

Shading Effects on Dry Matter Partitioning, Remobilization of Stored Reserves and Early Season Vegetative Development of Grapevines in the Year after Treatment

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ABSTRACT. Grapevines (*Vitis vinifera* L.) were covered with an 80% neutral shade cloth from flowering until harvest to investigate effects of shade on early season vegetative development in the year after treatment. Shading reduced root dry weight, the concentration of soluble sugars, and amino nitrogen in xylem sap at budbreak, and leaf area expansion in the following year. Dry weight of roots on both shaded and nonshaded vines declined by more than 50% in the first 3 weeks after budbreak and then began to increase, but still had not recovered to prebudbreak levels, 10 weeks after budbreak. Total leaf area per shoot was reduced in the year after shading due to both fewer and smaller leaves.

The influence of shade on grapevine (*Vitis vinifera*) leaf photosynthesis, vine yield, and berry composition has been reported extensively in the literature. Within-vine shading reduced the photosynthetic photon flux (PPF) within the interior of a grapevine canopy to 5% of ambient (Smart, 1974; 1985, 1987), a value below the light compensation point for individual grapevine leaves (Poni et al., 1993; Smart, 1987). Shade reduced the number of berries that set per cluster (Ebadi and Coombe, 1996; Ferree et al., 1998), decreased cluster and berry weight (Archer and Strauss, 1989; Morgan et al., 1985; Smart et al., 1988), delayed initiation of fruit softening and coloration (Dokoozlian and Kliewer, 1996), and inhibited accumulation of soluble solids in fruit (Morrison and Noble, 1990). Shade altered vine morphology and productivity, decreasing the specific leaf dry weight, soluble carbohydrate and starch content of leaves, vine yield, total fruit soluble solids, total leaf area per vine, number of axillary shoots per cane, and weight of winter pruning (Cartechini and Palliotti, 1995).

The vegetative responses of grapevines in the year after shading have not been reported extensively in the literature. Keller and Koblet (1995) imposed two light regimes to potted grapevines for 20 d beginning at the onset of flowering and found no effect of treatment on total pruning weight or the nitrogen content of pruning wood in the following winter. Vines grown under a low light regime were severely affected by inflorescence necrosis in the year of treatment (Keller and Koblet, 1994) and displayed earlier budbreak and faster rates of vegetative development in the year following treatment (Keller and Koblet, 1995). The dry weight of all tissues was reduced when measured after 20 d of growth under a low light regime (Keller and Koblet, 1995), but the possibility for recovery of dry weight in the remainder of the season was not considered in their study.

Resumption of shoot growth in the spring represents a critical stage of development for deciduous woody plants. Root reserves play important and specific roles in supplying substrates for shoot respiration and growth, especially in woody species that flower and begin fruit development before substantial canopy development (Loescher et al., 1990). Sugars synthesized in grapevine leaves after harvest in the previous year and converted to starch in the wood and roots, are the first carbohydrates used by emerging shoots in the spring (Scholefield et al., 1978). Changes in the concentration of carbohydrates within a tissue, such as the reduction in starch concentration that occurs in grapevine roots in spring (Winkler and Williams, 1945) does not in itself represent a measure of the net loss or gain of carbon from that system. Rather, the absolute amount of starch in root tissues is the product of their dry weight (grams) and starch concentration (milligrams per gram dry weight). A reduction in the starch concentration within root tissues may occur concomitantly with formation of new root tissues, conceivably resulting in a net increase in the absolute amount of starch within that system. Grapevine root tissues are also a major site for storage and remobilization of nitrogen to support early spring growth (Obbink et al., 1973; Peacock et al., 1989). Whereas arginine is the predominant form of nitrogen in the bark and wood of grapevines (Kliewer, 1967), 70% of the total nitrogen present in xylem sap of grapevines at budbreak is transported as glutamine (Andersen and Brodbeck, 1989a).

According to Hale and Weaver (1962) and Koblet (1969), the dependence of new shoot growth in grapevines on stored reserves lasts only until the first few leaves on the shoots are about half their final size, at which time they start exporting more photosynthate than they import. However, Stoev et al. (1959) and Yang et al. (1980) reported that grapevine roots continue to mobilize reserves to the aerial portion of the plant up to the period of flowering. Keller et al. (1995) concluded that the leaves are clearly not the only source organs during bloom and that remobilization of reserves from the perennial parts of the vine may be continued or reinitiated beyond bloom.

The present study compares changes in dry matter partitioning and root starch concentrations during the first 10 weeks of growth in grapevines that had been shaded or nonshaded the previous

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season. The information provides an indication of the pattern of carbohydrate depletion and recovery in the roots of grapevines and the carryover effects of shading on the delivery of carbon and nitrogen required for new tissue growth in spring. The hypothesis for the study is how long does carbohydrate depletion continue in the spring and do stressed and nonstressed vines respond similarly?

Materials and Methods

PLANT MATERIAL. The vines used were the same as those in an earlier study of irradiance and light quality effects on grapevine growth and fruit quality (Hummel and Ferree, 1997). Two-year-old vines growing in the greenhouse on their own roots in 8-L nursery pots in a medium of 1 soil :1 peat :1 perlite (by volume) were pruned to a single bud at the beginning of the 1995 growing season. The arising shoot was maintained at 15 nodes and all laterals removed at regular intervals throughout the growing season. Vines were thinned to leave the basal cluster on each vine. 'DeChaunac' and 'Seyval Blanc' grapevines (64 and 32 vines of each cultivar, respectively) were grown in the greenhouse and half the plants of each cultivar were covered with an 80% shade cloth from bloom until harvest. The treatment reduced *PPF* by 85% under bright, clear conditions and 75% under overcast conditions compared to the nonshaded vines (Hummel and Ferree, 1997). In November 1995 all vines were placed in a coldroom (4 EC), then moved to a greenhouse once chilling requirements had been met (April 1996). 'DeChaunac' grapevines were arranged into eight randomized complete blocks with four shaded and four nonshaded vines in each replication. 'Seyval' grapevines were arranged into eight randomized complete blocks with two shaded and two nonshaded vines in each replication. Vines received no fertilization but were watered as required. The vines were left nonpruned so that each vine consisted of a single cane 15 buds in length supported vertically by a stake.

PLANT SAMPLING. A single 'DeChaunac' vine was harvested from each replication at four different times, chosen to represent the potential period of carbohydrate depletion and recovery of the root system. The times corresponded to budbreak (17 Apr. 1996) and 3, 5 (bloom) and 10 weeks after budbreak. A single 'Seyval' vine was harvested from each replication 4 weeks after budbreak (7 May 1996) and again 10 weeks after budbreak. Root, trunk, cane, and shoot tissues from each plant were separated, frozen immediately in liquid nitrogen, and stored at -20 EC. Tissues were lyophilized, weighed, and ground in a Wiley (Arthur H. Thomas Co., Philadelphia, Pa.) mill to pass a 40 mesh (0.635 mm) screen. Xylem sap was collected from the eight replicate 'DeChaunac' vines that were destructively harvested at budbreak after cutting the cane, leaving a 2 cm section at the base for attachment of latex tubing. Sap was collected into 15 mL plastic screwcap tubes placed in ice over a 24 h period and samples were stored at -20 EC until later analyses.

TISSUE ANALYSES. Soluble sugars were extracted from a 100 mg dry weight sample of lyophilized root tissues in hot (70 EC) 80% ethanol three times (7.5 mL + 3.0 mL + 3.0 mL) in a shaking water bath. The combined ethanolic extracts were partitioned in 7 mL each of water and chloroform. The aqueous extract was dried in vacuo at 40 EC and resuspended in 7 mL deionized water before being passed through a polyvinylpoly-pyrrolidone (PVPP) column. Total soluble sugars were quantified using the phenol-sulfuric acid method (Dubois et al., 1956). Excess ethanol was evaporated from the pellet remaining after ethanol extraction.

The pellet was then boiled for 2 h in 5 mL 0.1 N sodium acetate (pH 5.0). After boiling, the sample was incubated with 11.5 U amyloglucosidase (EC 3.2.1.3, Sigma, St. Louis, Mo.) and 11.5 U isoamylase (EC 3.2.1.68, Sigma, St. Louis, Mo.) in a shaking water bath for 16 h at 55 EC. The glucose liberated after incubation was quantified using the phenol-sulfuric acid method. Starch and soluble sugars were expressed on a percent dry weight basis.

SAP ANALYSES. Soluble carbohydrates in xylem sap were quantified using gas chromatography. Briefly, 2 mL of sap was taken to dryness under a stream of air in a heating block at 45 EC. Solids were dissolved in 100 μ L STOX reagent containing 8-phenylglucose as an internal standard (Pierce Chemical Company, Rockford, Ill.), and samples were vortexed and incubated in a 70-EC heating block for 30 min with occasional vortex mixing. After cooling, 100 μ L hexamethyldisilazane (HMDS) and 20 μ L trifluoroacetic acid (TFA) were added, and allowed to react for 60 min before analysis. These reagents result in formation of trimethylsilyl (TMS) derivatives of the sugars in each sample after their stabilization as oximes. TMS-oximes in a 2 μ L injection volume were separated on a packed column of 3% OV-17 on Chromosorb WHP using a Hewlett Packard 5890 Series II (Hewlett Packard, Avondale, Pa.) gas chromatograph with a flame ionization detector. Oven temperature was programmed for a 150 EC hold for 8 min to 240 EC at 15 EC/min. Peak areas were quantified using a Hewlett Packard 3396A integrator. The nitrogen content of xylem sap was determined colorimetrically using the α -amino nitrogen assay (Nivard and Tesser, 1965). Duplicate 50- μ L aliquots of xylem sap from each vine were adjusted to 1 mL with double deionized, distilled water. Ninhydrin reagent (1.6 g Ninhydrin + 160 mg Hydrindantin dissolved in 40 mL ethylene glycol monomethyl ester, 40 mL 0.5 N sodium acetate buffer, pH 4.65, added when dissolved) was added to the sample and vortexed before placing in a boiling water bath for 15 min. After cooling, 4 mL 2% collidine in 1 ethanol : 1 water was added to each sample and vortexed for 20 s to stabilize color. Absorbance was determined at 570 nm and the amino nitrogen concentration calculated against a series of glutamine standards that were included with each assay.

LEAF AREA ESTIMATION. Lamina length and width were measured twice weekly until 50 d after budbreak for each leaf on a basal, mid, and apical shoot on each plant. Basal shoots were selected from the middle region of the lowermost five buds, the mid shoot from the middle region of buds 6 to 10 from the base of the cane, and the apical shoot chosen from the middle region of buds 11 to 15 from the base of the cane. The area of individual leaves was estimated from calibration curves of area versus the product of lamina length and width obtained from a destructive sample of at least 200 leaves for each cultivar. Calibration curves were obtained using regression analysis (SAS Inst., Inc., 1989). Total leaf area per shoot was calculated as the sum of the individual area of all leaves on a shoot. The increase in leaf area per shoot over time was described by fitting a sigmoid curve to the data using nonlinear regression analysis in the SAS statistical package (SAS Inst., Inc., 1989).

STATISTICAL ANALYSES. Dry weight, leaf area, and xylem solute data are presented as means \pm SE of eight replicate vines for each treatment and time of sampling. Effects of shade were analyzed using the GLM procedure in the SAS statistical package. Changes in the dry weight of tissues between budbreak and 3 weeks after budbreak, and between 3 and 10 weeks after budbreak, were analyzed using single degree of freedom contrasts. The GLM procedure in SAS was used to test the significance of shade on

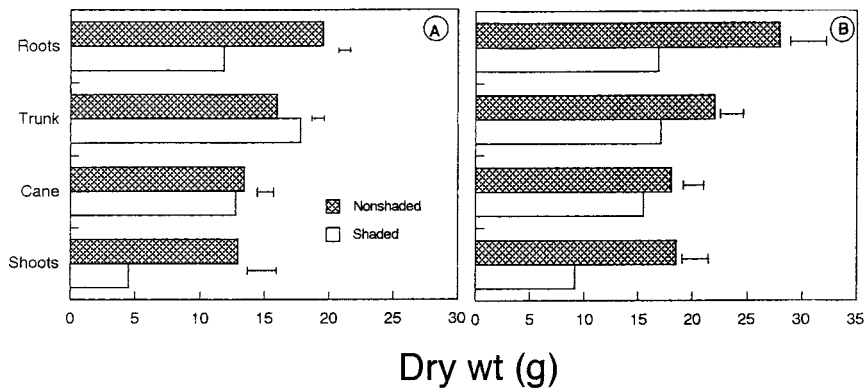


Fig. 1. Effects of shade in the previous year on dry matter partitioning of 'Seyval' grapevine tissues (A) 4 and (B) 10 weeks after budbreak. Data represent the mean of eight replicate vines. Horizontal bars represent the SE.

each of the parameters in the sigmoid function used to describe the increase in leaf area per shoot over time.

Results

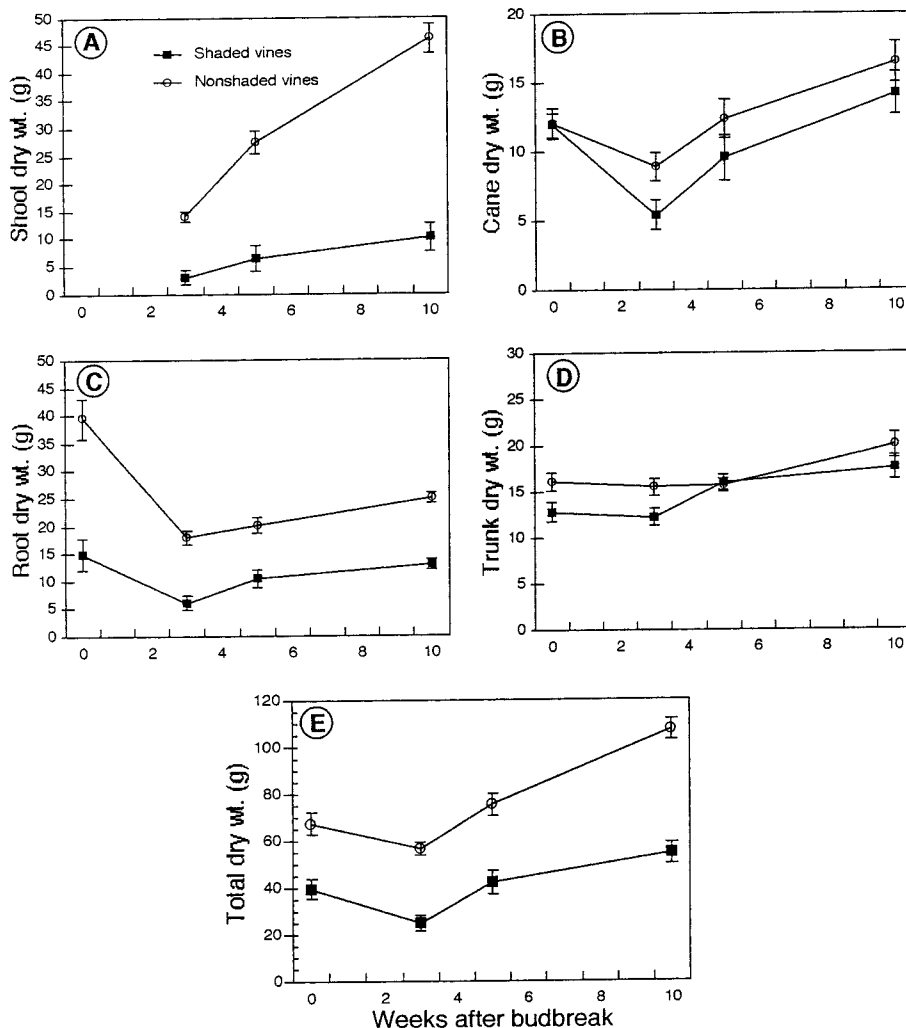
DRY MATTER PARTITIONING. Partitioning of dry matter between root, trunk, cane and shoot tissues is presented for 'Seyval' vines

4 and 10 weeks after budbreak (Fig. 1) and for 'DeChaunac' vines at budbreak and 3, 5, and 10 weeks later (Fig. 2). Shading reduced total plant dry weight of both cultivars. Four weeks after budbreak in the year following treatment dry weight of root ($P < 0.05$) and shoot ($P < 0.001$) tissues of shaded 'Seyval' vines was reduced compared to nonshaded vines (Fig. 1A). Dry weight of root ($P < 0.001$), trunk ($P < 0.05$) and shoot ($P < 0.05$) tissues was reduced 10 weeks after budbreak in the year following treatment in shaded compared to nonshaded vines (Fig. 1B). Dry weight of cane tissues on 'Seyval' vines was not affected by shading in the previous year. Total dry weight of 'DeChaunac' vines declined during the first 3 weeks of growth ($P = 0.06$) (Fig. 2). Root dry weight was significantly lower 3 weeks after budbreak compared to budbreak ($P < 0.001$), nonshaded and shaded vines losing 54% and 59% of their root dry weight, respectively. Root dry weight was significantly higher 10 weeks after budbreak compared to 3 weeks after budbreak ($P = 0.001$ and 0.07 for shaded and nonshaded vines, respectively). Dry weight of canes also declined on 'DeChaunac' vines during the first 3 weeks after budbreak ($P = 0.01$), with canes on vines that were shaded in the previous year losing 54% of their dry weight compared to a loss of only 26% on nonshaded vines. Cane dry matter was higher 10 weeks after budbreak compared to 3 weeks after budbreak ($P = 0.001$). Dry matter accumulation by new shoots occurred more rapidly

in nonshaded vines than vines that had been shaded in the previous year (Fig. 2).

Dry matter partitioning. The partitioning of dry matter between root, trunk, cane and shoot tissues is shown for 'Seyval' vines 4 and 10 weeks after budbreak (Fig. 1) and for 'DeChaunac' vines at budbreak and 3, 5, and 10 weeks later (Fig. 2). Shading reduced total plant dry weight of both cultivars. Four weeks after budbreak in the year following treatment dry weight of root ($P < 0.05$) and shoot ($P < 0.001$) tissues of 'Seyval' vines was reduced (Fig. 1A). The dry weight of root ($P < 0.001$), trunk ($P < 0.05$) and shoot ($P < 0.05$) tissues was reduced 10 weeks after budbreak in the year following treatment (Fig. 1B). The dry weight of cane tissues on 'Seyval' vines was not affected by shading in the previous year. Total dry weight of 'DeChaunac' vines declined during the first 3 weeks of growth (Fig. 2), although this effect was not statistically significant ($P = 0.056$). Root dry weight was significantly lower 3 weeks after budbreak compared to budbreak ($P = 0.0001$), unshaded and shaded vines losing 54% and 59% of their root dry weight

Fig. 2. Effect of shade in the previous year on dry matter partitioning of 'DeChaunac' grapevine tissue at budbreak (0), and 3, 5, 10 weeks later into the following dry weight components (A) shoot, (B) cane, (C) root, (D) trunk, (E) total. Each symbol is the mean of eight vines. Vertical bars represent ± 1 SE.



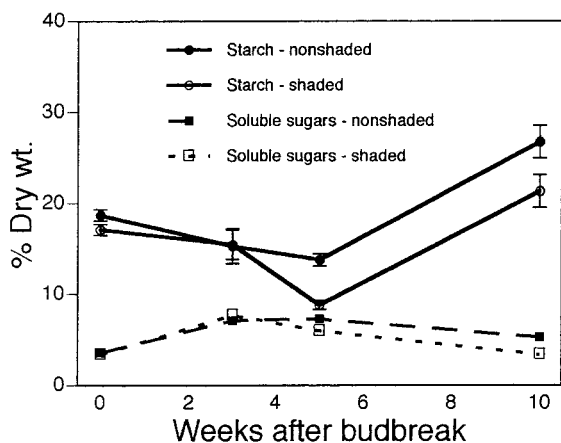


Fig. 3. Effects of shade in the previous year on starch and soluble sugars present in root tissues of 'DeChaunac' grapevines at budbreak and 3, 5 and 10 weeks later. Each symbol is the mean of eight vines. Vertical bars represent ± 1 SE. Error bars smaller than symbols have been omitted.

respectively. Root dry weight was significantly higher 10 weeks after budbreak compared to 3 weeks after budbreak ($P = 0.0007$, 0.0669 for shaded and unshaded vines respectively). The dry weight of canes also declined on 'DeChaunac' vines during the first 3 weeks after budbreak ($P = 0.01$), canes on vines that were shaded in the previous year losing 54% of their dry weight compared to a loss of only 26% on unshaded vines. Cane dry matter was higher 10 weeks after budbreak compared to 3 weeks after budbreak ($P = 0.001$). Dry matter accumulation by new shoots occurred more rapidly in unshaded vines than vines that had been shaded in the previous year (Fig. 2).

NONSTRUCTURAL CARBOHYDRATES IN ROOT TISSUES. The concentration of starch in root tissues of 'DeChaunac' vines at budbreak was 18% of the total dry weight for both nonshaded and shaded plants (Fig. 3). Root starch concentration declined during the first 5 weeks after budbreak and this decline was greater for shaded vines. Between 5 and 10 weeks after budbreak, root starch concentration increased, although 10 weeks after budbreak it was still lower in vines that had been shaded in the previous year compared to nonshaded vines. Soluble sugars increased in root tissues of both shaded and nonshaded vines to a maximum of 7.5% on a dry weight basis 3 weeks after budbreak and then declined until 10 weeks after budbreak (Fig. 3).

SOLUBLE CARBOHYDRATES IN XYLEM SAP. Mean rate of xylem sap flow over the 24 h period following cutting was 1.4 and 1.0 $\text{mL}\cdot\text{h}^{-1}$ for nonshaded and shaded vines, respectively (Table 1). Glucose was the predominant soluble carbohydrate present in xylem sap, accounting for 70% and 55% of the total soluble carbohydrates detected in sap from nonshaded and shaded vines, respectively. Fructose and myo-inositol were also present in xylem sap, along with trace amounts of sucrose. The concentrations of glucose, fructose, and myo-inositol were 5.9, 4.1, and 2.5 times higher, respectively, in xylem sap from nonshaded compared to shaded vines. Amino nitrogen was 1.8 times higher ($P < 0.05$) in xylem sap of nonshaded vines compared to shaded vines (Table 1).

LEAF GROWTH. Shoots that developed from apical buds were longer (data not presented) and had a greater leaf area than those growing from basal buds on canes. Basal shoots on 'Seyval' vines had a total final leaf area of 44 cm^2 , whereas shoots arising from mid and apical positions on the cane had 162 cm^2 and 276 cm^2

total final leaf area, respectively. Total final leaf area of 'DeChaunac' shoots occupying basal, mid, and apical positions were 176, 298, and 446 cm^2 , respectively. Since there was no interaction between shading treatment and shoot position for either cultivar, individual and total shoot leaf area data are presented for apical shoots only. There was a positive linear relationship between leaf area and the product of lamina length and width for each cultivar. 'DeChaunac' leaf area (cm^2) = $0.83 \times [\text{lamina length (cm)} \times \text{lamina width (cm)}]$, ($R^2 = 0.98$, $P < 0.001$); 'Seyval' leaf area = $0.86 \times [\text{lamina length (cm)} \times \text{lamina width (cm)}]$, ($R^2 = 0.99$, $P < 0.001$). Using the relationship for each cultivar, leaf area was estimated from the lamina dimensions measured on intact vines for each leaf on the three sample shoots per plant.

Total shoot leaf area was calculated as the sum of individual leaf areas for each shoot. A sigmoid growth curve of the form $y = a / (1 + e^{b-(c \cdot x)})$ was fitted to the data from each vine describing the increase in total leaf area per shoot with time, and each of the parameters in this model was tested for differences due to shading using analysis of variance procedures. Shoots on vines that were shaded in the previous year had decreased total leaf area (Fig. 4) as the result of fewer, smaller leaves (Fig. 5). Growth of the basal four leaves on 'Seyval' shoots was unaffected by shading in the previous year, whereas growth of the basal four leaves of shoots on 'DeChaunac' vines was inhibited by shading (Fig. 5). Analysis of the curves fitted to individual leaf growth data revealed that differences in final leaf area were explained by differences in the rate of expansion during the linear phase of leaf growth rather than by the duration of leaf area expansion (data not presented).

Discussion

Shading reduced total dry weight of grapevines at budbreak in the year after treatment by reducing dry weight of root and cane tissues. Shading reduced dry weight of roots of 'Seyval' vines by 47% and 'DeChaunac' vines by 51% when measured 4 and 5 weeks after budbreak. These data suggest that under shaded conditions fewer carbohydrates were available for formation of storage reserves in roots. Remobilization of carbohydrates from reserves in late winter and early spring provides one of the major substrates required for shoot respiration and growth of woody plants (Loescher et al., 1990). The importance of roots as a storage pool for reserves is illustrated in Fig. 2. Dry weight of 'DeChaunac' root tissues declined in the first 3 weeks after budbreak, as the glucose liberated from starch hydrolysis (Winkler and Williams, 1945) and glutamine synthesized from the nitrogen released after arginine catabolism (Roubelakis-Angelakis and Kliewer, 1992) were translocated to developing shoots in the xylem stream. Dry

Table 1. Effects of shade in the previous year on xylem sap flow rate, soluble carbohydrates, and amino nitrogen in sap of 'DeChaunac' grapevines at budbreak.

Xylem sap	Nonshaded ²	Shaded ²
Flow rate ($\text{mL}\cdot\text{h}^{-1}$)	1.4 ± 0.3	1.0 ± 0.1
Soluble carbohydrates (μM)		
Glucose	1079 ± 229	184 ± 49
Fructose	259 ± 90	64 ± 16
Sucrose	6 ± 2	9 ± 7
myo-Inositol	191 ± 26	77 ± 19
Amino nitrogen ($\mu\text{g}\cdot\text{mL}^{-1}$)	135 ± 16	74 ± 14

²n = 8 \pm SE.

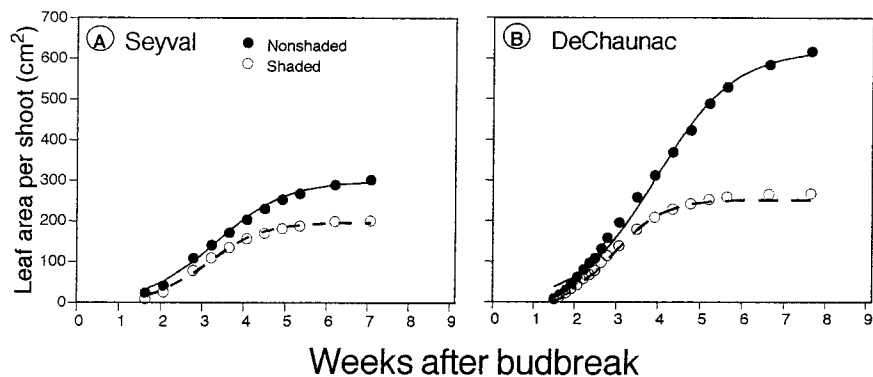


Fig. 4. Effects of shade in the previous year on total leaf area per shoot of (A) 'Seyval' and (B) 'DeChaunac' grapevines. Each line represents the mean of a single shoot on eight replicate vines. A sigmoid curve of the form $area = a / (1 + e^{-b \cdot (c - \text{days after budbreak})})$ was fitted to the data for each vine. 'Seyval': nonshaded, total leaf area per shoot (cm^2) = $299 / (1 + e^{-4.0 \cdot (-0.17 \cdot \text{days after budbreak})})$; shaded, total leaf area per shoot (cm^2) = $197 / (1 + e^{-4.8 \cdot (-0.22 \cdot \text{days after budbreak})})$; a, b, c, were significantly different at $P < 0.05$, 0.05 and 0.01, respectively. 'DeChaunac': nonshaded, total leaf area per shoot (cm^2) = $619 / (1 + e^{-4.3 \cdot (-0.16 \cdot \text{days after budbreak})})$; shaded, total leaf area per shoot (cm^2) = $252 / (1 + e^{-5.0 \cdot (-0.25 \cdot \text{days after budbreak})})$; a, b, c, were significantly different at $P < 0.001$, 0.05, or 0.001, respectively.

weight of roots on shaded and nonshaded 'DeChaunac' vines was reduced by 55% in the first 3 weeks after budbreak with only 20% of this reduction due to loss of starch. However, dry weight of root tissues began to increase again 3 weeks after budbreak, suggesting that translocation of current assimilates from shoot to root tissues in the phloem began as early as 3 weeks after budbreak. Even 10 weeks after budbreak, dry weight of root tissues had not recovered to their weight at budbreak. These trends in carbohydrate depletion and recovery in root tissues were evident in both shaded and nonshaded vines.

Dry weight of cane tissues showed a trend of initial decline and subsequent recovery similar to that observed in the root tissues, but the initial loss of dry weight from cane tissues was less than that in root tissues in absolute terms. These data suggest that while both cane and root tissue function as a pool of storage reserves, the root tissues are the larger pool from which reserves are drawn. Dry weight of 'DeChaunac' shoots increased more rapidly on nonshaded vines, possibly because of increased delivery of soluble carbohydrates and amino nitrogen in the xylem sap which resulted in faster rates of leaf growth. Subsequent accumulation of dry matter would be greater in nonshaded vines because of increased availability of current assimilates from the higher leaf area that developed compared to vines that were shaded in the previous year.

Starch concentrations in root tissues of both shaded and nonshaded vines declined for the first 5 weeks after budbreak, then increased. Root starch concentration at budbreak was 18% of the dry weight, declining to 9% and 14% 5 weeks later for shaded and nonshaded vines, respectively. During the same period, the concentration of soluble sugars in root tissues increased from 3.5% to 7% of the dry weight. The trend of initial depletion and subsequent recovery of carbohydrate reserves was demonstrated in grapevine roots by Winkler and Williams (1945). In this earlier study, root starch

concentration did not begin to recover until 3 months after budbreak, whereas vines in the present study began accumulating dry matter (Fig. 2) and starch (Fig. 3) 3 and 5 weeks after budbreak, respectively. The earlier accumulation of starch and dry matter by root tissues in the present study might have been due to the absence of any reproductive sinks on these vines.

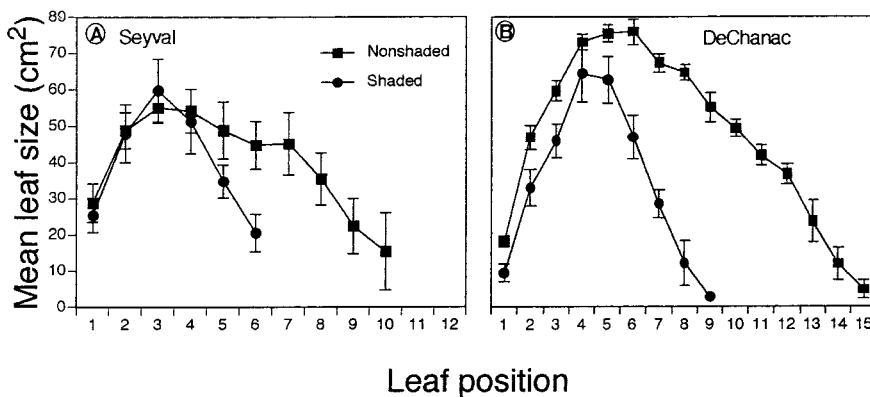
While root starch concentration continued to decline between 3 and 5 weeks after budbreak, total root dry weight actually increased over this period. These data suggest that soluble sugars translocated out of shoots and derived from current assimilates were being utilized for

new root growth rather than accumulating as starch. Five weeks after budbreak, however, the root starch concentration began to increase concomitantly with a continued increase in root dry weight, suggesting remobilization of carbohydrates from stored reserves had ceased. We believe developing shoots were photosynthetically autonomous 5 weeks after budbreak since depletion of starch in root tissues had ceased at this time. Furthermore, at this time the predominant direction of carbon flow through the roots had been reversed as the strategy of the vine changed from carbon remobilization to carbon accumulation.

Glucose was the predominant soluble carbohydrate detected in xylem sap. Other soluble carbohydrates detected, in decreasing concentrations, were fructose, myo-inositol, and sucrose. Sucrose and its reducing sugars have been reported in xylem sap from grapevines (Andersen et al., 1995; Andersen and Brodbeck, 1989b; Ohkawa, 1981; Stoev et al., 1959; Wormall, 1924). Myo-inositol has not been reported previously in xylem sap from grapevines, but in the present study accounted for 12% and 20% of the total soluble carbohydrates in sap from nonshaded and shaded 'DeChaunac' vines, respectively. Interestingly, myo-inositol was reported recently to provide a carbon source for induction of tracheary elements in lemon [*Citrus limonia* (L.) Burm.] fruit in preference over sucrose, D-glucose or glycerol (Kahn, 1995).

Concentrations of glucose and amino nitrogen were 6- and 2-fold higher, respectively, in xylem sap collected from vines that were nonshaded in the previous year compared to vines that had been shaded. More extensive starch hydrolysis in the larger root

Fig. 5. Effects of shading in the previous year on final leaf size at each position on the shoot of (A) 'Seyval' and (B) 'DeChaunac' grapevines. Symbols are the means of eight replications. Vertical bars represent ± 1 SE.



systems of nonshaded vines would result in higher concentrations of glucose in the xylem sap, higher positive root pressures, and faster rates of exudation. The ninhydrin reagent used to measure amino nitrogen in the present study does not detect nitrate nitrogen. However, nitrate nitrogen accounts for only 5% of the total nitrogen in xylem sap while glutamine is the major transport form of nitrogen in the xylem sap collected from grapevines (Roubelakis-Angelakis and Kliewer, 1992). Increased concentrations of soluble carbohydrates and nitrogen in xylem sap at budbreak would presumably result in greater availability of carbohydrates and nitrogen for new tissue formation and should lead to faster rates of vegetative development.

Shading in the previous year inhibited leaf area development of both cultivars, although this effect was more pronounced on 'DeChaunac' vines. Expansion of the first four leaves on shoots of 'Seyval' vines was unaffected by shading in the previous season. However, expansion of more apical leaves on these shoots was inhibited. Expansion of all leaves on 'DeChaunac' shoots was inhibited on vines that were shaded in the previous year. Slower rates of leaf area expansion may have been due to limited delivery of growth factors such as nitrogen (Roggatz et al., 1999) to the expanding tissues in the year after treatment, since we also measured a reduced concentration of amino nitrogen in bleeding sap. However, final leaf size has also been correlated with the number of cells present at the onset of unfolding (Milthorpe and Newton, 1963). Since we did not measure the number of cells within the leaf primordia at budbreak we cannot discount the possibility of a direct effect of light on differentiation of leaf primordia in the year of treatment. The effects of shade in the previous year on leaf area expansion were strikingly similar to the effects of imposing a low *PPF* during the expansion phase itself (Smith and Longstreth, 1994). Analysis of xylem sap collected from 'DeChaunac' vines at budbreak suggest this reduction in vegetative development might be related to the supply of soluble sugars and/or nitrogen from the roots, although the supply of other factors, such as hormones may also be responsible.

This study demonstrated that shading during a growing season reduced vegetative development in the year after treatment. Our study was extreme in reducing light levels, while shading reduced light levels to 20% of the level of ambient light over the whole season, reductions of light to only 5% of ambient levels have been measured in significant portions of the canopy interior of grapevines (Smart, 1974; 1985, 1987). Reduced delivery of soluble sugars and nitrogen to developing tissues in the spring are likely factors which mediate this response, although a direct effect of reduced light on differentiation of leaf primordia in the year of treatment may also have occurred. The importance of roots as a reserve tissue was demonstrated by the extent that root dry weight decreased in the first 3 weeks after budbreak. The root tissues of both shaded and nonshaded vines began to accumulate dry matter 3 weeks after budbreak, suggesting newly assimilated carbon was being translocated from shoots to the roots at this time. Despite this downward movement of carbon, dry weight of root tissues had not recovered even after 10 weeks of shoot growth. A reduction in the rate of expansion of individual leaves prior to flowering can be expected to reduce net carbon assimilation, and therefore limit the productivity of the vine, by reducing either fruit set or early fruit growth. Further work is needed to establish if early season vegetative development is limited by the delivery of growth regulators under stress conditions, and to determine the length of time after budbreak that individual shoots remain

dependent upon the delivery of reserves from storage tissues in perennial plants.

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