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## Predictive modelling of magnesium concentration in grapevine petioles as a basis for liming recommendations in vineyard acid soils

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### Summary

Soil acidification is a natural process which can either be accelerated by the activity of plants, animals and humans, but can be controlled through appropriate soil management. The main aim of this work was to develop a predictive modelling of magnesium concentration in grapevine petioles for liming amendment recommendation in vineyard acid soils. One liming material, dolomite, has been added to the soil at three doses: 0.9, 1.8 and 2.7 t CaCO<sub>3</sub> ha<sup>-1</sup>. Magnesium exchangeable content in soil surface and concentrations of this nutrient in petioles of leaf samples were investigated during three years. Exchangeable magnesium in soil tended to increase with increasing dolomite application rate. These increases were significant among all the doses and the control, except for the dose of 0.9. However, only between the highest dose and the control significant differences in magnesium concentration in petioles could be detected. In addition, one linear model has been proposed to make liming recommendations in vineyard acid soils based on petiole magnesium concentrations.

**Key words:** Amendment, dolomite, 'Mencía', veraison, linear.

### Introduction

Soil acidity is one of the most yield-limiting factors for grape production in high rainfall areas. Natural soil acidification is favoured in areas of high rainfall and ustic or udic moisture regimes, which facilitate the weathering and leaching of neutral and basic soil minerals and the release of hydrolytic cations in the soil solution, such as aluminium and iron (III). Aluminium (Al) toxicity is considered the main limiting factor for plant growth on acid soils (Kochian *et al.* 2005). Inadequate management increases soil acidity because of the greater solubilisation of Al. As the pH decreases, the Al<sup>3+</sup> species increases in concentration relative to the hydroxylated species, which favours a complete desorption and leaching of the rest of the exchangeable cations (Garzón *et al.* 2011). The low concentration of the macronutrient cations that results from this process is another limiting factor for grapevine growth on acid soils.

Liming is the most widely used long-term method of soil acidity amelioration. The application of adequate

quantities of liming materials to acid soils encourages various beneficial chemical and biological changes in the soil: liming improves the structural conditions (aeration) and increases the bioavailability of phosphorus (P), calcium (Ca), magnesium (Mg), and molybdenum (Mo) nutrients. Magnesium is less effectively bound than other bi- and trivalent cations, like Ca and Al; the low selectivity of exchange sites for Mg retention is one of the main reasons why acid soils are poor in adsorbed magnesium. Magnesium is a vital cofactor in the absorption of light energy by chlorophyll, being principally translocated in the phloem of vines (Zatloukalova *et al.* 2011).

In spite of the importance of the soil fertility, in vineyard production deficiency or surplus of macro- and micronutrients in grapevines is usually diagnosed by tissue testing, where the critical value is the nutrient concentration at which the vine no longer responds to further additions of that nutrient (White 2003). The dried blades/petioles are analyzed to detect the total amount of each macro- and micronutrient, and the results are compared with critical values.

The main objective of this investigation was to develop a predictive model to explain the content of magnesium in grapevine petioles as a basis for developing a liming recommendation system for Mediterranean conditions. This was carried out through a field study in which the effect of different doses of amendment dolomite, on the petiole magnesium contents at veraison stage in an acid soil dedicated to *Vitis vinifera* L. 'Mencía' cultivation, was studied. However, the effect of dolomite on the exchangeable magnesium content in soil surface has also been studied.

### Material and Methods

**Study site:** A commercial vineyard, which was located 560 m above sea level in the municipality of Villafraanca del Bierzo (León; Spain) with geographic coordinates of 42°35'N latitude and 6°46'W longitude, was selected as the study site. The area experiences a semi-arid climate according to the Thornthwaite classification. The mean annual reference evapotranspiration (FAO Penman-Monteith) is 922 mm, while the mean annual rainfall is 616 mm (estimation performed for the period 2000-2011) (SIAR, 2012). From a bioclimatic point of view, the site would be classified as upper meso-Mediterranean based on the thermotypes classification and subhumid according to the ombrotypes classification (IGME, 1995). The soil under

study corresponds to an Inceptisol, suborder Xerept, group Haploxerept according to soil taxonomy (IDEE 2012).

The research was conducted on the *Vitis vinifera* L. 'Mencia' variety grafted on a Richter 110 rootstock with an age of 75 years. Planting lines displayed an east-west orientation. The conduction system involved a head training, and there were 4-5 arms per plant. Winter pruning left a thumb-sized arm with two buds. There was no summer pruning. The vineyard had no irrigation system support. No fertilisers or extra amendments other than that proposed in this research were implemented during the study period. Similarly, during the years of monitoring, the soil on which the vineyard grew did not undergo any type of tillage.

**Liming dose:** The liming doses used in the trial were chosen from those routinely employed by local winegrowers. Doses of 0.9, 1.8 and 2.7 t CaCO<sub>3</sub>·ha<sup>-1</sup> were tested, which corresponded to 0.89, 1.79, and 2.68 t of dolomite·ha<sup>-1</sup>. The dolomite was in a powdery state. In March 2006, after tillage to a depth of approximately 25 cm, the amendment was manually applied and then incorporated into the soil using a second tillage pass. Dolomite was provided by Calfensa Projects, S. L. (Santa Comba; Lugo; Spain).

**Statistical design of the experiment:** Liming materials was applied at three doses (0.9, 1.8 and 2.7 t CaCO<sub>3</sub>·ha<sup>-1</sup>) with three repetitions per dose, providing 9 subplots (3 × 3). An additional three subplots were used as controls, yielding a total of 12 subplots. The treatments were distributed among the subplots in a completely random design. Each subplot was approximately 24 m<sup>2</sup> and consisted of eight vines planted in a line, although only the four middle plants were sampled to avoid edge effects. For the same reason, the 12 subplots were distributed in three plant lines that were separated by three untreated buffer lines.

**Leaf and soil samples:** After the amendment was added, the effect of the liming on the magnesium concentrations in soil (Mg<sub>s</sub>) and petioles (Mg<sub>p</sub>) was monitored for three years (2006, 2007 and 2008). Leaf sampling (30 petioles per sample from basal leaves opposite bunches) was carried out in these years at veraison (late August), whereas soil sampling was conducted by sampling soil (depth of 0-30 cm) at five different phenological stages that included budding, flowering, fruit set, veraison, and leaf drop. The leaf samples were analysed by inductively coupled plasma atomic emission spectroscopy (ICP-AES) after wet digestion.

Air-dried soil samples were sieved to a diameter of 2 mm Ø. Textural classes were determined by the Bouyoucos hydrometer method (1962). Official methods of analysis (MAPA, 1993) were used for the determination of (i) the organic matter (OM) (organic carbon was determined according to the wet oxidation method and the organic carbon was multiplied by 1.724 to estimate the organic matter), (ii) the pH in a soil:water (1:2.5) suspension and in a 1:2.5 suspension of soil in 1 N KCl, and (iii) the content of exchangeable cations (Ca, Mg, and K) by extraction of the cations with successive aliquots of 1 N ammonium acetate and subsequent analysis by atomic absorption spectroscopy (AAS). The Al exchange was determined by inductively

coupled plasma atomic emission spectroscopy (ICP-AES) using 1 N KCl as the extraction agent (Little, 1964). The P levels were determined by visible molecular absorption spectroscopy after extraction with successive aliquots of sodium bicarbonate following the method proposed by OLSEN *et al.* (1954). The percentage of base saturation was calculated from the relationship between the sum of the contents of the exchangeable cations (Ca, Mg, and K) and the ECEC. The ECEC was obtained from the arithmetic sum of the concentrations of the exchangeable Ca<sup>2+</sup>, Mg<sup>2+</sup>, K<sup>+</sup>, and Al<sup>3+</sup>.

**Statistical analysis:** Data from each of the liming doses were tested for normality using the Shapiro-Wilk test and probability-probability (P-P) plots displaying the distribution frequency of the data. In addition, identification of potential outliers was analysed. An analysis of variance (ANOVA) was performed to compare the means of Mg<sub>p</sub> and Mg<sub>s</sub> at each of the levels of liming dose factor. Tukey's HSD test (Honestly Significant Difference) values were calculated at the 5 % probability level to facilitate comparison between the treatment means. In addition, the variance of the Mg<sub>p</sub> variable was decomposed into levels of the independent variable (liming dose) in linear, quadratic, and cubic terms to find the most appropriate model (WEBSTER 2007). The statistical analyses were performed using IBM SPSS v19 software.

## Results and Discussion

**Characterization of the liming material and the soil before liming:** Tab. 1 shows the mineral composition of the liming material used in this study (VILLA 2005), whereas Tab. 2 shows the baseline analytical data for the first 30 cm of the soil profile subsequently sampled in the study. The aluminum saturation (Al/ECEC) was very high at the start of the investigation (76 %). The acidity and the presence of Al<sup>3+</sup> in the soil were clearly shown by the difference between the pH (H<sub>2</sub>O) and the pH (KCl). The difference between the two values is an indicator of interchangeable acidity, and this difference was close to one. Low concentrations of the exchangeable Ca and Mg were evident, and the concentration of P was suboptimal for the grapevine cultivation.

**Petiole and soil Mg concentrations:** Tab. 3 shows the mean values and standard deviations for Mg<sub>p</sub> and Mg<sub>s</sub> monitored in the test plots during the three years of the trial. Exchangeable Mg<sub>s</sub> increased with increasing dolomite application rate. Mg<sub>s</sub> in dose of 2.7 (0.54) was significantly higher than that in the others, whereas Mg<sub>s</sub> in

Table 1

Chemical composition of the liming material (dolomite) expressed as dry matter

	CaO <sup>†</sup>	MgO <sup>†</sup>	Na <sub>2</sub> O <sup>†</sup>	K <sub>2</sub> O <sup>†</sup>	Al <sup>‡</sup>	OM <sup>†</sup>
Dolomite	310.9	184.0	1.2	3.5	9529	0.0

<sup>†</sup>in g·kg<sup>-1</sup>; <sup>‡</sup>in mg·kg<sup>-1</sup>.

Table 2

Baseline soil characteristics and analytical data before liming for soil surface (0-30 cm)

Depth	Sand <sup>†</sup>	Silt <sup>†</sup>	Clay <sup>†</sup>	Textural class (U.S.D.A.)				OM <sup>†</sup>
0-30 cm	52.6	26.0	21.3	Sandy clay loam				0.77
	pH <sub>w</sub>	pH <sub>k</sub>	EC <sup>‡</sup>	P <sup>§</sup>	Ca <sup>p</sup>	Mg <sup>p</sup>	K <sup>p</sup>	Al <sup>p</sup>
0-30 cm	4.56	3.73	0.04	6.04	0.27	0.16	0.18	1.88

<sup>†</sup>in % g·kg<sup>-1</sup>; <sup>‡</sup>in dS·m<sup>-1</sup>; <sup>§</sup>in mg·kg<sup>-1</sup>; <sup>p</sup>in cmol(+)-kg<sup>-1</sup>.

Table 3

Magnesium concentrations in petioles (Mg<sub>p</sub>) and soil surface (Mg<sub>s</sub>). Means that are followed by the same letter do not differ at p = 0.05

Lime treatment	Dose <sup>†</sup>	Statistic	Mg <sub>p</sub> <sup>‡</sup>	Mg <sub>s</sub> <sup>§</sup>
Control	0	Mean	0.67b	0.18c
		Standard deviation	0.19	0.13
Dolomite	0.9	Mean	0.69ab	0.26bc
		Standard deviation	0.16	0.18
	1.8	Mean	0.75ab	0.31b
		Standard deviation	0.28	0.16
	2.7	Mean	0.96a	0.54a
		Standard deviation	0.21	0.31

<sup>†</sup>in t CaCO<sub>3</sub> ha<sup>-1</sup>; <sup>‡</sup>mean in %; <sup>§</sup>in cmol(+)-kg<sup>-1</sup>

dose of 1.8 (0.31) only was significantly higher than that in control (0.18). In addition Mg<sub>s</sub> in dose of 0.9 (0.26) was not significantly higher than that in control. The increase of the amount of exchangeable Mg by the application of dolomite to the soil surface horizons has previously been encountered in field tests by ILLERA *et al.* (2004) and VIDAL *et al.* (2006). Contents of exchangeable magnesium in subsoil were not determined. However, almost all reports on surface dolomitic liming experiments agree that these positive effects on Mg<sub>s</sub> concentrations are restricted to a few cm depth of the profile; in most cases to the upper few cm of the top mineral soil (MATZNER and MEIWES 1991).

Dolomite treatment had significant effect on Mg<sub>p</sub> between control (0.67) and the highest dose (0.96), although Mg<sub>p</sub> in 0.9 (0.69) and 1.8 (0.75) doses were not significantly higher than that in control. These results are in opposition with the pattern for Mg to decrease with liming according to the results shown by WOOLDRIDGE *et al.* (2010). However, the trial of these researchers was carried out with calcitic lime, so antagonism between Ca and Mg during root uptake must be taken in account in this case. The storage and mobilization of nutrients in the woody parts of the plant, would explain the occasional delay in vine response (Mg<sub>p</sub> levels) to dolomitic liming.

Development of the Mg<sub>p</sub> predictive model: A polynomial contrast for the Mg<sub>p</sub> was performed. Results are shown in Tab. 4. The value of the F statistic for the Mg<sub>p</sub> dose-dependent regression model

was significant, F = 3.4 (p < 0.05). Sum of squares was decomposed into a linear term and a nonlinear term. The linear term produced a significant F-statistic (p < 0.01), which suggested that a predictive model for the Mg<sub>p</sub> as a linear function of the dose would be appropriate. In addition, the deviation from linearity was not significant, indicating that a quadratic term does not explain a significant portion of the variance that remained unexplained after including only the linear term. Accordingly, a first order linear model was chosen as the most appropriate Mg<sub>p</sub> predictive model.

The predictive model is aimed at estimating the Mg concentration in leaf petioles from the CaCO<sub>3</sub> dose in the liming material. The linear model was developed, and the 95 % confidence intervals were calculated for the predicted values (Tab. 5). In the proposed models, the coefficient of determination R<sup>2</sup> represents the variance in the Mg<sub>p</sub> that was explained by the predictive model. The coefficient of determination obtained was low because of the disper-

Table 4

Analysis of variance for the Mg<sub>p</sub> response to four levels of lime after three years

Source	df	SS	MS	F	Sig.
Treatments	3	0.47	0.16	3.4	*
Linear regression	1	0.39	0.39	8.4	**
Deviations from linear regression	2	0.08	0.04	0.9	0.41
Quadratic regression	1	0.08	0.08	1.7	0.20
Residual	32	1.47	0.05		
Total	35	1.94			

df: degrees of freedom; SS: sum of squares; MS: mean square.

\*significant at the 0.05 probability level (p < 0.05);

\*\*significant at the 0.01 probability level (p < 0.01);

\*\*\*significant at the 0.001 probability level (p < 0.001).

Table 5

Linear model for prediction of Mg<sub>p</sub> content (in %)

Model	Equation	R <sup>2</sup>
95 % CI (Upper)	Mg <sub>p</sub> = 1.07 + 0.10 LQ	
Proposed model	Mg <sub>p</sub> = 0.63 + 0.10 LQ	0.20
95 % CI (Lower)	Mg <sub>p</sub> = 0.18 + 0.10 LQ	

CI: confidence interval; LQ: quantity of calcium carbonate in t·ha<sup>-1</sup>

sion of the petioles Mg concentrations data for each liming dose. Thus, in the model proposed, the CaCO<sub>3</sub> dose could explain 20 % of the variance in the Mg<sub>p</sub> values. If achieve a concentration of magnesium in petiole of 1 % is fixed as an adequate level in veraison to avoid nutritional deficiencies (FREGONI cited by BAVARESCO *et al.* 2010), our model predicts that the dose of CaCO<sub>3</sub> required would be 3.7 t CaCO<sub>3</sub>·ha<sup>-1</sup> that is far from the higher liming doses tested in the trial.

### Conclusions

The results from this field trial have shown the positive effects of dolomitic liming with respect to increasing soil exchangeable magnesium. However, effect on magnesium concentrations in petioles were significant only with the highest dolomite dose, which confirms that although nutrient availability is primarily dependent on the mineral makeup of the soil, nutrient uptake has a physiological basis.

The relationship between the magnesium concentration in petioles and the dolomite dose proved to be linear; therefore, the best-fitting model used a first order linear model. However, a more accurate model, in terms of variance explained, would require testing higher dolomite doses.

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