses and crop seasons (CQFS-RS/SC, 2004). This indicated that, even in soil lacking N fertilization, a sufficient quantity of N was released via the mineralization process of the organic matter that was taken up by the grapevines (BRUNETTO et al., 2009).

**Table 2.** Total nitrogen content in whole leaves collected at full flowering and at berry color change in grapevines subjected to the application of 0, 10, 15, 20, 40, 80 and 120 kg N ha<sup>-1</sup> year<sup>1</sup> during the 2008, 2009 and 2010 crop seasons.

Coon Sanan			Emerica	$\mathbb{R}^2$					
Crop Season	0	10	15	20	40	80	120	- Equation	K
				Full flow	ering (g kg	<sup>-1</sup> )			
2008/09	35.5	36.1	34.9	38.8	43.5	43.6	43.0	$y=34.270+0.2341x-0.0013x^2$	0.92*
2009/10	24.1	26.1	25.9	26.7	33.0	33.4	34.3	$y=23.654+0.2302x-0.0012x^2$	0.93*
2010/11	20.6	22.3	22.5	24.4	22.3	26.6	26.1	$y=21.198+0.0895x-0.0004x^2$	0.76*
			Cl	nange in co	lor of the b	erries (g kg	<sup>-1</sup> )	·	
2008/09	33.6	30.3	32.3	31.5	29.7	31.8	31.4	Ns	-
2009/10	19.5	21.1	21.1	22.0	21.7	19.6	22.8	Ns	-
2010/11	20.1	20.3	22.5	21.3	22.1	20.7	21.9	Ns	-

ns = not significant at 5% probability of error; \* = significant at 5% probability of error.

When the grape yield per hectare was compared to the total N content in the whole leaves collected at full flowering during the three crop seasons, the increase in the N content correlated with the increase in the N dose applied to the soil. This may be expressed using the quadratic equation (y=68.5208-3.6522x+0.0535x², R²=0.54\*). The highest grape yield was obtained with a total N content of 20.6 g kg⁻¹ (15.98 mg ha⁻¹) (Figure 1b). When N accumulated to greater than 20.6 g kg⁻¹ in the whole leaves, a reduction in grape yield was observed. Over the years, the N content increased in the leaves; however, grape yield was greater only in the years with less rainfall (Table 1).



**Figure 1.** The ratio between the grape yield and the dose of nitrogen applied to the soil (a) and between the yield and the total nitrogen in whole leaves collected at full flowering (b) in grapevines subjected to the application of 0, 10, 15, 20, 40, 80 and 120 kg N ha<sup>-1</sup> year<sup>-1</sup> during the 2008, 2009 and 2010 crop seasons. \* = significant at 5% probability of error.

#### Grape yield and its components

The application of increasing doses of N increased the grape yield per plant and per hectare in a quadratic manner in the 2008/09, 2009/10 and 2010/11 crop seasons. The yield was primarily increased due to an increase in the number of

clusters per plant and an increase in the weight of the clusters. There was no increase in the number of berries per cluster or in the 100-berry weight within the clusters (Table 3). The increase in grape yield and the positive impact on several parameters may be attributed to the N uptake from the soil from the applied urea. Brunetto et al. (2006b) reported using urea enriched with 15N isotopes and the N content in the whole leaf collected at full flowering was measured (Table 2). This is desirable because low organic matter content in the soil results in low N availability. Therefore, the annual application of N to the soil in the form of urea or an organic source (LORENSINI et al., 2012; NASCIMBENE et al., 2012) becomes necessary. This is especially important because approximately 22.28 kg N ha<sup>-1</sup> year-1 is exported annually via Cabernet Sauvignon grape clusters. The greatest grape yield per hectare in all crop seasons was obtained using a dose of 20 kg N ha<sup>-1</sup>, with a reduction in yield observed at greater doses (Figure 1a). These results are similar to those obtained by Brunetto et al. (2009) in Cabernet Sauvignon grapevines grown in an Udorthen soil subjected to N application in the Serra Gaúcha region of Rio Grande do Sul State, in which the authors also observed that the greatest grape yield was obtained in the plants provided with N applications of 15 to 30 kg N ha<sup>-1</sup>. The higher grape yield from the grapevines subjected to low doses of applied N, such as 20 kg N ha<sup>-1</sup>, may be attributed to the release of N derived from labile organic matter in the soil and from the residues of decomposition on the soil surface, such as pruned leaves and stems and decaying roots within the soil (BRUNETTO et al., 2011), because there was sufficient rain distribution, especially throughout the vegetative period of the grapevines (Table 1). It may also be attributed to biological N fixation by native or planted leguminous plant species that grow within the vineyards, as well as to the release of N contained in their tissues following decomposition on the soil surface (PATRICK et al., 2004; BRUNETTO et al., 2011).

It should be emphasized that doses greater than 20 kg N ha<sup>-1</sup> were not appropriate because they led to a reduction of grape yield per hectare (Figure 1a). This may be attributed to an increase in the vegetative growth of the plants, which may be measured by shoot growth or even by their mass at the time of winter pruning, and also to the high N content within the plant tissue, which increases the incidence of fungal diseases in the leaves and fertilized flowers (DUCHÊNE et al., 2001).

**Table 3.** Number of clusters per plant, average weight of the clusters, number of berries per cluster, 100-berry weight, grape yield per plant and grape yield per hectare in grapevines subjected to the application of 0, 10, 15, 20, 40, 80 and 120 kg N ha<sup>-1</sup> year<sup>-1</sup> during the 2008, 2009 and 2010 crop seasons.

C S			Dose	(kg N ha <sup>-1</sup>	year <sup>-1</sup> )			Farration	$\mathbb{R}^2$
Crop Season	0	10	15	20	40	80	120	Equation	K
					Yield	per plant (	kg)		
2008/09	2.5	2.7	3.4	3.2	2.7	2.6	2.6	$y=2.849+0.0033x-0.00005x^2$	0.25*
2009/10	1.0	1.1	1.1	1.2	0.7	0.6	0.6	$y=1.16-0.0097x 0.00004x^2$	0.70*
2010/11	3.6	3.9	5.1	5.0	4.8	3.4	3.4b	$y=4.1743+0.0185x-0.0002x^2$	0.34*
					Total	yield (Mg	ha-1)		
2008/09	9.4	10	12.5	11.8	9.9	9.5	9.7	$y=10.6406+0.0038x-0.0001x^2$	0.39*
2009/10	3.7	4.1	4.5	4.6	2.6	2.4	2.3	$y = 4.4060 - 0.0336x + 0.0001x^2$	0.82*
2010/11	13.2	14.6	18.9	18.6	17.9	12.6	12.5	$y=15.4344+0.0736x-0.0009x^2$	0.65*
					Numb	er of clust	ers per plant	t	-
2008/09	25.1	18.1	21.7	23.0	24.2	23.6	26.7	Ns	-
2009/10	34.1	30.7	28.3	32.9	23.9	24.0	28.6	$y=34.161-0.3006x+0.00212x^2$	0.77*
2010/11	31.7	29.9	33.9	35.0	33.2	24.2	31.6	$y=33.674-0.1094x+0.00065 x^2$	0.21*
					Weight	of the clu	sters (g)		
2008/09	99.6	149.2	156.7	139.1	111.6	110.2	97.4	$y=130.98+0.0771x-0.0032x^2$	0.34*
2009/10	29.3	35.8	38.9	36.5	29.3	25.0	21.0	$y=34.854-0.0465x-0.0006x^2$	0.70*
2010/11	113.6	130.4	150.4	144.6	144.6	140.5	109.7	$y=122.62+1.0564-0.0098x^2$	0.78*
					Numb	er of berri	es per cluste	er	-
2008/09	70.3	62.1	78.0	75.7	77.8	70.8	71.1	Ns	-
2009/10	36.2	43.5	43.4	43.0	38.0	34.0	34.0	Ns	-
2010/11	81.4	85.9	94.4	87.1	87.9	86.2	85.2	Ns	-
					100-berry	weight (g	)		-
2008/09	154.1	155.9	163.6	163.8	159.3	168.0	168.5	Ns	-
2009/10	118.0	109.4	114.8	108.6	103.4	100.1	102.6	Ns	-
2010/11	157.9	152.5	151.5	151.6	156.4	152.0	147.9	Ns	-

ns = not significant at 5% probability of error; \* = significant at 5% probability of error.

# **Must composition**

The application of N to the soil increased the N content in the must from the grape berries in a quadratic manner only in the second crop season evaluated, 2009/2010 (Table 4). Brunetto et al. (2007) obtained similar results in Brazil using must from Cabernet Sauvignon grapevines grown in a Typic Hapludalf soil that was subjected to increasing doses of applied N. Brunetto et al. (2008a) also achieved similar results utilizing must from two grapevine cultivars, Bordô and Couderc 13, which were subjected to applied N. Outside Brazil, in the United States (California), Ough et al. (1968) observed an increase in the total N content, ammoniacal N and biotin in grape must from grapevines subjected to N fertilization.

A higher N content in the must tends to reduce the probability of stuck fermentation because N has an impact on the microbial biomass, the fermentation rate and fermentation time (BELTRAN et al., 2005; JIRANEK et al., 1995). The total P content in the must was not affected by the N application during any of the three crop seasons, a result also obtained by Brunetto et al. (2007; 2008a). However, the total K contents in the must increased in the 2010/11 crop season as the dose of N applied to the soil increased. This may be because the plants supplied with additional N increase their leaf area, which thereby increases transpiration and forces K transport through the xylem and translocation via phloem to growing tissues. The K eventually reaches the berries because the plant has an

efficient uptake system for K, and its bioavailability is considered to be high (CECpH7.0 ≤ 5.0 cmolc dm-3 and available K 46-90 cmolc dm-3) in the soil utilized in the present study (CQFS-RS/SC, 2004). The potential increase in the K content in the must, in response to an increase in the N applied in the soil, is a disadvantage because higher K contents may lead to the formation of K bitartarate in the wine. The formation of K bitartarate may increase the pH of the wine, which will accelerate its oxidation and reduce its quality over time (MPELASOKA et al., 2003).

The application of N to the soil over the three crop seasons did not affect the pH or total acidity, or the tartaric and malic acid contents in the must of the Cabernet Sauvignon berries (Table 5). However, N application in the 2010/11 crop season led to an increase in the soluble solids in the must in a quadratic manner in which the highest values were observed at the lowest N doses (up to 20 kg N ha<sup>-1</sup>). Therefore, up to this dose level, there was an increase in the sugar values because the Brix scale represents approximately 90% of the sugars found in the must (BRUNETTO et al., 2009). The reduction in the soluble solids in the must of the berries from grapevines subjected to N doses higher than 20 kg N ha-1 may be due to increased vigor, especially of the aboveground grapevine tissues (SPAYD et al., 1994; DELGADO et al., 2004). The increased vigor promotes greater shading, and results in a less efficient photosynthetic area, which behaves as a photoassimilate sink (BORGHEZAN et al., 2014).

**Table 4.** Total content of nitrogen, phosphorus and potassium in the grape must of grapevines subjected to the application of 0, 10, 15, 20, 40, 80 and 120 kg N ha<sup>-1</sup> year<sup>-1</sup> during the 2008, 2009 and 2010 crop seasons.

0			Dose (	kg N ha <sup>-1</sup>	year-1)			Equation	R <sup>2</sup>
Crop season	0	10	15	20	40	80	120	•	
					Tota	al nitroge	n (%)		
2008/09	0.9	1.0	1.0	0.9	1.1	1.1	1.1	Ns	-
2009/10	3.4	3.0	3.7	4.3	3.6	6.0	3.0	$y=2.827+0.0687x-0.0005x^2$	0.51*
2010/11	1.9	2.1	2.7	3.1	2.7	2.8	2.2	Ns	-
					T	otal phos	ohorus (%)		
2008/09	0.1	0.1	0.1	0.1	0.1	0.8	0.1	Ns	-
2009/10	2.2	2.4	2.4	2.3	2.4	2.9	2.7	Ns	-
2010/11	0.2	0.2	0.2	0.2	0.2	0.2	0.2	Ns	-
					Tot	al potassi	um (%)		
2008/09	2.4	2.5	2.5	2.6	2.5	2.7	2.3	Ns	_
2009/10	0.9	1.0	1.0	1.0	1.0	1.0	1.0	Ns	-
2010/11	3.6	3.7	4.0	3.7	3.4	3.6	3.3	$y=3.728-0.0015x-0.00015x^2$	0.50*

ns = not significant at 5% probability of error; \* = significant at 5% probability of error.

**Table 5.** Soluble solids, pH, total acidity, tartaric acid and malic acid in the grape must of grapes from grapevines subjected to the application of 0, 10, 15, 20, 40, 80 and 120 kg N ha<sup>-1</sup> year<sup>-1</sup> during the 2008, 2009 and 2010 crop seasons.

0			Dose (kg l	N ha <sup>-1</sup> ye	ar <sup>-1</sup> )			P. J	R <sup>2</sup>
Crop season	0	10	15	20	40	80	120	Equation	R-
					So	luble sol	ids (°Brix)-		
2008/09	21.9	20.7	20.7	21.0	21.4	21.6	21.4	Ns	-
2009/10	15.4	18.8	18.8	18.1	16.9	17.8	18.5	Ns	-
2010/11	19.3	21.7	20.7	21.5	18.1	17.5	17.0	$y=21.022-0.0497x+0.000120x^2$	0.64*
						pH of	the must		
2008/09	3.7	3.8	3.9	3.8	3.8	3.8	3.8	Ns	-
2009/10	3.4	3.6	3.6	3.7	3.6	3.6	3.7	Ns	-
2010/11	4.0	4.0	4.0	4.0	4.0	4.0	4.0	Ns	-
					Tota	al acidity	(meq L-1)-		
2008/09	45.3	57.8	49.8	45.8	51.4	48.2	48.3	Ns	-
2009/10	122.1	117.3	109.2	83.9	87.5	83.7	91.3	Ns	-
2010/11	38.0	47.9	45.9	52.0	68.4	45.6	46.3	Ns	-
					Та	rtaric aci	d (g L-1)		
2008/09	4.4	4.8	4.2	4.7	4.6	5.6	4.4	Ns	-
2009/10	6.3	6.2	6.1	5.5	5.5	5.7	5.0	Ns	-
2010/11	6.3	6.2	6.1	5.5	5.5	5.7	5.0	Ns	-
					M	alic acid	(g L-1)		
2008/09	1.8	3.7	2.5	2.4	2.2	3.2	2.1	Ns	-
2009/10	6.6	5.9	6.6	5.6	4.8	4.9	4.5	Ns	-
2010/11	6.6	5.9	6.6	5.6	4.8	4.9	4.5	Ns	-

 $<sup>^{</sup>ns}$  = not significant at 5% probability of error;  $^{\star}$  = significant at 5% probability of error

The literature describing the effect of applied N on the composition of grape must, especially on pH, total acidity, malic acid and tartaric acid, is inconclusive because there are two reported scenarios. While some studies indicate that the addition of N may increase the pH, total acidity, malic acid and tartaric acid (BRUNETTO et al., 2009), other studies, such as that conducted by Spayd et al. (1994) in the central region of Washington in the United States (and which utilized increasing doses of N (0, 56, 112 and 224 kg N ha<sup>-1</sup>) on grapevines), indicate that the values of malic and tartaric acid in the must are not affected. The data from these latter studies are similar to the results of the study performed by Peacock et al. (1991), who reported that a dose of 50 kg N ha-1, applied in a parceled manner to grapevines in the United States during the phenological stages, did not affect the pH values in the must. In this study, the values for pH and total acidity were similar to those found in the must of the Cabernet Sauvignon cultivar in experiments carried out by and Rizzon and Sganzerla

(2007), which were carried out in the Serra Gaúcha region of Rio Grande do Sul State. The pH values of the must in the present study were above the indices considered to be satisfactory, i.e., from 3.1 to 3.6 (MOTA et al., 2009).

### Conclusion

Even in a sandy soil with a low organic matter content, the response to the application of N was low, and the highest Cabernet Sauvignon grape yield was obtained using an application of 20 kg N ha<sup>-1</sup> year<sup>-1</sup>. The application of N increased the nutrient content in the leaves collected at full flowering, but had little effect on the total nutrient content in the must. It did not affect the enological characteristics of the must, such as soluble solids, pH, total acidity, or the levels of malic acid and tartaric acid.

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