High Soil Moisture and Low Soil Temperature Are Associated with Chlorosis Occurrence in Concord Grape

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Abstract. Leaf yellowing (chlorosis) is not unique to Concord grape, yet occurs with great intensity in the arid, irrigated central Washington state growing region. Past research on nutrients has not shown a clear cause and effect relationship between soil and/or plant nutrient status and chlorosis. We investigated both nutritional and climatic conditions for their association with chlorosis occurrence. Six vineyard sites were selected, 2 each with no history of chlorosis (achlorotic), occasional chlorosis, and annually reoccurring chlorosis (chronically chlorotic) and monitoring sites in chlorotic and achlorotic areas were established. Nutrient elements K, Ca, Mg, Mn, and Cu plus the nonnutrient elements Na and Al were monitored in soil (surface, 0 to 30 cm, and subsurface, 30 to 75 cm, depths) and leaf tissue (both petioles and blades) prebud burst (soil only), at bloom, and preveraison at 650 degree days at all vineyard sites for the 2001, 2002, 2003, and 2004 growing seasons. In addition, both soil temperature and moisture were monitored. To evaluate the intensity of chlorosis at each site, chlorotic vines were GPS marked and mapped post-bloom each year. Overall, chlorosis incidence was more widespread in 2001 and 2003 than in 2002 or 2004. There were few relationships with soil or tissue nutrient concentrations. However, soil moisture was consistently higher and soil temperature lower in the period between bud burst and bloom in the chlorotic sites. This suggests that a cold, wet soil environment prior to bloom impedes grape root growth and/or function and triggers plant chlorosis. Yearly differences strongly support this finding.

Leaf yellowing, or chlorosis, appears in a number of perennial fruit crops grown in the irrigated areas of Central Washington State. In this growing region Concord grape (*Vitis labruscana* Bailey) shows symptoms every year, with variation in how widespread the occurrence is from year to year. Concord grape yield significantly declines in chlorotic vines as they lose leaves and, with time, vigor (Ahmedullah and Kawakami, 1983). Bavaresco et al. (2005) found a similar decline in *Vitis vinifera* L. 'Cabernet Sauvignon' when grafted to a lime-susceptible rootstock.

Research into the causes of this type of chlorosis has had limited success in identifying causes and developing management strategies. Korcak (1987) found widespread chlorosis in fruit trees when soil pH and calcium carbonate levels were high. Studies on grapevine have associated grape chlorosis occurrence with high levels of soil bicarbonates (Dow and Tukey, 1985; Mengel et al., 1984). However, Li et al. (2005) found that in addition to high soil carbonate/bicarbonate content, fruit tree chlorosis was also found when soil bulk density increased and DTPA-extractable

iron decreased in the 20 to 40 or 40 to 60 cm soil depth. Since soils throughout the central Washington growing region are typified as having high soil pH (>7.7) and presence of free calcium carbonates at relatively shallow depths (NRCS, 2005), the findings of Li et al. (2005) support the concept that some factor beyond pH and calcium carbonates are

involved in grape chlorosis in areas similar to our growing region.

Research on cranberry (*Vaccinium macrocarpon* Ait.) chlorosis found that the symptoms occur in response to water stress (Lampinen, unpublished data). The suggestion that some aspect of climate, including soil climatic conditions, may contribute to chlorosis is supported by the recent findings that the physiological disorder blackleaf in grape is triggered by climatic (including soil) conditions (Olmstead et al., 2005; Smithyman et al., 2001). The year to year variability in the extent of grape chlorosis further supports the concept of climate as a potential contributor to the extent of the disorder.

Although the symptoms are classical of a number of different nutrient deficiencies (Ahmedullah et al., 1983; Marschner, 1986), research trials of different foliar nutrient sprays have been shown to have limited success in either alleviating the symptoms or reducing vine vigor decline (Ahmedullah and Kawakami, 1983; Stevens, 1998). These findings are consistent with other research that found limited, and only short-term, success in alleviating chlorosis with supplemental iron fertilizers (Natt, 1992; Veliksar et al., 1995).

Thus, fruit tree and grape vine research suggest several different possible mechanisms to the development of grape chlorosis. The objective of this research was to study the following potential causes of grape chlorosis: 1) single element nutrient deficiency; 2) a multiple element nutrient deficiency or insufficiency (e.g., a transient deficiency [Marschner, 1986]); 3) high concentration of one nutrient element causing the exclusion of uptake of other nutrient elements; 4) physiological stress due to climatic (including soil) conditions; and 5) a combination of these factors. To this end, six vineyard sites in the Yakima Valley were intensively studied over

Fig. 1. Relative location of six vineyards for chlorosis study. Sites 2 and 3 are achlorotic, sites 1 and 6 occasionally, and sites 4 and 5 chronically chlorotic.

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a 4-year period to evaluate potential contributors to grape chlorosis.

Materials and Methods:

This study was initiated in April 2000 in six different vineyards in the Yakima Valley in Central Washington (Fig. 1). Vineyards were located in Prosser, Grandview, and Sunnyside, between $-119^{\circ}50'28''\text{W}$ and $-119^{\circ}41'1''\text{W}$ longitude and $46^{\circ}24'78''\text{N}$ and $46^{\circ}15'53''\text{N}$ latitude. All vineyards consisted of well established mature vines (>15 years old) and were sprinkler irrigated commercial operations. To study the array of chlorotic conditions typical of the region, two vineyards each of three different types were chosen: achlorotic vineyards, occasionally chlorotic vineyards, and chronically chlorotic vineyards. The soils at all sites were Warden fine sandy loam soils (coarse-silty, mixed mesic superactive mesic Xeric Haplocambid) except site 5, which was a Shano silt loam (coarse-silty, mixed mesic superactive mesic Xeric Haplocambid). By definition, sandy or silty loam soils can have as low as 0% clay and, in a low clay situation, the line between silty loams and sandy loams is largely a difference between the distribution of silt and sand (Brady and Weil, 1999). The loess derived soils in the Yakima Valley, including those in our study site, have very low clay content ($<3\%$) and typically between 35% and 65% silt (Davenport, unpublished data).

Chlorosis tracking and vineyard monitoring was established in April 2000 and continued through September 2004. Three monitoring areas were established throughout the vineyard block at the achlorotic vineyards, sites 2 and 4. Six monitoring areas were established at sites 1, 3, 5, and 6, with three each in areas that historically were chlorotic or achlorotic. To determine if there were any difference in nutrients of chlorotic and achlorotic areas in the vineyard blocks soil samples were collected three times (prebud burst, bloom, 650 degree days) and plant tissue samples (leaf blades and petioles) were collected at bloom and 650 degree days. The 650 degree day sampling was used as a preveraison sampling period based on the number of degree days (based 10°C) to be consistent with recent research on Concord grape (Davenport et al., 2003; Keller et al., 2004). At each sampling interval, 30 total soil or tissue samples were collected, with 12 to 18 chlorotic or achlorotic samples each year. Soil samples were collected from the standard surface soil depth (0 to 30 cm) and from the subsurface soil (30 to 75 cm), which has been identified as the area of root proliferation in Washington Concord grape (Wample et al., 2000). Soil samples were a composite of three cores taken within a 0.5-m radius of the sample vine. Leaves opposite the basal cluster were sampled on both chlorotic and achlorotic plants, sampling achlorotic tissue. Samples were analyzed using Atomic Absorption Spectroscopy (Wright and Stuczynski, 1996) by a commercial test lab (Cascade Analytical, Wentachee, Wash.) for Na, Ca, Mg, K, Cu, Fe, and Al for both soil and tissue. Soil pH was analyzed on the spring soil samples only.

In each of the monitoring areas, soil moisture and temperature were recorded. Soil moisture measurements were collected weekly to a depth of at least 1 m using a neutron probe, with readings in 20 cm increments. Soil temperature data was collected with two-channel ruggedized temperature data loggers (Onset Computers, Bourne, Mass.) which were buried in the soil adjacent to the neutron probe access tubes. Data was collected both at 5 and 30 cm below the soil surface on an hourly basis beginning at bud burst and continued until just prior to harvest.

In addition, weather data from 1999 and 2000 (the two years proceeding the study) through October 2004 was compiled (PAWS, <http://index.prosser.wsu.edu>). Data compiled were average, maximum and minimum air temperatures ($^{\circ}\text{C}$), precipitation (mm) and evapotranspiration (ET, mm). The data were used to evaluate weather trends in an 18 to 24 month period before the occurrence of chlorosis to establish if there is a characteristic set of climatic conditions associated with years when grape chlorosis is widespread.

Plant tissue, soil, and weather data were analyzed using ANOVA and regression analysis (PROC GLM, PROC REG) with PC SAS (SAS Institute, Cary, NC).

The studied portion or entire vineyard block was mapped to track the increase or decrease in site specific chlorosis with time and to verify

that monitoring zones were in chlorotic or achlorotic sites. This was conducted just after bloom, when chlorosis was clearly visible, using a Trimble Ag-122 Global Positioning System (GPS, Trimble Corp., Sunnyvale, Calif.). The entire row length was traveled and plants with any visible chlorosis symptoms were marked. Maps of the chlorosis extent were made using ArcView 3.2 software (ESRI, Redlands, Calif.). The two vineyards chosen as achlorotic sites did not develop chlorosis symptoms during this experiment despite several years where chlorosis was extensive in the region.

Results and Discussion

Chlorosis was associated with higher levels of soil Ca and Mg as well as lower levels of Fe and Mn (Table 1). Soil Al was statistically lower in the chlorotic sites but the actual value difference is small and unlikely to be biologically significant. Only Mn and Al were significantly different in leaf tissue, with Al higher and Mn lower in leaves from chlorotic plants (Table 2). Lower leaf tissue Mn in chlorotic plants has been identified in past work on chlorosis, yet foliar Mn supplements were not found to be effective at reversing chlorosis (Ahmedullah and Kawakami, 1983; Stevens, 1998). The lower level of Fe in soils but not in tissue in chlorotic plants may be explained by the method of measurement. Recently, Smith

Table 1. Average nutrient concentration for soils collected at three sample times (prebud burst, bloom, and 650 degree days) and two depths from 2001–04 in locations within vineyards with and without chlorosis.

Parameter	Concn (mg·kg ⁻¹)						
	K	Ca	Mg	Fe	Cu	Mn	Al
Presence of chlorosis							
Yes	292	4772	407	13	1.3	3.8	1.0
No	275	2455	354	35	1.8	6.2	1.1
Soil depth							
0–30 cm	378	3342	377	24	1.3	5.3	1.1
30–75 cm	190	2904	387	22	1.7	4.6	1.0
Level of significance							
Chlorosis (C)	NS	**	**	**	NS	*	**
Soil depth (S)	**	NS	NS	NS	NS	NS	NS
Sample time (T)	NS	NS	NS	NS	NS	NS	NS
C × K	NS	NS	NS	NS	NS	NS	NS
C × T	NS	NS	NS	NS	NS	NS	NS
L × T	NS	NS	NS	NS	NS	NS	NS
C × L × T	NS	NS	NS	NS	NS	NS	NS

NS, **Nonsignificant or significant at $P < 0.01$ or 0.001, respectively.

Table 2. Average nutrient concentration for leaf tissue at two sample times (bloom, 650 degree days) from 2001–04 in locations within vineyards with and without chlorosis symptoms.

Parameter	Concn (mg·kg ⁻¹)			Concn (%)			
	K	Ca	Mg	Fe	Cu	Mn	Al
Chlorosis present							
Yes	1.89	1.66	0.78	165	8.3	41	100
No	1.94	1.58	0.40	152	8.7	68	83
Tissue type							
Leaf blade	1.02	1.78	0.61	247	9.2	68	132
Leaf petiole	2.80	1.45	0.55	70	7.8	43	50
Level of significance							
Chlorosis (C)	NS	NS	NS	NS	NS	**	*
Tissue type (L)	**	**	NS	**	**	**	**
Sample time (T)	**	**	NS	**	**	**	**
C × K	ns	NS	NS	NS	NS	NS	NS
C × T	NS	NS	NS	NS	NS	NS	NS
L × T	NS	NS	NS	NS	NS	NS	NS
C × L × T	NS	NS	NS	NS	NS	NS	NS

NS, **Nonsignificant or significant at $P < 0.01$ or 0.001, respectively.

Fig. 2. Soil moisture in the root zone measured with neutron probe averaged across shallow (30 cm) and deep (75 cm) sampling depths for a) all 4 years (2001–04) and b) during the bud burst to bloom period in high (2001–03) vs low (2002–04) chlorosis pressure years. Bars are ± 1 standard error.

and Cheng (2005) reported that leaves of plants grown with different rates of soil applied Fe did not show differences in leaf tissue total Fe but did show differences when active Fe was extracted and measured. Regardless, the limited relationship between soil and plant nutrient differences associated with chlorosis occurrence suggests that neither nutrient abundance nor limitations alone trigger chlorosis.

Spring soil pH was significantly different by sample depth but not between chlorotic and achlorotic sites ($P = 0.0001$ and 0.1569 , respectively). The soil pH at these sites was typical of the region, ranging between 7.7 to 8.5 in the surface and 7.7 to 8.6 in the subsurface soils. Free calcium carbonate was not tested at these sites, but soil series descriptions indicate that the Warden soil becomes violently effervescent at about 40 cm depth

and Shano at about 80 cm (NRCS, 2005).

There were differences between sites in soil depth to impermeable layer, which affected how deeply soil moisture could be monitored. To determine if there were any differences between soil moisture at chlorotic or achlorotic monitoring locations, soil moisture data at 20 cm (midpoint of 0 to 30 cm depth) and 60 cm (midpoint of 30 to 75 cm depth) depths were used, since soil moisture measurements were possible to this depth at all monitoring locations. However, analysis of the data showed no consistent differences in soil moisture by depth ($P > 0.01$, data not given). Analyzing the data across all four years did show that soils at the chlorotic sites were wetter in the early part of the season (Fig. 2). Most importantly, this difference was pronounced during the period between bud burst and bloom across all four years (Table 3, Fig. 2).

Soil temperature measured across the entire growing season both at the surface (5 cm) and in the root zone (30 cm) did not show differences between chlorotic and achlorotic sites (data not given). However, when soil temperature for the period from bud burst through bloom was evaluated, the average maximum soil temperature (both surface and root zone) were lower in chlorotic sites than achlorotic sites (Fig. 3). Further, there was a trend of lower average soil temperatures in chlorotic areas. Lower soil temperatures have been documented to reduce root growth and increase the effective concentration of plant available nutrient in the soil needed for plant uptake (Marschner, 1986). In addition, there was a trend of higher minimum soil temperatures in chlorotic areas, suggesting that higher soil moisture in the chlorotic sites acted as a buffer and reduced soil temperature fluctuations.

There were no obvious patterns of differences in monthly average, maximum, minimum air temperatures or ET in any of the years that suggest a distinct set of climatological conditions associated with triggering chlorosis (data not shown). There were noticeable differences by year in cumulative precipitation. Total cumulative precipitation between dormancy and harvest was distinctly higher in 2000, the year prior to the study, and in 2003 (Fig. 4a) and during the period between bud burst and bloom, precipitation was lowest during 2002 and 2004 (Fig. 4b).

The extent of chlorosis, measured as plants with chlorosis symptoms post-bloom, varied by site and year (Figs. 5, 6, 7, and 8). Chlorosis was most extensive across sites 1 and 3. Site 1 was initially being chosen as an occasionally chlorotic site. In 2001 the irrigation system at this site was changed from furrow to overhead sprinkler irrigation and there was a concurrent increase in soil moisture at this site (Davenport et al., 2003; Mills, unpublished data). Chlorosis symptoms had not been seen on that vineyard prior to the change in irrigation system, however, it became widespread following the change in irrigation management (Fig. 5). Likewise, site 5 was chosen as a chronically chlorotic site yet had decreasing levels of chlorosis during the study period. The yearly maps of each vineyard clearly show that, with time,

Table 3. Day of year of key phenological stages for Concord grape during the time period from 2001–04.

Year	Bud burst	Bloom	650 Degree days	Veraison
2001	108	141	201	229
2002	108	161	199	238
2003	105	157	198	227
2004	99	153	198	233

Fig. 3. Surface (0–5 cm, s) and root zone (about 30 cm, rz) soil temperature expressed as daily average (avg), maximum (max), or minimum (min) measured with Hobo sensors over 4 years (2001–04). Solid bars show significantly different values between chlorotic and achlorotic sites at the $P < 0.05$ level. Inset table shows differences in maximum surface soil temperature in high (2001–03) vs low (2002–04) chlorosis pressure years.

Fig. 4. Cumulative monthly precipitation for the year preceding the study (2000) and the 4 years of the study (2001–04) a) for the entire season; and b) during the period between bud burst and bloom.

chlorosis occurs in a consistent area within a given vineyard.

Looking at the annual extent of chlorosis in the chlorotic vineyards, it is apparent that there was greater chlorosis presence in 2001 and 2003 when compared with 2002 and 2004 (Figs. 5, 6, 7, and 8). Total cumulative precipitation between dormancy and harvest suggests that 2003 was a wetter year and 2001 was a very dry year (Fig. 4a). In fact, 2001 was a drought year (Scott et al., 2002). However, due to drought conditions there were irrigation water restrictions in 2001 which influenced irrigation management. As a response to forecasts of drought conditions, fruit growers in the Yakima Valley typically irrigate at high rates in the spring, using the available cold snow melt water. In addition, cumulative precipitation between bud burst and fruit set was also higher in 2001 and 2003 than 2002 and 2004 (Fig. 4b).

Both soil moisture and soil temperature data collected from bud burst through bloom was classified into high (2001, 2003) or low (2002, 2004) chlorosis years. When compared for chlorosis incidence in the high pressure years, maximum surface soil temperature was highest at achlorotic sites whereas at chlorotic sites maximum soil temperature differed between high and low pressure years (Fig. 3). Soil moisture in the period leading up to bloom was higher in both chlorotic and achlorotic sites in the high pressure years when compared to the low pressure years (Fig. 2b). The same analysis was conducted on soil and tissue nutrient levels and no differences were found. Thus, since soil moisture directly impacts soil temperature, this suggests that high early season soil moisture, particularly during bloom, triggers chlorosis. This may be related to an impedance in root growth, a reduction of root function, or a requirement for higher nutrient concentrations in soil solution in the cool, wet environment (Huang et al., 2005; Marschner, 1986).

Fig. 5. GPS-GIS maps of chlorotic vines in occasionally chlorotic site 1 from 2001–04.

Fig. 6. GPS-GIS maps of chlorotic vines in chronically chlorotic site 3 from 2001–04.

Conclusions

The findings from this study suggest that high soil moisture conditions are critical to chlorosis occurrence. Figures 5 through 8 clearly demonstrate that there are areas of each vineyard that are subject to chlorosis every year and, in high chlorosis pressure years, these areas dramatically expand. Both Perret and Koblet's (1984) work finding chlorosis in compacted soil zones resulting in wet, reducing conditions and the work of Li et al. (2005) showing chlorosis associated with high soil bulk density supports this finding. Our

Fig. 7. GPS-GIS maps of chlorotic vines in chronically chlorotic site 5 from 2001–04.

not of soil Ca levels, between high and low chlorosis pressure years.

Chlorosis expansion in high pressure years may be related to factors such as depth of soil to an impermeable layer (e.g., hardpan) or free calcium carbonates, neither of which were measured in this study.

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results are also consistent with the published literature associating chlorosis to high levels of soil Ca (Baveresco et al., 2005; Korcak, 1987). However, our results suggest that in alkaline soils, early season moist soil conditions result in an environment that adversely affects grape root function, triggering chlorosis, which is further exacerbated by presence of high levels of soil Ca. This is supported by differences in soil moisture and temperature conditions, but

Fig. 8. GPS-GIS maps of chlorotic vines in occasionally chlorotic site 6 from 2001–04. (note: no chlorotic vines in 2004).