

Leaf mineral status and bud fertility of 'Centennial Seedless' grapevines

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KEY WORDS: *Vitis vinifera*, flowering, bud necrosis, nitrogen, potassium, boron, iron.

ABSTRACT

Twelve vineyards of cv 'Centennial Seedless' (*V. vinifera* L.) were chosen for leaf (lamina + petiole), lamina and petiole mineral analysis, over a two year period, in Jales, São Paulo State, Brazil. Twenty vine canes from each vineyard were sampled for bud fertility evaluation using a stereomicroscope. The relationship between bud fertility or bud necrosis and mineral contents were investigated. The correlations determined were weak when other factors were involved in bud fertility, such as temperature, light, shoot vigor and water deficiency. Nitrogen and potassium contents showed an inverse relationship with the percentage of bud necrosis. Iron and boron contents presented a quadratic relationship between the percentage of fertile buds and the percentage of bud necrosis. The possible involvement of these elements in root growth and, therefore, in cytokinin biosynthesis, could explain their influence on floral stimulus. (*Bulletin O.I.V.*, 2005, vol. 78, nº 897-898, pp. 737-749).

ZUSAMMENFASUNG

Mineralstatus der Blätter und Fruchtbarkeit der Knospen bei der kernlosen Rebsorte Centennial Seedless

Zwölf Weinberge, an denen die Rebsorte 'Centennial Seedless' (*V. vinifera* L.) angebaut wird, wurden für eine über 2 Jahre angelegte Mineralanalyse der Blätter (Blattspreite + Blattstiel) in Jales, São Paulo State, Brasilien, ausgewählt. An zwanzig Weinstöcken jedes Weinbergs wurden Proben zur Abschätzung der Fruchtbarkeit der Knospen mit Hilfe eines Stereomikroskops entnommen. Die Beziehung zwischen Fruchtbarkeit der Knospen oder Knospennekrose und Mineralgehalt wurde untersucht. Es wurden geringe Wechselbeziehungen festgestellt, sobald die Fruchtbarkeit der Knospen durch weitere Faktoren wie Temperatur, Licht, Triebkraft und Wassermangel beeinflusst wird. Der Stickstoff- und Kaliumgehalt stand in umgekehrtem Verhältnis zum prozentualen Anteil der Knospennekrose. Der Eisen- und Borgehalt stand in einem quadratischen Verhältnis zur Fruchtbarkeit der Knospen und dem prozentualen Anteil der Knospennekrose. Die mögliche Beteiligung dieser Elemente am Wurzelwachstum und infolgedessen an der Cytokinin-Biosynthese, könnte ihren Einfluss auf den Blühstimulus erklären. (*Bulletin O.I.V.*, 2005, vol. 78, nº 897-898, pp. 737-749).

RESUMEN

Estado mineral de la hoja y fertilidad de la yema de la variedad de vid Centennial Seedless

Se seleccionaron doce viñedos de cv. 'Centennial Seedless' (*V. vinifera* L.) para realizar un análisis mineral de las hojas (tejido foliar + pecíolo), del tejido foliar y del pecíolo, durante un período de dos años, en Jales, Estado de Sao Paulo, Brasil. Se tomaron veinte muestras de sarmiento de cada viñedo para evaluar la fertilidad de la yema mediante un estereomicroscopio. Se analizó la relación entre la fertilidad de la yema o necrosis de la yema y el contenido mineral. Las correlaciones determinadas fueron bajas al introducir otros factores en la fertilidad de la yema (temperatura, luz, vigor de los brotes y carencia de agua). El contenido en nitrógeno y potasio mostró una relación inversa con el porcentaje de necrosis de la yema. El contenido en hierro y boro presentó una relación cuadrática entre el porcentaje de yemas fértiles y el porcentaje de necrosis de la yema. La posible implicación de estos elementos en el desarrollo de la raíz y, por tanto, en la biosíntesis de la citoquinina, podría explicar su influencia en el estímulo floral. (*Bulletin O.I.V.*, 2005, vol. 78, n° 897-898, pp. 737-749).

RÉSUMÉ

État de la composition minérale des feuilles et la fertilité des bourgeons de la vigne Centennial Seedless

Ce travail a été réalisé sur douze vignobles du cépage 'Centennial Seedless' (*Vitis vinifera* L.) en analysant la composition minérale des feuilles (limbe + pétiole), limbe et pétiole pendant deux années, dans la commune de Jales, État de São Paulo, Brésil. Vingt sarments de chaque vignoble ont été pris et la fertilité des bourgeons a été évaluée par stéréomicroscopie. On a étudié le rapport entre la fertilité des bourgeons et la nécrose des bourgeons avec la nutrition minérale. Les corrélations établies ont été faibles, parce que d'autres facteurs ont des influences sur la fertilité des bourgeons, comme la température, la lumière, la vigueur des rameaux et le déficit hydrique. Les concentrations en azote et en potassium ont montré une relation inverse avec le pourcentage de bourgeons nécrosés. Les teneurs en fer et en bore ont présenté une relation de deuxième degré par rapport au pourcentage de bourgeons fertiles et au pourcentage des bourgeons nécrosés. La possibilité de ces éléments d'avoir influencé la croissance des racines et, partant, la biosynthèse de cytokinines pourrait expliquer son influence dans le stimulus floral. (*Bulletin O.I.V.*, 2005, vol. 78, n° 897-898, pp. 737-749).

RASSIUNTO

Status minerale del pampino e fertilità della gemmanelle viti "centennali senza semi"

Dodici vigneti del cv "centennale senza semi" (*V. vinifera* L.) sono state scelte per analisi minerale del pampino (lamina + petiole), del lamina e del petiole, per un periodo di due anni, in Jales, Stato di São Paulo, Brasile. Sono stati campionati venti pali da ogni vigna per valutare mediante stereomicroscopio la fertilità della gemma. È stato analizzato il rapporto fra la fertilità o la necrosi della gemma e il contenuto di minerale. Sono state riscontrate deboli correlazioni quando altri fattori sono intervenuti nella fertilità della gemma, quali la temperatura, la luce, il vigore del germoglio e la mancanza d'acqua. Il contenuto di potassio e d'azoto ha mostrato una relazione inversamente proporzionale alla percentuale di necrosi della gemma. Il contenuto di boro e ferro ha presentato un rapporto quadratico fra la percentuale delle gemme fertili e la percentuale di necrosi della gemma. La possibile partecipazione di tali elementi nello sviluppo della radice e, pertanto, nella biosintesi della citochinina, potrebbe spiegare la loro influenza sullo stimolo floreale. (*Bulletin O.I.V.*, 2005, vol. 78, n° 897-898, pp. 737-749).

INTRODUCTION

Grapevines (*Vitis spp.*) present a compound auxillary bud that includes lateral meristems, also denominated latent (Morrison, 1991). Latent buds containing inflorescence primordia are called fertile buds (Srinivasan and Mullins, 1981) and any unbalance in the factors involved in the differentiation of the inflorescence primordia can lead the uncommitted primordia to produce tendrils or shoots (Shikhamany, 1999; Mullins *et al.*, 2000).

According to Boss *et al.* (2003), after differentiation the immature inflorescences survive during winter in a latent stage in the dormant bud, and subsequently in bud sprouting. The following spring the development process continues until flower formation.

Several external factors may influence bud fertility, such as temperature, light, nutrient availability and water deficiency (Shikhamany, 1999; Dry, 2000; Mullins *et al.*, 2000). According to Thimann (1974), flowering is a process under hormonal control and these external factors can influence the hormonal balance through modification of physiology of the plant.

Sucrose, cytokinins and mineral nutrients are considered important components of floral stimulus, and the presence of these substances in optimum concentrations are necessary for the specific gene activity in the vegetative meristem (Vaz *et al.*, 2004).

Optimum nitrogen, phosphorus and potassium leaf contents have been correlated to maximum cytokinin production by grapevine roots, necessary for floral induction (Jako, 1976). Nevertheless, excessive nitrogen can reduce the formation of fertile buds (Keller and Koblet, 1995). Although it is possible that these mineral nutrients have a direct effect on cytokinin biosynthesis, it is much more likely they act directly via root growth and the induction of root primordia, the major site of cytokinin biosynthesis (Marschner, 1995).

There is a recommended optimal mineral content range in leaf, lamina and petiole for table grapes in São Paulo State (Brazil), although such levels are related to bloom or veraison stage after production pruning (Terra *et al.*, 2003). As in many tropical regions the formation pruning is carried out aiming to harvest in the winter, such recommendation is not valid, when no clusters remain in this first vegetative cycle.

The "double pruning-single cropping" system has currently been adopted in these regions. After harvest in winter, grapevines are forced to undergo rest for about a month, during which period water is withheld to help concentrate the reserves in the mature parts. All the fruiting canes are pruned back to spurs retaining only one basal node. This is called 'formation pruning'. Buds from these spurs grow and differentiate into floral primordial and the shoots mature in about five months. These mature shoots are pruned for fruiting before the onset of winter (March-April). This pruning is called 'production pruning'. Thus, in this system, a cycle of two pruning results in one crop.

Many vineyards have showed low bud fertility in the State of São Paulo, Brazil, although the reasons for this fact remain unclear (Motoyke, 1994). However, an inadequate use of fertilizers could be the reason for this problem.

The objective of this study is thus to verify the influence of mineral nutrition on bud fertility and bud necrosis of 'Centennial Seedless' grapevines.

MATERIAL AND METHODS

Leaf samples were collected for mineral analysis during two seasons (2002 and 2003), between October and November, from twelve 'Centennial Seedless' (*Vitis vinifera* L.) vineyards, located in the commune of Jales, State of São Paulo, Brazil (20°16'S and 50°24'W, 418 m a.s.l.). The vines were grafted onto IAC-572 'Jales' rootstock [*V. caribaea* x (*V. riparia* x *V. rupestris* 101-14)] and trellised by pergola system. Some characteristics of the vineyards sampled are showed in *table I*.

In Northwestern of São Paulo State, a region with mild winters, the vines are pruned very short (1 or 2 buds/cane) after harvest, between September and October, and no clusters are left. This practice is called formation pruning, just aiming the development of new canes for production pruning (10 to 15 buds/cane) in March, which allows harvests between June and August, during the winter in the Southern Hemisphere.

In each vineyard, two samples of one hundred leaves were taken for chemical analysis; one of them for leaf (lamina + petiole) and another, for a separate lamina and petiole analyses. A sample was taken of the youngest mature leaf (opposed to the cluster) of each shoot during bloom between November and December (Terra, 2003).

Sample preparation and chemical analyses methodology was described by Bataglia *et al.* (1983). The contents of the macro nutrients nitrogen, phosphorus, potassium, calcium and magnesium were expressed as percentage of dry weight, and the contents of micronutrients boron, copper, iron, manganese and zinc were expressed as ppm of dry weight. The results are presented in *tables II, III* and *IV*.

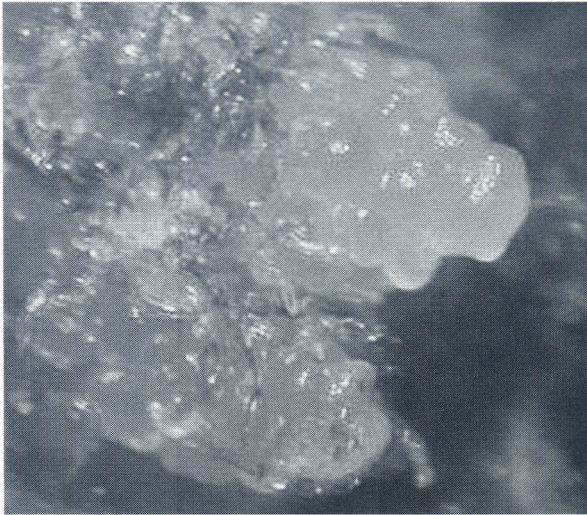
In March, at the end of the vegetative season before production pruning, twenty canes were randomly sampled from each vineyard. The fifteen basal buds from every cane were dissected and examined under a stereomicroscope with 45-fold zoom, to verify the presence of inflorescence primordia (*Figure 1*) or the occurrence of bud necrosis, according to the description of Mullins *et al.* (2000). Relationships between percentage of bud fertility, bud necrosis and mineral contents were determined by regression analysis using a statistical program SPSS 10.1 for Windows®.

RESULTS AND DISCUSSION

Data presented in *tables I, II* and *III* was verified to ascertain that mineral contents are too high in most of the cases, and could be considered excessive for the majority of vineyards, in accordance with recommendations for table grape vines in São Paulo State (Terra, 2003). The lamina-N content, for example, should be considered excessive (up to 3.4% of dry weight) for 62.5% of vineyards. The same could be said for petiole-K, where 100% of the areas showed excessive levels (up to 2.8%).

However, it is important to note that no clusters remain at this time in the Northwest of São Paulo State, and therefore no exportation from leaves is

FIGURE 1
Inflorescence primordia of 'Centennial Seedless' grapevines



expected, and higher nutrient levels are forecasted. The formation pruning did not allow cluster development in this first vegetative cycle because they had spurs instead of canes.

Some other elements are too high for other reasons. Manganese and copper are usually very elevated due to the fungicides applied in the vineyards.

There was no significant relationship between bud fertility or bud necrosis and the contents of phosphorus, calcium, magnesium, copper, zinc and manganese. However, the increase in lamina-N and petiole-K contents linearly reduced the percentage of bud necrosis (*Figure 2*). Nitrogen is an important element for carbohydrate metabolism, and low levels of this element can reduce the number of fertile buds in grapevines (Bowen and Kliewer, 1990; Mullins *et al.*, 2000). Probably, this happened because nitrogen has the most prominent influence on the production and export of cytokinins to the shoots and on root growth, where many phytohormones are synthesized (Marschner, 1995).

Cytokinins have been considered important components in floral induction. External stimulus on leaves by light, promotes the production and exportation of cytokinins to the stem meristem which have an important role in flower differentiation (Vaz *et al.*, 2004). Thereafter, cytokinins promote the establishment of sinks, acting directly at least in two essential proteins for nutrient translocation (invertase and hexose-carrier), which is necessary for floral stimulus (Peres and Kerbauy, 2004). Therefore, low nitrogen levels can also be harmful for bud formation because it can cause bud necrosis in grapevines.

FIGURE 3

Relationship between the percentage of fertile buds as a function of petiole-B content ($\bullet y = -72.27 + 4.71x - 0.046x^2, r^2 = 0.3460^*$) and lamina-B content ($\circ y = -85.75 + 3.52x - 0.023x^2, r^2 = 0.4300^{**}$) of 'Centennial Seedless' grapevines

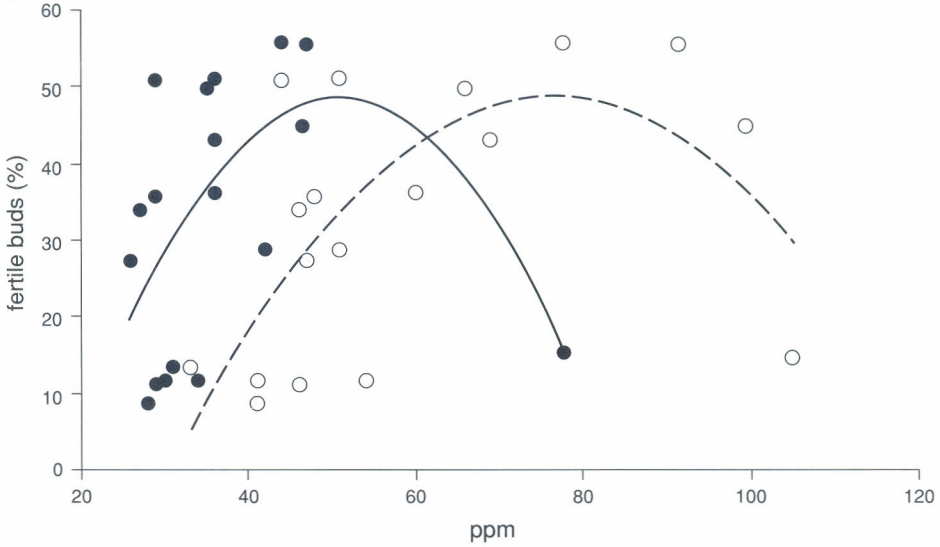
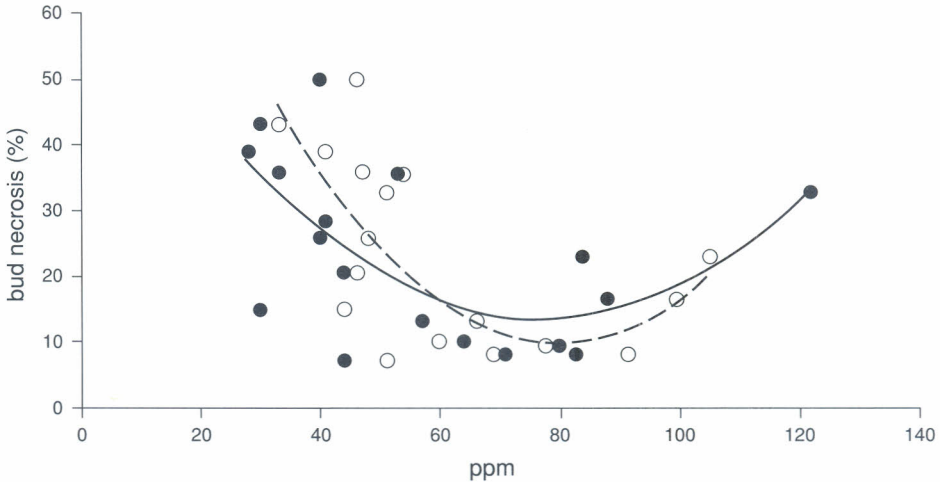


FIGURE 4

Relationship between the percentage of bud necrosis as a function of leaf-B content ($\bullet y = 72.29 - 1.52x + 0.0098x^2, r^2 = 0.4163^{**}$) and lamina-B content ($\circ y = 113.88 - 2.61x + 0.016x^2, r^2 = 0.4982^{**}$) of 'Centennial Seedless' grapevines

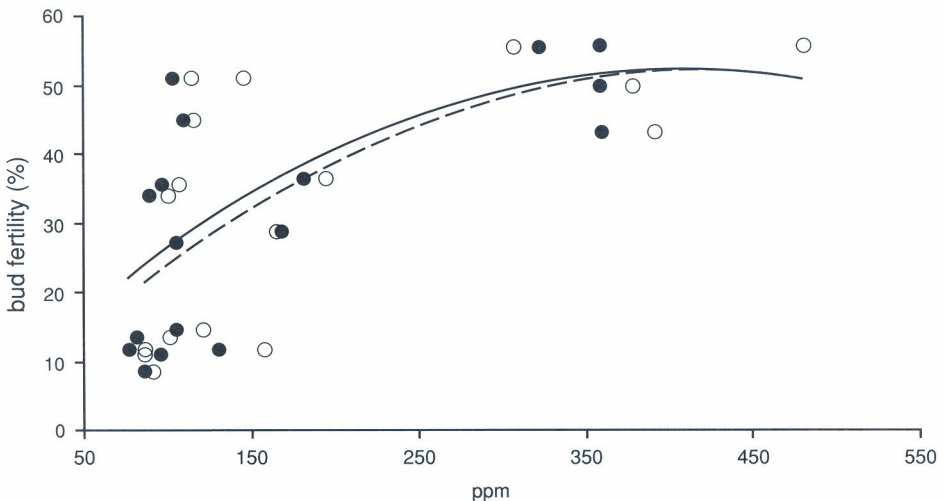


in nitrate reduction, positive correlations between iron supply, ferredoxin and nitrate reduction are to be expected (Marschner, 1985). As this element is associated with nitrogen nutrition, it might be also involved in flower induction. In Brazil, iron deficient soils are uncommon, but sometimes, elevated soil pH and water deficit can reduce plant uptake (Terra, 2003).

Excessive shoot vigor can increase bud necrosis and reduce bud fertility by mutual shading (Chadha and Shikhamany, 1999). **Therefore, it was studied the** relationship between fresh cane weight and mineral contents or bud fertility, but no significance was observed (data not shown).

It is also important to mention that application of gibberellin might be involved in bud necrosis in vines (Lavee *et al.*, 1993). All these facts corroborate the hypothesis of a direct effect of nutrients on hormonal balance, by and large cytokinins synthesis.

FIGURE 5
Relationship between the percentage of fertile buds as a function of leaf-Fe content ($\circ y = 5.15 + 0.24x - 0.0003x^2, r^2 = 0.3940^*$) and lamina-Fe content ($\bullet y = 2.72 + 0.24x - 0.0003x^2, r^2 = 0.4261^{**}$) of 'Centennial Seedless' grapevines.



CONCLUSIONS

The role of mineral nutrition in bud fertility of 'Centennial Seedless' grapevines in Brazilian vineyards was demonstrated. Nevertheless, the correlations verified were weak, once other factors are involved in bud fertility, such as temperature, light, shoot vigor and water deficiency. Vineyards deficient in nitrogen, potassium, boron and iron showed high incidence of bud necrosis and/or low bud fertility. Excessive boron levels were also detrimental to fertile bud formation. The possible involvement

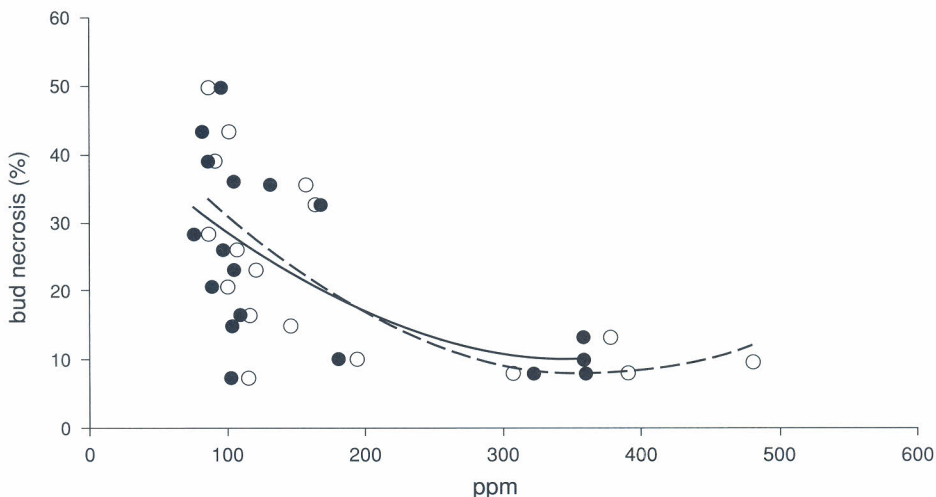
of these elements on root growth and, therefore, in cytokinin biosynthesis, could explain their influences in floral stimulus. Lamina analysis was the variable that more often showed a relationship to bud fertility.

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FIGURE 6

Relationship between the percentage of bud necrosis as a function of leaf-Fe content ($\circ y = 45.95 - 0.23x + 0.0005x^2$, $r^2 = 0.3693^*$) and lamina-Fe content ($\bullet y = 51.05 + 0.24x - 0.0003x^2$, $r^2 = 0.4345^{**}$) of 'Centennial Seedless' grapevines



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TABLE I
Characteristics of the Centennial Seedless vineyards
sampled in São Paulo State, Brazil

Vineyard	Vine Spacing	Age (years)
1	4.0 x 3.0 m	5
2	3.0 x 2.5 m	4
3	3.0 x 2.3 m	3
4	5.0 x 3.0 m	4
5	5.0 x 2.5 m	2
6	5.0 x 3.0 m	5
7	5.0 x 3.0 m	6
8	5.0 x 1.0 m	4
9	3.0 x 2.5 m	3
10	3.0 x 2.5 m	3
11	5.0 x 3.0 m	7
12	3.0 x 2.5 m	2

TABLE II

Mineral leaf content of macronutrients (% of dry weight) and micronutrients (ppm of dry weight) of 'Centennial Seedless' grapevines

Vine-yards	N	P	K	Ca	Mg	B	Cu	Fe	Mn	Zn
	Dry weight (%)					Dry weight (ppm)				
First Season (2002)										
1	2.9	0.36	1.8	1.5	0.41	44	10	103	260	25
2	2.8	0.29	1.7	1.7	0.40	40	25	96	529	30
3	3.9	0.47	2.1	1.2	0.23	41	299	77	80	26
4	3.3	0.40	2.4	1.3	0.37	53	16	131	655	104
5	3.9	0.27	1.8	2.3	0.40	70	278	360	765	60
6	4.1	0.26	1.3	1.5	0.26	82	515	322	196	22
7	3.8	0.39	1.7	3.4	0.56	87	18	110	361	35
8	3.3	0.39	1.7	1.6	0.40	64	20	181	759	54
9	3.2	0.42	2.5	1.5	0.46	57	285	359	383	50
10	3.8	0.41	1.7	2.0	0.41	44	629	89	294	29
11	4.2	0.43	1.6	2.8	0.42	79	42	359	153	95
12	3.3	0.37	1.7	1.6	0.27	28	396	87	233	32
Second Season (2003)										
1	2.5	0.26	2.0	2.1	0.49	30	48	104	308	32
2	3.5	0.40	1.1	2.0	0.28	33	15	105	443	73
3	2.7	0.30	1.7	2.3	0.48	122	13	168	583	59
4	3.4	0.45	1.6	1.8	0.33	40	15	97	293	45
5	3.6	0.45	2.6	2.4	0.48	84	104	105	578	43
6	3.3	0.43	2.1	1.4	0.31	30	136	82	365	80
7	3.1	0.32	1.4	1.9	0.33	29	288	121	162	41
8	3.8	0.41	2.4	1.4	0.40	36	11	96	858	118
9	3.3	0.36	2.1	1.4	0.39	90	10	115	240	56
10	3.5	0.43	2.3	1.6	0.45	65	355	170	184	32
11	2.8	0.34	1.4	1.8	0.38	53	7	176	264	43
12	4.7	0.31	1.6	3.9	0.44	70	27	488	312	38

TABLE III
Mineral lamina content of macronutrients (% of dry weight) and micronutrients (ppm) of 'Centennial Seedless' grapevines

Vine-yards	N	P	K	Ca	Mg	B	Cu	Fe	Mn	Zn
	Dry weight (%)					Dry weight (ppm)				
First Season (2002)										
1	3.2	0.34	1.5	1.6	0.41	51	10	115	306	22
2	3.2	0.29	1.6	1.8	0.42	46	29	87	403	31
3	3.8	0.47	1.5	1.3	0.22	41	219	87	90	33
4	3.4	0.40	1.7	1.3	0.34	54	19	158	765	123
5	4.3	0.27	1.4	2.2	0.35	68	277	391	725	56
6	3.9	0.26	1.3	1.3	0.34	91	450	307	213	23
7	4.2	0.39	1.2	3.3	0.53	99	19	116	429	35
8	3.7	0.39	1.4	1.6	0.40	60	22	194	806	54
9	3.7	0.42	2.1	1.5	0.48	66	267	378	418	52
10	3.7	0.41	1.2	2.1	0.39	46	594	101	397	36
11	4.7	0.43	1.5	3.6	0.47	77	53	481	180	110
12	3.1	0.37	1.3	2.7	0.39	41	545	91	385	37
Second Season (2003)										
1	3.2	0.26	1.4	1.8	0.38	44	65	146	270	32
2	3.5	0.40	1.1	2.4	0.37	47	14	105	526	104
3	3.1	0.30	1.2	2.0	0.40	51	8	165	480	48
4	3.1	0.45	1.4	1.7	0.48	48	18	107	309	38
5	3.8	0.45	2.2	2.5	0.45	105	176	121	782	53
6	3.3	0.43	1.5	2.3	0.38	33	208	102	532	46
7	3.4	0.32	1.3	1.7	0.29	28	224	114	149	46
8	4.1	0.41	1.8	1.4	0.40	39	13	137	962	133
9	3.6	0.36	1.4	1.4	0.38	57	12	128	242	59
10	3.6	0.41	1.8	1.8	0.51	76	342	172	209	29
11	3.5	0.36	1.4	1.9	0.40	55	9	184	265	42
12	4.7	0.35	1.5	3.6	0.49	77	22	445	325	39

TABLE IV

Mineral petiole content of macronutrients (% of dry weight) and micronutrients (ppm) of 'Centennial Seedless' grapevines

Vine- yards	N	P	K	Ca	Mg	B	Cu	Fe	Mn	Zn
	Dry weight (%)					Dry weight (ppm)				
First Season (2002)										
1	1.4	0.50	5.3	1.2	0.51	36	10	30	184	23
2	1.3	0.32	5.2	1.2	0.47	29	14	31	222	25
3	1.7	0.60	4.8	0.9	0.23	30	36	44	31	31
4	1.8	0.49	5.2	0.9	0.40	34	14	57	174	57
5	1.2	0.41	7.4	1.1	0.43	36	40	56	144	28
6	1.9	0.37	5.3	1.0	0.37	47	75	58	223	25
7	2.5	0.50	4.6	2.2	0.62	46	6	24	110	19
8	1.4	0.42	5.4	1.1	0.38	36	11	44	275	32
9	1.4	0.59	6.7	0.9	0.41	35	36	98	137	31
10	1.7	0.57	4.8	1.4	0.54	27	78	51	162	37
11	1.8	0.59	6.7	1.6	0.50	44	14	69	73	34
12	1.6	0.56	3.6	1.8	0.55	28	118	78	197	43
Second Season (2003)										
1	1.4	0.34	4.6	1.3	0.48	29	18	44	101	27
2	1.8	0.61	3.2	1.8	0.43	26	11	72	182	38
3	1.3	0.42	4.0	1.5	0.61	42	8	65	226	32
4	1.9	0.62	3.5	1.9	0.63	29	12	66	105	27
5	1.7	0.50	3.2	1.4	0.41	78	33	25	270	15
6	1.5	0.43	4.6	1.9	0.55	31	109	64	466	31
7	1.5	0.43	4.0	1.4	0.42	30	41	33	88	19
8	1.8	0.53	5.8	1.0	0.38	28	7	48	318	44
9	1.4	0.40	4.7	0.9	0.36	36	8	38	122	30
10	1.5	0.50	5.8	1.1	0.52	41	66	45	115	29
11	1.1	0.44	4.5	1.4	0.46	36	5	35	100	28
12	1.9	0.36	5.5	1.5	0.45	40	7	59	76	24