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Transpiration Efficiency and Apparent Cuticular Transpiration in Some C_3 and C_4 Plants

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Abstract. Amphistomatous C_3 (*Nicotiana tabacum* L., *Datura stramonium* L.) and C_4 (*Sorghum saccharatum* Pers. and *Zea mays* L.) species were examined to find how (if at all) their inherent differences in water-use economy are reflected in apparent cuticular transpiration or *vice versa.* Transpiration efficiency (TE) was calculated from steady state photosynthesis (A) and transpiration (E) rates estimated for the upper side of the leaf after light induction of stomata opening. Apparent cuticular transpiration (E'_c) was measured as the part of transpiration which was not eliminated by convective counteraction of the air stream passing across the amphistomatous leaf: total pressure difference (ΔP) across the leaf was increased and the minimal value of $E_{\Delta P>0}$ was taken as the apparent cuticular transpiration rate (E'_c). E'_c was treated relative to E at ΔP equal to zero ($E_{\Delta P=0}$), E'_{cr}. Measurements were carried out under two leaf-air vapour pressure differences (VPD).

E_r (*i.e.* $E_{\Delta P>0}/E_{\Delta P=0}$) versus ΔP patterns differed qualitatively between the investigated C_3 and C_4 plants. TE increased and E'_{cr} decreased from tobacco, stramony, maize to sorghum for both VPD of air. E'_{cr} and TE were approximately linearly related, the slope being dependent on VPD. The increase in VPD resulted in larger E and slightly smaller epidermal conductance (g) at ΔP equal to zero. Both E'_c and E'_{cr} decreased markedly at the same time especially, for species with high TE. The results were considered as an indirect confirmation that E'_c values estimated by the technique used reflect species-specific differences in external peristomatal and cuticular vapour loss, at least in a relative sense.

Stomatal guard cells represent terminal points for the transpiration stream of liquid water. Vapour escaping quard cell walls can follow one of the following pathways: (i) it may evaporate from the inner walls bordering the substomatal cavities, pass through the stomatal pores, then the boundary layer and escape into the ambient atmosphere; (ii) it may evaporate from the outer periclinal walls directly into the boundary layer without passing through the stomatal pores or (iii) it may escape via some intermediate places on the ventral walls bordering the central apertures. The respective vapour fluxes going through the pathways (i) and (ii) were termed internal and external parts of peristomatal transpiration (Maier-Maercker 1983). Up to now, we do not know exactly the distribution patterns of evaporating sites in the vicinity of the stomatal pore

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and, even more, the quantitative proportions of the water vapour fluxes (i) – (iii) (*e.g.* Appleby and Davies 1983, Sheriff 1984).

We proposed a technique to separate the cuticular vapour flux, including the external peristomatal flux, from the other vapour fluxes through the stomatal pore (Šantrůček and Slavík 1990a). The magnitude of what we termed apparent cuticular transpiration (E'_c), and what we believed to be largely the vapour flux passing not the stomatal pores, was shown to sharply increase during the initial phase of stomatal opening for maize. This allowed us to propose that the external peristomatal flux (ii) accounts for a great deal of the measured apparent cuticular transpiration. Once the stomatal opening had reached a certain size, E'_c magnitude seemed to be insensitive to further increases. Therefore, under such opening conditions, E'_c should represent a loss of vapour, obeying no further stomatal control. This should produce an increase in water cost relative to benefits in CO_2 fixation from the view of transpiration efficiency. On the contrary, transpiration efficiency should be codetermined by apparent cuticular transpiration.

The experiments below were designed to test the expected relationship.

MATERIAL AND METHODS

We used the following amphistomatous species of C_3 and C_4 plants which were expected to be different in their TE: tobacco (*Nicotiana tabacum* L.) and stramony (*Datura stramonium* L.) (C_3), maize (*Zea mays* L.) and sorghum (*Sorghum saccharatum* Pers.) (C_4). Plants were grown in garden soil in plastic pots placed in a greenhouse for five to eight weeks (from February to May), with daylight extended, when necessary, to 14 hours with fluorescent tubes. Air temperature and relative humidity were 25 ± 3 °C and 60 ± 10 %, respectively.

The middle parts of the mature leaves attached to the plants were investigated. A leaf surface (of area 6 cm^2 involving the midrib) was exposed on both sides in windows made in plastic transparent sheets. The exposed area was made air-tight on the edges of the windows and enclosed in a double-sided leaf chamber (LI-COR 6200 0.25 dm^3 chamber adapted as described by Šantrůček and Slavík 1990b). The abaxial (lower) leaf side was illuminated with a photo lamp (incident photosynthetic photon flux rate about 600 μmol m^{-2} s^{-1}). Water vapour, CO_2 exchange and temperature of the adaxial (upper) leaf side were measured under natural CO_2 concentration in the open gasometric system described by Šantrůček and Slavík (1990b).

Steady state photosynthesis and transpiration rates were estimated by increasing the air pressure difference (ΔP) between the two sides of the chamber in steps. The pressure difference and resulting convective flow of air across the leaf was used to minimize vapour diffusion out of the stomatal pore. Transpiration efficiency was calculated as the ratio of photosynthesis to transpiration rates measured prior to the application of the pressure difference. The minimal value of transpiration rate obtained during the stepwise increase of the total pressure difference ΔP was taken as the apparent cuticular transpiration rate. Measurements were made repeatedly with the same leaf under two various leaf-air vapour pressure differences (VPD). Five to seven replicates were made with different plants for each plant species.

RESULTS

Transpiration efficiency increased from tobacco to stramony, maize and sorghum for both leaf-air vapour pressure differences (see Table 1). A decrease in VPD led to a significant increase in TE for sorghum ($P = 0.001$) and maize ($P = 0.061$) but not for tobacco and stramony.

TABLE 1

Transpiration efficiency, TE, computed as the ratio of photosynthesis, A, to transpiration, E, rates measured under zero pressure difference across the leaf $(A/E)_{\Delta P=0}$ (means of six replicates). Means of apparent cuticular transpiration rates, E'_c, and of their values relative to whole epidermal transpiration, E'_{cr}.

	TE		E		E'_c		E'_{cr}	
	[mmol CO_2 . mol^{-1} H_2O]		[mmol . m^{-2} . s^{-1}]				[%]	
VPD [kPa]	2.7	2.0	2.7	2.0	2.7	2.0	2.7	2.0
tobacco	1.68	1.69	1.07	0.99	0.74	0.78	76.7	83.0
stramony	2.27	2.34	1.17	0.99	0.7	0.69	57.7	71.3
maize	3.53	4.53	1.01	0.87	0.32	0.43	29.2	53.8
sorghum	4.33	7.4	0.81	0.53	0.05	0.2	10.9	35.8

As in a previous report for maize (Šantrůček and Slavík 1990b), application of the pressure drop across the leaf blade of maize or sorghum, and the resulting convective flow of air, caused a fall in vapour loss from the upper side of the leaf (Fig. 1). On the other hand, transpiration decreased only slightly in tobacco and stramony and then rose as the total pressure difference (ΔP) increased (Fig. 1). The transpiration rate at high ΔP reached up to 1.5 times that of the initial rate at zero pressure difference for several tobacco plants. For all species, the smallest value of transpiration found during the pressure application was taken as the apparent cuticular transpiration E'_c. Its absolute values as well as its values normalized to steady state vapour loss at $\Delta P = 0$ $(E'_c/E_{\Delta P=0})$, E'_{cr}, are given in Table 1.

Transpiration efficiencies, TE = $(A/E)_{\Delta P=0}$, for individual measurements were plotted against the respective E'_{cr} values (Fig. 2a,b). The species with higher TE exhibited a smaller proportion of apparent cuticular transpiration for both VPD. Only greater variance in E'_{cr} by maize leaves exposed to smaller VPD disturbed this species-specific arrangement (Fig. 2b). Regression lines fitted to the experimental data differed in their slopes according to VPD (see equations in Fig. 2a,b).

DISCUSSION

Relevance of the apparent and actual rates of cuticular transpiration

Transpiration versus pressure difference patterns for maize and sorghum followed principally those predicted by the earlier model (Šantrůček and Slavík 1990a). On the contrary, tobacco and stramony responses were quite different:

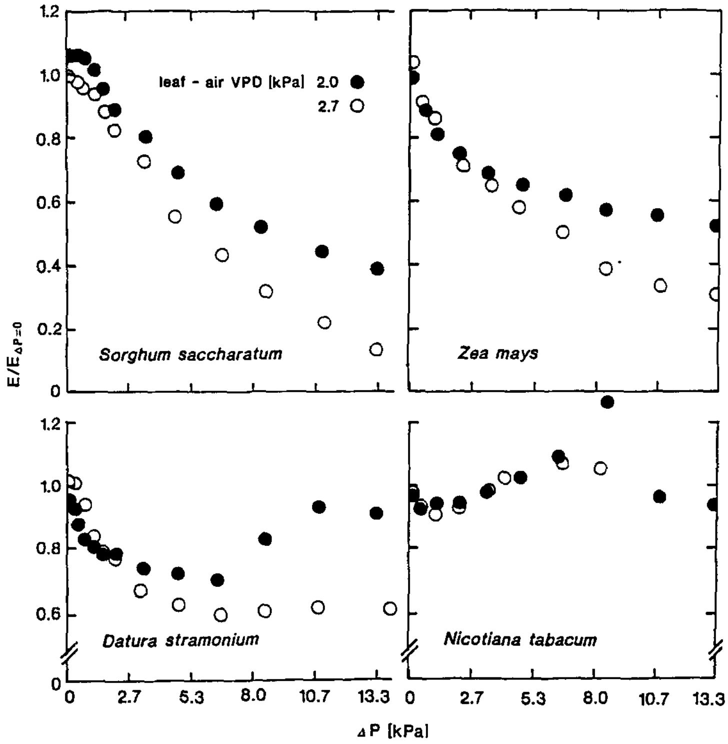

Fig. 1. Water loss, E, from the leaf surface exposed to various levels of the total pressure difference, ΔP, across the leaf blade. E values are expressed relative to the initial values at zero pressure difference, $E_{\Delta P=0}$. Measurements were made under two leaf-air vapour pressure differences, VPD, at incident photon flux rate (400–700 nm) 600 $\mu mol.m^{-2}.s^{-1}$. Means of five to seven replicates with different plants are presented.

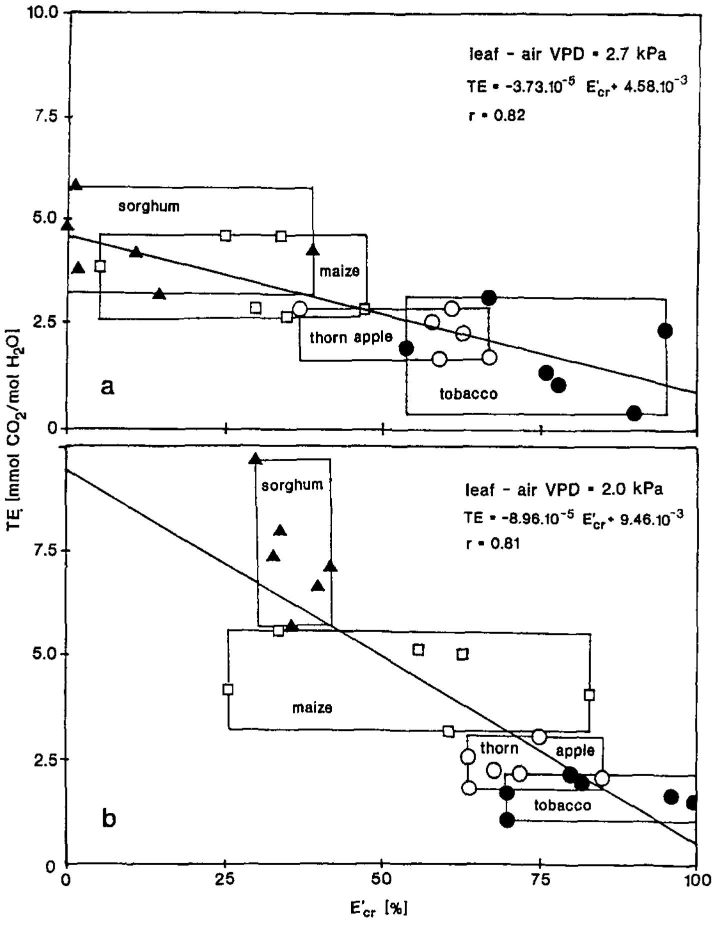

Fig. 2. Transpiration efficiency, TE, of four plant species versus relative apparent cuticular transpiration, E'_{cr}. E'_{cr} values were estimated as the minimal values of vapour loss from $E/E_{\Delta P=0}(\Delta P)$ relationships averaged in Fig. 1. Points resulted from measurements on individual plants under a leaf-air vapour pressure difference of 2.7 (Fig. 2a) and 2.0 kPa (Fig. 2b). Regression lines, equations and correlation coefficients, r, are given. Distribution patterns for individual species are outlined by rectangular figures for clarity.

transpiration decreased only slightly below the initial hundred percent and then rose with increasing pressure difference. In addition, the residual (minimal nonsuppressible) transpiration flux, which we interpreted as the apparent cuticular transpiration, was unexpectedly high, especially for tobacco and stramony. These findings raise doubts about the validity of the estimated E'_c values, at least in an absolute sense.

Two conditions must be met to get satisfactory agreement between the estimated (apparent) and the actual cuticular transpiration rates by the applied technique. First, vapour diffusion through the central aperture must be diminished under a sensitivity level of measuring device. Second, experimental treatment (overpressure) must affect only the above mentioned diffusive flux, not the vapour loss from the outer leaf surfaces.

The first condition was shown by the model to be met for sufficiently high viscous flow of air through the leaf and rather small stomata opening (Šantrůček and Slavík 1990a,b). Imperfections in the mechanistic model (*e.g.* a non-homogeneous air speed profile over the cross-sectional area of the stomatal aperture, or changes in diffusivity of the vapour in narrow pores) could quantitatively affect its predictions. The qualitative failure of the model for particular plant species only, as found here, can hardly be explained by general model imperfection. The concomitant increase in diffusive vapour loss from- and viscous flow of air through the pores could be explained by nonuniform stomata opening (stomata patchiness) induced probably by water stress caused by the viscous flow of air. The C_3 and C_4 – specific degree of non-uniformity of stomatal resistance was suggested by Cowan and Troughton (1971) as the means how the respective plants can optimize the relation of transpiration to CO_2 assimilation. Changes in their parameter α by tobacco and stramony leaves during our experimental treatment could reverse the uniform closing of stomata to nonuniform stomata response. It could account for the increase in the $E/E_{\Delta P=0}(\Delta P)$ course (see Fig. 1c,d) but it fails in an explanation of monotonous $A/A_{\Delta P=0}(\Delta P)$ fall by these plants (data not shown). Abscisic acid and water stress induced heterogeneity in the stomata opening over the leaf surface, as well as species-specific responses, were reported recently by Terashima *et al.* (1988), Downton *et al.* (1988) and Beyschlag and Pfanz (1990).

It is easy to check experimentally that the second condition holds under stomata closure. This was done for maize previously and for the other plants with similar results here (data not shown). However, to check it under stomata opening is difficult. Species-specific differences in stomata morphology could decide whether and to what extent turbulent streaming occurs in close vicinity to stomata for a given air convection across the pore. It could perhaps induce an extra vapour loss from exposed structures. This is the other possibility which could account for the observed increase in vapour loss by tobacco and stramony under high ΔP. Even if this artifact situation is physiologically implausible, the huge potential capacity of external epidermis surfaces to water vapour loss would be notable.

Apparent cuticular transpiration and transpiration efficiency

We proposed that the estimated apparent cuticular transpiration reflects the vapour loss which is not subject (partially, at least) to stomatal control. A verification test suggested negative correlation between relative apparent cuticular transpiration and transpiration efficiency. However, one important aspect must be taken into account: variability in TE must be induced by a factor, which *a priori* does not exclude the possibility of action of apparent cuticular transpiration and which does not confuse this action. *E.g.* TE can be greatly influenced by the leaf-air vapour pressure difference. But this factor is insufficient, because its primary mechanism of action on TE minimizes the possible effect of apparent cuticular transpiration. Besides, apparent cuticular transpiration itself was shown to be influenced by VPD in the opposite direction to stomatal transpiration (see Fig. 1). On the other hand, evolutionary based species-specific differences in water-use efficiency may at least be partially caused by inherent variability in cuticular transpiration (*e.g.* Plhák 1984, Richards *et al.* 1986). This is the reason why only species-specific differences in TE are correlated with E'_c in this report. Of course, the correlation may be expected to be different under various VPD.

The relative proportion of the apparent cuticular transpiration increased in more humid air according to the position of the given plant species on the TE(E'_{cr}) line. Species-specific humidity response occurred approximately in such a way that only the slope of the line has changed, not the intercept value of the minimal TE. Consequently, both the C_4 plants and especially sorghum become much more transpiration efficient under the increase in air humidity than do the C_3 plants.

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BOOK REVIEW

Studies of the Magnitude and Nature of Pesticide Residues in Stored Products Using Radiotracer Techniques. International Atomic Energy Agency, Vienna 1990, 143 pp, Austria Sch. 460,–

The well-publicized effects of pest-control chemicals on human health and environmental quality have increased public concern in the last three decades. The objective of this publication, based on contributions of a FAO/IAEA Coordination Meeting, held in Turkey, 1988, was to review analytical procedures and radiotracer techniques applied to investigate the presence of unwanted pesticide residues in stored food. As demonstrated, radiotracer techniques have shown to be superior in detecting, identifying and qualifying pesticide residues. Most of the research reported in the 16 contributions deal with the degradation of organophosphorus insecticides, malathion, pirimiphosmethyl and chlorpyriformethyl, and with the presence of fenvalerate, ethylene dibromide, and methyl bromide in a variety of stored commodities, *e.g.* wheat, barley, maize, faba beans etc. The publications include 2 model protocols for the determination of quantities of chemically bound and volatile residues that remain in the treated material. Of interest are Tables presenting data on pesticide residues remaining in agricultural products after various periods of storage.

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