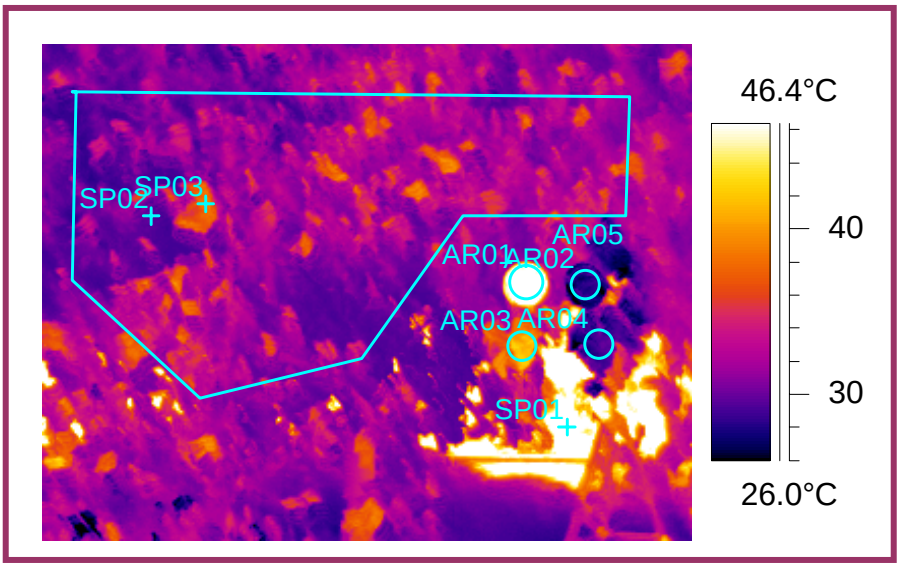
Australian Government

Grape and Wine Research and Development Corporation

When to water? Assessment of plant-based measurements to indicate irrigation requirements.



FINAL REPORT to

GRAPE AND WINE RESEARCH & DEVELOPMENT CORPORATION

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Research Organisation:	**CSIRO Plant Industry**
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Abstract

A number of plant-based measures of performance have been assessed with a view to developing a simple, cost-effective means to determine grapevine response to irrigation. Two methods in particular were able to reliably differentiate between vines receiving different irrigation treatments and show promise as methods capable of development into practical irrigation scheduling tools. These were measurement of pre-dawn leaf water potential with a pressure bomb and the calculation of a canopy conductance index (Ig) based on the temperatures of the leaf canopy and filter paper reference surfaces measured with an infrared thermometer. Many other techniques were also assessed and these included measurement of stomatal conductance with a porometer, determination of sap flow, measurement of midday stem and leaf water potential and dendrometry to determine the relationship between irrigation and trunk diameter. Techniques that relied on fixed pieces of equipment such as sap flow and dendrometry were at a disadvantage because they allowed only a small number of vines to be measured and the cost was high. On the other hand, techniques using portable equipment such as a pressure bomb or a porometer allowed us to make many measurements and to assess both the spatial and temporal variation in measured parameters. We were then able to estimate the most cost-effective sampling strategy to reduce the error to acceptable levels.

Non Technical Summary

To increase the efficiency of current irrigation practices it is desirable to have some way of assessing vine performance in response to changing environmental conditions. In most cases this is currently achieved by monitoring the rate of soil water depletion, which is related to plant water use (high crop water use means faster depletion of soil water). There are many methods available for soil moisture monitoring but they are all quite expensive and tend to be used at selected points within the vineyard which may not take into account the inevitable variability in soil depth and other properties which occur throughout most vineyards. Furthermore, there is no flexibility in where the measurements are made once the access tubes or sensors are put in place. Soil moisture measurement is really only a surrogate for what we really need to know, and that is the performance of the vine under various irrigation practices and current environmental and soil conditions. What is needed is a plant-based measure of vine performance with which to assess both short and long term responses to irrigation and environmental conditions, which could then form the basis of an irrigation scheduling program and a real-time assessment of vine health.

Our knowledge of grapevine physiological response to water deficits and changing environmental conditions allows us to interpret plant-based measurements and make decisions based on these measurements regarding irrigation scheduling. We report on a range of plant-based measures of vine performance to determine the most cost-effective way of irrigation scheduling to better match irrigation input to plant requirement. This is particularly important for new irrigation techniques such as PRD where discontinuities in soil moisture profiles are deliberately created, making scheduling based on soil water measurements more difficult. A number of benefits can flow from such a plant-based monitoring system. For example:

- Instrumentation which is mobile and can be used in multiple locations
- Assurance that plant function is normal
- Possibility of measurements on large areas of canopy to integrate responses, as opposed to the “point” measurements of soil moisture, which may not be representative.
- Irrigation systems which respond directly to the requirements of the crop
- Improved water use efficiency because water input is tightly coupled to crop requirement
- Reduced water wastage better for the environment

This project aims to assess a number of plant-based methods that have potential to be used as irrigation scheduling tools and we have made estimates of how variable these measures are in space and time so that realistic numbers of measurements are made to reduce the inevitable errors

associated with within-vine and between-vine variation. We have summarized below the types of measurements made and commented on the merits and disadvantages of each. Emphasis has been given to the development of techniques that can be readily employed with relatively inexpensive and robust equipment and does not require a high level of operator training.

Technique	Comment
Stomatal conductance	• A sensitive measure of vine performance but subject to rapid change in response to ambient conditions. Data interpretation may be difficult without specialized knowledge • High level of temporal and spatial variability requiring multiple measurements • Equipment not very robust under sustained field use ***A useful research tool but not yet developed for routine commercial use***
Sap flow	• Provides knowledge of the performance of the whole vine • Installations stable for extended periods without attention • Some uncertainty regarding the accuracy of absolute sap flow rates but capable of producing reliable long term relative numbers • Number of vines that can be measured may be limited by cost ***A robust reliable technique capable of further development for commercial use***
Midday leaf water potential (ψL)	• A well established method for assessment of plant water status, however the relationship between measured ψL and physiological function may not be so well established • Guidelines for the interpretation of ψL data have been developed • Equipment is robust and mobile so measurements throughout a vineyard are possible • A high level of spatial variation may require multiple measurements to reduce error • Interaction between stomatal function and ψL may lead to incorrect data interpretation • New loggable equipment may make this measurement more attractive, although then subject to the constraint of few measured vines ***Easily measured with reliable equipment by relatively unskilled personnel. Variability may require many measurements for reliable data. Skill required in data interpretation, especially with regard to varietal differences. New equipment needs development but shows promise.***
Midday stem water potential(ψstem)	• Closely correlated with ψL but less error due to reduced leaf to leaf variability may mean fewer measurements required

	• Advantages offset by time required for placement of bags ***Some advantages over ψL but subject to the same basic constraints***
Pre-dawn leaf water potential (ψpd)	• A sensitive measure of soil/plant water status capable of detecting differences on a daily basis • Easily measured but must be done before sunrise. • Eliminates short-term variability due to stomatal responses • Data interpretation may be compromised by discontinuities in soil moisture as the indicated ψpd will reflect the wettest soil ***The most sensitive measure of water potential for detecting irrigation status. Capable of continuous measurement with new equipment***
Canopy temperature	• Measurement of canopy temperature can readily differentiate vines on the basis of their irrigation status • Thermal imaging techniques demonstrate errors that can be introduced by non-vine components • High leaf to leaf variability eliminated by multiple measurements made from moving vehicle with cheap infrared thermometers • Large areas of vineyard can be measured in a short period of time • Canopy conductance index (Ig), calculated from measurements of canopy temperature and temperature of reference surfaces is a good indicator of vine response to irrigation treatment ***Further development of this methodology is required but it shows significant promise as a cheap, reliable method for large scale use.***
Dendrometry	• Reliable measurements can be made over a long period of time • Data can be logged • Data interpretation is difficult but attempts have been made to automate data analysis and decision making • Vines show different responses at different times of the season, again complicating data analysis • Rain events significantly disrupt the daily patterns of stem diameter changes • Constraints may be imposed due to the cost of equipment and the ability to measure relatively few vines ***Instrumentation can be made robust and reliable and capable of extended periods of measurement. With some more development dendrometry could be a useful tool applicable to grapevines in Australia***

Background to Project

During the past ten years the Australian winegrape industry has experienced a period of massive expansion fuelled by demand for high quality, reasonably priced wines in world markets. Since 1987 the area planted to grapevines has increased from 60,000ha to the current area of over 150,000ha. This expansion has occurred in a climate of decreasing availability of water for irrigation, an increase in its cost and a heightened awareness of environmental issues among growers, the public and governments. There is therefore considerable pressure on the winegrape industry to increase the efficiency of water use.

Irrigation practices always have the potential to be environmentally damaging through depletion of ground and surface water reserves, raising water tables, salinisation of soils, increasing soil sodicity and the leaching of nutrients and agricultural chemicals into water courses. Problems such as these may threaten the long-term sustainability of many irrigation areas. Some of the problems associated with irrigation can be minimized by improving the efficiency of water use, thereby reducing abstraction and runoff. However, as water inputs are reduced, there is an increased risk of inducing damaging water deficits in the crop. There is therefore increasing pressure on grape growers to improve irrigation efficiency. In addition to sound economic reasons for this there are equally pressing environmental issues to be addressed such as over-commitment and depletion of ground and surface water resources, rising water tables, salinity, and the need to be and be seen to be environmentally responsible. Irrigation efficiency has been increasing under these pressures as irrigation inputs are better matched to vine water requirements and further improvements in irrigation efficiency are also possible through implementation of techniques such as partial rootzone drying (PRD) which deliberately modify the physiology of the plant to make it more efficient in its use of available water.

To further increase the efficiency of irrigation it would be desirable to have some way of assessing vine performance in response to changing environmental conditions. In most cases this has been achieved by monitoring the rate of soil water depletion, which is related to plant water use (high crop water use means faster depletion of soil water). There are many methods available for soil moisture monitoring such as neutron probe, time domain reflectometry (TDR), gypsum blocks and capacitance sensors. However, they are all quite expensive and thus tend to be used at selected points within the vineyard which may not take into account the inevitable variability in soil depth and other properties which occur throughout most vineyards. Furthermore, there is no flexibility in where the measurements are made once the access tubes or sensors are put in place.

Soil moisture measurement is really only a surrogate for what we really need to know, and that is the performance of the vine under various irrigation practices and current environmental and soil conditions. What is needed is a plant-based measure of vine performance with which to assess both short and long term responses to irrigation and environmental conditions, which could then form the basis of an irrigation scheduling program and a real-time assessment of vine health.

Our knowledge of grapevine physiological response to water deficits and changing environmental conditions has become sophisticated enough to allow us to interpret plant-based measurements and make decisions based on these measurements regarding irrigation scheduling. In this document we report on a range of plant-based measures of vine performance to determine the most cost-effective way of irrigation scheduling to better match irrigation input to plant requirement. This is particularly important for new irrigation techniques such as PRD where discontinuities in soil moisture profiles are deliberately created, making scheduling based on soil water measurements more difficult. A number of benefits can flow from such a plant-based monitoring system. For example:

- Instrumentation which is mobile and can be used in multiple locations
- Assurance that plant function is normal
- Possibility of measurements on large areas of canopy to integrate responses, as opposed to the “point” measurements of soil moisture, which may not be representative.
- Irrigation systems which respond directly to the requirements of the crop
- Improved water use efficiency because water input is tightly coupled to crop requirement
- Reduced water wastage better for the environment
- Increased knowledge of vine physiology under deficit irrigation will help in refining the techniques for future use

Project Objectives

The margin for error in matching water input to plant requirement becomes smaller as the buffer provided by the high water inputs is removed. Furthermore, as the link between water deficit and grape quality is exploited to produce winegrapes to specified quality, there will be an additional need to monitor plant performance in relation to water input.
This project therefore proposes to assess a range of plant-based measures which will directly link irrigation input to vine performance, thereby increasing the efficiency of water use and simplifying the manipulation of plant water status. Priority will be given to the development of methods which offer cost-effective solutions to the objective of supplying the Australian winegrape industry with tools to further improve the efficiency of irrigation water use.

The performance of a grapevine canopy, in terms of its ability to support the development of a grape crop and its response to environmental variables such as soil moisture, ambient humidity, ambient CO_2 etc., can be described by a number of plant-based measurements. Some of these are described below:

Technical information concerning the issues to be addressed

Stomatal conductance

Stomatal conductance is tightly linked to availability of soil water but in addition shows rapid response (minutes) to potentially stressful conditions such as high ambient temperature, low relative humidity, high wind velocity, changed ambient CO_2 (climate change).

Leaf/canopy temperature

Evaporation of water in leaf sub-stomatal cavities leads to leaf cooling. Thus, transpiration rates can be inferred from the difference between leaf or canopy temperature and ambient air temperature. Water stressed leaves will have a temperature at or above ambient if transpiration is restricted by stomatal closure whereas transpiring leaves will have depressed temperature relative to ambient. Some irrigation practices such a RDI and PRD result in a degree of stomatal closure, so leaf temperature may also be indicative of the effectiveness of these techniques. Infrared thermometry is able to integrate large areas of canopy into one measurement and thermal imaging techniques can provide more information about variations in canopy temperature and authenticate measurements made with simple infrared techniques.

Shoot and trunk diameter

The diameter of shoots and trunks show typical diurnal change indicative of growth in water-abundant and water-restricted conditions. Interpretation of these patterns can allow an assessment of plant water requirements.

Sap flow

Measurement of the flux of sap from roots to shoot is an effective way to determine the integrated transpiration from the entire vine canopy, which is subject to perturbations caused by atmospheric or soil water conditions.

Leaf/stem water potential

As soil water availability declines so does soil water potential. This in turn influences the water potential of the aerial parts of the vine. Simple measures of leaf or stem water potential may therefore be indicative of soil and canopy water conditions, particularly if taken during the night when soil moisture is in equilibrium with canopy moisture potential. During daylight hours leaf water potential is subject to short term variation driven by

current stomatal aperture and the prevailing evaporative pressure described by air temperature, wind speed and humidity.

Temporal and Spatial Constraints

This project recognizes the variability in vine performance that exists in most vineyards. This variability can be caused by changes in vineyard characteristics such as soil properties, irrigation water delivery, topography etc. In addition, many of the plant-based characteristics which may potentially be useful indicators of plant performance show significant changes throughout the day. For example, stomatal conductance is low during the hours of darkness and in well watered vines reaches a maximum sometime around midday, similarly, leaf water potential tends to equilibrate with soil moisture potential during the night and falls rapidly as transpiration is driven up by increased temperature and solar radiation during the day. These diurnal changes may be superimposed on constraints imposed by vineyard physical characteristics resulting in both spatial and temporal influences on measured plant parameters. One of the project objectives was therefore to differentiate and quantitate these influences. The technique employed was to measure plant response at a fixed point in the vineyard and also at a number of designated vines scattered throughout the vineyard. The fixed point measurements will describe temporal variation and the measurements made throughout the vineyard will describe both temporal and spatial variation.

Details and justification of methods adopted

Experimental sites

Experiments were carried out at a number of locations (Table 1)

Table 1. Experimental sites and locations

Site	Location	
	S	**E**
University of Adelaide Coombe Vineyard	34.58.01	138.38.01
SARDI Nuriootpa Research Station	34.28.24	139.00.17
Michael Bowe Vineyards Woodside, Adelaide Hills	34.56.26	138.52.07
Rosemount Vineyard Langhorne Creek	35.19.37	138.59.80

University of Adelaide Coombe Vineyard

Experimental vines were own-rooted Cabernet Sauvignon and Shiraz trained as a vertically shoot positioned canopy. Rows were oriented north-south. Vine and row spacing were 1.8m and 3.0m respectively. Irrigation was controlled by the vineyard manager and the vines received between 0.7 and 1.0ML/ha.

SARDI Nuriootpa Research Station

Experimental vines were own-rooted Shiraz, Grenache and Cabernet Sauvignon. They were trained to a single cordon and no shoot positioning was employed. Rows were oriented east-west. Vine and row spacing was 2.0m and 3.0m respectively. A number of irrigation strategies were employed to allow independent control of water input to sections of the vine rows so that degrees of water stress could be imposed.

Michael Bowe Vineyards Woodside, Adelaide Hills

Experimental vines were own-rooted Chardonnay and Merlot. They were trained to a single cordon and vertically shoot positioned. Vine and row spacing was 1.8m and 2.8m respectively. The irrigation was controlled by the vineyard manager although sections of the row comprising either 20 or 4 vines could be isolated by appropriate by-pass tubing and taps. This allowed the vines in these sections to receive less irrigation and become stressed.

Rosemount Vineyard Langhorne Creek

The site chosen comprised a 7ha block of own-rooted Shiraz vines planted in 1992 (Figure 1). The site has been used extensively for other GWRDC-funded projects and has been selected as one of the sites in the new GWRDC Soil and water Initiative. Details of the site can be found in the report to GWRDC on project CSL01/01 by DM Lanyon and RGV Bramley "*Ameliorating soil constraints to the performance of established vineyards*". That project had established three treatments on the block: viz. control, mulched and ripped. The mulching treatment applied 150m^3/ha of composted grape marc along the vine rows. The deep ripping was intended to improve root penetration into the mid-row by incorporating 150m^3/ha of composted grape marc in the top 50mm along the wheel tracks and then ripping to 400mm. Each treatment comprised 4 rows replicated 15 times throughout the block. For the CSP02/02 experiments only the control vines were used. Vines from which measurements would be taken were selected and identified with bar code tags (Figure 2). The location of each tagged vine was determined using a differentially corrected global positioning system accurate to ±500mm. An automatic weather station was located on the headland adjacent to the vine block. Water meters were inserted in some of the irrigation laterals so that uniformity of water application could be checked.



Figure 1. Location of the experimental block at the Rosemount Estates Langhorne Creek vineyard (grey rectangle).

The site was originally chosen because of large variation in soil properties which limited vine performance, resulting in vines of generally low vigour (Lanyon and Bramley, 2004).

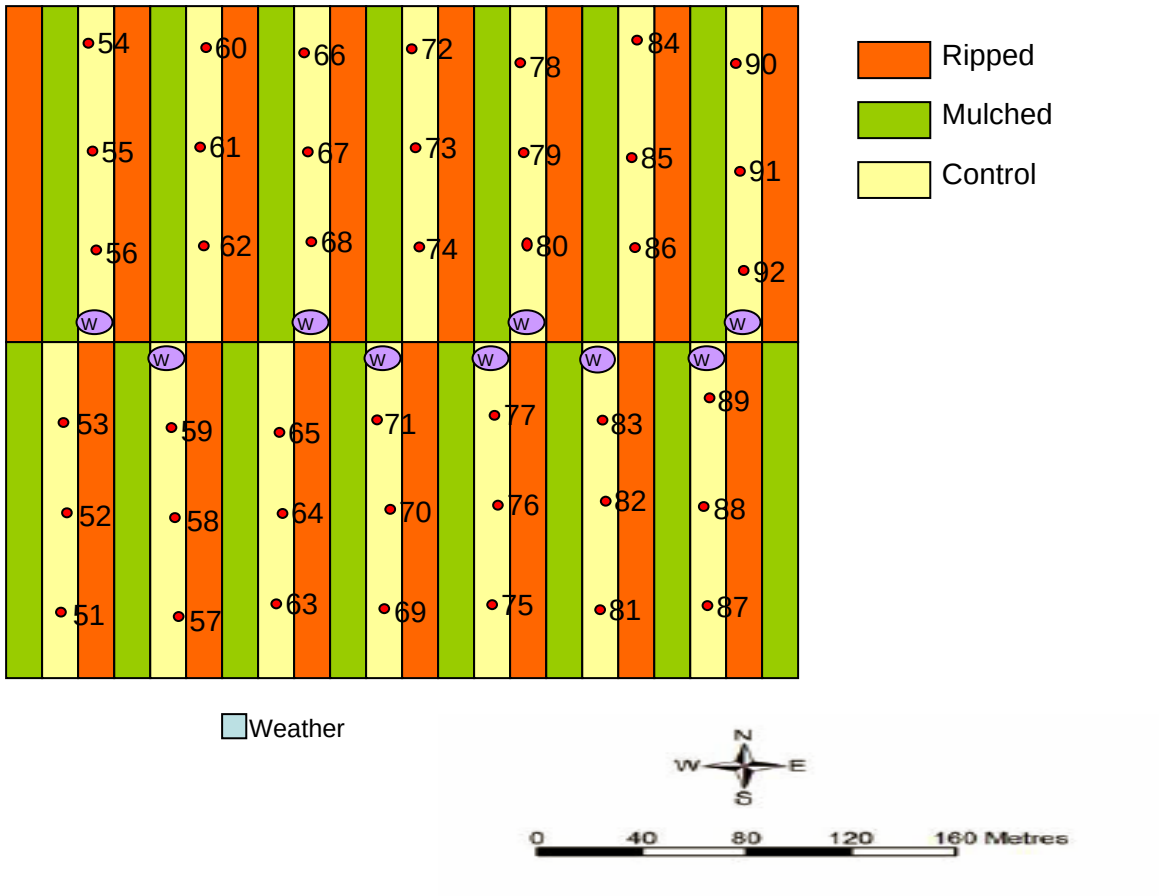


Figure 2. Location of experimental vines and their assigned numbers at the Langhorne Creek site. Also shown are the water meter and automatic weather station locations.

Plant measurements

Water potential

Fully sun exposed leaves were selected and enclosed in a plastic bag before excising the petiole 10-12mm from its point of attachment. Leaf water potential was measured within 20s of excision with a 3000 series Plant Water Status Console (Soilmoisture Equipment Corp, Santa Barbera, USA) using industrial grade nitrogen to pressurize the chamber. The rate of pressurization was adjusted to approximately 0.5MPa/min and this rate was retained for all measurements. The instrument had been modified to include a needle valve for pressurization rate adjustment and replacing the analogue pressure gauge with an electronic digital pressure

transducer and readout, reading to 0.01MPa. The electronics were designed so that the maximum reading (leaf water potential) was retained until the unit was depressurized. This eliminated errors which can occur when manually transcribing output from an analogue gauge. In some experiments two pressure bombs were required and in these instances the instruments were cross-calibrated by simply connecting them together. Appropriate corrections were applied where necessary to equalize the readings. Stem water potential was measured by enclosing selected leaves in double layer plastic bags comprising an inner black layer and an outer white layer. This ensured the exclusion of light and the minimization of radiative heating. The bags were applied the evening before measurement. In the case of pre-dawn water potential, it was necessary to ensure that leaf water potential had equilibrated with soil water potential and a series of measurements were made at the Langhorne Creek site from midnight onwards. No further change in water potential occurred after about 02.30 and subsequent measurements of pre-dawn water potential were made between 03.00 and first light.

One of the disadvantages of leaf water potential as a measure of plant water status is the traditionally measurements have been made manually with a pressure bomb as described above. However, during the course of this investigation we became aware of a system for leaf water potential measurement based on psychrometric sensors which can be continuously logged (Psψpro logger and L51 sensors, Wescor Inc. Logan, Utah). These were tested and compared with traditional measurement methods using a pressure bomb. There are significant safety issues associated with the use of traditional pressure bomb equipment pressurized with bottled gas in that it is possible to over pressurise the chamber to the point where failure of seals could result in explosive expulsion of plant and seal material. As an alternative we have tested a manually pressurized pressure bomb (PMS Instrumant Company, Albany USA) and one pressurized with a small electric tyre pump. The latter would be appropriate for pre-dawn water potential measurement where values would be unlikely to be less than -1.0MPa which was the pressure limit of the electric pump.

Stomatal conductance

Stomatal conductance was measured with Delta-T AP3 porometers calibrated according to the manufacturer's instructions. Conductance measurements were also made with a PPSystems steady state porometer. This instrument was modified so that data were output through a serial interface to a Symbol Palm computer which incorporated a barcode reader. This combination

allowed real-time calculation of conductance data and linking of individual measurements to identification bar code tags attached to each vine. All porometers were cross-calibrated by multiple sequential measurements of leaves.

Sap flow

Xylem sap velocity was measured with Greenspan SF100 sensors. These were installed in vine trunks by drilling radial holes completely through the trunk, passing through the trunk centre. The probes were pushed through the trunk to emerge on the far side. Probe tips were located such that the thermistor sensors were 5mm below the surface. This method of installation provided physical support for the probes and accurate placement of the thermistor elements. The protruding sensor supports were insulated with glass fibre insulation and the whole sensor body and vine trunk wrapped in reflective aluminized insulation. Data were logged at 30min intervals.

Infrared thermometry

Canopy temperature was estimated with infrared thermometers. Instruments used were an Everest 1100 and a Raytek Ranger MX. Temperature was measured on the sun-exposed or shaded part of the canopy by making multiple hand-held measurements or with the instruments attached to a data logger. The analogue output from the thermometer, along with 5 thermistor sensors, was logged at 4Hz using a custom-built logger system (Measurement Engineering Australia). Four thermistor sensors were assigned to appropriate reference surfaces and the fifth measured ambient air temperature in a small aspirated screen. Measurements were made with the operator sitting in the tray of a utility vehicle or on a quad bike. The infrared thermometer was aimed at the vine canopy as the vehicle was driven slowly along the vine row. In this way several hundred meters of vine row could be sampled in a few minutes. Details of how this equipment was used are further described in the appropriate section of the report.

Thermal imaging

Images of vine canopies were captured using a Thermacam SC2000 handheld long-wave thermal imager (FLIR Systems, Basingstoke) and these images were processed using Thermacam Researcher software.

Results and Discussion

Leaf water potential

Leaf water potential (ψL) is relatively easily measured with cheap, robust equipment. It does not require a high degree of skill to obtain the measurements. Guidelines for interpreting ψL have been developed in the United States where this is commonly used as a tool to assist in irrigation scheduling.

Table 2. Values of midday leaf water potential used as a guide for irrigating winegrapes in California

Midday leaf water potential (MPa)	Stress condition
More than -1.0	No stress
-1.0 to -1.2	Mild stress
-1.2 to -1.4	Moderate stress
-1.4 to -1.6	High stress
Less than -1.6	Severe stress

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Under Australian conditions, if the aim was to have non-stressed vines, attaining ψL more than than -1.0MPa would be difficult, and even well watered vines in the Riverland of South Australia, receiving in excess of 6ML/ha would be classified as moderately or highly stressed, having midday ψL of less than -1.4MPa (McCarthy 1997, Soar, unpublished). Chardonnay vines growing in the Barossa Valley under irrigation conditions typical of the region had a minimum midday water potential close to -1.8MPa (Figure 3) which would be classified as severely stressed according to the US scheme. These Barossa vines yielded on average 12.9 t/ha which is

higher than the national average of 12 t/ha for Chardonnay and considerably more than the Barossa average of 8.5t/ha (ABS figures) and so the low ψL had not compromised yield.

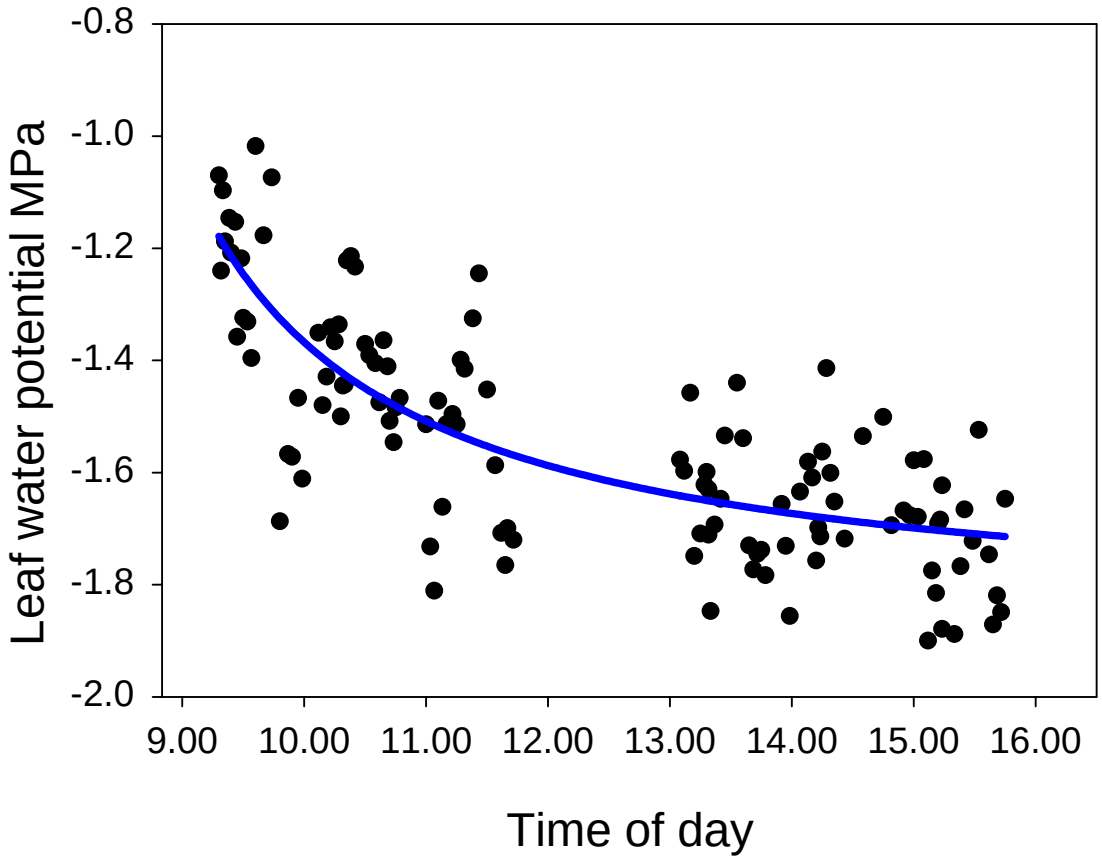


Figure 3. Diurnal changes in leaf water potential (ψL) of Chardonnay vines growing in the Barossa Valley. Measurements made February 1st 2003.

There is considerable scatter in these data and later we will present analysis of ways to reduce this "noise" for minimum time cost. A measure of ψL less subject to temporal and spatial variation may be pre-dawn leaf water potential (ψpd). ψpd is considered to be a surrogate for soil matric potential since during the night when stomata are closed and transpiration pressure is low leaf water potential will become equilibrated with soil water potential, or at least the wettest part of the soil that the roots of that plant are in contact with. At the Langhorne Creek site it was found that ψL rose during the hours of darkness and showed only minor changes after 2am (Figure 4). All pre-dawn water potential measurements reported hereafter were therefore made between 3am and dawn.

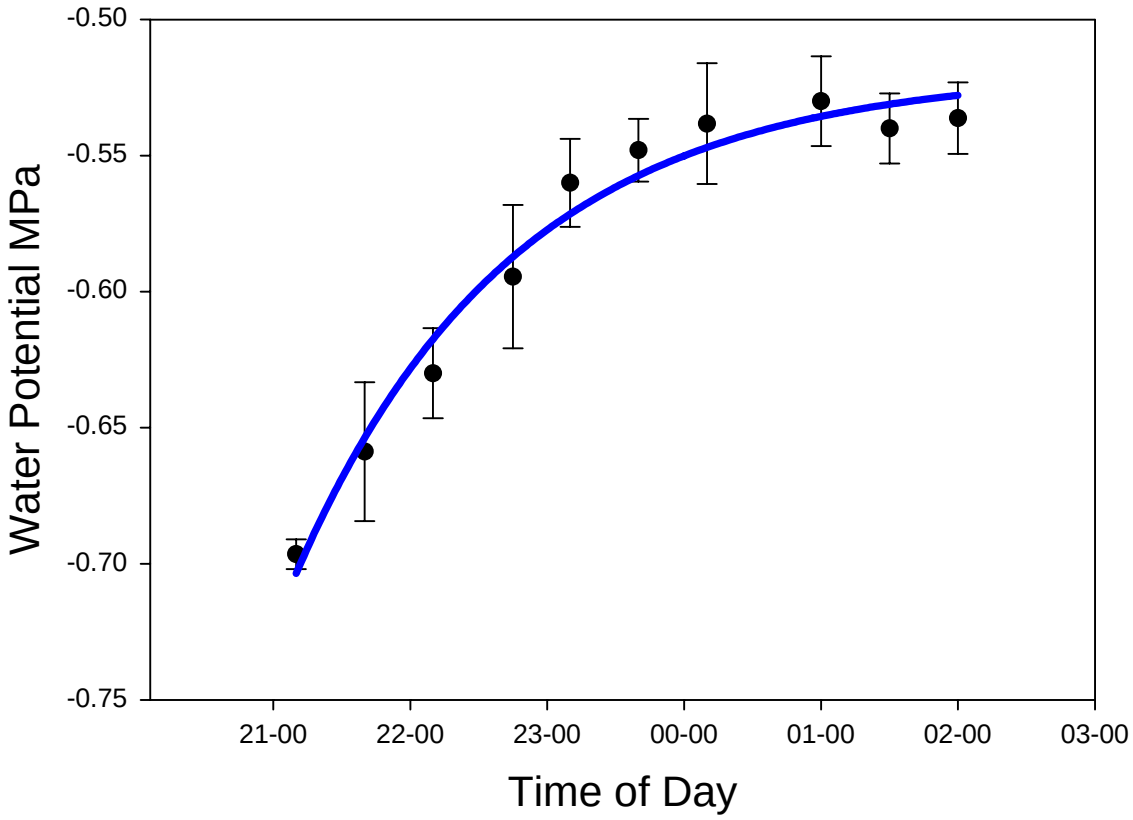


Figure 4. Leaf water potential of Shiraz vines at Langhorne Creek. Figures at each time point are mean (n=10) and standard error. All measurements are of leaves from a single vine in the centre of the block.

Pre-dawn leaf water potential responded sensitively to irrigation status. In an experiment on Merlot vines in the Adelaide Hills at Woodside, a 10 vine section was isolated from others in that row by switching the water supply from the normal drip line to a by-pass line with no drippers. Measurements were made on the centre 2 vines. Water was withheld for an 8 day period beginning one day after the vines were last irrigated. The remaining vines in the row, some of which served as controls, were irrigated every second day. ψpd fell progressively during the measurement period (Figure 5). Midday leaf water potential for the same period changed little however despite significant changes in VPD during the measurement period (Figure 6).

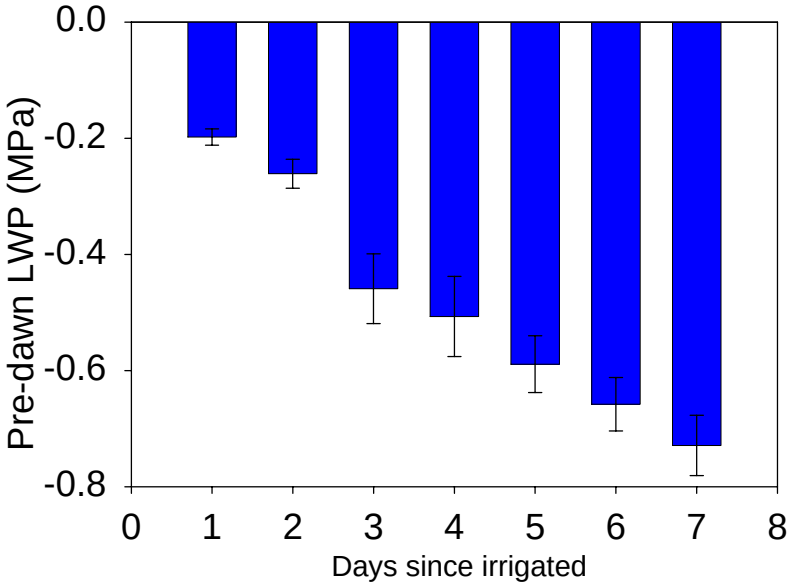


Figure 5. Pre-dawn leaf water potentials of Merlot vines in the Adelaide Hills. Water was withheld on day 0. Figures are means and standard errors (n=10).

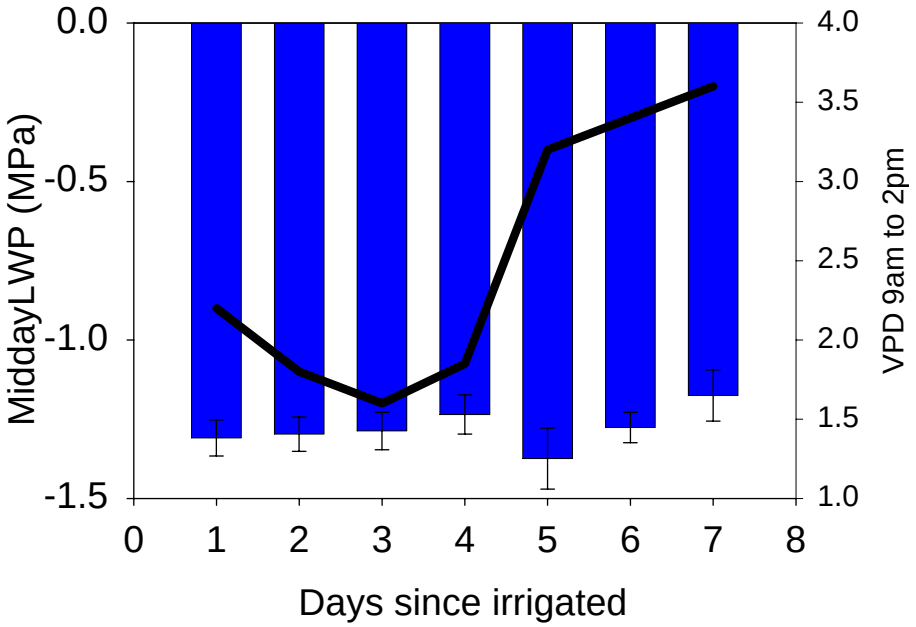


Figure 6. Midday leaf water potentials of Merlot vines in the Adelaide Hills. Water was withheld on day 0. Bars are means and standard errors (n=10) and solid line is mean VPD (KPa) between 9am and 2pm.

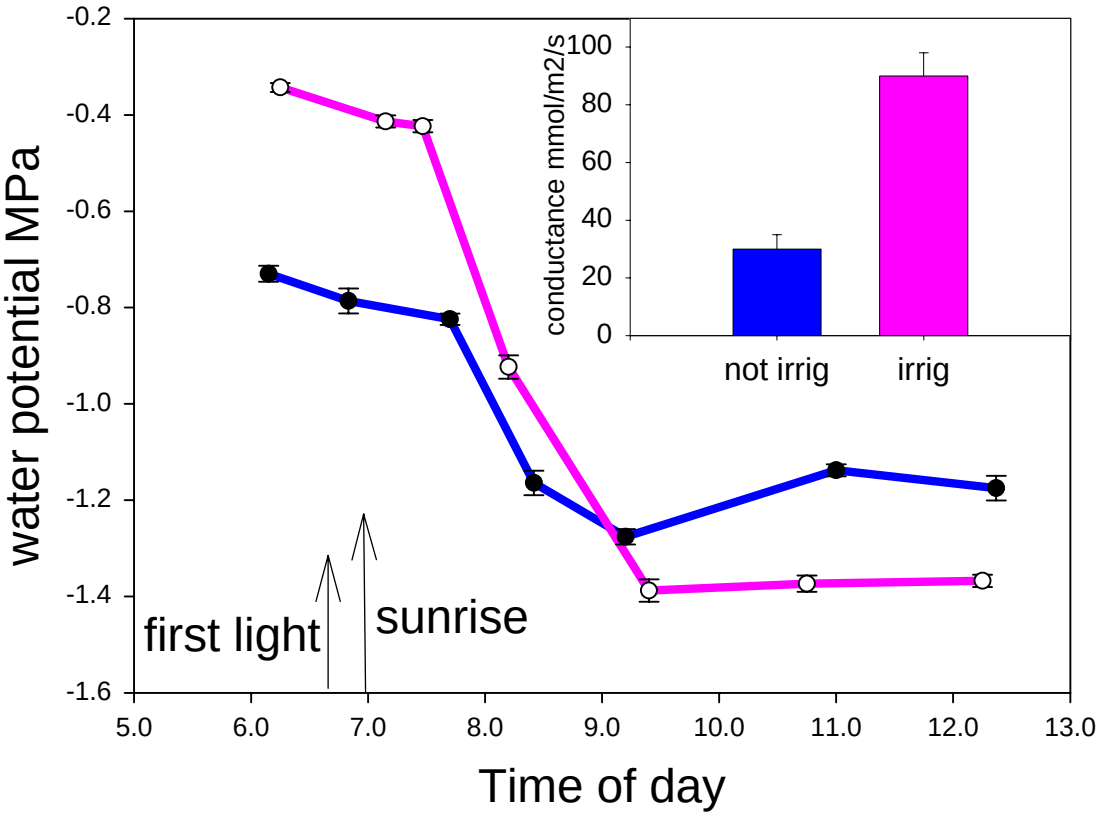


Figure 7. Leaf water potential of irrigated (pink) and not irrigated (blue) Merlot vines. Data points are means and bars are standard errors (n=10). Inset shows stomatal conductance of the same vines measured between 11.00h and 13.00h (n=20).

On day 8 of the experiment measurements of ψL were continued from pre-dawn until after midday (Figure 7). ψL of the non irrigated vines was significantly lower than that of the irrigated vines at pre-dawn and these values changed little until the leaves received direct sunlight. Thereafter ψL fell rapidly in both treatments. However, the fall was more rapid in the irrigated vines than in those that had not received irrigation for 8 days. By 11am the ψL of the irrigated vines was significantly lower. This difference was the result of higher transpiration in the irrigated vines. The inset in Figure 7 shows that stomatal conductance was significantly lower in the non-irrigated vines.

These data strongly endorse pre-dawn leaf water potential as a measure to indicate vine water status and questions the potential of midday ψL to be a meaningful indicator of vine water status. In the absence of a knowledge of ψpd the midday values of ψL data alone would have suggested

that the vines that had not received water for 8 days, and would by other criteria have been considered to be seriously water stressed, were in fact better hydrated than the vines that had been recently irrigated. This paradox was due entirely to the fact that stomatal conductance reflected the irrigation status of the vines and that greater transpiration in the irrigated vines reduced ψL under the high VPD conditions of the day. In the absence of this additional information ψL alone can be misleading.

In this experiment irrigation was withheld from a block of 10 vines and measurements made on the centre 2 vines. There was thus a significant buffer between the measured vines and those within the same row receiving irrigation. The behavior of ψpd, ψL and stomatal conductance is consistent with the interpretations given above but other experiments on these same rows of vines used blocks of 4 vines, with measurements made on the centre 2 vines. Data from these experiments were consistent with an interpretation that the measured vines were influenced by irrigation events outside of the experimental vine block. In these experiments (data not shown) ψpd did not fall as expected after water was withheld but nevertheless the vines showed signs typical of water stress such as leaf yellowing and reduced stomatal conductance. It is likely that these vines were insufficiently buffered from adjacent vines receiving a different irrigation treatment. ψpd became equilibrated with the small portion of the vines roots that must have had access to water. However, during the day this water was rapidly depleted and the vines became stressed. This problem was not apparent when an adjacent row received a different irrigation treatment, suggesting that root access to the adjacent row was restricted by soil compaction in the wheel tracks. The lesson from this is that vines will behave during the day according to the moisture status of the bulk of the roots. In other words, the relatively small block size would have given meaningful data if the intention was to relate soil moisture to canopy performance, but measurements of ψpd will be influenced by discontinuities in soil moisture such that the values will reflect the wettest roots, irrespective of their amount relative to total root mass.

We have seen above that ψpd can be made as soon as equilibrium with soil moisture occurs, in the case of the vines at Langhorne Creek this occurred at about 3am and that this value will change little until ψL is driven down by increased transpiration after sunrise. Whenever this measurement is made it involves working during the hours of darkness and this is unlikely to be popular with vineyard staff (or research workers) who also have plenty to do during daylight hours. Some method of making these measurements automatically would therefore be desirable.

Towards the end of this project we became aware of sensors sold in the United States designed to measure ψL , the output from which can be logged using a purpose-built power supply and logger (PSψPRO water potential system, Wescor Inc). The sensors are miniature thermocouple psychrometers that can be pressed against a leaf surface for ψL measurement. The sensors actually measure the relative humidity of the air in equilibrium with the leaf. The relationship between water potential and relative humidity (from the Wescor manual) is give by

$\psi = (RT/V_w)e/e_0$

Where:

ψ = total water potential

R = universal gas constant

T = absolute temperature

V_w = molar volume of water ($1.8x10^{-5}m^3.mol^{-1}$)

e/e_0 = relative humidity expressed as a fraction

The water potential that supports plant growth is from 0 to -1.5MPa (1.0 to 0.993 RH)

Some of these sensors were obtained and tested on vine leaves. Since vine leaves have a relatively thick and water-proof cuticle equilibration of the relative humidity between the inside of the leaf and the sensor chamber was slow or did not occur at all. This was true whether the sensor was applied to the abaxial or adaxial leaf surface since in the darkened environment of the chamber the stomata were closed. By lightly abrading the leaf surface with 600 grade abrasive paper the cuticle barrier was removed and RH equilibrium was attained (Campbell and McInnes, 1999). After leaf abrasion it did not matter to which leaf surface the sensors were applied. During the pre-dawn period indicated water potential was approximately -0.4MPa (Figure 8), which is consistent with pressure bomb readings taken on these vines (eg. Figure 7). The changes in water potential recorded by the sensors throughout the rest of the day are consistent with pressure bomb readings with a minimum water potential being reached at approximately 3pm. The minimum value of -1.2MPa was also consistent with pressure bomb readings for these vines. The sensors therefore appeared to provide believable quantitative and temporal data. Eight sensors were attached to some Chardonnay vines at the Adelaide Hills site, four were on vines receiving regular irrigation and four were on vines from which water was withheld from day 77 (Figure 9).

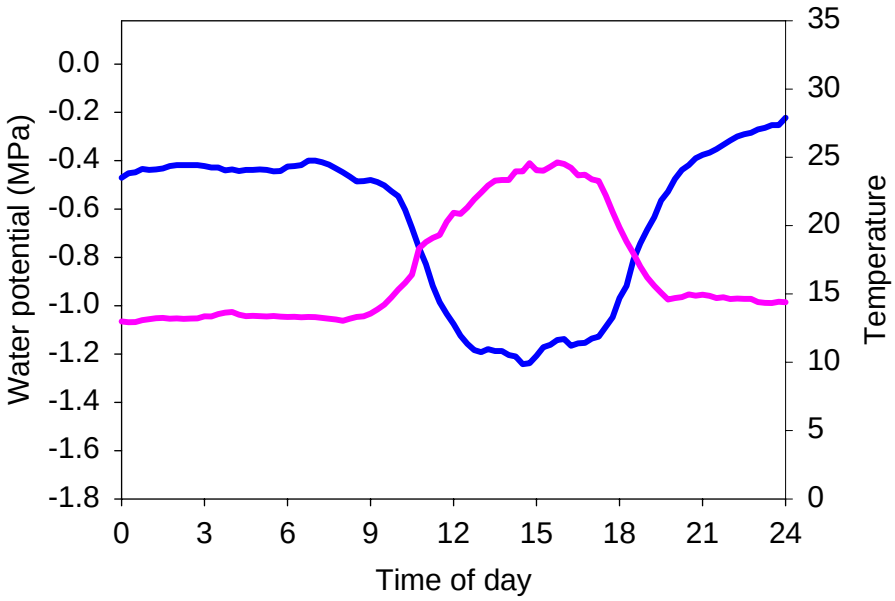


Figure 8. Water potential of Merlot leaf as indicated by Wescor sensor (blue line) and ambient temperature (pink line).

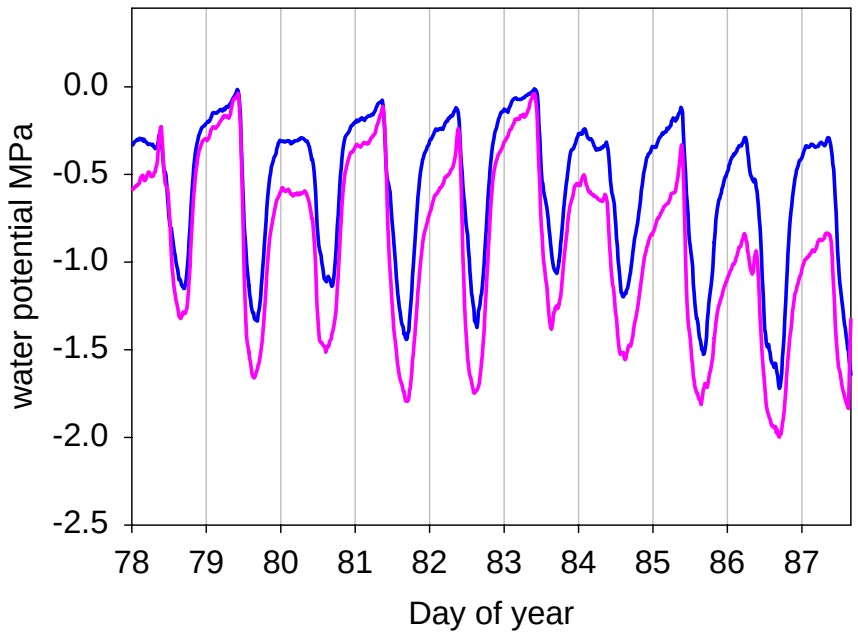


Figure 9. Water potential of Chardonnay leaves as indicated by Wescor sensors. Control irrigated vines (blue) and not irrigated from day 77 (pink). Each line mean of 4 sensors.

All sensors showed consistent diurnal trends with minima in late afternoon and maxima sometime during the hours of darkness. The minimum water potential of the non-irrigated vines was on average about 0.4MPa lower than the irrigated vines. On most days the maximum predawn water potential was lower in the non-irrigated vines and this tendency became more apparent towards the end of the measurement period as the non-irrigated vines became more stressed. There was also a tendency for the pre-dawn water potential in the non-irrigated vines to take longer to reach a stable value. This preliminary evaluation of Wescor sensors suggests that they are capable of producing data that is entirely consistent with measurements made manually with a pressure bomb. And they have the obvious advantage that they can accumulate data unattended. One disadvantage is that the sensors can become dislodged from the leaves due to wind or vineyard operations and data will be lost. At the suggestion of Professor Steve Tyerman we have tried installing the sensors in the trunks of trees. This puts the sensor in direct contact with the xylem and should have a number of advantages over installation on leaves. These include more protection of the sensors, the data should represent the water status of the whole vine and not individual leaves and they should be more stable over time. It was found that an installation on a vine leaf only gave reliable data for a maximum of about 10 days due to deterioration of the leaf under the sensor. This deterioration occurred much more quickly if the leaf was abraded too vigorously. A guide seemed to be that if the abraded area looked dark compared with surrounding tissue then it would have a very short life under the sensor. The object was to remove the cuticle without damage to the underlying cells. Because the idea of placing the sensors in trunks rather than on leaves did not occur before the vines were senescent we tested them instead in trunks of *Eucalyptus camaldulensis*. An 11mm hole was drilled with a standard twist drill in the trunk to a depth of about 12mm (Figure 10A). The standard drill bit was then replaced with one that had had its point removed. The sensor was then inserted into the resulting flat-bottomed hole in direct contact with the wood. The sensor was sealed into the hole with silicon sealant (Figure 10 B). The sensors were then wrapped in glass-fiber insulation and the part of the trunk where the sensors were installed was wrapped in aluminized roof insulation material.

The water potential measured by the sensors showed normal diurnal trends (Figure 11). Pre-dawn water potential was about -0.2MPa and this began to fall slowly commencing at about 8am and more sharply after midday to reach a minimum at about 4pm. The pre-dawn value was regained at about 9pm. Given that the tree was close to a water source (Figure 10A) this pre-dawn value seemed reasonable. The most negative value recorded by this sensor was -4.8MPa which seems



Figure 10. Installation of Wescor sensors in trunk of *Eucalyptus camaldulensis*. A. drilling access hole. B. Finished installation showing sensors sealed into trunk.

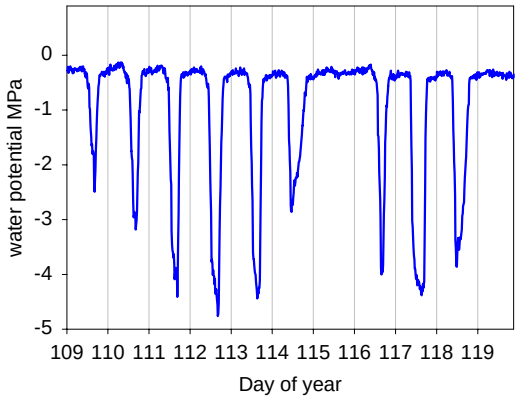


Figure 11. Water potential indicated by Wescor sensor in trunk of *E. camaldulensis* tree.

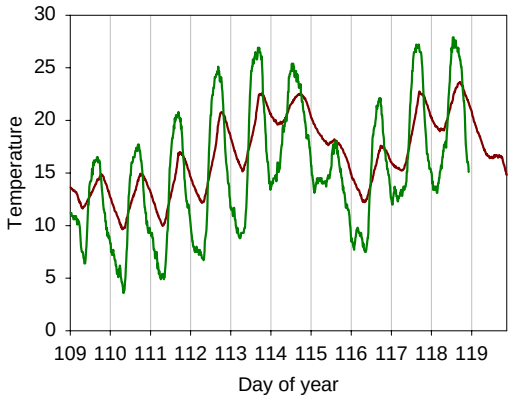


Figure 12. Sensor (brown) and ambient (green) temperature.

rather low and these low minima were a feature of most sensors in this tree. The minimum values of water potential appeared to be more closely related to ambient air temperature than to sensor temperature (Figure 12), giving some confidence that there was minimal temperature effect on the actual reading. This conclusion is further justified by consideration of data on day 115 when there was heavy cloud cover and water potential did not fall below -0.3MPa. The sensors remained in the tree for over 2 months and three out of the four sensors were functioning normally at the end of that period. In summary, this short test of Wescor L51 thermocouple psychrometer sensors suggested that they are a viable alternative to the pressure bomb as a means of measuring leaf water potential. They have advantages of relatively low cost compared with a pressure bomb, ability to log data unattended for extended periods. When used to measure stem water potential there are further advantages of more stable and long-lived installation but some work will be required to fully understand the quantitative nature of the delivered data. It is intended to install some in grapevine trunks during the 05/06 season and validate the readings with measurements of leaf water potential using a pressure bomb.

Spatial and temporal components

As outlined above, one of this project's main objectives was to determine the feasibility of making plant-based measurements to aid the decision making process with regard to irrigation scheduling. The questions we have asked are:

- Would it be feasible to make measurements on a relatively small number of vines and extrapolate the results to the whole vineyard?
    - If the answer is YES, how many measurements would we need to make and over what time interval?
    - If the answer is NO, how many vines throughout the vineyard would need to be measured and over what time interval?
- What is the best measurement to make?

In trying to answer these questions we have concentrated on well developed techniques that can be accommodated with readily available instruments, can be carried out by personnel with a minimum of training and with a minimum of data interpretation. Leaf water potential (ψL) would seem to fit these criteria quite well, especially as some guidelines for its use have already been developed, but as shown above there may be difficulties in data interpretation because of the tight link between ψL and current stomatal conductance. To reduce the dependence of ψL on

conductance it has been suggested that a better measure of leaf water status would be stem water potential (ψstem). ψstem is determined by covering the leaf for a period before measurement to inhibit transpiration. The assumption is made that the measured water potential reflects that of the stem (or cane in the case of grapevine) and would be a better value to represent integrated canopy water status. In practice leaf transpiration is inhibited by placing a plastic bag over the leaf the night before measurement.

ψL was measured at the Langhorne Creek site on a number of occasions. A representative data set is shown in Figure 13

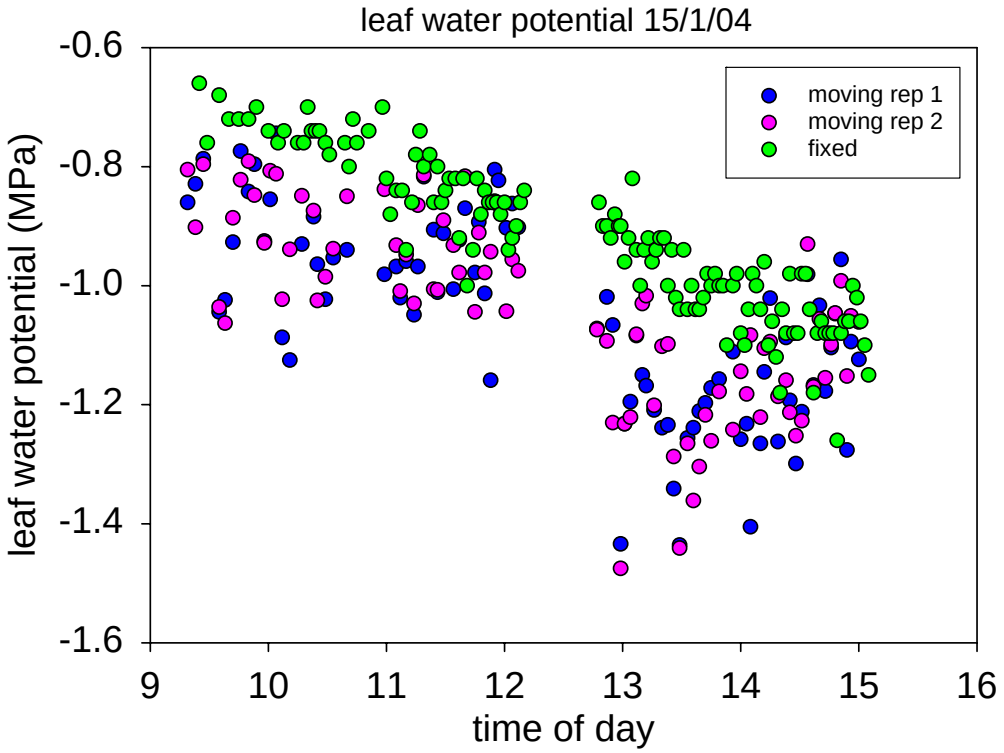


Figure 13. leaf water potential measured 15/1/04 at one point in the vineyard (green symbols) and throughout the vineyard. At each vine throughout the vineyard two readings were taken (blue and red symbols) between 9.00h and 12.00h and then again between 13.00h and 15.00h. The fixed readings were taken near vine 70 shown in Figure 2.

ψL fell during the measurement period (9am to 3pm) in both fixed and moving sets and it is obvious that the variability was much greater in the moving than in the fixed set. On average, the ψL of the fixed vines was higher than that of the vines in most of the rest of the vineyard. This is

consistent with the vineyard soil properties reported by Lanyon and Bramley (2004) as the fixed measurement vine was located in a part of the vineyard with relatively high clay content and high readily available water in the soil profile from 15-45cm. The error components of the fixed and moving data sets are reported fully in Appendix 1and summarized in Table 3 where it can be seen that having removed components of temporal variation the error associated with the data from the vineyard as a whole (Vine) was far greater than that from the data derived from the fixed vine.

Table 3. Estimates of variance components derived from the data in Figure 13.

Source	Estimate	SE
Vine	1.0797	0.2651
Measurement error	0.447	0.056

It is further possible to model the errors expected from the sampling strategies for varying numbers of vines and varying the numbers of readings taken from each vine (Figure 14). For a single vine strategy the error is minimized by increasing the numbers of readings, but there is little improvement after 5 leaves have been measured (red line). However, substantial reductions in error are achieved by increasing the number of vines sampled to say 5 (yellow line) even with a single measurement per vine although a small further reduction could be achieved by taking 2 readings per vine. One strategy in optimizing a sampling scheme is to achieve a given degree of precision for a minimum cost. In the data described in this report there is no estimate of cost other than the time taken for a given set of measurements, and so this will be used as a suitable surrogate. The time taken for various measurements and optimized numbers of vines and measurements per vine to achieve a standard error of no more than 5% of the mean are summarized in Table 19 of Appendix 1. For the data shown in Figure 13 the optimum strategy was to sample 6 vines with a single measurement at each vine at a time cost of approximately 13 minutes. By contrast, stomatal conductance, with a much greater error component, may have required making 2 measurements on each of 57 vines to achieve the set level of accuracy at a time cost of approximately 3.5 hours. Stomatal conductance data will be discussed in detail later.

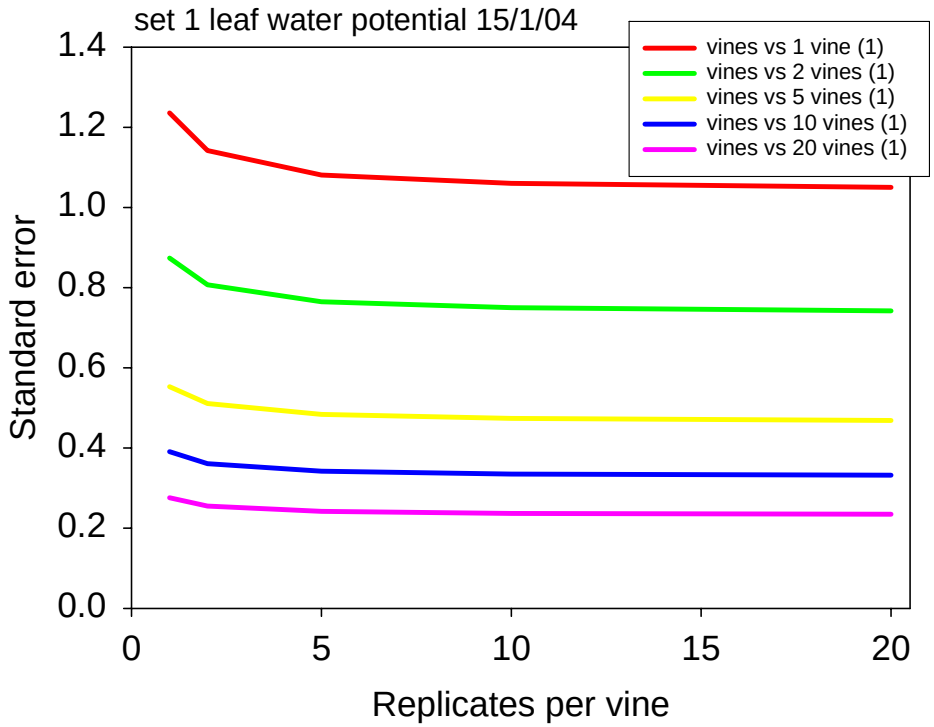


Figure 14. Effect of increasing number of replicates per vine for different numbers of vines

This analysis of spatial and temporal components of variation showed that temporal variation in measures of water potential or stomatal conductance for example can sometimes be quite short term, being influenced by such things as irradiance or wind speed. In theory, measuring stem water potential in place of leaf water potential should eliminate some of these short term fluctuations. Figure 15 shows a comparison of stem and leaf water potentials measured on 8/2/05. Both measures showed the same distinct diurnal trend with a rapid fall during the morning and a minimum achieved around midday. The removal of the temporal component and application of the model to derive minimum time cost to achieve a relative standard error of 5% showed that this could be achieved by measuring stem water potential in about 6.5 minutes compared with 13 minutes for leaf water potential (Table 19, Appendix 1). However, this estimate does not take into account the time expended applying the bags the day before measurement. This would have been considerably more than the time required to measure the same leaves, suggesting that it would be more economic in terms of time expended to measure the leaf water potential of more vines without the time overhead of having to apply the bags. This position is further strengthened by the close correlation between leaf and stem water potential (Figure 16), suggesting that petiole hydraulic conductivity must be high in cv. Shiraz, rapidly equilibrating stem and leaf water

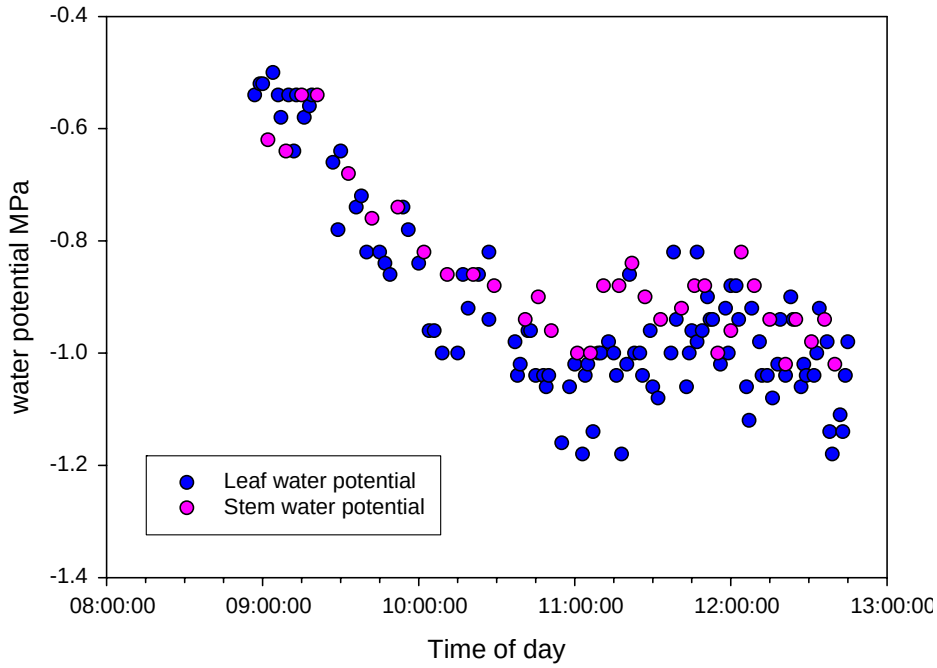


Figure 15. Stem and leaf water potentials measured at Langhorne Creek 8/2/05.

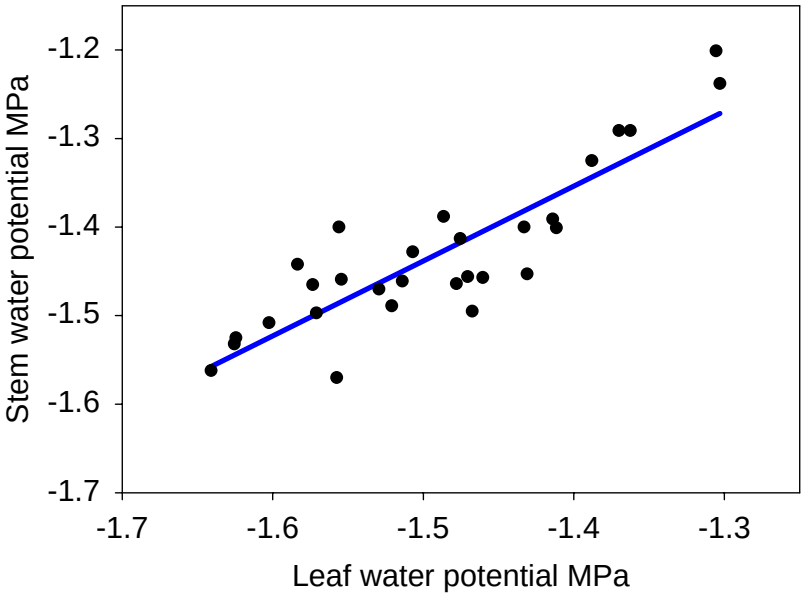


Figure 16. Correlation between leaf and stem water potential. Measurements made 4/2/04. r^2=0.759

potentials. However, we may need to be mindful of the potential for there to be varietal differences in hydraulic conductivity that may impact on this conclusion. For example Schultz (2002) has suggested that differences in the potential for xylem cavitation, and therefore hydraulic conductivity, may be at the root of differences in stress response on Shiraz and Grenache.

Temporal variation in water potential is obviously a complicating factor in the interpretation of leaf water potential data and as we have seen above (Figure 5) pre-dawn leaf water potential (ψpd) can provide an estimate of vine water status that is minimally influenced by time, provided there is equilibrium between canopy and soil water potential, and for normally irrigated vines this appears to be true any time after about 3am and daybreak. An estimate of the spatial variation in ψpd was obtained at the Langhorne Creek site (Figure 17).

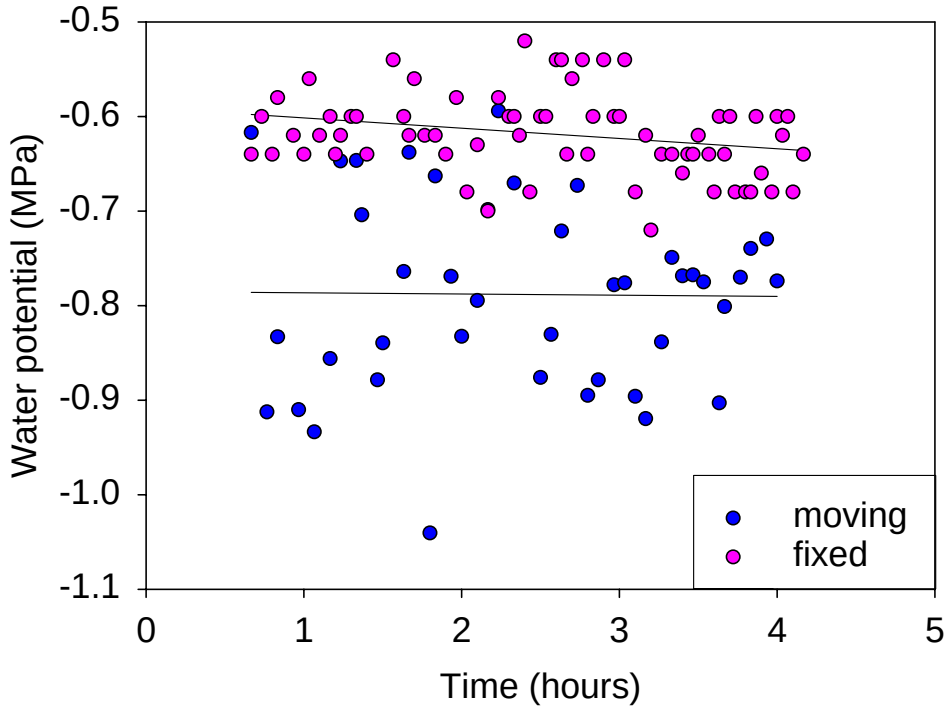


Figure 17. Pre-dawn leaf water potential at the Langhorne Creek site 13/2/04. The fixed measurements were made on 2 vines between marked vines 69 and 70 (Figure 2). Linear regression lines are shown for the moving and fixed data.

The values of ψpd were stable in the fixed position after about 1am. The variation is obviously much greater in the vines sampled through the vineyard than of those of the fixed vine. The stationary readings indicated a weak trend with time. The trend was reflected in the moving

readings as shown in Figure 17. While the trend was the same, the stationary readings were typically 0.17 MPa less negative than the moving values, with respective means being -0.618MPa and -0.788MPa. However, overall there was no relationship between the temporal trend of the stationary readings and the moving readings. The variation can therefore be considered due to spatial and measurement error. The relative standard deviation of the moving and static measurements was 13% and 7% respectively.

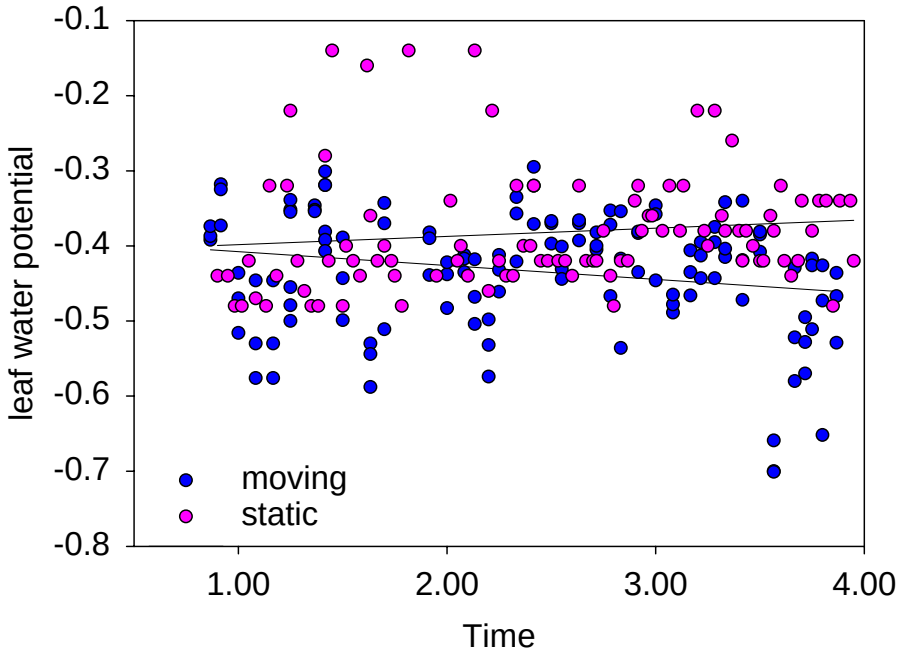


Figure 18. Pre-dawn leaf water potential at the Langhorne Creek site 14/1/05. The fixed measurements were made on 2 vines between marked vines 69 and 70 (Figure 2). Linear regression lines are shown for the moving and fixed data.

The same series of measurements was made in 2005 and again there was no significant trend with time (Figure 18). In contrast to the 2004 measurements the variation among the static and moving data was high and the means similar, with relative standard deviations for the static and moving figures of 20% and 18% respectively. The mean values were much higher than in 2004, possibly reflecting the average temperature of the preceding day (21.5 and 17.6 for 2004 and 2005 respectively) or perhaps when the measurements were made relative to the irrigation cycle.

All leaf water potential measurements reported so far were made with a bomb pressurized with bottled air or nitrogen. There are some OH&S considerations with this type of equipment as the



Figure 19. A, measuring leaf water potential with a manually pressurized bomb. B, using a pressure bomb with a 12V tire pump to measure pre-dawn water potential.

bomb can in theory be pressurized to a limit imposed only by the regulator on the gas cylinder and this may extend to 7 or 8 MPa. At this pressure serious harm could be done to an operator in the event of seal failure. With these problems in mind we have tested several alternatives. Firstly we tested a manually pressurized bomb (Figure 19A). This instrument consists of a simple piston pump attached to the base of the leaf chamber. One stroke of the pump pressurizes the chamber by about 0.05MPa. The operator is required to stop and observe the cut petiole after every stroke or two. The error can be high due to failure to observe the point at which the water potential is balanced by the chamber pressure but more importantly the time taken per reading is about 5-times longer than with a conventional bomb, especially one fitted with a digital readout. However, with careful use it is capable of precision of a similar order to the conventional instrument. We also experimented with a conventional bomb pressurized with a 12V tyre pump (Figure 19B). This has the disadvantage that it is limited to a pressure of 1MPa but this is quite adequate for pre-dawn water potential measurements where the water potential would not be

expected to be below -1.0MPa. It would not be suitable for midday leaf water potential measurements which may require pressures as high as 2MPa. The speed of measurement and the precision was no different to a conventional bomb.

Stomatal conductance

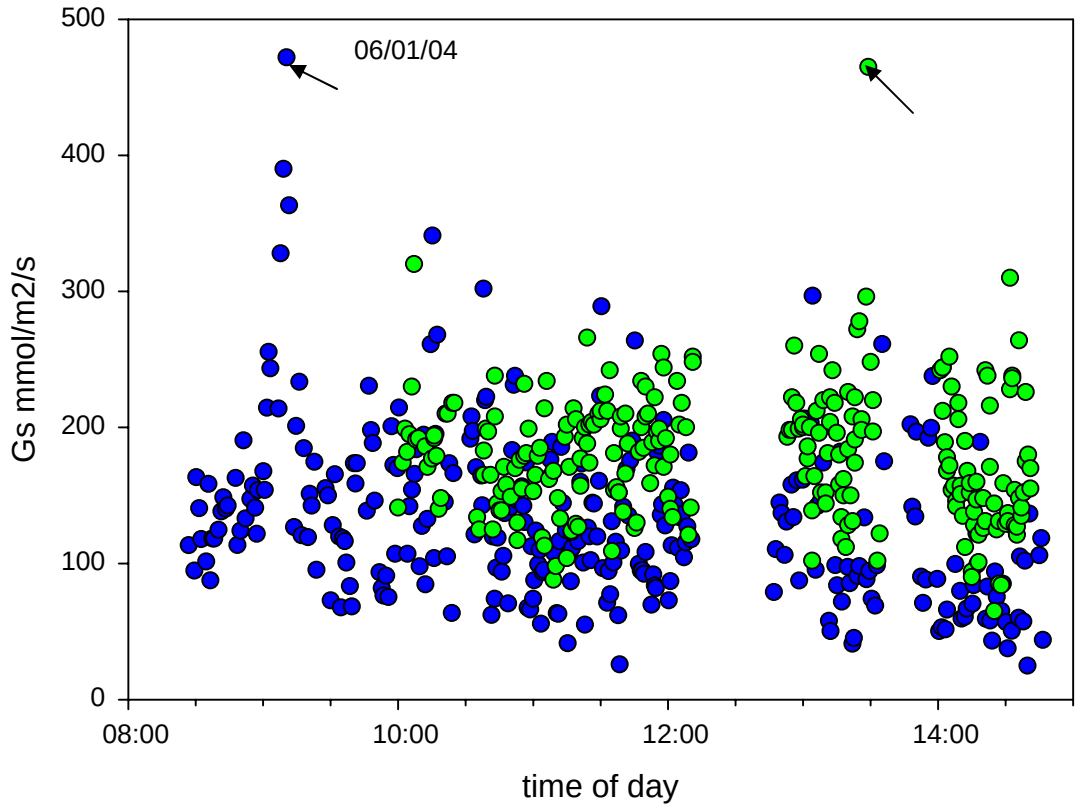


Figure 20. Stomatal conductance (Gs) measured at Langhorne Creek 6/1/04. Static vines (green) measured were between vines 69 and 70 (Figure 2). The vines throughout the vineyard (blue) were measured twice, between 08:30and 12:00 and then again between 12:45 and 15:00. Outliers (arrowed) were removed from the analysis.

There was no significant trend with time in the static measurements but there was a slight trend for a reduction in Gs as the day progressed in the moving measurements (r^2=0.1125). This may have been biased by the fact that the moving measurements started a little earlier than the static ones and during this early period there were a number of high readings. Analysis of the moving readings showed that the temporal trend had no significant effect on the readings. Estimates of between vine error and measurement error were used to model the most appropriate sampling

strategy (Figure 21). Unlike measurements of leaf water potential error estimates were very high, leading to a sampling strategy calling for 57 vines with 2 samples per vine at a time cost of nearly 4 hours (Table 19, Appendix 1).

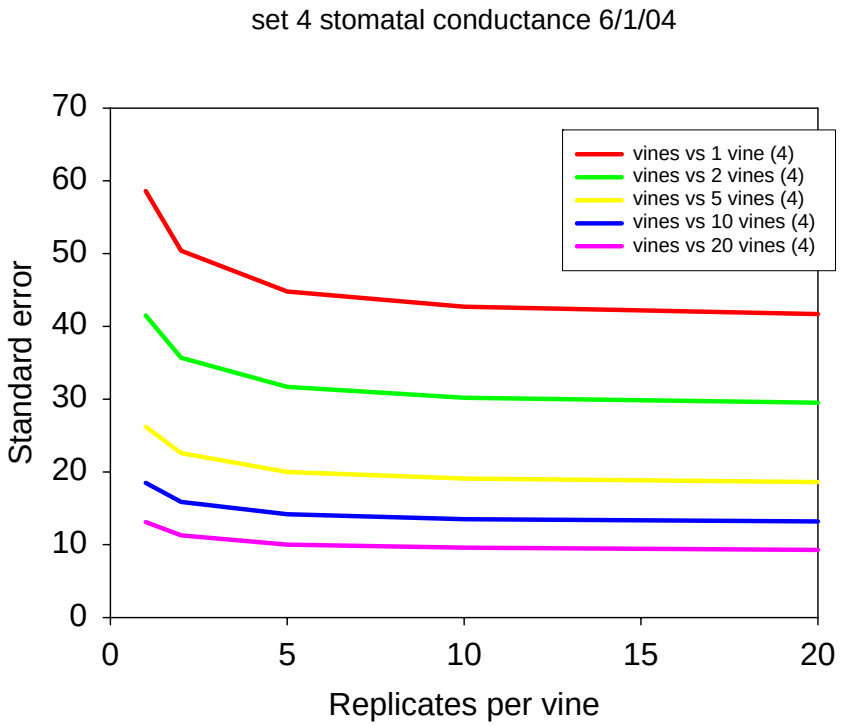


Figure 21. Effect of increasing number of replicates per vine for different numbers of vines.

These measurements were repeated later in the season (Figure 22) with similar results. There was little temporal trend evident in either the static or moving measurements. Thus the moving readings can be described as random fluctuations about a mean, where the mean is 189 with a variance of 9839 ± 674 (SE = 99). The analyses indicate that in this example there is no need to sample a series of plants. Rather the standard error of the measure can be estimated from the equation SE = 99/sqrt(*r*), where *r* is the number of replicates.

The optimal strategy is therefore to take many readings on each vine, and to visit few vines (Appendix 1).

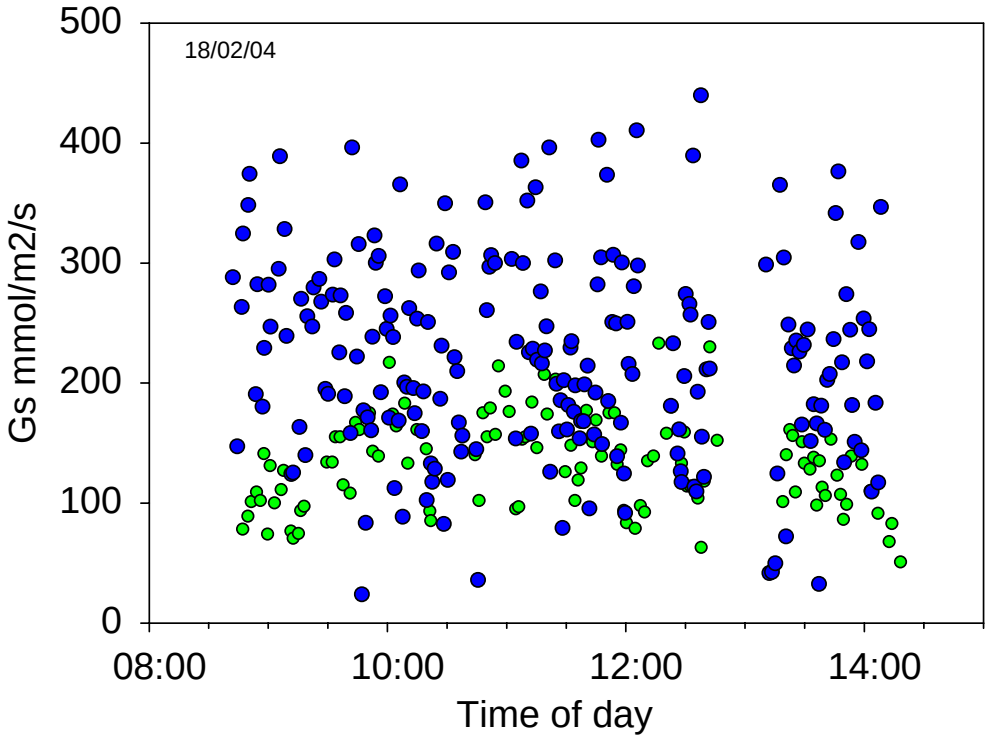


Figure 22. Stomatal conductance (Gs) measured at Langhorne Creek 18/02/04. Static vines (green) measured were between vines 69 and 70 (Figure 2).

Because of this apparent contradiction between these two data sets the experiment was repeated early in the next season (Figure 23). There was strong linear trend towards lower conductance later in the day in both moving and static measurements. These linear regressions accounted for 40% and 70% of the variation respectively. For the moving measurements the optimum sampling strategy to obtain a relative standard error of 5% was to sample 32 vines with 4 measurements per vine for a time cost of just over 3 hours (Table 19 Appendix 1).

These three sets of stomatal conductance data are characterized by a high degree of variability, requiring many measurements to be made to reduce the error. The data shown in Figure 22 and to a lesser extent in Figure 20 are not what would have been expected in that there was little or no temporal trend. Figure 23 is more representative of what would

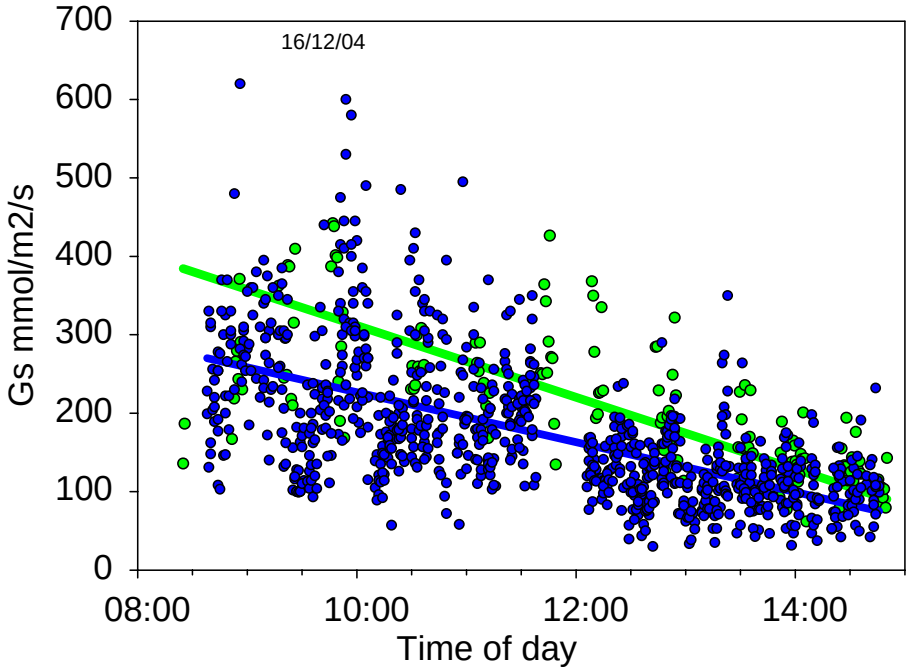


Figure 23. Stomatal conductance (Gs) measured at Langhorne Creek 16/12/04. Static vines measured were between vines 69 and 70 (Figure 2). Linear trends in moving (blue) and static (green) measurements are shown.

have been expected, with a decrease in conductance through the day. The reasons for a lack of a trend in 2 out of the 3 data sets are unclear. The weather during the measurement day and for three days prior to the measurement day may give some clue. Evaporation for these 4 days was 2.7mm, 5.0mm and 1.9mm respectively. The more stressful conditions of the February measurements may have resulted in differential expression of stress responses through the vineyard. Although the measurement day was not particularly stressful, with maximum air temperature of about 26^{o}C there had been a 44 ^{o}C day on February 14th, 2 days before the measurements were made.

Leaf and canopy temperature

It has been recognized for many years that transpiration results in evaporative cooling of the leaf lamina and that this depression in temperature, relative to ambient, can be a useful indicator of plant performance. Thus, leaf temperature can be a surrogate for stomatal conductance. In the section above we have shown that stomatal conductance is an extremely variable parameter, requiring many measurements to obtain a reasonably acceptable standard error. By measuring canopy temperature it is possible to integrate a large number of leaves into the measurement, thus effectively reducing the error associated with leaf to leaf or vine to vine variation. The idea of using canopy temperature as a measure of vine performance is also attractive because the infrared thermometer equipment required is relatively inexpensive and can be operated by relatively unskilled personnel, although data interpretation may present some difficulties. One of the potential problems with using infrared thermometry to measure canopy temperature is that it is non-selective. That is, when the instrument is pointed at the canopy it will measure not only the leaves but also things like trellis posts, soil or sky appearing through gaps in the canopy. Readings from these items can seriously bias the average temperatures ascribed to the canopy since their temperature can be significantly above or below that of the leaves. Trellis posts and soil can reach temperatures of 60^{o}C and clear sky may register as -40^{o}C. This part of the project sought to assess the feasibility of using canopy temperature, measured with an infrared thermometer, as an alternative to other measures such as stomatal conductance and to compare data from simple infrared thermometers with date from a thermal imaging system.

We have previously validated IR measurements of grape leaf temperature by simultaneously measuring a patch of canopy of about 1 m^2 with an infrared thermometer and a large number of very fine thermocouples attached to the underside of the leaves. Figure 24 shows the relationship between several thousand such measures. There was a very close 1/1 relationship between the IR and thermocouple measurements, showing that IR thermometry can be used with some confidence as a measure of true leaf temperature. As a further test of the ability of infrared thermometry to differentiate grape canopies on the basis of their transpiration rates we simultaneously measured canopy temperature and stomatal conductance of Shiraz and Grenache vines receiving either normal or RDI irrigation. The RDI irrigation regimes resulted in significantly lower stomatal conductance and higher canopy temperatures (Table 4).

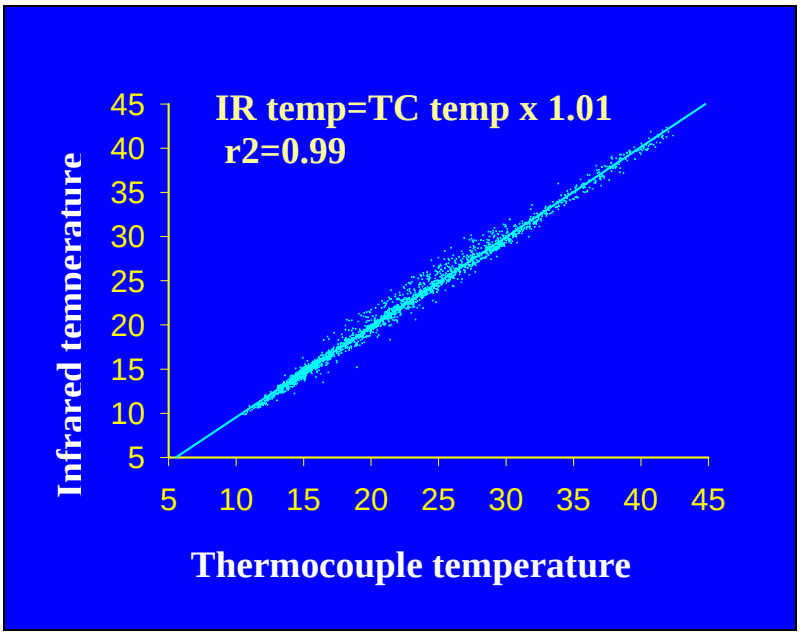


Figure 24. Relationship between leaf temperature measured with an infrared thermometer and thermocouples pressed against the underside of the leaves.

Table 4. Stomatal conductance and canopy temperature of Shiraz and Grenache under control or RDI irrigation regimes. Figures are means and standard error. Different letters within a row signify significant differences (P<0.001)

	Control		**RDI**	
	Conductance mmol/m^2/s	Temperature ^oC	Conductance mmol/m^2/s	Temperature ^oC
Shiraz	257±13 a	30.32±0.25 x	59±4.9 b	32.86±0.25 y
Grenache	197.4±14.2 a	27.96±0.18 x	114.1±12.9 b	29.19±0.04 y

These IR readings were made with a hand-held thermometer (Everest) and some effort was made to avoid non-canopy components such as posts and gaps, thus reducing the error which may be associated with the inclusion of these in the mean value. As a means of further validating these data we were able to use a thermal imaging camera to differentiate canopy and non-canopy components.

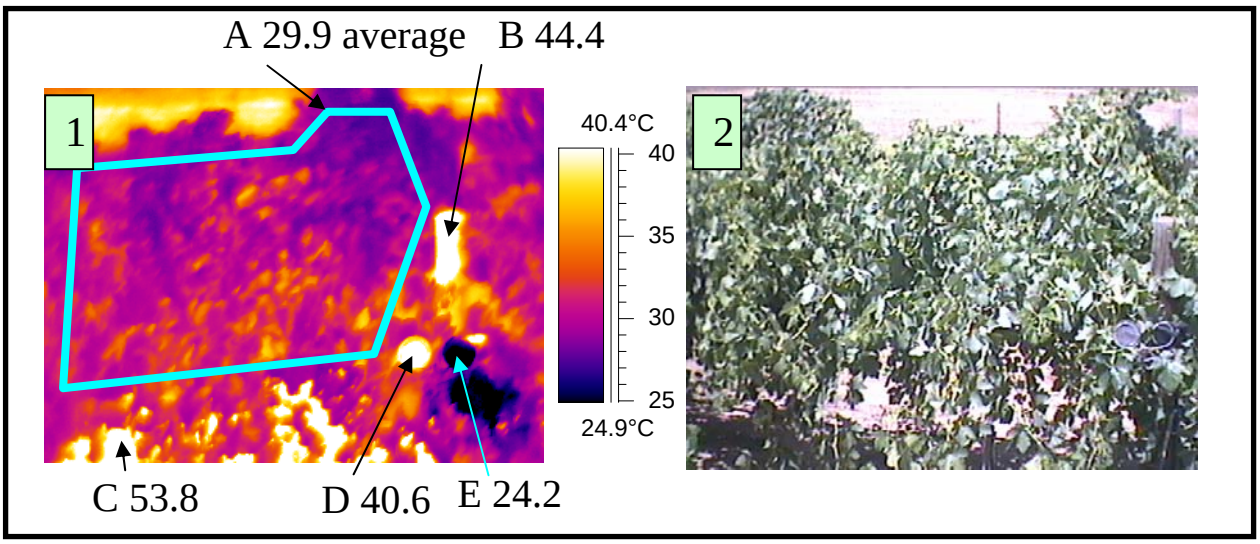


Figure 25

1. Thermal image of a section of Shiraz canopy. A, average temperature of leaves within the irregular figure; B, trellis post; C, gap in canopy; D, dry reference surface; E, wet reference surface
2. Real colour image of the same portion of canopy
3. Enlargement of reference surfaces seen on the right of (2). These comprised 2 filter paper disks, one dry and one kept moist by spraying with water and 2 reference leaves below, the left one covered in petroleum jelly to prevent transpiration and the other kept moist by spraying with water.



The real colour image shows a typical Barossa canopy with gaps allowing sight of the ground of the inter-row and part of a trellis post. The thermal image of this canopy illustrates the temperature of these components. The trellis post registered 44.4^{o}C, gaps in the canopy over 50 ^{o}C. The wet and dry reference leaves registered 24.2 ^{o}C and 40.6 ^{o}C respectively, illustrating the power of evaporation to cool a leaf. The average temperature of the intact canopy (1A) was 29.9 ^{o}C.

Jones (1999) has shown that by using the temperatures of the canopy and reference surfaces it is possible to derive a figure (Ig) that is an index of canopy conductance.

$$Ig = (T_{dry} - T_{canopy}) / (T_{canopy} - T_{wet})$$

Where:

Ig = canopy conductance index

T_{dry} = temperature of dry reference surface

T_{canopy} = temperature of canopy

T_{wet} = temperature of wet reference surface

Using this formula it is possible to calculate Ig for many thermal images taken of canopies of irrigated and RDI vines. Figure 26 shows an example of the areas of canopy used in typical assessments.

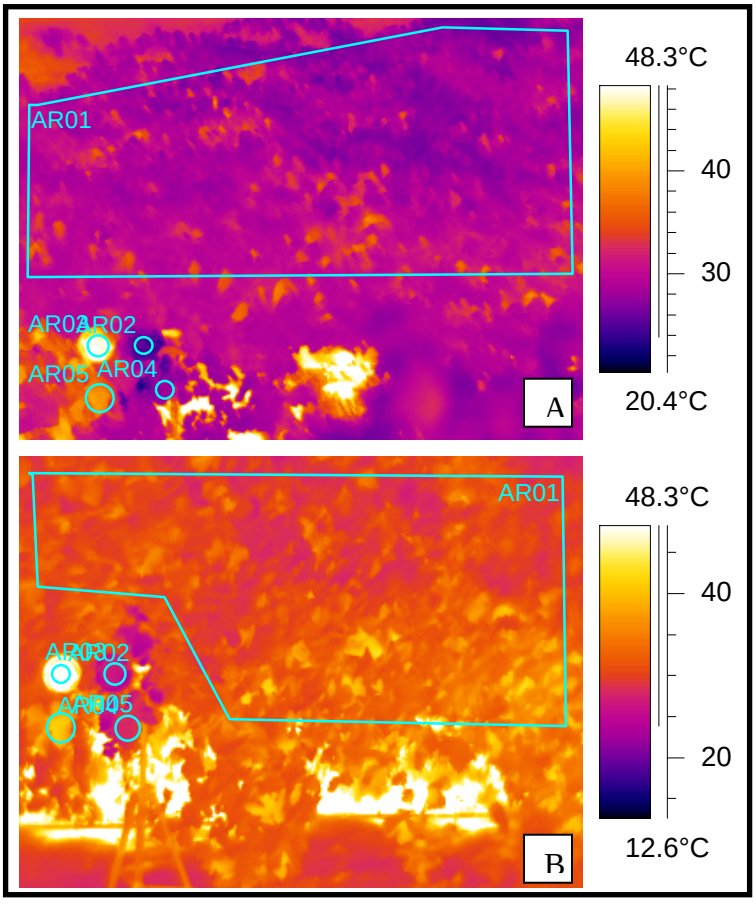


Figure 26.

A. Thermal image of irrigated Shiraz canopy showing areas of canopy and reference surfaces selected for temperature measurement.

B. Thermal image of RDI Shiraz canopy.

Images made on the sunny side of the canopy 20/01/2004

Mean sun-exposed and shaded canopy temperatures were significantly affected by the irrigation treatment (Table 5). Values of Ig derived from both filter paper and leaf references were also significantly different in the sun exposed situation although only the values derived from the leaves were significantly different on the shaded side of the canopy.

	Canopy Temperature	Ig $_{disk}$	Ig $_{leaf}$
Irrigated (20/1/04)	29.85	5.01	4.91
RDI (20/1/04)	31.00	4.01	2.63
P, n=6	0.026	0.025	0.034
Irrigated (22/1/04)	18.15	3.55	0.68
RDI (22/1/04)	18.85	3.04	0.27
P, n=9	0.0069	ns	0.0076

Table 5. Canopy temperatures and derived values of Ig for sun exposed (20/1/04) and shaded (22/1/04) canopies of Shiraz vines at Nuriootpa. Ig $_{disk}$ values calculated using measures of canopy temperature and temperature of reference filter paper disks. Ig $_{leaf}$ calculated using canopy temperature and temperature of reference leaves.

These analyses required an expensive thermal imaging camera and considerable time investment for data processing. We have shown above that it is possible to detect temperature differences between canopies of vines at different stages of an irrigation cycle using an inexpensive hand-held infrared thermometer and it would be preferable if such an instrument could be used for the calculation of Ig. However, we have drawn attention above to the errors that could be introduced into measurements of canopy temperature by non-vine components such as trellis posts, gaps in the canopy and sky. To assess the possible effects of these non-vine components and to simulate the measurement of canopy temperature with a hand-held thermometer we used multiple thermal images and derived the mean temperature of a simple rectangle superimposed on the canopy as would occur if the image field of an IR thermometer were passed over the canopy. We included

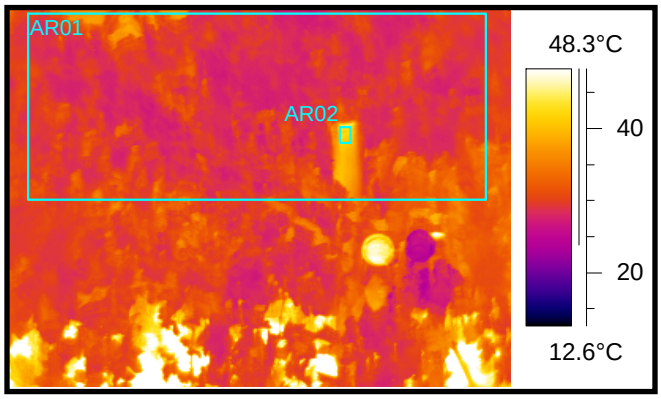


Figure 27. Thermal image of Shiraz canopy including a trellis post. The mean temperature of the indicated rectangle was 29.8°C, including the trellis post which indicated by itself 41.9°C

all non-vine components such as gaps and trellis posts (Figure 27). The mean canopy temperature of the irrigated vines was 29.76^{o}C and of the RDI vines 30.42^{o}C for 21 comparisons (P=0.022, analysis on reciprocal transformed data). Thus, a simulated measurement with a hand-held infrared thermometer, including non-vine components yielded a significant difference in temperatures between canopies from irrigated and RDI vines, in much the same way as the actual measurements made with an infrared thermometer (Table 4). The effect of the non-vine components was minimal because even though their temperatures are considerably elevated compared with the canopy, their area is small and their contribution to temperatures averaged over the whole canopy were small. In order to obtain meaningful temperature data from large areas of canopy we have tested a system where an infrared thermometer is held by an operator in or on a vehicle. In practice we have used a quad bike or a utility (Figure 28).



Figure 28. Measuring canopy temperature with an infrared thermometer.

The output from the thermometer is captured in a data logger at a rate of 4 readings per second. The vehicle drives slowly past the vines with the operator pointing the thermometer at an angle of about 30^{o} to the vine row. It was found that, compared with measurements made with the thermometer pointed at 90 degrees to the canopy, this eliminated some of the error caused by gaps in the canopy as these were not visible at this angle. Temperatures of wet and dry filter paper references and ambient temperature inside a small aspirated enclosure were measured with

thermistors and also logged with the same data logger. In this way very large data sets can be accumulated and many meters of vine row can be measured in a short time. The relationship between canopy temperature and stomatal conductance was determined in a number of ways at a number of locations. At Nuriootpa varying amounts of water were applied to a row of Shiraz and a row of Cabernet Sauvignon. Each row contained approximately 60 vines. One third of the vines at the western end of each row received irrigation on a daily basis (wet treatment), the middle one third received water until the end of December and were unirrigated thereafter (drying treatment) and one third of the vines on the eastern ends of the rows had remained unirrigated for the whole of the 2004/2005 season until after the present experiments were completed (dry treatment). 140mm of rain fell from the beginning of November 2004 to the end of January 2005. Of this total, 26mm fell on January 19th, five days before these experiments commenced. None of the vines showed any signs of water stress, but the dry vines were noticeably less vigorous than the wet and drying vines.

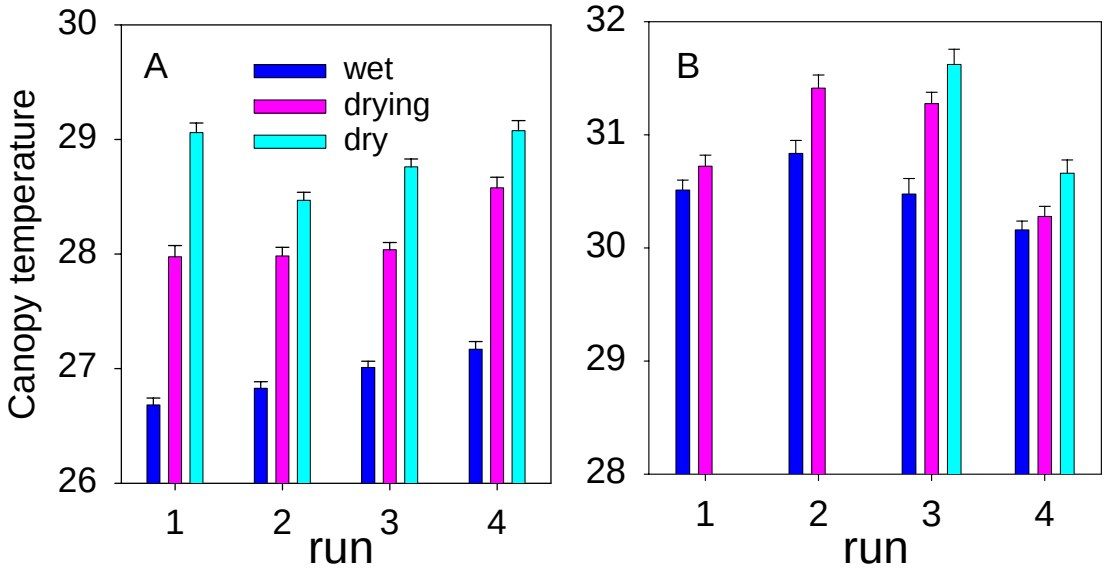


Figure 29. Canopy temperature measured with an Everest infrared thermometer. Each vine row containing the three irrigation treatments was measured four times (run 1-4). Each set of 4 measurements took between 2 and 3 minutes.
A. Shiraz south facing canopy. B. Shiraz north facing canopy.
Instrument error caused some data loss in B. Bars are standard errors (n=120).
Measurements made between 11.50 and 12.20 24/01/2005

The temperature of the fully irrigated vines was significantly lower than that of the drying vines which was, in turn, significantly lower than that of the dry vines on the southern (shaded) side of the canopy (Figure 29A). Similar results were obtained for each of the 4 runs. On the sunlit side

of the canopy temperatures were higher but differences between treatments were less marked but the same trend existed, with the canopy of the irrigated vines cooler than the drying and dry vines.

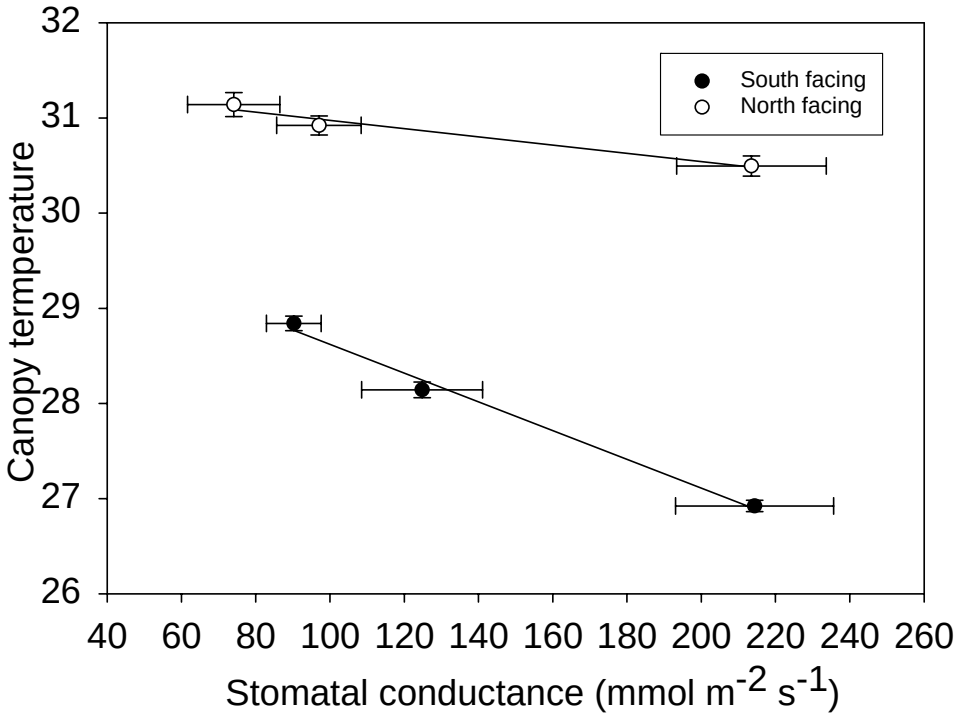


Figure 30. Relationship between stomatal conductance and canopy temperature of Shiraz vines.

There was a linear relationship between stomatal conductance and canopy temperature (Figure 30). The slope of the relationship was greater for the south facing side of the canopy. A canopy

conductance index (Ig) was calculated as described above for each data set, using temperatures of

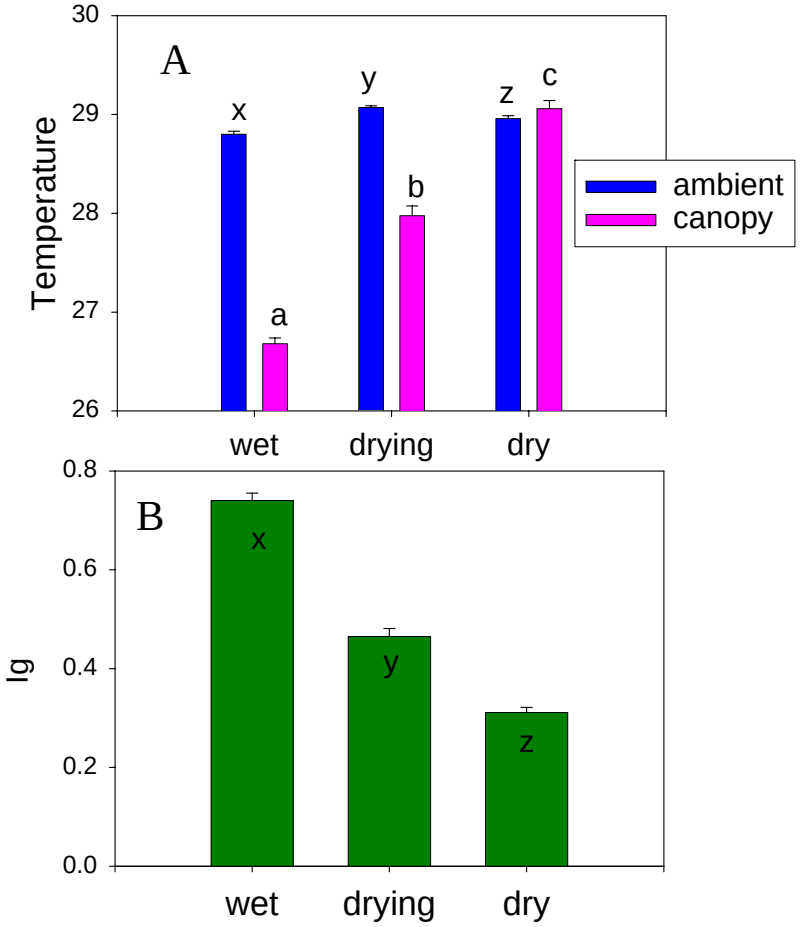


Figure 31. A. Temperature of south facing canopy of Shiraz vines, ambient temperature and B, canopy conductance index (Ig). Bars are standard errors (n=120) and different letters associated with each data type indicate differences significant (P<0.01)

wet and dry reference surfaces and canopy temperature. Representative data are shown in Figure 31. Ambient air temperature is also shown on this figure. The temperature of the canopy of wet and drying vines was lower than ambient but that of the dry vines was very close to ambient. Essentially the same results were obtained with Cabernet Sauvignon, although the differences in canopy temperature between treatments were sometimes smaller (Figure 32), reflecting the smaller range of stomatal conductances (Figure 33). Figure 33 also illustrates the error associated with each of these measurements. The standard error bars on the conductance measurements are

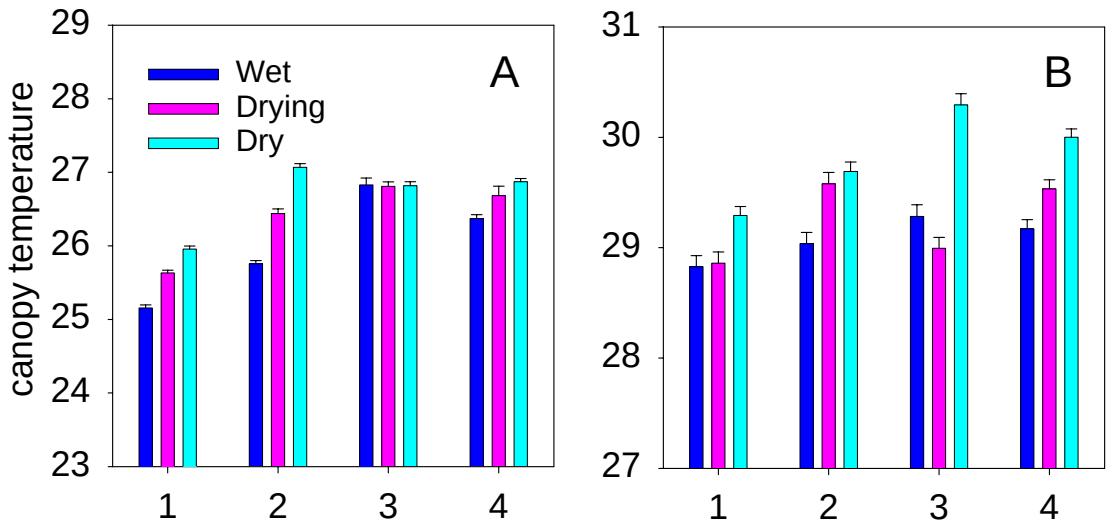


Figure 32. Canopy temperature measured with an Everest infrared thermometer. Each vine row containing the three irrigation treatments was measured four times (run 1-4). Each set of measurements took between 10 and 20 minutes.
A Cabernet Sauvignon south facing canopy. B. Cabernet Sauvignon north facing canopy. Bars are standard errors (n=120). Measurements made between 11.10 and 11.30 24/01/2005

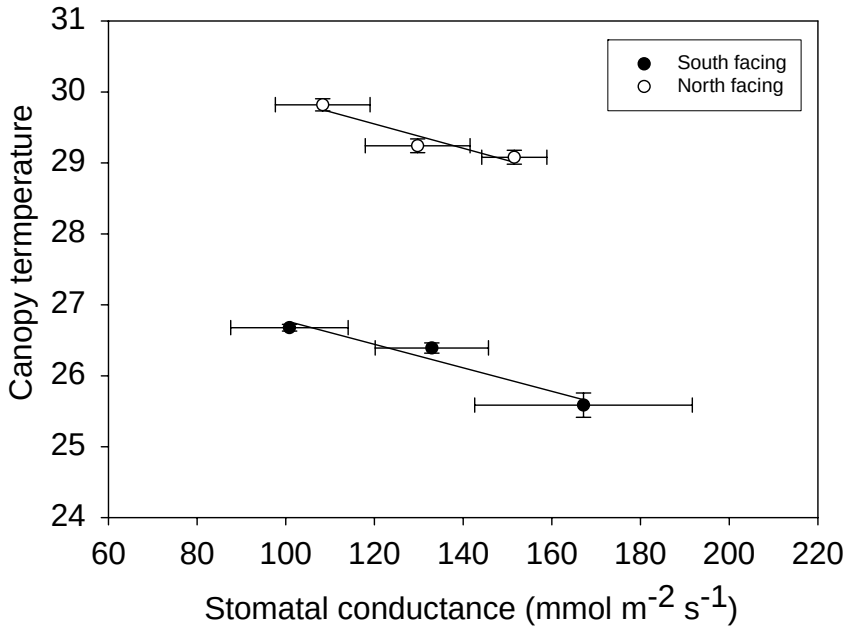


Figure 33. Relationship between stomatal conductance and canopy temperature of Cabernet Sauvignon vines.

much greater than for the canopy temperature measurements. This is the result in part of the relatively small number of conductance measurements that can be made and the high leaf to leaf variability of this parameter. This was well illustrated in the section above on conductance measurements. Nevertheless, the strong linear relationships between conductance and canopy temperature suggests that the latter will be a good surrogate for rapid assessment of the former. Canopy temperature is determined by the balance between incoming radiation, ambient air temperature and the cooling effect of leaf transpiration. The latter is determined by stomatal conductance and current evaporative conditions (air temperature, humidity, wind speed etc). However, in grapevine, transpiration is not very tightly coupled to evaporative conditions because of rapid stomatal adjustments. This has been graphically shown by Lu et al (2003) and Yunusa et al (2000) who showed that transpiration was changed little by large changes in VPD. This can also be concluded from sap flow data shown in this report. Thus a consideration of canopy temperature and ambient temperature alone may provide a reasonable estimate of grapevine transpiration. This can be further refined by the calculation of a canopy conductance index as described by Jones et al. (2002) that takes into account the current environmental conditions that may be influencing transpiration.

To further test the robustness of the relationships between canopy temperature, calculated conductance index (Ig), stomatal conductance and ambient minus canopy temperature difference one panel (3 vines) of fully irrigated Shiraz vines was sprayed with abscisic acid (ABA) to induce stomatal closure. For comparison, several panels of vines to the east of the sprayed vines and several to the west were also measured. The vines were measured as soon as the spray had dried from the leaves. The temperature of the ABA-sprayed leaves was significantly higher than either of the control panels (Figure 34 A). The temperature of the control panel to the west of the ABA-sprayed vines was significantly higher than the other control panel and this was reflected in the stomatal conductance figures (Figure 34 C). Calculated Ig values changed in concert with the conductance and ambient minus canopy temperature differences (Figure 34 C grey bars). These results are in agreement with previous measurements reported above, although the absolute values of Ig differed significantly. The higher stomatal conductances in this experiment were chiefly responsible for this. Further experiments would probably be required to more clearly define a minimum value of Ig that could be used to trigger an irrigation event. Nevertheless, the method appears to be capable of robustly estimating canopy conductance with relatively small error when compared with other methods such as measurement of stomatal conductance or leaf water potential. A major factor contributing to this reduction in error is the ability to make a large

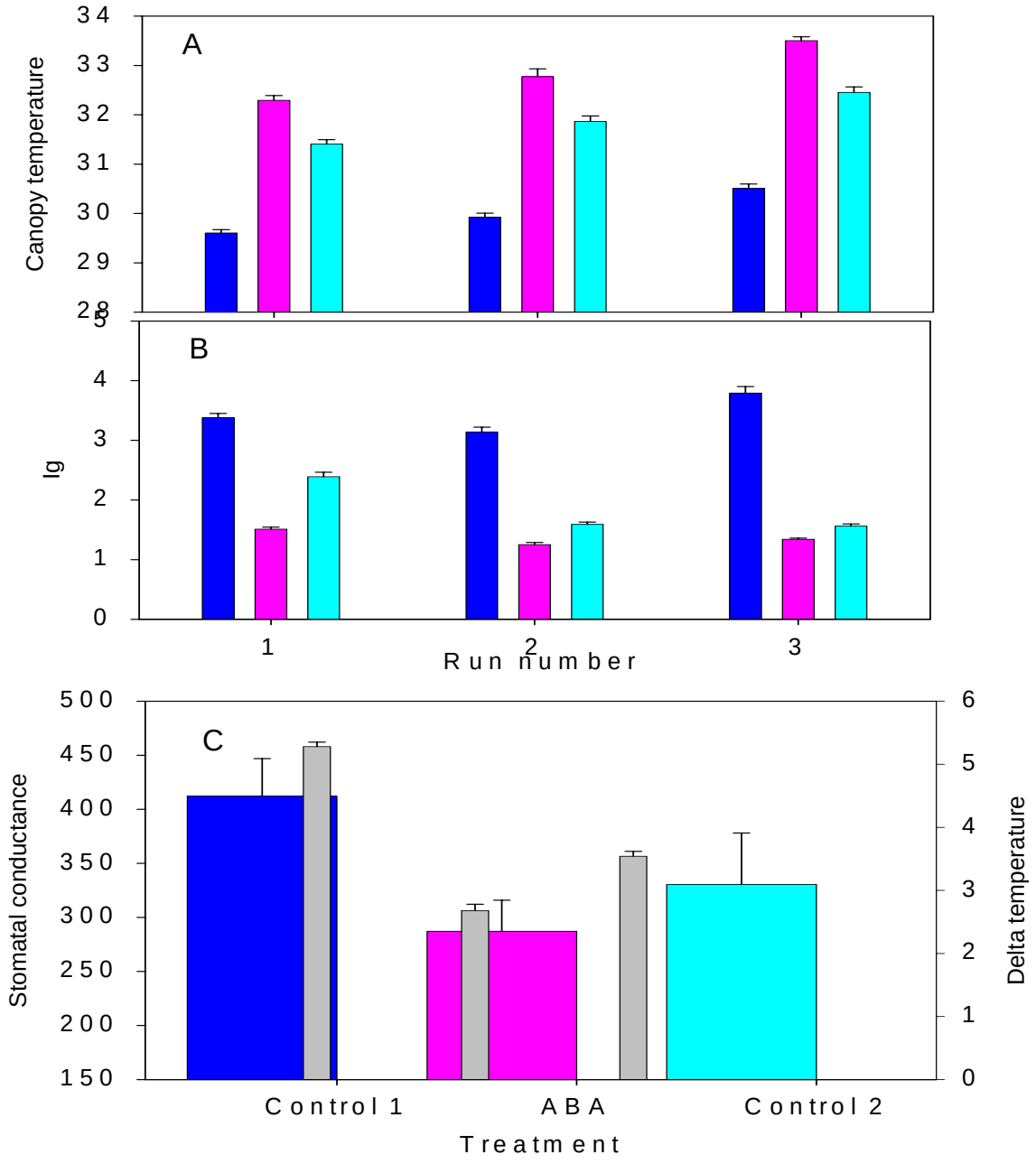


Figure 34. Effect of spraying leaves with abscisic acid (400 mg/L). A. Canopy temperature of fully irrigated Shiraz vines measured with Everest infrared thermometer. Treatments were: unsprayed vines to the east of sprayed vines (dark blue), ABA sprayed vines (pink), unsprayed vines to west of sprayed vines (pale blue). B. Conductance index (Ig). C. Stomatal conductance for the three treatments and the respective canopy-ambient temperature differences (grey bars).

number of measurements in a short period of time, thus eliminating many of the problems associated with temporal changes in plant physiological parameters that can occur in a relatively short time scale.

The assessment of Ig requires the measurement of the temperatures of wet and dry reference surfaces. In these experiments we used black filter paper disks but real vines leaves have also been used with some success (Jones et al., 2002). In the case of the vine leaves the “dry” leaf was covered in petroleum jelly to prevent evaporative cooling and the wet leaf was kept so by spraying with water. This arrangement obviously imposes some limitations on the practical application of this technique for the assessment of vine performance. Nevertheless, Ig does seem to be a robust surrogate for the direct measurement of conductance. A more readily measured parameter that is also related to vine transpiration is the depression of leaf temperature, relative to ambient. Consequently a relationship between canopy temperature depression and Ig was sought. Figure 35 shows this relationship for a number of measurements of these parameters on Shiraz vines showing a wide range of stomatal conductance.

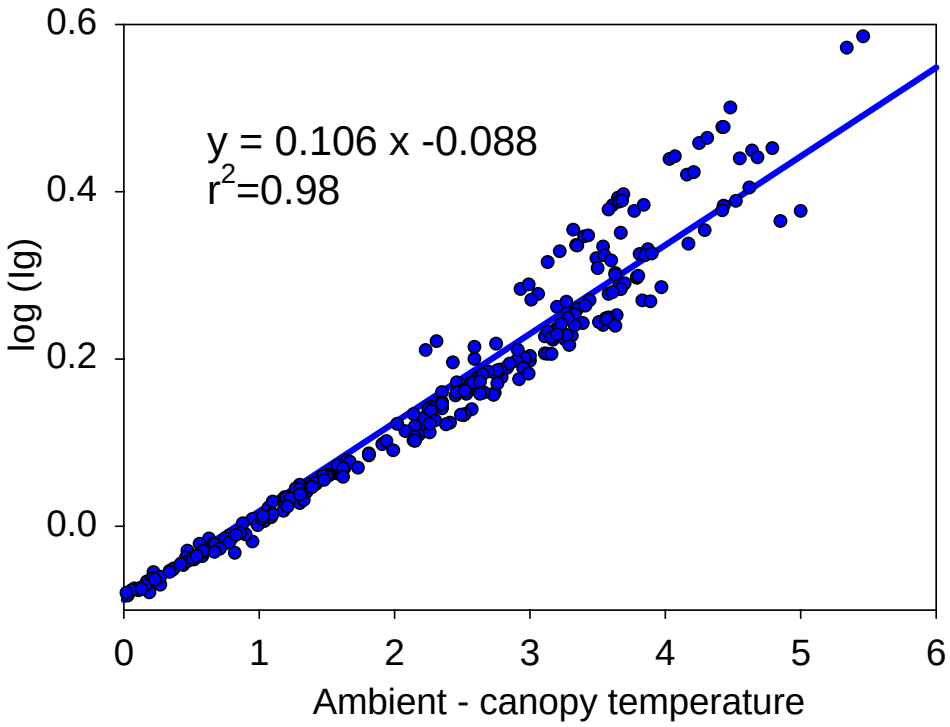


Figure 35. Relationship between canopy temperature depression and conductance index (Ig) for Shiraz.

It may therefore be possible to find some unique relationship between some easily measured variable and canopy conductance.

Most of the data collected for the preparation of this section of the report were consistent and predictable (it should be noted that the data presented here represents only a small proportion of the total available, perhaps as little as 10%), however not all of the data were so easy to interpret. For example, one set of data collected from the north face of a Shiraz canopy on Jan 24th at Nuriootpa illustrates some of the potential problems encountered in this work.

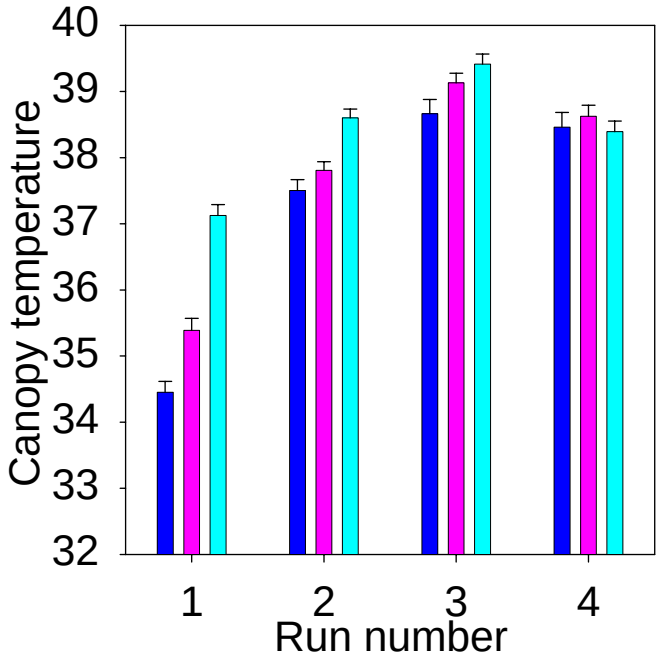


Figure 36. Canopy temperature of north-facing Shiraz canopy. Irrigation treatments are as described above. Measurements made with a Cole Palmer infrared thermometer between 14.45 and 15.00 24/01/2005.

Within each of the first three runs the relative canopy temperatures were as seen previously, with the fully irrigated vines (dark blue) having lower canopy temperature than either the drying (pink) or dry (light blue). However, the temperatures increased with each run. In the fourth run there were no temperature differences between the treatments. There could be a number of explanations for these anomalies. For example:

- The increase in temperature over the first three runs could have been the result of increasing radiation over the few minutes it took to make the measurements, due perhaps to variable cloud cover. This was unlikely as the weather station situated within a few meters of the vines and logging at 1 minute intervals, recorded a cloudless day although at the time the measurements were made (14.45 to 15.00) radiation was beginning to

decline after its maximum. The recorded canopy temperatures would be more consistent with an increase in radiation, not a decrease.

- Stomata frequently close partially in the middle of the day. This would cause a general increase in canopy temperatures but this would have been unlikely to have occurred over such a short time span and so consistently throughout the canopies.
- The measurements were made with a Cole palmer infrared thermometer. This was identical to the Raytek Ranger MX instrument described in the materials and methods section. The Cole Palmer and Raytek instruments had a much narrower field of view than the Everest instrument used for most of the measurements. This would result in slightly more variability in recorded temperatures as gaps in the canopy would be seen "in total" instead of being integrated with surrounding foliage by the wider field of view of the Everest instrument. This is evident from the slightly greater relative standard deviation of measurements made with the Cole Palmer or Raytek when compared with the Everest. However, this is unlikely to have caused the anomalies described here. A more likely cause would have been a systemic instrument fault caused perhaps by a low battery voltage. We saw no other evidence for this however and readings taken with the same instrument on another part of the canopy appeared to be fairly consistent and in agreement with earlier reported data.

Having considered all these possibilities no clear reason for the errors are apparent. There were no major changes in ambient conditions of radiation or humidity during the measurement period although close inspection of the weather station data did show a rapid increase in air temperature and wind speed during the period of measurement (Figure 37) although neither rise was extreme. Such short term plant-based changes seem unlikely so we are left with the possibility that there was a temporary adjustment in instrument sensitivity or that minor changes in ambient conditions are having a disproportionate effect on canopy temperature. Instrument error seems the most likely cause as the difference between indicated canopy temperature and ambient air temperature recorded by the weather station may be incorrect. The latter showed between 31.3^{o}C and 32.5 ^{o}C while the canopy temperature was between 34.5 ^{o}C and 39 ^{o}C for the wet vines, which should have had a temperature lower than ambient. Clearly, the reasons for these anomalies need further investigation.

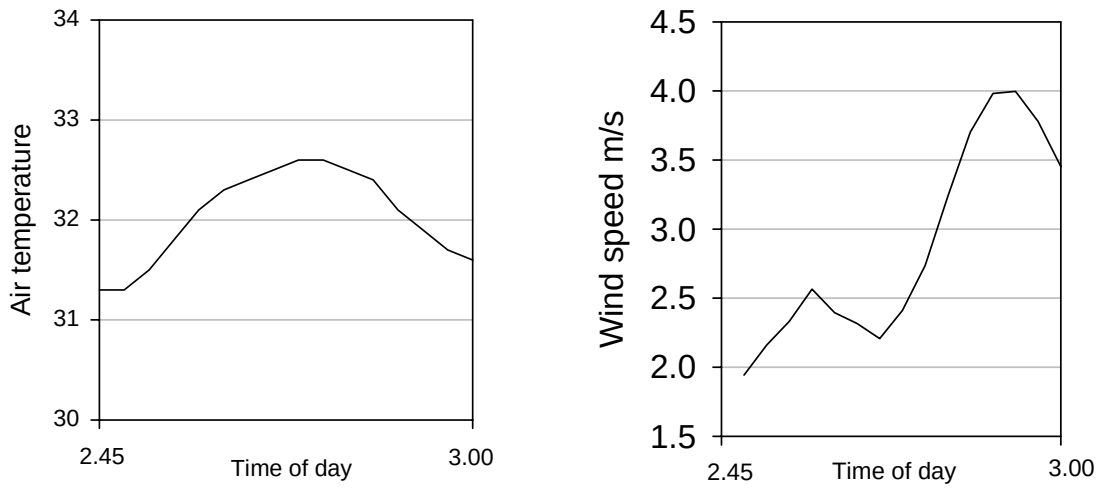


Figure 37. Air temperature and wind speed during the time that measurements shown in Figure 36 were made.

Measurement of sap flow

There are a number of methods available for measuring sap flow in trunks and canes of grapevines. We have used heat pulse sensors. Each sensor has three 2mm stainless steel probes that are inserted into holes drilled into the vine trunk (Figure 38). The upper and lower probe contain thermistor temperature sensors and the centre probe contains a heating element. A measurement cycle is initiated by energising the heater for about 2 seconds. Heat from the probe passes through the wood and is detected by the thermistor sensors. The time taken for the probe temperatures to return to the pre-heat pulse condition is influenced by sap flow; the higher the sap flow, the quicker the re-equilibration. Sap flow can then be calculated from this time and various constant values that describe the size of the trunk and the properties of the wood.



Figure 38. Heat pulse sap flow sensor.

Data from the sap flow sensors are typically logged at 30 minute intervals. Some representative data from a single day are shown in Figure 39. The low rate of sap flow during the night may or may not be real. No “time out” value was elected in processing the data. If a timeout value is set it can mask real changes that do occur in sap flow at night. Under conditions of low VPD nocturnal sap flow tends to be close to zero but during nights of high temperature and VPD sap flow can

increase significantly. Sap flow increases rapidly after first light and reaches a maximum at about 3pm (Figure 39).

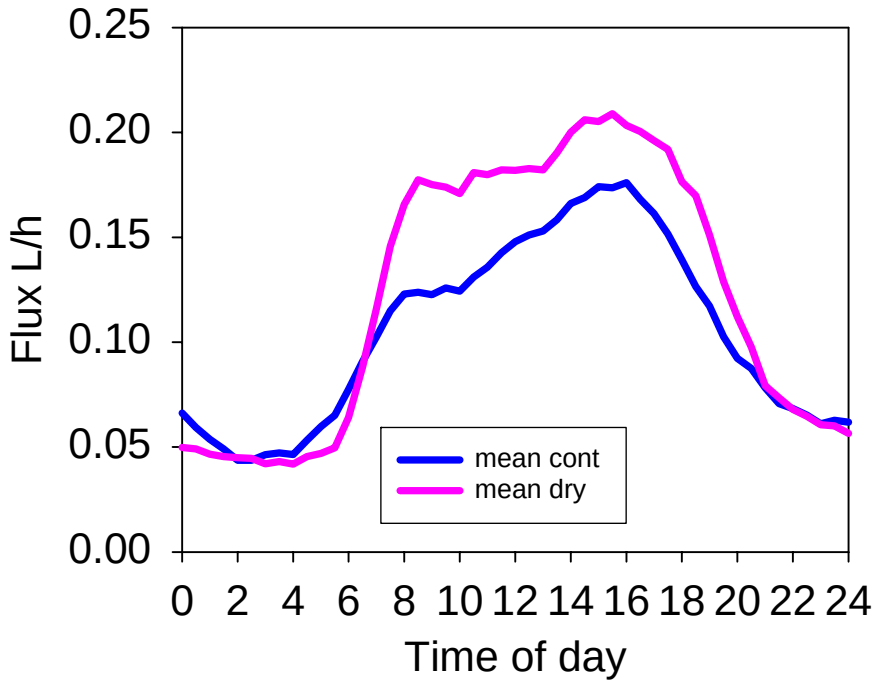


Figure 39. Sap flow estimates for Merlot vines on January 19th 2005. Data for each line were derived from 2 sensors in each of 2 vines. At the time of these measurements all vines were irrigated. Later on in the experiment water was withheld from those designated as "dry".

Figure 40 shows that sap flow changed little from day to day despite large changes in evaporative demand (VPD). A combination of low radiation and rain on Feb 2nd, Feb 9th, March 5th and March 15th had a marked effect on sap flow however. Whereas daytime sap flow appeared to be relatively insensitive to changes in VPD, higher nighttime sap flow was associated with periods of high night temperatures and VPD. For most of the experimental period when all vines were receiving the same irrigation the average sap flow of the control vines was between 80 and 90% of that of the "dry" vines. This was probably the result of vine-to-vine variation caused perhaps by slight differences in canopy size or sap flow sensor placement. All vines were irrigated on March 8th and over the next 5 days sap flow in both control and dry vines slowly declined (Figure 41) despite generally rising VPD (Figure 40). On March 13th the control vines were irrigated and their sap flow increased whereas that of the dry vines remained unaltered. The control vines were again irrigated on March 15th. It also rained that day and sap flow in both control and dry vines was very low.

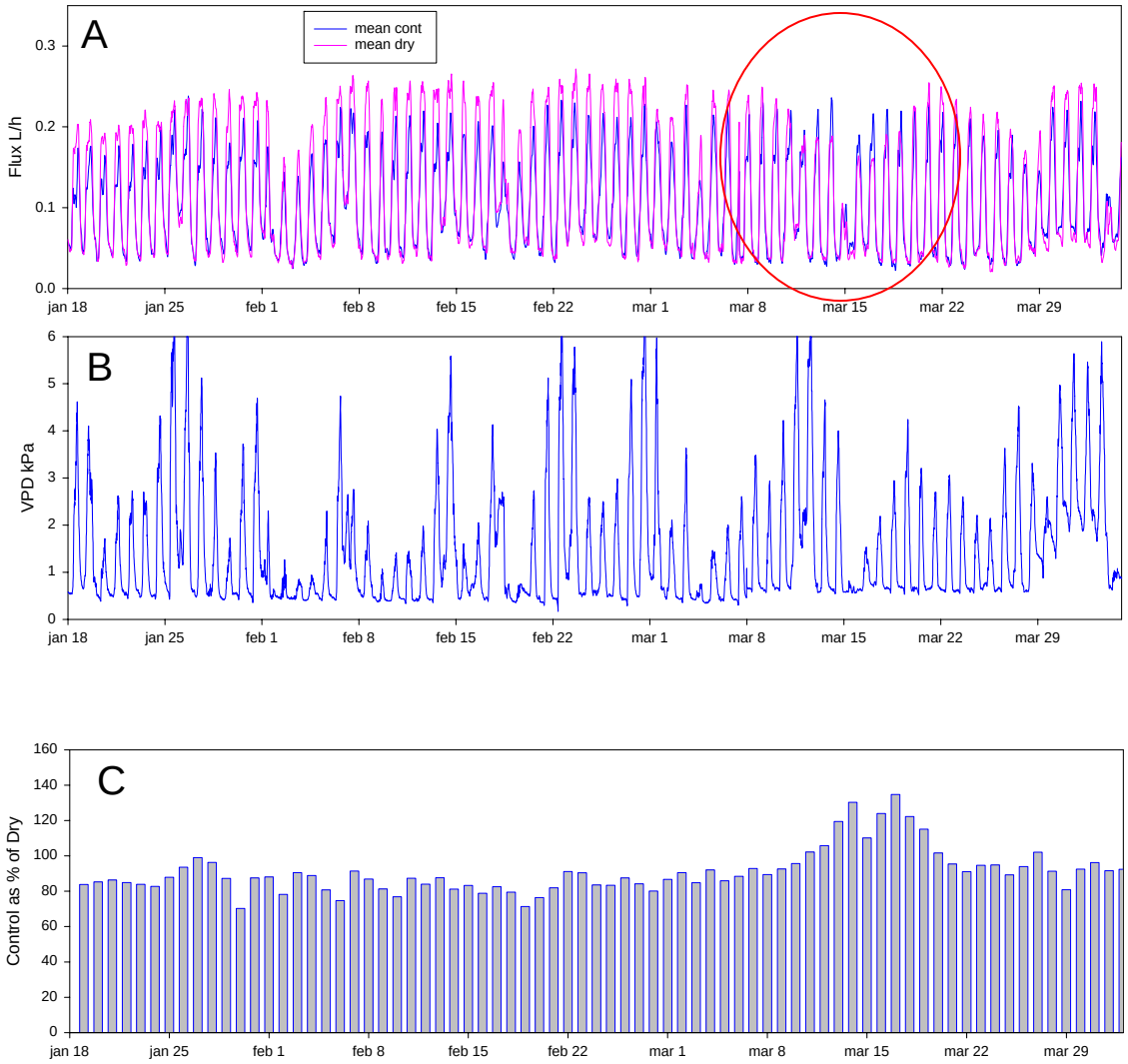


Figure 40. A. sap flow of irrigated (control, blue) and not irrigated (dry, pink) Merlot vines. Each line is the mean of the outputs from 4 sensors. B. VPD during the same period. C. Daily maximum sap flow of the control vines expressed as a percentage of the maximum flow in dry vines. The control and dry vines received the same irrigation until March 8th when water was withheld from the dry vines. The red circle indicates this period and this is shown in more detail in Figure 41.

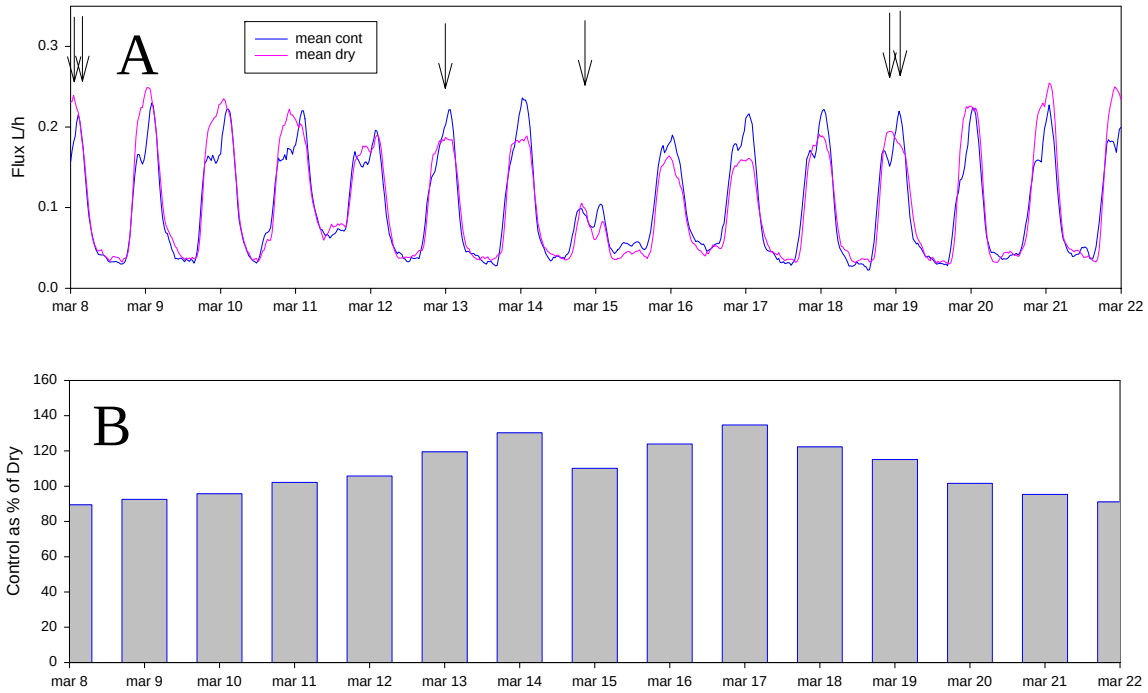


Figure 41. A. Sap flow of Merlot vines. All vines were irrigated on March 8th (double arrow). Single arrows indicate irrigation on Control vine only. All vines were again irrigated on March 19th (double arrow). B. Maximum daily sap flow of control vines expressed as a percentage of that of non-irrigated.

The difference in sap flow between control and dry vines was maintained over the next few days until all vines were again irrigated on March 19th. Despite being without irrigation for 11 days the maximum reduction in sap flow, relative to controls, was only about 20 to 30% (Figure 41). Midday water potentials were measured on March 11th , with control and dry vines recording values of -1.61 and -1.57MPa respectively (P=0.37) and again of March 17th when the values were -0.94 and -1.54 MPa respectively (P<0.0001), the water potential of the control vines having risen in response to the irrigation while that of the dry vines remained low. In summary, the sap flow of well irrigated vines changed little in response to large changes in ambient evaporative conditions, probably as a result of stomatal adjustment. After irrigation was withheld differences in sap flow between the non-irrigated and control irrigated vines was evident almost immediately but this was due to an increase in the control rather than any short-term change in the sap flow of the dry vines.

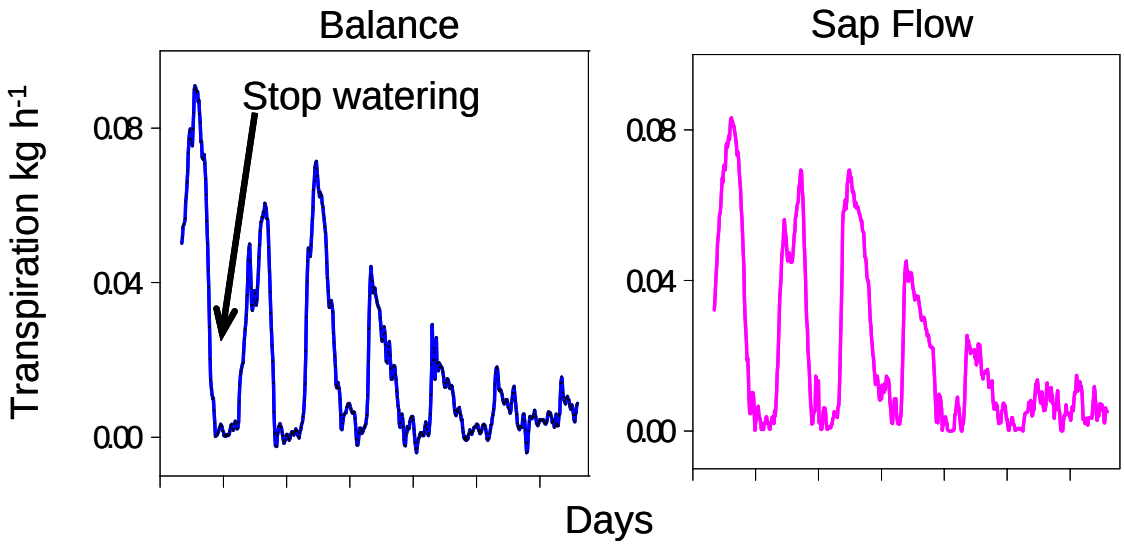


Figure 42.Transpiration of potted Sultana vine measured with heat pulse sap flow sensors (pink) and with an electronic balance (blue). Water was withheld on day 1.

Although the sap flow sensors installed in Merlot vines in the field failed to convincingly detect major changes in transpiration following cessation of irrigation, this probably reflects our incomplete understanding of the factors driving transpiration of these vines rather than a failure of the technology. We have shown on a number of occasions that heat pulse sap flow sensors can sensitively detect changes in transpiration. For example, in one experiment we installed sensors in a cane of a potted Sultana vine. The vine was placed on an electronic balance and the output from the sap flow sensors and balance were logged at 15 minute intervals. The output from the sap flow sensors accurately reflected the actual transpiration measured with the electronic balance (Figure 42), even reproducing fine detail in daily changes in water use. For example, on day 2 there was a reduction in transpiration in the middle of the day, probably caused by a reduction in radiation and this was shown by both balance and sap flow sensors. In this example sap flow was normalized to the absolute values provided by the electronic balance. Without prior knowledge of real transpiration rate values reported by the sap flow system may be in error, especially with the relatively small conducting cross sectional area of a grapevine cane. Nevertheless, this example does illustrate that the heat pulse sap flow system is capable of faithfully reporting changes in transpiration on a day to day basis.

Dendrometry

Verdtech, a Spanish irrigation equipment company, manufacture trunk diameter sensors for perennial horticultural crops and these can be integrated with an existing remotely accessed soil water monitoring system that is in use in Australia.

Following a meeting with Dr Moshe Cohen of Verdtech, in Spain it was arranged that he would visit Australia and install a number of the trunk diameter sensors developed and manufactured by Verdtech. Six sensors were installed in an existing PRD experiment on Chardonnay grapes located on the SARDI Research Centre, Nuriootpa (Figure 43).



Figure 43. Verdtech trunk diameter sensors on Chardonnay vines, Barossa Valley

The sensors were connected to an existing remotely accessed soil water monitoring system and trunk diameter was recorded every 15 minutes. There were few maintenance problems with the sensors and all six sensors operated satisfactorily throughout the entire growing season. Data were exported from the Verdtech software supplied, compiled into a Microsoft Access database for interrogation and derivation of the maximum daily skrinkage as recommended by Dr Cohen and selected data graphically presented in SigmaPlot®.

Seasonal pattern of change in trunk diameter.

All vines exhibited a similar pattern of change in trunk diameter between budburst and harvest. There was a daily expansion and contraction throughout the season and between about budburst and flowering there was no overall increase in trunk diameter (Figure 44). Commencing soon after flowering the daily expansion was greater than the daily contraction resulting in an overall increase in trunk diameter during this eight week period. Maximum trunk diameter occurred in mid January followed by a period of reduction in trunk diameter prior to harvest (Figure 45).

Irrigations applied during this period were detected by the sensors, however trunk diameter continued to retract during this period. Post harvest, trunk diameter stabilised and remained similar throughout the winter period (data not presented).

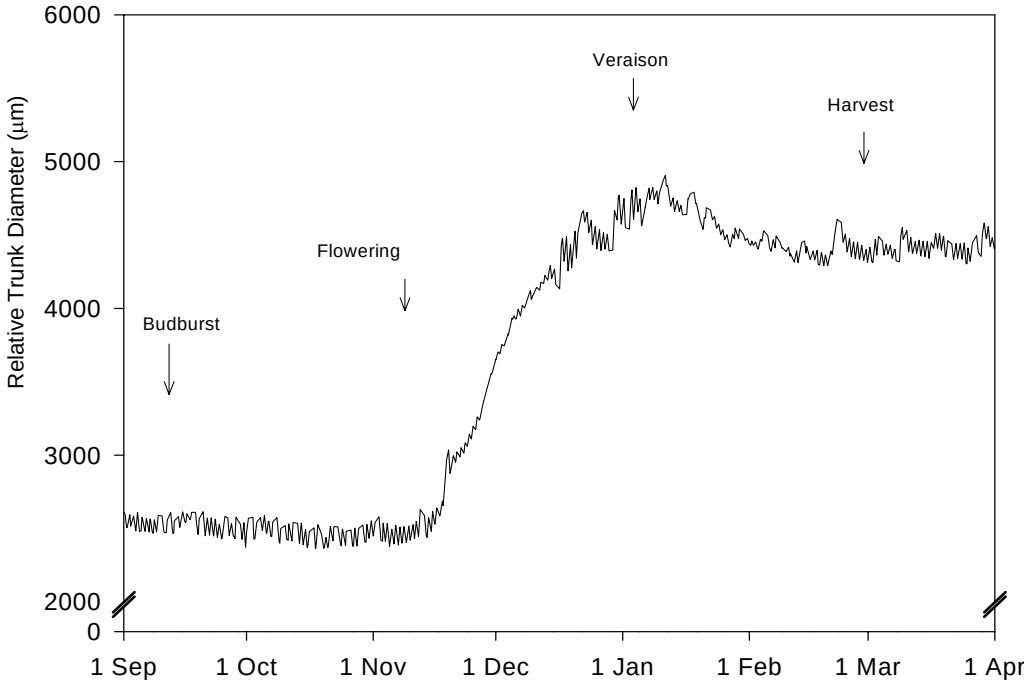


Figure 44 Change in trunk diameter of a single vine between September and April.

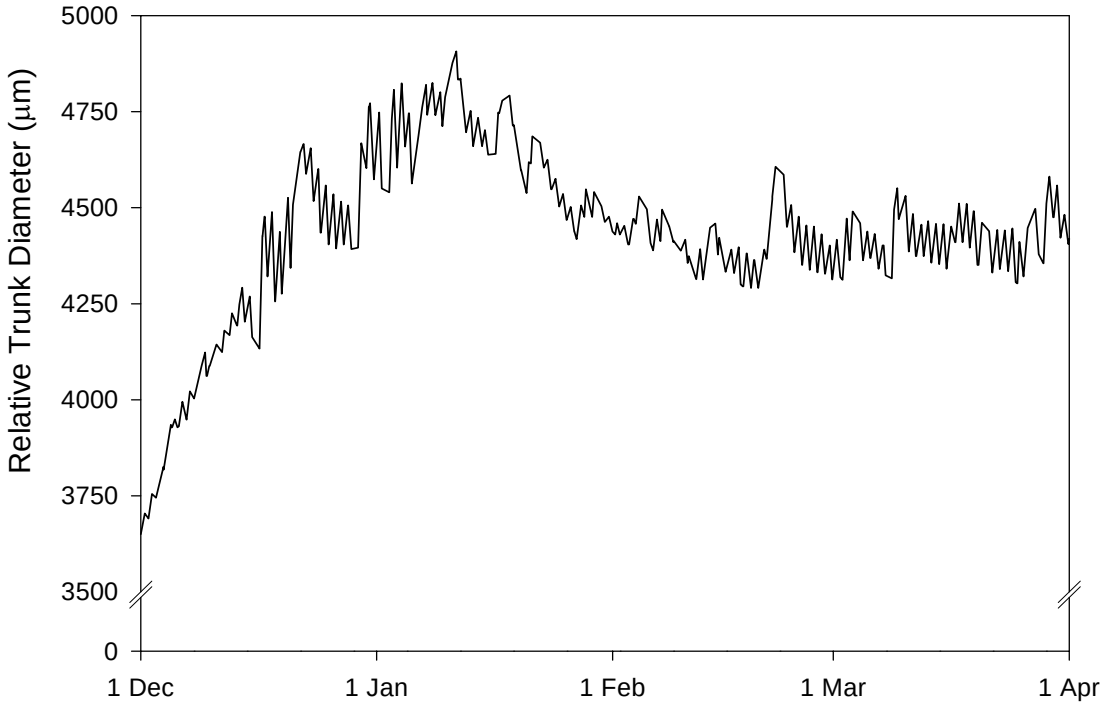


Figure 45. Change in trunk diameter of a single vine between December and April showing period of reduction in trunk diameter during January-February.

Daily pattern of trunk diameter evolution

Maximum trunk diameter was recorded around sunrise each day (Figure 46 and Figure 47) and daily minimum at about 4:00pm (Central Standard Summer time) each afternoon. Weather conditions influenced the maximum daily shrinkage (MDS) with greater MDS on hot days with high evaporative demand (Figure 46) compared with milder days (Figure 47)

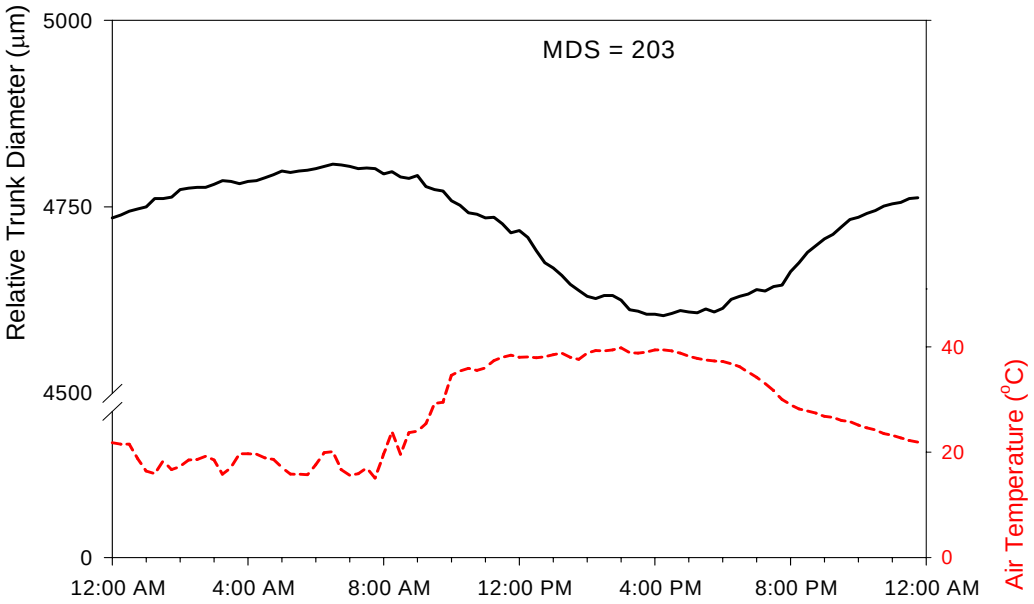


Figure 46. Change in trunk diameter over a 24 hour period during which maximum screen temperature was 39.8°C at 3:00pm Central Standard Summer Time.

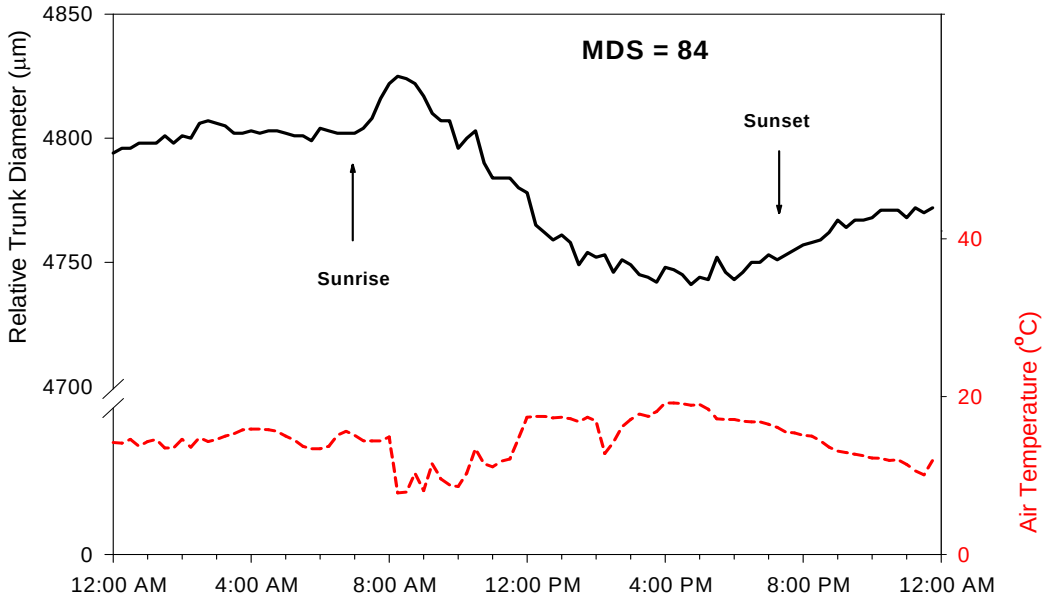


Figure 47. Change in trunk diameter over a 24 hour period during which maximum screen temperature was 19.2°C at 4:00pm Central Standard Summer Time.

Trunk diameter exhibited a high sensitivity to rainfall events. On a broad time scale noticeable increases in trunk diameter were associated with rainfall events at any stage of

the growing season. On a 24 hour time scale, in association with 15 minute rainfall data from an automatic weather station located approximately 10 meters from test vines, it can be seen (Figure 48) that a sudden increase in trunk diameter in late October coincided almost exactly with rainfall. This response could be the result of rapid foliar uptake of water and vine hydration or simply due to bark expansion as a result of direct wetting or the increase in air humidity associated with rainfall. This rainfall event only produced a small increase in soil water content and no change was detected below the 20cm deep sensor. A simple experiment involving direct wetting of the trunk bark or foliage wetting only should be undertaken.

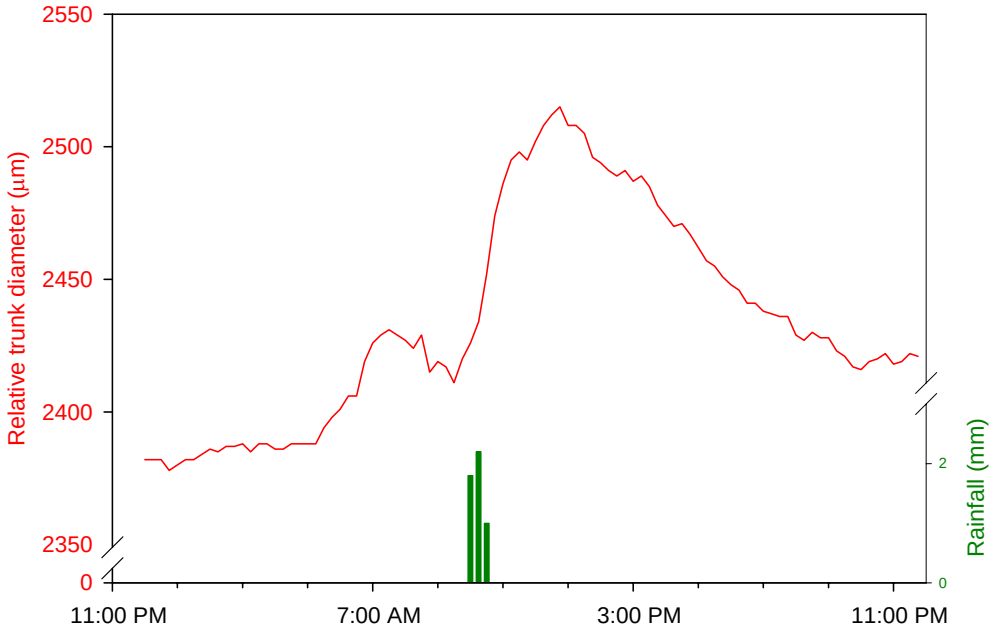


Figure 48. Change in trunk diameter over a 24-hour period during which a rainfall event occurred after the attainment of early morning maximum trunk diameter

Can trunk diameter be used as an irrigation scheduling tool?

For any device to be used as an irrigation scheduling tool it should reflect the need for irrigation including some predictive ability as to the timing of the next irrigation and also respond to irrigation. In grapegrowing regions such as the Barossa Valley where winter rainfall is usually sufficient to fill the vine rootzone to field capacity, irrigation scheduling tools must be able to track the decline in soil water content or change in plant water status during the early part of the growing season and define the stage when irrigation should commence as both too early or too

late will have negative impacts on yield and quality. Tools that indicate an irrigation requirement after the threshold are not suitable. As many Australian vineyards are now managed using some form of deficit irrigation vines are under some degree of water stress at key phenological stages. It could be hypothesised that trunk behaviour under such conditions would vary from that of a fully hydrated vine and may not be suitable for irrigation scheduling under deficit conditions. This, combined with the seasonal changes and sensitivity to rain reported here, may add too many complexities to the immediate adoption of this technology.

Irrigation responses during period of trunk expansion

In the first half of December 2003 there was steady increase in trunk diameter until the 14th December when trunk recovery at night was reduced (Figure 49). A similar pattern occurred over the 2 succeeding nights. Based on soil water content estimated from capacitance probes and data collected from previous seasons, an irrigation was applied on the 16th December and there was an immediate response in trunk diameter. This 3-day period of trunk shrinkage coincided with higher than average temperatures with a maximum of 40^{o}C recorded on the 16th December, the day an irrigation was initiated. The maximum temperature recorded the following day was 10^{o}C cooler and there was a large increase in trunk diameter during that 24 hr period. It is unclear if the observed trunk shrinkage was in response to above average air temperature or water deficit.
In late December there was another period of trunk shrinkage. However, based on maximum daily shrinkage data between about the 22nd and 29th December (not presented) it was difficult to schedule the timing of irrigation using these data and soil water data were used to schedule the irrigation applied on the 29th. There was a large increase in trunk diameter following this irrigation.

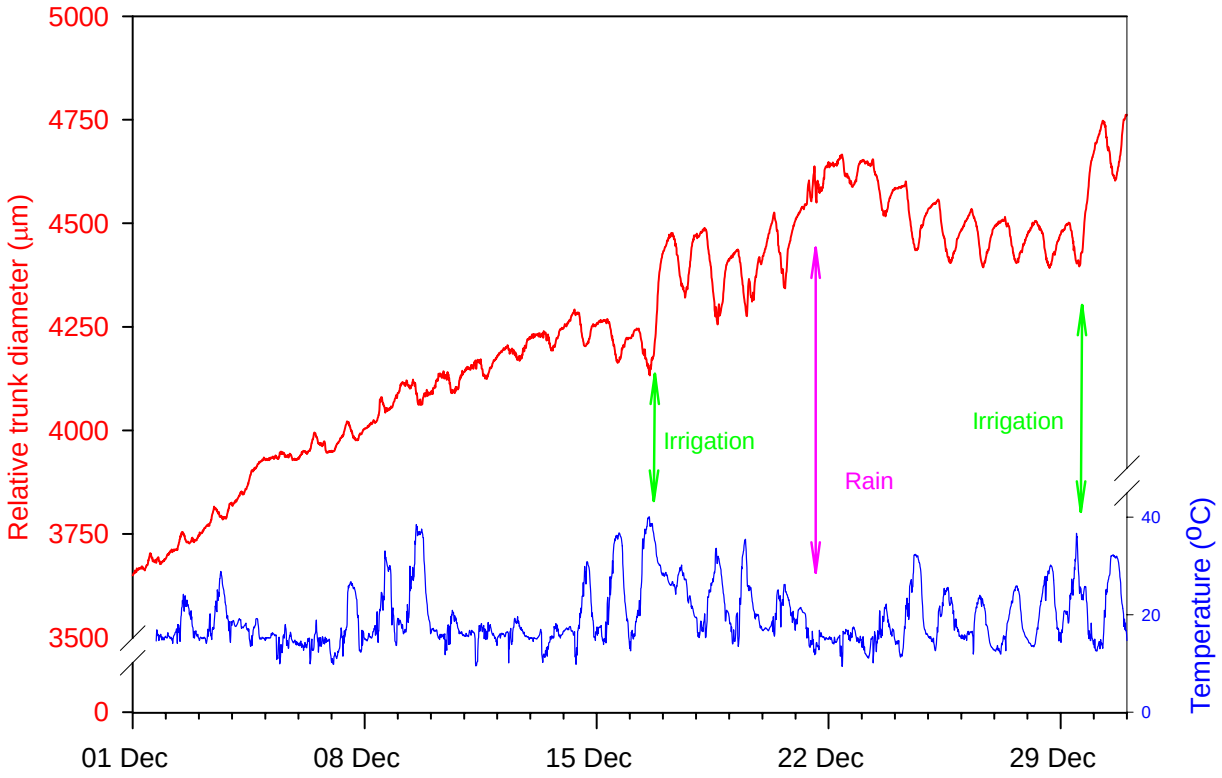


Figure 49. Change in trunk diameter of a single vine and screen air temperature during December. Arrows indicate the commencement of irrigation or rain fall events

Irrigation responses during period of trunk shrinkage.

Commencing in early January (Figure 45) the steady decrease in trunk diameter added to the difficulty in predicting the need for irrigation. Maximum Daily Shrinkage (MDS) was also highly variable (Figure 50) during this period with daily MDS being greater than changes after an irrigation. The large MDS recorded on 12th January does not appear to be associated with abnormal weather conditions (temperature, wind, humidity) and highlights the potential difficulty of using this technology to schedule irrigations.

MDS could perhaps be used as a predictor of irrigation if the seasonal loss in trunk diameter, (expressed on a daily basis) was subtracted from MDS. Data collected from well irrigated vines (irrigated nightly) and vines with the entire crop removed exhibited the same pattern of shrinkage (data not presented). While experimentally this approach is feasible it would be impractical for growers wishing to use changes in trunk diameter to schedule irrigation.

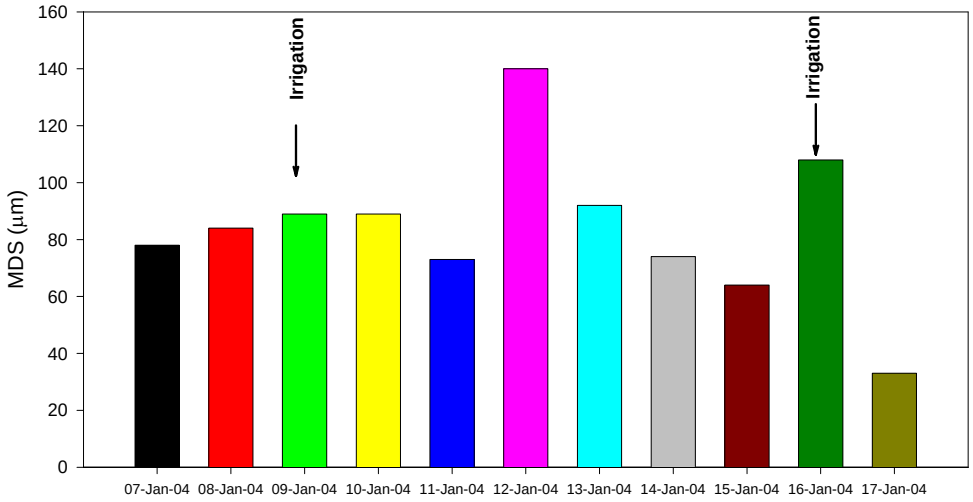


Figure 50. Daily maximum daily shrinkage (MDS) of a single Chardonnay vine during January 2004. Arrows indicate timing of irrigation events.

Conclusions

- Reliable measurements can be made over a long period of time using the trunk diameter sensors supplied by Verdtech. The data could be logged.
- The large amount of data generated required considerable processing before an interpretation was possible. This needs to be automated.
- Vines show different responses at different times of the season, again complicating data analysis
- Rain events significantly disrupt the daily patterns of stem diameter changes and daily changes in trunk behavior did not always reflect water status
- Constraints may be imposed due to the cost of equipment and the ability to measure relatively few vines

Implications and Recommendations

In carrying out this project we have chosen techniques for measuring plant function that are reasonably well established and understood and could be implemented using relatively unskilled operators at a reasonable cost. Resource constraints have also limited the scope and range of techniques that could be investigated. There are other techniques in existence or that could be further developed that have not been addressed in this project. For example, there are reports of the development of an irrigation control system based on the sensing of leaf thickness. This may be analogous to the continuous sensing of leaf water potential as discussed in this report. The leaf thickness sensor has been developed into a commercial product by a company in Israel (Leaf-Sen Irrigation Systems Ltd). We have not considered changes in canopy reflectance that might be measured through remote sensing, either from aerial or satellite images, as a means of measuring canopy performance. It was felt that there was a requirement for assessments virtually on a daily basis. Irrigation cycles, particularly in the hotter regions such as the Riverland and Sunraysia where the soils are freely draining, can be just a few days, making imaging that would have to obtained from a third party impractical and expensive. Furthermore, the ability of these technologies to detect early changes in canopy properties is limited and probably would not offer sufficient early warning of the effects of water stress (Cifre et al (2005). These authors also identify remote sensing of chlorophyll fluorescence as a means of assessing vine response to water deficit. However, the development of specialized instruments and knowledge would be required to make this work and it is beyond the scope of the present investigation.

As an early warning of impending water stress, stomatal conductance probably has no equal, but the dynamic way in which the stomata are able to respond not only to plant water status but also to environmental variables such as ambient humidity, light and wind speed make the measurement and interpretation of conductance data difficult. Our assessments of spatial and temporal variation in stomatal conductance were characterized by a high level of error, requiring many measurements to be made to bring these errors within acceptable limits. This would require considerable time investment and to some extent this is self-defeating since we are dealing with a variable known to change significantly within the time frame required to make enough measurements to reach the acceptable error level. The equipment required for the measurement of stomatal conductance is reasonably expensive and not particularly robust for field use. We have found to our cost that the capacitive sensors used to measure relative humidity that are employed

in many of these instruments can be permanently damaged by exposure to sulphur. This would be of major concern since sulphur treatments form the basis of most spray programs aimed at powdery mildew.

In assessing the various plant-based measures of vine performance we have concentrated mainly on commercially important varieties such as Shiraz, Cabernet Sauvignon and Merlot. Work in other projects is drawing attention to the fact that there is significant variability in the way that different varieties respond to water deficits. For example, Soar (2004) has shown that under relatively mild environmental conditions Shiraz and Grenache are not readily distinguished on the basis of their stomatal behavior. However, under more stressful conditions of high temperature and VPD it becomes possible to draw a distinction between these varieties on the basis of their expression of isohydric and aniosohydric characteristics. This means that Shiraz is able to maintain relatively high rates of gas exchange on stressful days at the expense of stored soil water. Grenache, on the other hand, has stomatal control that is more tightly coupled to current ambient conditions and it tends to exhibit reduced rates of gas exchange under these conditions, thus minimizing its use of stored soil water. Knowledge of these distinctions would be important in the interpretation of stomatal conductance data because a low conductance in Grenache on a hot day might not reflect a need for additional water. Conversely, a relatively high conductance in Shiraz may not mean that it does not need additional water resources. These subtleties in data interpretation, the high level of variability in conductance data that requires multiple measurements over an extended period and the sensitivity of some sensors to a common agricultural chemical probably preclude the widespread use of stomatal conductance measurement as means of indicating irrigation water requirement.

One reason for the high level of variability in conductance measurements is that each leaf is able to independently modify its stomatal behavior. Indeed, it is likely that small groups of stomata (and there are several hundred per square mm of leaf surface) are able to operate independently of other groups on the same leaf. Variability could be reduced if the conductance of the whole vine, or groups of vines, could be measured. By measuring the flow of sap in the trunk or cane of a vine it is possible to integrate the water loss from each individual leaf on that vine and we have shown in this report that heat pulse sap flow sensors are capable of producing estimates of xylem sap flow over extended periods with little maintenance other than battery changing and even this could be eliminated with the use of solar panels to maintain battery function. These sensors produce an estimate of instantaneous sap flow in L/h and the accuracy of this measurement is

difficult to validate in a field situation without sacrificing the vine. The figures obtained in this study with mature well watered Merlot vines suggested daily water use of about 3L per vine which is probably not unreasonable. However, because the sensors gave stable outputs over an extended period they would probably be used in a relative way to show trends in water use rather than absolute amounts. The data obtained in this study showed that in Merlot, sap flow was fairly consistent from day to day, despite large differences in evaporative demand. They clearly showed rain events however, when transpiration would have been reduced to very low levels. With experience in data interpretation it is likely that sap flow sensors could be used as a basis for irrigation scheduling and their long term stability make them attractive for this purpose. This potential has been recognised by a number of others (Ginestar et al 1998; Escalona et al 2002). However they are quite expensive and suffer from the problem that it is only possible to measure relatively few vines within a vineyard.

Plants are able to operate effectively over a relatively narrow range of leaf water potentials (ψL) and for grapevine this may be from zero to about -2MPa. In the United States midday ψL is commonly used as a guide to vine irrigation status and appropriate guidelines are available. However, as we have shown above anomalies can be generated because of the strong dependence of ψL on current stomatal conductance. In isohydric varieties where stomatal conductance is tightly linked to evaporative conditions or to irrigation pre-history ψL may not accurately reflect the water requirement of the vine and may in fact indicate the exact opposite. In situations where stomata have been induced to close through root-sourced chemical signals midday ψL of non-irrigated vines may be higher than those that had been fully irrigated. Similarly, the type of physiological mechanisms used by a vine to deal with potentially stressful conditions can influence measured ψL. We have seen above that isohydric vines will tend to close their stomata at higher values of ψL than anisohydric varieties, especially on hot dry days, so even with similar levels of soil water availability ψL may not be the same. Like stomatal conductance, ψL is very variable when the whole vineyard is considered and multiple measurements are required to bring errors to an acceptable level. Midday ψL is therefore subject to many influences some of which can result in serious anomalies if we assume a simple relationship between ψL and water requirement. Pre-dawn ψL on the other hand is not subject to many of these influences and may be considered as a surrogate for a measurement of soil water potential. Our experiments indicate that pre-dawn ψL can be a sensitive indicator of irrigation status and is much more stable than any of the other measures we investigated on a whole vineyard basis. Data interpretation may be

compromised, however, if the vines have access to even small amounts of water. In this situation the measured leaf water potential may reflect the water potential of the wettest soil to which the roots have access. This small amount of water may be rapidly depleted once the vines begin to transpire, resulting in the development of water stress. The disadvantage with pre-dawn ψL is implicit in its name, that is, it has to be measured during the hours of darkness but loggable sensors are now available for measuring ψL and with some further development these may prove to be an acceptable alternative to the traditional pressure bomb method of manual measurement. One of the disadvantages of these sensors was the relatively short period of time for which it was possible to measure a particular leaf before it deteriorated to the point where the data became unreliable. It may be possible to overcome this problem by installing the sensors in the trunk where it may be possible to make a direct measure of xylem water potential. Like sap flow in the trunk this would provide an integral measurement of the whole vine rather than for individual leaves which can be subject to significant variability due to shading, hydraulic connection to the cane or senescence. Stem water potential can also be estimated by covering leaves to prevent transpiration and then measuring ψL in a conventional pressure bomb. Leaf to leaf variability was certainly reduced by this method but the close correlation between ψL and stem water potential suggested that there was no more information to be gained with this method. The time saving in making fewer measurements to obtain an acceptable error level was offset by the time taken to apply the bags the day before. Given the close relationship between ψL and stem water potential it was suggested that petiole hydraulic conductivity must be high. However, Schultz (2003) has suggested that differences in hydraulic properties may be important in differentiating Grenache and Shiraz. We should therefore be mindful of the potential for there to be varietal differences in the relationship between ψL and stem water potential.

Subject to the provisions imposed by the differentiation based on isohydric/anisohydric behavior, we have confirmed transpiration as a plant-based measure with potential to yield information about the physiological health of a vine but its use may be precluded by the high degree of leaf to leaf variability and the consequent high time investment required to obtain estimates with a sufficiently low error component. However, it is well known that transpiration is linked to canopy temperature through the cooling effect of water evaporating from the leaf and many attempts have been made to use leaf temperature to define plant health. Recent advances in thermal imaging technology have allowed a better understanding of the relationships between canopy temperature and current environmental conditions (Jones et al 2002). Jones (1999) describes the derivation of a canopy conductance index (Ig) from measurements of canopy temperature and the temperatures

of wet and dry reference surfaces. We have shown that this appears to be a robust way to describe vine water use under a range of conditions and it readily differentiated between Shiraz, Merlot and Cabernet vines at various stages of their irrigation cycle. Like most other plant-based measures there is a high degree of leaf to leaf temperature variability but to a large extent this is overcome with infrared thermometry because the instruments are capable of integrating large areas of canopy in a single measurement. We have taken this one step further by making many thousands of measurements over the course of a few minutes. A disadvantage of this is that data management becomes a significant issue and for this to become a usable system more effort will need to be devoted to custom-written software to deal with the data. Also, the maintenance of the wet and dry temperature references needs improvement. The tight linear relationship between log(Ig) and the more easily derived canopy temperature relative to ambient temperature needs confirmation under a range of ambient temperature and humidity conditions, but it does suggest that there may be new ways of dealing with these data for a simplified outcome. While most of the datasets collected conformed to expectation there were some anomalies that were most readily explained by instrument instability. Nevertheless, a consideration of the possible causes of these anomalous data did highlight the need for a better understanding of the effects of rapidly changing ambient conditions on the robustness of the Ig calculation and the way that temperature data are collected.

Table 6. Summary and conclusions

Technique	Comment
Stomatal conductance	• A sensitive measure of vine performance but subject to rapid change in response to ambient conditions. Data interpretation may be difficult without specialized knowledge • High level of temporal and spatial variability requiring multiple measurements • Equipment not very robust under extended field use ***A useful research tool but not yet developed for routine commercial use***
Sap flow	• Provides knowledge of the performance of the whole vine • Installations stable for extended periods without attention • Some uncertainty regarding the accuracy of absolute sap flow rates but capable of producing reliable long term relative numbers • Number of vines that can be measured may be limited by cost ***A robust reliable technique capable of further development for commercial use***

Midday leaf water potential (ψL)	• A well established method for assessment of plant water status, however the relationship between measured ψL and physiological function may not be so well established • Guidelines for the interpretation of ψL data have been developed • Equipment is robust and mobile so measurements throughout a vineyard are possible • A high level of spatial variation may require multiple measurements to reduce error • Interaction between stomatal function and ψL may lead to incorrect data interpretation • New loggable equipment may make this measurement more attractive, although then subject to the constraint of few measured vines ***Easily measured with reliable equipment by relatively unskilled personnel. Variability may require many measurements for reliable data. Skill required in data interpretation, especially with regard to varietal differences. New equipment needs development but shows promise.***
Stem midday water potential(ψstem)	• Closely correlated with ψL but less error due to reduced leaf to leaf variability may mean fewer measurements required • Advantages offset by time required for placement of bags ***Some advantages over ψL but subject to the same basic constraints***
Pre-dawn leaf water potential (ψpd)	• A sensitive measure of soil/plant water status capable of detecting differences on a daily basis • Easily measured but must be done before sunrise. • Eliminates short-term variability due to stomatal responses • Data interpretation may be compromised by discontinuities in soil moisture as the indicated ψpd will reflect the wettest soil ***The most sensitive measure of water potential for detecting irrigation status. Capable of continuous measurement with new equipment***
Canopy temperature	• Measurement of canopy temperature can readily differentiate vines on the basis of their irrigation status • Thermal imaging techniques demonstrate errors that can be introduced by non-vine components • High leaf to leaf variability eliminated by multiple measurements made from moving vehicle with cheap infrared thermometers • Large areas of vineyard can be measured in a short period of time • Canopy conductance index (Ig), calculated from measurements of canopy temperature and temperature of reference surfaces is a good indicator of vine response to irrigation treatment

	Further development of this methodology is required but it shows significant promise as a cheap, reliable method for large scale use.
Dendrometry	• Reliable measurements can be made over a long period of time • Data can be logged • Data interpretation is difficult but attempts have been made to automate data analysis and decision making • Vines show different responses at different times of the season, again complicating data analysis • Rain events significantly disrupt the daily patterns of stem diameter changes • Constraints may be imposed due to the cost of equipment and the ability to measure relatively few vines ***Instrumentation can be made robust and reliable and capable of extended periods of measurement. With some more development dendrometry could be a useful tool applicable to grapevines in Australia***

IP Issues

Most of the techniques used in these studies are simple plant physiology tools with a long history of use. There is no IP associated with their use. We have identified the need for knowledge about varietal differences in interpreting data obtained. This knowledge may not exist but this knowledge gap is being addressed in a new GWRDC project (CSP05/02).

One of the most promising areas identified for development by this project is the application of canopy temperature measurement to irrigation scheduling. While the idea of using infrared thermometry for this purpose has been public knowledge for many years, the development of the so called "*canopy conductance index*" (Ig) has been has been carried out by one of the coauthors of this report (Professor HG Jones of the University of Dundee). The visit of Professor Jones to Australia for the purpose of carrying out this project has been covered by an agreement, copies of which are held by CSIRO, GWRDC and University of Dundee.

Mr Andrew Skinner of Measurement Engineering Australia has been a collaborator and has contributed expertise in data logging and data manipulation.

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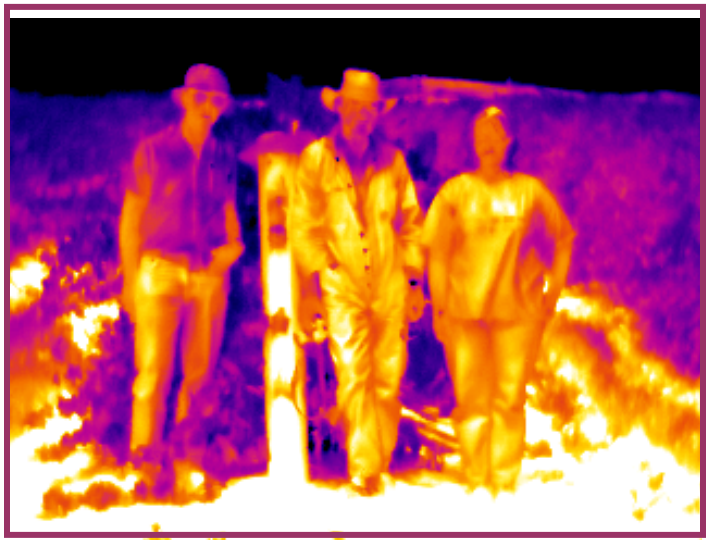
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Appendix 1.

Statistical Analysis of Spatial-Temporal Variation in Leaf Water in a Vineyard

05/143
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Report prepared for Dr Brian Loveys
CSIRO Plant Industry
September 15, 2005

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Executive Summary

In conclusion, the following points are made

1. Temporal occurred for all measures except in the pre-dawn water potential measures. Measures at other times will be difficult to compare both across sites and between times;
2. Stomatal conductance measures are much more variable than stem and leaf water potentials;
3. It is necessary to include a number of vines to obtain stable measures for all parameters – for some parameters, especially stomatal conductance, savings can be made by taking multiple (5-10) measures on the same vine;
4. A method for determining an optimal strategy that gives the smallest cost of obtaining a required standard error is given;
5. For water potential, no more than two measures per vine should be taken;
6. Small standard errors cannot be achieved by sampling one or two vines only as shown in Figure1.

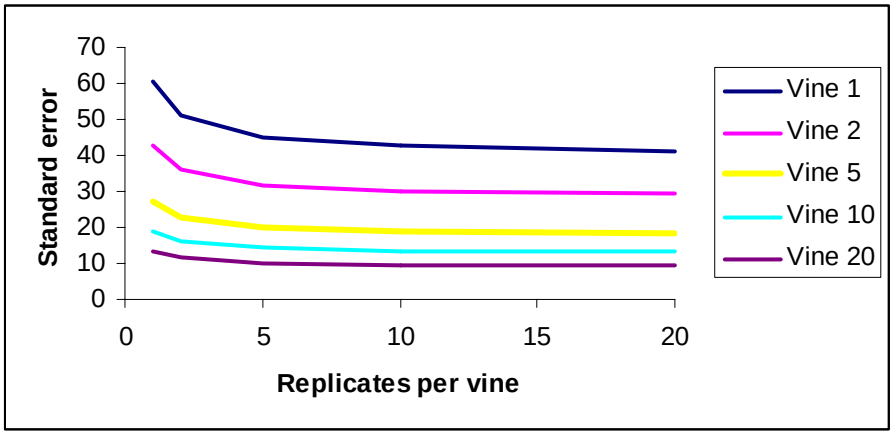


Figure 1 Effect of increasing number of replicates per vine for different numbers of vines, (Data from Set 6, stomatal conductance, 16/12/04)

Introduction

Water potential is known to be one of the limiters of plant growth. However, good water is often a limitation in viticulture. Suitable sampling measures are therefore required to obtain a cost-effective measure of the water potential in the vineyard.

The formulation of a suitable monitoring scheme requires an understanding of the types of variation. Basically two types of variation need to be considered.

Spatial variation

Spatial variation will arise in a vineyard from two main sources

Variation due to location in the vineyard.

No vineyard is totally uniform. Typically there are trends in a range of variables including soil texture at different levels, depth to the water table and soil thickness to name a few. Typically there will also be irrigation that is likely to be unevenly applied both at a dripper scale and at the vineyard scale. This variation across the vineyard is a major contributor to the spatial variation.

Between vine variation

A second form of spatial variation arises due to differences between vines. These differences would be due to very local effects and perhaps a residual nursery effect, a planting effect etc. Such local effects can be reinforced by inter-plant competition. The between vine effects is approaching the 'nugget' effect of Kriging.

When a few vines are considered, and these are well dispersed across the vineyard, the gross spatial effect would be almost completely confounded with the between vine effects. When this situation occurs, the vines can be considered as random samples from the vineyard. The current study, which would consider only less than 100 vines from a vineyard, this approximation would be satisfactory.

Temporal variation

Temporal variation can also be considered at a range of scales

Effect of minute-scale effects

These short term variations can be caused by changes of incoming radiation due to clouds or changes in wind speed. Over a scale of minutes these effects would be more or less consistent across the entire vineyard.

Diurnal effects

These effects occur at the scale of hours in a fairly predictable manner. Often these effects can be described by a harmonic model.

Day to day effects

Day to day effects are mainly caused by such factors as change in weather patterns, irrigation schedules.

Seasonal effects

Seasonal effects are a result of changes in weather patterns together with changes in phenology such as leaf loss, bud burst etc.

Other sources of variation

There will be variation other than the temporal and spatial components. This variation could include components from
Difference between plant parts;
Instrument error;
Operator effect and bias.
For convenience, these other sources of variation will be termed measurement error (or noise).

Statistical methods

Variance components

The variance of a singe measurement can be considered as the sum of the variance of each of component. We write this as

$V(y) = Vs + Vt + Vn$

where

Vs is the spatial component;

Vt is the temporal component; and

Vn is the noise component.

Variance of mean measurement

Often several measures are taken on the same vine. The variance of a measurement can then be written as

$V(y) = Vs + Vn/r$

The variance of the mean of y is then

$V(\text{mean}(y)) = (Vs + Vn/r)/s.$

The variance of the mean of several observations is less because of the replication

Isolating the components of variation

The variation between samples taken from different vines will typically be a mixture of all three sources of variation. An understanding requires a measure of these components. Several techniques are available for this.

Estimation of measurement error (Vn)

An approximate estimate of the noise of the component can be obtained by taking multiple readings on a single plant in quick succession. An analysis of variance using the vine-time units as classifying factors can be used here. The error mean square of this analysis would provide a measure of the noise.

The between units mean square can be used to derive a variance component that is a mixture of both spatial and temporal variation.

Estimation of temporal variation

A system of stationary readings can be used to give a measure of the temporal and noise components of the variance.

Estimation of spatial variation.

An estimate of the spatial variation can be obtained by considering the readings from the moving samplers, and then correcting for temporal variation.

Data available

Set 1 Leaf water potential (15/1/04)

The data contain a series of measures in duplicate of approximately 40 vines, observed at two times of the day.

A series fixed measures were taken over the period, but at not exactly the same times.

Set 2 Pre-dawn water potentials (14/1/05)

There was a series of continuous (stationary) measures taken. Over the same period data were collected in triplicate over 40 vines

Set 3 Pre-dawn water potentials (13/2/04)

There was a series of continuous (stationary) measures taken. Over the same period data were collected without replication over 40 vines

Set 4 Stomatal conductance (6/1/04)

In this data set there were morning values taken in pentrupicate and afternoon values were taken in duplicate. There was also a set of stationary measures.

Set 5 Stomatal conductance (18/2/04)

In this data set single readings were taken on each of 40 vines in three cycles. At the same time a set of stationary measures was also taken

Set 6 Stomatal conductance (16/12/04)

This data set contained both fixed and moving readings. Typically the moving readings consisted of 10 individual measures on each plant.

Set 7 Stem and leaf water potential (8/2/05)

Any gross outliers observed in the data were discarded

Results of Analyses

Set 1 Leaf water potential (15/1/04)

The stationary data showed a trend with time which could be described by scatter around a spline (Error! Reference source not found.). The variance of the scatter about the spline was 0.25. There were duplicate readings taken on each vine. This enabled a two-way analysis of variance to be performed giving an estimate of the error mean square of 0.33.

The spline was used to give an estimate of the temporal effect for each time of the moving readings. This estimate was used as an input to a REML model (Thompson and Paterson, 1971).

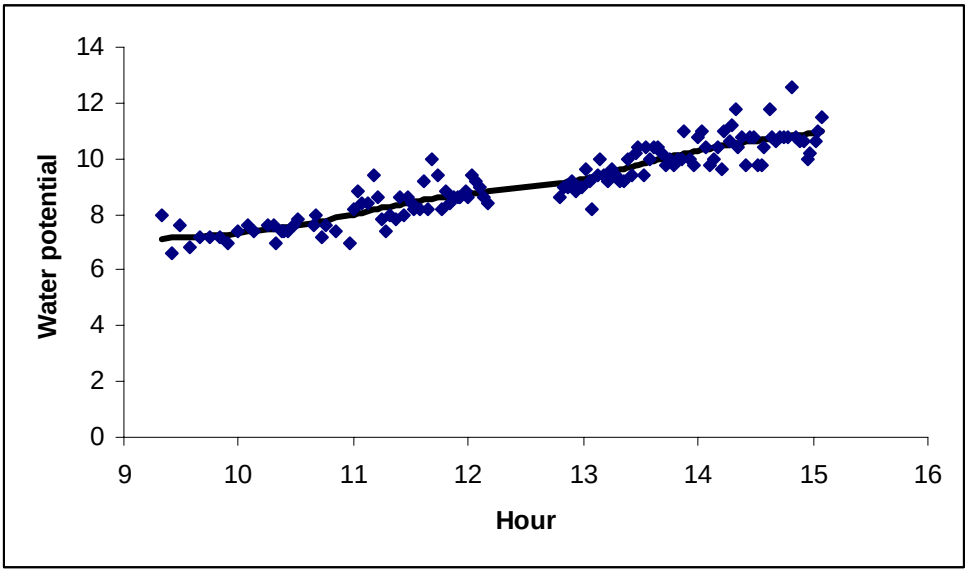


Figure 2 Change of water potential with time for Data Set 1 together with a fourth order spline. *Note: water potential data need to be multiplied by -0.1 to bring to MPa*

The REML analysis produces the following estimates of the variance components as shown in Table 2. Using those data it is easy to derive standard errors when varying numbers of vines and replicates per vines are tested using the formula

SE=Sqrt [(Between vine variance + noise/replicates per vine)/number of vines]

Table 1 Variance components for Set 1

Source	Estimate	SE
Vine	1.0797	0.2651
Measurement error	0.447	0.056

Table 2 Standard Errors of Estimates using Varying Numbers of Vines and Replicates per Vines for Set 1

Vines	1	2	5	10	20
Replicates					
1	1.236	0.874	0.553	0.391	0.276
2	1.142	0.807	0.511	0.361	0.255
5	1.081	0.765	0.484	0.342	0.242
10	1.060	0.750	0.474	0.335	0.237
20	1.050	0.742	0.469	0.332	0.235

Set 2 Pre-dawn water potentials (14/1/05)

There was no apparent effect of time on the stationary water potentials. There was a variance of 0.0060 about mean.

A REML analysis indicated that there was a vine effect present as well as noise.

Table 3 Variance components for Set 2

Source	Estimate	SE
Vine	0.004363	0.001128
Measurement error	0.00216	0.00033

Table 4 Standard Errors of Estimates using Varying Numbers of Vines and Replicates per Vines for Set 2

Vines	1	2	5	10	20
Replicates					
1	0.081	0.057	0.036	0.026	0.018
2	0.074	0.052	0.033	0.023	0.016
5	0.069	0.049	0.031	0.022	0.015
10	0.068	0.048	0.030	0.021	0.015
20	0.067	0.047	0.030	0.021	0.015

Set 3 Pre-dawn water potentials (13/2/04)

The stationary readings indicated a weak trend with time that could be described by a spline with variance about the spline of 0.00155. The trend of the spline was reflected in the moving readings as shown in Figure 3. While the trend was the same, the stationary readings were typically 0.17 less negative than the moving values. However, overall there was no relationship

between the temporal trend of the stationary readings and the moving readings. The variation can therefore be considered spatial and noise.

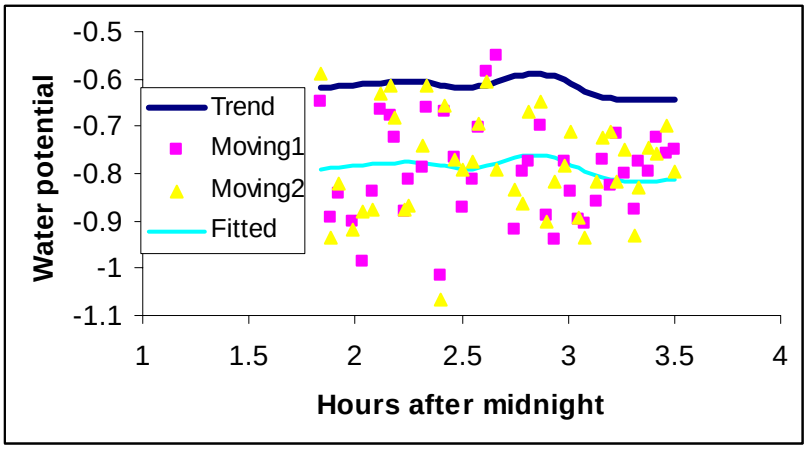


Figure 3 Trend of the stationary readings, the two moving readings and a bias corrected trend.

The variance estimates from the REML analysis are shown in Table 6. Those data were then used to produce the predicted standard errors when different numbers of readings per vine and numbers of vines are sampled.

Table 5 Variance components for Set 3

Source	Estimate	SE
Vine	0.00878	0.00223
Measurement error	0.00216	0.00245

Table 6 Standard Errors of Estimates using Varying Numbers of Vines and Replicates per Vines for Set 3

Vines	1	2	5	10	20
Replicates					
1	0.106	0.075	0.047	0.034	0.024
2	0.100	0.071	0.045	0.032	0.022
5	0.096	0.068	0.043	0.030	0.022
10	0.095	0.067	0.042	0.030	0.021
20	0.094	0.067	0.042	0.030	0.021

Set 4 Stomatal conductance (6/1/04)

Subset A

Two fixed values (10.117hour, 320) and (13.283 hour, and 465) were clear outliers and were excluded from the data. The remaining data had a mean and standard deviation of 132.7 and 61.6.

The stationary values could be described by a high order spline as shown Figure 4. That spline was used as an estimate of the effect of time for the moving readings.

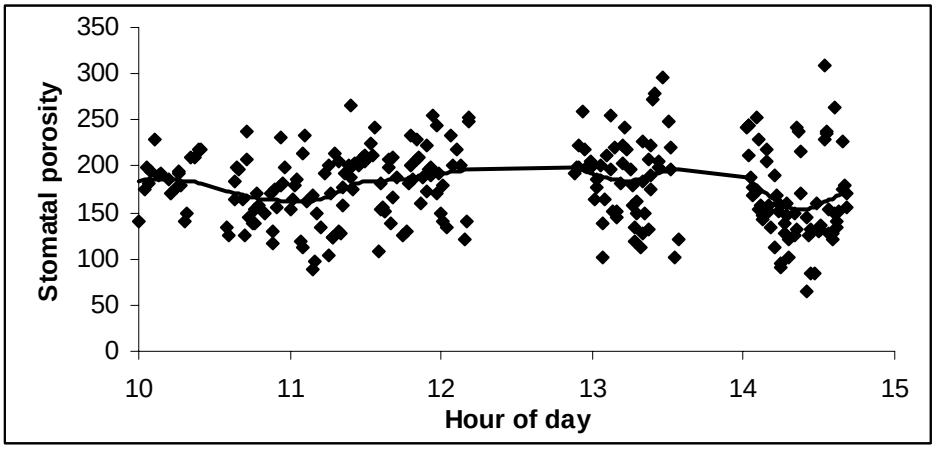


Figure 4 Stomatal conductance with a fitted trend

Subset B

There was no linear trend present (estimate of slope was only -1.7) in the data from the stationary samples, but there was a trend apparent in the moving values (Figure 5).

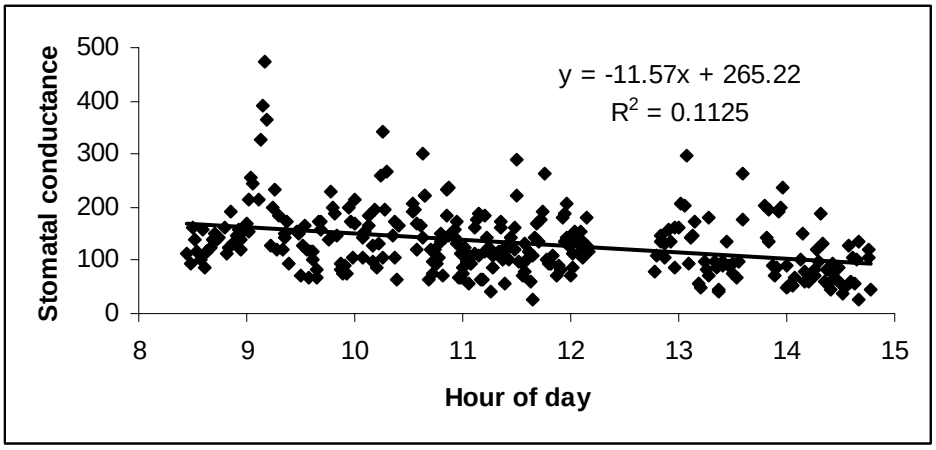


Figure 5 Moving values from Set 4 plotted against hour of day

A REML analysis was conducted on the moving readings. That analysis found that the temporal trend (derived from the stationary data) had no significant effect on the stomatal conductance, but hour of day was significant ($P<0.001$). The analysis provided estimates of the between vine variance and also an estimate of noise.

Table 7 Variance components for Set 4a

Source	Estimate	SE
Vine	1648	422
Measurement error	1790	160

The estimates shown in Table 7. were then used to derive the predicted standard errors shown in Table 8.

Table 8 Standard Errors of Estimates of Stomatal Conductance using Varying Numbers of Vines and Replicates per Vines for Set 4a

Vines	1	2	5	10	20
Replicates					
1	58.6	41.5	26.2	18.5	13.1
2	50.4	35.7	22.6	15.9	11.3
5	44.8	31.7	20.0	14.2	10.0
10	42.7	30.2	19.1	13.5	9.6
20	41.7	29.5	18.6	13.2	9.3

The time required for the measurements was 48 ± 11 seconds for measures on the same vine and 129 ± 73 seconds for measures between vines.

Set 5 Stomatal conductance (18/2/04)

There was little trend in the stomatal conductance as shown in Figure 6. Furthermore there was no relationship between the trend obtained from the stationary readings and the fixed values (r = 0.000).

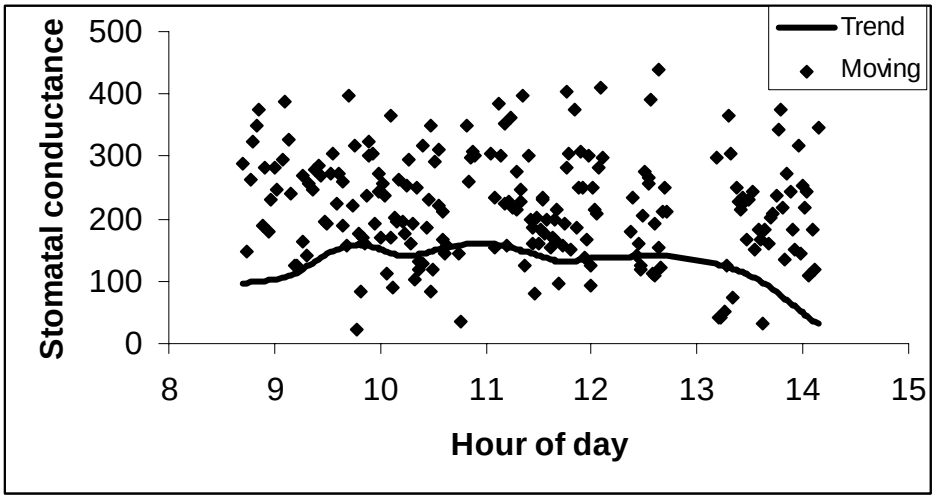


Figure 6 Changes of stomatal conductance from moving samples, and trend from stationary samples

A REML analysis indicated that there was no relationship between the time trend and stomatal conductance. Surprisingly there was no effect of the vines. Thus the moving readings can be described as random fluctuations about a mean, where the mean is 189 with a variance of 9839 ± 674 (SE = 99).

The analyses indicate that there is no need to sample a series of plants. Rather the standard error of the measure can be estimated from the equation

$$SE = 99/\mathrm{sqrt}(r),$$

where r is the number of replicates.

The time for a within plant reading was 51 ± 52 seconds and for the between vines was 141 ± 204 seconds. The optimal strategy is therefore to take many readings on each vine, and to visit few vines.

Set 6 Stomatal conductance (16/12/04)

Set A

There was a strong linear trend that could be described by the simple linear regression

Gs = 603.7 ± 24.3 – 35.39 ± 2.06

Which accounted for 40% of the variation. Fitting splines up to order 15 did not significantly improve on this relationship.

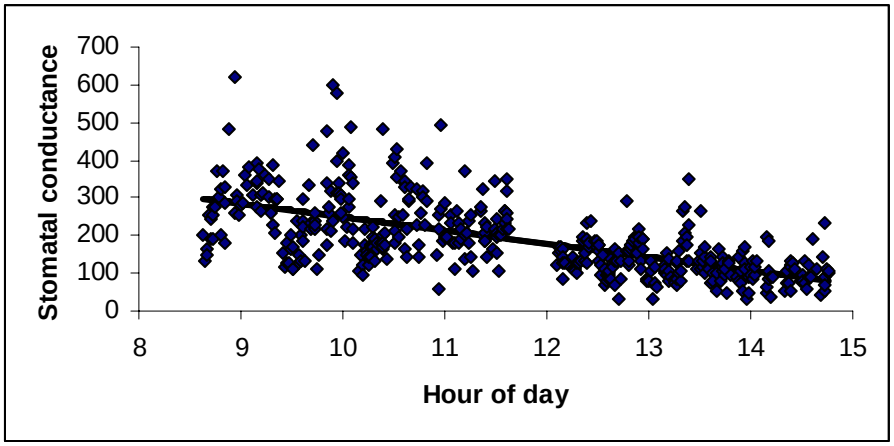


Figure 7 Set 6A changes in stomatal conductance across a day. The trend is represented by a linear regression

The REML analysis indicated a very significant trend. The components of variance (between groups and between leaves) are shown in Table 9. These estimates were used to derive the predicted standard errors shown in Table 10.

Table 9 Variance components for Set 6a

Source	Estimate	SE
Vine (Group)	1924	517
Leaf	3975	281

Table 10 Standard Errors of Estimates of Stomatal Conductance using Varying Numbers of Vines and Replicates per Vines for Set 6a

Vines	1	2	5	10	20
Replicates					
1	76.8	54.3	34.3	24.3	17.2
2	62.5	44.2	28.0	19.8	14.0
5	52.1	36.9	23.3	16.5	11.7
10	48.2	34.1	21.5	15.2	10.8
20	46.1	32.6	20.6	14.6	10.3

Set 6B

There was a temporal trend in the stationary measurements that was described by a fifth order spline as shown in Figure 8.

The REML analysis indicated a very significant trend. The components of variance (between groups and between leaves) are shown in Table 11. These estimates were used to derive the predicted standard errors shown in Table 10.

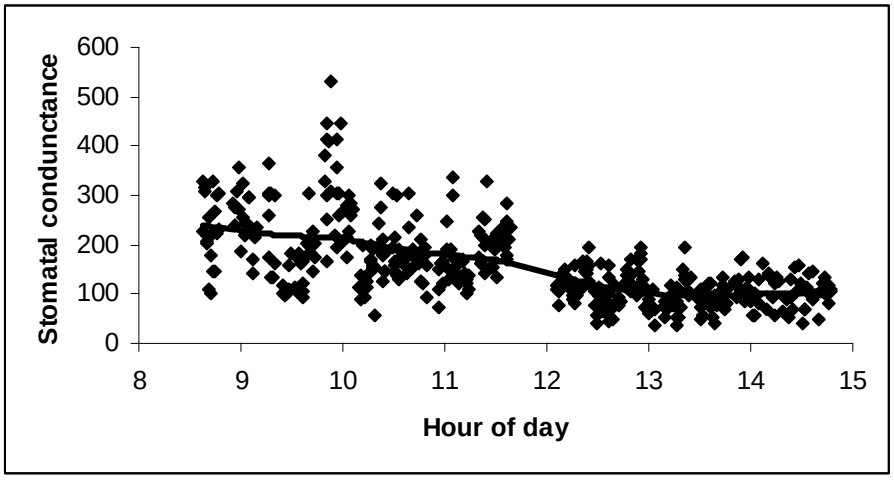


Figure 8 Set 6B changes in stomatal conductance across a day. The trend is represented by a 5th order spline

Table 11 Variance components for Set 6b

Source	Estimate	SE
Vine (Group)	1608	405
Leaf	2051	146

Table 12 Standard Errors of Estimates of Stomatal Conductance using Varying Numbers of Vines and Replicates per Vines for Set 6b

Vines	1	2	5	10	20
Replicates					
1	60.5	42.8	27.1	19.1	13.5
2	51.3	36.3	22.9	16.2	11.5
5	44.9	31.8	20.1	14.2	10.0
10	42.6	30.1	19.0	13.5	9.5
20	41.4	29.2	18.5	13.1	9.2

Table 10 and Table 12 indicate that as the number of replicates per vine increases, there becomes a limit when further replicates give no decrease in the standard error. This can be better seen in Figure 1.

Set 7 Stem and leaf water potential (8/2/05)

A comparison of the first and second readings of the stem and leaf water potential indicated that the first and second reading were very similar, with a standard deviation of the average value of each being 0.086.

A comparison of the stem and leaf water potential indicated that the leaf water potential was on average more negative by 0.039 ± 0.017.

There were distinct changes with time in the stationary measures as indicated in Figure 9. The same trend was apparent in the moving readings but the magnitude of the effect was much less.

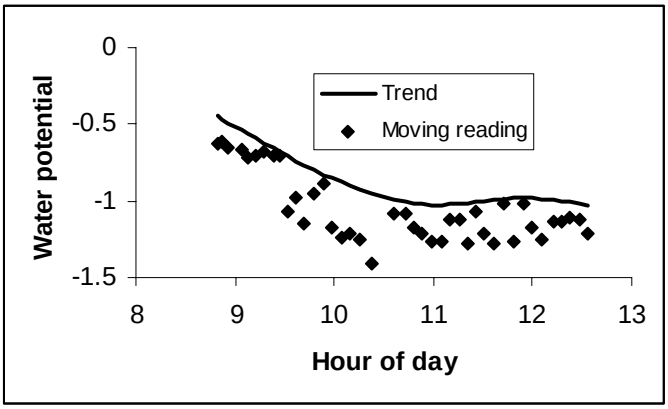


Figure 9 Comparison of temporal trend and observed changes in the moving readings of leaf water potential

The times required for the set of moving readings was seconds whereas the stationary set required 130 seconds.

The REML analysis for stem water potential indicated a strong relationship between the trend (as measured by the stationery readings) and the observed data. After that trend was removed the estimates of the variance components were as shown in Table 13.

Table 13 Variance components for Set 7 (stem water potentials)

Source	Estimate	SE
Vine	0.00384	0.00138
Noise	0.00390	0.00086

Similarly, for the leaf water potential there was a very significant trend component. After that trend was removed the estimates of the variance components were as shown in Table 14.

Table 14 Variance components for Set 7 (leaf water potentials)

Source	Estimate	SE
Vine	0.00978	0.00246
Noise	0.00321	0.00050

Table 15 Standard Errors of Estimates of Stem Water Potential using Varying Numbers of Vines and Replicates per Vines for Set 7

Vines	1	2	5	10	20
Replicates					
1	0.088	0.062	0.039	0.028	0.020
2	0.076	0.054	0.034	0.024	0.017
5	0.068	0.048	0.030	0.021	0.015
10	0.065	0.046	0.029	0.021	0.015
20	0.064	0.045	0.028	0.020	0.014

Table 16 Standard Errors of Estimates of Leaf Water Potential using Varying Numbers of Vines and Replicates per Vines for Set 7

Vines	1	2	5	10	20
Replicates					
1	0.114	0.081	0.051	0.036	0.025
2	0.107	0.075	0.048	0.034	0.024
5	0.102	0.072	0.046	0.032	0.023
10	0.101	0.071	0.045	0.032	0.022
20	0.100	0.071	0.045	0.032	0.022

Implications

The water potentials Set 1 had a large between vine component. To reduce the standard error it was necessary to sample many vines with few replicates on each vine. There was therefore no need to take more than one or two replicates per vine. By contrast Set 5 had little (estimated zero) between vine component. A comparison of the data sets is given in Table 17.

One strategy in choosing a sampling scheme is to achieve a given degree of precision for a minimum cost. No direct measure of cost was available, but time was considered a suitable surrogate. Estimates of time required for a repeat measurement on the same vine and for measures on different vines could be deduced for some data sets where times were recorded for every measurement. The times used are shown in Table 18.

In other cases time was recorded only once for a vine so it was then not possible to partition the time into between vine and within vine measures. To enable these not available (NA) data were substituted by values of 120 and 30 seconds respectively. Note that it is the ratio of these values that is important rather than the absolute values.

A minimisation routine can then be used to choose the number of samples that give the required accuracy for the minimum cost. These results are shown in Table19.

Table 17 Comparison of between vine and within vine variance components

Set	Measurement type	Between Vine	Measurement	Vine:Measurement
1	LWP	1.0797	0.447	2.42
2	PD - LWP	0.00436	0.00216	2.02
3	PD -LWP	0.00878	0.00216	4.06
4	SC	1648	1790	0.92
5	SC	0	9839	0.00
6a	SC	1924	3975	0.48
6b	SC	1608	2051	0.78
7a	Stem WP	1924	3975	0.48
7b	Leaf WP	1608	2051	0.78

Table 18 Information required for optimising sampling scheme

Set	Measurement type	Between Vine Time	Measurement time	Measurement mean
1	LWP	NA	NA	10.49
2	PD - LWP	NA	NA	-0.437
3	PD -LWP	NA	NA	-0.79
4	SC	129	48	133
5	SC	141	51	220
6a	SC	208	34	190
6b	SC	209	34	157

7a	Stem WP	NA	NA	-1.01
7b	Leaf WP	NA	NA	-1.05

Table 19 Optimal numbers of vines and samples per vine to achieve a standard error of no more than 5% of the mean

Set	Measurement type	Number of vines	Number per vine	SE(mean)	Measurement mean	Relative accuracy	Cost (time)
1	LWP	6	1	0.504	10.49	4.8%	780
2	PD - LWP	14	1	0.022	-0.437	4.9%	1820
3	PD -LWP	7	1	0.040	-0.79	5.0%	910
4	SC	57	2	6.679	133	5.0%	12825
5	SC	8	10	11.090	220	5.0%	5208
6a	SC	32	4	9.549	190	5.0%	11008
6b	SC	37	3	7.870	157	5.0%	11507
7a	Stem WP	2	3	0.051	-1.01	5.0%	380
7b	Leaf WP	4	3	0.052	-1.05	5.0%	760

Discussion

We desire to make recommendations of how to sample various parameters associated with water balance in a vineyard. One approach is to minimize the standard error of that measure for a given cost of measurement. Alternatively, we can choose a standard error that is acceptable to the worker and then seek a combination of numbers of vines and number of readings per vine that will achieve that standard error for a minimum cost.

Cost of taking additional measurements will be effectively time proportional to the time required. We can therefore in effect use measurement time as a surrogate of the cost of measurements. For each additional measure at a particular vine there is a small overhead - say half a minute. To move to another vine might cost say 2 minutes. These numbers will vary with the reading type.

Temporal variation was important in some cases, but not in others. For example, the pre-dawn water potential exhibited no temporal trend. This was as expected by the client. ON the other hand, stomatal conductance was affected by the time of measurement. This temporal trend could be removed to give a more precise set of measurements. In some cases the trend was simple and could be simply described by a linear trend. This trend can be removed in retrospect. Removal of a more complex trend can be achieved using a spline, but that requires a series of measures from a stationery station.

The presence of temporal trends poses the question as to at what time of the day should measures be taken. While inconvenient, pre-dawn water potential measures have the distinct advantage of not being affected by small changes in time.

It is also noted that the measures of variance components that have been estimated are only estimates, and the errors of these estimates that are quoted in this report have large error estimates. Whereas over estimation of the numbers of vines sampled will cost extra resources, an underestimate will give unstable estimates. It would therefore be wise to sample more than the 'optimal' number of vines to obtain stable estimates.

Conclusions

In conclusion, the following points are made

7. Temporal trends do occur except in the pre-dawn measures. Measures at other times will be difficult to compare both across sites and between times;
8. Stomatal conductance measures are much more variable than stem and leaf water potentials;
9. It is necessary to include a number of vines to obtain stable measures for all parameters – for some parameters, especially stomatal conductance, savings can be made by taking multiple (5-10) measures on the same vine;
10. For water potential, no more than two measures per vine should be taken.

References

Genstat for Windows. Version 7.0.1.198 Lawes Agricultural Trust.

Paterson HD and Thompson R (1971). Recovery of inter-block information when block sizes are unequal. Biometrika 58: 545-551.

Appendix 1a

Details of analysis of Set 1

Table 20 Analysis of moving readings

ANOVA

Source of Variation	SS	df	MS	F	P-value	F crit
Rows	402.7088	83	4.851913	14.67859	7.77E-28	1.437879
Columns	0.590486	1	0.590486	1.786408	0.185017	3.955961
Error	27.43511	83	0.330544			
Total	430.7344	167				

There was no significant effect between replicates.

Noise estimated as 0.3305 from Table 20.

Fitting a spline to the fixed data left a residual mean square of 0.25.

Set 6a had a value of -4324. This was replaced by the average of the remaining values in the group.

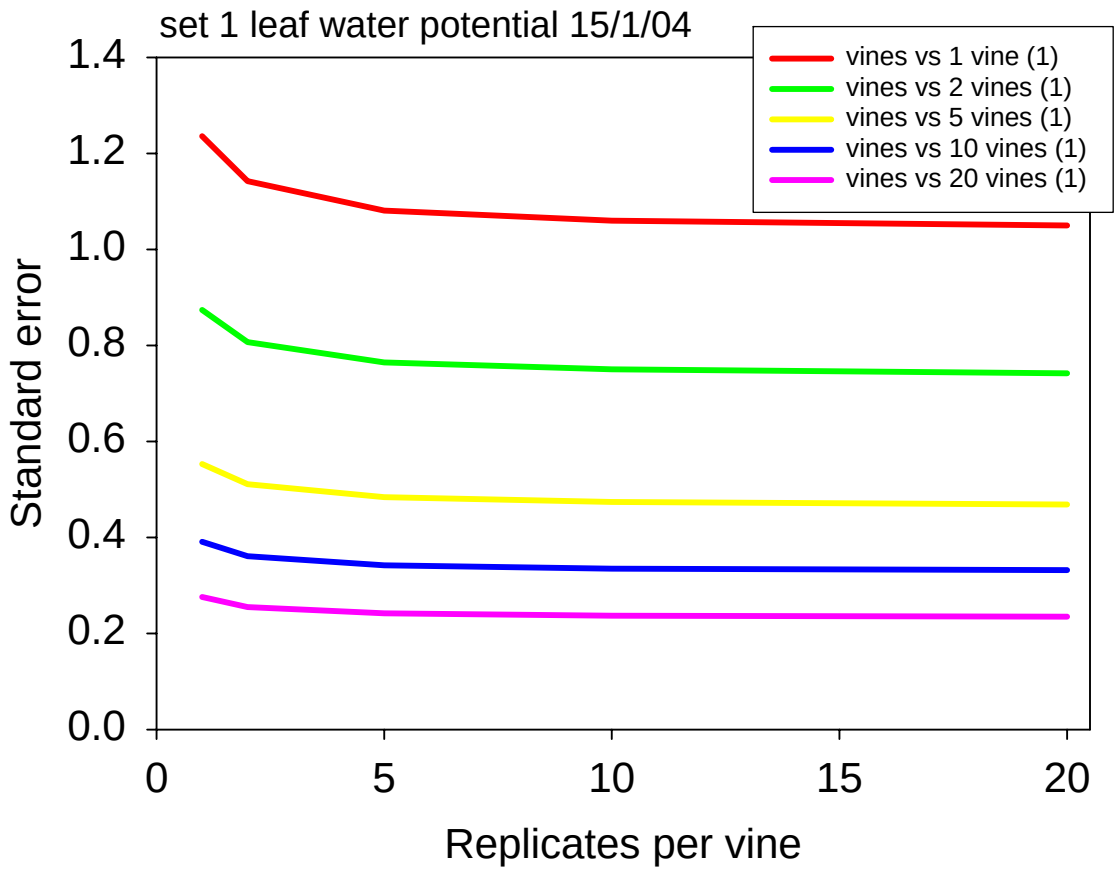
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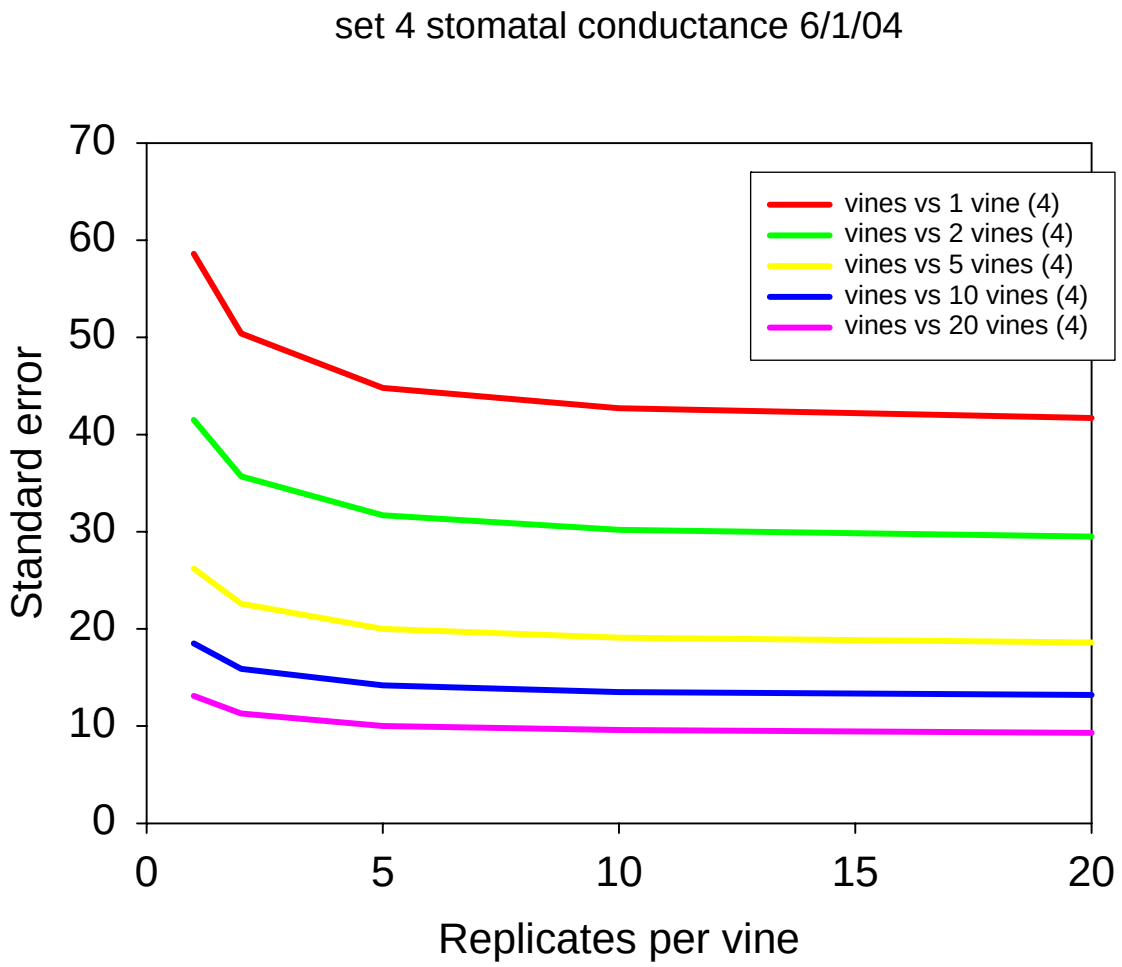
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vcomponent[spline=Time]
reml Fixed

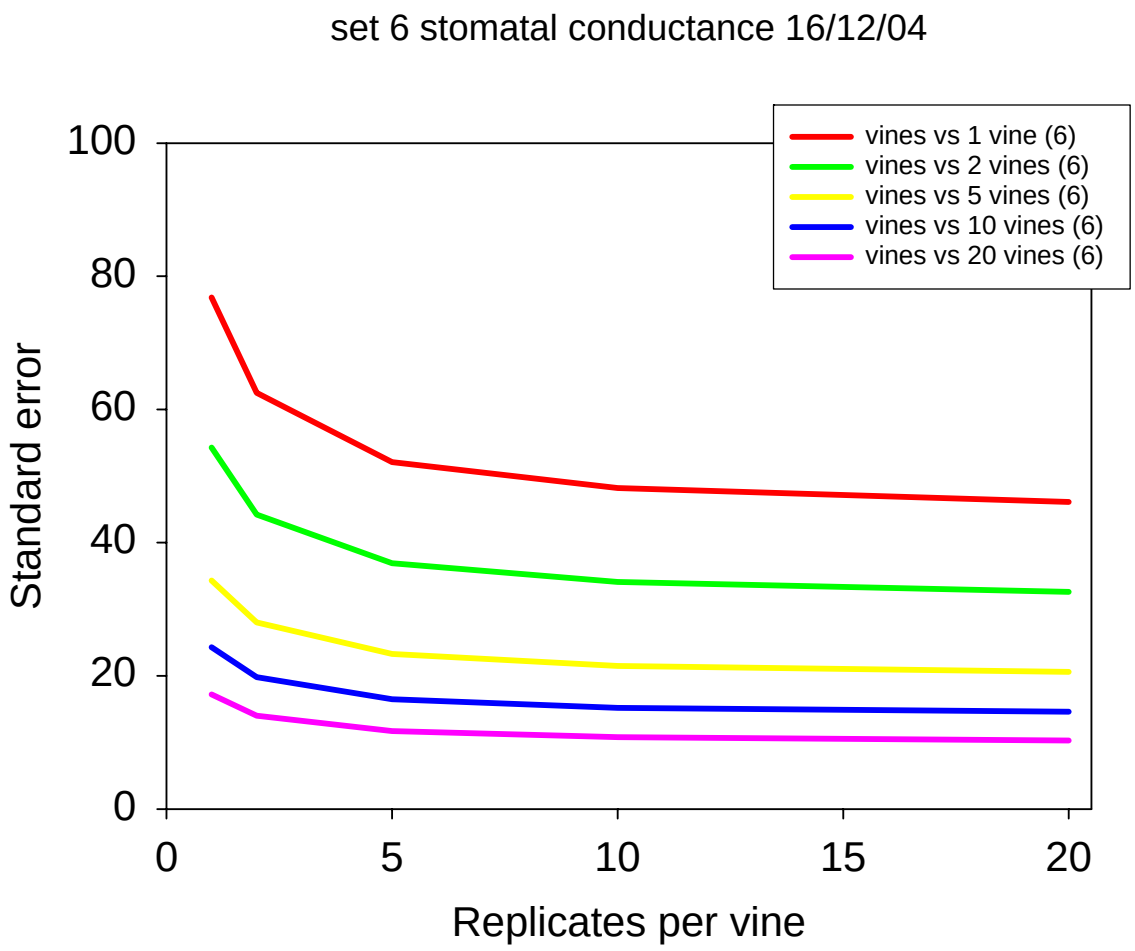
model Fixed;f=f
for d=2...10
fit sspline(Time;d)
endfor

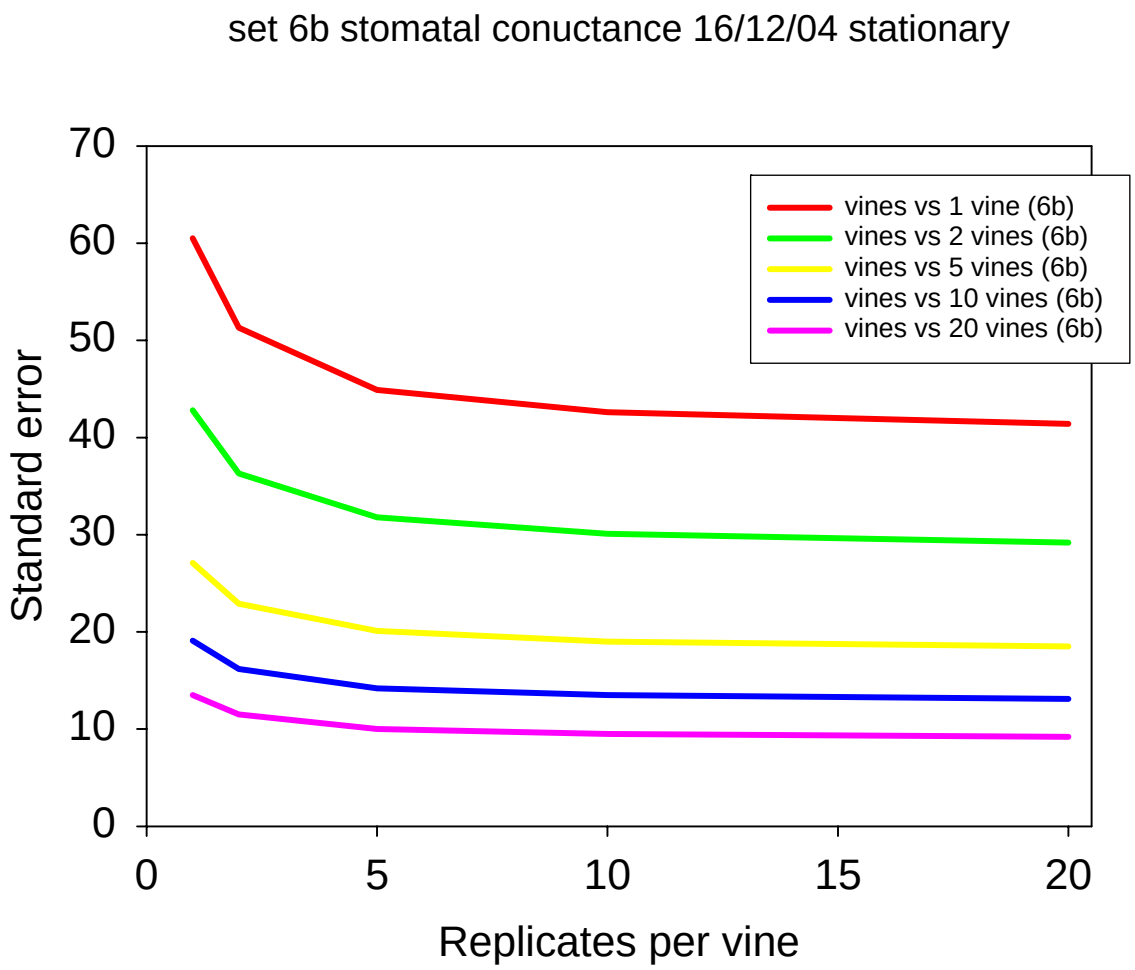
fit sspline(Time;5)
print Time,Fixed,f
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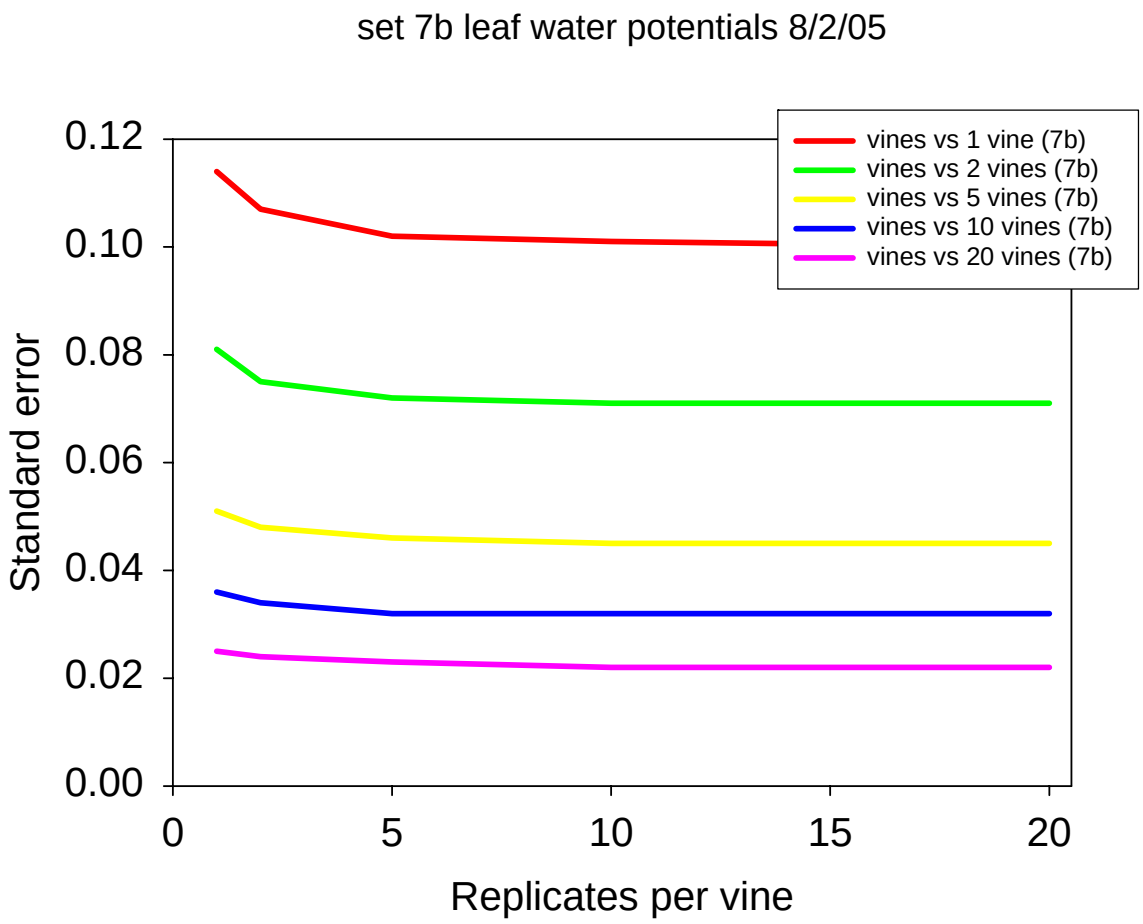
There follows graphical representation of the relationships between the effect of increasing number of replicates per vine for different numbers of vines, as outlined in the tables above.




set 4 stomatal conductance 6/1/04
Standard error
70
60
50
40
30
20
10
0
0
5
10
15
20
Replicates per vine
vines vs 1 vine (4)
vines vs 2 vines (4)
vines vs 5 vines (4)
vines vs 10 vines (4)
vines vs 20 vines (4)


set 6 stomatal conductance 16/12/04
vines vs 1 vine (6)
vines vs 2 vines (6)
vines vs 5 vines (6)
vines vs 10 vines (6)
vines vs 20 vines (6)
Standard error
100
80
60
40
20
0
0
5
10
15
20
Replicates per vine


set 6b stomatal conuctance 16/12/04 stationary
Standard error
Replicates per vine
0
10
20
30
40
50
60
70
0
5
10
15
20
vines vs 1 vine (6b)
vines vs 2 vines (6b)
vines vs 5 vines (6b)
vines vs 10 vines (6b)
vines vs 20 vines (6b)


set 7b leaf water potentials 8/2/05
vines vs 1 vine (7b)
vines vs 2 vines (7b)
vines vs 5 vines (7b)
vines vs 10 vines (7b)
vines vs 20 vines (7b)
Standard error
0.12
0.10
0.08
0.06
0.04
0.02
0.00
0
5
10
15
20
Replicates per vine

Appendix 2. Copy of certificate given to staff at Rosemount Estate Vineyards at Langhorne Creek in appreciation of the significant help given in the completion of this project

