ANALYSIS OF CANOPY TRANSPIRATION BASED ON THE SAP FLOW MEASUREMENT

Jiří Kučera

Environmental Measuring System Ltd.

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1. Foreword

There are many papers and textbooks describing the water regime of plants from the point of view of physiology and also a lot of literature on the methodology of measurement of related variables. However, there are only a few papers that link these two sides of the coin on the same level. Please consider this work as an attempt in this field.

2. Aim of the work

The main objective of my work is to describe the different strategies of different species under different meteorological conditions and their resistance to limited water availability in the soil. Both phenomena will be accessed by analyzing stomatal responses to environmental conditions.

Since stomata are the only means by which plants can influence their function at a certain growth stage, stomatal behavior is a key factor for production on the one hand and the ability to survive water stress on the other.

Finally, knowledge of plant response to climatic conditions can provide a theoretical basis for theoretically informed forest management. The current approach is likely to have to change under current global warming, as the optimal areas for certain species will change. In such a situation, stomatal characteristics of trees – response to solar radiation and vapor pressure deficit - should help to optimize the selection of suitable species.

3. Introduction

3.1. Canopy transpiration

Estimation of canopy transpiration is one of the key inputs used in hydrology, plant physiology and climatology (Sinclair & Ghanem 2020). Transpiration in the context of potential evapotranspiration and soil water availability leads to the analysis of canopy conductance and subsequently plant response to evaporative demands and drought stress (Allen et al. 1998). From this point of view, it should help forestry in selecting tree species according to their physiological parameters. Other areas of interest are agriculture, horticulture and also plantations with a short rotation period for energy purposes.

Principally, there are two methods of direct estimation of transpiration:

- Methods based on the theory of turbulent fluxes – eddy covariance, aerodynamic method, Bowen ratio energy balance, surface renewal, scintillometers. The basic problem of these methods is the measurement of the sum of transpiration and evaporation without the possibility of their differentiation.
- Measurement of sap flow of sample trees with scaling up to the whole canopy and subsequent conversion to transpiration in [mm]. On the contrary to above mentioned methods the evaporation does not affect this measurement.

The main difference between the two methods is that the turbulent flux measurement gives evapotranspiration values, i.e. the sum of transpiration and soil evaporation. In the case of significant understorey, it is not possible to distinguish between the transpiration of the two layers. Under such conditions, the measurement of sap flux is the only way to help at this point.

The most common method of measuring turbulent flows - eddy covariance (EC) - is based on pure physics but has its limitations. It requires a relatively large area of homogeneous canopy and fails at still air. Also, because of the need to install a measurement system above the canopy, tall masts must be erected in full-grown forests. EC systems are already widely used but remain relatively expensive.

3.2. Measurement of sap flow

The measurement of water consumption of entire plants is usually based on measuring the volume of water consumed by the plant over time by measuring the volume (potometers) or weight of transpiring water. This is relatively easy by small plants but impossible on large trees.

Since the transpiration means the evaporation from leaves, there is a possibility of direct measurement on the leaf level in situ or by weighting of cut twigs and following upscaling on the whole crown (Schulze et al. 1985). Leaf transpiration can be measured by porometers (Scholander et al. 1965) or by means of small leaf chambers (LiCor, Inc., USA, company literature). However, none of those methods can't be used for long time measurements.

Measuring of sap flow is a way of estimation of water uptake in large trees where other methods such as weighing or potometers are excluded. Measuring the amount of water passing through the trunk towards the branches and leaves is the only realistic approach for measuring the long-term water consumption of mature trees.

Sap Flow measurement has a more than 90-year history of development, during which many measurement methods have been established. Several theoretical papers have been published that attempt to shed light on this complex problem, but it is questionable to what extent the input assumptions correspond to the actual situation in different tree species.

Principally, there are two groups of measuring methods based on

- measurement of the sap velocity by means of markers, moving together with streaming water. It can be dye, heat, radioactive elements. The basics method which uses the injection of heat methods were given by Huber (1932) and several other methods have since been developed on the same principle
- volumetric water flow based on heat balance of the compartment inside the xylem through which the water passes. It assumes an energy input that is split between the conduction losses and the heat dissipated with the moving water.
- methods that are outside the two groups mentioned above and that compare heat transfer in different directions around an artificial heating element - in the direction of water flow and perpendicular to it.

According to that, there are some methods which are commonly used to measure the sap flow of mature trees:

- Heat Pulse Velocity (HPV; Huber 1932, Swanson 1981)

- Tissue Heat Balance (THB; Čermák et al. 1973; Kučera a kol. 1977)
- Stem Heat Balance (SHB; Sakuratani 1981; Lindroth et al. 1995, Cienciala 1995, Urban 2012)
- Heat Dissipation (HD; Granier 1985)
- Heat Ratio Method (HRM; Swanson and Whitfield, 1981, Burgess et al. 2001).
- Heat Field Deformation (HFD; Nadezhdina a kol. 1998)

Heat Pulse Velocity

The HPV method is based on the principle of periodic pulse heating and measurement of the rate of temperature propagation up and down from the heater, unlike some other methods (HD; Granier 1985, THB; Čermák et al. 1973; Kučera et al. 1977, SHB; Sakuratani 1981, Lindroth et al. 1995, Cienciala 1995, Urban 2012) that require continuous heat input to the device and measurement of heat loss around the heater. Thus, transpiration is not assessed based on sap flux per se, but rather on heat transfer and movement in the xylem (Forster 2017). The lower limit of stem diameters reported in published stem studies is ≈ 5-6 mm (Cohen et al. 1988, Cohen et al. 1990).

Heat balance techniques - there are currently at least three sensors based on that technique:

Stem Heat Balance (Dynagage Sap Flow sensor, Dynamax Inc., USA).

It uses the principle of the measurement of heat loses in three directions: Axial up and down along the stem and the lateral loss through the outer insulation of the cylindrical heater. The method was originally proposed by Sakuratani (1981). His approach supposes the rectangular shape of the heated filed along the stem which is, however, far from reality. The actual design of the sensor also causes stem strangulation during longer installation on plants with high stem growth.

Tissue Heat Balance - Environmental Measuring Systems, Ltd., CZ (EMS Brno)

Intended for large stems, this sensor uses for internal heating of stem tissues (therefore named as Tissue Heat Balance) the original idea of direct heating (Čermák et al. 1973; Kučera a kol. 1977). On the opposite to all other methods is the xylem heated by the alternating electrical current passing between the set of three flat stainless electrodes. The main advantage of this ingenious idea in that there is excluded the heat transfer between a heating element and the water-active xylem which makes hardly defined error under higher sap flow rate. Also, if the length of electrodes covers at least approximately the expected depth of sapwood, the measured values are nearly independent of the radial profile of hydro active xylem. The sensors based on this principle gives sap flow values in kilograms per 1 cm of stem circumference at cambium level. The value for the whole tree is obtained by multiplying by the stem circumference. This method is suitable for tree trunks with diameter bigger that ca 12 cm at cambium level.

An important feature of this sensor is the way of operation. The heating power is controlled a way which keeps the temperature difference between the ambient and the water leaving the heated space on the constant level. The measured value of heating power is that directly proportional to the volumetric water flow. Since the xylem temperature does not change with the sap flow rate, the heat capacity of the xylem does not affect the dynamic properties of the measurement (Kucera 1977).

Stem heat balance (EMS Brno)

Similarly as SHB sensors (Dynamax Inc., USA) this sensors also uses external heating with a cylindrical heater but it measure the temperatures inside the stem. The heat balance is not calculated at each time of the measurement but it uses for estimation of heat losses from the heated space the value measured during under zero sap flow – typically at predawn. The senor also operates with constant temperature difference.

Both EMS sensors are based on exact physical principle (Kučera 1977). They do not use any empirical coefficients or species-related parameters. Except for sensors for larger stems, where a basic knowledge of sapwood depth is required to select one of the three available electrode lengths. The output values are always in kilograms per hour.

Heat Dissipation – Granier sensors.

In principle, it is also a heat balance method. The output variable is sap flow density in terms of [kg.m^2.h]. The calculation is based on the measurement of the temperature difference between heated and unheated needle inserted is the hydro active xylem (Granier 1985). The method in not based on a clear physical background but it is widely used because of simple hardware. The calculation of the whole tree sap flow needs a good knowledge of radial velocity profile in the sapwood.

Note: All heat balance methods are based on the same principle but the main difference among them is the definition of the heated space under different sap flux density. The estimation of cross section area and the shape of the heated space affected by streaming water is the main issue related to those methods.

Heat Ratio Method (ICT International, Australia)

This sensor represents the latest sophisticated version of the heat pulse method described by Swanson and Whitfield (1981). This method estimates the sap flow velocity according to the difference in spreading of the short pulse of heating energy upwards and downwards the stem. The heat pulse is released in a needle inserted into the stem between two other needles with temperature sensors placed in a row along the stem. Sap flow values are calculated according to the course of heat response to the pulse. However, the calculation needs the knowledge of some tree specific parameters, like thermal diffusivity, wood density and moisture content (Steppe et al 2010).

Heat Field Deformation (ICT International, Australia)

This sensor in mainly intended for the analysis of radial sap velocity profile. Three needle sensors measure the temperature around the heating needle in both vertical and horizontal directions in more depths. Similarly to HRM sensor, the calculation of volumetric sap flow needs the knowledge of tree specific parameters (Nadezhdina 1998.).

Note: There is an article that compares all of the above sensors except EMS sensors. The authors used a cut tree trunk through which water was pumped (Steppe 2010). Apart from the unfathomable reason why they left out one worldwide commercially available sensor with a long history of development, I have doubts about the methodology of this experiment. In the early 1990s, a similar effort was made to use cut beech and spruce logs for THB sensor calibration. The only result of this challenging work was the discovery that the water-conductive part of the plant loses its natural pressure conditions after cutting and therefore loses its original properties (Čermák 1992).

Notes and comments:

There are some issues with needle sensors which should be mentioned:

- Xylem water potential. The main problem in measuring sap flow is the negative pressure in the hydroactive xylem - the more invasive parts of the measuring sensor are used, the more the conducting vessels or tracheas are affected. In the case of an extremely sophisticated measuring system that takes into account all possible shapes of radial conductivity profiles and all their specificities, the sensor accurately measures an object that is already significantly different from the original one, mainly due to disturbance of original water pathways with temperature sensors or heaters. Therefore, the actual sensor design is always a compromise between accuracy and the effect on the measured object. There are also sensors that do not use any invasive parts, but their sensitivity to the water flow passing in deeper layers of the conductive system is a matter of discussion (Dynagauge).

- Depth of sensing parts. The accuracy of the measurement of temperature difference with needle sensors is strongly dependent on the same depth of inserting of needles. The stem is not a cylinder, it has only a cylindrical shape. The heterogeneous wood density and bark roughness makes the real stem far from the ideal cylinder. Therefore, the propagation of changes of ambient temperature could be different at different places on the stem surface. The radial temperature gradient in sap wood areas in the morning and in the evenings, when the change of ambient temperature is the fastest, it reaches even degree per cm. Consequently, the needles with temperature sensors installed at the distance of few centimeters far from each other can give false dT even without an artificial heating and it could significantly affect the measurement at the low dT levels.

- Alinement. From technical reasons and due to the wood heterogeneity it is not easy to drill parallel holes. The smaller needle diameter, the more likely the drill bit tends to bend. Then, the measuring method which depends of the distance between needle sensors can suffer with errors due to needle alinement.

- Influence of high thermal conductive steel could bias the measurement of temperature profile with sensors placed close to each other in one steel needle.

- Heat transfer from the needle heating element to the surrounding xylem. Due to the small surface area of the needle, the amount of heat lead out with streaming water is under higher sap flow rates not directly proportional to its velocity.

There is a paper (Tatarinov et al. 2005) analyzing the theoretical dis- and advantages of different ways of heating with tin line, rectangular plate or in space. The result of the analysis shows the advantages of space heating - the larger the space heated, the smaller the errors due to uncertainty factors (thermal conductivity of the wood, inhomogeneity of the radial sap flow profile and external temperature gradients), which were strongly influenced by the size of the heated area.

3.3. Related measurement

Since the main reason for measuring sap flux is to calculate canopy transpiration and consequently canopy conductivity based on the parameterization of the P-M equation (Monteith 1965), some additional environmental variables in the soil-surface-atmosphere continuum are required.

3.3.1. Global solar radiation

These sensors should measure the total incoming shortwave radiation within the range 300 to 3000 nm. There are two main types of sensors:

- Thermopile pyranometer based on the thermal principle - covers the entire spectrum of solar radiation.

- Photovoltaic pyranometer - uses light-sensitive semiconductor components. The measuring range is usually 400 to 1100 nm, but the spectral sensitivity is not the same in different parts or spectra. These sensors are considerably cheaper than thermocouple sensors. They are calibrated for sky radiation under the assumption that the incoming radiation spectrum is more or less stable. They must not be used to measure radiation with a spectrum distorted by reflection or passing through partially transparent substances (leaves, water).

3.3.2. Net radiation

Two types of sensors are used to measure net radiation according to the output quantities - simple ones that indicate the difference between incoming and outgoing radiation in the short-wave and long-wave spectral range. Four-part sensors are used to study in detail the incident and reflected radiation in both spectral ranges separately. All these types of sensors work on the thermal principle. The tricky part of differential sensors is the protection of the sensing part, which must not change the whole measured spectrum. Some manufacturers use a polyethylene hemisphere (Schenk 8110), some do not cover the sensing part at all (OTT HydroMet, NR lite2).

3.3.3. Air temperature

Recently, the temperature was measured mostly with Resistance Temperature Detector sensors (RTD), which uses the thermal dependence of the resistance of the metal resistors on the temperature. Due to the excellent resistance against aging and corrosion the platinum wires are used for those sensors.

Currently, resistive sensors are being replaced by electronic sensors based on the dependence of the threshold voltage on the semiconductor transition. These sensors often have a digital output. Due to individual factory calibration, they have excellent accuracy, which is at least equal to that of platinum sensors. As they are still relatively young, their long-term stability will be tested in the coming years.

3.3.4. Relative air humidity

Air humidity sensors suitable for long-term continuous measurement solely use the change of dielectric constant (permittivity) of a capacitor exposed by ambient air. These sensors passed serious development in last decades with respect to long-time stability and accuracy. However, they are sensitive to some chemicals (ammonia, methanol, ethanol, aromatic hydrocarbon). Currently they are also available as integrated circuits, mostly together with temperature sensors. Lifetime of humidity sensor is quite high, however, recalibration or sensor replacement after two-three year is recommended.

3.3.5. Wind speed and direction

With the falling price of ultrasonic anemometers, they are slowly replacing mechanical transducers with vane and Robinson cross transducers. Due to the absence of mechanical parts, sonic anemometers require no maintenance. In addition to the wind direction in degrees, they often indicate the N-S and E-W components of the wind direction. This solves the problem of averaging readings in degrees when the vane oscillates close to north, when the average of the measured directions gives false readings.

One exception sensor suitable for sturdy environment should be mentioned with mechanical anemometers – the "aircraft" type of sensor manufactured by R.M. Young Company - Heavy Duty Wind Monitor-HD 05108.

3.3.6. Precipitation

Measuring of precipitation is one of the trickiest tasks. Besides of problems caused by wind, frost, etc., the main issue is regional pattern of precipitation and consequent impossibility to interpolate the values from neighboring measuring stations. Simply – missing data are missing forever.

There are many types of precipitation collectors – from simple barrels to sophisticated laser *sensors*. The most common sensors are based on mechanical principle with the periodically emptied bin filled with water collected from the funnel of known size.

Some models use a kind of spoon, others use a tipping bucket with two identical chambers. The output of these sensors are short pulses generated when the moving part moves.

3.3.7. Soil water content

The volumetric water content of soil [$m^3.m^{-3}$] is one of the most important variables related to plant water relations, but also for many hydrological, climatic and water management reasons (Hillel 2004). This measurement is quite complex business mainly due to wide range of soil structure and the spatial variability of the water content.

The only accurate results can be obtained by the gravimetric method based on the difference between the fresh and dry weights of the soil samples. However, this method is neither continuous nor convenient and obviously cannot be used for long-term monitoring. Sensors which are commonly used for the measurement are based on electromagnetic principle where the physical measured value is relative permittivity of soil which mainly depends on the water content (Topp et al. 1980).

Unfortunately, the relation between the soil water content and the relative permittivity is soil specific. Therefore, most of sensor with digital outputs gives the relative permittivity also as a separate variable. Thus, it opens the space for calibration in certain soil type.

There are two groups of methods for measuring soil water content based on the electromagnetic principle. One group of sensors measures the capacitance of a capacitor made up of sensor parts where the soil is the dielectric (Capacitance, Atkins et al. 1998; Frequency Domain Reflectometry, Kelleners 2004). The second group of sensors is based on the speed of propagation a high-frequency electromagnetic pulse along a two-wire antenna or coaxial line – Time Domain Reflectometry (Topp and Davis 1985; represented by Campbell CS650/655 sensors), Time Domain Transmissometry (Topp et al. 1980; Topp et al. 2001; Truebner SMT-100 sensors).

The most sophisticate method "Coaxial Impedance Dielectric Reflectometry" uses Stevens Water Monitoring Systems (Hydra Probe). On the contrary to other methods is the measurement based on the voltage difference generated and reflected high frequency signal. This sensor is less dependent on the soil salinity and temperature, soil variability and inter-sensor variability than other methods.

Some above-described measuring method are more or less dependent of the soil type or electric conductivity (salinity), nevertheless, the main problem is common for all sensors (Hillel 2004): Since the relative permittivity of water is 80 and relative permittivity of air is only 1, the air gaps between the sensor active part and the soil makes significant error. Therefore, proper sensor installation is the most important condition of getting good results. The variability of measured values in soil with normal electrical conductivity depends more on the correct installation of the sensor than on its type.

3.3.8. Matric soil water potential

Considering sap flow rate as a function of leaf-soil pressure difference and resistance of water conductive paths, soil water potential is from definition much more related to the transpiration that soil moisture. Soil water potential has always negative values (Aslyng 1963). The lower it is, the lower must be also leaf water potential in order to get water from soil to leaves and consequently evaporate in vapor phase (Fitter a Hay 2001).

The measuring methods are based on direct measurement of negative pressure or on indirect measurement via water content in a body with well-defined hydraulic characteristics (Gardner 1986; Topp and Ferré 2002).

- Direct pressure measurement (tensiometers) is applicable in the range above - 0.1 MPa, when the difference to atmospheric pressure is zero. The pressure sensor senses the vacuum above the water column by connecting the sensor to a ceramic porous pot.

- Measurements based on volumetric water content are made in porous material. When installed in soil, its water potential is equal to the water potential of the soil and therefore its water content of sensor depends on the matrix water potential of the surrounding soil. If the retention curve of this porous material is known, its water content is a function of the water potential (Hillel 2004). The water content can be measured by a method similar to the measurement of the water content of soil: electromagnetic properties, electrical conductivity or thermal conductivity of the sensor.

Sensors based of the measurement of *relative dielectric permittivity* looks partially similar as soil sensors which are instead of soil surrounded with a porous material (Hillel 2004). The minimum range of those sensors is mostly up to - 2 MPa.

Measurement of the *direct electrical conductivity* of a porous material requires excitation with AC voltage or bidirectional pulses to prevent polarization of the electrodes. Since the electrical conductivity depends on the chemical composition of the water in addition to the water content, a buffering agent, usually gypsum, is used inside the porous material (Hillel 2004). The lifetime of this type of sensors is mostly limited to two years of the reliable measurement due to slow dissolution of gypsum. The maximum range of this sensor group is -2 MPa.

- Sensors that use the *thermal conductivity* of the porous sensor body. An internal electric heater releases short energy pulses that cause the sensor temperature to rise. The slope of the rising temperature is proportional to the water content of the sensor and, due to the known retention curve, gives sensor water potential values that are equal to the water potential of the soil. The measurement is by definition discontinuous, the minimum range of soil water potential is approximately 0.2 MPa. A soil-specific calibration is required.

The main problem with all these sensors is the size of pores. Sensitivity at low potential requires relatively large pores and, conversely, the smallest pores determine sensitivity at low potential. The pore size distribution is the factor that influences the quality of soil water potential sensors.

3.3.9. Soil heat flux

The sensors are based on the measurement of the temperature difference between both sides of a disk with defined thermal conductivity. The disk is placed horizontally into the soil. Output of sensor is calibrated in [W.m^2] and it is used in the calculation of energy balance.

Measuring the heat flux of the soil is necessary to calculate the energy stored - released from the soil. In theory, it should be located at the soil surface, but for proper operation it should be placed in a homogeneous soil profile as close to the soil surface as possible (Jansen et al. 2011).

3.3.10. Stem diameter

High-resolution measurement of stem diameter is not only related to growth, but it also reflects the water potential of the xylem (Zweifel et al., 2005). Stem shrinkage caused by limited water availability in the soil is one of the best indicators of drought stress (Fernández and Cuevas, 2010). Together with sap flow measurements, it provides a synergistic view of the tree's water status and its response to environmental variables (Devine and Harrington, 2011). Automatic dendrometers are used to measure shrinkage of stem, which allow continuous measurement of water status and tree growth while being non-destructive and automatically recording in real time with a high level of accuracy (Améglio and Cruiziat, 1992; Zweifel et al., 2001; Zweifel et al., 2006). There are two types of commonly used sensors:

- *point sensors*. These sensors are screwed down the xylem where no movement due to growth or water status are not expected. The measuring part – mostly a linear potentiometer or LVDT sensor it pointed in radial direction towards the stem. The main disadvantage of point dendrometers is the fixation by screw into the stem (for the radial type) or the limited thickness of the trunk (Dobbertin et al. 2013).

- *band dendrometers*. These sensors measure changes in the circuit and therefore the output of the sensor is relative to the entire shank. The sensors are non-invasive in principle and the installed sensor does not interfere with growth around the sensor. The main disadvantage of the tape dendrometer is the handling of the sharp tape and its precise location on the tree trunk (Cattelino et al. 1986).

3.3.11. Leaf water potential

The current leaf water potential (LWP) is highly dependent on environmental conditions. Since the transpiration rate is small or almost zero at night, LWP equilibrates with the water potential of the soil at night. LWP values measured just before dawn usually have the highest LWP value during the day. The pressure chamber method is most often used to measure LWP (Scholander 1965).

3.3.12. Xylem water potential

The measurement of the xylem water potential is quite tricky. The sensor must be placed at the cambium level and well protected again drying out. The measuring method is based on psychrometric principle (Dixon 1984). The temperature sensors inside the sensor are extremely tiny and they need gentle handling.

3.4. Data using and processing

Sap flow data calculation is performed according to the Sap Flow System EMS methodology (EMS Brno) supported with Mini32 software.

As was already mentioned, the main reasons for the measurement of sap flow are:

- Detail physiological studies focused on tree behavior under different soil and climatic conditions, response to CO_2 elevation, fertilizing, bark beetle attacks etc.
- Hydrological issues, mostly water balance of catchments where the forest closes the total water balance - runoff, water drain, soil water content, precipitation, transpiration from open area.
- Distinguishing between the evapotranspiration estimated by methods of turbulent fluxes and tree transpiration. The difference covers both soil evaporation and understorey transpiration.
- Canopy conductance as a difference between potential (PET) and actual transpiration (E).

The easiest task is the first point – function of individual trees. All other three tasks need a scaling from sample trees to the canopy level. This issue is complicated in mixed forest and in sparse canopies.

The best tree parameter for proper scaling is the one most closely related to the tree's function.

The parameters in order of functionality:

- Ratio between the number of trees per ha and number of measured tree samples
- Ratio between the cross section area of all trees per ha and all sample trees
- Ratio between the sapwood area of all trees per ha and all sample trees
- Scaling curve – the dependence between the tree parameter and sap flow rate. Such a parameter can be DBH, cross section area, number of leaves, leaves effective area (Čermák et al. 2004) etc.
- Scaling according to solar equivalent leaf area (Čermák 1989)

In praxis – scaling curve based on DBH is preferred if such a dependence has reasonable coefficient of determination. Otherwise, a simpler parameter must be accepted - mostly number of trees. This problem will be described in detail later.

Canopy conductance:

The most sophisticated and commonly used is the method of parametrization of Penman-Monteith (P-M) equation of estimation aerodynamic and canopy conductance. It is based on the original Penman's equation of PET, describing the calculation of evapotranspiration from different surfaces (water, well-watered lawn, bare soil) according to main meteorological variables. The equation is composed of two parts, from which the second part includes the empirical part describing the dependence on the wind speed. Later, Monteith (Monteith 1965) significantly upgraded the Penman theory according to the theory of turbulent fluxes and introduced both conductances to calculation of transpiration.

The parametrization of P-M equation is based on the knowledge of transpiration and meteorological variables. The aerodynamic conductance is calculated according to the theory of turbulent fluxes from wind speed and wind parameters of the canopy. Therefore, the only missing variable is canopy conductance.

The canopy conductance can be described as a function of environmental factors - mainly global radiation and vapor pressure deficit (Lohamar 1980). This function varies among tree species and in knowledge is the key factor of understanding the response of trees to evaporation requirements and soil water status.

4. Material and methods

The following methodical approach regards to analysis of transpiration of mature forests. It is based on my own experience and on data sets measured under my supervision. The data processing uses mostly the software Mini32 which has been created and developed by Environmental Measuring Systems Ltd. This software is intended for the effective processing of long-term data series and is uses graphical view as a user interface.

The methodology describes the whole process the analysis of the canopy transpiration from the selection of sample trees until the explanation of the response of canopy conductance to environmental factors.

The canopy in here considered as a homogeneous relatively large mono-species forest which matches theoretical assumptions of big-leaf concept (Martin at al 1997).

4.1. Method of the sap flow measurement

Regarding the measurement method, we use only THB sensors with direct heating of the stem tissues. This method established in 1974 and it has been improved significantly over the last thirty years. During that time most of the issues have been discovered and successfully solved. In the current arrangement the method does not need any empirical, tree- or species-specific parameters. It is almost independent of the radial conductivity profile and directly outputs volumetric water flow values (Kučera 1977, Čermák 2004).

Sensors based on this method are commercially available and used worldwide (Sap flow system EMS81, EMS Brno, Ltd., CZ).

4.2. Tree samples selection

The number of tree samples obviously affects the reliability of the canopy transpiration estimation (Čermák et al. 2004). It is always a trade-off between accuracy and available budget. Three trees are the bare minimum, five to eight trees is a good number, and twelve sample trees is close to the optimum.

Proper selection of sample trees depends on the canopy structure – they should cover all tree diameter classes with respect to their social status (predominant, dominant, co-dominant). It is important for later scaling up to the canopy. On the other hand, the contribution of small trees to the canopy transpiration is rather small and there is mostly a tree class (around DBH 10 to 20 cm) which contribution is close to zero.

The choice of individual trees is always subjective and depends on experience. Since we tend to select only well looking trees, a random approach is an option. For example, all trees along a 1 m wide transect in a random direction were selected as sample trees (Cienciala et al. 1999). On the other hand, severely damaged and visually dying trees should be excluded from the selection.

Sample trees should have undamaged stem without unusual irregularities. Significant wounds on stem contributes to irregular sap flow around the circumference what affect the scaling from the measuring point to the whole tree sap flow.

4.3. Location of the sensor on the stem

The number of sensors installed on larger stems has long been a matter of debate. The historical sensor arrangement, which used five electrodes and needle temperature sensors inserted to the xylem between the electrodes, was quite sensitive to the shape of the radial temperature profile. Also the uncertainty of needle depth measured towards the bark surface relative to the real temperature isolines caused errors under strong radial temperature gradient. As a result, the accuracy of the measurements was unstable and the differences between sensors installed on opposite sides of the trunk were larger. Therefore, two sensors on the stem seemed like a good idea.

However, the currently used sensor consisting of three electrodes with needles placed in grooves in the electrodes gives much more stable results and the acceptable differences between sensors installed on opposite sides allow the installation of only one sensor per tree. Last but not least, it is also a matter of budget. More trees measured with a bit lower accuracy is better solution than one half of trees measured with two sensors because of expected bigger differences between sample trees than between two sensors placed on the same tree.

Another point is the height and orientation of the measuring point. Both of these factors are related to the need of maximum homogeneity of sap flow around the circumference of the stem and to minimum influence of environmental factors:

Ambient temperature gradient

It should be noticed that heat balance methods are always based on the measurement of temperature difference, mostly in vertical direction. Ambient vertical gradient clearly interferes with the artificial one which comes into the calculation. The THB method used uses only a temperature difference of 1 K to 3 K. The reason for this is to save the energy required for heating and to minimize the impact on the plant tissue. The protective insulation of the sensors prevents the influence of short-term environmental disturbances such as direct sunlight, wind etc., but there is no insulation that can eliminate the vertical gradient caused by the temperature difference between ground and air. This gradient is the biggest close to the ground. From this point of view is good idea to place the sensors as high as possible.

Sap flow homogeneity

With respect to supposed complex water pathways close to the root swelling, the installation of sensor far above this level is recommended. Also – the higher the better. For the same reason the sensors must be placed below the first branching.

Sensor assembly

The sensors are composed not only from the active parts but also from other parts and/or cooperating sensors. Insulation against direct sunshine, stem increment sensors, data transmitter, IR thermometers measuring the temperature of the stem surface all require their space.

Strangers

Higher installation far from the stranger’s hands is also an issue which should be considered in measuring sites located close to populated places.

Fieldworker's view

Sensor installation made from ladder is quite tough. The most comfortable, with respect to above mentioned limits and still generally acceptable is the installation ca 150 cm above the ground.

Sensor orientation

There is no reason to consider orientation. The most important thing is to choose a place free of uneven spots, branches, etc. Since the measuring point should be reinstalled after two growing seasons, there should be left room for future sensor placement.

4.4. Installation process

4.4.1. Sensor in detail

For correct installation and subsequent data processing, it is necessary to understand the principle of operation of the sensor. Below is a detailed description of the sensor operation, which is based on publications: Čermák et al. (2004) and manufacturer literature (EMS Ltd., CZ).

The sensor consists of the part installed in the tree trunk and the electronics built into a box suspended below the measuring point.

The base of the measuring point consists of three flat plates - electrodes - used to connect the heating AC current to the xylem, and a fourth, reference plate below them. The bottom electrode does not participate in the heating and therefore does not need the insulated part below. However, it is necessary for thermal symmetry with respect to the upper electrodes. The electrodes are available in three lengths covering the radial conductivity profile at 25-, 35- or 45 mm depth. Special tools must be used to drive the electrodes into the trunk so that they are inserted in parallel at the correct depth. The part of the top three electrodes that will be in contact with the highly electrically conductive phloem is insulated with a special adhesive tape.

A set of four needle temperature sensors (1 mm in diameter) is used to measure the temperature difference at the measuring point. The thermocouple junctions are located about 2 mm from the needle tip. The needles are inserted into grooves in the electrodes, which are also 1 mm wide. The depth of the grooves ensures that the temperature sensors are placed in the center of the non-insulated part of the electrodes, which is intended to cover the entire conductive water profile. Together with the high thermal conductivity of the electrodes, this arrangement represents the average temperature in the radial direction. This arrangement also prevents drilling holes in the xylem and possible clogging of the capillaries by wood dust. The temperature sensor forms a common assembly with the clamps for connecting the electrodes. This assembly is suspended from the central electrode and also carries the electronic module via a cable with waterproof Switchcraft connector. This arrangement avoids the installation of any other fixings on the tree trunk.

Note: Since the needles are in direct contact with electrodes where the peek voltage reaches more than 100 V and the sensor output voltage is in order of tens µV, the sensors in needles are thoroughly electrically insulated.

The electronic module contains quite sophisticated electronics which keeps the temperature difference (dT) in the measuring point on the preset level by means of the feedback between the heating power and the average dT of temperature sensors. The sap flow related output is the power necessary for maintaining this difference under different sap flow rates divided to dT in order to make the output values independent on the dT setting. One of the most important features of the measuring system is its dynamics in terms or response of power to sap flow rate changes. It should be aperiodic, that means fast enough but without an overshooting.

There are two main advantages of the strategy to keep dT and control heating power.

- Correct measurement of sap flow dynamics (Kučera et al. 1977). Since the temperature of the heated part of the xylem does not change, there is no measurement lag behind the actual sap flow changes due to the inertia of the xylem tissues.

- Energy consumption. A certain amount of energy is necessary to cover the heat loss through the line from the measuring point independent of the water flow, but the main part of the energy dissipated with the flowing water is directly proportional to the sap flow. This makes obvious advantage for the solar powering because both sap flow and the power source are proportional to sunshine.

- Accuracy - for systems maintaining constant output, the measured temperature difference is lowest at the highest sap flow. However, the lower the temperature difference, the greater the influence of disturbing factors such as the ambient temperature gradient. In systems with controlled dT, the ratio between artificially and naturally induced temperature differences remains constant and the accuracy of sap flow measurement during the day is not reduced.

- Tissue overheating. The target temperature difference at the measurement site must not fall below a reasonable limit with respect to the required accuracy. However, with constant power and no water movement, the temperature of the heated parts can reach ten times higher values and disturb the plant tissue. Maintaining a constant temperature allows a good compromise between the effect on the plants, reasonable accuracy and acceptable energy consumption.

Besides of sap flow values, the sensor gives on the output even more variables:

- Power supply voltage
- AC resistance between electrodes
- Temperature inside the electronic module
- Operating status (error codes)
- dT on each electrode
- Stem increment value – if the compatible dendrometer is connected

There are two sensor modification with respect of operation – datalogging sensors and sensors ready for connection to SDI-12 network. The SDI-12 sensors operated according to Serial-Digital Interface Standard for Microprocessor-Based Sensors Version 1.3.

Datalogging sensors require cooperation with Mini32 software, developed by EMS for handling with all sensors and dataloggers manufactured by EMS company and for advanced data processing, mainly manipulation with files in terms of easy preparing the database of variables measured with more systems even with different reading period.

4.4.2. Sensor installation

The procedure for installing the sensor is described in detail in the operating instructions, the following notes highlight the most important issues:

There are never enough reminders to follow all the details of the installation procedure. There is a reason for every detail - underestimating the details could affect the reliability of the measurement. In particular, the insulation needs to be placed correctly on the trunk.

It is also important to keep in mind the limited life of measurement points, especially for fast-growing trees. It is recommended to reinstall the measuring point on the same tree after one or two years (Cienciala et al. 1999). Then, due to growth, the electrodes could get stuck deep in the xylem and are subsequently difficult to remove. Also, sap in rapidly growing trunks tends to detach from the electrodes, which could underestimate the measured sap flow values.

4.4.3. Sensor configuration

There are two important changeable configuration parameters

- Measurement time and averaging and writing time to memory. The difference between these periods is a trade-off between the size of the data sets and the resolution of the measurements. Since the dynamics of sap flow are not that high, the most common setting is to measure every minute and storing ten-to-thirty-minute averages.

- Preset temperature difference at the measuring point. A higher dT reduces the influence of various disturbing factors but increases the power consumption approximately in proportion to the dT - increasing the dT from 1 K to 2 K also doubles the power consumption. Also, highly transpiring species (mostly deciduous) could hit the limit of the maximum heating power. The higher dT is recommended in case of suspicious strong influence of external temperature gradients. In such a case is a good idea to set higher dT for couple of days and compare measured data between and after dT increase. If there is not a significant difference, set the dT back to recommended optimal 1 K.

5. Data processing

Processing of sap flow variables has following steps:

- Cleaning the data
- Baseline subtraction
- Scaling up to the canopy level – transpiration
- Parametrization of Penman-Monteith equation

5.1.1. Cleaning of data from artefacts and invalid variables

Each data file originated from the measurement of sap flow contains values which should be deleted for some reasons. The most common reasons for invalid data are:

- Stem flow - water is passing downwards along sensor below insulation. It mostly causes something which looks like a sap flow at time when the evaporation demands are close or equal to zero. Those values should not be deleted but set to zero during the baseline subtraction procedure explained later.

- False values caused by external factors in the time period without leaves when there is no sap flow from the definition. From the analysis (Tatarinov et al. 2005) the effect of disturbing ambient factors rises with decreasing sap flow rate. Zero flow is the most vulnerable to this.

- Instrumental issues due to low powering voltage, sensor or datalogger malfunction, broken cables etc.

- Missing values due to electrical resistance between electrodes outside the technical limits. In this case, the sensor stops the measurement. Then, it checks the resistance every 20 minutes, waits for an acceptable value and tries to resume the measurement.

- The power required to maintain the temperature difference is at the upper limit. If a long warm-up at full power lasts more than 10 minutes, the sensor stops working for half an hour. The main reason for this is that the system cannot distinguish between an extremely high sap flow and a failure of the temperature sensors when a long heating with maximum power for a long time could damage living tissues with high temperature.

If the heating of the sensor has been interrupted for a longer period of time, the system needs some time to reach a steady state with the set temperature difference. During this time, the measured power value does not correspond to reality. Even a short power interruption during a battery change will cause a visible response.

Therefore, all values affected by these artifacts must be removed from the data set. Mini32 offers a "delete" option, but the final decision on what to delete is up to the user and their experience.

5.1.2. Baseline subtraction

As it was mentioned earlier, the power necessary for maintaining the temperature difference is proportional to volumetric sap flow magnitude but it also covers the conductive heat loss from the heated space. The value of heat loss depends on many factors which are hardly estimated, like xylem water content, sapwood depth, growth, bark wetness etc. The used measuring method is not able to measure this loss so an indirect methods of its estimation must be used. The simple method is just to connect the expected lowest values of daily sap flow patterns – mostly at 4:00 a.m. - into a broken or strait line which represents the value of heat losses in terms of sap flow - sometimes it is named as a "fictive flow" – and then to subtract them from measured values.

Therefore, there is a question if to consider each night value as a part of a broken line designed through the whole season or to set a long strait line involving the most of nearest point? Or something in between these approaches?

After many years of thought and data analysis, it has been confirmed that the daytime values of sap flow are not affected by the short fluctuations of the nighttime values. The final solution combines both approaches (Kučera et al. 2019) - Exponential Feedback Weighting method. The method considers sap flow values each day at 4:00 a.m. Such values are smoothed using exponential moving average. This approach was tested on many sample data sets and confronted with both above mentioned approaches and with the estimation based on my experience taking into consideration many ambient factors like air temperature and humidity, precipitation, dew point temperature. This method was incorporated into the Mini32 software although it leaves a space for a manual operation.

5.1.3. Scaling up

The goal of scaling the data measured on the sampled trees to the whole stand is to find the coefficient by which we must multiply the sap flow of the sampled trees to obtain the sap flow of all trees growing in the 1 ha canopy area represented by the sampled trees (Čermák et al. 2004). The total sap flow of all these trees divided by 10 000 gives then the canopy transpiration in [mm].

The first task before the scaling procedure is checking uniformity among sampled trees throughout the season. The good idea is to create the correlation matrix describing the relationship between trees. It should be performed on data sets with the original resolution. This takes into account not only the amplitude but also the diurnal patterns caused mainly by the different social position of the sampled trees.

The correlation matrix clearly distinguishes between the mainstream group of similarly behaving trees and trees that are out of the mainstream. The reasons for this are usually social status, health status or limited availability of soil water. If there is a sample tree whose sap flow record is out of mainstream, it could be removed from the dataset. However, the reason for the unusual behavior of such a tree needs to be understood.

Limited water supply is the most common issue. Given the sap flow proportional to the difference between leaf and soil water potential, low values of soil water potential require also lower leaf water potential. Below a certain value of leaf water content, the leaf water potential is not able to compensate decreasing soil water potential. The stomata get closer and transpiration and consequently sap flow are reduced. However, this limit does not occur at the same time and with the same intensity in all trees. Therefore, a scaling procedure based on a time period of limited water availability is almost useless for analyzing differences between different species or canopies under their normal physiological conditions since the scaling procedure assumes stable relationship between trees.

On the other hand, for detail analysis of canopy response to drought the scaling procedure can be done at any time period of reduced water availability separately.

The second step of procedure already supposes

- Well prepared database of sample trees. Sap flow in [$kg.h^{-1}$].
- DBH of all living trees from a defined area of known size around the sample trees. Approximately ¼ ha at least.

Process:

From the database of sampled trees, a period of couple (five to twenty) complete days with readily available soil water, fully developed leaf area, high evaporative demand and with minimum rainy days should be selected. In this time period, mean values of all trees is calculated ("Variable statistics" from pop-up menu in Mini32 graphics).

In the next step, a graph describing the dependence of the above values of the average sap flow on the DBH of sample tree is created (in Excel®) and the regression parameters are calculated. If a reasonable correlation exists between these variables, the scaling procedure can be based on the regression parameters from the graph. Such a regression is expected to be linear and to intersect the DHB axis at approximately 10 to 20 cm according to overall size of trees. This is the size of the "smallest tree in the canopy" whose contribution to the entire transpiration is negligible (Čermák et al. 2004).

Then, the estimated sap flow values of each tree in selected area are calculated according to regression parameters and then summarized. This way the sap flow of all trees growing on the sampling area in selected time period is calculated. Multiplying the ratio between 1 ha and the size of the sampling area gives the sap flow of all trees per 1 ha.

Finally, the ratio between the calculated sap flow of all trees per hectare and the sum of the sample trees gives the scaling factor. Multiplying the sum of the sap flow of the measured sample trees by this factor and dividing by 10 000 (m^2 per ha) gives the canopy transpiration in [mm]. However, this calculation works only under the same soil water (and phenological) status as it was at the time period of scaling.

If the regression between DBH of the sample trees and sap flow values is insignificant, another ratio between all trees and the sample trees (DBH, stem cross-sectional area, sapwood area, etc.) must be used as a scaling factor.

5.1.4. Related variables.

Besides standard meteorological variables:

- global solar radiation [$W.m^2$]
- net radiation [$W.m^2$]
- air temperature [°C]
- air humidity [%]
- wind speed and direction [$m.s^{-1}$, deg.]
- soil heat flux [$W.m^2$]

also vapor pressure deficit and a potential transpiration are required for the further data processing:

Saturated vapor pressure is calculated from the air temperature:

$$e_s = 6.112 * exp\left(17.62 * \frac{\mathrm{T}}{(\mathrm{T}+243.12)}\right) * 100 \qquad [1]$$

Vapor pressure deficit

$$D = e_s.(1 - \frac{r.h.}{100}) \qquad [2]$$

Potential transpiration (Penman 1948)

$$\lambda.E_p = \frac{\delta(R_n - G) + \gamma Da(1 + b.w)}{\delta + \gamma} \qquad [3]$$

where (besides of those already described):

e_s - saturated vapor pressure [Pa]

r.h. – relative humidity [%]

γ - psychrometric constant [Pa/K]

δ – slope of saturation water vapor pressure deficit [Pa/K]

E_p – potential transpiration [$kg.m^{-1}.s^{-1}$]

λ – water heat capacity [J/kg]

a, b – empirical parameters according to evaporating surface

5.1.5. First look at the data set

It is recommendable to take a look at the annual courses of measured data set in order to see the relationship between transpiration and related variables – mainly radiation, vapor pressure deficit, potential transpiration. It helps to see the similarity between PET and transpiration, start and end of growing seasons, possible time period of limited water supply.

It is good idea to compare daily patterns of both transpiration and PET variables in cloudless or nearly cloudless days throughout the season. This is the way how to get familiar with the whole data set before next calculations.

5.1.6. Parametrization of Penman-Monteith equation.

P-M equation is based on the original Penman equation (Penman 1948) describing the evaporation from different wet surfaces. The Penman equation [3] consists of two parts – first one includes (besides of constants and parameters) net solar radiation, soil heat flux and temperature. All those variables are relatively easy measurable. However, the second part includes air humidity (required for calculation of VPD) but also the effect of wind as important factor driving evaporation. Since his theory was developed quite long ago, the contribution of wind was estimated empirically for different surfaces.

Later, when the theory of turbulent diffusion became more common, the empirical part of the Penman equation was put on a theoretical basis. Two more variables were included into calculation – aerodynamic conductance (g_a) and canopy conductance (g_c):

Penman-Monteith equation:

$$\lambda . E = \frac{\Delta(R_n - G) + \rho c_p D g_a}{\Delta + \gamma\left(1 + {}^{g_a}/{}_{g_c}\right)} \qquad [4]$$

where (besides of those already described):

E – canopy transpiration [mm]

g_a - calculated from wind speed with the knowledge of aerodynamic parameters of the canopy:

$$g_a = \frac{k^2 u}{\ln\left[{(z-d)}/{z_{oh}}\right] * \ln\left[{(z-d)}/{z_{om}}\right]} \qquad [5]$$

where (besides of those already described):

c_p – specific heat of air [J.m^3]

ρ – density of dry air [kg.m^3]

g_a – aerodynamic conductance [s.m^{-1}]

g_s – canopy (stomatal) conductance [s.m^{-1}]

k – von Karman constant [-]

d – zero plane displacement [m]

z_{om} – canopy roughnes (for momentum) [m]

z_{oh} – canopy roughnes (for heat and vapor) [m]

z – wind speed measurement height [m]

u – wind speed [m.s^{-1}]

Note: Aerodynamic parameters can be measured using at least two anemometers installed above the canopy, but they can be derived from the height and structure of the canopy. Also, the anemometer measuring the wind speed for the calculation should be placed above the canopy.

Supposing that we are able to measure the canopy transpiration, canopy conductance is the only unknown variable. The current standard method of estimation of g_c is based on the inversion of the P-M equation such a way, that the g_c is calculated as a function of measured variables (Martin).

However, g_c is also supposed to be driven by meteorological variables. The commonly used Lohammar equation (Lohammar et al. 1980) describing influence of R_g and VPD to stomatal conductance is written in the form assuming stomata opening during solar activity and stomata closing due to high evaporating demands:

$$g_c = \frac{R_g}{R_g + R_0} \cdot \frac{g_{cmax}}{1 + aD} \qquad [6]$$

where:

Rg – global solar radiation [W.m^2]

D – vapor pressure deficit [Pa]

g_{cmax} – maximal conductance [s.m^{-1}]

R_0, a – parameters

In the next step, the parameters of this equation are determined by regression analysis of both variables (Cienciala et al. 1999, Martin et al. 1997).

The particular approach used in this methodology is based on a direct regression analysis of the P-M equation with the Lohamar formula already included, but in a different form that gives the second part more degrees of freedom (parameter "b"). Also, a minimal stomatal conductivity is introduced as g_{min} (Urban, personal communication):

$$g_c = \frac{R_g}{R_g + R_0} \cdot g_{lim}.\left(0.5 - \frac{1}{\pi}.arctg\left(\frac{D}{a} - b\right)\right) + g_{min} \qquad [7]$$

The final equation entering the fit module of Mini32 looks like this:

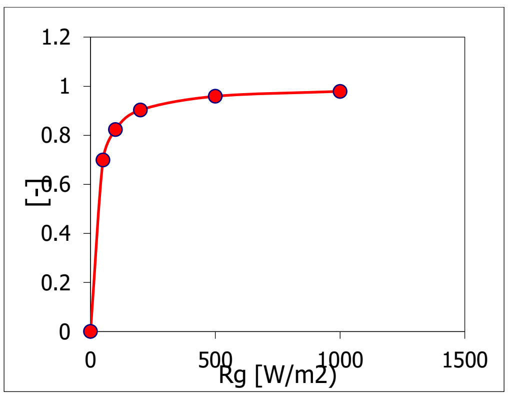
$$E = \frac{(\Delta.(R_n - G) + \rho.c_p.D.g_a)/\lambda}{\Delta + \gamma.(1 + \frac{g_a}{\frac{Rg}{Rg+R_0}.g_{lim}.(0.5-\frac{1}{\pi}.arctg\left(\frac{D}{a}-b\right)+g_{lim})})} \qquad [8]$$

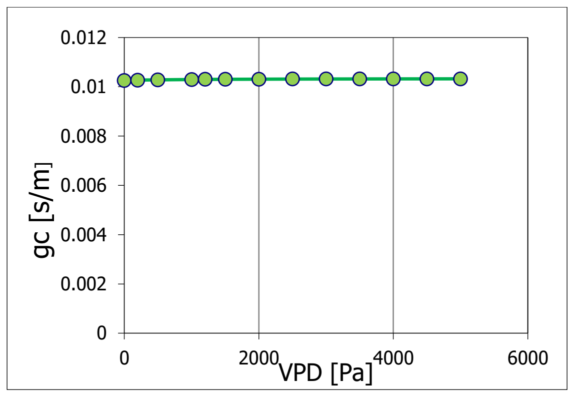
Note: The g_{lim} here is not equal to g_{max}. It must be calculated later as a value for VPD = 0.

After the fitting procedure, the unknown parameters of equation [8] – R_o, a, b and g_{min} are found and the chart describing stomata closure as a function of Rg and VPD can be created.

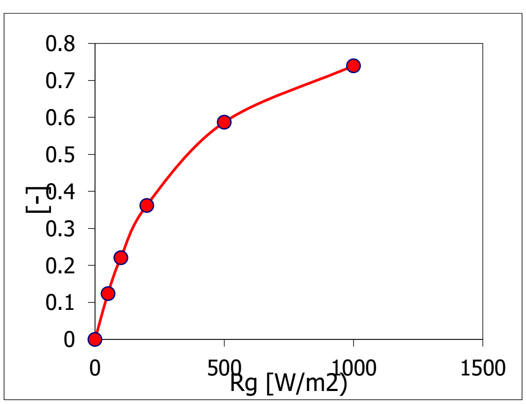
Example - comparison of stomatal control strategies of two forest tree species under unlimited soil water supply.

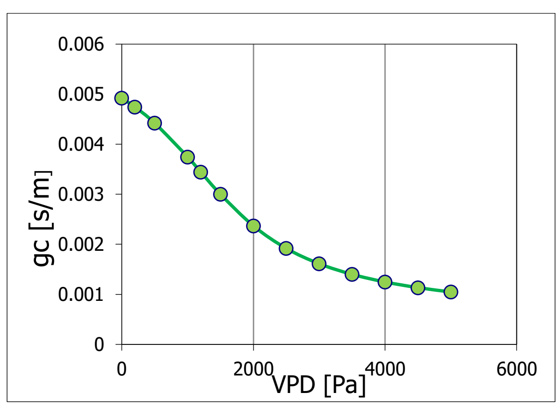
Beech





Spruce





6. Summary

The aim of all the measurements and data processing described in this text is to understand the response of plants to environmental variables. Manipulation of stomata is the only way to protect the conducting parts of the plant from damage due to the extremely low water potential of the xylem. Different plant species have different strategies when it comes to regulating stomatal conductance (Jones 1983). An analysis of their strategies should help forestry and agriculture achieve maximum efficiency, regardless of whether the strategy is one of yield or simple survival under adverse conditions.

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