Protocol

Sap flow

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Summary
Sap flow measurements provide a powerful tool for quantifying plant water use and monitoring qualitative physiological responses of plants to environmental conditions. As such, sap flow methods are widely employed to investigate the agronomic, ecological, and hydrological outcomes of plant growth. The following summary provides an overview of thermodynamic sap flow measurement techniques used to quantify volumetric water use in plants. A variety of sap flow techniques have been described in the literature, but here we will focus on the most commonly used methods suitable for measuring sap flow in stems, branches and roots of woody and herbaceous plants.

Definition
Thermodynamically-based sap flow methods utilize heat as a tracer to quantify the rate at which sap (water) passes through the xylem tissue of a plant.

Overview of Measurement Approaches
We briefly discuss the following five sap flow measurement techniques (citations included for original development and methodological improvements):

1. Heat Pulse Velocity
   - Swanson 1994; Green & Clothier 1988; Burgess et al. 2001; Green et al. 2003

2. Trunk Segment Heat Balance

3. Stem Heat Balance
   - Sakuratani 1981, 1984; Sakuratani et al. 1999; Baker & Van Bavel 1987

4. Thermal Dissipation Probes
   - Granier 1985, 1987; Clearwater et al. 1999

5. Heat Field Deformation
   - Nadezhdina & Čermák 1998; Nadezhdina et al. 1998

Background
Heat Pulse Velocity (HPV)- this method measures sap flow by determining the velocity of a short pulse of heat carried by convection in the transpirational stream. HPV methods, which were developed by Huber and colleagues in the 1930's, were the first sap flow techniques to utilize heat as a tracer of sap movement. A theoretical framework for HPV methods was further developed in the 1950's by Marshall (1958), and numerical corrections to
account for departures from idealized heat transport theory were developed several decades later by Swanson and Whitfield (1981) and Green et al. (2003). Briefly, HPV methods use needle-like, line heater and temperature sensor probes inserted into conducting sapwood. Temperature probes often contain one or two temperature sensors (thermocouples or thermistors) for point measurements at different depths into the sapwood. A short pulse of heat (1-6 sec) is released into the sap stream and sapwood temperature is monitored at points upstream and downstream from the heater.

Several HPV techniques have been developed to accommodate a variety of flow conditions. These techniques include: the Compensation Heat Pulse Method (CHPM), the $T_{\text{max}}$ method, and the Heat Ratio Method (HRM). Briefly, the CHPM (Swanson 1962) uses two temperature probes placed asymmetrically on either side of the heater. After a heat pulse is released, the temperature of the closer, upstream probe immediately rises due to the conduction of heat through the xylem tissue, while the downstream probe remains cool. The temperature of the downstream probe then rises due to the convection of heat via the moving sap stream. The time delay required for an equal temperature rise at both probes represents the time required for convection to move the peak of the heat pulse to the mid-point between the two probes. Heat pulse velocity is then calculated based on a velocity = distance/time function. The HRM (Burgess et al 1998, 2001) uses two temperature probes placed symmetrically on either side of the heater, and is based on a derivative of Marshall’s (1958) heat transport equation that relates heat pulse velocity to the ratio of temperature increases of the upstream and downstream probes following the release of the heat pulse. The Tmax method (Cohen et al 1981) uses a single temperature probe downstream from the heater and relies on measuring the time taken for this probe to reach a maximum rise in temperature following the release of a heat pulse, from which heat pulse velocity is calculated using an alternative derivative of Marshall’s (1958) heat transport equation. All HPV methods require theoretically derived correction factors to be applied to correct heat pulse measurements for probe-induced wound effects, and to convert heat pulse velocity to sap velocity taking into account the physical properties of sapwood, without which heat pulse velocity remains a significant underestimate of sap velocity. Volumetric rates of sap flow are then calculated as the product of sap velocity and conducting sapwood area. HPV methods demand accurate probe spacing and thermal homogeneity in the sapwood, and it is highly recommended that calibrations be performed to validate quantitative measurements.

The CHPM is the most widely used and works well under high flow conditions, is remarkably robust, and suitable for field measurements in a range of species, but researchers have found that $T_{\text{max}}$ works better under some conditions (Green et al 2003). However, both the CHPM and Tmax methods have inherent difficulty resolving flows slower than 2-10 cm/h, and are incapable of resolving zero or negative (reverse) flow rates. The HRM was developed specifically to resolve zero and reverse rates of flow with great success, but the technique has unresolved limitations under high flow conditions (Bleby et al. 2008), which is the subject of current research. In the meantime, recent efforts have been made to combine techniques (i.e. CHPM and HRM) into a dual sensor that would work well under, high, low and reverse flow conditions through the addition of one more temperature sensing probe (Bleby et al 2004; McElrone et al. 2008).

Until recently, HPV was used only on woody stems because of the need to drill holes in insert needles in the woody tissue. van der Velde et al (2006) successfully modified heat pulse sensors for use in squash, a non-woody herbaceous plant, and Clearwater et al. (2009) recently adapted the technique for external use on smaller plant organs, including those of herbaceous plants. One advantage to using the HPV method is the low-power requirement (relative to the constant heating required for other techniques-see below), which can be beneficial under remote field conditions. The main disadvantage is the damage caused by drilling holes for sensor installation; this problem applies to all probe based sap flow measurement techniques and may be particularly important to consider for high-value perennial crops.

**Thermal Dissipation Probes (TDP)**- this method was developed by Granier (1985, 1987), and consists of two
needle probes (diameter ~2mm) inserted radially into a trunk and placed one directly above the other at ~10.0cm spacing along the trunk axis. The upper needle functions in both heating and temperature sensing (contains a thermocouple), while the lower needle functions as the reference temperature sensing probe. Constant power is applied to the heater in the upper needle, and the difference in temperature (dT) between the two probes is dependent on the rate of sap flow. As sap flow rates increase, dT between the probes decreases as heat is more rapidly dissipated away from the heater.

Granier et al. (1990) suggested that this empirical method could be used without the calibration needed for other sap flow methods because sap flux density calculations are independent on the characteristics of the wood anatomy. Clearwater et al. (1999) completed calibrations with additional species and confirmed that Granier's original calibration held when the entire heated probe was in contact with conducting sapwood xylem. However, the method underestimated sap velocity when part of the probe was in contact with non-conducting xylem or bark; the underestimation was proportional to the percent of the sensor in contact with non-conductive xylem. Clearwater et al. (1999) developed a correction factor that could be used to correct for sensor placement errors when the proportion of contact with non-conducting tissue was known, and recommended the use of probes <20mm and with multiple thermocouple depths to avoid these errors. Recent work has also shown that the original calibration of Granier is not universally applicable to all species and xylem types and that previous estimates of absolute rates of water use for ring-porous species obtained using the original calibration coefficients may be associated with substantial error, and independent calibration data should be collected where possible (Bush et al. 2010).

Trunk Segment Heat Balance (TSHB)- a summary of the historical development of this technique has been presented thoroughly by Čermák et al. (2004) and Smith and Allen (1996), and we refer readers to these articles for additional details. The original TSHB method was designed for use in large trees (diameter >12cm), where a section of the trunk is heated from the inside by an electric current passing through xylem tissue. The current is supplied by several electrodes (3-5 stainless steel plates ~25mm wide and ~1mm thick) pounded into the trunk sapwood with the center electrode oriented radially. In earlier designs, four temperature sensing probes (thermocouple needles ~1mm diameter) were inserted radially into the sapwood and even with the top of five electrode plates. The five electrode plates were even spaced from one another, creating four measurement sections of sapwood. The middle two thermocouple probes were positioned in the middle of the two sapwood sections immediately adjacent to the central electrode. The two additional thermocouple probes at this height were positioned 6cm outside of the outer two electrodes to account for any external temperature gradients outside of the heat field. An additional four thermocouple probes were also positioned outside the heat field at 10 cm directly below the upper electrode array thermocouples, and served as the reference needles to determine the temperature difference (dT) between the lower and upper thermocouple probes.

A commercially available modern version of the sensor utilizes 3 upper electrode plates that receive a constant heating power of (0.6W) supplied by an alternating current. An additional reference plate is positioned 10cm directly below the central upper electrode, and the dT between the heated upper plates and the lower reference plate is measured by needle probes containing several thermocouple points for radial profiling. In this setup the temperature sensing needles are embedded directly in the center of each plate. The TSHB method is then used to calculate the heat balance of a defined heated space where the energy input is split between the warming of the convective water moving through the trunk and the conductive heat losses (see equation details below). A miniature TSHB sensor has been developed for use in smaller diameter stems (0.6-2cm), and utilize an external heating (supplied by a resistive wire) and temperature sensing (see product details at EMS website listed in Table 1 below).

TSHB sensors are usually installed at least 0.5m above the ground to reduce the effects of steep temperature gradients that can occur below this point. As with other sap flow techniques, these temperature gradients can be
compensated for by installing additional temperature sensing needles outside of the heat field.

**Stem Heat Balance (SHB)** - similar to the TSHB technique, SHB quantifies sap flow through stems (and other smaller diameter organs as small as 4mm diameter) by balancing heat fluxes of into and out of a heated section of stem. Heat is applied to the entire circumference of the stem typically with a flexible heater (width varies according to the segment diameter) that is wrapped around the stem and covered with layers of cork, foam insulation, and reflective shielding. The foam insulation and shielding are extended well above and below the sensor hub to help minimize the effects of external temperature gradients, which seem to be unavoidable for some applications (see Shackel et al. 1990). A thermopile, formed with pairs of thermocouples connected in series across the cork layer, is used to measure radiant heat loss away from the heater. Two additional thermocouples are placed in parallel along the axis and against the surface of the stem at equal distance above and below the heater. The SHB method benefits from non-invasive installation of probes into the plant segment, but this setup also presents problems for determining the radial flow profile into the segment. Sap flow is determined with SHB sensors by balancing heat inputs and outputs using the equations listed below.

**Heat Field Deformation (HFD)** - a summary of the historical development of this technique has been presented thoroughly by Čermák et al. (2004), and we refer readers to this article for additional details. HFD is based on the measurement of the deformation of a heat field extending out from a constantly powered, needle-based linear heater inserted radially into the trunk of large trees. Under zero flow conditions, the heat field looks like a symmetrical ellipse with the longer axis of the ellipse oriented axially along the stem due to greater heat conduction of stem tissue in the axial relative to tangential directions. When sap is flowing, the heat field becomes a deformed ellipse elongates with increasing flows. Sap flow is calculated with this method using the ratio of temperature gradients measured by two pairs of thermocouple probes placed around the linear heater in the axial and tangential directions. The axial thermocouples are positioned above and below the heater and allow for bidirectional flow measurements in the acropetal and basipetal directions and for very low flows (similar to HRM described above). In the commercially available sensors, two more thermocouples are positioned equidistant (5-10mm) on either side of the heater (to the right and left). Multiple thermocouple depths are used in each temperature needle to determine the radial flow profile into the stem.

**Equations**

**HPV Methods:**

**CHPM:**

\[ v_h = \left[ \frac{X_d + X_u}{2t_z} \right], \]

where the temperature rise following the pulse of heat is measured at distances \( X_d \) (m) downstream and \( X_u \) (m) upstream from the heater and \( t_z \) (s) is the time delay for the temperatures at points \( X_d \) and \( X_u \) to become equal (i.e. how long does it take the heat pulse to reach the midway point between the two temperature sensors)

**\( T_{\text{max}} \) method:**

\[ v_h = \left[ \frac{\sqrt{X_d^2 - 4kt_m}}{t_m} \right], \]
where the time required \( (t_m, s) \) for a maximum temperature rise to be recorded by a single sensor located a distance \( X_d(m) \) downstream from a line heater, and the thermal diffusivity \( (\kappa) \) of the xylem matrix is determined under zero flow conditions using the following equation: \( \kappa = \frac{X_d^2}{4t_m} \)

**HRM:** \( \nu_h = \frac{[\kappa \ln(\nu_1/\nu_2)]}{X} \)

where a ratio of the increase in temperature \( (\nu_1/\nu_2) \), following the release of a heat pulse, is measured at distances \( (X, m) \) equidistant downstream and upstream from a line heater.

After obtaining \( \nu_h \) values, all heat pulse techniques require correction for wounding effects and probe misalignments before conversion to volumetric water use determined from the radial profile of flows and active sapwood area. See details for these corrections in each of the reference papers for these techniques.

**TDP Method:** \( m_v = 0.000119Z^{1.231} \)

where \( m_v \) is the volumetric sap flux density \( m^3m^{-2}s^{-1} \) and \( Z \) is determined from \( (dT_0 - dT)/dT \). \( dT_0 \) is the maximum temperature difference measured under conditions of zero flow (at night after extended rainy period). The mass flow rate of sap is then calculated using the following equation:

\[
F_m = \frac{r_m A_s}{s_v}
\]

where \( r_s \) is the density of sap, and \( A_s \) is the cross sectional area of sapwood (see details on the determination of active cross sectional area below).

**TSHB Method:** \( P = Q^*dT^*c_w + dT\lambda \)

where \( P \) is the heat input (W), \( Q \) is the sap flow rate \( (kg \cdot s^{-1}) \), \( dT \) is the temperature difference between the upper heated needles and lower reference needle(s) \( (K) \), \( c_w \) is the specific heat capacity of water \( (J \cdot kg^{-1} \cdot K^{-1}) \), and \( \lambda \) is the coefficient of heat losses away from the measuring point. \( \lambda \) can be partially eliminated by using foam insulation and reflective shielding around the sensor installation (these practices can be useful for all sap flow techniques described here to reduce the impact of environmental temperature gradients external to the sensor).

**SHB Method:** \( P = q_v + q_r + q_l \)
where $P$ is the heat input supplied by the heater, $q_v$ is the rate of vertical heat loss by conduction in the stem (measured with axial thermocouples placed above and below the heater), $q_r$ is the radial heat loss by conduction measured with the thermopile, and $q_f$ is heat transferred convectively by the transpiration stream - the remaining balance of the heat inputs and outputs. Additional details for the calculations of $q_v$ and $q_r$ can be found in literature by Sakuratani, Baker, and van Bavel. The mass flow rate of sap is then calculated using:

$$F_m = \frac{2q_f}{c_s(dT_a + dT_b)},$$

where $c_s$ is the specific heat capacity of water, $dT_a$ and $dT_b$ are the temperature changes, respectively.

### HFD Method:

$$Q_w = f[k_{geom}, k_{st}, c_w(dT_{sym}/dT_{as})],$$

where the temperature gradient ratio ($Q_w$) is proportional to the rate of sap flow, and constants are the geometry of the measuring point ($k_{geom}$), the physical properties of the conducting system including stem heat conductivity ($k_{st}$) and the specific heat capacity of water ($c_w$), and the temperature gradients measured away from the line heater in the axial/symmetrical (above and below the heater) direction ($dT_{sym}$) and the tangential/asymmetrical (to the sides of the heater) direction ($dT_{as}$).

### Equipment requirements:

Numerous researchers construct sap flow sensors themselves, while others have sold patent/design rights to companies that now supply sap flow sensors. All five techniques (described above) are now commercially available in one form or another, and have been adapted for a variety of applications (see product information and images for various suppliers listed in Table 1). Many of these commercially available products are also packaged with plug and play datalogging devices making the technology accessible and approachable to a broader audience. Alternatively, sensors can be interfaced with user-built measurement systems that utilize other commercially available dataloggers or computers. A major advantage of user-built systems is that other components and other sensors (e.g. soil moisture sensors, weather sensors etc) can be mixed and matched to meet specific needs, while a major disadvantage is the significant amount of time and the technical skill and experience required to construct and maintain such systems. If you are new to the use of sap flow methodology, we recommend utilizing one of the commercially available products. We have not provided details on sensor construction on this site. However, if you are looking to refine or adapt existing methods or develop new techniques for your specific application, we suggest contacting researchers most commonly linked to each technique for construction details.
### Table 1: List of commercially available sap flow sensors for each of the major types of sensors and contact information for the companies

<table>
<thead>
<tr>
<th>Method</th>
<th>Manufacturer</th>
<th>Company Website</th>
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<tbody>
<tr>
<td><strong>Heat Pulse Velocity</strong></td>
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<td></td>
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<tr>
<td></td>
<td>Armidale NSW</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Australia</td>
<td></td>
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<tr>
<td><strong>CompensationHPV (CHPV)</strong></td>
<td>Advanced Measurement &amp; Controls, Inc.</td>
<td><a href="http://www.advmnc.com">www.advmnc.com</a></td>
</tr>
<tr>
<td></td>
<td>WA, USA</td>
<td></td>
</tr>
<tr>
<td><strong>CHPV, Tmax &amp; Tz</strong></td>
<td>Tranzflo NZ Ltd</td>
<td><a href="http://www.tranzflo.co.nz">www.tranzflo.co.nz</a></td>
</tr>
<tr>
<td></td>
<td>Palmerston North</td>
<td></td>
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<tr>
<td></td>
<td>New Zealand</td>
<td></td>
</tr>
<tr>
<td><strong>Custom Made</strong></td>
<td>East 30 Sensors, Inc.</td>
<td><a href="http://www.east30sensors.com">www.east30sensors.com</a></td>
</tr>
<tr>
<td></td>
<td>Pullman, WAUSA</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Brno, Czech Republic</td>
<td></td>
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<tr>
<td><strong>Stem Heat Balance</strong></td>
<td>Dynamax, Inc.</td>
<td><a href="http://www.dynamax.com">www.dynamax.com</a></td>
</tr>
<tr>
<td></td>
<td>Houston, TX, USA</td>
<td></td>
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<tr>
<td><strong>Thermal Dissipation Probes</strong></td>
<td>Phytech, Ltd.</td>
<td><a href="http://www.phytech.com">www.phytech.com</a></td>
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<td></td>
<td>Kibbutz Yad Mordechai, Israel</td>
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<td>Dynamax, Inc.</td>
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<td>Houston, TX, USA</td>
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<td></td>
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<td></td>
<td>Kibbutz Yad Mordechai, Israel</td>
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Terminology

Edwards et al. (1997) proposed a unified nomenclature for sap flow measurements in an effort to overcome fundamental misunderstandings about the physics of heat and sap movement in wood. We refer readers to this summary that primarily covers heat pulse techniques but can be applied more broadly to other sap flow methods. More recently Lemeur et al. (2009) revisited sap flow nomenclature after confusion arose over the use of symbols, units, and physical quantities during the 7th International Workshop on Sap Flow; these authors suggested discontinuation of some commonly used terms. Below we present a list of terms that have been and are still commonly used in the sap flow literature.

- **Conduction**- the passage of energy, particularly heat or electricity, through something

- **Convection**- the transfer of heat energy associated with the movement of fluid

- **Corrected heat pulse velocity** \( (v_c) \)-raw heat pulse velocity that has been corrected for wounding effects associated with sensor probes disrupting flow in xylem

- **Cumulative sap flux** \( (Q_c) \)- cumulative sap flux integrated over a period of time

- **Flux**- the amount of some quantity flowing across a given area, which is often perpendicular to the flow, per unit time

- **Heat pulse velocity** \( (v_h) \)- the rate at which a heat pulse is convected downstream in the xylem sap from a line heater moves; also referred to as raw \( v_h \) \( (\text{m s}^{-1} \text{or cm hr}^{-1}) \)

- **Mass flow**- fluid is transported by mass flow alone
**Sap flow velocity** \( (v_p) \)-refers to the speed of sap measured at a point or along a needle probe \( (m \text{ s}^{-1}) \); Lemeur et al. (2009) suggested this term be discontinued, however it is often useful when qualitative assessments of sap flow are the primary focus of a given study.

**Sap flow rate**-refers to the volume mass of sap flowing through a stem per unit time; this value is integrated to account for radial flow profiles across the active cross sectional sapwood area \( (m^3 \text{ s}^{-1}) \); Lemeur et al. (2009) suggested this term be discontinued.

**Sap flux** \( (Q) \)- integration of sap flux density over a given surface

**Sap flux density** \( (F_d \text{ or } J \text{ or } J_p) \)- volume of sap flowing across an area of sapwood perpendicular to the direction of flow per unit time \( (m^3 \text{ s}^{-1} \text{ m}^{-2}) \)

**Wounding corrections coefficients**- multipliers that are applied to raw heat pulse velocities \( (v_h) \) to correct for the effects of wounding imposed by the placement of probes within the sapwood matrix; these values are applied to the raw \( v_h \) before sap flow velocity is converted to volumetric sap flow.

**Range of values**

Reported values vary significantly between studies because of the variety of units that have been to express sap flow (as described by Edwards et al 1997 and Lemeur et al. 2009).

**Scaling from Single Sensors to the Whole Plant Volumetric Water Use**

In order to scale measurements from individual sensors to whole plant estimates of water use, one must accurately determine the active cross-sectional area of sapwood and how flow changes across this tissue. Sap flow sensors with multiple radial depths, xylem mobile dyes, and tree core inspections are often used to determine the active cross sectional area of sapwood. Gross wood cross-sectional area is measured using the radius of the organ (measured under the bark), and discounting the non-conductive heartwood based on clear discoloration (visual inspection of cores extracted with an increment borer at the same height as sensor installation) or by staining the sapwood. To stain the active sapwood under transpiring conditions, dye is injected into deep holes made with an increment borer, and the xylem tissue 2-4 cm above the injection point is sampled with a borer 1-2 hrs after the injection (Goldstein et al. 1998). The depth of the active sapwood is then determined from these subsequent cores. Other researchers have attempted to eliminate the effects that emboli have on dye infiltration by completing chisel incisions into the wood under the dye solution; the plant segment is submerged in the solution using a flexible plastic collar sealed to the trunk.

For large trees with deep sapwood, several point measurements of sap flow from different depths across the conducting area may be required to describe the radial pattern of flow. Radial flow measurements then must be integrated to estimate whole-plant water use. Common approaches for integrating radial measurements include the fitting of polynomial mathematical functions to point measurements and integrating this equation across the sapwood profile, or the use of...
‘weighted average’ type integrations where sapwood is divided into concentric bands and volumetric flow across each band is determined from corresponding point measurements of sap flux density, and whole-plant water use is subsequently calculated as the sum of flows from all bands (Hatton et al 1990).

Woody plant xylem is not uniform, and many sub-surface features, not obvious based on visual inspection from the outside of the plant (i.e. knots and dead wood), can disrupt flow and create non-conductive regions of the tissue. In practice, it is not possible to avoid all of these hidden features, but careful sensor installation and appropriate sampling (i.e. multiple radial depths to account for profiles and enough sensors to account for circumferential variability) can help to overcome these limitations and generate reliable data.

Health, safety, and hazardous waste disposal considerations

Care must be taken in handling electrical power inputs needed to supply heat for these sensors. When making the sensors yourself, care should be taken to solder junctions under well ventilated conditions while using eye protection.

Literature references


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