Whole plant- versus leaf-level regulation of photosynthetic responses after partial defoliation in *Eucalyptus globulus* saplings

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Supplementary Material

Method S1. All sugar standards were purchased from Sigma Aldrich, St. Louis, MO, USA. The identification of 8 soluble sugars - fructose, galactinol, gentiobiose, glucose, myo-inositol, raffinose, stachyose and sucrose was achieved by comparison of retention times, MS and MS/MS data with those obtained for each respective standard. No xylose, pinitol or quercitol was detected. All inositols are reported here as a group. The instrument was a Waters Acquity H-series UPLC coupled to a Waters Xevo triple quadrupole mass spectrometer. A Waters Acquity UPLC BEH amide column (2.1 x 50mm x 1.7 micron particles) was used. The solvents were 0.4% ammonia in water (Solvent A) and acetonitrile (Solvent B). Two μL of each sample was injected. The UPLC program was 20% A:80 % B to 45% A:55% B at 6 minutes, and this was followed by immediate re-equilibration to starting conditions for 3 minutes. The flow rate was 0.35mL/min, the column was held at 50 °C and the sample compartment was at 7 °C. Retention times under these conditions were: xylose, 0.83 min; fructose, 0.98 min; pinitol, 1.00 min; quercitol, 1.10 min; glucose, 1.20 min; sucrose, 1.90 min; myo-inositol, 2.21 min; trehalose, 2.53 min; gentiobiose, 2.76 min; raffinose, 3.44 min; galactinol, 3.72 min and stachyose, 4.69 min. The mass spectrometer was operated in negative ion electrospray mode with a needle voltage of 2.3 kV, and selected ion monitoring (SIM) was used to detect all analytes. The ion source temperature was 130°C, the desolvation gas was nitrogen at 950 L/hr, the cone gas flow was 50 L/hr and the desolvation temperature was 450°C. Data was processed using MassLynx software. SIM ions and cone voltages were as follows; xylose *m/z* 149.1, 14V; quercitol *m/z* 163.1, 15V; fructose, glucose and myo-inositol *m/z* 179.1, 17V; pinitol, *m/z* 193.1, 22V; sucrose, trehalose, gentiobiose and galactinol *m/z* 341.1, 24V; raffinose *m/z* 503.2, 30V; stachyose *m/z* 665.3, 40V. Dwell time was 50ms per channel.

The quantification of the positively identified sugars was based on a five-point standard curve over the range 0 to 500 ppm generated for each sugar using trehalose as the internal standard, and the amount was expressed as mg/g dry matter (DM). The relative quantification of minor unidentified sugars was based on the closest related response factor.

The minor monosaccharide was based on the response factor of fructose, the partially co-eluting inositol was combined with myo-inositol and the same response factor was used. Correlation coefficients ranged from 0.997 for myoinositol to 0.999 for fructose and glucose. A 50 ppm quality control sample was run after every 10 samples. Nine individual carbohydrates were detected in *E. globulus* foliage and they were (in decreasing order of abundance): fructose (39.8%), glucose (35%), the inositol group (9.8%), galactinol (4.6), gentiobiose (4), raffinose (3.2%), sucrose (2%), unknown monosaccharide (1.5 %) and stachyose (0.5%).

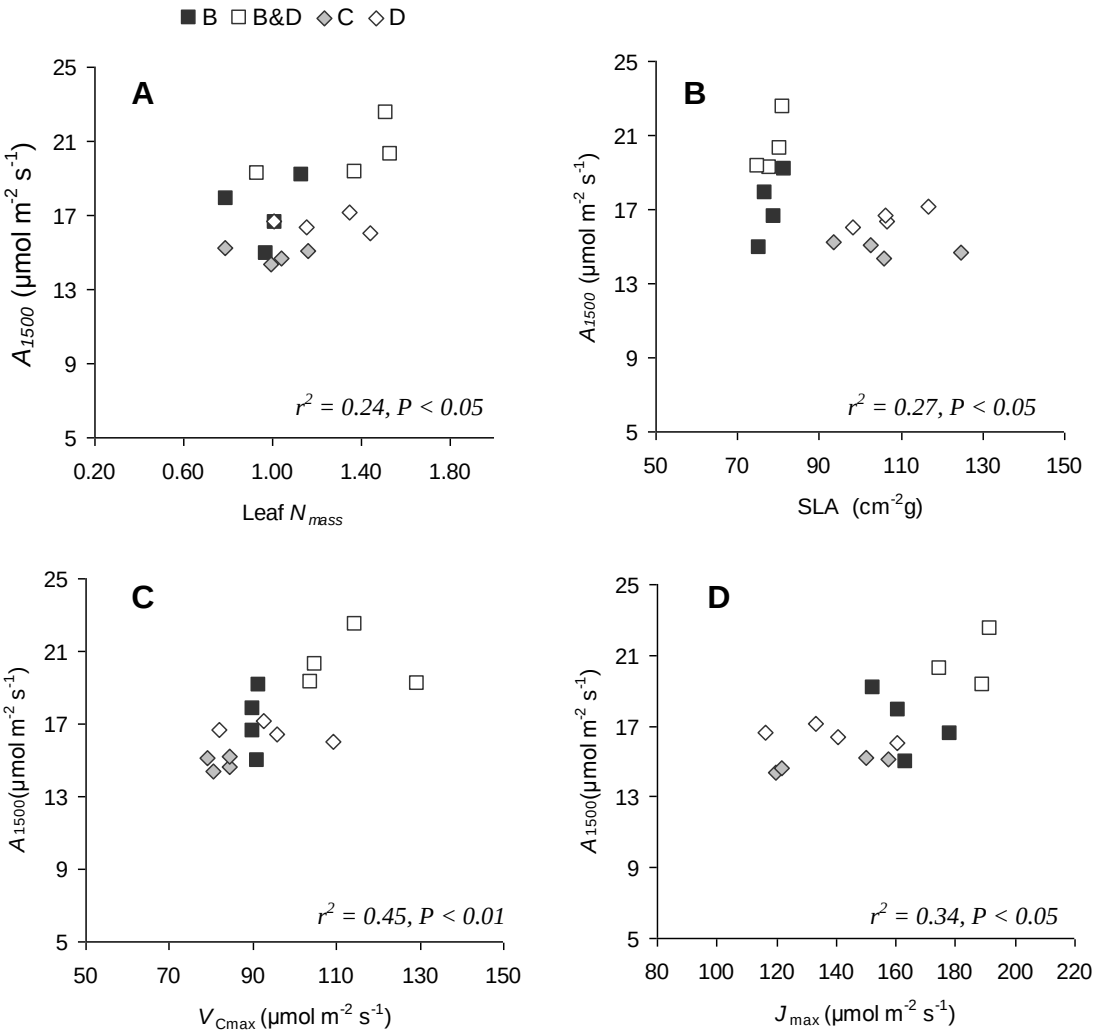

Fig. S1. Relationship between A_{1500} (measured directly) with leaf-level N_{mass} (A), SLA (B), V_{Cmax} (C) and J_{max} (D) of *Eucalyptus globulus* following treatments (B = bud damage; B&D = bud damage and defoliation; C = control; D = 50% defoliation) in week 5. The significant regressions shown are described by the following equations: (a) $A_{1500} = 5.2_{Leaf\ N} + 11.3$, (b) $A_{1500} = -0.08_{SLA} + 24.7$ (c) $A_{1500} = -0.12_{Vcmax} + 6.13$ and (d) $A_{1500} = 0.046_{Jmax} + 9.96$.