

Thermographic measurements on plant leaves

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ABSTRACT

An important process of plant physiology is the transpiration of plant leaves. It is actively controlled by pores (stomata) in the leaf and the governing feature for vital factors such as gas exchange and water transport affixed to which is the nutrient transport from the root to the shoot. Because of its importance, the transpiration and water transport in leaves have been extensively studied. However, current measurement techniques provide poor spatial and temporal resolution. With the use of one single low-NETD infrared camera important parameter of plant physiology such as transpiration rates, heat capacity per unit area of the leaf and the water flow velocity can be measured to high temporal and special resolution by techniques presented in this paper. The latent heat flux of a plant, which is directly proportional to the transpiration rate, can be measured with passive thermography. Here use is made of the linear relationship between the temperature difference between a non transpiring reference body and the transpiring leaf and the latent heat flux. From active thermography the heat capacity per unit area of the leaf can be measured. This method is termed active, because the response of the leaf temperature to an imposed energy flux is measured. Through the use of digital image processing techniques simultaneous measurements of the velocity field and temporal change of heated water parcels travelling through the leaf can be estimated from thermal image sequences.

Keywords: Transpiration Rate, Heat Capacity, Active Thermography, Passive Thermography, Motion Detection

1. INTRODUCTION

Through the advances and availability of modern low noise infrared cameras the wide spread application of thermographic measurements to a number of scientific fields become feasible. This allows for an unobtrusive measurement of physical parameters to a high spatial and temporal resolution. Plants leaves can be thought to consist mainly of water. Their water content ranges from 70% - 98%, depending on the species.¹ To maintain such a high water content the transport of water from the root to the shoot is very important. Affixed to this transport of water is the transport of vital nutrients and minerals, indispensable for a functioning metabolism of the organs. As plants are sessile organism, they cannot escape from unfavorable environments. Hence they have to adapt to the temperature in their habitat or develop means of only growing when favorable conditions are present. Examples of the latter are annual plants that survive unfavorable conditions as seeds or biannual species that survive cold periods as bulbs or rhizomes.²

In order to maintain a temperature most favorable for their metabolism, plants have developed a number of thermoregulation functions. Passive thermoregulation involves altering the proportion of reflected radiation (e.g. by white hairs, salt crusts, etc.) or varying the inclination of leaves. The only short term thermoregulation available to plants is their *transpiration*. This actively controlled loss of water vapor occurs through pores (stomata) formed by a complex of cells in the outermost cell layer of leaves. This transport is driven though the concentration gradient of water vapor in the leaf-internal gas space and the surrounding atmosphere. The cells that form the stomata control transpirational water loss by altering the aperture of the pores the conductance of the leaf for water vapor.³

Apart from their thermoregulation function, stomata present the main link in between external atmosphere and leaf-internal gas space. Gas exchange of CO₂ and O₂ takes place through the same stomata. The transpiration stream provides the major route of nutrient transport from the root to the shoot. Thus the transpiration rate is directly connected to the

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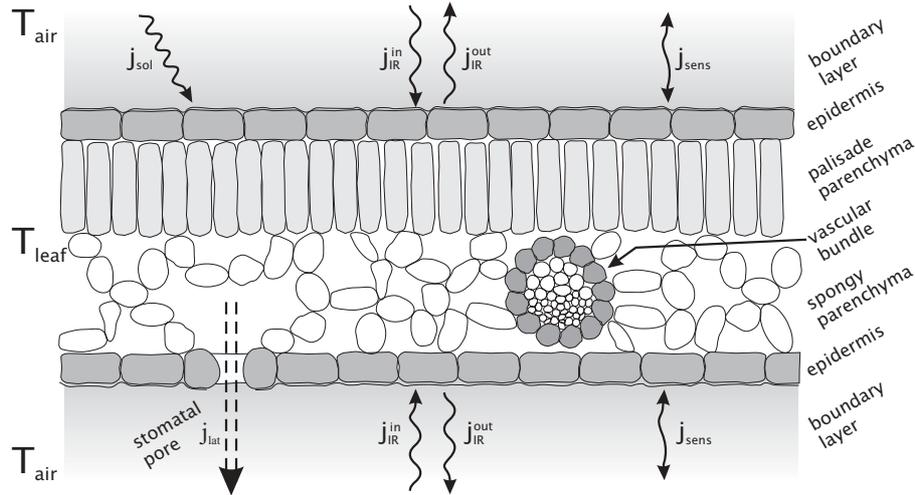


Figure 1. Schematic cut through a leaf. It shows the constituent fluxes of the net heat flux as well as the distinctive functional layers.⁵

nutrient transport and indirectly to the gas exchange of the plant leaf. Due to this integration of many vital transport phenomena in plant leaves, the stomata are highly controlled by external and internal factors. The transpiration of plants has been studied extensively owing to their importance to in the field of plant physiology. However, currently available techniques possess the major drawback of being point measurements only, with long integration times.⁴

Thermographic schemes for measuring the water relations in plants become feasible due to the high absorption of water in the infrared wave spectrum. In conjunction with the high water concentration of typical plant leaves this leads to radiation in the infrared being absorbed almost exclusively by the water. Plant leaves possess a complex heterogeneous internal structure as can be seen from Figure 1. Due to this inhomogeneity different parts of the leaf will contain different amounts of water per area, affecting its thermal properties. The spatially resolved thermal properties of leaves have not been studied intensively yet, due to the lack of appropriate techniques.

Novel quantitative thermographic techniques for measuring the heat capacity of leaves, the latent heat flux and the water velocity will be presented in this paper. In Section 2 the underlying equations will be presented.

2. HEAT FLUXES IN PLANT LEAVES

Following the law of conservation of energy, the energy efflux of a body has to equal the energy influx plus energy stored in the body. The energy stored in leaf consists of *photosynthesis*, other metabolic processes and changes in the leaf temperature. Typically, photosynthesis and other metabolic processes account for less than 1% of the influxes³ and can thus be neglected. Thus, the net heat flux density j_{net} at the plant leaf is given by

$$j_{net} = j_{lat} + j_{sens} + j_{IR} + j_{sol}, \quad (1)$$

with the *latent heat flux density* j_{lat} , the *sensible heat flux density* j_{sens} and the *radiative heat flux density* made up of the *longwave and shortwave flux densities*, denoted by j_{IR} and j_{sol} respectively. In the following influxes are represented as positive and effluxes as negative numbers. Fluxes will be expressed as flux densities in the unit Wm^{-2} .

Of interest is the *transpiration rate*, which is expressed as the *mass flux of water vapor* j_{wv} , given by

$$j_{wv} = -j_{lat}/\lambda, \quad (2)$$

where λ is the latent heat of evaporation of water and the negative sign comes about due to the loss of energy caused by an efflux of water vapor. In biological applications the water vapor flux is expressed in molar units. The conversion is achieved by dividing j_{wv} with the molecular weight of water M_{H_2O} leading to

$$j_{wv}^{mol} = -\frac{j_{lat}}{\lambda M_{H_2O}}. \quad (3)$$

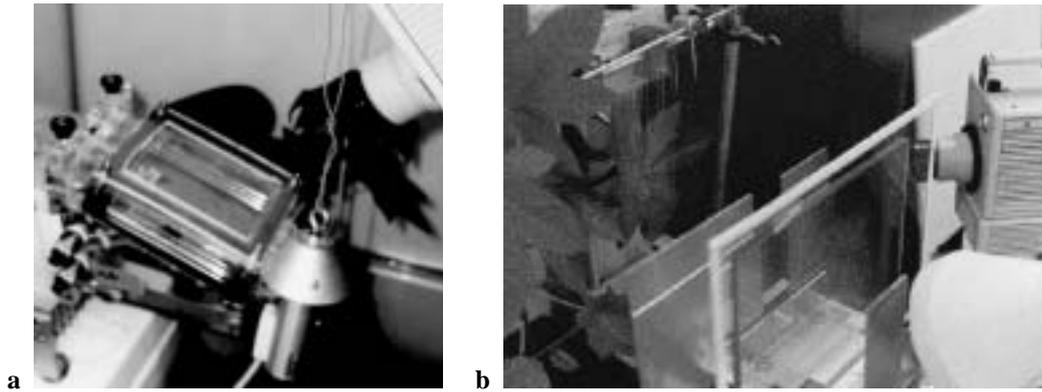


Figure 2. **a** The set-up of the passive thermographic method. The IR camera in the upper right corner is imaging the leaf of the plant fixed inside the cuvette. **b** The set-up of the active thermographic technique. The IR radiator illuminates the leaf through a set of screens and filters.⁵

In order to measure this transpiration rate one has to accurately estimate the latent heat flux j_{lat} . This quantity can be derived from *Fick's first law* for a known difference of the relative humidity between leaf and atmosphere. Measuring the transpiration rate in this fashion has the drawback of only allowing point measurements with relatively long integration times. By employing a modern low noise mid range IR camera this drawback can be circumvented. By noting that the constituent fluxes of Equation (1) are the same for non-evapotranspiring reference body with roughly the same optical properties as the leaf under investigation as for the leaf itself. However, the latent heat for such a body will vanish. By comparing such a reference body with the transpiring leaf the latent heat and thus the transpiration rate can be deduced. This technique for measuring the transpiration rate is termed *passive thermography* and will be described in Section 3.2.

3. EXPERIMENTAL TECHNIQUES

3.1. Set-up

In the following sections experimental techniques relying on passive and active thermography will be introduced. The set-up of those techniques is very similar. The tip of the central lobe of the leaf of a *Ricinus communis* plant is clamped between the two halves of a standard gas exchange cuvette made from acrylic glass, as can be seen in Figure 2. The whole set-up is placed in a 1.5 m³ thermally insulated chamber with humidity and temperature control and an artificial light source to allow for reproducible conditions.

A gas exchange system provides control of the relevant parameters such as temperature, wind speed, humidity and gas concentration inside the cuvette. With the cuvette all important parameters such as internal and external air temperature, air humidity, light intensity and gas concentration such as CO₂ are measured. To allow a bulk estimate of the exchange rate at the leaf, concentrations and humidity are measured at the input and output gas paths of the cuvette.

The passive thermographic technique relies on relating the temperature of the transpiring leaf to a reference body. As such a small brass plate (1 cm²) is coated with Tetonal Photolack, a black varnish with an infrared emissivity of 97%. The plate is then placed inside the cuvette next to the leaf.

For the active thermographic technique an additional energy flux $j_{act}(t)$ has to be imposed on the leaf. As an energy source a medical infrared radiator (Philips 150 E R95E) is used. In order to avoid reflexes in the thermal images the wavelength range of the radiator has to be limited to ranges outside the wavelength window the IR camera is sensible to. This is achieved by a set of filters placed in the optical path of the radiator. The radiator and the IR camera are controlled by the same computer thus facilitating synchronization of the heat source with the image acquisition.

The infrared camera used for the measurements described in this paper is an Amber Radiance I MWIR camera. The InSb detector is sensitive to a wavelength range of 3-5 μm. It has to be actively cooled to 70 K by a Stirling cooler. The noise level of the camera is characterized by a NEDT = 25 mK. The detector is a square FPA with a resolution of 256 × 256 pixel. The camera is capable of a frame rate of 60 Hz.

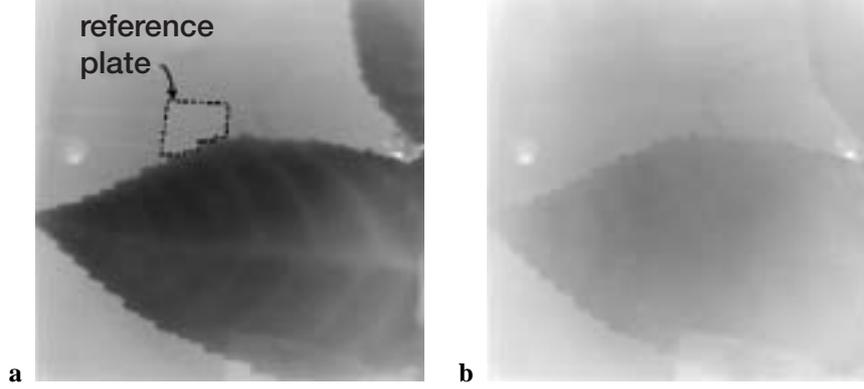


Figure 3. Images of the passive thermography. **a** Low temperature of the leaf as compared to the reference plate indicates high transpiration rates. **b** The temperature of the leaf is almost the same as the reference plate because almost all stomata are closed.

3.2. Passive thermography

The underlying principle of the passive thermography is observing both the leaf and a non-transpiring reference body. This technique is termed *passive* as both leaf and reference body are observed only, with no external heating employed.

The reference body should have the same optical properties as the leaf. Due to the experimental set-up, both leaf and reference body will be subject to the same energy fluxes. Hence all the fluxes are equal, except the flux of latent heat. For both, reference body and leaf, the equations of the type (1) can be solved for the transpiration rate j_{wv}^{mol} , resulting in⁶

$$\begin{aligned}
 j_{wv}^{\text{mol}} = & \frac{1}{\lambda M_{H_2O}} \left\{ (8\epsilon_{\text{IR}}^{\text{leaf}} \sigma T_{\text{ref}}^3 + 2\rho c_p k_{\text{heat}}^{\text{leaf}}) \Delta T \right. \\
 & + \left(\frac{C}{A} \right)_{\text{leaf}} \frac{d}{dt} T_{\text{leaf}} - \frac{\epsilon_{\text{IR}}^{\text{leaf}}}{\epsilon_{\text{IR}}^{\text{ref}}} \left(\frac{C}{A} \right)_{\text{ms}} \frac{d}{dt} T_{\text{ref}} \\
 & + 2\rho c_p \left(k_{\text{heat}}^{\text{leaf}} - \frac{\epsilon_{\text{IR}}^{\text{leaf}}}{\epsilon_{\text{IR}}^{\text{ref}}} k_{\text{heat}}^{\text{ref}} \right) (T_{\text{air}} - T_{\text{ref}}) \\
 & \left. + \left(\epsilon_{\text{sol}}^{\text{leaf}} - \frac{\epsilon_{\text{IR}}^{\text{leaf}}}{\epsilon_{\text{IR}}^{\text{ref}}} \epsilon_{\text{sol}}^{\text{ref}} \right) j_{\text{in}} \right\}, \quad (4)
 \end{aligned}$$

where use was made of the approximation $T_{\text{ref}}^4 - T_{\text{leaf}}^4 \approx 4T_{\text{ref}}^3 \Delta T$, which is valid for small $\Delta T = T_{\text{ref}} - T_{\text{leaf}}$ ($\delta T < 5$ K). In this equation T_{ref} and T_{leaf} are the absolute temperatures of the reference body and the leaf, σ is the Stefan-Boltzmann constant, ϵ_{IR} and ϵ_{sol} are the emissivities for infrared and solar radiation respectively, ρ is the density of air, c_p is the specific heat capacity of air, (C/A) is the heat capacity per unit area, j_{in} is the incident radiation flux density and k_{heat} is the heat transfer velocity, defined as

$$k_{\text{heat}} = -\frac{D}{\Delta x}. \quad (5)$$

Here D is the diffusion coefficient and Δx the diffusion length.

From Equation (4) it becomes evident that there exists a linear relationship between the transpiration rate j_{wv}^{mol} and the temperature difference ΔT . The terms in row two to four represent an offset which shall be denoted by X . Equation (4) can thus be shortened by writing

$$j_{wv}^{\text{mol}} = b k_{\text{heat}}^{\text{leaf}} \cdot \Delta T + X. \quad (6)$$

By comparing this equation with Equation (4) it becomes evident that the factor b is known or can be measured quite easily. By taking some experimental precautions the offset term X can be simplified somewhat. If the reference body is chosen so that its optical properties are very similar to those of the leaf and its temperature is the same as that of the air, then this offset term can even be neglected. This is of course only true if the temperature of both reference body and leaf change slowly, which is a valid assumption under laboratory conditions.

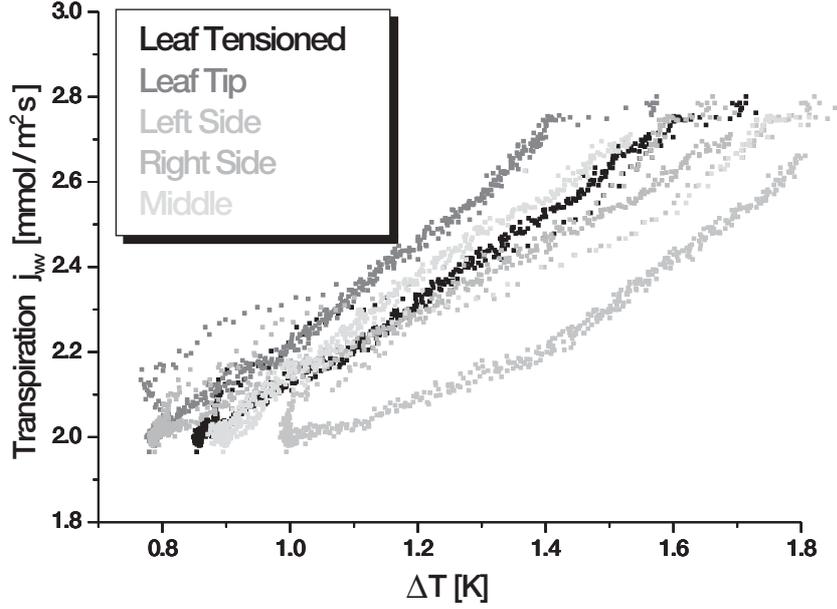


Figure 4: The transpiration rate j_{wv} plotted against the temperature difference ΔT for different parts of a leaf.⁷

In Figure 3 two images of a typical measurement are shown. The transpiring leaf is much colder than the reference plate, which is not the case for a leaf with closed stomata. As can be seen the temperature difference ΔT between reference body and leaf can be computed for every individual pixel of the leaf, thus allowing a spatially resolved estimate of the transpiration rate j_{wv}^{mol} for a known heat transfer velocity. The importance of spatially resolved measurements is emphasized in Figure 4. Here the transpiration rate is calculated from the difference of water vapor concentration in between the input and output gas paths of the cuvette. The net transpiration rate is then plotted against measures of ΔT at different locations of the leaf. Only through a spatially resolved measurement are strongly diverging measurements apparent depending on the location of measurement at the leaf.

3.3. Active thermography

The heat capacity of the leaf can be measured spatially resolved by active thermography. This technique is called active, as the leaf's thermal response to an imposed heat flux is analyzed. The energy budget of the leaf is given by (1), or expanding the constituent fluxes

$$\left(\frac{C}{A}\right)_{\text{leaf}} \frac{d}{dt} T_{\text{leaf}} = \epsilon_{\text{sol}}^{\text{leaf}} j_{\text{in}} + 2\epsilon_{\text{IR}}^{\text{leaf}} \sigma (T_{\text{surr}}^4 - T_{\text{leaf}}^4) + 2k_{\text{heat}}^{\text{leaf}} \rho c_p (T_{\text{air}} - T_{\text{leaf}}) + k_{wv} \lambda c_{wv} (h_{\text{leaf}} - h_{\text{air}}). \quad (7)$$

Assuming that $T_{\text{surr}} = T_{\text{air}}$, this equation can be simplified by substituting $T(t) = T_{\text{air}} - T_{\text{leaf}}(t)$

$$-\frac{d}{dt} T(t) \approx \left(\frac{A}{C}\right)_{\text{leaf}} \left\{ \epsilon_{\text{sol}}^{\text{leaf}} j_{\text{in}} + j_{\text{lat}} + j_{\text{act}}(t) + (8\epsilon_{\text{IR}}^{\text{leaf}} \sigma T_{\text{air}}^3 + 2\rho c_p k_{\text{heat}}^{\text{leaf}}) T(t) \right\}, \quad (8)$$

where use was made of the approximation $T_{\text{air}}^4 - T_{\text{leaf}}^4(t) \approx 4T_{\text{air}}^3 T(t)$, which is valid for small $T(t)$. In Equation (8) an additional heat flux $j_{\text{act}}(t)$ is introduced, accommodating the actively controllable heat source.

Equation (8) can be written as

$$\frac{d}{dt} T(t) = -\frac{1}{\tau} T(t) - b - z(t), \quad (9)$$

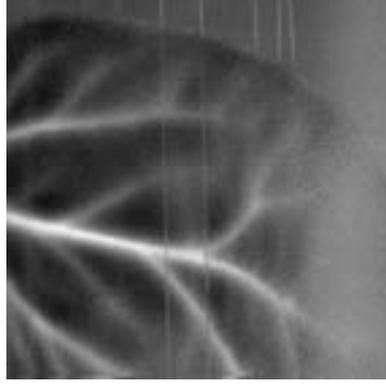


Figure 5: Parameter image of the time constant τ from the constant flux method.⁵

where the following substitutions have been introduced

$$\tau^{-1} \equiv \left(\frac{A}{C}\right)_{\text{leaf}} (8\epsilon_{\text{IR}}^{\text{leaf}} \sigma T_{\text{air}}^3 + 2\rho c_p k_{\text{heat}}^{\text{leaf}}) \quad (10)$$

$$b \equiv \left(\frac{A}{C}\right)_{\text{leaf}} (\epsilon_{\text{sol}}^{\text{leaf}} j_{\text{in}} + j_{\text{lat}}) \quad (11)$$

$$z(t) \equiv \left(\frac{A}{C}\right)_{\text{leaf}} j_{\text{act}}(t). \quad (12)$$

The differential equation (9) can be solved with an appropriately chosen $z(t)$. In the following two choices of this function will be discussed: a step change in the heat flux $j_{\text{act}}(t)$ referred to as the *constant flux method* and a periodic variation hence called *periodic flux method*.

3.3.1. Constant flux method

For the constant flux method the incident heat flux $j_{\text{act}}(t)$ is switched on at the time $t = 0$. For this case the solution to Equation (9) is found to be

$$T(t) = T^\infty + \Delta T e^{-t/\tau}, \quad (13)$$

where T^∞ is the temperature difference between air and leaf in the new steady state and ΔT the temperature increase of the leaf due to the additional flux. It can be shown that ΔT is given by $\Delta T = z\tau$.⁵ For actually measuring the temperature response of a leaf with respect to the incident heat flux j_{act} , Equation (13) can be expressed in absolute temperatures. Substituting $T = T_{\text{air}} - T_{\text{leaf}}(t)$ leads to

$$T_{\text{leaf}}(t) = T_{\text{leaf}}^\infty - \Delta T e^{-t/\tau}. \quad (14)$$

The goal of the constant flux method is to gain spatially highly resolved measures for the parameters T_{leaf}^∞ , ΔT and τ . This can readily be achieved by employing a single low noise infrared camera. By acquiring an image sequence starting synchronized with switching on the incident heat flux j_{act} at the time $t = 0$, the temperature change of the leaf $T_{\text{leaf}}(t)$ is recorded to a spatial resolution limited only by the infrared camera. The exponential function of Equation (14) can then be fitted to the temporal development of each individual pixel's temperature using the *Levenberg-Marquardt* nonlinear least squares fit.⁸ In this fashion spatially highly resolved estimates of the parameters T_{leaf}^∞ , ΔT and τ are achievable. The parameter image of τ obtained in this fashion is shown in Figure 5.

We are set out to measure the heat capacity of the leaf under observation. This can readily be achieved by solving Equation (10) for the heat capacity, resulting in

$$\left(\frac{C}{A}\right)_{\text{leaf}} = \tau (8\epsilon_{\text{IR}}^{\text{leaf}} \sigma T_{\text{air}}^3 + 2\rho c_p k_{\text{heat}}^{\text{leaf}}). \quad (15)$$

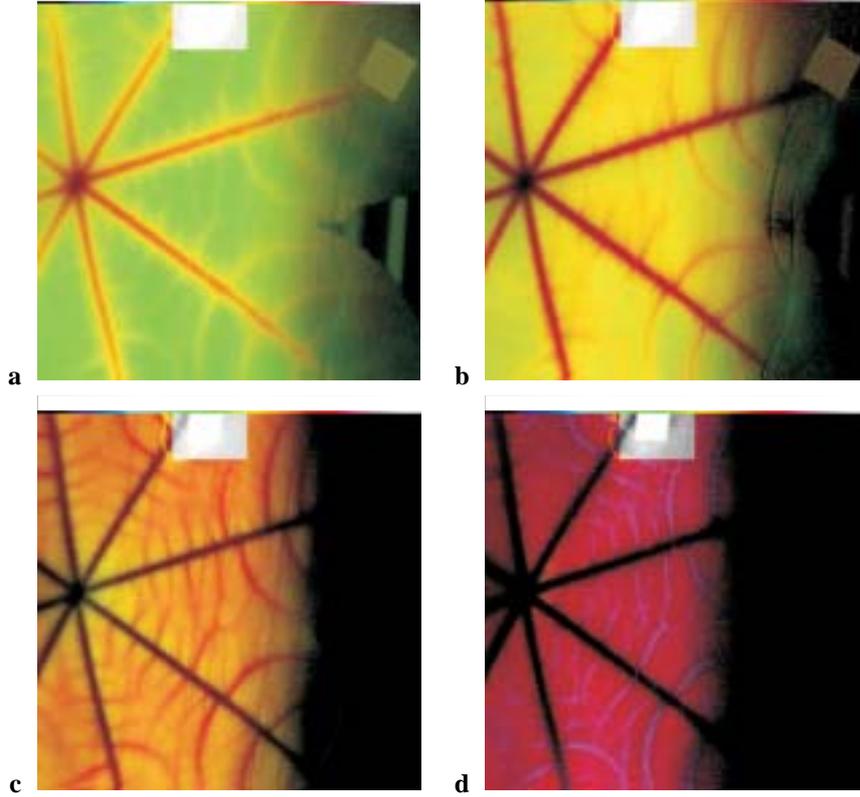


Figure 6. Parameter images depicting the phase $\varphi(\omega)$ and amplitude for different input frequencies ω : **a** $\omega = 2\pi 0.0039$ rad/s, **b** $\omega = 2\pi 0.0078$ rad/s, **c** $\omega = 2\pi 0.0156$ rad/s, **d** $\omega = 2\pi 0.0313$ rad/s.⁶

The heat capacity can thus be obtained from the estimate of τ if the transfer velocity $k_{\text{heat}}^{\text{leaf}}$ is known. All the other quantities in Equation (15) can easily be measured or are known constants ($\epsilon_{\text{IR}}^{\text{leaf}} = 0.975$, $\rho = 1.2 \text{ kg m}^{-3}$, $c_p = 1007 \text{ J kg}^{-1}\text{K}^{-1}$). As was explained in Section 3.2, the transfer velocity $k_{\text{heat}}^{\text{leaf}}$ can be measured spatially resolved from from passive thermography. Hence all the unknowns in Equation (15) can be estimated from thermography and thus the heat capacity computed. In the interveinal area typical values for the heat transfer velocity are found to be $k_{\text{heat}}^{\text{leaf}} = 8.84 \text{ mms}^{-1}$ with $\tau_{\text{iv}} = 25 \text{ s}$. In these areas the heat capacity is then

$$\left(\frac{C}{A}\right)_{\text{leaf}}^{\text{iv}} = 826 \frac{\text{J}}{\text{Km}^3}. \quad (16)$$

This value for the heat capacity can be verified by calculating the thickness d_{iv} of a water layer with the same heat capacity. This is found to be

$$d_{\text{iv}} = \frac{\left(\frac{C}{A}\right)_{\text{leaf}}^{\text{iv}}}{c_p^{\text{water}}} = 197.8 \mu\text{m}. \quad (17)$$

Here c_p^{water} is the specific heat capacity of water ($c_p^{\text{water}} = 4165 \cdot 10^6 \text{ JKm}^{-3}$). This computed water layer thickness is in good agreement with microscopy data of the same species.

3.3.2. Periodic flux method

The second method of determining the heat capacity of leaves by active thermography represents the periodic flux technique. This method is similar to the *Lock-In* technique employed in non-destructive test engineering.^{9, 10} Here the incident heat flux $j_{\text{act}}(t)$ is varied periodically. Under these conditions the differential equation (9) can be solved in the

frequency domain. It is known from the *Fourier transform* theorem that a time derivative of a function in the time domain $f(t)$ is equivalent to the complex multiplication of the Fourier transform $\hat{f}(k)$ with $i\omega$ in the frequency domain, or

$$\frac{\partial}{\partial t} f(t) \longleftrightarrow i\omega \hat{f}(k). \quad (18)$$

Applying this theorem to Equation (9) leads to

$$i\omega \hat{T}(\omega) = -\frac{1}{\tau} \hat{T}(\omega) - \hat{b} - \hat{z}(\omega). \quad (19)$$

Under most experimental conditions it is safe to assume that latent heat flux j_{lat} as well as the incident radiation flux j_{in} and air temperature T_{air} remain constant during the measurement. Equation (19) can then be solved for \hat{T}_{leaf} , yielding

$$\hat{T}_{\text{leaf}}(\omega) = \frac{\hat{z}(\omega)}{\frac{1}{\tau} + i\omega}. \quad (20)$$

The phase φ and amplitude of this complex quantity is given by

$$\varphi(\omega) = -\arctan(\omega\tau) \quad \text{and} \quad |\hat{T}_{\text{leaf}}(\omega)| = \sqrt{\frac{|\hat{z}(\omega)|^2}{\frac{1}{\tau^2} + \omega^2}}. \quad (21)$$

It becomes apparent from this equation, that τ can be derived both from the phase and the amplitude. Since the phase $\varphi(\omega)$ is independent of $\hat{z}(\omega)$ it is preferable to employ it for computing τ . For periods ω of the actively controlled heat flux $j_{\text{act}}(t)$ that are much longer than the time constant of the leaf τ , that is $\omega \ll 1/\tau$, the phase can be approximated by

$$\varphi(\omega) \approx -\omega\tau. \quad (22)$$

The phase can thus be computed at every pixel of the image sequence. In the same fashion as in the constant flux method in Section 3.3.1, the heat capacity of the leaf $(C/A)_{\text{leaf}}$ can be computed following Equation (15).

Experiments were conducted with different frequencies of $j_{\text{act}}(t)$ ranging from $3.9 \cdot 10^{-3}$ Hz to $3.13 \cdot 10^{-2}$ Hz, corresponding to period lengths of 256 s to 32 s respectively. The amplitude and phase can be represented in a single color image. The phase is color coded and the amplitude is indicated by the brightness. The parameter images at the different frequencies are shown in Figure 6. It can be seen that the phase shift of the veins is larger than the shift of the interveinal fields. From Equations (15) and (22) this indicates the greater heat capacity of the veins.

3.3.3. Water flow velocity

Another technique of active thermography can be used for estimating the water velocity in plant leaves. The set-up differs from that of previously described active techniques in that not the whole leaf is heated by an energy source. Instead only a part of the leaf is heated by a laser or heating wire as can be seen in Figure 7. The movement of the heated water parcel is then visualized by the infrared camera. Apart from moving the water parcel will equilibrate with its surroundings. From a newly developed image processing algorithm both the movement and the temperature decline can be accurately estimated.¹¹

The temperature decline is due to diffusion, which can be thought of to be isotropic as a first approximation. The differential equation of such a process can be written as

$$\frac{d}{dt} g = \frac{\partial}{\partial t} g + u \frac{\partial}{\partial x} g + v \frac{\partial}{\partial y} g = D \Delta g, \quad (23)$$

where $\vec{f} = (u, v)^T$ is the optical flow, x and y are the axes of the two dimensional image frame of reference, D is the constant of diffusion and $\Delta g = (\partial^2 g / \partial x^2 + \partial^2 g / \partial y^2)$ is the Laplace operator. Here the computations are conducted in

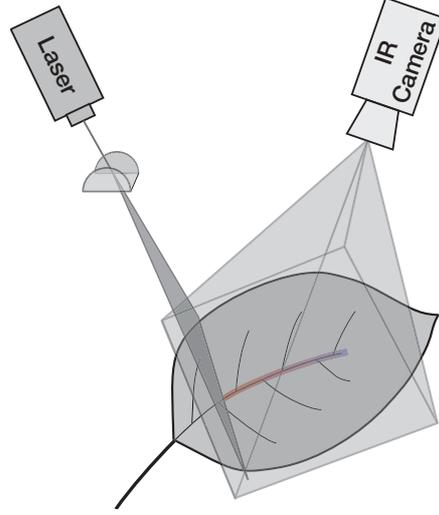


Figure 7: Sketch of the experimental set-up of the active thermography technique for estimating the water flow velocity.

image intensities g which can be mapped to absolute temperature T from radiometric calibration. This equation is under-determined and thus does not possess a unique solution. By assuming local consistency of the parameters D , u and v this equation can be extended to this local neighborhood consisting of n pixels, resulting in the following set of equations:

$$\begin{pmatrix} -\Delta g_1 & g_{x,1} & g_{y,1} & g_{t,1} \\ -\Delta g_2 & g_{x,2} & g_{y,2} & g_{t,2} \\ \vdots & \vdots & \vdots & \vdots \\ -\Delta g_n & g_{x,n} & g_{y,n} & g_{t,n} \end{pmatrix} \cdot \begin{pmatrix} D \\ u \\ v \\ 1 \end{pmatrix} = \vec{D} \cdot \vec{p} = 0, \quad \text{with } \vec{D} \in \mathbb{R}^{n \times 4}, \vec{p} \in \mathbb{R}^4 \quad (24)$$

where $g_{x,1}$ is the partial derivative of the i th pixel grey value with respect to x . Δg_1 is the Laplace operator acting on the i th pixel grey value.

The set of equations (24) can be solved in a total least squares sense,¹² yielding the sought after parameters D , u and v . The real world velocities can then be computed from the optical flow \vec{f} from a geometric calibration of the infrared camera.

The results of a typical measurement are shown in Figure 8. The water velocity and the constant of diffusion is computed highly spatially and temporally resolved, limited only by the resolution and the frame rate of the infrared camera.

The estimated water velocity provides some indication of the transpiration rate. Writing Equation (23) not as image intensities but as absolute temperature results in

$$j_{\text{net}} = \left(\frac{C}{A} \right)_{\text{leaf}} \frac{d}{dt} T_{\text{leaf}} = D \Delta T_{\text{leaf}}. \quad (25)$$

From estimating the temperature change $d/dt T_{\text{leaf}}$ the net heat flux j_{net} can be estimated from the knowledge of (C/A) which can be estimated from the techniques presented in Sections 3.3.2 and 3.3.1.

4. CONCLUSION

On the basis of thermal image sequence analysis three remote sensing techniques have been developed for measuring transpiration rate, water content and water flow velocity, as well as net heat flux in plant leaves. All three techniques have in common a high spatial and temporal resolution, which is only limited by the resolution and frame rate of the infrared camera used.

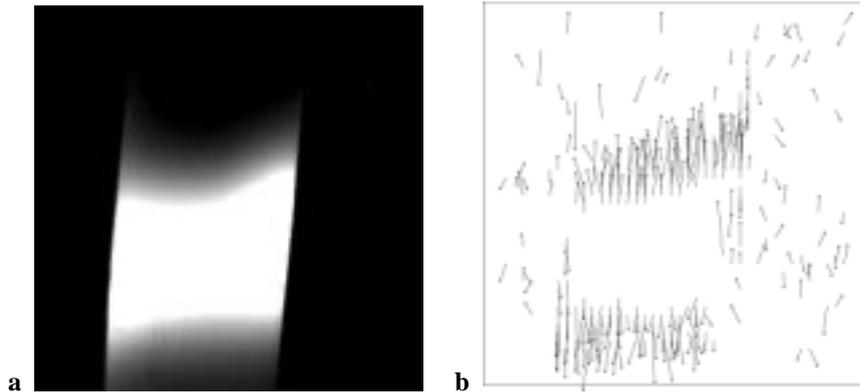


Figure 8: **a** An image of the thermographic sequence of a moving heat pulse. **b** the velocity field.

It has been shown that high spatial resolutions are indispensable if a profound knowledge concerning transpiration rates and parameters concerning them are to be gained. It is known that the stomata are not opened or closed homogeneously, an effect known as stomatal patchiness. This and other effects call for highly resolved measurement techniques, both spatial and temporal. Only through the application of modern thermographic techniques are highly accurate unobtrusive measurements feasible. This will improve our understanding of dynamic processes in water relations and in the development of plants.

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