

Is transpiration efficiency a viable plant trait in breeding for crop improvement?

Thomas R. Sinclair

Crop Science Department, North Carolina State University, Raleigh, NC 27695-7620, USA.
Email: trsincla@ncsu.edu

Abstract. Increased transpiration efficiency – commonly the ratio of mass accumulation to transpiration – is often suggested as a critical opportunity for genetic improvement for increased crop yields in water-limited environments. However, close inspection of transpiration efficiency (*TE*) shows that it is a complex term that is explicitly dependent upon both physiological and environmental variables. Physiological variables include leaf photosynthetic capacity, biochemical composition of the plant productions and possible hydraulic limitation on water flow in the plant. Environmental variables include atmospheric CO₂ concentration and atmospheric vapour pressure deficit. To complicate the resolution of transpiration efficiency, a weighted integration over the daily cycle and over the dates of interest needs to be resolved. Consequently, it is concluded that transpiration efficiency is not a variable easily resolved for use in many breeding programs. Instead, component traits contributing to *TE* need to be studied to increase the effective use of available water through the growing season to ultimately maximise growth and yield of the crop.

Additional keywords: ¹³C discrimination, leaf photosynthetic capacity, plant biochemical composition, vapour pressure deficit.

Received 6 September 2011, accepted 12 March 2012, published online 24 April 2012

Introduction

Developing crop cultivars for water-limited conditions is often voiced as a major objective for future crop improvement in both the popular press and in scientific reviews. ‘More crop per drop’ is a popular phrase heralding the possibilities for increasing crop yields when water is limiting. From a historical perspective, the empirical evidence is that crop yields have increased without any change in the precipitation received by crops, i.e. more yield per unit water input. It is appealing to conclude that focussed effort on the ratio of yield to water use will continue the trend for increasing this ratio.

Assessment of possibilities for future increases in ‘water use efficiency’ is critical in understanding possibilities for breeding for crop improvement. When water use efficiency is defined in terms of yield per unit of precipitation, then the many genetic and environmental factors contributing to yield formation confound the understanding of plant water use. To narrow the focus to plant processes, Passioura (1996) suggested the conceptual framework to define crop yield (*Y*) as a function of crop harvest index (HI, ratio of grain mass to total plant mass), the amount of transpired water (*T*) and crop transpiration efficiency (*TE*) defined as the ratio of accumulated plant mass (*M*) to water transpired.

$$Y = HI \times T \times TE. \quad (1)$$

Equation 1 has become a critical conceptual reference and is frequently used to highlight the importance of *TE*. A common

conclusion based on Eqn 1 is that identifying genotypes with increased *TE* would result directly in yield increase. As discussed in this paper, the difficulty is that *TE* is not a simple variable readily amenable to genetic selection as is often assumed. Indeed, *TE* is a very complex term dependent both on physiological traits of the plant and on the environment.

Mechanistic expression of transpiration efficiency

A more complete understanding of the dependence of plant growth on water use than offered by Eqn 1 is to use the mechanistic expression for daily crop growth (*G_d*) obtained by rearranging equation 4 given by Sinclair *et al.* (1984):

$$G_d = T_d k_d / (e_a^* - e)_d, \quad (2)$$

where *T_d* is daily transpiration rate, *k_d* is a mechanistic coefficient discussed below and (*e_a^{*} – e*)_d is a daily atmospheric vapour pressure deficit weighted for the daily cycle of transpiration rate. Equation 2 can be integrated over the growing season and multiplied by HI to get yield so that the resultant expression has a form equivalent to Eqn 1. In this case, *TE* is replaced by an integral of the more explicitly defined variables *k_d*/(*e_a^{*} – e*)_d.

An explicit definition of *TE* as *k_d*/(*e_a^{*} – e*)_d is also deceptively simple. Each of these terms is complicated by the fact that they need to be integrated over the day and then even more challenging integrated over the growing season (Tanner and Sinclair 1983). The *k_d* term was defined by Tanner and Sinclair

(1983) and is predominantly a physiological term, although it has environmental components. The variable $(e^*_a - e)_d$ is the difference between saturated vapour pressure (e^*_a) and the actual vapour pressure (e) of the air weighted for transpiration rate through the daily cycle. Although $(e^*_a - e)_d$ is often calculated as a simple arithmetic mean over a day, it must be weighted for the variation in transpiration through the daily cycle. As a result $(e^*_a - e)_d$ is highly dependent on the environment and the physiological behaviour of the plant.

Consequently, the definition of TE as $k_d/(e^*_a - e)_d$ illustrates that TE is not a simple term dependent on single readily identifiable traits of plants amenable to genetic improvement. Without knowledge about both the environment and the specific physiological traits of the plant, it is difficult to resolve TE for a cultivar. Consequently, the short answer to the question of the title of this paper is, 'No, the dependence of TE on both environmental and physiological variables may make it too complex as a readily accessible characteristic for practical breeding efforts'.

Even though the conclusion is discouraging in regards to using general approaches to increase TE , more detailed focus on the component plant traits that directly impact TE may allow targeted phenotyping in contributing to development of desirable genotypes for water-limited environment. In particular, the above analysis shows that the component traits determining $TE - k_d$ and $(e^*_a - e)_d$ are traits at a lower biological scale that might prove useful in resolving traits for breeding. Each of the component terms defining TE is examined to understand their role in crop water use and to assess the possibility of applying them in breeding for crop improvement.

Increase k_d ?

The variable k_d as defined by Tanner and Sinclair (1983) actually represents a collection of variables:

$$k_d = 1.6abcP_aL_D/L_T, \quad (3)$$

where a is the molecular weight ratio between (CH_2O) and CO_2 , i.e. 0.68, b is the conversion ratio from hexose to plant biomass, c reflects the CO_2 partial pressure diffusion gradient between the interior of the leaf and the external atmosphere, $(1 - C_i/C_a)$, P_a is the partial pressure of atmospheric CO_2 and L_D/L_T is the ratio of leaf area in direct beam radiation to effective 'transpiring' leaf area ($\sim 1.4/2.2$).

The nominal values of k_d based on Eqn 3 are ~ 5 Pa for high-energy C_3 crops such as soybean (*Glycine max* (L.) Merr.), 6 Pa for low-energy C_3 crops such as wheat (*Triticum aestivum* L.) and 9–10 Pa for C_4 crops including maize (*Zea mays* L.) and sorghum (*Sorghum bicolor* L.). The two terms in the definition of k_d that account for much of the difference among species are b and c . The conversion of hexose to plant mass, i.e. b , depends directly on the nature of the plant mass being synthesised. Plant mass that is composed mainly of carbohydrate, e.g. starch and cellulose, has a large b value compared with plant mass that is high in protein and lipid content. Therefore, large differences in b and consequently k_d , exist among species and could exist among genotypes within a species. The difficulty is the practical utility in increasing b to increase k_d . For vegetative tissue, the main source of variation in b is protein concentration.

Decreasing protein concentration of vegetative tissue would increase b but this would likely result in decreased photosynthetic capacity of leaves and less stored nitrogen for eventual transfer to growing grains. Increasing b for grain by decreasing seed protein and lipid concentration is likely possible by genetic selection. However, the commercial value of grain is linked in large part to their composition. Although genotypes of wheat or soybean could be selected for decreased concentrations of protein and increased b , the marketplace has economic penalties for low protein concentration grain. Hence, for most crops increasing b does not seem to be a realistic target for genetic alteration.

In comparing species, the major difference in c is that $(1 - C_i/C_a)$ is high in species with the C_4 assimilation pathway as compared with those with only the C_3 pathway. The maximum value of c for those species dependent only on the C_3 photosynthetic pathway is ~ 0.3 . Although those species with the high CO_2 -affinity C_4 pathway have maximal values of ~ 0.7 . Genetic variation within a species for c , i.e. $(1 - C_i/C_a)$ and the impact on k_d has been studied in only a few cases. Xin *et al.* (2009) measured C_i/C_a of 25 sorghum lines and found transpiration efficiency normalised for $(e^*_a - e)$, i.e. k_d and found a very significant ($r^2 = 0.904$), negative linear relationship between the two variables, as predicted by Eqn 3. Similarly, the data reported by Monneveux *et al.* (2006) for five cultivars of durum wheat gave the same highly significant ($r^2 = 0.99$) negative linear relationship between a measure of k_d and C_i/C_a for data obtained from both well-watered and rainfed treatments.

Clearly, if commercial cultivars are not exhibiting near maximal photosynthetic capacity, there is a major opportunity to increase k_d . Wright *et al.* (1988) concluded from their study of eight genotypes of peanut (*Arachis hypogaea* L.) that differences among genotypes in the ratio of accumulated plant mass to transpired water were a result of differences in photosynthetic capacity. Unfortunately, no measurements were presented that allowed direct comparison of c values. Gilbert *et al.* (2011) compared the photosynthetic capacity of soybean and identified genotypic differences that could have a small but important impact on k_d .

A few studies have attempted measurements allowing direct determination of k_d . Zur and Jones (1984) used a canopy enclosure to measure gas exchange in the field of one soybean cultivar (Williams) under field conditions. The estimate of k_d was independent of the watering regime and over 5 days k_d was stable at 4 Pa. In rice (*Oryza sativa* L.), Haefele *et al.* (2009) calculated from regression analysis that over a diversity of experiments that k_d ranged from 1.3 to 5.0 Pa. Hammer *et al.* (1997) determined k_d for 49 sorghum genotypes representing a range of genetic backgrounds. Although several genotypes had very low k_d , the average for all genotypes was 8.5 kPa and the highest k_d obtained for any genotype was 9.8 Pa. Significantly, a very close correspondence ($r = 0.98$) was found between accumulated plant dry mass and transpiration. Therefore, rice and sorghum genotypes were identified with inferior values for k_d , but none exceeded the range of the anticipated upper values.

Kemanian *et al.* (2005) provided an extensive assessment of the value of k_d for barley (*Hordeum vulgare* L.) and wheat. Based on their field experiments with barley, they estimated k_d

to be 6.2 Pa in one year and 6.9 Pa in the second year. They also presented values of k_d calculated for eleven published studies with barley and wheat. The values of k_d ranged from 2.8 to 7.0 Pa with many of the estimates in the range from 4.2 to 6.0 Pa. In further analysis of the data, Kemanian *et al.* (2005) concluded that k_d decreased with increasing vapour pressure deficit. Although this is a possibility, their analysis overlooked the possibility that the estimate of vapour pressure deficit was potentially a dynamic variable and the evaluation of $(e_a^* - e)_d$ may need to be adjusted depending on the environment. This confounding situation is discussed later in the paper.

Overall, the experimental estimates of maximal values of k_d do not differ greatly from the original estimates by Tanner and Sinclair (1983). That is, no species or genotype has been identified that greatly exceeds the original estimate for k_d . Examination of the two critical variables defining k_d , i.e. b and c , does not offer much encouragement for substantial improvement in k_d predicted for a species. Considering the commercial restrictions on increasing b and the likely high values already present in c for current crop cultivars, in most cases it appears that k_d is not a likely target for major alteration to improve TE . It is anticipated that any crop that has been subjected to substantial breeding and selection has already been optimised to a large extent for b and c for commercial production. Of course, if b and c have not yet been maximised then there could be major reward in shifting these variables to higher values and increase k_d .

Decrease $(e_a^* - e)_d$?

In the original derivation by Tanner and Sinclair (1983), they emphasised that vapour pressure for a season had to be weighted for the changes in transpiration rate and that an 'average' vapour pressure deficit is likely not appropriate. Although they were applying this argument for estimating seasonal vapour pressure deficit, the same logic applies over the daily cycle. For each day, there may be a challenge in quantifying a weighted vapour pressure deficit through the daily cycle to obtain $(e_a^* - e)_d$. Tanner and Sinclair (1983) suggested the following relationship:

$$(e_a^* - e)_d = 0.75(e_{\max}^* - e_{\min}^*), \quad (4)$$

where e_{\max}^* is saturated vapour pressure at daily maximum temperature and e_{\min}^* saturated vapour pressure at daily minimum temperature. Abbate *et al.* (2004) examined the appropriateness of Eqn 4 for environments in Argentina. They found a slightly different coefficient of 0.72 was optimum for their conditions.

At first glance, $(e_a^* - e)_d$ appears to be a purely physical term and would not be subject to genetic manipulation. However, closer examination of $(e_a^* - e)_d$ indicates that genetic differences might actually have a large influence on this term. Since $(e_a^* - e)_d$ is to be weighted for transpiration rate, if plants regulate transpiration so that transpiration rate may be restricted under some conditions, then the effective value of $(e_a^* - e)_d$ is altered. There are at least two plant traits that result in restricted transpiration under high, midday vapour pressure deficits both resulting from restricted plant hydraulic conductance. When hydraulic conductance becomes limiting to water transport, the plant is unable to meet the transpiration demand at high vapour pressure deficit and this response results

in decreased stomata conductance to match transpiration rate to water flux in the plant. For those plants with limited hydraulic conductance, even under well-watered conditions the possibility exists for midday stomata closure under high vapour pressure deficit resulting in a lowered effective $(e_a^* - e)_d$. Another trait responding to limited hydraulic conductance is the expression of decreased stomata conductance with soil drying. The combination of limited water flux due to soil and plant conductance can result in a decrease in transpiration to match hydraulic flux. The expression of the sensitivity to soil drying would again be midday stomata closure and a decrease in effective $(e_a^* - e)_d$.

The existence of genetic variation in the plant hydraulic conductance limiting transpiration rate under high vapour pressure deficit conditions has become apparent in recent studies. Those genotypes expressing this trait under well-watered conditions have unrestricted transpiration rates at low $(e_a^* - e)$, but above a threshold $(e_a^* - e)$ exhibited little or no further increases in transpiration rate (Fig. 1). Genetic variation for this response has been reported for soybean (Fletcher *et al.* 2007; Sadok and Sinclair 2009), peanut (Devi *et al.* 2010), sorghum (Gholipour *et al.* 2010), pearl millet (*Pennisetum glaucum* (L.) R.Br.) (Kholova *et al.* 2010b), chickpea (*Cicer arietinum* L.) (Zaman-Allah *et al.* 2011) and maize. The threshold of $(e_a^* - e)$ for the initiation of the limitation on transpiration rate is commonly in the range from 1.6 to 2.4 kPa. A consequence of the restriction of transpiration rate at high $(e_a^* - e)$ during the daily cycle is that daily weighted $(e_a^* - e)_d$ is decreased. Hence, those genotypes expressing this limited-transpiration trait would have increased TE under well-watered conditions.

The basis for the expression of maximum transpiration rate has been shown to be linked to limited hydraulic conductance in soybean (e.g. Sinclair *et al.* 2008). Sadok and Sinclair (2010) found differences among soybean genotypes in response of their leaf transpiration rate when fed silver ions, which are potential inhibitors of aquaporins. Their suggestion was that those genotypes exhibiting insensitivity to silver likely had a different population of aquaporins than other genotypes that caused them to have a lower hydraulic conductance in the plant. Carpentieri-Pipolo *et al.* (2012) tracked the silver-sensitivity trait in soybean to four quantitative trait loci, indicating a possibility for readily breeding for the restricted maximum transpiration trait and hence, an increased TE .

A negative consideration in applying the maximum-transpiration trait is that partial decrease in stomatal conductance also results in elevated leaf temperature and a limitation on photosynthetic rate. The increase in leaf temperature as a result of the decreases in stomatal conductance is likely to be very modest. In an analysis of the possible influence of decreased stomatal conductance on leaf temperature as a result of elevated CO_2 concentration, Allen (1990) predicted leaf temperature increases of only 2°C or less in soybean as a result of a 41% decrease in stomatal conductance. Experimental studies under elevated CO_2 resulted in leaf temperature increases in soybean of no more than 0.7°C even though stomatal conductance decreased by up to 50% (Allen *et al.* 1998). These results do not indicate that temperature increase is a major problem as a result of partial

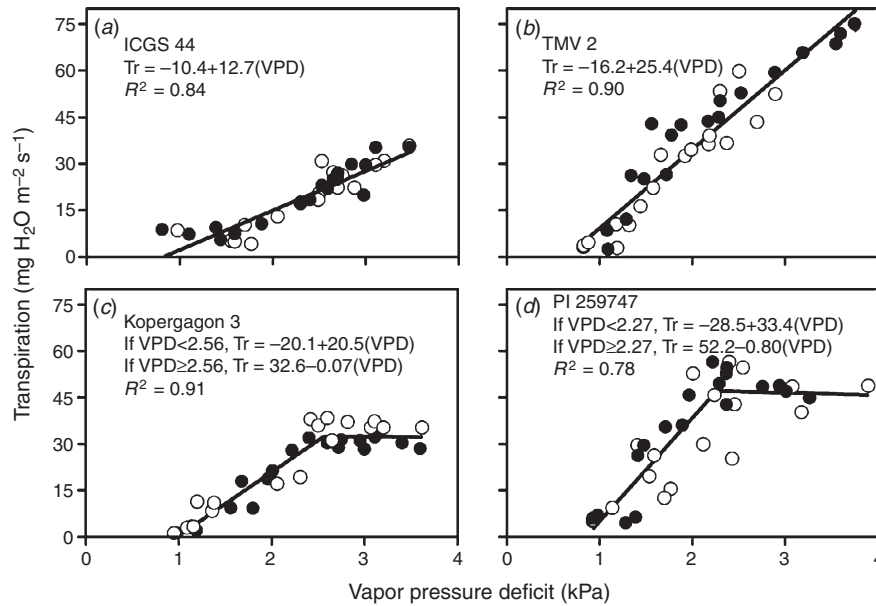


Fig. 1. Plant transpiration rate response of four genotypes of peanut to changes in atmospheric vapour pressure deficit. Genotypes ICGS 44 and TMV2 exhibited continually increasing transpiration rates with increasing vapour pressure deficits while genotypes Koperagon 3 and PI 259747 exhibited an essentially constant transpiration rate at high vapour pressure deficit. The two sets of symbols in each graph represent different sets of data, which were combined in the regression analysis (Devi *et al.* 2010).

decreases in stomatal conductance under high vapour pressure deficit conditions.

However, partial stomata closure will clearly result in decreased growth potential. This problem can be offset by the benefit of limited transpiration rate, especially early in the growing season, resulting in soil water conservation that can contribute to crop development and growth during late-season dry periods. Simulation studies with sorghum in Australia (Sinclair *et al.* 2005) and soybean in USA (Sinclair *et al.* 2010) have indicated that the dual benefit of decreased effective $(e_a^* - e)_d$ and conserved soil water could result in yield gains in 75–85% of the growing seasons for each of these two crops.

Another possibility for increasing midday stomata closure and hence, decreased $(e_a^* - e)_d$, is altered sensitivity of plants to soil drying. Assuming that the initial response to soil drying is a decrease in midday stomata conductance as water deficit develops in the plant, the value of $(e_a^* - e)_d$ would be decreased. Hence, those genotypes that are more sensitive to soil drying by initiating decreased stomatal conductance at higher soil water content have the possibility of exhibiting a lower $(e_a^* - e)_d$ and increased TE . Sensitivity of plants to soil drying can be expressed as a function of the fraction of transpirable soil water (FTSW) remaining in the soil (Fig. 2). Commonly, limitations on transpiration rate develop when FTSW decreases to roughly one third (Sadras and Milroy 1996). However, genetic variability has been observed for this trait in soybean (Hufstetler *et al.* 2007), pearl millet (Kholova *et al.* 2010a), chickpea (Zaman-Allah *et al.* 2011), sorghum (Gholipour *et al.* 2012) and maize. For example in peanut, Devi *et al.* (2009) found in a comparison of 19 genotypes that the FTSW threshold for the decrease in

transpiration rate with soil drying ranged from 0.22 to 0.71 (Fig. 2). Further, they found that TE when the plants were subjected to soil drying was higher for those genotypes with the higher FTSW thresholds.

Early stomatal closure with soil drying also has the major benefit of conserving soil water as the crop is entering drought. Decreases in stomata conductance at high soil water contents allow initiation of water conservation at an earlier stage in soil drying. As a result, the imposition of severe water deficit stress on the plant is delayed. Simulations models with maize (Sinclair and Muchow 2001) and soybean (Sinclair *et al.* 2010) have shown this to be a beneficial trait. The combined benefit of water conservation and increased TE because of early decreases in stomata conductance was simulated in soybean to result in a yield increase in more than 80% of the growing seasons in most regions of the USA.

Further, the early decrease in stomatal conductance with soil drying is the opposite response proposed for osmotic adjustment in leaves. If osmotic adjustment sustains gas exchange to lower soil water content, then the crop is at risk of severe drought damage before it again receives water. Indeed, the simulations studies by Sinclair and Muchow (2001) and Sinclair *et al.* (2010) have shown yield loss because of the proposed osmotic adjustment response.

Approaches to identify TE traits

Above, the specific traits leading to increased TE have been identified. A major challenge now is to identify genetic variation within a species for these traits that can then be exploited in breeding programs. Consequently, rather simple methods

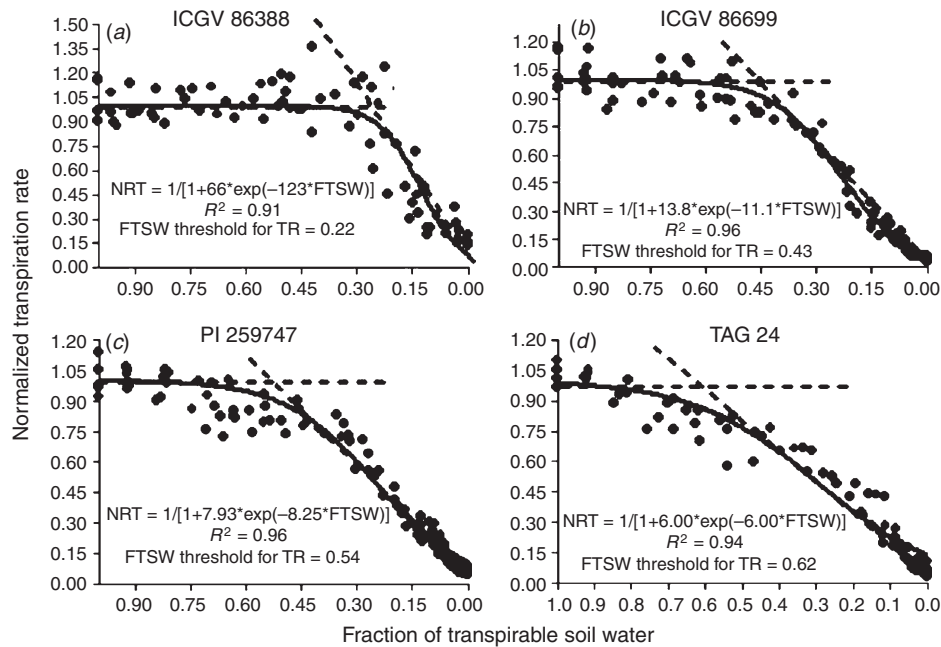


Fig. 2. Normalised plant transpiration rate response of four genotypes of peanut to changes in soil fraction of transpirable soil water (FTSW). The response is represented by an exponential function (solid line) and a two-segment linear regression (dashed line) (Devi *et al.* 2009). Among these four genotypes the threshold for the decreased in transpiration based on the linear regression ranged from 0.62 for TAG 24 to 0.22 for ICGV 86388.

for phenotyping for these specific are needed to allow screening of large numbers of genotypes. Here, extant techniques that have been applied in the search for genetic variation in *TE* are reviewed.

$\Delta^{13}C$

Farquhar *et al.* (1982) derived an expression for determining the isotope discrimination in photosynthesis of $^{13}CO_2$. The discrimination $\delta^{13}C$ reflected a linear dependence on the ratio of $^{13}C_i/^{13}C_a$. The empirical relationship found by Farquhar and Richards (1984) for wheat was,

$$\Delta^{13}C = 4.4 + 22.6^{13}C_i/^{13}C_a. \quad (5)$$

Hence, $\Delta^{13}C$ is intimately linked to the key variable *c* in the definition of the variable k_d influencing *TE*. Low values of $^{13}C_i/^{13}C_a$ resulting in low values of $\Delta^{13}C$ would result in high *c* and hence, high *TE*.

A positive correlation between $\Delta^{13}C$ and $^{13}C_i/^{13}C_a$ has been reported in comparing genotypes grown in greenhouses for peanut ($r^2=0.64$, Hubick *et al.* 1988) and wheat ($r^2=0.61$, Condon *et al.* 1990). However, such a relationship has not always been reported. In a comparison of eight cultivars of wheat grown in the field under irrigated conditions, Morgan *et al.* (1993) found a positive relationship ($r^2=0.69$) in one growing season and a negative relationship ($r^2=0.62$) in a second growing season. Monneveux *et al.* (2006) found no correlation between $\Delta^{13}C$ and C_i/C_a in a comparison of five durum wheat cultivars grown in the field under irrigated conditions in Montpellier, France, even though there was genetic variation in C_i/C_a . Similarly, in a comparison of four

hard red winter wheat (Arapahoe, Cheyenne, Karl 92 and Scout 66) grown in the field at Lincoln, Nebraska, Xue *et al.* (2002) failed to find a correlation between $\Delta^{13}C$ and measured in one year and $\Delta^{13}C$ in the second year.

Variability of $\Delta^{13}C$ under field conditions appears to place a limitation on the use of the carbon discrimination technique in some environments. For example, Hall *et al.* (1994) compared the relative ranking of $\Delta^{13}C$ obtained for cowpea (*Vigna unguiculata* L. Walp.) cultivars grown in four divergent locations. They found no correlation in genetic ranking of $\Delta^{13}C$ among locations. Condon *et al.* (1990) found no correlation for $\Delta^{13}C$ measured in the greenhouse and in the field for 16 wheat genotypes. It appears that weather variability in the field, especially in non-arid locations, results in variability in $^{13}C_i/^{13}C_a$ over time that introduces variability in $\Delta^{13}C$. Certainly such factors as water-deficit and vapour pressure deficit as discussed above can result in decreased stomatal conductance limiting CO_2 flux.

Despite the potential limitations of the $\Delta^{13}C$ technique under field conditions, it has been successfully applied in Australia to develop higher yielding wheat cultivars for dryland conditions. Rebetzke *et al.* (2002) described screening of plants in the greenhouse under well-watered conditions for low $\Delta^{13}C$. Field tests of superior $\Delta^{13}C$ lines from the breeding program and selection based on $\Delta^{13}C$ measurements and yield resulted in the selection of a genotype that had an average yield increase of ~11%, i.e. 0.11 t ha^{-1} at average dryland yields of 1 t ha^{-1} . At greater yield levels, the percentage advantage decreased linearly. The cultivar 'Drysdale' developed in this study has been released for production in dryland areas.

TE based on pot studies

A common approach is to grow plants in pots and track their water use and vegetative growth over a period of time. These studies are commonly conducted under controlled environments. One attempt to better represent field conditions is a system set up at ICRISAT, Hyderabad, India in which large pots (0.2 m diameter \times 1.2 m tall) are placed in trenches outdoors (Vadez *et al.* 2008). Polyethylene beads are put on the soil surface to greatly restrict water loss by water evaporation from the soil surface. A rain shelter is moved over the plants in the event of rain. The pots are weighed periodically to calculate transpiration rates over short time intervals. Plant harvests at the beginning and end of the interval of interest gives the increase in mass. Accumulated plant mass divided by total transpiration is defined as *TE*.

Certainly, these pot experiments offer direct measures of variation in *TE*. However, the challenge remains to apply these results in a breeding effort. As the preceding discussions have indicated, *TE* is a consequence of both several physiological traits and the environment. Hence, the *TE* measured in pot studies is not a genetic trait *per se*, so accurate extrapolation of pot results depends on several variables. The conditions for expression of high *TE* in one situation may result in low *TE* in another environment. To breed for increased *TE*, it is likely necessary to examine a specific trait and use variability of the specific trait in the breeding program. Selection of genotypes for high *TE* under well defined or measured environmental conditions can be employed as a very useful initial screen of a large number of genotypes. However, those selected lines need to be studied further to document the specific trait leading to increased *TE* under the test conditions.

Comparisons of pot *TE* among genotypes if the soil water is allowed to dry is even more challenging because of possible genetic differences in the threshold where stomatal conductance declines in response to drying soil. If no other potentially confounding factor is involved, *TE* on drying soil is likely to be greater for the genotype with a higher FTSW threshold for decrease in transpiration rate. For example, little variation in *TE* in peanut has been found when grown under well-watered conditions, but large genetic variation has been identified when plants were subjected to water-deficit treatments (Wright *et al.* 1994; Devi *et al.* 2009). Indeed, Devi *et al.* (2009) found, as hypothesised, that increased *TE* was associated with a high threshold for the decrease in transpiration with soil drying.

The duration of the time in which the plant is exposed to the drying soil is also important. If the pots are taken to terminal drought then presumably the genotypes having a high threshold will be delayed in reaching the end point. Assuming the environment for the genotypes with the longer drought period is unchanged, the additional days of low stomatal conductance will result in increased *TE*. If the experiment is not taken to terminal drought, then the results are very difficult to resolve because the severity of drought and its influence on *TE* differs among the genotypes.

Field benefit of increased *TE*

The above discussion indicates that options do, in fact, exist for increasing *TE* assuming the breeding is focussed on component

traits. The difficulty is that these options do not offer opportunities for dramatic increases in *TE* with the exception of improvements in species with particularly poor intrinsic photosynthetic capacity. The conclusion that there are limited chances to increase *TE* in a practical situation is consistent with historical evidence. DeWit (1958) analysed the relationship between growth and transpiration normalised for pan evaporation using data collected across North America and Europe beginning in the early 20th century. He found that within a species, regardless of genotype, soil, watering regime and fertility conditions there was a common *TE* among all the results.

Although the analysis presented here indicates that improvement in *TE* is possible, the impact for increasing crop yield is limited. Indeed, this analysis supports the original conclusion by DeWit (1958) that a close relation between growth and transpiration should be commonly expected. Although some improvements of *TE* can be made for some crops under certain conditions, the complexity of the traits contributing to *TE* does not make *TE* a strong candidate as a breeding objective for increasing crop yield under water-limited conditions.

Moreover, Blum (2009) made the argument that *TE* is only a concern under water-limited conditions and in that the real challenge is to increase the effective use of water for transpiration when water is limiting. In this view then, specific traits that enhance the effective use of water become critical. In actuality, the two water-conservation traits discussed above, i.e. increased sensitivity in stomata conductance to vapour pressure deficit and in soil drying, both contribute to increasing effective use of water. Although these traits enhance *TE*, their contributions to soil water conservation may be more important, allowing plants to remain physiologically active longer following the onset of soil water-deficit conditions. Overall, breeding efforts may be more rewarded by examining specific traits that are likely to allow crops to be effective in use of water than attempts to resolve increases in *TE*.

References

- Abbate PE, Dardanelli JL, Cantarero MG, Maturano M, Melchiori RJM, Suero EE (2004) Climatic and water availability effects on water-use efficiency in wheat. *Crop Science* **44**, 474–483.
- Allen LH Jr (1990) Plant responses to rising carbon dioxide and potential interactions with air pollutants. *Journal of Environmental Quality* **19**, 15–34. doi:10.2134/jeq1990.00472425001900010002x
- Allen LH Jr, Valle RR, Jones JW, Jones PH (1998) Soybean leaf water potential responses to carbon dioxide and drought. *Agronomy Journal* **90**, 375–383. doi:10.2134/agronj1998.00021962009000030010x
- Blum A (2009) Effective use of water (EUW) and not water-use efficiency (WUE) is the target of crop yield improvement under drought stress. *Field Crops Research* **112**, 119–123. doi:10.1016/j.fcr.2009.03.009
- Carpentieri-Pipolo V, Pipolo AE, Abdel-Haleem H, Boerma HR, Sinclair TR (2012) Identification of QTLs associated with limited leaf hydraulic conductance in soybean. *Euphytica*, in press.
- Condon AG, Farquhar GD, Richards RA (1990) Genotypic variation in carbon isotope discrimination and transpiration efficiency in wheat: leaf gas exchange and whole plant studies. *Australian Journal of Plant Physiology* **17**, 9–23. doi:10.1071/PP9900009

- Devi MJ, Sinclair TR, Vadez V, Krishnamurthy L (2009) Peanut genotypic variation in transpiration efficiency and decreased transpiration during progressive soil drying. *Field Crops Research* **114**, 280–285. doi:10.1016/j.fcr.2009.08.012
- Devi MJ, Sinclair TR, Vadez V (2010) Genotypic variation in peanut for transpiration response to vapor pressure deficit. *Crop Science* **50**, 191–196. doi:10.2135/cropsci2009.04.0220
- DeWit CT (1958) 'Transpiration and crop yields. Verslag. Landbouwk. Onderz. 64.6.' (Institute of Biological and Chemical Research on Field Crops and Herbage: Wageningen, The Netherlands)
- Farquhar GD, Richards RA (1984) Isotopic composition of plant carbon correlates with water-use efficiency of wheat genotypes. *Australian Journal of Plant Physiology* **11**, 539–552. doi:10.1071/PP9840539
- Farquhar GD, O'Leary MH, Berry JA (1982) On the relationship between carbon isotope discrimination and the intercellular carbon dioxide concentration in leaves. *Australian Journal of Plant Physiology* **9**, 121–137. doi:10.1071/PP9820121
- Fletcher AL, Sinclair TR, Allen LH Jr (2007) Transpiration responses to vapor pressure deficit in well-watered 'slow-wilting' and commercial soybean. *Environmental and Experimental Botany* **61**, 145–151. doi:10.1016/j.envexpbot.2007.05.004
- Gholipour M, Prasad PVV, Mutava RN, Sinclair TR (2010) Genetic variability of transpiration response to vapor pressure deficit among sorghum genotypes. *Field Crops Research* **119**, 85–90. doi:10.1016/j.fcr.2010.06.018
- Gholipour M, Sinclair TR, Prasad PVV (2012) Genotypic variation within sorghum in transpiration response to drying soil. *Journal of Agronomy and Crop Science*, in press.
- Gilbert ME, Zwieniecki MA, Holbrook NM (2011) Independent variation in photosynthetic capacity and stomatal conductance lead to differences in intrinsic water use efficiency in eleven soybean genotypes before and during drought. *Journal of Experimental Botany* **62**, 2875–2887. doi:10.1093/jxb/erq461
- Haefele SM, Siopongco JDLC, Boling AA, Bouman BAM, Tuong TP (2009) Transpiration efficiency of rice (*Oryza sativa* L.). *Field Crops Research* **111**, 1–10. doi:10.1016/j.fcr.2008.09.008
- Hall AE, Thiauw S, Krieg DR (1994) Consistency of genotypic ranking for carbon isotope discrimination by cowpea grown in tropical and subtropical zones. *Field Crops Research* **36**, 125–131. doi:10.1016/0378-4290(94)90061-2
- Hammer GL, Farquhar GD, Broad IJ (1997) On the extent of genetic variation for transpiration efficiency in sorghum. *Australian Journal of Agricultural Research* **48**, 649–655. doi:10.1071/A96111
- Hubick KT, Shorter R, Farquhar GD (1988) Heritability and genotype x environment interactions of carbon isotope discrimination and transpiration efficiency in peanut (*Arachis hypogaea* L.). *Australian Journal of Plant Physiology* **15**, 799–813. doi:10.1071/PP9880799
- Hufstetler EV, Boerma HR, Carter TE, Earl HG (2007) Genotypic variation for three physiological traits affecting drought tolerance in soybean. *Crop Science* **47**, 25–35. doi:10.2135/cropsci2006.04.0243
- Kemarian AR, Stockle CO, Huggins DR (2005) Transpiration-use efficiency of barley. *Agricultural and Forest Meteorology* **130**, 1–11. doi:10.1016/j.agrformet.2005.01.003
- Kholova J, Hash CT, Kakkera A, Kocova M, Vadez V (2010a) Constitutive water-conserving mechanisms are correlated with terminal drought tolerance of pearl millet (*Pennisetum glaucum* (L.) R.Br.). *Journal of Experimental Botany* **61**, 369–377. doi:10.1093/jxb/erp314
- Kholova J, Hash CT, Kumar PL, Yadav RS, Kocova M, Vadez V (2010b) Terminal drought-tolerant pearl millet (*Pennisetum glaucum* (L.) R.Br.) have high leaf ABA and limit transpiration at high vapour pressure deficit. *Journal of Experimental Botany* **61**, 1431–1440. doi:10.1093/jxb/erq013
- Monneveux P, Rekika D, Acevedo E, Othmane M (2006) Effect of drought on leaf gas exchange, carbon isotope discrimination, transpiration efficiency and productivity in field grown durum wheat genotypes. *Plant Science* **170**, 867–872. doi:10.1016/j.plantsci.2005.12.008
- Morgan JA, LeCain DR, McCaig TN, Quick JS (1993) Gas exchange, carbon isotope discrimination, and productivity in winter wheat. *Crop Science* **33**, 178–186. doi:10.2135/cropsci1993.0011183X003300010032x
- Passioura JB (1996) Drought and drought tolerance. *Plant Growth Regulation* **20**, 79–83. doi:10.1007/BF00024003
- Rebetzke GJ, Condon AG, Richards RA, Farquhar GD (2002) Selection for reduced carbon isotope discrimination increases aerial biomass and grain yield of rainfed bread wheat. *Crop Science* **42**, 739–745. doi:10.2135/cropsci2002.0739
- Sadok W, Sinclair TR (2009) Genetic variability of transpiration response to vapor pressure deficit among soybean cultivars. *Crop Science* **49**, 955–960. doi:10.2135/cropsci2008.09.0560
- Sadok W, Sinclair TR (2010) Genetic variability of transpiration response of soybean (*Glycine max* (L.) Merr.) shoots to leaf hydraulic conductance inhibitor AgNO₃. *Crop Science* **50**, 1423–1430. doi:10.2135/cropsci2009.10.0575
- Sadras VO, Milroy SP (1996) Soil-water thresholds for the responses of leaf expansion and gas exchange: a review. *Field Crops Research* **47**, 253–266. doi:10.1016/0378-4290(96)00014-7
- Sinclair TR, Muchow RC (2001) System analysis of plant traits to increase grain yield on limited water supplies. *Agronomy Journal* **93**, 263–270. doi:10.2134/agronj2001.932263x
- Sinclair TR, Tanner CB, Bennett JM (1984) Water-use efficiency in crop production. *Bioscience* **34**, 36–40. doi:10.2307/1309424
- Sinclair TR, Hammer GL, van Oosterom EJ (2005) Potential yield and water-use efficiency benefits in sorghum from limited maximum transpiration rate. *Functional Plant Biology* **32**, 945–952. doi:10.1071/FP05047
- Sinclair TR, Zwieniecki MA, Holbrook NM (2008) Low leaf hydraulic conductance associated with drought tolerance in soybean. *Physiologia Plantarum* **132**, 446–451. doi:10.1111/j.1399-3054.2007.01028.x
- Sinclair TR, Messina CD, Beatty A, Samples M (2010) Assessment across the United States of the benefits of altered soybean drought traits. *Agronomy Journal* **102**, 475–482. doi:10.2134/agronj2009.0195
- Tanner CB, Sinclair TR (1983) Efficient water use in crop production: research and re-search? In 'Limitations to efficient water use in crop production'. (Eds HM Taylor, WR Jordan, TR Sinclair) pp. 1–27. (American Society of Agronomy: Madison, WI)
- Vadez V, Rao S, Kholova J, Krishnamurthy L, Kashiwagi J, Ratnakumar P, Sharma KK, Bhatnagar-Mathur P, Basu PS (2008) Root research for drought tolerance in legume: *Quo vadis?* *Journal of Food Legumes* **21**, 77–85.
- Wright GC, Hubick KT, Farquhar GD (1988) Discrimination in carbon isotopes of leaves correlates with water-use efficiency of field-grown peanut cultivars. *Australian Journal of Plant Physiology* **15**, 815–825. doi:10.1071/PP9880815
- Wright GC, Nageswara Rao RC, Farquhar GD (1994) Water-use efficiency and carbon isotope discrimination in peanut under water deficit conditions. *Crop Science* **34**, 92–97.
- Xin Z, Aiken R, Burke J (2009) Genetic diversity of transpiration efficiency in sorghum. *Field Crops Research* **111**, 74–80. doi:10.1016/j.fcr.2008.10.010
- Xue Q, Soundararajan M, Weiss A, Arkebauer TJ, Baenziger PS (2002) Genotypic variation of gas exchange parameters and carbon isotope discrimination in winter wheat. *Journal of Plant Physiology* **159**, 891–898. doi:10.1078/0176-1617-00780
- Zaman-Allah M, Jenkinson DM, Vadez V (2011) Chickpea genotypes contrasting for seed yield under terminal drought stress in the field differ for traits related to the control of water use. *Functional Plant Biology* **38**, 270–281. doi:10.1071/FP10244
- Zur B, Jones JW (1984) Diurnal changes in the instantaneous water use efficiency of soybean crop. *Agricultural and Forest Meteorology* **33**, 41–51. doi:10.1016/0168-1923(84)90092-3