Application of Sugar Foam to Vineyard Acid Soils under Mediterranean Conditions

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Calcium (Ca) deficiency and aluminium (Al) toxicity are considered major chemical constraints that limit vine growth in vineyard acid soils under Mediterranean conditions. The main aim of this work was to evaluate the effects of three doses of sugar foam (900, 1 800 and 2 700 kg $CaCO_3/ha$) on five soil properties (pH in water, Ca, Mg, K and Al exchangeable contents) and Ca concentrations in petioles and canes of one wine grape variety. Research was conducted over three seasons (2006 to 2008) in a random block design at León (Spain). The results for plants and soils were subjected to general lineal model analysis. As a result, one linear model, validated through a cross-validation procedure, has been proposed to predict exchangeable calcium levels in vineyard acid soils at véraison. Based on the results for the soils, the higher doses of sugar foam (1 800 and 2 700 kg $CaCO_3/ha$) were the most effective doses in decreasing Al exchangeable and increasing Ca exchangeable contents. In addition, liming increased Ca content in petioles and decreased Ca content in canes, but in both cases these differences were not significant.

INTRODUCTION

Soil acidity is a major limitation to soil productivity in many regions around the world. The adjustment and maintenance of soil acidity to a level suitable for grape production is an important first step in the management of an acid soil dedicated to *Vitis vinifera* L. Soil acidity can affect various factors that contribute to the biological, chemical and physical properties of soil that make it best able to fulfil its desired purpose. Grape production systems undergo accelerated soil acidification as a consequence of anthropogenic inputs and outputs. The resultant decline in soil pH associated with grape production systems may be sufficient to cause moderate to severe Al and manganese (Mn) toxicity, thereby affecting the long-term economic viability of vineyard production and resulting, in some cases, in permanent degradation of the soil as a resource base (Sumner & Noble, 2003).

Aluminium toxicity is recognised as the main limiting factor for plant growth on acid soils (Kochian *et al.*, 2005). Although there are no identifiable symptoms of Al toxicity suitable for use as a diagnostic indicator, the most apparent effects are a reduction in both root and shoot growth (Menzies, 2003). However, the Al concentration in the soil solution is not the only important growth-limiting parameter in grape production on acid mineral soils. Instead,

toxicity is modulated by Al speciation and the presence of Ca (Marschner, 2012). Thus, because Ca deficiency in grapevine occurs primarily on soils with a very low pH because the highly toxic Al becomes very soluble at pH < 5.0 and blocks Ca uptake, liming is an important practice to achieve profitable grape production in vineyards established on acid soils.

Liming is the most widely used long-term method of soil acidity amelioration. The addition of limestone materials to the soil increases the pH above 5.5, which promotes the precipitation of Al as $Al(OH)_3$ and causes its elimination from the soil solution and the exchange complex. In addition to reducing the Al content of the soil, 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), Ca, magnesium (Mg), and molybdenum (Mo) (Fageria & Baligar, 2008).

Modern viticulture requires the implementation of efficient, sustainable and environmentally sound management practices. In this context, sugar foam is a relatively recently used organic liming material that has emerged because of the development of the beet sugar industry (García *et al.*, 2009). Sugar foam is highly rich in active lime. In addition,

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it contains abundant organic matter, as well as other nutrient elements such as P, iron (Fe), copper (Cu) and zinc (Zn) (Vidal *et al.*, 2006). In recent years, this residue obtained from the manufacturing of sugar has increasingly been recognized as a serious alternative as a liming material in Spain to improve the agro-soil-plant system in its chemical (García *et al.*, 2009; Pérez de los Reyes *et al.*, 2013a) and physical properties (Pérez de los Reyes *et al.*, 2011).

The main objective of this study was to evaluate the effect of different doses of sugar foam amendment on the pH in water and the exchangeable cation contents, as well as on petiole Ca contents at véraison in an acid soil dedicated to *Vitis vinifera* L. cv. 'Mencía' cultivation over three years. In addition, a predictive model to explain the content of exchangeable Ca at véraison as a basis for developing liming recommendations and determining cane Ca contents at leaf fall was evaluated.

MATERIALS AND METHODS **Study site**

A commercial vineyard, located 560 m above sea level in the municipality of Villafranca del Bierzo (León, Spain), with geographic coordinates of 42°35'N latitude and 6°46'W longitude, was selected as the study site. The mean annual reference evapotranspiration (FAO Penman-Monteith) is 922 mm, while the mean annual rainfall is 616 mm; the mean monthly temperature varies from 20.0°C (July) to 4.5°C (January) (estimations performed for the period 2000 to 2011) (SIAR, 2012). From a climatic point of view, the grape-growing region would be classified as Region I (\leq 1 390 Celsius degree-days) based on the system devised by Amerine and Winkler (Jackson, 2008). The soil under study corresponds to an Inceptisol, suborder Xerept, group Haploxerept according to the Soil Survey Staff (2010), and to Cambisol Dystric according to the FAO (2006).

The research was conducted on the *Vitis vinifera* L. cv. Mencía variety grafted onto a Richter 110 rootstock with an age of 75 years. Planting lines displayed an east-west orientation. The conduction system involved head training, and there were four to five spurs 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 and during the years of monitoring, and the soil on which the vineyard grew did not undergo any type of tillage. A traditional vineyard soil management practice of local growers in vine rows (using herbicide in the spring before budburst) was implemented annually to alleviate weed competition.

Sugar foam dose

The liming doses used in the trial were chosen from those routinely employed by local winegrowers. Doses of 900, 1 800 and 2 700 kg calcium carbonate equivalent (CCE, defined as the acid-neutralising capacity of the material regarding pure $CaCO_3$) per ha were tested, which corresponded to 1 177, 2 354 and 3 531 kg of sugar foam per ha. The sugar foam consisted of variable diameter aggregates, which were manually disaggregated before

incorporation into the soil. 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. The sugar foam was provided by the La Bañeza (Leon, Spain) sugar factory, which is owned by British Sugar.

Statistical design of the experiment

The sugar foam was applied at three doses (900, 1 800 and 2 700 kg CaCO₃/ha) with three repetitions per dose, providing nine subplots (3×3). In addition, three subplots were used as controls, yielding a total of 12 subplots. The treatments were distributed among the subplot in a completely randomised block design. Each subplot was approximately 24 m² and consisted of eight vines planted in a line, although only the four middle plants in every subplot 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. The soil and plant data were tested for univariate normality (by the Shapiro-Wilk test), and then subjected to univariate analysis of variance using the general linear means procedure. The statistical analyses were performed using R 3.0.1. (R Core Team, 2013).

Soil and plant analysis

After the sugar foam was added, the effect of the liming on the following soil properties was monitored for three years (2006 to 2008): pH in water (pHw) and exchangeable Ca, Mg, K and Al saturation (CaECEC, MgECEC, KECEC and AlECEC, respectively). This monitoring was conducted by sampling soil a at depth of 0 to 30 cm at the véraison phenological stage. A total of three samplings (late August) were done over the study period. The 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) soil organic matter (SOM), (ii) pHw and electrical conductivity in a soil:water (1:2.5) suspension, and (iii) exchangeable bases, which were extracted with 1 M NH, AcO at pH 7 and quantified by atomic absorption spectrometry (Ca and Mg) or flame emission spectrometry (K). The exchangeable Al 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 effective cation exchange capacity (ECEC) was obtained from the arithmetic sum of the concentrations of the exchangeable Ca²⁺, Mg²⁺, K⁺, and Al³⁺, whereas CaECEC, MgECEC, KECEC and AlECEC, were calculated from the relationship between the content of each of the exchangeable cations and the ECEC.

Calcium concentrations in petioles (Ca_p) were evaluated through leaf sampling (30 petioles per subplot from basal leaves opposite bunches) at véraison (late August), whereas calcium concentrations in canes without bark (Ca_c) were evaluated through cane sampling (15 canes per subplot) at leaf fall (early November). Both leaf and cane samples were analysed by inductively coupled plasma atomic emission spectroscopy (ICP-AES) after wet digestion.

RESULTS AND DISCUSSION

Characterisation of the liming material and the soil before liming

Table 1 shows the mineral composition of the liming material used in this study (Villa, 2005). The Ca content of the sugar foam is due mainly to the presence of Ca in the form of slaked lime $[Ca(OH)_2]$ and, to a lesser extent, as carbonate $(CaCO_3)$ (Espejo, 2001).

Table 2 shows the baseline analytical data for the first 30 cm of the soil profile subsequently sampled in the study. The aluminium saturation (Al/ECEC) was very high at the start of the study (76%) for the first 30 cm of the soil profile. In addition, low percentages of the exchangeable Ca and Mg saturation were evident.

Soil exchange complex

There was a highly significant effect of sugar foam dose on levels of CaECEC [F(3, 32) = 12.2, P < 0.001] and AlECEC [F(3, 32) = 6.6, P < 0.01]. F-ratios were large enough to be statistically significant in these soil properties, so post hoc tests (Tukey's honestly significant difference test) were done to know where the differences lay between groups. Sugar foam dose had no significant effect on pHw [F(3, 32) = 2.1, P = 0.12], MgECEC [F(3, 32) = 2.6, P = 0.07] and KECEC [F(3, 32) = 1.8, P = 0.17].

Table 3 shows the post hoc tests between the soil property means. At a depth of 0 to 30 cm, CaECEC tended to decrease significantly in the sequence 2 700/1 800 > 900/control, whereas AIECEC tended to increase significantly in the sequences 2 700 < 900/control and 1 800 < control. The

sugar foam doses had no significant effects on pHw, MgECEC and KECEC at the depth (0 to 30 cm) studied. The overall effects of the higher doses of sugar foam (2 700 and 1 800 kg CaCO₂/ha) on the exchange complex are consistent with those found by Illera et al. (2004) and Vidal et al. (2006). Although the pHw does not change significantly, we can conclude that these agro-industrial products have a positive final effect on the quality of the acidic soil and can be considered as effective alternatives for traditional liming materials. In addition, although vine root depth was not measured, it could conceivably be hypothesised that an increase of CaECEC at the studied depth (and probably in the subsoil by leaching of Ca) would be able to promote the penetration of vine roots into subsoil layers. The latter is in accordance with the fact that the uptake of Ca required for root growth is particularly problematic in acidic soils because of (i) poor physiological status of roots due to Al toxicity and (ii) relatively low Ca concentration in the soil solution (Marschner, 2012).

The potential impact of the toxic trace elements accumulated in the so-called sugar foam in soils used for grape production is undoubtedly of interest. It is important to bear in mind that high levels of trace elements in viticultural soils, such as cadmium (Cd), chromium (Cr), nickel (Ni) or lead (Pb), among others, can reduce vine growth or establish a serious risk for contamination of the food chain through bioaccumulation in the grapes (Naidu *et al.*, 2004). Although it was not possible to investigate if the addition of sugar foam caused an increase in the content of soil trace elements, previous research by Pérez de los Reyes *et al.* (2013) with

TABLE 1

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|----------|-------------|--------------|----------------|--------|--------------|
| Chemica | COMPOSITION | of the sugar | foam expressed | i as c | II V Matter. |
| | | | | | |

| Treatment | CaO | MgO | Na ₂ O | K ₂ O | Al | OM | CCE |
|------------|--------|--------|-------------------|------------------|---------|--------|--------|
| | (g/kg) | (g/kg) | (g/kg) | (g/kg) | (mg/kg) | (g/kg) | (g/kg) |
| Sugar foam | 403.8 | 14.5 | 0.4 | 0.9 | 2 469 | 79.0 | 757 |

OM: Organic matter; CCE: Calcium carbonate equivalent.

TABLE 2

Baseline soil characteristics of soil surface (0 to 30 cm) before liming.

| Depth | Sand (%) | Silt (%) | Clay (%) | Textural class (USDA) | | | SOM (%) | |
|---------|-----------------|----------|----------|-----------------------|------------|-----------|----------------|-------------------|
| 0-30 cm | 52.6 | 26.0 | 21.3 | Sandy clay loam 0.77 | | | 0.77 | |
| | рН _w | EC (d | dS/m) | CaECEC (%) | MgECEC (%) | KECEC (%) | AIECEC (%) | ECEC (cmol(+)/kg) |
| 0-30 cm | 4.56 | 0. | 04 | 10.70 | 6.40 | 7.60 | 75.30 | 2.49 |

TABLE 3

Effect of the sugar foam dose (in kg CaCO₃ per ha) on pHw and exchangeable complex, 2006 to 2008.

| Dose | pHw | CaECEC (%) | MgECEC (%) | KECEC (%) | AIECEC (%) |
|---------|------------------|-----------------|-----------------|----------------|-------------------|
| Control | $4.56\pm0.05a$ | $14.6\pm2.1b$ | $6.2 \pm 1.2a$ | $8.0 \pm 1.7a$ | $71.2 \pm 3.9a$ |
| 900 | $4.73\pm0.08a$ | $20.0\pm4.2b$ | $8.6 \pm 1.9a$ | $6.3 \pm 0.8a$ | 65.2 ± 6.3 ab |
| 1800 | $4.74\pm0.07a$ | $32.4 \pm 3.3a$ | $9.2 \pm 1.0a$ | $5.4 \pm 0.6a$ | $53.0 \pm 3.8 bc$ |
| 2700 | $4.77 \pm 0.06a$ | $36.4 \pm 1.3a$ | $11.1 \pm 0.3a$ | $4.9 \pm 0.5a$ | $47.6 \pm 1.6c$ |

Means (\pm standard error) in the same column that are followed by the same letter do not differ at P = 0.05.

sugar foam showed that, after an amendment of an acidic vineyard soil (Tipic Palexerult according to Soil Survey Staff, 2010), levels of barium (Ba), rubidium (Rb) and strontium (Sr) were increased in the amended soil in comparison to the original soil. In accordance with viticultural concepts of vineyard soil quality and sustainability, future studies on the current topic therefore are recommended.

Development of the CaECEC predictive model

A predictive model to explain the content of calcium in the exchange complex at véraison as a basis for developing a liming recommendation system was developed. We proceeded to model the CaECEC as a function of the sugar foam dose in kg CaCO₃/ha. The sum of squares was decomposed into a linear, quadratic and cubic term. Powers of dose beyond a one-order polynomial did not make significant contributions to explaining the variation in Ca in the exchange complex at véraison. Only linear terms produced a significant F-statistic (P < 0.001) (Table 4).

The Akaike Information Criteria (AIC) provides another method for comparing models. Models with AIC not differing by 2 should be treated as equally adequate, but the one with the lower AIC should be adopted (Chatterjee & Hadi, 2006). In addition, the selection of a final regression model involves a compromise between predictive accuracy and parsimony. Thus, a linear model (AIC = 262.2) was selected as being more adequate than a quadratic model (AIC = 264.1) and a cubic model (AIC = 264.6).

In the proposed model, the coefficient of determination, R^2 , represents the variance in the CaECEC that was explained by the predictive model. Thus, in the linear model proposed, the sugar foam dose could explain 51% of the variance in the CaECEC values. Fig. 1 shows a plot of the fitted linear model with prediction (inner) and confidence (outer) bands. The equation for the linear model including the sugar foam doses (D, in kg CaCO₃/ha) is:

$$CaECEC (\%) = 14.1 + 0.016 D$$
 (1)

When prediction is the primary goal in a regression equation, the selection and interpretation of the regression model does not signal the end of the labour. Instead, methods for assessing model generalisability should be considered. Thus, to assess the generalisability of the linear model proposed in this work, a cross-validation method was developed. In k-fold cross-validation, the initial data are randomly partitioned into k mutually exclusive subsets or folds, each of approximately equal size. Training and testing are performed k times. We used the commonly accepted 10-fold application (Harrell, 1998), so, in this situation, the model was trained using nine of the tenths and prediction error was computed on the remaining one tenth. The root of the cross-validation residual sums of squares, which is a corrected measure of prediction error averaged across all folds, from the CaECEC levels obtained from the proposed model was 8.9%.

If achieving a percentage of CaECEC in ECEC of 70% is fixed as a desirable level for grapevine performance (Abbott, cited by Hazelton & Murphy, 2007), our model predicts that the dose of sugar foam required would be 3 494 kg $CaCO_3$ /ha, that is far from the higher liming doses tested in the trial.

However, because soils are extremely diverse and their characteristics have large time and spatial variability, it would be necessary to verify all the results obtained in this work for their suitability for practical application through long-term field experiments. In addition, soil processes are strongly dependent on weather conditions in a given year (Körschens, 2006). Thus, although the findings of this work may help us to understand the practice of liming with sugar foam in terms of making decisions about the need for lime, more research on this topic therefore is recommended to understand how environmental changes or soil variability may influence the results.

Petiole and cane Ca concentrations

Table 5 shows the mean values for Ca_p and Ca_c monitored in the test plots during the three years of the trial. Sugar foam dose had no significant effect on Ca_p [F(3, 32) = 2.2, P = 0.11] and Ca_c [F(3, 32) = 0.5, P = 0.72]. However, Ca_p increased with increasing sugar foam application rate in the sequence Control < 500 < 1 000 < 1 500, whereas Ca_c decreased with increasing sugar foam application rate in the sequence Control > 900 > 1 800/2 700.

In the same way as in this research, Wooldridge *et al.* (2010) previously reported an increase in the calcium petiole concentration by the application of liming materials in field tests. However, the trial of these researchers was conducted at fruit-set. If achieving a concentration of calcium in the petiole of 1.5% is fixed as an adequate level at véraison to

TABLE 4

Analysis of variance for the CaECEC response to four levels of sugar foam over three years, 2006 to 2008.

| Source | Degrees of freedom | Sum of squares | Mean square | F | Significance |
|----------------------|---------------------------|----------------|-------------|------|--------------|
| Sugar foam dose | 3 | 2 852 | 951 | 12.2 | *** |
| Linear regression | 1 | 2 740 | 2 740 | 35.3 | *** |
| Quadratic regression | 1 | 3.99 | 3.99 | 0.05 | 0.82 |
| Cubic regression | 1 | 108 | 108 | 1.38 | 0.25 |
| Residual | 32 | 2 485 | 77.7 | | |
| Total | 35 | 5 337 | | | |

* 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).

avoid nutritional deficiencies (Fregoni, cited by Bavaresco *et al.*, 2010), none of the petiole Ca concentrations in the test plots were in adequate nutritional ranges. This is consistent with the suboptimal percentages of exchangeable calcium (CaECEC) in all the sugar foam doses tested.

Figure 2 shows the relationship between Ca_p concentration and CaECEC at véraison with dose of sugar foam (in kg CaCO₃/ha) as a conditioning variable. As can be

seen, Ca_p increases as CaECEC increases. Consequently, the largest concentrations of Ca_p were obtained with sugar foam doses that were more effective in increasing Ca saturation in the soil exchange complex, concomitant in the subplots with a major decrease of Al saturation in the soil exchange complex.

Figure 3 shows the average total Ca_c concentration for each sugar foam dose obtained in the three cropping years

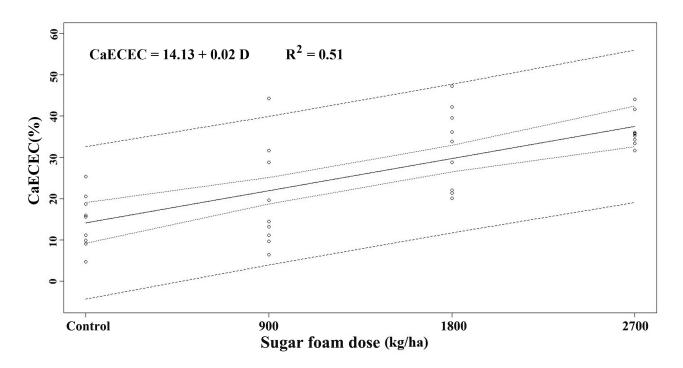
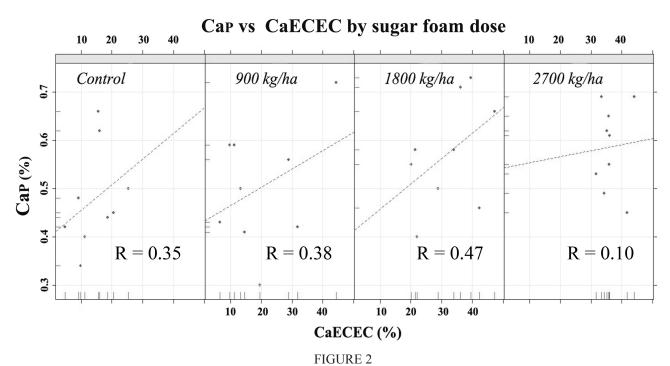


FIGURE 1 Linear model of exchangeable Ca (CaECEC) prediction with confidence and prediction limits (2006 to 2008). Sugar foam dose (D) in kg CaCO,/ha.



Relationship between calcium concentration in petioles (Ca_p) and exchangeable calcium (CaECEC) at véraison by sugar foam dose (kg CaCO₃/ha), 2006 to 2008. R is the bivariate correlation coefficient.

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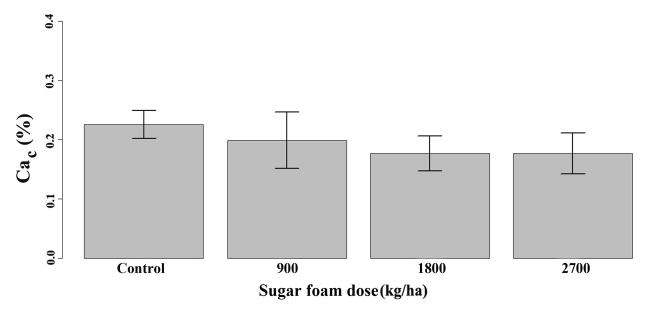


FIGURE 3

Average values of calcium concentrations in canes (Ca_c, as dry matter percentage) during the three cropping years (2006 to 2008) by sugar foam dose (kg CaCO,/ha). Standard errors (SE mean) are shown as bars ($\pm 1 \times$ SE mean).

| TABLE 5 |
|---|
| Effect of the sugar foam dose (in kg CaCO ₃ per ha) on Ca _p |
| and Ca _c , 2006 to 2008. |

| Dose | Ca _p (%) | Ca _c (%) |
|---------|---------------------|---------------------|
| Control | $0.48\pm0.03a$ | $0.23 \pm 0.02a$ |
| 900 | $0.50\pm0.04a$ | $0.20\pm0.05a$ |
| 1800 | $0.57\pm0.04a$ | $0.18\pm0.03a$ |
| 2700 | $0.59\pm0.03a$ | $0.18 \pm 0.03a$ |

Means (\pm standard error) in the same column that are followed by the same letter do not differ at P = 0.05.

(2006 to 2008). In contrast to the effect that normally would be expected after liming, Ca_c decreased with increasing sugar foam dose. However, the effects of liming on the calcium levels of canes have not been studied to date. Notwithstanding the foregoing, these data must be interpreted with caution because it is important to bear in mind the chemical digestion carried out in this investigation (an acid mixture of perchloric, sulphuric and nitric acid). It therefore may be the case that another procedure of cane digestion (pressure digestion or microwave digestion) will show different results on Ca_c than those obtained in this work. Thus, future research should be concentrated in this area.

CONCLUSIONS

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The field application of sugar foam increased the amount of Ca and decreased that of Al in the exchange complex of the surface horizon in an acid soil dedicated to grapevine cultivation. A linear model is proposed to represent the variance in the CaECEC at véraison. The coefficient of determination obtained was moderate. A more accurate model, in terms of variance explained, would require testing higher sugar foam doses. Based on the results of the threeyear experiment, liming with sugar foam did not significantly affect Ca contents in petioles and canes. However, the calcium content in the petioles was higher in soils treated with sugar foam. In this way, the use of sugar foam is an interesting industrial by-product alternative to other liming materials. The results of the Ca content in canes were controversial. Future research therefore should concentrate on the investigation of this area.

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