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Evaluating the potential of a novel dual heat-pulse sensor to measure volumetric water use in grapevines under a range of flow conditions

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Abstract. The aim of this study was to validate a novel, dual sap-flow sensor that combines two heat-pulse techniques in a single set of sensor probes to measure volumetric water use over the full range of sap flows found in grapevines. The heat ratio method (HRM), which works well at measuring low and reverse flows, was combined with the compensation heat-pulse method (CHPM) that captures moderate to high flows. Sap-flow measurements were performed on *Vitis vinifera* L. (cvv. Thompson seedless, Chardonnay and Cabernet Sauvignon) grapevines growing in a greenhouse and in three different vineyards, one of which contained a field weighing lysimeter. The combined heat-pulse techniques closely tracked diurnal grapevine water use determined through lysimetry in two growing seasons, and this was true even at very high flow rates (>6 L vine^{-1} h^{-1} for Thompson seedless vines in the weighing lysimeter). Measurements made with the HRM technique under low flow conditions were highly correlated (R^2 ~ 0.90) with those calculated using the compensated average gradient method that is used to resolve low flow with the CHPM method. Volumetric water use determined with the dual heat-pulse sensors was highly correlated with hourly lysimeter water use in both years ($R^2 = 0.92$ and 0.94 in 2008 and 2009 respectively), but the nature of the relationship was inconsistent among replicate sensors. Similar results were obtained when comparing grapevine water use determined from sap-flow sensors to miniaturised weighing lysimetry of 2-year-old potted vines and to meteorological estimates for field-grown vines in two additional vineyards. The robust nature of all of the correlations demonstrates that the dual heat-pulse sensors can be used to effectively track relative changes in plant water use, but variability of flow around stems makes it difficult to accurately convert to sap-flow volumes.

Additional keywords: heat pulse velocity, sap flow, *Vitis vinifera*, weighing lysimetry.

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Introduction

Quantification of crop water use can be done using a variety of techniques that vary in precision, reliability and cost. Weighing lysimetry is considered a gold standard for crop evapotranspiration (ET_c) measurements, providing excellent accuracy and sensitivity to measure *ET* directly. A large weighing lysimeter has been used to measure water use of the Thompson seedless grapevines (*Vitis vinifera* L.) during vineyard establishment and at maturity (Williams *et al.* 2003*a*, 2003*b*) and for estimates of water use for grape production throughout California growing regions (Williams and Ayars 2005*a*). In this latter study, it was demonstrated that grapevine water use and the crop coefficient (K_c) were linear functions of the percent shaded area on the vineyard floor measured throughout the growing season. This method of deriving K_c values has since been used in studies to determine the effects of applied water amounts, at various fractions of estimated ET_c, on productivity and fruit quality of raisin, table and wine grapes at numerous locations throughout California (Williams 2010; Williams *et al.* 2010).

Although weighing lysimeters provide a direct measure of grapevine water use, they are expensive to build and much time is needed to ensure measurement accuracy. Sap-flow sensors provide an alternative and portable means to measure water use of many vines at one time and could be useful in viticulture (Lascano *et al.* 1992; Ginestar *et al.* 1998*a*, 1998*b*; Braun and Schmid 1999). However, the reliability of sap-flow sensors, especially on large vines, has been questioned (Tarara and

Ferguson 2001). This is illustrated in work by Ginestar *et al*. (1998*a*, 1998*b*), Lu *et al.* (2003), Patakas *et al*. (2005) and Yunusa *et al*. (2000) in which the diurnal time course of transpiration for well irrigated vines was generally maximised in the late morning, remained constant until late in the afternoon, and then decreased. This is in contrast to data obtained in the weighing lysimeter in which it was shown that the diurnal course of ET of well watered vines continues to increase from sunrise until solar noon and then decreases as the sun goes down in the afternoon (Williams *et al*. 2003*b*; Williams and Ayars 2005*a*). These previous results suggest that the flow rates of these vines were so great after mid-morning that the sensors were unable to accurately measure the flux of sap above a certain value. Preliminary measurements of sap flow were conducted recently on large vines growing at the Kearney Agricultural Center using commercially available Granier-type sap-flow sensors (M Fidelibus and L Williams, unpubl. data). These sensors were unable to resolve the high rates of midday sap-flows measured by the lysimeter and exhibited plateaus similar to those found in the other studies mentioned above.

The aim of this study was to determine the potential of measuring water use of grapevines using a novel dual heat-pulse sap-flow sensor that combines two heat-pulse techniques to measure volumetric water use over the full range of sap flows found in grapevines. The heat ratio method (HRM; Burgess *et al*. 2000, 2001), which works well at measuring low and reverse flows was combined in a single set of sensor probes with the compensation heat-pulse method (CHPM; Green and Clothier 1988) used to capture moderate to high flows. A recent review of sap-flow methods concluded that heat-pulse methods have a strong theoretical basis and they are likely to outperform empirical continuous heating methods (such as the thermal dissipation method) as they do not require specific calibrations and are less affected by natural temperature gradients (Vandegehuchte and Steppe 2013). However, heat-pulse methods remain to be validated over the complete range of flows found in field grown grapevines that have been reported to be as high as 60 L day^{-1} and in excess of 8 L vine^{-1} h^{-1} (Williams *et al.* 2003*a*, 2003*b*; Williams and Ayars 2005*b*; Williams and Baeza 2007). Water use measured with this dual sensor was compared against actual water use of mature Thompson seedless grapevines growing in the Kearney field station's grape weighing lysimeter and small Merlot grapevines growing in pots. In addition, water use measured with the dual heat-pulse sensor was compared with estimates of water use based on shaded area (Williams and Ayars 2005*b*) under mature Cabernet Sauvignon and Chardonnay grapevines grown in Napa Valley and Davis, CA respectively.

Materials and methods

Sap-flow sensor specifications and construction

Sap flow was measured with both commercial sensors (East 30 Sensors Inc., Pullman, WA, USA) and with sensors built in our laboratory. Commercial sensors consisted of 35 mm long, 1.2 mm diameter needles housing either Type E point thermocouples or a 36 Ω nichrome wire heater. Thermocouple needles each contained two measurement junctions, located 10 mm apart. Our in-house sensors consisted of three, 20 mm long, 1.2 mm diameter needles, each with one Type E point thermocouple and a fourth needle containing a coiled 18 Ω nichrome wire heater. Homemade sensor needles were formed by cutting off the tip of a 35 mm long hypodermic needle (18G1.5 short bevel, Becton Dickinson and Co., Franklin Lakes, NJ, USA) forming a 20 mm tube. The cut end of the tube was then sealed with solder. Either a thermocouple or a nichrome heater was then inserted into the tube, and sealed in place with an epoxy resin. Heaters were formed by coiling a length of nichrome wire around itself. The exposed ends of the heater wire were connected via extension wire, through a control switch, to a 12 V battery.

Commercial and in-house sensors were installed in vines such that thermocouple needles were placed 6 mm upstream (–) and 6 and 18 mm downstream (+) from the heater. The thermocouples located –6 mm upstream and +6 mm downstream from the heater were used for HRM measurements (see details in work by Burgess *et al*. 2001), whereas thermocouples –6 mm upstream and +18 mm downstream were used for the compensation heat-pulse method (CHPM) measurements.

We also compared HRM measurements under low flow conditions to those derived from the calibrated average gradient (CAG) procedure. This procedure, developed recently by Testi and Villalobos (2009), consists of averaging the DT measurements from the –6 and +18 mm thermocouples after the heat pulse, then obtaining the heat-pulse velocity function in its linear domain where it is still measurable with the traditional CHPM. We then used this function to calculate velocities below the minimum measurable by the CHPM. See details in work by Testi and Villalobos (2009).

A dremel tool (Robert Bosch Tool Corporation, Dremel, Racine, WI, USA) with a 1.25 mm diameter drill bit was used at low speed to drill into vines for needle installation. Consistent needle spacing's were ensured by using a drill guide. Sensor needles were coated with grafting wax (East 30 Sensors Inc.) as they were inserted into drill holes in order to improve thermal contact and to prevent fungal growth.

The heat-pulse sensors were wired with extension cable to AM25T multiplexers controlled by CR10X dataloggers (Campbell Scientific Inc., Logan, UT, USA), and measurements were taken every 30 min. Heaters were activated from a 12 V power supply for 2–6 s, depending on the sensor, in order to deliver similar strength heat pulses for the commercially manufactured and in-house sensors.

Field weighing lysimeter calibration

The weighing lysimeter was located at the University of California, Kearney Agricultural Research and Extension (KARE) Center near Parlier, California (36°48′N, latitude 119°30′W, longitude). A detailed description of the lysimeter can be found in work by Williams *et al*. (2003*a*). Briefly, two *Vitis vinifera* L. cv. Thompson seedless (clone 2A) grapevines were planted in 1987 in the 2 × 4 × 2 m deep lysimeter. The vines now have trunk diameters of ~10 cm. The lysimeter is located inside a 1.4 ha vineyard, with vine and row spacing of 2.15 and 3.51 m, respectively, oriented east to west. An overhead trellis was used with the foliage support wires located at a height of 1.83 m above the soil surface. The supports extended

to midway between rows. The trellis for the vines in the lysimeter was self-contained within the lysimeter to ensure it was part of the lysimeter mass. Two vines to the east and to the west of the lysimeter were also trained to the overhead trellis, and four vines in the rows directly to the north and to the south of the lysimeter were also trained in a similar manner. Vines were irrigated at 100% of ET_c with water being replaced whenever 16 L were lost from the lysimeter (8 L vine^{-1}). Two layers of 30-gauge, clear PVC film (Goss Plastic Film Corp., Los Angeles, CA, USA) were placed on the soil surface of the lysimeter at various times during the two growing seasons to minimise soil evaporation. To reflect the addition of the plastic covering to the lysimeter's soils surface, water use of the vines in the lysimeter was referred to as ET_{Lys} in comparisons with grapevine water use estimated with sap-flow sensors. A datalogger (21X Micrologger, Campbell Scientific) recorded the weight of the lysimeter every minute and averaged these values hourly.

Two sap-flow sensors were installed in each of the lysimeter vines in 2008. Three sensors were installed in each of the lysimeter vines in 2009 to better account for variability around the trunk's circumference. Sensors were positioned around the circumference of each lysimeter vine's trunk as close as possible to 180° apart in 2008. In 2009, after extensive dye infiltration experiments were performed on numerous adjacent vines we realised that the grapevine trunks were hydraulically sectored (i.e. packets of the trunk xylem were functionally connected to portions of the canopy) and that certain sections exhibited much higher flows – thus we strategically placed the sensors around the trunks to target primary sectors connected to extensive leaf area and to ensure adequate coverage around the trunk. Sensors were removed after the 2008 season and reinstalled in a different location in 2009. Sensors and grapevine trunks were covered with reflective shielding down to the soil surface to insulate against large temperature fluctuations due to solar radiation on the sensor.

The canopies of both lysimeter vines were covered with 95% shade cloth at several intervals throughout the day on 17 and 18 September, 2008. The shade cloth was supported by a frame mounted on the trellis of vines located to the east and west of the lysimeter such that no weight was added to the lysimeter. Photon flux density (PFD) was measured with a Sunfleck Ceptometer (Decagon, Pullman, WA, USA) above and beneath the shade cloth.

The response of heat-pulse velocity to water availability was investigated by terminating irrigation from 21 July until 9 August 2008 to Thompson seedless grapevines growing to the west of the lysimeter (in the same row). Two of the vines were trained to the overhead trellis and the other two to a 1.2 m 'T' trellis (cross arm was 0.6 m in width). Irrigation had been withheld before the start of heat-pulse measurements (11 July) and resumed on 6 August. These vines were instrumented with sap-flow sensors using methods described above.

Greenhouse mini-lysimeter experiment

Seven greenhouse grown, 2-year-old *V. vinifera* (cv. Merlot) vines grown in pots were used to compare water use measured with sap flow and by weight loss of the potted vines. Our in-house heat-pulse sensors were installed in these vines as described above and water use by weight loss was determined by tracking changes in the weight of the pots with an electronic balance (Ohaus CD-11, resolution of 1 : 5000 LFT, 1 : 20 000 non-LFT, Ohaus Corporation, Florham Park, NJ, USA). The balance was connected to a CR-1000 datalogger (Campbell Scientific Inc.), and its output was recorded once every minute and changes in weight over a 15 min interval were tallied to coincide with each heat-pulse measurement. The duration of data collection ranged from 5–49 days among the seven replicates, and vine water use was modified by supplying water or allowing the pot to dry down. Each pot was sealed in a plastic bag to prevent water loss from the soil surface.

Estimation of active sapwood areas

To limit damage to the two Thompson seedless vines growing in the field lysimeter, the active sapwood area of these vines was inferred from dye infiltrations performed on similar grapevines grown elsewhere in the same vineyard using a procedure similar to that by Sano *et al*. (2005). Briefly, a plastic collar was wrapped around the trunk of each vine and sealed at the bottom with putty. The collars were filled with a xylem mobile dye (0.1% solution of acid fuchsin) and a wood chisel was used to make incisions below the level of the dye to prevent embolisms. Chisel incisions were made on three sides of the vine at ~35° relative to the trunk axis and were made deep enough to be at least 40 mm below the trunk's surface. It was previously found that three chisel incisions were sufficient to stain active tissue around the entire circumference of the vine. The collars were left on the vine for ~90 min, at which time the majority of the canopy was noticeably stained. During this period, the collars were monitored and refilled with dye as needed to prevent the trunks' tissue being exposed to air. The trunks were then cut at various trunk heights parallel to the soil surface, and the exposed trunk cross-sections were photographed. These images were processed using the Leica Application Suite (Leica Microsystems GmbH, Wetzlar, Germany) to determine the maximum and minimum stem diameter at the cut, the percent stained area and the depth of staining. Correlations between these parameters were determined to estimate the active cross-sectional area of sap flow of the lysimeter vines, at each sensor, from measurements of minimum and maximum vine diameter made at each sensor location. Vines subjected to dye infiltration had an average stained depth of 6.7 mm below the cambium. The stained depth was variable around the trunk circumference and was often deeper in 3–4 cm wide packets of tissue. These packets noticeably bulged from the vine trunks and could often be traced to individual arms in study vines.

The cross-sectional areas of active sapwood of the vines in the greenhouse experiment were determined similarly following water use measurements. The base of each vine stem was submerged in a 0.1% solution of acid fuchsin and cut 10–15 cm below the site of the heat-pulse sensor. Vines remained in the solution for an hour, after which most leaves were stained. The stem was then cut, and digital images were taken of the cut surface to quantify the area stained. Wounding sizes around each sensor needle were also evaluated from the dye infiltrations, as described below.

Water use at Oakville and Davis, CA

Ten sensors were installed in five *V. vinifera* (cv. Cabernet Sauvignon) vines (two sensors per vine) at the UC Davis, Department of Viticulture and Enology's Oakville field station (38°26′N, latitude, 122°25′W, longitude) located in Napa Valley. The vines were trained to a vertical shoot positioning (VSP) trellis. Vine and row spacing were 2.0 and 2.5 m, respectively, with rows oriented NW to SE. Ten sensors were installed in five *V. vinifera* (cv. Chardonnay) vines at the UC Davis, Department of Viticulture and Enology's experimental vineyard (38°32′N, latitude, 121°47′W, longitude) with two sensors in each of three vines and one sensor in each remaining vine. The vines were trained to a 'T' trellis (crossarm width 0.6 m). Vine and row spacings were 2.44 and 3.66 m, respectively, with rows oriented east to west. Water use was determined every half hour with the sap-flow sensors. Sap-flow measurements were averaged hourly and compared with hourly estimates of vineyard ET_c using the following equation:

$$ET_c = ET_o \times K_c. \quad (1)$$

Reference ET was obtained from California Irrigation Management Information System (CIMIS) weather stations at the Oakville field station and 3 km from the vineyard in Davis. Shaded area beneath the vines was determined by placing a grid beneath the vines at solar noon. The percentage of shade within each square of the grid was estimated in increments of 10%. The K_c at each site was estimated by converting the percent shaded area ((shaded area/area per vine) × 100) using the equation from Williams and Ayars (2005*b*).

Wounding corrections and conversion of heat-pulse velocity to sap velocity

Corrections are required to adjust recorded sap flows for deviations from the idealised heat-pulse equation caused by interruption of the sap stream due to sensor installation. Heat-pulse velocities obtained from the HRM were corrected for this wounding effect using the equation developed by Burgess *et al*. (2001) for a sensor with thermocouple needles spaced 6 mm from the heater. Wounding corrections for the CHPM were derived using a Fortran model provided by S Green and modified for our specific sensor installations as described by Green and Clothier (1988) and Green *et al*. (2003). The width of xylem inactivated by sensor installation and the resultant wounding response was determined from the dye infiltrations as described above and by performing this on six additional Thompson seedless vines in the same vineyard as the weighing lysimeter at the KARE Center. The sensors had been installed and operating for several weeks. After dye infiltration, the bark and cambium around the drill holes of a sensor were removed with a razor blade. The width of unstained tissue around the drill holes was measured to determine the wounding size. The average wounding width of these sensors (2 mm) was then used to correct heat-pulse velocities made in the lysimeter as well as for vines at Oakville and Davis. The wounding size of each sensor in the greenhouse grown vines was determined from the unstained width around the sensor needles following a measurement cycle after dye infiltration. Spacing errors were minimised using a drill guide and assessed using data collected when it was assumed that flow was at or very near to zero (at night, under cool, humid, windless conditions) as in Burgess *et al*. (2001).

Heat-pulse velocities (cm h^{-1}) obtained from the HRM and CHPM and corrected for wounding effects were converted into sap velocities (cm h^{-1}; numerically equivalent to sap-flux density, $cm^3\ cm^{-2}\ h^{-1}$) using measurements of wood properties as described by Burgess *et al*. (2001) and adjusted for variable moisture content using the methods by Vandegehuchte and Steppe (2012). The wood properties used for this conversion, namely density of dry and fresh tissue and moisture content, were obtained from cores in the weighing lysimeter vineyard on 8 and 10 August, 2009. Cores were taken to a depth of 1 cm below the cambium. The average moisture content was 0.855 (kg water per kg wood), the density of dry tissue or basic density was 582.4 kg m^{-3} and the density of fresh tissue or green density was 1079 kg m^{-3}. Values in this range were previously shown to have negligible effects on the sap-flux density (Vandegehuchte and Steppe 2012), which was consistent with our findings.

Measurements of vine water status

Midday leaf water potential (Ψ_l) was measured on various dates throughout the experiments with a pressure chamber (PMS Instrument Co., Corvallis, OR, USA) using two fully expanded, sun exposed leaves per vine. Midday leaf water potential was determined following a procedure similar to that described by Williams and Araujo (2002).

Data analyses

Relationships between actual water use determined with the weighing lysimeter for the greenhouse-grown vines and estimated water use for vines at Davis and Oakville were determined using linear regression. Coefficients of determination were calculated for each relationship.

Results

Patterns of heat-pulse velocity and lysimeter water use

Heat-pulse velocity tracked the daily water use patterns of grapevines grown in the weighing lysimeter when using both the HRM and CHPM sensors (Fig. 1). Heat-pulse velocities measured by the HRM tracked ET_{Lys} well in the morning and at night, but, as ET_{Lys} increased, the HRM readings reached a plateau at velocities ranging from 15–35 cm h^{-1}. The CHPM resolved heat-pulse values >30 cm h^{-1} and mimicked the diurnal pattern of ET_{Lys} across this range. There appeared to be no upper limit to the CHPM, even at very high flow rates (Fig. 1). The slight decrease in maximum ET_{Lys} over the 3 days (Fig. 1) was mimicked in relative magnitude by a decrease in heat-pulse velocity from the CHPM sensor. Combined output from the HRM and CHPM sensors produced a very strong correlation between the daily course of heat-pulse velocity and ET_{Lys} ($R^2 = 0.97$ on these dates). The HRM sensors also effectively tracked low flows that occurred on occasion at night and are likely associated with capacitance refilling or night-time transpiration. For most sensors, data output from the two methods required no manipulation to merge (Figs 1, 2); the plateaued portion of the HRM diurnal curve could be simply removed, and the low flows measured with HRM in

the morning and at night lined up well with higher values obtained with CHPM. When a data gap did exist between the two methods we extrapolated by averaging values at either end of the data gap.

Irrigation was terminated on 11 July, 2008 to the vines in the row growing outside the weighing lysimeter. Midday Ψ_l on this date was –0.84 MPa and it decreased to –1.14 MPa on 22 July and down to –1.34 MPa on 5 August. Daily maximum values of heat-pulse velocity decreased 60% from 21 July until 6 August, 2008 (Fig. 2). A total of 55 L of water were applied to the vines on 5 August between 1400 and 1700 hours. Midday heat-pulse velocities rapidly increased to values similar to those before when irrigation was terminated. During this period, the portion of daily heat-pulse velocity measured by the HRM (and not discernable by the CHPM) increased from 6 to 21% from 21 July to 6 August.

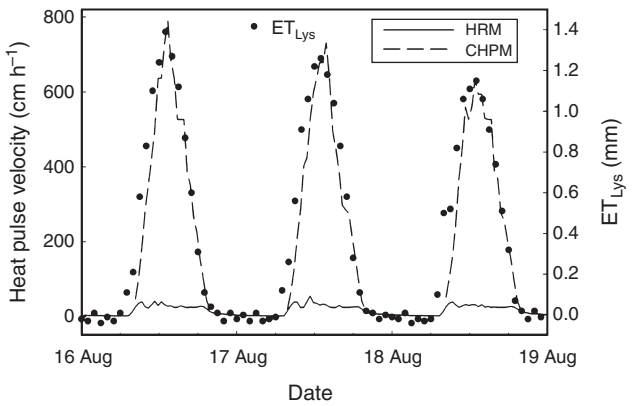

Fig. 1. Diurnal heat-pulse velocities determined from the heat ratio method (HRM) or compensation heat-pulse method (CHPM) sensors compared with hourly water use for grapevines growing in a weighing lysimeter (ET_{Lys}) in 2009. The soil surface of the lysimeter was covered with plastic during the time of the measurements. Daily lysimeter water use was 38.6, 37.3, and 35.0 L vine^{-1} on 16, 17, and 18 August respectively. Hourly values of ET_{Lys} in mm multiplied by four is equivalent to L vine^{-1} h^{-1}.

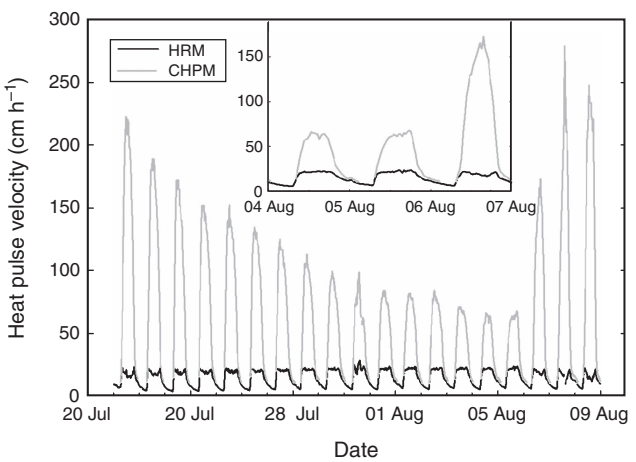

Fig. 2. Corrected heat-pulse velocity measured on vines growing outside the lysimeter in response to the termination of irrigation on 11 July, 2008. Water was withheld until 5 August when irrigation resumed at 1400 hours for 3 h and then irrigated normally after that day. Daily average vapour pressure deficit (VPD) remained the same throughout this period.

Comparison of low flow determined by CAG versus HRM

HRM measurements under conditions of low flow were highly correlated and near unity with velocities derived using the CAG procedure for CHPM (Fig. 3). Three of the five sensors evaluated for this analysis had slopes for the correlation of CAG and HRM very close to 1.0 with $R^2 \geq 0.9$ (Fig. 3). The slope of this relationship for the other two sensors were 0.82 and 0.72 (both with $R^2 > 0.91$), where the CAG method exhibited slightly lower values than the HRM.

Comparison of vine volumetric water use measured by the weighing lysimeter and sap-flow sensors

Substantial variation in sap velocity was observed among sensors within a vine, between vines, and across years for sensors in the weighing lysimeter vines (Table 1). The majority of sensor values in 2009 were much greater than those in 2008 despite a similar range of ET_{Lys}. Sensors in 2009 also had a larger spread in values than in 2008 (Table 1). Despite differences in regression equations, the volumetric sap flow predicted from each sensor (aside from sensor 8, which was apparently positioned in necrotic trunk tissue) was strongly related to lysimeter water use.

Greenhouse study

Volumetric sap flow of potted Merlot vines in the greenhouse study was highly correlated with water use measured with the balance ($R^2 > 0.94$) for all seven of the vine replicates (Table 2). While the intercepts of each regression were close to zero, the slope of the regressions varied from 0.62 to 1.27, with an average of 0.88 (Table 2).

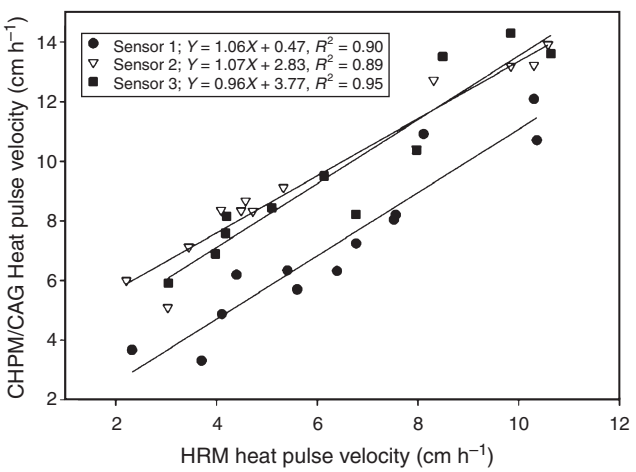

Fig. 3. Comparison of heat-pulse velocity measurements measured with the heat ratio method (HRM) and estimated using the calibrated average gradient method (CAG). Data from three sensors are shown along with the corresponding regression information for each of the three sensors. An additional three sensors were evaluated and showed similar patterns to those presented here.

Table 1. Linear regression parameters for the relationship between water use measured on vines growing in a weighing lysimeter (ET_{Lys}) and the dual heat-pulse sensors (ET_{Lys} (*x*-axis) *vs* volumetric sap flow (*y*-axis), both in L h^{-1})

Sap-flow sensors 1–4 were used for measurements in 2008, while sensors 5–10 were used in 2009

Sensor	Slope	Intercept	R^2
1	0.43	0.18	0.86
2	0.63	0.31	0.92
3	0.36	0.20	0.96
4	0.39	0.25	0.95
5	0.83	–0.02	0.92
6	2.00	–0.42	0.94
7	2.21	–0.16	0.97
8	0.02	0.05	0.30
9	1.90	–0.44	0.96
10	3.30	–1.05	0.89
Mean	1.19	–0.10	0.86

Table 2. Linear regression parameters for the relationship between water use measured with a digital balance (*x*-axis) and the dual heat-pulse sensors (*y*-axis) for potted greenhouse grown grapevines

Sap flow measured by dual heat-pulse sensors was recorded every 30 min. Weight loss measured by the balance was recorded every minute, and water use for comparison with sap flow was determined from the sum of weight changes measured in a 15 min interval centred on each sap-flow measurement

Sensor	Slope	Intercept	R^2
1	0.88	0.0001	0.95
2	1.27	0.0002	0.99
3	0.84	0.0017	0.97
4	0.96	–0.0008	0.98
5	0.63	–0.0010	0.95
6	0.62	0.0010	0.94
7	0.94	–0.0023	0.95
Mean	0.88	–0.0004	0.96

Comparison of sensor water use and estimated ET_c *at two diverse locations*

Estimated ET_c and water use determined from sap-flow sensors were compared at the Oakville and Davis sites (Fig. 4). Similar to previous results above, the R^2 values of the regressions were high (>0.92), but the slopes differed. The average midday Ψ_l for vines at Davis from 22 June to 22 September, 2009 was -1.27 ± 0.09 MPa. The average midday Ψ_l for the experimental vines in Oakville from 13 May 2009 to 21 September 2009 was -0.88 ± 0.14 MPa (Table 3). The greater stress exhibited in the vines in Davis could have contributed to the greater overestimation of ET_c as the CIMIS based estimates assume a well watered crop. There was substantial variation between sap flow measured by different sensors at Davis and Oakville, similar to that at the Kearney Agricultural Center.

Circumferential variation in heat-pulse velocity within grapevines

The variation in heat-pulse velocity around the circumference of vines in Davis was investigated by comparing concurrent readings from five sensors placed around the circumference of a vine's trunk (Fig. 5). Sensors varied significantly with midday velocities differing by as much as 80–90 cm h^{-1}.

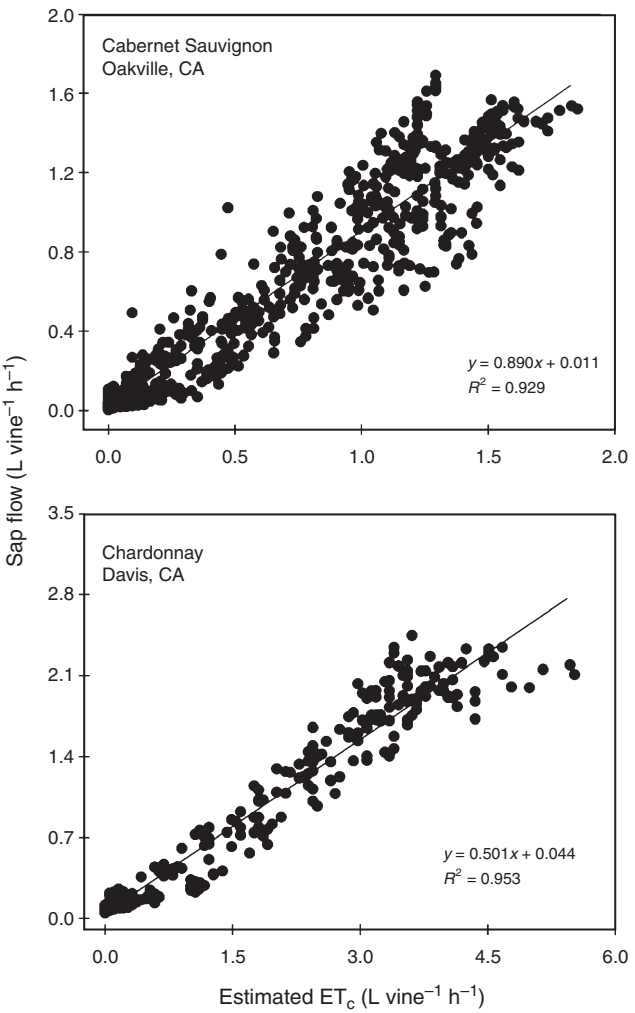

Fig. 4. Relationships between estimated ET_c and sap flow from the Davis and Oakville sites. Estimated ET_c was calculated as the product of ET_o and K_c. Sap flow was the average of five vines at each site, taken every half hour, averaged to hourly values. Data is from three 2 week periods at each site.

Discussion

The dual technique heat-pulse sensors effectively tracked diurnal, seasonal and experimentally-induced changes in sap flow. In almost all situations the sensor measurements were highly correlated with other estimates/measurements of vine water use (i.e. weighing lysimeter, weighing potted plants, and CIMIS based estimates), but the sensors varied considerably even when installed in the same vine. Data from the two techniques typically blended together very smoothly. While variability between sensors has been demonstrated before (Vertessy *et al.* 1997; Lu *et al.* 2000), the high variability shown here likely makes it impractical to use sap-flow measurements to accurately measure volumetric water use for use in automated irrigation. In contrast, the sensors do a good

Table 3. Estimated crop coefficients (K_c) and midday leaf water potentials (Ψ_{leaf}) from Davis and Oakville, CA

Values of K_c were derived from shaded area measurements and the relationship found in fig. 10 of Williams and Ayars (2005*b*). Midday leaf water potential was obtained from 10 leaves, two per vine, at each site. All measurements were made in 2009

Date	K_c	Ψ_{leaf} (MPa)
	Davis	
22 June		–1.31
06 July		–1.37
08 July	0.60	–
23 July		–1.28
24 July	0.57	–
14 Aug	0.58	–1.15
03 September		–1.13
11 September	0.61	–1.22
18 September		–1.34
22 September	0.63	–1.36
	Oakville	
13 May	0.22	–0.70
10 June	0.40	–
15 June	0.41	–
26 June	0.31	–
09 July	0.35	–
21 July	0.36	–0.92
07 August	0.45	–0.79
02 September	0.47	–
03 September	–	–1.05
21 September	0.50	–0.95

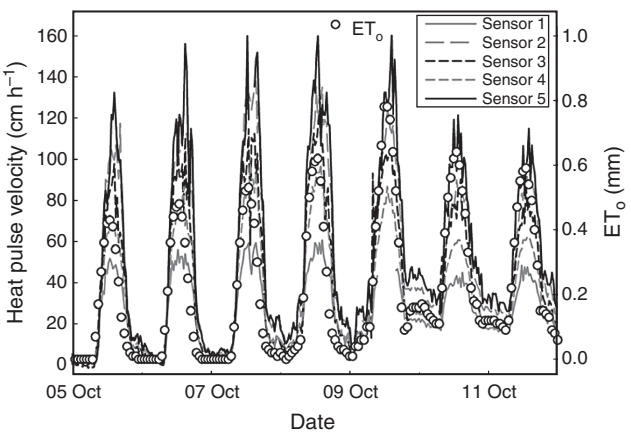

Fig. 5. Corrected heat-pulse velocity from five sensors placed around the circumference of a Chardonnay grapevine grown at the Davis site in 2008. Reference *ET* (ET_o) is also given in the figure as hourly values.

job of tracking changes in vine water use that could be used to evaluate relative changes (i.e. qualitative aspects) in water demands associated with changing microclimatic conditions across the season and by stress imposed by soil water limitations. The novel dual heat-pulse sensors evaluated here show excellent promise for research based qualitative analyses particularly when they can be evaluated against other techniques.

Heat-pulse velocity vs *lysimeter water use*

Comparisons of the heat-pulse velocities obtained from the HRM and CHPM with lysimeter water use indicate that both techniques can be used to effectively capture the full range of sap flow in grapevines. Although the HRM was able to follow ET_{Lys} at low sap-flow rates, it could not measure heat-pulse velocities greater than ~35 cm h^{-1}, which is similar to the upper measurement limit of the HRM reported by Bleby *et al.* (2008). Although not effective under high-flow conditions, the HRM technique does provide accuracy as a research tool for detecting low flow rates that often occur at night (i.e. due to capacitance refilling and/or night-time transpiration) and reverse flow (due to hydraulic redistribution) that are known to occur in grapevines. Inclusion of the HRM technique in a dual sensor enables better approximation of zero flow conditions (assuming accurate probe spacing upon installation) and provides a means to measure dynamic changes in sap-flow patterns documented in roots (e.g. Bleby *et al.* 2010). Our analysis of the CAG procedure demonstrated that CHPM could be used to reliably resolve low and high flows, but HRM would still be needed under conditions of reverse flow (see also other recent efforts along this line that could serve as alternative methods – Green and Romero 2012; Romero *et al.* 2012).

The CHPM sensor was able to follow diurnal lysimeter water use for heat-pulse velocities greater than 30 cm h^{-1} (nearing the upper limit of the HRM sensor but still providing overlap between the techniques), and did not appear to have a maximum recordable value. A decrease in maximum hourly ET_{Lys} from 5.6 to 4.8 L $vine^{-1}$ h^{-1} over a 3 day period was matched with a proportional decrease in heat-pulse velocity as measured by the CHPM (Fig. 1). This hourly value of ET_{Lys} was the highest measured in this study and it demonstrated that the CHPM sensor can match lysimeter water use even under very high flow rates, which appeared as midday plateaus in previous sap-flow studies (Ginestar *et al.* 1998*a*, 1998*b*; Yunusa *et al.* 2000; Lu *et al.* 2003; Patakas *et al.* 2005). The Thompson seedless vines growing in the weighing lysimeter had very high rates of hourly and daily volumetric water use due to the fact that the vines were well watered, grown in an arid environment with high evaporative demand, and trained to an overhead trellis. K_c values of up to 1.3 are not unusual for vines trained to an overhead trellis with the canopy covering a large amount of area (Williams and Ayars 2005*b*).

The CHPM has not been calibrated to independent measures of grapevine water use with flow rates as high as reported here. It is especially important to ensure that a sensor is still accurate at high flow rates, as errors in this range would have the greatest effect on cumulative water use measurements (Shackel *et al.* 1992). When sap velocity is high, the time it takes for the heat pulse to reach the midpoint between the thermocouples is reduced. The same magnitude in error of this time measurement will cause a larger percentage error in heat-pulse velocity when the measurement time is smaller. Therefore, it is not a given that agreement between the CHPM and independent measures of water use found under low flow conditions (Eastham and Gray1998; Yunusa *et al.* 2000; Petrie *et al.* 2009; Collins *et al.* 2010) could be extrapolated to high flows because of its inherent errors. However, we found that heat-pulse velocities determined

from our sensors were proportional to lysimeter water use over a large range including very high flow rates. Furthermore, our dye infiltration experiments confirmed extremely high sap-flow rates in vines surrounding the lysimeter; we found that dye introduced into the base of vine trunks was detected in the leaves at a distance of 4.11 m away from the insertion point within 35 min (AJ McElrone and LE Williams, unpubl. data).

Field and greenhouse calibrations

Volumetric water flow determined from heat-pulse sensors used in this study was highly correlated with ET_{Lys}, but commonly deviated from a 1 : 1 relationship. There are several factors that could have caused the sap velocity to misrepresent actual water use measured by lysimetry including: (1) the heterogeneous nature of grapevine xylem violates assumed conditions in which the heat-pulse velocity method was developed (Marshall 1958), and (2) border effects on measured heat-pulse velocity (Swanson 1983). However, the strong correlations found in the field lysimeter experiment over two seasons indicate that such factors were not substantial enough to cause our sensor measurements to fail to track actual vine water use.

Substantial variation was identified among the heat-pulse velocities for numerous sensors installed around the circumference of single grapevines (e.g. Fig. 5). Circumferential variation in sap flow has been found to be minimal for some species (Bleby *et al.* 2004), whereas other studies have found high variability among measurements taken from multiple sensors positioned uniformly around tree trunks (Vertessy *et al*. 1997; Lu *et al*. 2000). The 23-year-old vines growing in the lysimeter had sensors measuring very high velocities (e.g. sensor #10, Table 1), and others measuring low or no flow (e.g. sensor #8, Table 1). It has been estimated that single point sap-flow sensors measure the flow across 1.5–2 cm of trunk circumference (i.e. ~1 cm on either side of heater needle) (Marshall 1958). For a mature grapevine with a trunk diameter of 10 cm (like those in the lysimeter), a sap-flow sensor measuring 2 cm of circumference would only be sampling <7% of the sapwood. The weighing lysimeter is also located in a Thompson Seedless vineyard with a high prevalence of the grapevine wood disease Esca (LE Williams, pers. obs.), which is common in mature vineyards in California. Variability of functional xylem induced by trunk diseases within and across mature grapevines may prove to make it very difficult, if not impossible, to accurately represent active cross-sectional area of vines for volumetric sap-flow estimation in this growing region, and may help to explain the contradictory results of sapwood area estimation among studies of grapevines (Braun and Schmid 1999; Fernández *et al*. 2008; Petrie *et al*. 2009). Conversely, the sensors installed on young vines growing in the greenhouse exhibited much lower variability among sensors and regression slope values closer to one for balance versus sap-flow measured water use relationships likely resulting from healthy and uniform sapwood in these vines. This suggests that similar problems in functional xylem variability may be minimal for field installation in young vineyards. Non-destructive techniques to estimate the active sapwood area would certainly improve these conversions and facilitate sensor installation targeting the most active tissue.

Between the 2008 and 2009 seasons, the sensors in the lysimeter vines were moved to different locations to limit the wounding response, which has been shown to progressively deactivate tissue around installed sensors (Swanson 1983). In 2009, the sensors were also moved to 'packets' of xylem bulging out from the trunk's surface that could often be traced to individual arms. Sectored packets of xylem have been shown to supply water to individual segments of the plant canopies (Dye *et al*. 1991); therefore, sap flow in these segments can be different from one another depending on the evaporative demand and the portion of the canopy they supply. Targeting of these sectors likely contributed to the large increase in heat-pulse velocities measured in 2009 compared with those of the previous year. The variability of functional xylem as described above made sensor installation difficult to ensure representative sampling of the entire cross-section.

Distinct regression equations of volumetric sap flow and vine water use were also obtained at the Davis and Oakville sites (Fig. 4). Despite a high degree of variability among sensors, combined volumetric water use of all vines was highly correlated with estimated ET_c at each site. Results from Oakville and Davis indicate that the difficulties in scaling point measurements of sap flow to total vine water use found in the weighing lysimeter experiment are pervasive in grapevines, but that sensors consistently correlate well with estimates of ET_c. Our results are similar to those reported by Intrigliolo *et al*. (2009), who compared canopy gas exchange measurements with sap flow on six grapevines. For five of the six vines, there was a strong linear relationship between water use measured by either technique. However, these authors found that the sap-flow sensors underestimated flow and that the slope of the regression of hourly water use measurements was inconsistent between vines. Differences between sap-flow measurements and *ET* estimates at these two sites could also be due to the water status of the vines at the two locations. Grapevine ET_c at both locations was estimated as the product of ET_o and the K_c. The K_c values used in that equation are for non-stressed crops cultivated under excellent agronomic and water management conditions and achieving maximum crop yield (i.e. standard conditions) (Allen *et al.* 1998). Therefore, the estimated values of ET_c calculated at both sites were for non-water stressed vines. The greater slope between sap flow and estimated ET_c at Oakville compared with that at Davis may indicate that the vines at Oakville were less stressed for water than at Davis. This is supported by midday Ψ_l values measured at both sites (Table 3). Daily grapevine water use has been shown to be significantly correlated with midday Ψ_l; water use decreased linearly as midday Ψ_l decreased from –0.8 to –1.25 MPa (Williams *et al.* 2011).

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