Canopy and environmental control of root dynamics in a long-term study of Concord grape

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Summary

- Below-ground carbon allocation represents a substantial fraction of net photosynthesis in plants, yet timing of below-ground allocation and endogenous and exogenous factors controlling it are poorly understood.
- Minirhizotron techniques were used to examine root populations of *Vitis labruscana* Bailey cv. Concord under two levels of dormant-season canopy removal and irrigation. Root production, pigmentation, death and disappearance to a depth of 110 cm were determined over two wet and two dry years (1997–2000).
- There was continual root production and senescence, with peak root production rates occurring by midseason. Later in the season, when reproductive demands for carbon were highest and physical conditions limiting, few roots were produced, especially in dry years in nonirrigated vines. Root production under minimal canopy pruning was generally greater and occurred several weeks earlier than root production under heavy pruning, corresponding to earlier canopy development. Initial root production occurred in shallow soils, likely due to temperatures at shallow depths being warmer early in the season.
- Our study showed intricate relationships between internal carbon demands and environmental conditions regulating root allocation.

Key words: below ground, carbon allocation, Concord grape, exogenous vs endogenous control, lianas, root dynamics, root production, *Vitus labruscana*.

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Introduction

Understanding environmental factors and source–sink relationships controlling root growth is critical to understanding how plants may adapt to a changing climate, as well as being essential to efficient agricultural management of woody crops. Although it has been demonstrated that a substantial fraction of a plant's carbohydrate supply can be allocated below ground (Lambers, 1987; Jackson *et al.*, 1997), greater knowledge of the timing of root growth and death and factors regulating this timing is crucial to understanding how plants function in the environment. It is recognized that internal carbon demands in plants (endogenous factors) should interact with environmental factors (exogenous factors) to regulate seasonal changes in root systems (e.g. Tierney *et al.*, 2003). However, our current understanding of how endogenous or exogenous factors regulate root dynamics is limited, particularly under field conditions.

There is particularly limited understanding of how plants balance carbon allocation among different tissues. Although ideas such as optimization theory suggest that plant carbon will be deployed to those absorptive tissues whose resource acquisition most limits plant growth (e.g. Bloom *et al.*, 1985), certain carbon sinks within a plant, such as reproductive organs, demand substantial carbon even though they are not effective resource-acquiring organs. In particular, reproductive organs and roots are often considered to compete for plant carbohydrates (Hansen, 1977). For example, leaf and fine root recovery in response to canopy removal was at the expense of fruit production (Eissenstat & Duncan, 1992). However, there can be trade-offs between reproductive and below-ground allocation, with high reproductive allocation reducing below-ground allocation (Palmer, 1988; Forshey & Elfving, 1989; Rosecrance *et al.*, 1996 and citations therein; Berman & DeJong, 2003). Even in young plants during establishment, high reproductive allocation can decrease below-ground allocation (McLean *et al.*, 1992; Schreiner, 2003). It is unclear, however, how trade-offs in reproductive and root allocation might interact with below-ground resource limitations.

Carbon allocation of woody plants is complicated by their capacity for storage, which is a necessary part of their perennial life strategy for dealing with fluctuations in growing conditions. Woody plants with alternate-year mast cycles store resources to be used in alternate years (Rosecrance *et al.*, 1998). Mild water stress can decrease vegetative shoot allocation without affecting reproductive allocation (Dry *et al.*, 2001; Bryla *et al.*, 2003). Consecutive years of stress, however, could certainly lead to strong negative effects if carbon reserves are debilitated and never replenished. The balance among shoot, root and reproductive growth with resource limitations poses challenging but interesting issues.

Water availability interacts with root growth in a complex fashion. When soil moisture is high and aeration is adequate, root growth can be rapid owing to the abundance of water and lower soil impedance typical at higher soil water contents (reviewed by Richards, 1983). Moderate soil water stress can also enhance root growth, shifting allocation below ground to reduce water limitation for overall plant growth (Freeman & Smart, 1976; Richards, 1983; van Zyl, 1984; Bloom *et al.*, 1985). Even partial drying of root systems can lead to decreased vegetative shoot allocation (Dry & Loveys, 1999). Shoot growth may be more strongly affected by water limitations than is reproductive growth, which could cause carbohydrate reserves to be allocated for reproductive and root growth. Under severe soil moisture stress, however, limited root growth may occur (van Zyl, 1984) because of very low soil moisture availability and high soil impedance (Taylor & Gardner, 1963; Cornish *et al.*, 1984).

Whereas several environmental factors are known to affect root production under field conditions (e.g. Tierney *et al.*, 2003), environmental factors are seldom examined along with seasonal changes in plant carbon balance. Portions of the balance between shoot and root growth and timing of root growth are clearly under genetic control and are part of the life history strategy of plants (Oleksyn *et al.*, 2000). However, most studies of root dynamics have not been able to address both internal and environmental factors. Few studies of below-ground carbon allocation and timing of root growth and death have been of sufficient length to discern relative strengths of endogenous and exogenous factors affecting root dynamics under varying weather conditions. A notable exception was the study by Norby *et al.* (2004), which found the carbon balance of woody plants to be very responsive to elevated CO_2 over 6 yr. Sweetgum plantations more than doubled root production under elevated CO_2 while root mortality remained constant, effectively increasing the size of standing root populations. Long-term field studies are needed to discern such effects, which can be variable from year to year.

In its native range in the north-eastern USA, *Vitis labruscana* grows vigorously, sustaining high yields, and is typically grown on its own root system, unlike many wine grape varieties, which are grafted on rootstocks. In the wild state, grape vines allocate resources mostly to vegetative growth, especially in the shady understory, with large allocation to reproduction only when exposed to full sun (Possingham, 1994). In most viticultural production systems, pruning of up to 90% of the cane (dormant shoots) during the winter season is used to control above-ground vegetative growth, to reduce shading of the fruit and flower buds, and to restrict high reproductive allocation that may stress vines (Possingham, 1994). Minimal canopy removal has recently increased in popularity for native US grapes, resulting in larger early-season vine canopies but similar final canopy sizes and greater reproductive allocation compared with heavily pruned vines. These viticulture systems thus provide the opportunity to examine root dynamics in response to different patterns of above-ground growth in woody plants with intense competition for carbon between vegetative and reproductive organs.

Because patterns of root population dynamics and distribution in the soil profile have rarely been described in relation to shoot phenology and seasonal patterns, our objective was to examine the basic dynamics of root population development over several seasons in a reproductive woody plant with strong trade-offs between vegetative and reproductive growth. We examined root production, pigmentation, mortality and distribution in the soil profile of mature Concord grapevines (*V. labruscana*) under treatments of heavy and minimal dormant-season cane pruning, with and without irrigation. Treatments were ongoing for 6 yr before the study, ensuring that the vines were equilibrated to the treatments. Root dynamics were observed over four years that varied in rainfall, allowing the effects of fruit production on root dynamics to be assessed under different environmental conditions. We tested the hypotheses that: (1) compared with heavy pruning, minimal pruning promotes early-season root development corresponding to the earlier canopy development under minimal pruning; (2) non-irrigated vines produce more roots at deeper depths in dry years than vines receiving supplemental irrigation, owing to water being more available at deeper depths in dry years; (3) amongst the four treatments, vines exposed to minimal pruning and no irrigation produce the fewest roots, owing to their greater reproductive allocation; and (4) root production is inversely related to reproductive allocation because carbon allocated for reproduction limits the carbon available for root growth.

Materials and Methods

Study site and plant material

The study site was located at Cornell University's Vineyard Laboratory in Fredonia, NY, USA. Soils were a very well-drained, Chenango gravelly loam that was at least 2 m deep with no apparent restrictive layers. Vines were mature, 25-yr-old *Vitis labruscana* Bailey cv. Concord trained to a high-wire bilateral cordon system at 1.8 m above ground, spaced 2.4 m between vines and 2.7 m between rows. An experimental unit comprised five adjacent vines within a row, with an additional buffer vine on each end that received the same treatment but was not measured. Experimental units were randomly assigned to each treatment combination: one of two pruning treatments (heavy or minimal pruning), and one of two supplemental irrigation treatments (with and without irrigation) in a 2 × 2 factorial design (4 treatment combinations). The vineyard was divided into four blocks to account for soil heterogeneity, with each treatment combination occurring once in each block.

Pruning and irrigation treatments were initiated in 1991. Dormant vine pruning was conducted every winter. Balanced pruning, which requires heavy pruning, has been commonly used for grape production in the Lake Erie region because this regime balances size of the crop to vine growth, limiting crop size but leading to more consistent fruit maturation. Minimal pruning has recently gained popularity in this region because it is less labor-intensive.

Balanced pruning in the winter left 44 buds per kg of pruned canes (representing 10–15% of all buds produced), whereas minimal pruning entailed no pruning except for a hedge undercut at a 1-m height, to keep shoots off the ground, and cuts necessary to maintain vine individuality.

Irrigated vines received supplemental drip irrigation when necessary as determined by measurements of soil moisture (via neutron attenuation), shoot growth rates, vine midday stem water potentials and potential evapotranspiration taken at the site. The goal was to maintain soil moisture at levels necessary for sustaining leaf function, and measurements of leaf gas exchange showed that this goal was met in irrigated treatments. Soil moisture measurements were also taken in 1999 and 2000 in the vicinity of root observation tubes using time-domain reflectometry (TDR) to assess differences between irrigation treatments (Anderson *et al.*, 2003). Dates of bud break, bloom, veraison (fruit color change from green to red, indicating the start of fruit ripening) and harvest for experimental vines were recorded each year. Annual fruit yields for each experimental unit were measured at harvest, which occurred in late September or early October.

Root observations

Root populations were monitored through transparent plastic (cellulose acetate butyrate) root observation tubes (minirhizotrons), 183 cm long and 5.7 cm outside diameter, etched with a column of 127 numbered, 1.0 × 1.5 cm^2 windows. Although the type of plastic used in root observation tubes can affect root dynamics in some species (Withington *et al.*, 2003), relative differences within a species are not likely to be affected. Tubes were installed in early fall of 1996 at 30° from the vertical, perpendicular to the rows. A tube was installed in between adjacent experimental vines of each treatment so that each tube had at least two vines receiving the same treatment (including buffer vines) between it and a vine receiving a different treatment. Eight tubes for each pruning and irrigation treatment were examined, two tubes per block, selected randomly among installed tubes.

Images of roots visible in the windows were collected every 2 wk with a miniature video camera system (Bartz Technology, Santa Barbara, CA, USA) beginning in March 1997 through the growing season as described previously (Comas *et al.*, 2000). After fall dormancy, images were collected approximately once a month, unless the soil surface was snow- or ice-covered, until the following March, when collections were made again every 2 wk. Dates that individual roots were produced, turned noticeably brown without loss of turgor (pigmented), turned black and shrivelled (died), and disappeared were recorded (see Comas *et al.*, 2000 for details). Root stages of brown pigmentation and black/shrivelled were previously linked with sharp decline in metabolism and cessation of all activity in roots, respectively (Comas *et al.*, 2000). Root observations continued through October 2000.

Seventy-four windows with 1–17 roots were chosen at random to determine the relationship between numbers of roots in a window and root length. Total root length in these windows was measured with RooTracker (Duke University, Durham, NC, USA) and a regression was established between root number and length ($R^2 = 0.86$). Root production was then expressed as new root length per surface area of window.

Root starch concentration

Woody roots between 2 and 7 mm were collected in 1997 on October 23 from vines located next to those with root observational tubes and under the same management. Periderm was removed from oven-dried samples before grinding with a sample mill equipped with a 0.5-mm screen (Cyclotech 1093, Tecator, Sweden). Samples were boiled with deionized H_2O for 10 min, and were then cooled. For each sample, two subsamples were digested with 0.5 M sodium acetate (pH 4.8), one subsample having 5 units of amyloglucosidase and 2.5 units of α-amylase and the other without. Finally, glucose concentration was determined with colormetric analysis of the reduced glucose from supernatant extracted from each subsample, and the two subsamples were compared to determine the amount of glucose that was incorporated in starch (Nelson, 1944; Somogyi, 1952).

Statistics

Root observations from the two tubes in each experimental unit in each block were pooled. Data for all the tests run were found to be normally distributed using the Shapiro–Wilk test. Residuals indicated homogeneity of variance. To examine treatment effects on overall root production, total annual root production was tested with an ANOVA using a 2 × 2 factorial design between pruning and irrigation, with four blocks and four replicated years. Root production during vine phenological periods of the early and late season (*early*: early spring growth through flowering; *late*: fruit development through harvest) was examined separately with an ANCOVA in a factorial design of pruning and irrigation, including block and replicate year in the model and prior and current season yields as covariates (SAS Institute, Cary, NC, USA) to evaluate potential effects of fruit carbon demands. To examine the timing of root production specifically, root production between May and September in each of the top two depth intervals (0–20 cm and 21–40 cm) was examined with a MANOVA with repeated measures by month in a 2 × 2 factorial design of pruning and irrigation, with four blocks and two replicated years. Only data from wet years (1997 and 2000) were used in this comparison because only in these years were enough roots produced to allow splitting of root production by depth and month in this statistical analysis. The distribution of root production at different depth intervals over all four years was tested with a MANOVA with repeated measures for depth interval and a factorial of pruning and irrigation, including four blocks and replicate years. Two- and three-way interactions between year, pruning and irrigation with $P > 0.20$ were dropped from the models. Differences at $P = 0.05$ were considered clearly significant. Differences between $P = 0.10$ and $P > 0.05$ were considered marginally significant because of the limited statistical power typical of field root studies in which the variability and expense of replication are high.

Results

Environmental conditions and vine growth

Over the four years of the study, two years (1997 and 2000) were cool and wet and two years (1998 and 1999) were warm and dry (Table 1). There was a greater shortage in rainfall in 1999 than in 1998 (Table 1), which led to greater drought stress during the growing season in 1999 than 1998 because the number of degree-days was similar (data not shown). Spring was later in 1997 than in the other three years, causing all vine developmental stages in 1997 to occur from 10 to 40 d later than in the other three years (Table 1). Estimates based both on light interception (Fig. 1a) and shoot growth (Fig. 1b) indicated that minimally pruned vines develop their canopy more quickly than heavily pruned vines, although final canopy development was similar in the two treatments (Lakso *et al.*, 1997; Lakso, 1999a). Minimally pruned vines averaged *c.* 350 shoots per vine, whereas heavily pruned vines averaged *c.* 90 shoots per vine. Minimally pruned vines depleted soil water more rapidly early in the season (Lakso *et al.*, 1999), and had *c.* 35% higher annual yields on average compared with heavily pruned vines (Lakso, 1999b) (data not shown).

Table 1 Year-to-year differences in important stages of vine phenology, precipitation and mean daily temperature during the study

Stage*	Time	1997	1998	1999	2000
Early development	April 1 to bloom	Apr 1–Jun 28	Apr 1–Jun 5	Apr 1–Jun 9	Apr 1–Jun 12
Flowering/fruit set	Bloom to 30 days post bloom	Jun 29–Jul 28	Jun 6–Jul 5	Jun 10–Jul 9	Jun 13–Jul 11
Fruit development	30 days post bloom to veraison	Jul 29–Sep 9	Jul 5–Aug 17	Jul 10–Aug 14	Jul 12–Aug 22
Fruit ripening	Veraison to harvest	Sep 10–Oct 21	Aug 18–Sep 15	Aug 15–Sep 28	Aug 23–Oct 11
Shoot dormancy	Harvest to March 31 of following year	Oct 22–Mar 31	Sep 16–Mar 31	Sep 29–Mar 31	Oct 12–Oct 31
Precipitation (mm)	June 1–August 30	352	232	191	299
% of normal precipitation	June 1–August 30	136	90	74	115
Mean max. temperature (°C)	June 1–August 30	24.6	26.0	27.1	25.0

In the final year of the study, root observations ended soon after shoot dormancy. Precipitation for each year relative to average precipitation from 1926 to 2004 was used to determine precipitation for each year as a percentage of normal precipitation.

*'Early development' refers to the period just before budbreak about May 1 and includes the development of the current flower clusters and shoots to a leaf plastochron index of *c.* 10.

'Flowering/fruit set' refers to the flowering period and the initial fruit development via cell division to about half of final volume, including the final set of berries. Shoot growth is active.

'Fruit development' refers to the slowing of growth of the fruit before the beginning of ripening and the gradual decline of shoot growth.

'Fruit ripening' refers to a strong growth of the fruit in volume and in accumulation of sugar to final ripeness; usually accompanied by little growth of the rest of the plant.

'Veraison' refers to fruit color change from green to purple, indicating the start of fruit ripening.

'Shoot dormancy' refers to the postharvest period that leads to leaf senescence, and ultimate leaf fall by mid-November, and the winter dormancy period.

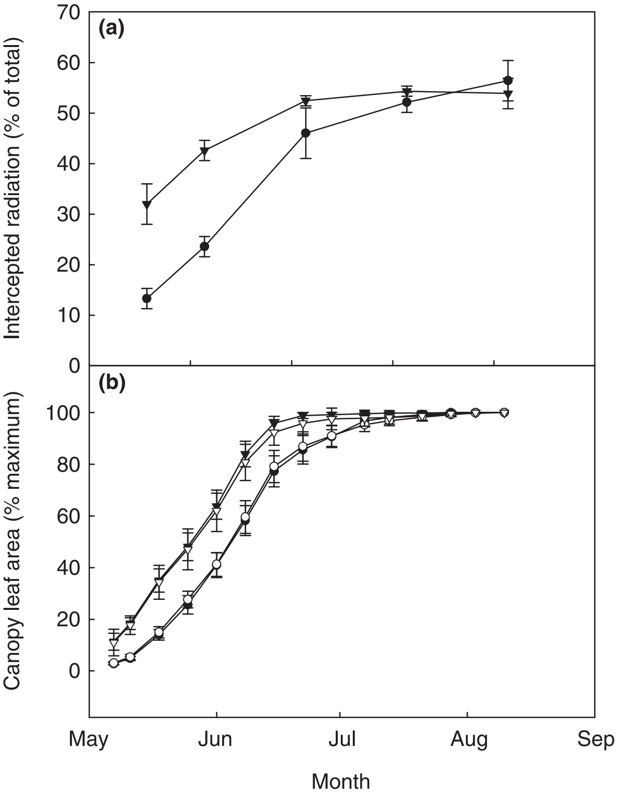

Fig. 1 Seasonal light interception and canopy size of vines compared among treatments. (a) Different patterns of seasonal radiation interception of balance-pruned and minimally pruned Concord grapevines in the same trial in 1993. Interception was estimated by the point-grid shadow area method as described by Wünsche *et al.* (1995). Each point of each treatment is the average of four experimental blocks. (b) Example of seasonal pattern of relative leaf area development in 1999. Forty shoots per treatment were marked and monitored for length at intervals. Leaf area per shoot was estimated by a length to leaf area regression obtained by shoots harvested from buffer vines at intervals. Error bars are standard errors. Filled circles, Bal/NI; open circles, Bal/Irr; filled triangles, Min/NI; open triangles, Min/Irr.

Standing root populations

Nondestructive estimates of root populations visible with the minirhizotrons indicated substantial year-to-year variability among the treatments (Fig. 2). In all treatments, the total population of roots present was continuously composed of both white and brown roots, rather than a complete change from populations of white roots to those of brown (Fig. 2a,b). Large populations observed in 1997 directly following the fall 1996 installation of tubes may have been due to initial effects of installation stimulating root growth, as previously reported elsewhere (Joslin & Wolfe, 1999). There was generally a rapid rise in root populations until veraison, with a unimodal peak in root population size (Fig. 2c). Peaks in brown root populations generally followed those in white root populations by approximately 3 wk (Fig. 2a,b). Although there was some evidence that in dry years (1998 and 1999), shifts in percentage of white to brown roots occurred sooner in nonirrigated treatments than in irrigated treatments, these patterns were not statistically significant (Fig. 2a,b; however, see Anderson *et al.*, 2003). Total root populations of nonirrigated treatments in dry years peaked sooner than those of irrigated treatments because there was limited later-season growth seen in irrigated treatments (Fig. 2c; see the next section). In wet years (1997 and 2000), there was little apparent difference in timing of root populations peaks between irrigated and nonirrigated vines (Fig. 2c). The percentage of brown roots dominated that of white roots later in the season, particularly after October and over winter, although there was always some quantity of white roots present (Fig. 2a,b).

Root production and mortality related to above-ground development

Patterns of root production in relation to vine phenology and fruit development were not consistent from year to year, but peak root production generally occurred during flowering/fruit set or fruit development depending on the treatment and year examined (Fig. 3a). Peak root production of irrigated vines in wet years occurred during flowering/fruit set but was slightly delayed until fruit development in dry years. Peak root production of nonirrigated vines was similar to that of irrigated vines in the first wet year (1997), was extremely low during the dry years (1998 and 1999), and was delayed compared with irrigated vines until fruit development in the wet year (2000), which followed the two dry years.

Minimally pruned vines generally produced more roots than heavily pruned vines in early stages of crop development (early development through flowering/fruit set), although this pattern was weaker in 1999 and 2000 ($F_{1,51} = 5.00$, $P = 0.05$; Figs 3a and 4). The earlier root production in minimally pruned vines coincided with earlier canopy development in these vines, which we observed to have little vegetative shoot growth past bloom compared with heavily pruned vines that have shoot growth for several weeks post-bloom (Fig. 1). Irrigation effects on root production during the early season weakly interacted with year with irrigated vines producing slightly more roots in 1998–2000 ($F_{3,51} = 2.17$, $P = 0.10$; Fig. 3a). In late stages of shoot phenology (after the initiation of fruit development), root production in dry years (1998 and 1999) was extremely limited without irrigation, and, within each pruning treatment, led to an overall significant effect of irrigation ($F_{1,54} = 4.90$, $P = 0.05$; Fig. 3a). The difference in root production between irrigated and nonirrigated vines in the dry years was largest during fruit development and was greater in the second dry year, 1999, than in the first, 1998 (Fig. 3a). There were no effects of pruning on late-season root production ($F_{1,54} = 0.38$, $P = 0.54$; Fig. 3a). In all years but 1998, root production virtually ceased after the beginning of fruit ripening.

Similar to root production patterns, patterns of peak root mortality in relation to vine phenology and fruit development

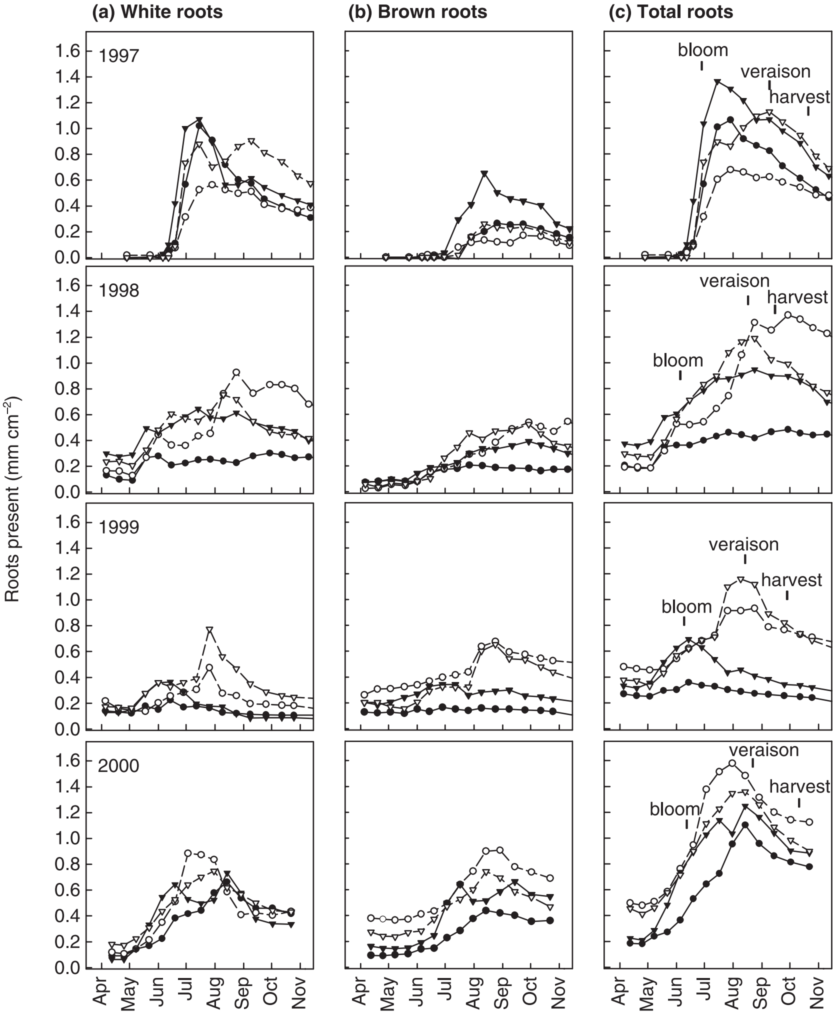

Fig. 2 Root population size visible using minirhizotrons (root length per observational area) of (a) white roots, (b) brown roots and (c) total roots (white + brown) in heavily pruned/nonirrigated (Bal/NI, filled circles), heavily pruned/irrigated (Bal/Irr, open circles), minimally pruned/nonirrigated (Min/NI, filled triangles) and minimally pruned/irrigated (Min/Irr, open triangles) Concord grape vines. Each point of each treatment is the average of root populations from four experimental blocks. White and total root population size was calculated from cumulative root production minus cumulative root pigmentation and death (black/shrivelled stage), respectively. Tick marks on the time axis indicate the beginning of the month. Across all dates and treatments, average SE of total roots present = 0.06, white roots present = 0.11, and brown roots present = 0.13, respectively. 1997 and 2000 were wet years; 1998 and 1999 were dry.

also varied from year to year, generally following peaks in root production by one stage in vine phenology (Fig. 3a,b). Peaks in mortality most often coincided with fruit development or fruit ripening (Fig. 3b). There were two exceptions when peaks in mortality did not follow that of production with the same offset. In 1998, root mortality of minimally pruned nonirrigated vines was delayed, following peak root production by two stages in shoot phenology rather than one stage. In 2000, peak root mortality of minimally pruned nonirrigated vines coincided with peak root production at the stage of fruit development, possibly indicating a shortage in carbohydrate supply.

Root distribution in the soil profile

Across all four years, pruning significantly affected total annual root production by depth, with minimally pruned vines producing more shallow roots than heavily pruned vines (significant interaction between pruning and depth; $F_{4,192} = 2.50$, $P = 0.05$). This effect was more pronounced in wet years (1997 and 2000; Fig. 4) than in dry years (1998 and 1999, data not shown). When root production was examined monthly for timing at different depths, peak root production under minimal canopy pruning was observed approximately 1 month earlier in shallow depths than root production under heavy pruning ($F_{4,24} = 2.74$, $P = 0.05$; Fig. 4; statistical analysis was limited to the top two depth intervals in 1997 and 2000 owing to limited root production for analysis on this fine scale). This early root production in minimally pruned vines in the shallow layers corresponded to the earlier canopy development of minimally pruned vines. Root growth in deeper soil layers may be limited by soil temperatures early in the season (data not shown).

Irrigation affected the overall vertical distribution of root production in dry years but not in wet years, leading to a significant interaction between irrigation and year ($F_{12,119} = 2.26$, $P = 0.05$; Fig. 5). In the first dry year (1998), root production of nonirrigated vines was limited in the 20–40 cm depth, and slightly higher in the 61–80 cm depth compared with root production of irrigated vines. In the second dry year

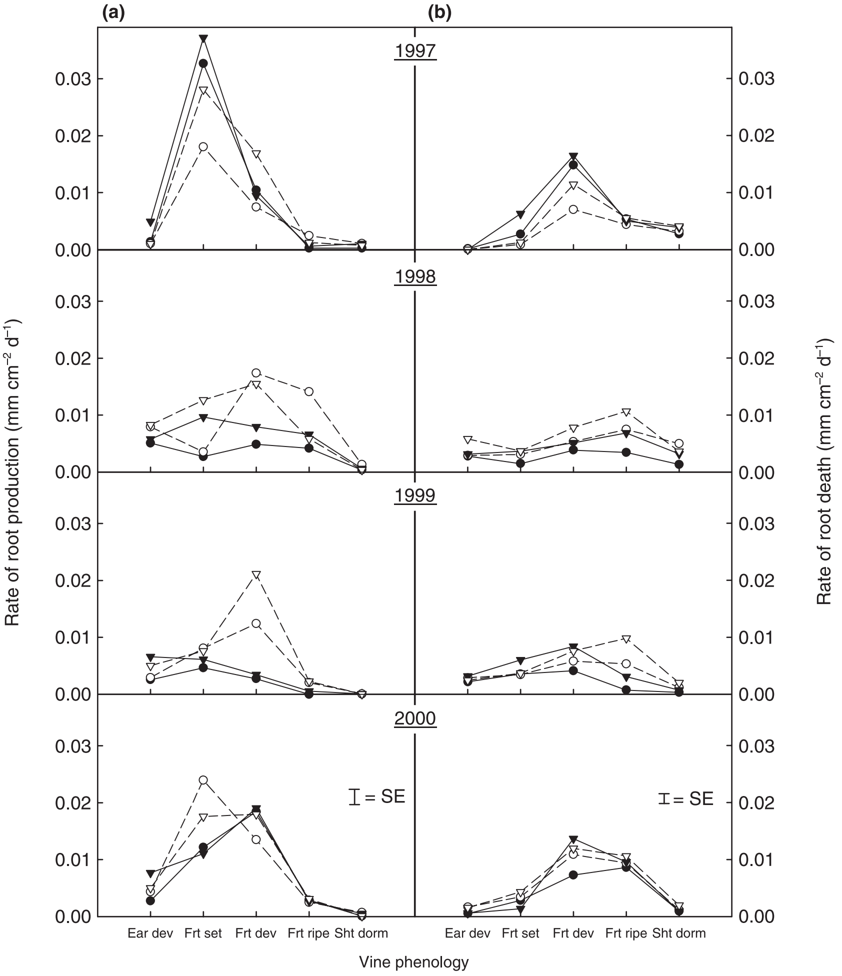

Fig. 3 Rates of root production and mortality over the season. (a) Average daily root production (root length per area of observational window per day) during five key vine phenological stages described in Table 1: early development (Ear dev), flowering/fruit set (Frt set), fruit development (Frt dev), fruit ripening (Frt ripe) and shoot dormancy (Sht dorm). In the early season (early development through fruit set) pruning had a significant effect on root production ($F_{1,51} = 5.16$, $P = 0.05$), while irrigation had a significant effect on root production in the late season (fruit development through shoot dormancy; $F_{1,54} = 4.90$, $P = 0.05$). (b) Average daily root mortality rate (root length per observational area per day) during the same five phenological stages. Early seasonal stages generally occurred before mid-July, where as late seasonal stages generally occurred after mid-July. Error bars indicate average standard errors (SE) across all dates and treatments for root production and death rate. Filled circles, Bal/NI; open circles, Bal/Irr; filled triangles, Min/NI; open triangles, Min/Irr.

(1999), root production of nonirrigated vines was limited over a wider depth interval (20–60 cm), and slightly higher than root production of irrigated vines in the 0–20 and 81–110 cm depths. When dry years (1998, 1999) were analyzed separately from wet years (1997, 2000), irrigation had a significant effect on root distribution in dry years ($F_{4,19} = 2.93$, $P < 0.05$). Surprisingly, root distribution of the dry years did not persist into the wet year of 2000, which followed the two dry years. Thus, fine root distribution did not appear to have lasting effects from year to year. Consequently, irrigation had marginal, if any, effect on root distribution when wet years were analyzed alone ($F_{4,19} = 2.24$, $P = 0.10$; Fig. 5).

Annual root production

Across the four years of the study, minimally pruned vines had 26% higher total-season root production than heavily pruned vines ($F_{1,52} = 4.19$, $P = 0.05$; Fig. 6a). There were no significant interactions between pruning and irrigation treatments, nor between pruning or irrigation and year of measurement. While minimally pruned vines always produced more roots than heavily pruned vines in nonirrigated treatments, heavily pruned vines occasionally produced more roots than minimally pruned vines in irrigated treatments, although these occurrences did not affect the overall statistical relationship (Figs 2a and 3a). There was a significant interaction in root production between irrigation treatments and year of measurement ($F_{3,52} = 2.77$, $P = 0.05$; Fig. 6b). In dry years (1998 and 1999), irrigated vines had 87% higher annual root production than nonirrigated vines. However, in wet years (1997 and 2000), root production was similar between irrigation treatments.

Across all four years, when vines had heavy reproductive growth in the prior year, they tended to produce more roots in the early stages of vine phenology in the following year, leading to a significant covariate of prior-year fruit production with early root production ($F_{1,51} = 3.80$, $P = 0.05$). Across all treatments, vines with heavy reproductive growth in 1997 had low starch concentrations in woody roots at the end of 1997 ($r = 0.62$; $n = 16$). Correlations within minimal and heavy pruning treatments followed the overall general correlation but correlations were weaker due to fewer observations (minimal: $r = 0.45$; heavy: $r = 0.20$; $n = 8$ in both). Low starch levels measured at the end of 1997 were associated with high early-season root production across all treatments in 1998

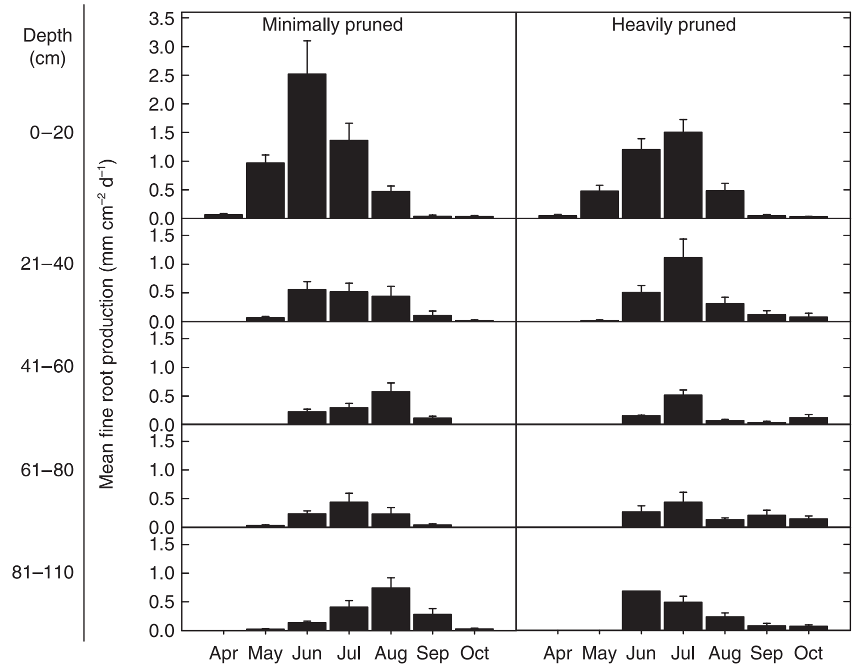

Fig. 4 Average root production (root length per observational window per day) by depth intervals in wet years (1997 and 2000). Each bar is the mean daily root production for each month per depth averaged over both irrigation treatments in four blocks for the two wet years. The earlier root production in the top 40 cm in minimally pruned vines compared with those heavily pruned and the overall shift in root production for both pruning treatments with depth as the season progressed were significant ($P < 0.05$). Statistical analysis was limited to roots produced in the top two depth classes (0–40 cm) due to small root populations in lower depths constraining analysis on a fine scale.

($r = 0.36$; $n = 16$) but were very weak within pruning treatments owing to few observations. Starch was not measured in subsequent years. Over all four years, correlations between prior-year reproductive growth and early-season root production within nonirrigated treatments were more positively correlated than those within irrigated treatments (heavily pruned/nonirrigated: $r = 0.56$, minimally pruned/nonirrigated: $r = 0.33$; heavily pruned/irrigated: $r = 0.14$, minimally pruned/irrigated: $r = 0.13$; Fig. 7). Correlations within nonirrigated treatments of heavily and minimally pruned vines resulted from overall differences in reproductive growth and early-season root production among all blocks in any given year, rather than particular blocks supporting higher early-season root production for a given prior-year yield.

At late stages of shoot phenology, no relationship was detected between prior-season reproductive growth and current root production nor between current-season reproductive growth and root production across all four years ($F_{1,54} = 0.00$, $P = 0.95$; $F_{1,54} = 0.00$, $P = 0.95$, respectively; data not shown). Likewise, no relationship could be discerned between prior or current season reproductive growth and total annual root production ($F_{1,51} = 1.47$, $P = 0.23$; $F_{1,51} = 0.14$, $P = 0.71$, respectively; data not shown).

Discussion

We found partial support for our hypotheses on factors affecting root production in Concord grape. Minimal pruning promoted earlier spring root development, which coincided with the earlier canopy development of minimally pruned vines compared with those heavily pruned. Size of root populations among the pruning and irrigation treatments of vines fluctuated between years and different times in the season, governed by endogenous and as well as exogenous factors at various times. Compared with minimal dormant pruning, we found that vines under heavy pruning produced fewer fine roots. Irrigation allowed more root production in dry years and affected the vertical distribution of roots in the soil profile. Heavy reproductive growth was generally associated with lower starch reserves in woody roots, implying that stored reserves may have been used for reproductive growth. In the latter part of the season, few roots were produced once reproductive development reach stages of high carbon demand on the vines. Across different years, heavy reproductive growth in a given year was associated with higher fine root production in the early part of the following year, indicating that greater reproductive allocation did not entirely hamper allocation to roots.

Environmental cues may be part of a signal for initial root production (Fitter *et al.*, 1999; Tierney *et al.*, 2003) but at least a portion of root production appears to be regulated by endogenous factors, possibly linked to photosynthetic supply. Whereas spring root production in all treatments was initiated around the time of bud break (Fig. 3), root flushing generally occurred more quickly in minimally pruned vines (Fig. 2a), corresponding to their faster canopy development (Fig. 1). Furthermore, within pruning treatments (and therefore independent of canopy development), we found additional evidence of endogenous control on root production with treatments that had larger reproductive allocation allocating more resources to root production in the early season of the following year. Biological reasons for increasing allocation below ground could include the facts that: (1) when vines grow

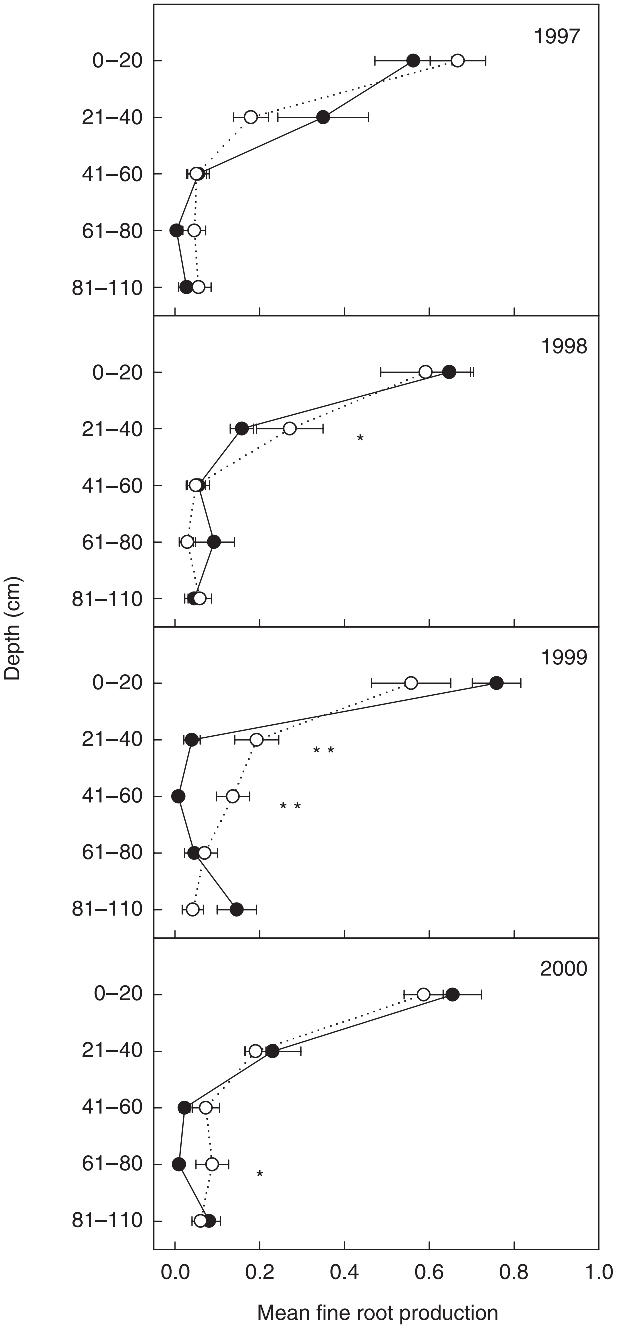

Fig. 5 Annual root production as a fraction of total production by depth in irrigated (open circles) and nonirrigated (filled circles) treatments. Each point is an average across heavy and minimal pruning treatments and four blocks for each year of the study. One asterisk (*) denotes significance at $P < 0.10$; two asterisks (**) denote significance at $P < 0.05$ for irrigation affects from individual factorial models analyzed for each depth and year.

vigorously and support heavy reproductive growth, they may also be able to support more root growth; (2) large reproductive allocation may have required more water and nutrients so that in periods following heavy reproductive growth, vines may have been stimulated to increase allocation to roots, which acquire water and nutrients; or (3) after a season of heavy reproductive growth when vines may not have been able to allocate many resources to roots, vines may have increased allocation to roots to make up for limited allocation during the prior period. Although vines with large reproductive growth had lower starch reserves in roots at the end of one season, increased root production in the early portion of the following year may have still been supported by starch reserves, which were low but not depleted, and by current photosynthates. Research tracking carbohydrate allocation with radioactive isotopes has demonstrated that root growth can be supported by current photosynthate (e.g. Thompson & Puttonen, 1992). Although optimization theory suggests that plants selectively allocate resources to acquire a limiting resource, shifts in

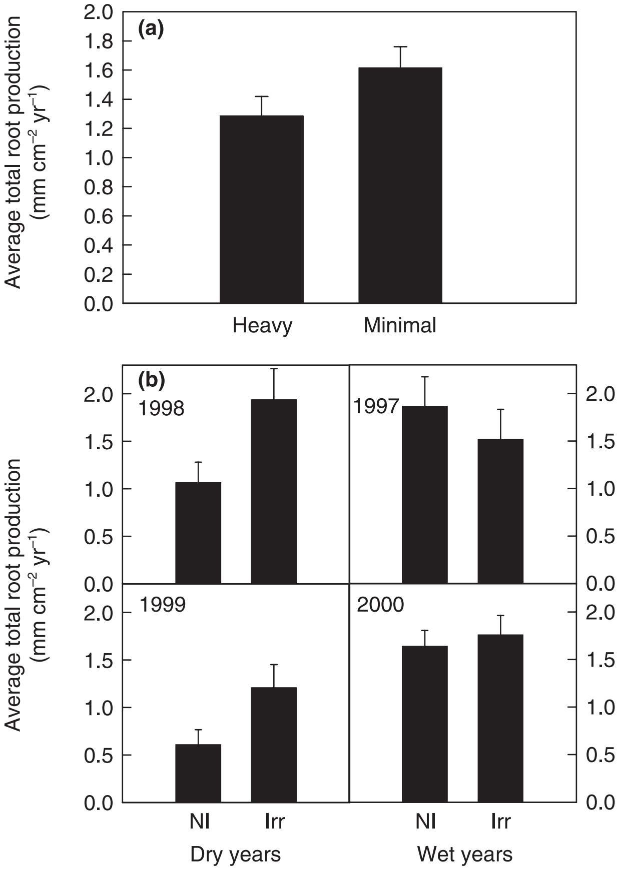

Fig. 6 Average yearly root production (root length per observational window per year) over four years in a Concord grape vineyard. (a) Root production in heavily pruned (Heavy) and minimally pruned (Minimal) treatments, averaged over irrigation treatments. Minimally pruned vines exhibited significantly higher annual root production than heavily pruned vines over the four years of the study ($P < 0.05$). (b) Root production in dry (1998 and 1999) and wet (1997 and 2000) years in irrigated (Irr) and nonirrigated treatments (NI), averaged over pruning treatments. There was a significant interaction of irrigation with year ($P < 0.05$). In dry years (1998 and 1999), root production in irrigated treatments (Irr) was higher than in nonirrigated (NI), but there was no general pattern in wet years.

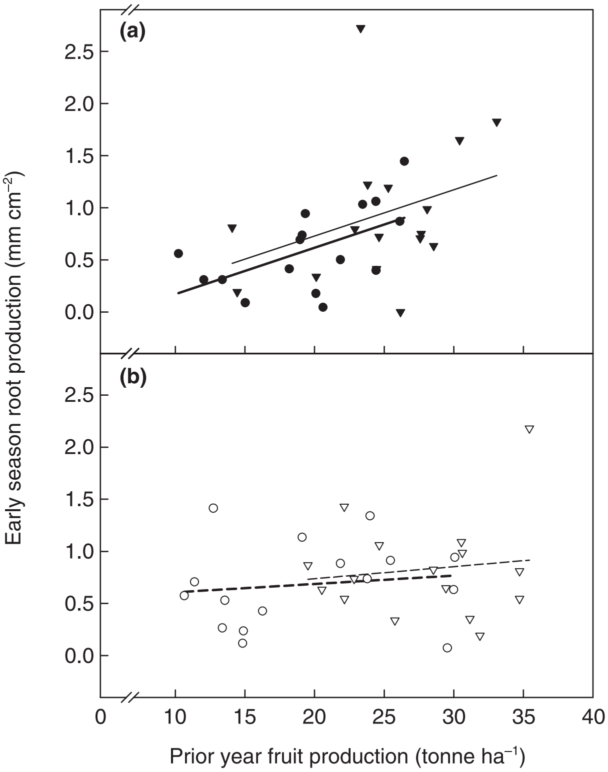

Fig. 7 Correlation of early-season root production (root length per observational window from April 1 through flowering) with prior-year fruit production (yield per area of vineyard) over four years in nonirrigated (a) and irrigated treatments (b). Early-season root production is defined as all roots produced during the time of bud and flower development, which generally occurs before mid-July Each point represents one block of each year. Over all four treatments, greater early-season root production was correlated with higher reproductive allocation in the prior year ($r = 0.32$; $P < 0.05$), although correlations within nonirrigated treatments were more positively related than within irrigated (heavily pruned/nonirrigated: $r = 0.56$, minimally pruned/nonirrigated: $r = 0.33$; heavily pruned/irrigated: $r = 0.14$, minimally pruned/irrigated: $r = 0.13$). Filled circles, Bal/NI; open circles, Bal/Irr; filled triangles, Min/NI; open triangles, Min/Irr.

allocation may only occur at times of the year, such as the early season, when strong competition from reproductive sinks are not present.

The internal carbon balance of the vines may have interacted with irrigation effects, leading to a diminished white root population in minimally pruned vines after two dry years. Minimally pruned vines, which had greater reproductive allocation than heavily pruned vines, did not have reduced capacity to produce roots in a single dry year following a wet year, but after two consecutive dry years, capacity for root production was diminished. Total root populations in minimally pruned vines without irrigation were still greater than those of heavily pruned vines in the second dry year, owing to minimally pruned vines having a large number of brown roots (Fig. 2). However, the metabolic activity of brown roots is low compared with white roots (Comas *et al.*, 2000).

Both endogenous and exogenous factors may have been responsible for limiting root growth during dry years. First, the second dry year (1999) had more intense drought than the first, which likely limited all root production without irrigation in the dry part of the season. Root production in dry conditions could be retarded owing to environmental conditions such as the soil being too dry to allow for root penetration or carbon limitation for root growth under these conditions. While photosynthesis is often reduced under dry soil conditions and could lead to carbon limitations on root growth, root respiration and growth are also greatly reduced, often leading to an increase of starch reserves in plants experiencing drought (Bryla *et al.*, 1997). Root growth of woody plants in climates with seasonal water patterns is often limited at dry times in the season when water is not available (e.g. Katterer *et al.*, 1995). Second, in 1999, reproductive allocation was 70 and 30% higher for heavily pruned and minimally pruned vines than in 1998, which, combined with reduced photosynthesis, may have greatly limited supply of current photosynthates for root growth. The delay in root production in nonirrigated vines during the wet spring of 2000 when environmental conditions should have been optimal for root growth might be indicative of carbon stress in vines in nonirrigated treatments after two dry years. Thus, it appears that a combination of factors may have limited root production in nonirrigated vines in dry years, with soil impedance possibly physically restricting root production in dry soil layers, and reduced photosynthesis eventually leading to limiting carbon availability for root growth.

Root lifespan affects standing populations of roots as much as root production. We had previously reported on the effects of pruning and irrigation on root lifespan over this 4-yr period (Anderson *et al.*, 2003). Irrigation did not affect root lifespan in dry years but slightly decreased root lifespan in wet years. Vines may have retained roots longer in years when root production was limited by dry soil and more readily shed roots selectively in wet years of high root production. Compared with heavily pruned vines, minimally pruned vines had longer-lived roots in wet years but shorter-lived roots in dry years, when minimally pruned vines may have been more stressed (Anderson *et al.*, 2003). Because root population sizes differ between wet and dry years, these interactive effects of pruning and irrigation on root lifespan suggest an intricate interplay between root production and lifespan.

Optimization theory would suggest that nonirrigated vines would have higher allocation to roots than irrigated treatments if plants maximize resource acquisition by allocating more resources to tissues acquiring limiting resources (e.g. Bloom *et al.*, 1985). There was greater stimulation in early-season root production after heavy reproductive growth in the previous year in nonirrigated vines of both pruning treatments (Fig. 7), possibly supporting the optimization theory. However, we did not find that nonirrigated vines had the largest root populations, possibly owing to physical limitations on root production from the soil environment under drought conditions. Nonetheless, as a fraction of net photosynthesis, *relative* allocation may have increased in nonirrigated vines even though we did not detect any increase in absolute root allocation.

While acknowledging plant control of root production and mortality, soil temperature is widely recognized as an important environmental cue for timing of root dynamics (Lyr & Garbe, 1995; Tierney *et al.*, 2003; Majdi & Ohrvik, 2004). Root production can be restricted when soil temperatures are low in the early spring and late fall. In grape, root growth generally occurs when temperatures are above 6°C and is optimum around 30°C, which is similar to many other temperate plants (reviewed by Richards, 1983). Root production in some woody species has been observed to occur continuously deeper in the soil profile while slowing at shallow depths as the season progresses (Lyr & Hoffmann, 1967). Soil temperatures fluctuate more widely at shallower than deeper depths; thus, soil temperature as a cue for root production is a complex signal affected by depth in the soil profile. Because carbon supply and sinks in a plant change over a season, soil temperature probably only exerts strong effects on root production at soil temperature extremes (e.g. below 10°C and above 35°C) (Richards, 1983; B. Huang, A.N. Lakso & D.M. Eissenstat, unpublished data).

In our study, seasonal production of roots appeared to be governed by a balance of both endogenous and exogenous factors. There was little evidence that either root production or root standing populations exhibited a consistent bimodal pattern, as reported previously for grape in more Mediterranean-type climates (van Zyl, 1988; Mullins *et al.*, 1992). Rather, root production was consistently unimodal for all treatments in wet years and irrigated treatments in dry years but varied in nonirrigated treatments in dry years. Bimodal root production in grape, similar to many temperate woody plants, typically has a large peak in the spring and a secondary peak in the fall (e.g. Mullins *et al.*, 1991). For example, in South Africa, root growth of Colombar/99R exhibits one peak at flowering and another peak at harvest (van Zyl, 1988). In this study as well as ours, root production tapered during fruit ripening. The lack of root flushes in fall in our system may result from the relatively short season, which ends very quickly following harvest as compared with other grape-producing regions.

In conclusion, our study along with others illustrates that the periodicity of root flushes may be jointly regulated by exogenous and endogenous factors: warming temperatures, moisture availability and carbohydrate supply from the shoot triggering root growth in spring; soil moisture limitations and competing carbon sinks restricting root growth in summer; and, in fall, moisture availability and carbohydrate supply from the shoot following harvest, triggering root growth as long as vines do not go immediately into dormancy. Our detailed examination of root production in Concord grape indicated that timing and quantity of root production was closely associated with canopy development when environmental conditions were favorable. There was little consistency in timing, however, of either peak root production or peak root standing populations from year to year, possibly owing to interactions between the carbon balance in the vines and climatic conditions. Simple predictions of timing of root production or standing population with shoot development, consequently, may not be possible. This study also illustrates the need for multiple years of root observations under field conditions to thoroughly investigate patterns of root dynamics associated with plant carbon balance or climatic conditions; only by understanding year-to-year variation can we interpret the relative strengths of endogenous and exogenous factors.

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