

## Vivianite (ferrous phosphate) alleviates iron chlorosis in grapevine

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### Summary

Synthetic vivianite [ $\text{Fe}_3(\text{PO}_4)_2 \cdot 8\text{H}_2\text{O}$ ] has been reported to alleviate iron (Fe) chlorosis in crops growing on calcareous soils. To test the effectiveness of vivianite in grapevine we carried out three-year (2003 to 2005) experiments in vineyards located in six different areas of Spain with Denomination of Origin (Rioja, Ribera del Duero, La Mancha, Montilla-Moriles, Condado de Huelva, and Jerez), which differed in grapevine rootstock/variety, climate, and soil properties. In all cases there was at least one treatment in which a suspension of vivianite was injected into the soil at the beginning of the experiment in spring, one control (“-Fe”, no Fe fertilizer added) treatment, and one or more treatments with Fe chelate (FeEDDHA) or an Fe(II) salt applied yearly. The concentration of chlorophyll per unit leaf area was estimated with a portable chlorophyll meter (readings in SPAD units). The SPAD value and the trunk perimeter increment of the vines fertilized with vivianite were significantly higher than those of the control (-Fe) vines through the three years in all fields except the Jerez one. Vivianite was not significantly more effective than Fe-sulfate (in Rioja field) or Fe chelate (in La Mancha field). Our results suggest in summary that vivianite is effective in improving the Fe nutrition of vine and has a significant long-lasting effect of at least three years. This is ascribed to vivianite being incongruently dissolved to produce a poorly crystalline Fe oxide phase (lepidocrocite), which is considered to be a good source of Fe to plant. Vivianite is effective, readily prepared in the field, not easily leached from the soil, cheap, and environmentally safe, constituting thus an adequate Fe fertilizer for grapevine.

**Key words:** calcareous soil, grapevine, iron chlorosis, iron phosphate, vivianite.

### Introduction

Iron (Fe) chlorosis is commonplace in grapevine cultivated in calcareous soils. The typical symptom of Fe chlorosis is a lack of chlorophyll (yellowing) in the interveinal tissue of the youngest leaves during the active growth period. Growth depression can also occur (BAVARESCO *et al.* 1993, GRUBER and KOSEGARTEN 2002), even before leaves become chlorotic. NIKOLIC and RÖMHELD (2002) also observed inhibited leaf expansion in grapes grown under se-

vere Fe deficiency. Eventually, leaves become entirely yellow, necrotic spots appear, yields are reduced, and plants die (REYES *et al.* 2006).

Many vineyards in the Mediterranean region are on calcareous soils and thus under risk of Fe chlorosis. Selecting tolerant rootstocks (BAVARESCO *et al.* 2005) is a common strategy to prevent Fe deficiency in grapevine. The most tolerant rootstocks have originated from American *Vitis* species, which are also resistant to phylloxera and fungal diseases. However, even the most tolerant rootstocks can develop symptoms of Fe chlorosis when grown in problematic soils. This frequently requires the use of Fe fertilizers; ideally, such fertilizers should have no adverse effects on the environment (e.g. groundwater contamination and eutrophication). Previous studies in Spain and Italy revealed that injecting a suspension of ferrous phosphate [an analogue of the mineral vivianite ( $\text{Fe}_3(\text{PO}_4)_2 \cdot 8\text{H}_2\text{O}$ )] into the soil reduced Fe chlorosis in chickpea (EYNARD *et al.* 1992), pear trees (DEL CAMPILLO *et al.* 1998), olive trees (ROSADO *et al.* 2002), and kiwifruit (ROMBOLÀ *et al.* 2003). Moreover, in contrast to the commonly used Fe chelates, the effect of vivianite lasted several years (DEL CAMPILLO *et al.* 1998, ROSADO *et al.* 2002). The purpose of this work was to assess the short- and long-term effectiveness of vivianite in correcting Fe chlorosis in grapevine. To this end, we conducted three-year field experiments.

### Material and Methods

**Study areas and plant materials:** The experimental fields were located in six major wine producing areas with a Denomination of Origin in Spain, namely: Rioja, Ribera del Duero, La Mancha, Montilla-Moriles, Condado de Huelva and Jerez. The situation and local climate of the fields in the 2002-2003, 2003-2004 and 2004-2005 growing seasons, and the characteristics of the plant material, are shown in Tab. 1. Only the Ribera del Duero field was drip-irrigated (~80 L/vine yearly). In all the experimental fields, plants had exhibited symptoms of Fe chlorosis in the previous season but not of any other nutrient deficiency.

Composite soil samples were collected from the 20-70 cm deep layer, where most of the active roots grew. Samples were air-dried, ground to < 2 mm and analyzed for particle size distribution (GEE and BAUDER 1986), organic carbon (OC, dichromate oxidation; WALKLEY and BLACK 1934), pH (1:2.5 soil:water suspension) and electrical conductivity (EC, 1:5 soil:water suspension). The total

Table 1  
Location, climatic data and plant material in the six experimental fields studied

Field	Latitude	Longitude	Altitude (m)	Precipitation† (mm)					Mean Temperature† (°C)					Age (yr)			
				Mean	2002–2003		2003–2004		2004–2005		2002–2003		2003–2004		2004–2005		
						2002	2003	2003	2004	2004	2005	2002	2003		2003	2004	2004
Rioja	42°34'N	2°32'W	614	400	512	469	351	12.8	12.5	12.3	12.3	12.3	12.3	12.3	12.3	4	
Ribera del Duero	41°34'N	4°07'W	765	435	596	425	223	13.4	12.3	12.2	12.2	12.2	12.2	12.2	12.2	12	
La Mancha	39°04'N	3°04'W	690	400	421	436	173	15.2	14.3	14.6	14.6	14.6	14.6	14.6	5		
Montilla–Moriles	37°35'N	4°40'W	348	500	579	630	267	18.2	17.6	17.4	17.4	17.4	17.4	17.4	6		
Condado de Huelva	37°33'N	6°53'W	110	490	574	799	269	18.1	17.7	17.6	17.6	17.6	17.6	17.6	4		
Jerez	36°43'N	6°13'W	64	600	724	756	312	18.2	17.7	17.3	17.3	17.3	17.3	17.3	7		

† 1 October to 30 September

CaCO<sub>3</sub> equivalent (CCE) was determined by weight loss upon treatment with 6 M HCl (VAN WESEMAEL 1955) and the active CaCO<sub>3</sub> equivalent (ACCE) or “active lime” with neutral NH<sub>4</sub> oxalate (DROUINEAU 1942). Available K was determined by extraction with 1 M NH<sub>4</sub>OAc buffered at pH 7 and available P by extraction with 0.5 M NaHCO<sub>3</sub> buffered at pH 8.5 (OLSEN *et al.* 1954). DTPA-extractable Fe (Fe<sub>DTPA</sub>) was determined according to LINDSAY and NOR-

VELL (1978) and acid NH<sub>4</sub> oxalate-extractable Fe (Fe<sub>ox</sub>) according to SCHWERTMANN (1964) except that the soil:solution ratio was 1:200 in order to prevent a significant pH rise in the extractant due to presence of carbonate. Citrate/ascorbate-extractable Fe (Fe<sub>ca</sub>) was determined according to REYES and TORRENT (1997).

**Experimental design and treatments:** A randomized block design with four blocks (six for the Rioja field) was adopted. The plots contained four plants each and were separated by one plant. The treatments were as follows: (a) Four treatments [control (-Fe), 250 and 375 g vivianite/vine, and 500 g FeSO<sub>4</sub>·7H<sub>2</sub>O/vine/yr] in the Rioja vineyard; (b) Three treatments [control (-Fe), 250 and 375 g of vivianite/vine] in the Ribera del Duero, Condado de Huelva and Jerez vineyards; (c) Four treatments [control (-Fe), 250 and 375 g vivianite/vine and 20 g FeEDDHA/vine/yr] in La Mancha vineyard; and (d) Three treatments [control (-Fe), 250 g vivianite/vine, and yearly application of a dissolution of 100 g FeSO<sub>4</sub>·7H<sub>2</sub>O + 10 g citric acid·l<sup>-1</sup> with a brush on the pruning cuts (Rességuier method) (REYNIER 2002) in the Montilla-Moriles vineyard]. It should be noted that the Rességuier method and the applications of iron sulfate and iron chelate (at the doses used in the these experiments) were those traditionally used by the vine growers in the corresponding regions to prevent Fe chlorosis.

Vivianite was applied in January 2003 in the Montilla-Moriles, Condado de Huelva, and Jerez fields, and in March 2003 in the other fields. The vineyards were fertilized with the amount of NPK fertilizer commonly used by the grapevine growers in each area. As per the leaf appearance, no deficiency symptoms other than the typical Fe chlorosis ones were observed during the experiment in any field; this suggested that, except for Fe, the local fertilizer application practices were essentially appropriate.

**Synthesis and application of vivianite:** Vivianite was prepared in a tank containing 100 L of continuously stirred water to which 2.5 kg of monoammonium phosphate [(NH<sub>4</sub>)H<sub>2</sub>PO<sub>4</sub>] was added until complete dissolution, followed by slow addition of 7.5 kg of ferrous sulfate (FeSO<sub>4</sub>·7H<sub>2</sub>O). The resulting thick suspension was initially white but turned rapidly into a greenish blue color typical of partly oxidized vivianite. Continuous stirring was needed to prevent the vivianite particles (2–10 µm in size) from settling on the bottom of the tank. The suspension, which contained about 50 g vivianite·l<sup>-1</sup>, was immediately injected into the soil by using a T-shaped injector connected to the tank. The volume required for the prescribed rate was applied in 5–10 points at a distance of 20–50 cm from the trunk and a depth of 30–45 cm.

**Plant analysis:** The chlorophyll concentration of young leaves was estimated from the “SPAD” readings acquired with a SPAD 502 portable chlorophyll meter (Minolta Camera Co., Osaka, Japan) at full bloom, veraison and harvest in the three growing seasons. The four youngest fully expanded leaves in each vine were collected for this purpose. Previous experiments had shown that the chlorophyll contents per unit surface area as determined by extraction with ethanol (WINTERMANS and DE MOTTS 1965) from the leaves of ‘Pedro Ximénez’ (Montilla-Moriles

vineyard), 'Zalema' (Condado de Huelva vineyard) and 'Palomino' (Jerez vineyard) were highly correlated with the SPAD readings ( $r = 0.88$ ,  $r = 0.93$ , and  $r = 0.94$ , respectively;  $P < 0.001$  in all cases).

Two leaf samples (including petiole) were selected from the main shoots of each vine at full bloom in the Montilla-Moriles field in 2003, and the Condado de Huelva and Jerez fields in 2003 and 2004. The petioles were washed with distilled water (0.01 % Tween 20) and then dried at 60 °C for 72 h before digestion with nitric/perchloric acid (ZAZOSKI and BURAU 1977). Calcium, Mg, Fe, Mn, Cu and Zn in solution were determined by atomic absorption spectrophotometry, K by flame emission, and P with the Molybdenum Blue color method of MURPHY and RILEY (1962). Mineral elements were determined as described above. Trunk perimeters were measured at the beginning and end of the experimental period (33-36 months), and yield was recorded at harvest.

**Statistical analyses:** The analysis of variance (ANOVA) was performed with Statistix 8.0 (ANALYTICAL SOFTWARE, Tallahassee, FL, USA). ANOVA for yield was done by using the initial trunk perimeter as covariable. In order to ensure normality, the trunk perimeter increment data were logarithmically transformed prior ANOVA for the Ribera del Duero and La Mancha fields. Unless otherwise stated, the word "significant" is used here to indicate significance at the  $P < 0.05$  level. Means were separated via the LSD test. For some measurements repeated at different times (Tab. 3) the mean plus the standard error for each treatment and time is shown.

## Results

**Soil properties:** The properties of the soils from the six experimental fields are shown in Tab. 2. All soils were strongly calcareous (CCE ranged from 423 to 705 g·kg<sup>-1</sup> soil and active lime from 91 to 286 g·kg<sup>-1</sup> soil). The pH in water ranged from 8.1 to 8.6, the organic carbon content was < 7.5 g·kg<sup>-1</sup> soil, and the clay content ranged from 156 to 365 g·kg<sup>-1</sup> soil. DTPA-extractable Fe ranged from 1.7 to 4.1 mg·kg<sup>-1</sup> soil [*i.e.*, it generally fell below the critical level of 4 mg·kg<sup>-1</sup> proposed by LINDSAY and NORVELL (1978)]. Fe<sub>ox</sub> and Fe<sub>ca</sub>, which are good predictors for the risk of Fe chlorosis in various plants (LOEPPERT and HALLMARK 1985, DEL CAMPILLO and TORRENT 1992; BENÍTEZ *et al.* 2002), ranged from 122 to 420 mg·kg<sup>-1</sup> soil and from 220 to 1071 g·kg<sup>-1</sup> soil, respectively. The Fe<sub>ox</sub>/ACCE ratio was lower than the critical level proposed by REYES *et al.* (2006) ( $25 \times 10^{-4}$ ), except for the Ribera del Duero field (Tab. 2); however, the grapevines in this field exhibited severe symptoms of Fe chlorosis (Figure, B).

**Effectiveness of vivianite:** The Figure shows the time course of the SPAD values in the 2003-2005 period. The control (-Fe) vines exhibited symptoms of Fe chlorosis (SPAD < 20) from the beginning of the experiment. SPAD generally increased from full bloom to harvest, partly because leaves become thicker with age and SPAD is obtained from reflectance rather than transmission measurements. The Ribera del Duero field exhibited the

Table 2

Selected properties† of the soils in the six experimental fields studied.

Field	Clay (g·kg <sup>-1</sup> )	OC (g·kg <sup>-1</sup> )	CCE (g·kg <sup>-1</sup> )	ACCE (g·kg <sup>-1</sup> )	pH (H <sub>2</sub> O)	EC (1:5) (dS·m <sup>-1</sup> )	Olsen P (mg·kg <sup>-1</sup> )	Available K (mg·kg <sup>-1</sup> )	Extractable Fe forms			
									Fe <sub>DTPA</sub> (mg·kg <sup>-1</sup> )	Fe <sub>ox</sub> (mg·kg <sup>-1</sup> )	Fe <sub>ca</sub> (mg·kg <sup>-1</sup> )	(Fe <sub>ox</sub> / ACCE)·10 <sup>4</sup>
Rioja	203	6.6	545	153	8.5	0.13	9	98	4.1	392	1071	26
Ribera del Duero	328	7.2	475	91	8.4	0.11	24	262	3.6	420	348	46
La Mancha	183	4.9	666	214	8.3	0.14	8	90	1.7	122	220	6
Montilla-Moriles	189	2.8	705	218	8.6	0.12	14	137	2.0	189	379	9
Condado de Huelva	156	4.1	423	161	8.1	0.10	9	90	2.2	125	265	8
Jerez	365	7.5	551	286	8.2	0.17	21	348	3.9	388	353	14

† OC: organic carbon; CCE: calcium carbonate equivalent; ACCE: active calcium carbonate equivalent (active lime); EC: electrical conductivity; Fe<sub>DTPA</sub>: DTPA-extractable Fe; Fe<sub>ox</sub>: oxalate-extractable Fe; Fe<sub>ca</sub>: citrate/ascorbate-extractable Fe

lowest SPAD values in the control (-Fe) vines and the Rioja the highest (Tab. 3), even though the variety/rootstock

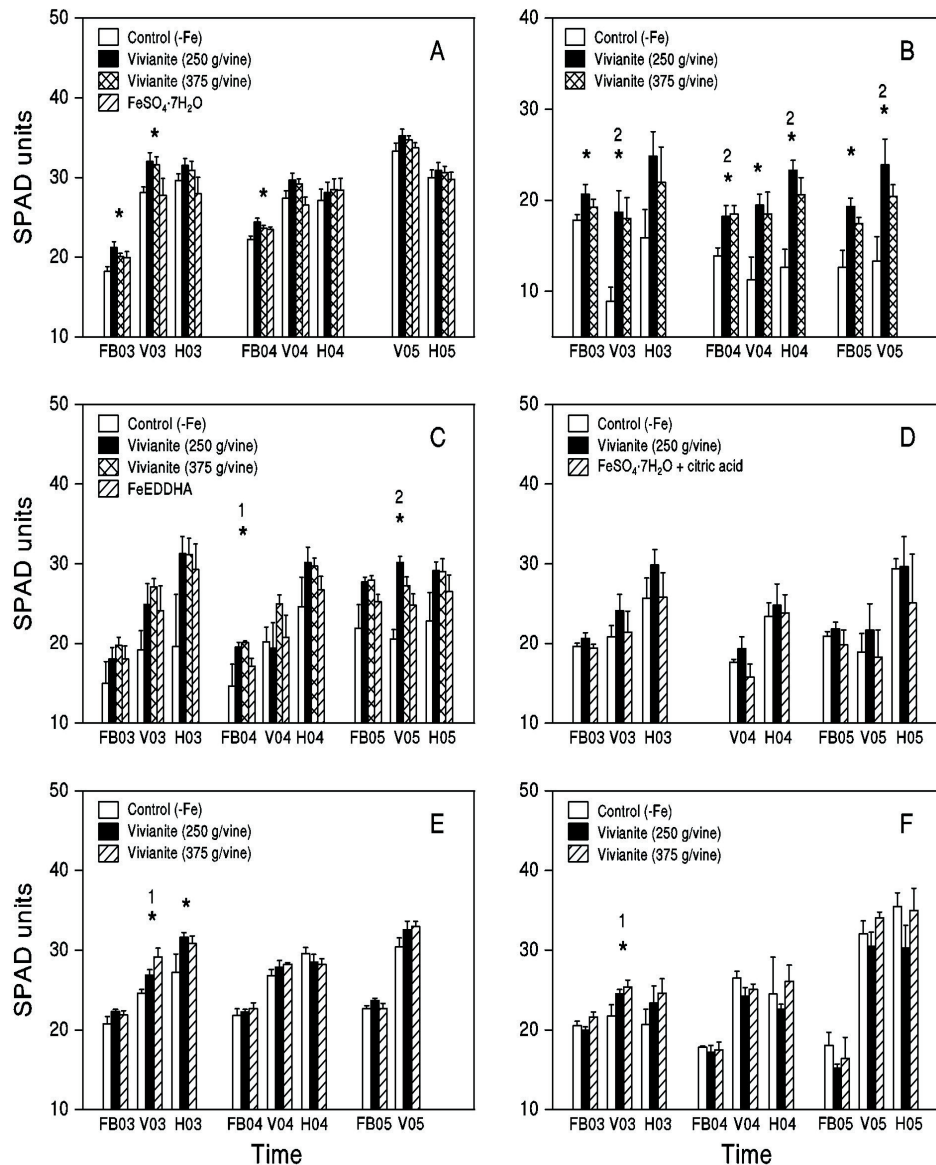


Figure: Time course of SPAD in vineyards established in the Rioja (A), Ribera del Duero (B), La Mancha (C), Montilla-Moriles (D), Condado de Huelva (E), and Jerez (F) in 2003, 2004 and 2005. FB: full bloom, V: veraison and H: harvest. Bars represent the standard error. An asterisk indicates those fields and times when the SPAD values of the control and vivianite-treated plants differed significantly ( $P < 0.05$ ). Significant differences for only the 375 g vivianite rate and for both the 250 and 375 g vivianite rates are indicated by numbers "1" and "2" above the asterisk, respectively.

was the same. Iron chlorosis was more severe in 2003 than it was in 2004 and 2005. This can be ascribed to the higher precipitation in October 2002 to September 2003 given that excess water in soil - which was never measured during the study - can aggravate Fe chlorosis (DAVENPORT and STEVENS 2006). The SPAD values for the vivianite-treated plants were higher than those of the control (-Fe) plants in 45 of the 50 measurements made in the Rioja, Ribera del Duero, La Mancha, Montilla-Moriles, Condado de Huelva and Jerez vineyards (Figure); however, differences were significant only in 15 measurements. The 3-year mean SPAD values at full bloom, veraison and harvest for the vines fertilized with vivianite at a rate of 250 g·plant<sup>-1</sup> were higher than those for the control (-Fe) vines in all but the Jerez field (Tab. 3). On the other hand, SPAD at the high vivianite application rate (375 g·plant<sup>-1</sup>) was not sig-

nificantly different from that obtained at the low rate (250 g·plant<sup>-1</sup>) in any field. If one takes into account the volume of soil explored by the active roots, 250 g vivianite/plant is a rate similar to that of 1 g vivianite·kg<sup>-1</sup> soil that proved to be effective to prevent iron chlorosis in previous experiments (EYNARD *et al.* 1992).

The yield of Fe-fertilized vines was not significantly different from that of control (-Fe) vines. Also, the yield of vines treated with Fe sulfate (Rioja field) or Fe-EDDHA (La Mancha field) was not significantly different from that of vivianite-fertilized vines (Tab. 3).

The trunk perimeter increment (TPI) in vivianite-treated plants was generally higher than that in control (-Fe) vines. TPI for the vivianite-fertilized vines in La Mancha, Jerez and Ribera del Duero fields (250 g·plant<sup>-1</sup>) was significantly different from that for the control (-Fe) vines.



Table 3

Effect of Fe fertilizers on SPAD at full bloom, veraison and harvest, and on the total yield and trunk perimeter increment in the six vineyards (mean  $\pm$  standard error)

Treatments	Field					
	Rioja	Ribera del Duero	La Mancha	Montilla-Moriles	Condado de Huelva	Jerez
	Mean <sup>†</sup> SPAD at veraison					
Control (-Fe)	20 $\pm$ 1	15 $\pm$ 1	17 $\pm$ 2	20 $\pm$ 0	22 $\pm$ 1	19 $\pm$ 1
Vivianite (250 g/vine)	23 $\pm$ 1	20 $\pm$ 1	22 $\pm$ 1	21 $\pm$ 1	23 $\pm$ 0	17 $\pm$ 1
Vivianite (375 g/vine)	22 $\pm$ 1	19 $\pm$ 1	23 $\pm$ 1		22 $\pm$ 0	19 $\pm$ 1
Other Fe Fertilizer <sup>‡</sup>	22 $\pm$ 1		20 $\pm$ 1	19 $\pm$ 1		
	Mean <sup>†</sup> SPAD at harvest					
Control (-Fe)	30 $\pm$ 1	11 $\pm$ 1	20 $\pm$ 1	19 $\pm$ 1	27 $\pm$ 1	27 $\pm$ 2
Vivianite (250 g/vine)	32 $\pm$ 1	21 $\pm$ 1	25 $\pm$ 2	22 $\pm$ 1	29 $\pm$ 1	26 $\pm$ 1
Vivianite (375 g/vine)	32 $\pm$ 1	19 $\pm$ 1	26 $\pm$ 1		30 $\pm$ 1	28 $\pm$ 1
Other Fe Fertilizer <sup>‡</sup>	30 $\pm$ 1		23 $\pm$ 1	18 $\pm$ 2		
	Mean <sup>†</sup> SPAD at harvest					
Control (-Fe)	29 $\pm$ 1	14 $\pm$ 1	22 $\pm$ 3	26 $\pm$ 1	28 $\pm$ 1	27 $\pm$ 3
Vivianite (250 g/vine)	30 $\pm$ 1	24 $\pm$ 1	30 $\pm$ 1	28 $\pm$ 2	30 $\pm$ 1	25 $\pm$ 2
Vivianite (375 g/vine)	30 $\pm$ 1	21 $\pm$ 2	30 $\pm$ 1		30 $\pm$ 1	29 $\pm$ 2
Other Fe Fertilizer <sup>‡</sup>	29 $\pm$ 1		27 $\pm$ 1	25 $\pm$ 2		
	Total Yield <sup>§¶</sup> (kg/vine)					
Control (-Fe)	10.8 ab	4.5 a	7.9 a	16.4 a	2.1 a	17.0 a
Vivianite (250 g/vine)	11.9 b	4.7 a	8.1 a	15.8 a	1.9 a	13.7 a
Vivianite (375 g/vine)	9.6 a	5.0 a	10.4 a		0.9 a	13.2 a
Other Fe Fertilizer <sup>‡</sup>	10.8 ab		13.0 a	13.6 a		
	Trunk Perimeter Increment <sup>§</sup> (%)					
Control (-Fe)	32 a	15 a	22 a	32 a	32 a	12 a
Vivianite (250 g/vine)	39 a	29 b	43 b	30 a	31 a	19 b
Vivianite (375 g/vine)	39 a	28 ab	28 ab		40 a	15 ab
Other Fe Fertilizer <sup>‡</sup>	42 a		24 a	30 a		

<sup>†</sup> Mean of the 2003, 2004 and 2005 data.

<sup>‡</sup> Other Fe fertilizers: 500 g FeSO<sub>4</sub>·7H<sub>2</sub>O/plant/year in Rioja; 20 g FeEDDHA/plant/year in La Mancha; and Rességuier method in Montilla-Moriles.

<sup>§</sup> Values followed by different letters were significantly different at  $P < 0.05$  in the LSD test.

<sup>¶</sup> 2003 and 2004 data in Condado de Huelva.

However, the TPI values for vines fertilized yearly with FeSO<sub>4</sub>·7H<sub>2</sub>O (Rioja field) or FeEDDHA (La Mancha field) were not significantly different from those for the control (-Fe) vines (Tab. 3). Finally, yield was not significantly affected by the Fe treatments.

Because the concentrations of the different elements in the petioles were not significantly affected by the Fe treatment, only the means for the experimental fields and nutrients studied are shown (Tab. 4). Such concentrations were all higher than the corresponding critical levels established by JONES *et al.* (1991).

## Discussion

The effect of vivianite on SPAD and TPI was observed across a variety of plant materials, climatic conditions and soil properties. It should be stressed that a single application of vivianite was effective in reducing Fe chlorosis for three years. Such a long-term effectiveness of vivianite was observed in previous field experiments with pear (five

years; DEL CAMPILLO *et al.* 1998) and olive trees (three years; ROSADO *et al.* 2002). The effectiveness of vivianite in correcting Fe chlorosis can be ascribed to its high content in Fe (~30%) and its slow, incongruent dissolution in calcareous media to give poorly crystalline lepidocrocite (ROLDÁN *et al.* 2002). Phosphate suppresses the formation of more crystalline Fe oxides, probably because their precursor phases [e.g. green rust in Fe (II) systems and ferrihydrite in Fe (III) systems] readily absorb phosphate ion. Thus, the oxidation and hydrolysis of Fe (II) sulfate resulted in the formation of poorly crystalline lepidocrocite in the presence of phosphate, but crystalline goethite and little lepidocrocite in its absence (CUMPLIDO *et al.* 2000).

The effectiveness in correcting Fe chlorosis of the poorly crystalline lepidocrocite resulting from the alteration of vivianite in calcareous media is consistent with the current contention that the main sources of available Fe to plants in calcareous soils are poorly crystalline Fe oxides. This contention is supported, among others, by the studies of LOEPPERT and HALLMARK (1985) in sorghum, DEL CAMPILLO and TORRENT (1992) in chickpea and sunflower, YAN-

Table 4

Mineral element concentrations in petioles at full bloom in the Montilla-Moriles, Condado de Huelva and Jerez experimental fields (mean  $\pm$  standar error)

Field	Petioles (g·kg <sup>-1</sup> )				Petioles (mg·kg <sup>-1</sup> )			
	P	K	Ca	Mg	Fe	Mn	Cu	Zn
Montilla-Moriles†	1.9 $\pm$ 0.4	12 $\pm$ 1	17 $\pm$ 1	5.4 $\pm$ 0.2	26 $\pm$ 1	26 $\pm$ 3	54 $\pm$ 3	30 $\pm$ 3
Condado de Huelva‡	5.2 $\pm$ 0.3	19 $\pm$ 1	31 $\pm$ 1	3.9 $\pm$ 0.0	28 $\pm$ 1	16 $\pm$ 1	28 $\pm$ 4	28 $\pm$ 1
Jerez‡	9.2 $\pm$ 0.3	19 $\pm$ 2	34 $\pm$ 2	6.3 $\pm$ 0.3	47 $\pm$ 6	47 $\pm$ 4	36 $\pm$ 4	58 $\pm$ 6

† Data from 2003.

‡ Mean of 2003 and 2004 data.

GUAS *et al.* (1997) in peach, and DE SANTIAGO and DELGADO (2006) in white lupine.

Based on the SPAD values and yield, a single application of vivianite was as effective as yearly applications of FeEDDHA in preventing Fe chlorosis in grapevine. One obvious reason for the long-term effectiveness of vivianite is that its particles, which are 2 to 10  $\mu$ m in size, cannot be easily leached from soil. By contrast, Fe chelates are easily leached, so they are less effective than vivianite in rainy years or in irrigated systems (ROMBOLÀ *et al.* 2003); for this reason they are usually applied once or twice yearly.

Because of its low mobility in soil, vivianite should be applied in a soil volume containing a significant fraction of the plant active roots. Thus, the best results are obtained when plant roots are confined to a small volume - into which the vivianite suspension is to be injected. This accounts for the high effectiveness of vivianite in the Ribera del Duero and La Mancha fields (Tab. 3), where the rooting depth was limited by drip irrigation in the former and a compact petrocalcic horizon at 50-60 cm in the latter. The other experimental fields were rainfed, so roots grew to a greater depth in response to the seasonal drought spells typical of the Mediterranean climate. Grapevine yield was not significantly affected by fertilization with Fe. Such a lack of response was probably due to plants having relatively large mass and nutrient reserves by virtue of their age (4 to 12 years). Similar results were previously obtained by ROMBOLÀ *et al.* (2003) for kiwifruit.

In summary, vivianite seems to be an interesting alternative to other Fe fertilizers used to prevent Fe chlorosis in grapevine judging by its effectiveness and long-term fertilizing effect. Moreover, it is not easily leached from the soil, inexpensive, easy to prepare, and environmentally safe (RODADO *et al.* 2002).

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