

A non-invasive plant-based probe for continuous monitoring of water stress in real time: a new tool for irrigation scheduling and deeper insight into drought and salinity stress physiology

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ABSTRACT: The non-invasive, magnetic leaf patch clamp pressure probe (also termed ZIM-probe) allows for the first time to measure continuously turgor pressure changes of plant leaves over long periods of time with high precision and in real time. The probe has become an important tool in plant physiology, molecular biology and ecology, but also in agriculture because the probe is very robust and user-friendly. Growers receive the information about the water status of their plants by wireless telemetry, mobile network and internet on an as-needed basis and can thus adjust very precisely both the timing of irrigation and the quantity of water to apply. Effects of air and leaf temperature, relative humidity, illumination and wind on turgor pressure can be monitored very sensitively both under indoor and outdoor conditions. Even the effects of blue and red light as well as of oscillations of stomata aperture on turgor pressure can be monitored by the probe with high sensitivity. Similarly, water deficit due to increase of the osmotic pressure in the nutrition solutions resulted in significant changes of the probe signals. Multiple probe readings open up new possibilities to resolve (together with other techniques) the mechanisms of short- and long-distance water transport, particularly how plants can cope with water shortage. The applications of the magnetic probe are numerous and one can expect highly interesting developments in plant water relations in the nearest future.

KEYWORDS: drought, leaf patch clamp pressure probe, osmotic pressure, salinity, water relations, ZIM-probe.

INTRODUCTION

Increasing worldwide shortages of fresh water, the continuous increase in water consumption by agriculture due to climate change, the increased demand for crop production to feed the world's increasing population and the progressive salinization of arable land provoked by irrigation are global problems. The ways in which plants respond to water shortage and other environmental constraints depends on their hydraulic architecture which controls plant water relations, in particular the balance between transpirational water loss, internal water reallocation and soil water uptake. The broad arrays of interacting mechanism controlling water uptake through the roots, long-distance transport in the xylem, water storage in tissue cells and fruits, and transpirational water losses have highlighted the need of sensor technology for monitoring continuously and in real time the water supply of herbaceous plants and trees, especially under quite variable field conditions.

The water status of plants is usually determined by using the pressure chamber (Scholander et al. 1965). An excised leaf or leafy twig is placed into a steel chamber with the cut end protruding through a pressure-tight seal to the ambient atmospheric pressure. Gas pressure is applied to the specimen and the overpressure at which water appears at the cut surface, the so-called balancing pressure, P_b (defined in relation to atmosphere), is postulated to be numerically equal to the xylem tension, P_x , (that existed before cutting) or to the leaf water potential, $P_c - \pi_c$ (where P_c is the turgor pressure and π_c the cellular osmotic pressure) assuming local equilibrium between the xylem and the tissue cells. The method is simple and thus very popular, but massively invasive and time-consuming. It does usually only allow spot measurements (e.g., at predawn or midday), but not continuous recordings. Furthermore, the number of leaves that can be measured is rather limited and, therefore, data can be misrepresentative of the overall *in situ* conditions (due to variability in height, light irradiation exposure, microclimate conditions, oscillations of stomata aperture etc.). Most important, and frequently ignored, the pressure chamber readings do not yield absolute, but rather relative values of xylem pressure or leaf water potential (Rüger et al. 2010a). Xylem pressure and cell turgor pressure (being linked hydraulically to each other in turgescence cells) can quite accurately be measured by the microcapillary probe techniques pioneered by Zimmermann and co-workers (Zimmermann et al. 1969, Balling and Zimmermann 1990). Even though these highly sophisticated techniques are minimal-invasive and allow very accurate measurements on the single xylem vessel or cell level of intact plants, they are not suitable for long-term outdoor applications. The same applies to ball tonometry, a non-destructive method by which cell turgor pressure is measured by application of an external pressure (Lintilhac et al.

2000, Geitmann 2006). Measurements of leaf thickness have also been used as an indirect indicator of the turgor pressure of leaves (e.g., Búrquez 1987, McBurney 1992, Malone 1993). However, these techniques have not found widespread applications for indoor and outdoor experiments because of the frequent insensitivity of leaf thickness to changes in the water status of the leaves and the anisotropy of leaf shrinkage (Jones 2007).

The non-invasive leaf patch clamp pressure probe (so-called ZIM-probe) recently introduced by Zimmermann and co-workers (Zimmermann et al. 2008, Westhoff et al. 2009a) has apparently overcome the impasse in measuring continuously the temporal and spatial dynamics of plant water relations in real time both and with high precision under laboratory and field conditions. The probe measures relative changes in leaf turgor pressure, i.e. the key water relation parameter that determines growth, fruit productivity and quality. The magnitude of turgor pressure is dictated by root water uptake, transpirational water loss and by the cellular osmotic pressure which, in turn, depends on intracellular solute (fertilizer) accumulation.

Innumerable plant water relation and field case studies performed in the last four years worldwide after the disclosure of the magnetic probe technology to the scientific world have emphasized the enormous potential of the magnetic probe technology for basic research in plant physiology and ecology as well as for water management in agriculture and forestry. Probing of leaves (together with dendrometer and sap flow measurements) provides a powerful key to decode short- and long-distance water transport in wild type plants and (genetically engineered) mutants, to screen efficiently for drought-tolerant genotypes (phenotyping in plant breeding) and to improve greatly irrigation scheduling in agriculture by saving water and increasing productivity while simultaneously reducing soil salinization.

In this communication the authors will focus on some of the most important results obtained in last few years using the magnetic probe technology. It is hoped that the evidence reviewed here will stimulate further experimentation. Needless to say, the applications are manifold and exciting because the surface has barely been skimmed.

PRINCIPLE OF THE MAGNETIC TURGOR PRESSURE PROBE

The measuring principle of the magnetic turgor pressure probe (ZIM-probe, ZIM Plant Technology GmbH, Hennigsdorf, Germany) is quite simple (Figure 1): a small three-dimensional leaf patch is used as an "artificial sensing compartment" for measuring relative changes of turgor pressure in the entire leaf tissue (Zimmermann et al. 2008, Westhoff et al.

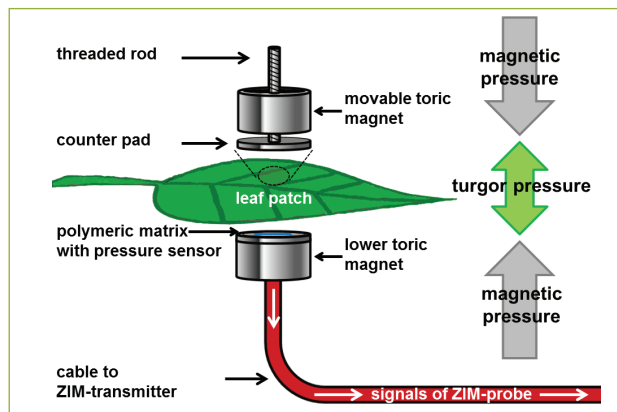


Figure 1. Schematic diagram of the measuring principle of the magnetic ZIM-probe.

2009a). This requires that the stomata of the patch are closed and that the patch is in hydraulic contact with its surrounding. Patches of (partly) dried-out leaves may not meet these demands because air prevents or at least reduces rapid pressure exchange between the patch and its surrounding. The “sensing compartment” is achieved by clamping an intact leaf between two planar circular pads made up of non-transparent material (metal or plastic). The pads are integrated into two toric magnets. One pad contains a receptacle in which the pressure sensor chip coated with a blue dye-stained polymeric substance is integrated. The magnet of the counter-pad contains an inside thread that could be moved along a threaded rod (see Figure 1). The variation in the distance between the counter-pad and the movable magnet allows adjustment of the applied magnetic force, P_{clamp} according to leaf rigidity and elasticity.

Leaf turgor pressure is determined by measuring the pressure transfer through the leaf patch, i.e. by measuring the output pressure, P_p , upon application of the constantly kept, magnetic clamp pressure, P_{clamp} (up to 450 kPa). A detailed thermodynamic analysis (Zimmermann et al. 2008) has shown that P_p is a power function of the turgor pressure P_c (i.e. both parameters are inversely coupled to each other):

$$P_p = \left(\frac{b}{aP_c + b} \right)^{\frac{1}{a}} \cdot F_a \cdot P_{\text{clamp}} \quad (1)$$

where a and b are constants. F_a is a leaf-specific attenuation factor which takes into account that only a constant fraction of P_{clamp} is arriving at the cell level because of attenuation of the pressure due to compression of leaf-specific, turgor pressure-independent structural elements (e.g., air spaces, cuticle and cell walls). F_a is dimensionless and is usually of the order of 0.2 to 0.4. Due to the design of the probes and the properties

of the polymer F_a is practically constant at turgor pressures >50 kPa. Below ca. 50 kPa the large air-filled spaces in the leaf becomes the dominant factor and control mostly the probe signal (see further below).

Even though the different parts of the probes exhibit different extension coefficients with temperature, the systematic error is <2 kPa when subjected to temperature regimes ranging from 10 °C to 30 °C in a climate chamber. Consistent with this, pressure probes mounted in a non-contact mode close to the leaf surface under field conditions showed negligible pressure changes over large temperature ranges.

Eq. 1 shows that the patch pressure, P_p is inversely coupled with leaf turgor pressure, P_c as expected since P_c is opposing P_{clamp} . This means that P_p is small when P_c is large and *vice versa* large, when P_c is small. This relationship could be verified by calibration of the ZIM-probe against the microcapillary cell turgor pressure probe (see review article Zimmermann et al. 2004). However, it is important to note that Eq. 1 only holds, i.e. that the probe signals reflect only turgor pressure changes, if the contact between leaf patch and the pads is homogenous. Placing of the measuring, blue-stained polymeric area of the probe on the mid-vein or other elevated leaf surface elements results in erroneous measurements because thickness changes of these structural elements are superimposed on the changes of the turgor-pressure dependent pressure transfer through the leaf.

The ZIM-probes are connected by cable to a transmitter. If desired, probes for measuring air temperature, relative humidity and light intensity close to the plant, as well as soil temperature and moisture, can also be connected to the transmitters thus giving a complete view about the microclimate and the soil properties. ZIM-probe and environmental data are sent wireless by the transmitter (telemetric units) over a distance of up to 1500 m to a controller. The controller contains a GPRS modem which is linked to an Internet server *via* the local mobile phone network. From the server the data can be downloaded by smartphones, tablets or laptops. Thus, leaf water status is available for the user in real time and a grower can make adequate irrigation decisions on an objective basis when they are needed.

MAGNETIC PROBE VERSUS PRESSURE CHAMBER MEASUREMENTS

Even though patch pressure, P_p measurements are performed on a single leaf level, whereas the balancing pressure values, P_{ch} measured with the pressure chamber data are based on multiple leaves, there is generally a quite good overall correlation between both parameters (Figure 2; Westhoff et al. 2008, Zimmermann et al. 2010, Bramley et al. 2012). Concomitant direct measurements of the cell turgor on many plant species (using the microcapillary cell turgor pressure

probe) have revealed that the dependency of both parameters on cell turgor pressure can be fitted by Eq. 1, demonstrating that both techniques are measuring relative changes in turgor pressure (and xylem pressure, respectively) or relative changes in leaf water potential (provided that the osmotic pressure can be assumed to be constant; R ger et al. 2010a, Bramley et al. 2012). This is at variance with the common interpretation of balancing pressure values. In the literature P_b values are usually interpreted in terms of absolute values of leaf water potential or xylem pressure values.

Concomitant probe and pressure chamber measurements on many plant species have shown that the pressure chamber gives a rough overview about the water status of plants. However, the spatial and temporal dynamics of water loss and water reallocation within the plant as well as regulatory processes of stomata aperture that can be measured with high accuracy by the ZIM-probe system (see below) cannot be resolved by the pressure chamber. This technique also fails completely, if small-sized plants (e.g. *Arabidopsis thaliana*) are investigated. It also leads to erroneous results if the xylem vessels contain gel-like compounds (mucopolysaccharides) and/or the leaves contain mucilage containing epistomatal plugs by which water loss and moisture uptake from the atmosphere is regulated (Zimmermann et al. 2004, 2007, Westhoff et al. 2009b). Excessive overpressures are needed to squeeze out water from mucilage-containing leafy twigs. Balancing pressure can also not be determined in plants exhibiting resin exudation [for example maple (*Acer* spp.); see also Ritchie and Hinckley, 1975].

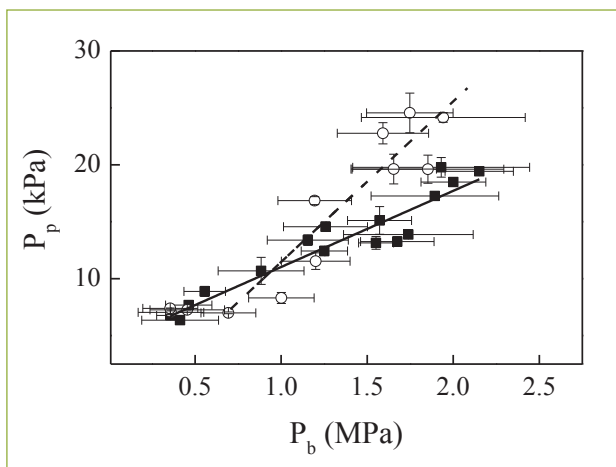


Figure 2. Typical plots of patch pressure (P_p) values measured with the ZIM-probe on beech (*Fagus sylvatica*, filled squares) and eucalyptus (open circles) trees under field conditions against the corresponding balancing pressures (P_b) values measured with the pressure chamber. Bars of the P_b and P_p values represent the SD of at least three measurements (beech: $r^2=0.97$; eucalyptus: $r^2=0.78$).

* Note that pressure probes mounted in a non-contact mode close to the leaf surface showed no P_p changes indicating that they resulted from effects of temperature on stomata and in turn, on turgor pressure.

EFFECTS OF MICROCLIMATE ON TURGOR PRESSURE

The regulation of stomata aperture by plant water status, illumination and internal CO_2 availability were extensively studied, while the response of stomata to elevated ambient temperature, T , is far less documented (Feller 2006). Climate models predict for the next decades in average increased temperatures worldwide. Effects of ambient temperature on plant stomata and subsequently on leaf turgor pressure are, therefore, of high scientific and agricultural interest. Temperature effects on turgor pressure can be recorded precisely and in real time (see measurements on sugarcane (*Saccharum* spp.) in Figure 3A) and they can easily be distinguished from light (as well as from relative humidity) effects as shown for wheat (*Triticum aestivum*; Figure 3B)*. The graphs show that an increase in temperature increases opening of the stomata as manifested in an increase of the P_p values. However, the effects are less than those of illumination (at least in the investigated temperature range). Detailed studies of the light response of the guard cells have shown that there are two separated reactions: a specific blue-light response and a photosynthetic reaction in the chloroplasts of the guard cells (Schwartz and Zeiger, 1984). This could be verified for the wild type *Arabidopsis thaliana* by measuring the response of P_p to low blue-light photon flux density under a saturated red-light background (Figure 3C; R ger 2008; republished by Ache et al. 2010). Stomata opening of *A. thaliana* also increases with increasing ambient temperature and decreases with increasing relative humidity, as demonstrated by probe measurements (R ger 2008, Barrag n et al. 2012). NHX *Arabidopsis* mutants showed opposite responses to illumination (and to changes in relative humidity). The results were in agreement with the inability of *nhx1* and *nhx2* mutant plants to osmoregulate, demonstrating that the magnetic probe is a valuable tool for monitoring the physiological response of *Arabidopsis* mutants (Barrag n et al. 2012).

EFFECTS OF STOMATA APERTURE OSCILLATIONS ON TURGOR PRESSURE

Stomata aperture oscillations lead to oscillations in the transpiration rate (Farquhar and Cowan 1974, Raschke 1975). Plants save water by the periodic reduction of transpiration, but at the expense of photosynthesis. This affects biomass production and ultimately crop yield. Stomata aperture oscillations are reflected in oscillations of xylem and cell turgor pressure. This could be verified by measurements using the minimal-invasive microcapillary cell turgor and xylem pressure probes

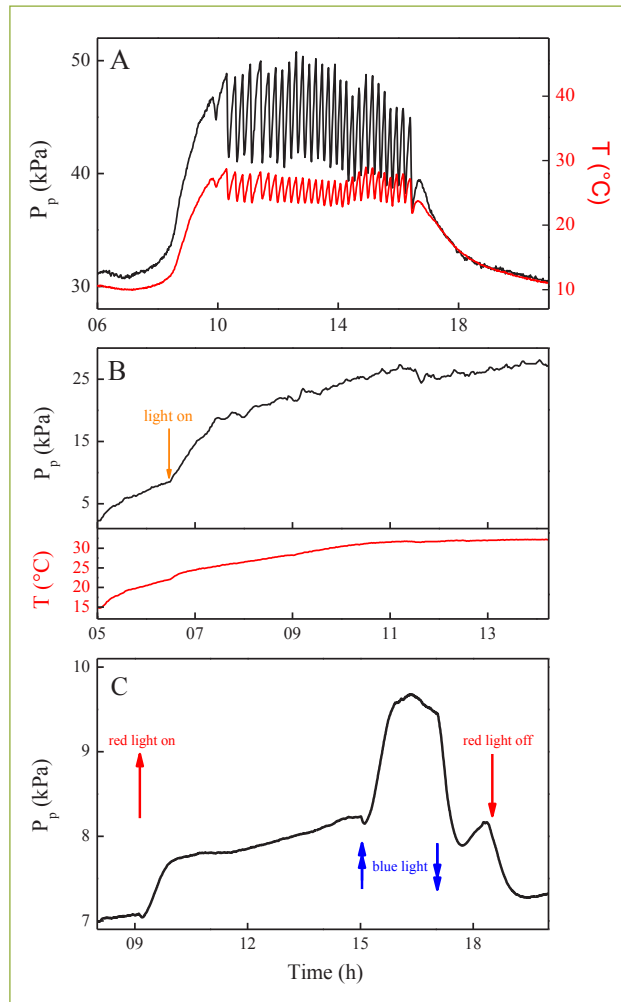


Figure 3. Light and temperature effects on the patch pressure (P_p) recorded under laboratory and greenhouse conditions. **A:** Effects of temperature (T ; red line) fluctuations on P_p measured on sugarcane. **B:** P_p changes of wheat leaves in response to temperature and light changes (top panel) and the corresponding ambient temperature (lower panel; red line). The orange arrow indicates when the light was switched on. Note that the slope of the P_p curves change significantly after switch on of the light. **C:** P_p changes of *Arabidopsis* leaves upon illumination with red ($200 \mu\text{mol m}^{-2} \text{s}^{-1}$; red arrows) light and superimposed blue ($18 \mu\text{mol m}^{-2} \text{s}^{-1}$; double-headed blue arrows) light. For further details, see Ruger (2008; republished by Ache et al. 2010).

(Zimmermann et al. 2002, 2004). As indicated in Figure 4 the ZIM-probes provide the same information as the highly sophisticated microcapillary probes, but with the important difference that measurements can be performed in the field (Zimmermann et al. 2010, 2013). Studies on banana plants (*Musa acuminata*) and grapevine (*Vitis vinifera*) as well as on citrus (*Citrus* spp.) and olive trees (*Olea europaea*) showed that there was a strong correlation between wind speed and the amplitude of the P_p oscillations (Zimmermann et al. 2010,

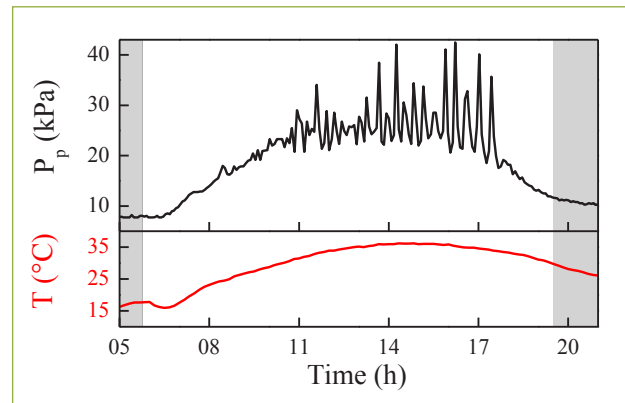


Figure 4. Typical P_p oscillations (period: 20.3 ± 3.3 min; $n=14$) measured on a leaf of an orange tree irrigated with low salinity water (21.6 mM) together with the corresponding changes in temperature (lower panel; red line). Nocturnal hours are marked by grey bars.

unpublished data). Accordingly, plants growing in the centre of the plantations and protected against the wind did not — if any — show strong oscillations.

For water management and irrigation improvement in agriculture it is interesting to note that immediate watering of plants after the onset of stomata aperture oscillations has resulted in a significant increase in harvest (up to 31%, unpublished data). For scientific reasons and for the application of the probe in drought stress physiology, probe measurements open up new possibilities to study the cascade of secondary physiological reactions triggered by the stomata-induced xylem and turgor pressure changes. For example, Wegner and Zimmermann (1998; see also review article of Zimmermann et al. 2004) using the miniaturised xylem pressure-potential probe have demonstrated for wheat and maize (*Zea mays*) that the xylem and turgor pressure changes induce synchronous changes in the trans-root electrical potential, presumably due to electrokinetic effects on the cell/tissue level close to the xylem. Electric potential changes have also been detected recently between the base of the stem and upper leaf of avocado trees (*Persea americana*) in response to drought, irrigation and diurnal light/dark regimes (Fromm 2006, Davies 2006, Gurovich and Hermosilla 2009). Gurovich and others discuss propagation of the electric signals through the tree, but ignore that pressure propagation through the xylem vessels and tissue is most likely the primary event that leads subsequently to electrical changes on the cellular level (see also Westhoff et al. 2008, 2009b). No doubt, concomitant probe and electrical potential measurements will contribute to the elucidation of signals causing stomata closure, i.e., primary hydraulic event and the secondary electrical responses. The results of such measurements may also have a great impact on the controversial discussion about the mechanism involved in long-distance wound signalling.

LEAF TEMPERATURE AND DROUGHT

Even though ambient air temperature is usually measured, leaf temperature is the essential parameter from a physiological point of view that affects stomata opening as well as the physiological processes of the leaves. Transpiring leaves have a temperature lower than the temperature in the surrounding because of the cooling effect by transpiration. By contrast, the temperature of non-transpiring leaves or sun-exposed leaves can exceed air temperature under some circumstances (see e.g. Ribeiro et al. 2005, Feller 2006). Leaf temperature is, therefore, a good estimate for the transpiration rate. Leaf temperature is usually measured with infrared thermometer or with infrared thermography, i.e. with quite sophisticated technology. Problems can arise — among other things — when the leaves are quite small (e.g. in the case of wheat) because of interferences of leaf transpiration with evaporation of water from the soil. These disadvantages are not shared by ZIM-probes in which a temperature sensor is integrated. Preliminary experiments on leaves of oak trees (*Quercus robur*) have shown that the surface temperature of transpiring leaves were by about six degrees lower than the surface temperature of non-transpiring leaves which was practically as high as the air temperature (Figure 5). Apart for the physiological relevance, simultaneous and continuous acquisition of turgor pressure changes together with leaf temperature obviously provides a strong basis for making irrigation decisions on an as-needed basis.

TURGOR PRESSURE RECOVERY POTENTIAL AFTER TRANSPIRATIONAL WATER LOSS

The rate of the decrease in patch pressure after noon peaking (or after switch off of the light under laboratory conditions) increases with proceeding drought and is, therefore, a very good indicator for water stress. The turgor pressure recovery phase can usually be fitted by an exponential function. The relaxation time, τ (the time constant of the exponential return of P_p to the night

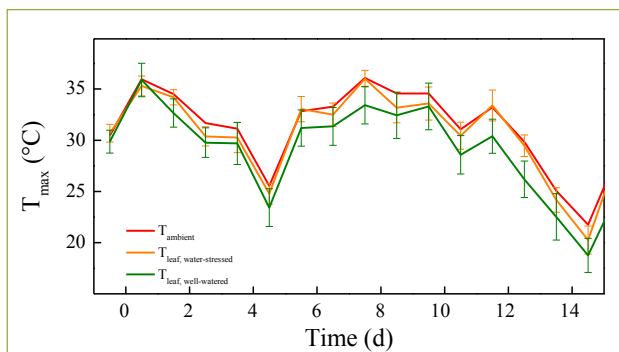


Figure 5. Plot of the local daily maximum air temperatures (red line) and the daily maximum leaf temperatures measured with the modified ZIM-probe. Measurements were performed on well-watered (green line) and water-stressed (orange line) oak trees growing in a forest ($n=8 \pm SE$ per treatment).

value; 63% of the value of P_p peaking) or the half-time $T_{1/2}$ (50% of the value of P_p peaking) are quantitative parameters that can be used for the characterisation of the water status (Zimmermann et al. 2010, Bramley et al. 2012). Plots of P_p versus air temperature or — more accurate — leaf temperature gives also fast insight into the turgor recovery potential of the leaves after noon P_p peaking. Depending on the water supply of the leaves different patterns between P_p and T as the temperature increased in the morning and decreased in the afternoon are measured (Figure 6): (1) $P_p = f(T)$ curves measured during the morning hours coincide with those measured during the afternoon hours within the limits of accuracy (no inverse hysteresis) and (2) turgor pressure recovery occurs significantly faster with decreasing temperature during the afternoon than turgor pressure loss with increasing temperature during the morning hours. Faster return to low P_p values relative to temperature during the afternoon indicates that sufficient water is available for turgor pressure recovery. Different levels of inverse hysteresis indicate different degree of water availability (for quantification, see Bramley et al. 2012). However, it has to be noted that an instantaneous drop of P_p in the afternoon to low values (pretending a high inverse hysteresis and, in turn, high water availability) are a clear-cut indication that the leaves contain a lot of air and/or that the xylem vessels are blocked by embolism (Ehrenberger et al. 2012).

DROUGHT AND SALINITY EFFECTS ON TURGOR PRESSURE

Proceeding drought stress manifests as increasing daily P_p amplitudes and frequently as a rise in the night P_p values. An example is given in Figure 7A for a premium grapevine in Chile. Irrigation leads to a decrease of the patch pressure in the following days. Simultaneously, the time constant, τ , of the turgor pressure recovery phase is increasing (see above). The daily increase in the P_p values has been observed in a huge number of crop species (e.g., wheat, maize, canola (*Brassica napus*), grapevine, citrus, olive, banana, forest trees etc.; Westhoff et al. 2008, Ruger et al. 2010b, Zimmermann et al. 2010, 2013, Bramley et al. 2012) and can clearly be correlated with increasing loss of leaf turgor following watering termination. The upward shift in the baseline signal, i.e. an increase in the minimal values at night indicates incomplete leaf rehydration. Salinity or, generally speaking, high osmotic pressure of the nutrition solutions or of the water used for irrigation leads to identical changes in the P_p values and τ . Figure 8 shows model experiments performed on sugarcane by using PEG as an osmoticum.

In the case of some species, e.g. olive, orange (*Citrus sinensis*), almond (*Prunus dulcis*), eucalyptus (*Eucalyptus pilularis*) and avocado trees, a change in the shape of the P_p curves occurs upon approaching turgor pressure zero, i.e. P_p assumes minimum values

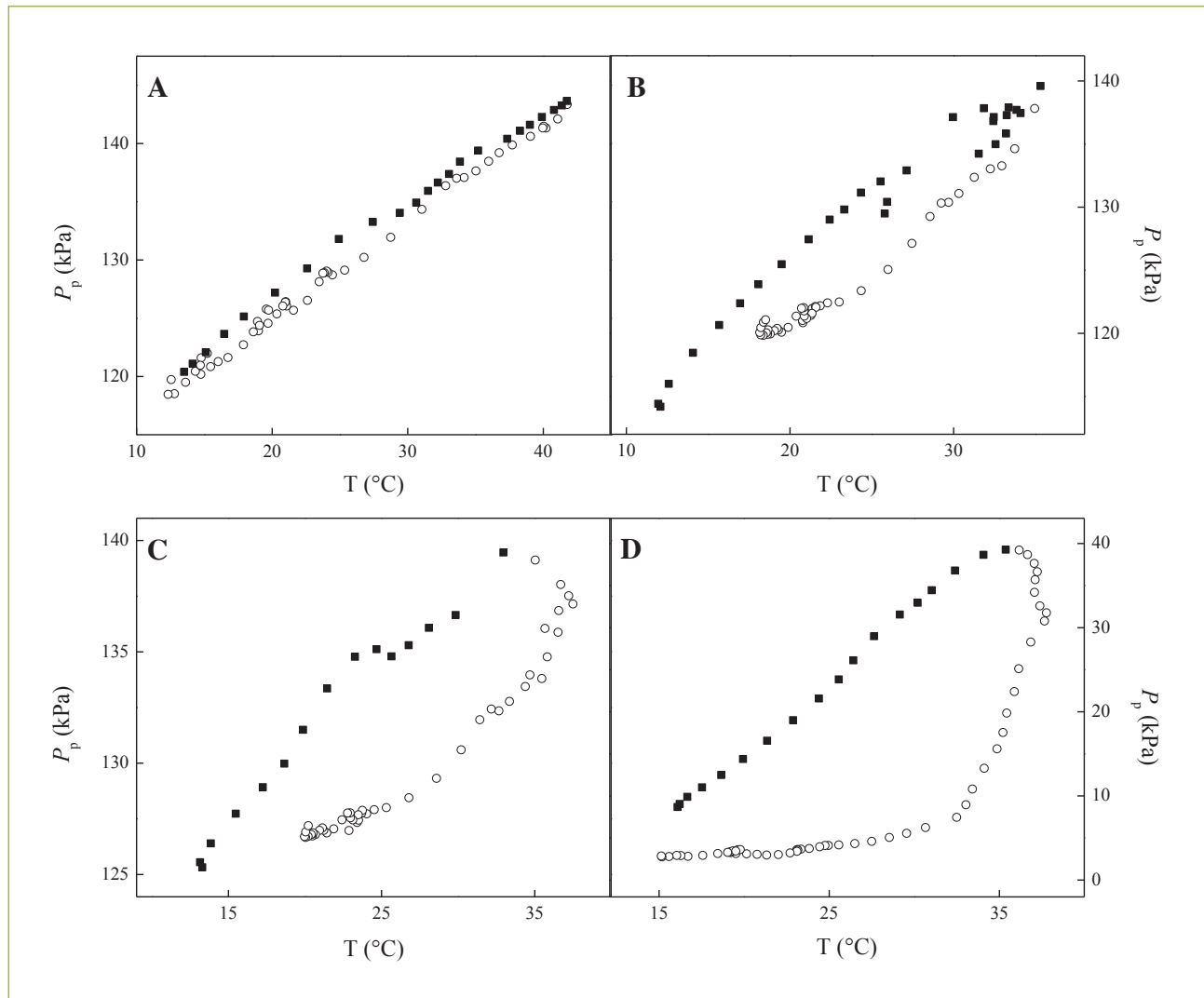


Figure 6. Typical schematic patterns between P_p and T as the temperature increased in the morning (filled squares) and decreased in the afternoon (open circles). **A:** $P_p = f(T)$ curves recorded on a plant subjected to mild water stress (field conditions; no inverse hysteresis). **B and C:** Increased levels of inverse hysteresis indicate increased degree of water availability or stress. **D:** $P_p = f(T)$ curves recorded on a leaf subjected to severe water stress as indicated by the rapid drop of P_p with decreasing T after noon peaking (presumably due to the disruption of the continuous water columns in the xylem by cavitation and/or embolism). For evaluation of the inverse hysteresis see Bramley et al. (2012).

during the day and maximum values during the night (Figure 7B). The reason for this is most likely a very unfavourable water to air ratio within the leaves (for details, see Ehrenberger et al. 2012). At turgor pressure values close to zero the compression of the large air-filled spaces mostly determines the response of the probe; the attenuation factor F_a in Eq. 1 is no longer constant and becomes the dominant factor. Transition states which can be recorded for a couple of days (depending on the species and drought) are usually observed before the inverse P_p curves are recorded (half-inverse curves, see e.g., Fernández et al. 2010, Bramley et al. 2012). Interestingly, these changes in the P_p curves are not reflected in sap flow as shown by concomitant sap flow measurements on olive trees (Rodríguez-Domínguez et al. 2012).

From a water management standpoint, the occurrence of (half-) inverse P_p curves is an important indicator for irrigation, because leaves and/or branches are irreversibly damaged.

HYDRAULIC PLANT FUNCTIONING

Multiple probing of leaves opens up new avenues to explore the temporal and spatial dynamics of water transport and reallocation in plants, particularly in trees. The patch pressure P_p displayed diurnal patterns in the field which depended on height, sun-exposure and water reallocation (Figure 9). Among other things, measurements on avocado, banksia (*Banksia menziesii*) and eucalyptus trees in Australia showed (Rüger et al. 2010a, Zimmermann et al. 2013, Bader et al, unpublished results) that

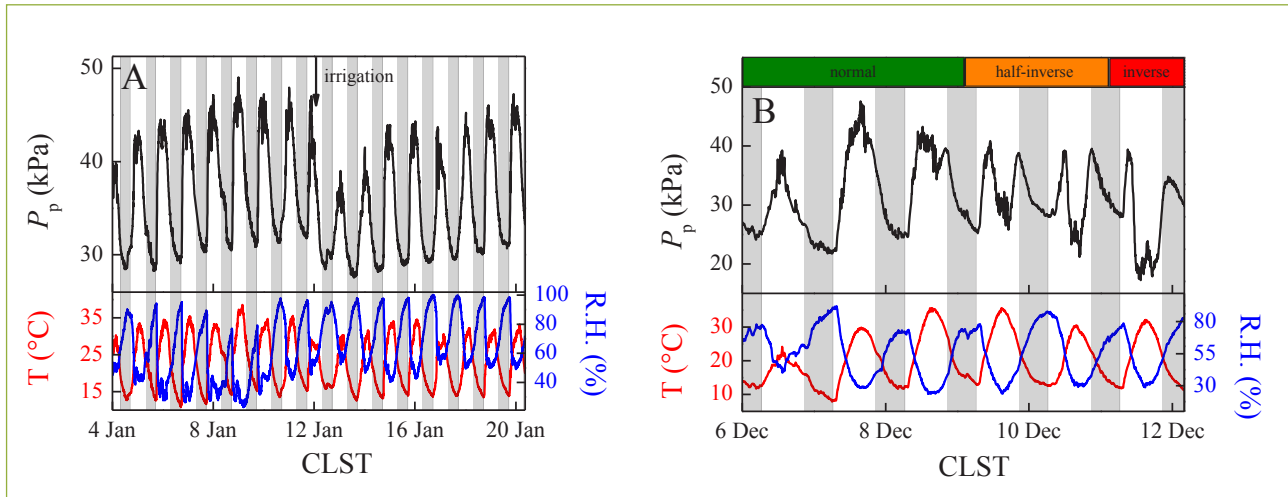


Figure 7. Part of a 3-month P_p recording (upper panel) performed in a grapevine orchard in Chile (A) and on an insufficiently watered almond tree in Chile (B) together with the corresponding profiles of local air temperature (lower panel; red line) and relative humidity (lower panel; blue line). Nocturnal hours are marked by grey bars. Arrow indicates irrigation. Note the effect of irrigation and non-irrigation on the amplitude of the P_p peak around noon and on the minimum P_p value during the night. For further details, see text. (CLST=Chile Summer Time).

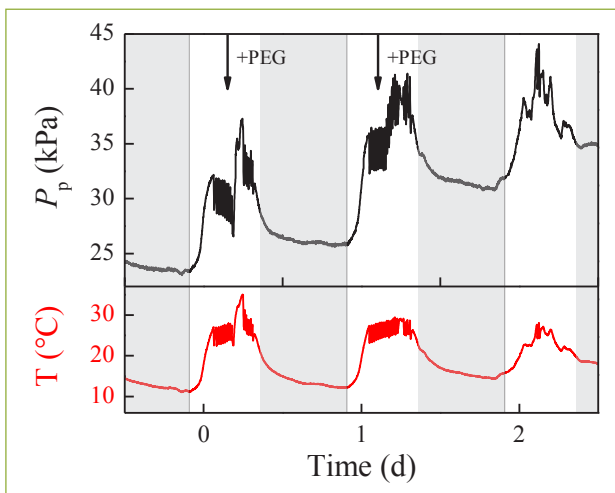


Figure 8. P_p changes of a sugarcane leaf upon addition of the osmoticum PEG (arrows: day 0: osmotic potential -0.25 MPa; day 1: osmotic potential -0.50 MPa) together with the corresponding ambient temperature (lower panel; red line). Note the increase of the amplitude of the P_p values at noon and during the night upon progressing osmotic stress. Nocturnal hours are marked by grey bars.

upper leaves (not shaded by surrounding leaves) on the south side developed drought stress symptoms which were comparable to those recorded on the north side, even though north-facing leaves experience longer and more intense sun exposure than south-facing leaves on the southern hemisphere. This is clear-cut evidence that trees compensate temporal water deficits by withdrawal of water from other sites of the trees. Furthermore, it was found that temporary drought stress was very often limited to

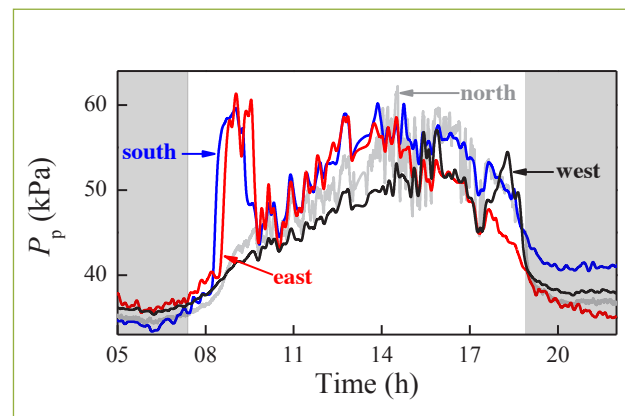


Figure 9. Multiple probe readings on lower leaves of a five meter tall well-watered avocado tree (30 January 2008, plantation close to Port Macquarie, Australia) performed on a sunny day. P_p values were normalised to the P_p range of the respective probes located on the north side. Nocturnal hours are marked by grey bars. Four probes were clamped in the cardinal directions of the compass (south: blue line; east: red line; north: grey line; west: black line). For details, see text and Ruger et al. (2010b).

individual branches. Concomitant dendrometer and probe measurements on small oak trees have demonstrated that cavitation and embolism in xylem vessels can easily be detected by determining the time lag between P_p peaking of the leaves and minimum stem diameter. The time lag between these two parameters gradually increases with progressing drought and is thus a very good indicator for (reversible or irreversible) blockage of the water pathways by water vapour and air.

CONCLUSIONS

The data reported here demonstrate that the ZIM-probe technology obviously exceeds the pressure chamber technique in all relevant performance and accuracy criteria, particularly under field conditions. The probes are non-invasive and therefore very robust in comparison to all other minimal-invasive technologies used or suggested for irrigation. These techniques require the insertion of microcapillaries and/or electrodes (Green et al. 2003, Fernández et al. 2001, 2006, Nadler et al. 2003, 2006, Zimmermann et al. 2004, Gurovich and Hermosilla 2009). Even though minimal-invasive techniques have a high impact for basic research, they are not suitable for outdoor measurements. Additionally, clogging and material deposits on the inserted microcapillaries and/or electrodes that occur with time leads to malfunctions.

The probes provide direct access to the turgor pressure, i.e. the key parameter for growth and fructification in real time and with high precision. The possibility to measure simultaneously leaf temperature broadens the range for precise irrigation control. Application of the correct amount of water at the right time saves not only water, but increases crop quality and yield potential, reduces leaching of fertilisers, reduces crop stress and susceptibility to disease and pests and increases the long term sustainability and profitability of the farmland. According to the

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broad basis of data, it is needless to say, that the probe technology is also superior to soil moisture methods frequently used in agriculture for water management because it allows tailored irrigation according to the very variable needs of plant species (independent of soil properties).

We also anticipate that the magnetic probe will find numerous applications in plant physiology and ecology. Advances in plant physiology and also in ecology have always been promoted by new technologies. A good example is the introduction of the various variants of magnetic resonance imaging a decade ago (literature quoted in Zimmermann et al. 2004). The recording of turgor pressure effects induced by stomata aperture oscillations and induced by blue/red light illumination shows the high sensitivity of the probe technology and will certainly stimulate a broadly based research in future that will lead to a better understanding of hydraulic plant functioning.

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