
# Lower Soil Moisture Threshold for Transpiration Decline under Water Deficit Correlates with Lower Canopy Conductance and Higher Transpiration Efficiency in Drought Tolerant Cowpea

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Terminal drought tolerance implies that plants have enough water to fill grains. Water saving traits, measured in tolerant and sensitive cowpea lines, showed that tolerant lines have developed several constitutive mechanisms, closely related to one another, which reduces the rate of water use and delay drought effects. This opens the possibility to decipher their genetic basis towards the development of drought tolerant cowpea cultivars.

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## Abstract

Water availability being critical for reproduction and grain filling, terminal drought tolerance may involve water saving traits. Experiments were undertaken under different VPD and water regimes (water stress [WS] and well-watered [WW]) to test genotypic differences and traits relationships in the fraction of transpirable soil water [FTSW] at which transpiration declines, canopy conductance [TR, g $H_2O$ $cm^{-2}$ $h^{-1}$], canopy temperature depression [CTD, °C], transpiration efficiency [TE, g $kg^{-1}$], growth parameters, using fifteen contrasting cowpea genotypes. Under WW conditions at vegetative and early podding stage, plant mass and leaf area were larger under low than under high VPD conditions and was generally lower in tolerant than in sensitive genotypes. Several tolerant lines had lower TR under WW conditions and restricted TR more than sensitive lines under high VPD. Under WS conditions, transpiration declined at lower FTSW in tolerant than in sensitive lines. Tolerant lines also maintained higher TR and CTD under severe stress than sensitive lines. TE was higher in tolerant than in sensitive genotypes under WS conditions. Significant and close relationships were found between TR and TE, CTD, and FTSW in both environments under different water regime conditions. In sum, traits that condition how genotypes manage limited water resources discriminated tolerant and sensitive lines. Our interpretation is that a lower canopy conductance limits plant growth and plant water use, and allows tolerant lines to behave like non-stressed plants until the soil is drier and maintains a higher transpiration under severe stress. A lower TR at high VPD leads to higher transpiration efficiency.

**Keywords**: Canopy temperature depression, Drought stress, Fraction of transpirable soil water, Canopy conductance, Transpiration efficiency, *Vigna unguiculata*.

## Introduction

Cowpea [*Vigna unguiculata* (L.) Walp.], a protein-rich grain legume is widely cultivated by resource-poor farmers in the semi-arid tropics of Africa, Asia, and Latin America where it is immensely important for its central role in the diet and economy of millions people (Singh *et al.* 2003; Dadson *et al.* 2005; Muchero *et al.* 2009). Despite its capacity to withstand water deficits, significant differences exist among cowpea genotypes for their response to terminal drought, i.e. water deficit stress occurring at the end of the growing season (Mai-Kodomi *et al.* 1999*a*; Muchero *et al.* 2008). In Africa, cowpea is commonly grown in the Sudanian and Sahelian semi-arid regions, where climate change is likely to make drought stresses even more severe in the future (Hall *et al.* 2003; Wittig *et al.* 2007; Vadez *et al.* 2011). Therefore, the identification of drought tolerant cowpea cultivars adapted to these agro-ecological zones is needed (Van Duivenbooden *et al.* 2002; Kholová *et al.* 2010*a*).

Extensive research has been carried out on the screening for mid- and late-season drought tolerance in cowpea, focusing on carbon isotope discrimination, chlorophyll stability index, leaf gas exchange, relative turgidity, relative water content, water use efficiency, and water potential (Hall *et al.* 1990; Cruz de Carvalho *et al.* 1998; Ashok *et al.* 1999; Singh and Matsui 2002; Ogbonnaya *et al.* 2003; Anyia and Herzog 2004; Hall 2004; Onwugbuta-Enyi 2004; Padi 2004; Slabbert *et al.* 2004; Souza *et al.* 2004; Hamidou and Braconnier 2007).

Nevertheless, only very few studies have used these indices to select parental genotypes in further genetic studies (Mai-Kodomi *et al.* 1999*b*; Muchero *et al.* 2009). We argue that, despite the complexity of the drought response, simple hypotheses based on water needs can be developed to guide the selection of critical traits (Vadez *et al.* 2007). Here, we test one of these hypothesis, i.e. that water saving traits are important for terminal drought adaptation, by comparing a range of contrasting lines.

Because water availability is critical for the reproduction and grain filling period, plant traits involved in a conservative use of soil water even if water is not limiting are indeed likely to be relevant for yield improvement under limiting water (Vadez *et al.* 2011). This has been shown in chickpea (Zaman-Allah *et al.* 2011*a, b*) and pearl millet (Kholova *et al.* 2010*a, b*). Recent findings showed that leaf area was lower in tolerant chickpea (Zaman-Allah *et al.* 2011*a*), peanut (Ratnakumar and Vadez 2011), and this logically limits plant water use. Significant variations in canopy conductance were also found among contrasting genotypes under non-limited water conditions in cowpea (Hall and Shulze 1980), chickpea (Zaman-Allah *et al.* 2011a), soybean (Purcell and Specht 2004; Fletcher *et al.* 2007; Sadok and Sinclair 2009), peanut (Bhatnagar-Mathur *et al.* 2007), sorghum (Gholipoor *et al.* 2010), and pearl millet (Kholova *et al.* 2010*b*).

There is also water saving option by having different soil moisture threshold where transpiration begins to decline upon progressive exposure to water deficit. For instance, the transpiration decline occurred in wetter soil (higher soil moisture threshold for transpiration decline) in tolerant than in sensitive chickpea genotypes (Zaman-Allah *et al.* 2011*a*). Genotypic differences for this trait were also found for transpiration response to progressive water deficit stress in several other crops (Vadez and Sinclair 2001; Bhatnagar-Mathur *et al.* 2007; Hufstetler *et al.* 2007; Devi *et al.* 2010;). This characteristic offers the opportunity to reduce water use and such information is not available in cowpea. Nevertheless, tolerant pearl millet had a lower FTSW threshold for the transpiration decline (Kholova *et al.* 2010*a*). This was interpreted to be a consequence of the lower canopy conductance and then lower plant transpiration of tolerant genotypes under well-watered conditions, which helped maintain the relative transpiration of water stressed plants to a level similar to well-watered plants until the soil was dryer. Whether these thresholds relate to the canopy conductance under well-watered conditions is an important question to resolve. Whether these canopy conductance differences would also relate to genotypic differences in transpiration efficiency [TE], which is a major source of crops yield variation under drought stress (Condon *et al.* 2004; Sheshshayee *et al.*

2006; Krishnamurthy *et al.* 2007), is another one. None of these questions has been tested in cowpea and they are addressed in here.

According to Gwathmey *et al.* (1992) and Gwathmey and Hall (1992), another important morphological trait that may contribute to drought adaptation of cowpea is a delayed leaf senescence [DLS] under water stress, which would enhance plant survival after a mid-season drought and limit damages to the first flush of pods. Cultivars with DLS also have enhanced production of forage because their leaves remain green and attached to the plant until harvest. Moreover, DLS can be easily measured by visual scoring using an appropriate scale as used by Muchero *et al*. 2008 to discriminate fifteen cowpea genotypes that exhibit significant genetic variation for drought tolerance.

In summary, the overall objective of the present study was to assess whether cowpea genotypes contrasting for their response to terminal drought in the field differ in their response to progressive soil drying conditions. Specific objectives were to: (i) evaluate growth and canopy conductance in different atmospheric VPD conditions, and test whether drought tolerant lines differ from sensitive one; (ii) compare whether tolerant and sensitive cowpeas differ in their growth response to progressive exposure to drought stress; (iii) determine whether there are variations in the soil moisture thresholds [FTSW] where transpiration declines across genotypes and environments; (iv) Assess possible relationships between some of these water saving traits.

## Material and methods

*Plant growth and description of experiments*

Experiments were simultaneously carried out under different vapor pressure deficit [VPD], by setting experiments in glasshouse and outdoor environments at ICRISAT-Patancheru in India (17° 30' N; 78° 16' E; altitude 549 m) during the post-rainy season between March and May 2010. During the crop growing period, the VPD was lower in glasshouse than outdoors, where air temperature was higher and relative humidity lower. The air temperature, relative humidity and resulting VPD varied between 24-40°C, 45-85%, 0.55-4.15 kPa, respectively in glasshouse while varying between 25-50°C, 20-70%, 0.85-7.45 kPa, respectively outdoors (Suppl. Fig. 1). Fifteen cowpea genotypes, contrasting for their response to drought stress under field and controlled environment conditions (Belko N., Cisse N. *et al*. unpublished), were selected for this investigation (Table 1). The work leading to this classification was conducted in well managed experimental station field in Senegal, Nigeria, Burkina Faso and California, and in controlled environments glasshouse and growth chamber in India, in seasons when the VPD was high.Seeds were received from the Department of Botany and Plant Sciences of the University of California Riverside, USA.

Plants were grown in plastic pots [20 cm diameter x 20 cm tall] filled with 5.5 kg of sandy clay loam Alfisol collected from the ICRISAT farm and fertilized with di-ammonium phosphate at the rate of 0.3 g $kg^{-1}$ soil and with farm-yard manure (1:50 v/v). The day before planting, the top soil of each pot was added with 2 g carbofuran to prevent seeds damage from soil-borne pests. Each pot was sown with 3 seeds and thinned to one seedling a week later. For each environment [glasshouse and outdoor], twenty one plants of each genotype were grown under well watered conditions until 30 days after sowing [the time when water treatments imposition started]. Then, fifteen most uniform plants of each genotype were selected to design the experiments in both environments. A thermo-hygrograph sensor (Tinytag Ultra 2 TGU-4500 Gemini Dataloggers Ltd., Chichester, UK) was positioned within the plants canopy in both glasshouse and outdoor environments for regular records of the air temperature and relative humidity throughout the crop growth and measurements period.

The day before water treatments imposition, pots were watered, allowed to drain to reach field capacity, and then late in the evening, pots were bagged with transparent plastic bag wrapped around the plant stem to prevent soil evaporation during the evaluation of plants transpiration. The fifteen plants were divided in three sets of five plants: the first set was kept under well-watered conditions [WW] and used for assessing the plants transpiration rate [TR, in g water loss $cm^{-2}$ $h^{-1}$, i.e. used as a simple proxy for canopy conductance] response to natural change of atmospheric VPD during the course of an entire clear day then harvested to measure the initial plant biomass [pre dry-down]. The second set was maintained under well-watered [WW] conditions and the third set was gradually exposed to water stress [WS] (see below). The experimental layout was a randomized complete block design with treatment-set as the main factor and genotypes as sub-factor randomized five times within each block.

*Transpiration rate in response to VPD*

The rate of water-loss per unit of leaf area [TR] was assessed on WW plants from the first set (see above) under natural variations of VPD during the course of an entire sunny day in both glasshouse and outdoor conditions. The plant transpiration was measured gravimetrically from the losses in pots weight between consecutive weighings. Pots were weighted with 0.01 g precision scale (PE 12, Mettler Toledo, Schweiz-GmbH, Germany) hourly between 7:00 am and 5:00 pm (India Standard Time). At the end of the day, plants were harvested and the leaf area measured (LI-3100, Licor, Lincoln, Nebraska, USA). Transpiration and leaf area data were used to estimate the transpiration rate, i.e. leaf water loss per unit of leaf area [TR, g $H_2O$ $cm^{-2}$ $h^{-1}$]. The plants parts were dried in an oven at 60°C for 3 days then, their dry masses were recorded. The specific leaf area [SLA, $cm^2$ $g^{-1}$] was calculated as the ratio between the leaf area [LA] and the leaf dry weight.

*Plant exposure to progressive water deficit stress (dry-down)*

After bagging, pots were weighed around 9:00 am at 31 days after sowing [DAS] to have the initial pot weight and thereafter pots were weighed every day in the morning to calculate the daily plants transpiration. Well watered plants were maintained as such by daily re-watering up to 80% field capacity, i.e. by bringing the pot weight to 200 g below the field capacity weight every day. Water stress plants were exposed to stress by partially compensating plant water loss from transpiration, i.e. plants were allowed to lose no more than 70 g each day. Therefore, any transpiration in excess of 70 g was added back to the pots, as previously described by Vadez and Sinclair (2001), to allow a progressive development of water-deficit stress over approximately two weeks.

The transpiration values were normalized to facilitate comparison as previously described by Kholova *et al*. (2010*a*). First, the daily transpiration ratio (TR) for each plant was calculated as the ratio of the transpiration rate of each individual WS plant divided by the average of transpiration rate for the five WW plants of that genotype. Secondly, the TR data were normalized by dividing each TR value over time by the average of the TR value for the first 3 days of the experiment when there was still no water limitation. This second normalization aimed to remove variation resulting from differences in plant size among WS plants within a genotype. This gave the normalized transpiration ratio (NTR) which accounted for plant to plant variation in transpiration within each genotype. When the NTR of stressed plants fell below 0.10, i.e. when the transpiration of WS plants was < 10% of that of WW plants, all the plants were harvested and their different parts were dried in an oven at 60°C for

3 days and then their dry mass were measured. The genotypes reached all that stage within two days from one another.

After the final harvest, the daily fraction of transpirable soil water [FTSW], i.e. the amount of soil water available for transpiration, was back-calculated on each day of the experiment. First, the total transpirable soil water [TTSW] available to support plant transpiration in each pot was calculated as the difference between the initial and final pot weight which was defined as the weight at the end of the experiment (Sinclair and Ludlow 1986). The FTSW values were calculated as:

$$FTSW = (\text{Daily Pot}_{\text{weight}} - \text{Final Pot}_{\text{weight}}) / TTSW \quad (1)$$

Since the plants were allowed to transpire no more than 70 g water per day, all the genotypes were exposed to similar stress intensities, at least from the viewpoint of the soil water content. Changes in NTR during the soil drying cycle were expressed as a function of FTSW which was used as the indicator of the stress intensity (Ritchie 1981).

*Canopy temperature depression [CTD], transpiration efficiency [TE], and leaf scoring*

The day before the end of the dry-down leaf temperatures were recorded on five replicates plants for both WW and WS treatments in both environments between 8:00 and 9:00 am with an IR-thermometer (Fluke 574, Fluke Thermography, Annapolis Lane Plymouth, MN, USA). Air temperature was recorded from a temperature and relative humidity recorder (Gemini Tiny Tag Ultra 2 TGU-4500 Data logger), which was located at the crop canopy level. In each plant, temperatures were recorded on three leaves at the top of the canopy and averaged. The canopy temperature depression [CTD] was calculated as the difference between the air temperature and the leaf temperature (CTD = $T_{air} – T_{leaf}$).

Transpiration efficiency [TE, g biomass $kg^{-1}$ water transpired] was calculated for each control and stressed plants in both environments as the ratio between the increase in plant biomass over the course of the dry-down and the total water transpired during the same time:

$$TE = (\text{Final Harvest}_{\text{biomass}} – \text{Pre Dry-down}_{\text{biomass}}) / \text{Total Water}_{\text{transpired}} \quad (2),$$

where the pre dry-down biomass was the biomass of plants used to assess the TR response to VPD and harvested at the beginning of the dry-down. The final harvested biomass was that of

WW and WS plants harvested at the end of the dry-down. The total water transpired was the sum of daily transpiration measured by daily weighing of pots during the dry-down.

Leaf senescence due to water-deficit stress was scored at the end of the dry-down in both glasshouse and outdoors. The state of leaf senescence was rated on a scale from 1 to 5, with 1 = totally green and turgescent, 2 = green and slightly wilted, 3 = green-yellow and wilt, 4 = yellow-green and severely wilt and 5 = completely yellow to brown / almost died.

*Statistical analysis*

Analyses of variance (Anova) were done using the statistical package program SAS (SAS Institute, Inc., 1988, Cary, NC, USA). One-way Anova was run to test the genotypic differences within each water treatment for plant growth parameters, transpiration rates, canopy temperature depression, transpiration efficiency and visual scores. The Tukey–Kramer test was used for the analysis of differences between genotype means. The relationships between TE and TR, TE and the FTSW thresholds, TR and the FTSW threshold, and CTD and TR were also tested.

For the FTSW thresholds analysis for each genotype, each NTR value was plotted against its corresponding FTSW value for each day of the experiment. A plateau regression procedure of the SAS program was used to estimate a specific FTSW threshold value where NTR initiated its decline (Ray and Sinclair 1997). This analysis provided a standard error and 95% confidence interval for each threshold value for each genotype. A non-linear regression analysis was done using GraphPad Prism (GraphPad 2.01, San Diego, CA, 1996) to fit the exponential model presented by Muchow and Sinclair (1991) (NTR = 1 / [1+A * exp (B * FTSW)]. The regression result obtained using this equation was compared among genotypes based on 95% confidence intervals of coefficients A and B. The plateau regression attempted to fit two linear segments where one segment is a plateau at Y = 1 and the second regression is a linear change in Y with respect to X. A key output from this analysis is the FTSW threshold for the two segments and the confidence intervals for this threshold. The averages of threshold values were compared across the genotypes using Tukey-Kramer method of Genstat (Genstat 12.1 VSN International Ltd., Hemel Hempstead, UK).

For the TR versus CTD relationship, the data were analyzed with the split line regression option of Genstat (9.0), which provides a breakpoint value where the slope of the fitted regression significantly changes.

## Results

*Genotypic variation for plant growth under WW conditions at 30 DAS*

Under glasshouse conditions, most tolerant lines had lower vigor than sensitive lines (Table 2). Higher root dry weight was found in five out of seven sensitive lines than in seven out of eight tolerant lines. All sensitive genotypes, except IT89KD-288, produced higher plant biomass than five tolerant lines. This was related more to difference in leaf dry weight (all but one sensitive genotypes had higher leaf dry weight than five out of eight tolerant) than in stem dry weight. The specific leaf area (SLA, $cm^2$ $g^{-1}$) varied between genotypes but did not discriminate tolerant from sensitive lines. Leaf area ($cm^2$ $plant^{-1}$) was the smallest in all drought tolerant lines, except IT97K-499-39 and KVx-61-1, than in all drought sensitive, except IT89KD-288 (Table 2).

Under outdoor conditions, growth parameters varied significantly among genotypes but did not clearly discriminate tolerant from sensitive lines, although the leaf area of five out of seven sensitive genotypes was higher than five out of eight tolerant ones (Table 2). In addition to the significant genotypic variations for all the growth parameters, there were highly significant differences between the glasshouse and outdoors environments for these growth attributes. Also, a significant effect of the interaction between genotype and environment [G x E] on the variation of the growth parameters was found, explaining a variance close to that for genotypic effect (Table 2).

In summary, tolerant genotypes had low early vigor for the majority of them but their differences with the sensitive lines for growth parameters were not clearly expressed under high VPD conditions outdoors as compared with the glasshouse environment.

*Response of leaf transpiration rate to changing atmospheric VPD*

Under glasshouse conditions at 30 DAS, canopy conductance [TR, g $H_2O$ $cm^{-2}$ $h^{-1}$] closely followed the diurnal pattern of atmospheric VPD, which ranged from 1.10 to 4.08 kPa during the day. Canopy conductance was significantly lower in most tolerant genotypes [IT84S-2049, IT99K-124-5, Mouride, Suvita 2] than in most of the sensitive ones [Bambey 21, IT82E-18, IT89KD-288, UC-CB46]. The largest differences between tolerant and sensitive lines for the canopy conductance were recorded between 11:00 and 15:00 where the VPD was above 3.5 kPa (most representative genotypes shown in Fig. 1A). TR, averaged for the whole day, was then about 40% lower in tolerant than in sensitive lines (data not shown). The total water transpired per plant throughout the day was significantly lower in five out of eight tolerant lines than in six out of seven sensitive lines in the glasshouse conditions (Fig. 2A).

Under outdoor conditions at 30 DAS, similar results for the canopy conductance to those under lower VPD conditions in the glasshouse were obtained: (i) canopy conductance was significantly lower in the most tolerant genotypes than in sensitive lines, (ii) largest differences were recorded at VPD above 6.5 kPa (Fig. 1B), (iii) average TR for the whole day was 30% lower in tolerant than in sensitive lines (data not shown). The total water transpired per plant during the whole day was also significantly lower in four out of eight tolerant genotypes than in six out of seven sensitive genotypes under well watered conditions outdoors (Fig. 2B).

*Effect of drought exposure on plant growth and transpiration efficiency*

Under glasshouse conditions at the end of the dry-down experiment under WW conditions, the root, stem, leaf, and plant biomasses of tolerant genotypes [IT84S-2049, Mouride, Suvita 2, KVx-61-1] remained lower than that of the sensitive ones [IT82E-18, IT83D-422, IT93K-93-10, IT97K-556-6]. The same applied to a lesser extent in the WS treatment (Table 3). Biomass increase, total water transpired, and TE under WW conditions did not discriminate tolerant from sensitive lines (Table 4). In the WS treatment, total water transpired was higher in six out of eight tolerant lines than in five out of seven sensitive lines (Table 4). By contrast, at the end of the drydown treatment, all genotypes had extracted a similar amount of water from the soil (TTSW, data not shown). TE values did not discriminate tolerant from sensitive lines.

Under outdoor conditions, in the WW treatment, all tolerant lines had lower total water transpired than five out of seven sensitive lines. TE was also higher in five out of eight tolerant lines than in five out of seven sensitive lines. In the WS treatment, there was genotypic variation for the biomass increase, total water uptake, and TE, but no discrimination between tolerant and sensitive lines (Table 4).

Overall, at the end of the dry down, the most tolerant lines showed lower biomass than sensitive ones, especially under WW and to some extent under WS stress conditions in the low VPD conditions of the glasshouse. Cowpea accumulated more biomass under low VPD than under high VPD conditions and, as expected, TE was lower under high VPD as compared with the low VPD conditions, for both water treatments. However, several drought tolerant lines [Mouride, Suvita 2 and IT84S-2049] maintained TE at higher level as compared with all the sensitive lines under high VPD conditions outdoors and especially in the WS treatment.

*Response of leaf gas exchange to progressive soil drying*

In the glasshouse, the FTSW thresholds for transpiration decline were lower in six out of eight tolerant than in five out of seven sensitive lines. The FTSW thresholds varied between 0.44 and 0.70 (Table 5) with the lowest thresholds recorded in the tolerant genotypes [Mouride, IT84S-2049, Suvita 2] and the highest threshold showed by the sensitive lines [Bambey 21, IT83D-442, IT93K-93-10]. A typical transpiration response discriminating tolerant from sensitive lines is presented in Fig. 3A&B. In outdoor conditions, similar results were obtained, with six out of eight tolerant lines having lower FTSW thresholds than six out of seven sensitive ones (Table5; Fig. 3C&D).

*Genotypic differences in canopy temperature depression in response to drought*

Under glasshouse conditions in the WW treatment, the canopy temperature depression [CTD] did not discriminate tolerant from sensitive lines at the end of the dry-down [45 DAS] (Fig. 4A). In contrast under WS conditions, CTD varied among genotypes and was lower in sensitive lines (average of -0.03 °C) than in tolerant lines (average of 1.39 °C) (Fig. 4B). Only one sensitive and one tolerant line differed from this. Under outdoor conditions, similar results were obtained. In the WW treatment, there was no clear CTD discrimination between tolerant and sensitive genotypes (Fig. 4C). Under WS conditions, the CTD was lower in sensitive lines (average of -0.74 °C) than in tolerant lines (average of 1.82 °C) (Fig. 4D).

*Scoring for stay green under water-deficit*

Leaf senescence caused by drought stress varied across genotypes under both glasshouse and outdoor conditions (Table 6), and several cowpea genotypes preserved stem and leaf greenness more than others (Suppl. Fig. 2). Tolerant Mouride, Suvita 2, IT84S-2049, and IT97K-499-39 kept greener (lower scores) than sensitive Bambey 21, IT82E-18, IT97K-556-6, and UC-CB46 (higher scores). There was a close agreement between the two environments for leaf damage visual rating.

## Discussion

Several traits related to plant growth and patterns of soil water use under both well watered and water stressed conditions discriminated terminal drought-tolerant from sensitive genotypes and that in both glasshouse and outdoors environments.

*Plant growth under non-limited water and drought stress conditions*

At 30 DAS under WW conditions, most tolerant genotypes had lower growth than sensitive lines under low VPD conditions in the glasshouse. These growth differences were not clearly expressed under high VPD conditions outdoors, where growth was depressed, likely because of a depressive VPD effect on leaf expansion (Tardieu *et al.* 2000). These early growth differences were explained by two different mechanisms: (i) a lower leaf area of tolerant line; (ii) a lower canopy conductance (TR, g $cm^{-2}$ $h^{-1}$). These present results are consistent with previous study in chickpea (Zaman-Allah *et al.* 2011*a*) and pearl millet (Kholova *et al.* 2010*a*). We interpret that under situations of terminal drought, high early vigor and development of large leaf areas could lead to rapid water depletion, and leave plants facing water scarcity while completing their cycle. Therefore, lower early growth by decreased LA and lower canopy conductance under WW conditions, as found in some tolerant lines, could be important adaptive response against late season drought stress, as previous hypothesized (Hammer 2006). Our findings in cowpea, added to the earlier one on chickpea, pearl millet, or sorghum facing similar stress, clearly indicate that limiting plant growth is a common mechanism across crops facing terminal drought stress. Of course, limiting plant growth would limit potential yield in those years or locations where the stress is mild.

At the end of the dry-down experiment, the biomass increase under WS was higher in the tolerant than in the sensitive lines. This was related to the higher soil moisture thresholds where transpiration declined in sensitive genotypes. Similar findings have been reported in peanut (Bhatnagar-Mathur *et al.* 2007; Devi *et al.* 2009), maize (Ray *et al.* 2002) and millet (Kholova *et al.* 2010*a*). This could also relate to the fact that, although transpiration efficiency decreased considerably under high VPD conditions across all genotypes, this decrease was relatively less in drought tolerant genotypes. Similar result were obtained in wheat, where tolerant lines maintained higher growth, biomass increase, water extraction and TE than sensitive lines under water stress (Condon *et al*. 2004).

*Genotypic differences in the transpiration rate response to natural change of VPD*

Tolerant lines had lower canopy conductance than sensitive lines and these consistent results were observed under both low VPD (glasshouse) and high VPD (outdoors). The largest differences between tolerant and sensitive lines were recorded around midday when the VPD was above 3.5 kPa and 6.5 kPa in the glasshouse and outdoors, respectively. Then, the TR computed for the whole day of experiment, was about 40% and 30% lower in tolerant than in sensitive lines under low VPD and high VPD conditions, respectively. These lower TR values led to, overall, lower total water transpired per plant per day in the majority of tolerant genotypes than in the sensitive lines under WW conditions in both environments. These results are in agreement with similar findings of lower canopy conductance in terminal drought-tolerant lines of pearl millet where both mechanisms were found: (i) a low canopy conductance at low VPD; (ii) a further restriction of canopy conductance at high VPD (Kholova *et al*. 2010*a*). Terminal drought tolerant chickpea also had constitutively lower TR than sensitive lines, but tolerant and sensitive had response of TR to VPD (Zaman-Allah *et al.* 2011*a*). In the previous work in pearl millet, we interpreted that the rapid changes in canopy conductance upon VPD increase could only be mediated by hydraulic signals. Our results are, as far as we know, the first evidences of a possible hydraulic limitations to the transpiration under high VPD in cowpea (Fig. 1), and genotypic differences associated to it that open the possibility of exploiting that feature towards breeding for drought adaptation.

Therefore, in genotypes that would restrict TR, especially at high VPD, there is a scope for water saving which would then be available and essential for grain filling late in the season (Sinclair *et al.* 2005; Ghoolipoor *et al.* 2010; Kholova *et al.* 2010*a*, *b*; Zaman-Allah *et* *al.* 2011*a*, *b*). It was then argued that a lower canopy conductance would lead to higher transpiration efficiency (Sinclair *et al.* 2005). There was indeed a close relationship between a lower TR and a higher TE under high VPD conditions and both water treatments ($R^2 = 0.40$ and 0.76 under WW and WS respectively; Fig. 5C&D), but this relationship was weak or non-significant under low VPD conditions (Fig. 5A&B). Our interpretation is that, in agreement with the theory, plants that would be capable of suppressing transpiration at high VPD would have an increased transpiration efficiency, and logically the capacity to limit transpiration at high VPD would be more beneficial in environments where high VPD conditions are more common, like the outdoor conditions of this work. It should be mentioned that a lower TR could also lead to yield penalties, for example under mild stress or non-limiting water supply (Sinclair and Muchow 2001; Cho *et al.* 2003; Richards *et al.* 2007; Sinclair *et al.* 2010), and could be here the reason for the lower biomass of tolerant lines. Thus, both traits as above described are important to consider only for the breeding of crops with enhanced terminal drought tolerance for regions with high VPD and low water supply.

*Variation in FTSW threshold and transpiration efficiency under drought conditions*

One of the key findings of this investigation was that FTSW threshold for transpiration was lower in most tolerant lines than in most sensitive lines in both glasshouse and outdoors. Therefore, upon progressive exposure to water deficit, transpiration declined in relatively dryer soil (lower FTSW) in the tolerant lines than in the sensitive ones in both low and high VPD conditions. The basis for the calculation of FTSW threshold is the total transpirable soil water (TTSW), which is the amount of water that can be extracted to support transpiration from a same volume of soil. This trait did not vary between cowpea genotypes, which also agree with our findings in other crops species. There is often confusion between TTSW and the total water transpired, which is the sum of TTSW and the water added to the WS plants in the course of the drydown. The water added of course varies between genotypes, and reflects growth differences between genotypes, and the very purpose of using a WW control is to normalize these differences. The differences in the FTSW thresholds where transpiration declines were in agreement with data obtained in groundnut (Bhatnagar-Mathur *et al.* 2007; Devi *et al.* 2009), soybean (Vadez and Sinclair 2001; Hufstetler *et al.* 2007), maize (Ray *et al.* 2002) and pearl millet (Kholova *et al.* 2010*a*). However, these results were different from those obtained in chickpea, where sensitive lines had a decline of transpiration in dryer soils than tolerant lines (Zaman-Allah *et al.* 2011*a*). Sinclair and colleagues (2010) showed that a higher FTSW threshold would contribute to grain yield increase in soybean. Our finding of large genotypic contrast for the FTSW thresholds in cowpea opens a scope to use that trait in breeding. Here, the FTSW thresholds for the decline in transpiration with soil drying were similar across VPD conditions. These results agreed with those reported in maize hybrids (Ray *et al.* 2002), although they differ from earlier assumption from Denmead and Shaw (1962) who make the assumption that FTSW threshold for the decline of transpiration upon imposition of water deficit should increase if the imposition of water deficit took place in conditions of higher evaporative demand.

*Relationship between TR, CTD, TE, and the FTSW thresholds for transpiration decline*

Since the largest TR differences between tolerant and sensitive lines were achieved at the time of the day when the VPD was the highest, a first question was then whether these large TR differences could lead to differences in TE, as hypothesized above. TE and TR were indeed closely related but the relationships were significant only in outdoors conditions, where the VPD was high (Fig. 5). The interpretation is that the low TR at high VPD was caused by a partial stomata closure under high VPD, as it has been shown in other crops (Kholova *et al.* 2010*b*; Devi *et al.* 2010). Therefore, the effective VPD for the transpiration in these plants is shifted to a lower value, leading to a higher TE according to the definition of TE (Tanner and Sinclair 1983). Also recent report indicates that soybean having transpiration sensitivity to high VPD reduced stomatal conductance under high VPD, but this was not accompanied by a proportional decrease in photosynthetic activity. This led to differences in intrinsic water use efficiency (Gilbert *et al.* 2011).

A second question was whether the differences in FTSW thresholds for the transpiration decline were related to the lower TR under WW conditions. There was indeed a tight positive relationship between the FSTW thresholds for transpiration decline and TR under both low and high VPD conditions ($R^2$ = 0.66 and 0.71 respectively; Fig. 6A&B). Our interpretation is that a lower TR, which leads in part to a lower absolute transpiration (Fig. 2), makes drought-stressed plant function like well watered ones until the soil has become dryer, as it was previously found and discussed (Kholova *et al.* 2010*a*). This then leads to having a lower FTSW threshold where transpiration drops upon progressive exposure to water deficit stress.

Since TR and the FTSW thresholds and TR and TE are both related, the third question was then whether these FTSW threshold differences could be related to TE. Under low VPD conditions, the relationship between TE and the threshold for transpiration decline was not significant (Fig. 6C) By contrast, under high VPD conditions there was a negative trend between the thresholds and TE (Fig. 6D). This agreed with the fact that no difference were observed in TE among genotypes under low VPD conditions but under high VPD conditions there were substantial TE differences among genotypes. These results were different from those found in peanut (Devi *et al.* 2009), although the polynomial relationship in the 0.2-0.6 FTSW range in this study was relatively poor ($R^2$=0.39). By contrast, the results presented here are in agreement with more recent results showing also a strong negative relationship ($R^2$=0.88) between the FTSW thresholds and TE (Devi *et al.* 2011).

Our overall interpretation on these three questions is that the lower TR of tolerant lines during the time of the day when the VPD is the highest, which is related to a partial closure of stomata, had two consequences: (i) first the lower TR of tolerant plants at high VPD led to increasing TE level, especially in those conditions with high VPD such as outdoors here. This is what we find here in the negative relationships between TE and TR in outdoor conditions (Fig. 5C&D). (ii) A lower TR saved water and allowed these plants, when exposed to stress, to function like fully irrigated plants for a larger part of the drying cycle. This is our interpretation of the positive relationships between the FTSW threshold for the transpiration decline and TR (Fig. 6A & B), where plants with low TR have indeed low FTSW values for transpiration decline. It is also illustrated by Figure 3, in which NTR of tolerant lines remains at a value of 1 up until lower FTSW values, i.e. for a longer time during the drying period. These two consequences are then the causal factors behind the relationship between the FTSW thresholds for the transpiration decline and TE at high VPD (Fig. 6D). Therefore, these FTSW thresholds become a very powerful tool to select plants that have the capacity to restrict transpiration at high VPD, itself leading to increasing TE.

At the end of the experiment under WS conditions, tolerant genotypes showed higher CTD than sensitive lines, which indicated that at these late stages of stress, tolerant lines likely maintained transpiration activity and this was well related to the lower leaf senescence scoring in these lines. CTD was also closely and positively related to TR, in a broken stick regression that described this relationship under both low and high VPD (Fig. 7). Therefore, the measurement of canopy temperature could become an easy way to assess TR in cowpea.

## Conclusion

For enhancing crops terminal drought tolerance water availability during reproduction and grain filling is crucial (Vadez *et al.* 2007; Zaman-Allah *et al.* 2011*a*). Lower early vigor, lower TR under WW conditions during the vegetative stage, lower leaf area development, sustained transpiration until the soil was relatively drier, and lower canopy conductance under high VPD conditions, appeared to be the main features discriminating tolerant from sensitive genotypes. Also, significant and close relationships were found between TR and: (i) TE under both WW and WS treatments outdoors; (ii) CTD under water stress conditions in both environments; and (iii) FTSW thresholds for transpiration decline. These results support the importance of TR regulation in explaining the differences in adaptation between tolerant and sensitive lines, especially under high VPD where lower TR, explained by a partial stomata closure, contributes to water saving and increase water use efficiency.

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**Table 1**. List of cowpea genotypes compared for their growth, transpiration rate, soil moisture thresholds [FTSW], transpiration efficiency, and canopy temperature depression in response to progressive soil drying.

| | Genotypes | Origin | Maturity |
|---|---|---|---|
| **Drought sensitive** | Bambey 21 | ISRA | Early |
| | IT82E-18 | IITA | Early |
| | IT83D-442 | IITA | Medium |
| | IT89KD-288 | IITA | Medium |
| | IT93K-93-10 | IITA | Early |
| | IT97K-556-6 | IITA | Medium |
| | UC - CB46 | UC Davis | Early |
| **Drought tolerant** | IT84S-2049 | IITA | Early |
| | IT97K-207-15 | IITA | Medium |
| | IT97K-499-39 | IITA | Early |
| | IT98K-128-2 | IITA | Medium |
| | IT99K-124-5 | IITA | Medium |
| | KVx-61-1 | INERA | Early |
| | Mouride | ISRA | Medium |
| | SuVita2 | INERA | Medium |

**Table 2.** Dry mass of plant parts (g plant$^{-1}$) of cowpeas grown under WW conditions and harvested at 31 DAS, before the initiation of the dry-down, in both glasshouse (top) and outdoor (bottom) environments. LA stands for leaf area (cm$^2$) and SLA for specific leaf area (cm$^2$ g$^{-1}$). Values shown with means are SE of five replicated plants per genotype. Lower case letters following means discriminate genotypes for each parameter based on Tukey's method at significance level 0.05. Outputs from the analysis of genotype, environment, and genotype x environment interaction effects on the different growth parameters are presented at the bottom of the table.

***Glasshouse***

| | Genotypes | Root dw (g) | Stem dw (g) | Leaf dw (g) | Plt dw (g) | LA (cm2) | SLA (cm2/g) |
|---|---|---|---|---|---|---|---|
| **Drought sensitive** | Bambey 21 | 1.73±0.10 c | 3.36±0.12 bc | 4.56±0.16 b | 7.91±0.11 c | 1265±69.89 a | 278±20.86 c |
| | IT82E-18 | 2.56±0.31 a | 3.40±0.21 b | 4.30±0.23 b | 7.70±0.21 c | 1320±87.51 a | 307±27.26 b |
| | IT83D-442 | 2.66±0.12 a | 3.60±0.23 b | 3.60±0.16 c | 7.20±0.16 c | 1096±66.77 b | 305±26.84 b |
| | IT89KD-288 | 1.21±0.18 d | 2.40±0.10 c | 2.41±0.11 d | 4.82±0.16 f | 499±21.16 f | 207±13.56 d |
| | IT93K-93-10 | 2.26±0.09 b | 4.18±0.21 a | 4.41±0.15 b | 8.59±0.25 b | 1166±24.83 ab | 264±16.31 c |
| | IT97K-556-6 | 2.00±0.06 b | 4.45±0.20 a | 4.19±0.15 b | 8.64±0.22 b | 1186±59.29 ab | 284±12.94 c |
| | UC - CB46 | 2.03±0.07 b | 3.37±0.10 bc | 3.87±0.16 c | 7.25±0.13 c | 1295±59.33 a | 335±19.03 ab |
| **Drought tolerant** | IT84S-2049 | 1.12±0.11 d | 1.68±0.19 e | 1.57±0.11 e | 3.25±0.17 g | 569±16.24 e | 364±33.06 a |
| | IT97K-207-15 | 1.66±0.13 c | 3.74±0.18 b | 4.43±0.18 b | 8.16±0.19 b | 835±19.86 d | 189±18.83 e |
| | IT97K-499-39 | 2.16±0.08 b | 4.23±0.18 a | 5.29±0.12 a | 9.51±0.14 a | 1119±38.84 b | 212±10.41 d |
| | IT98K-128-2 | 1.66±0.17 c | 3.48±0.17 b | 2.83±0.18 d | 6.31±0.17 d | 867±33.13 d | 307±11.59 b |
| | IT99K-124-5 | 1.60±0.16 c | 2.91±0.18 c | 2.50±0.20 d | 5.40±0.34 e | 1016±33.38 c | 409±28.17 a |
| | KVx-61-1 | 1.73±0.07 c | 3.83±0.24 b | 3.61±0.19 c | 7.44±0.20 c | 1205±30.46 ab | 334±18.96 ab |
| | Mouride | 1.01±0.06 d | 2.22±0.09 d | 2.63±0.12 d | 4.84±0.13 f | 588±22.01 e | 224±11.70 d |
| | SuVita2 | 1.08±0.05 d | 2.65±0.14 c | 2.62±0.14 d | 5.27±0.12 e | 882±47.61 d | 337±20.35 ab |
| | ***Sensitive Mean*** | ***2.06*** | ***3.54*** | ***3.91*** | ***7.44*** | ***1118*** | ***283*** |
| | ***Tolerant Mean*** | ***1.50*** | ***3.09*** | ***3.18*** | ***6.27*** | ***885*** | ***297*** |

***Outdoor***

| | Genotypes | Root dw (g) | Stem dw (g) | Leaf dw (g) | Plt dw (g) | LA (cm2) | SLA (cm2/g) |
|---|---|---|---|---|---|---|---|
| **Drought sensitive** | Bambey 21 | 1.26±0.05 cd | 1.01±0.04 c | 1.19±0.06 c | 2.20±0.06 b | 334±18.55 a | 280±24.08 a |
| | IT82E-18 | 1.53±0.04 c | 1.11±0.07 b | 1.40±0.09 b | 2.52±0.08 b | 388±16.99 a | 278±13.15 a |
| | IT83D-442 | 1.39±0.05 c | 1.01±0.06 c | 1.59±0.06 b | 2.60±0.07 b | 328±29.72 a | 206±21.46 c |
| | IT89KD-288 | 1.44±0.07 c | 1.40±0.05 a | 1.53±0.04 b | 2.93±0.05 b | 248±23.84 bc | 162±12.84 d |
| | IT93K-93-10 | 1.17±0.10 d | 1.00±0.06 c | 1.45±0.08 b | 2.45±0.20 b | 347±27.53 a | 240±12.69 b |
| | IT97K-556-6 | 0.99±0.06 e | 1.11±0.06 b | 1.17±0.05 c | 2.28±0.10 b | 229±20.97 c | 197±14.63 c |
| | UC - CB46 | 1.68±0.07 b | 1.27±0.06 ab | 2.17±0.08 a | 3.43±0.10 a | 367±12.31 a | 170±13.44 d |
| **Drought tolerant** | IT84S-2049 | 0.73±0.05 f | 0.76±0.04 d | 0.76±0.04 e | 1.53±0.07 d | 215±14.01 c | 283±11.98 a |
| | IT97K-207-15 | 1.09±0.10 d | 1.14±0.06 b | 1.25±0.11 bc | 2.39±0.11 b | 250±20.98 bc | 200±12.88 c |
| | IT97K-499-39 | 1.84±0.05 a | 1.30±0.05 a | 1.61±0.06 b | 2.91±0.08 b | 362±30.61 a | 224±13.07 b |
| | IT98K-128-2 | 1.08±0.03 d | 1.12±0.04 b | 1.31±0.09 bc | 2.43±0.12 b | 296±18.09 b | 227±13.08 b |
| | IT99K-124-5 | 1.29±0.09 cd | 1.15±0.07 b | 1.52±0.06 b | 2.67±0.06 b | 328±21.72 a | 216±11.79 bc |
| | KVx-61-1 | 1.10±0.06 d | 1.32±0.06 a | 1.46±0.08 b | 2.78±0.08 b | 249±18.34 bc | 170±10.42 d |
| | Mouride | 0.92±0.05 e | 1.03±0.04 c | 0.99±0.04 d | 2.02±0.06 c | 282±14.25 b | 285±19.43 a |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| | SuVita2 | 1.27±0.06 cd | 1.33±0.09 a | 1.52±0.07 b | 2.85±0.08 b | 348±12.45 a | 230±18.64 b |
| | ***Sensitive Mean*** | ***1.35*** | ***1.13*** | ***1.50*** | ***2.63*** | ***320*** | ***219*** |
| | ***Tolerant Mean*** | ***1.17*** | ***1.14*** | ***1.30*** | ***2.45*** | ***288*** | ***229*** |
| | | **Root dw** | **Stem dw** | **Leaf dw** | **Plt dw** | **LA** | **SLA** |
| **Env** | **F Value** | 944.14 | 11175.20 | 11513.60 | 33483.70 | 14623.80 | 599.36 |
| | **Pr > F** | < 0.0001 | < 0.0001 | < 0.0001 | < 0.0001 | < 0.0001 | < 0.0001 |
| **Gen** | **F Value** | 122.90 | 113.51 | 242.58 | 470.39 | 200.44 | 55.49 |
| | **Pr > F** | < 0.0001 | < 0.0001 | < 0.0001 | < 0.0001 | < 0.0001 | < 0.0001 |
| **G X E** | **F Value** | 47.78 | 94.55 | 161.03 | 334.36 | 121.45 | 49.67 |
| | **Pr > F** | < 0.0001 | < 0.0001 | < 0.0001 | < 0.0001 | < 0.0001 | < 0.0001 |

**Table 3.** Dry mass of plant parts of cowpeas subjected to WS and WW conditions and harvested at the end of the dry-down experiments in both glasshouse (top) and outdoor (bottom).Values shown with SE are means of five replicated plants per genotype. Lower case letters following means discriminate genotypes for each parameter based on Tukey's method at significance level 0.05. Outputs from the analysis of genotype, water treatment, and genotype x treatment interaction effects on the growth parameters are presented at the bottom of the table.

| ***Glasshouse*** | **Root dw (g)** | | **Stem dw (g)** | | **Leaf dw (g)** | | **Plt dw (g)** | |
|---|---|---|---|---|---|---|---|---|
| **Genotypes** | WW | WS | WW | WS | WW | WS | WW | WS |
| Bambey 21 | 3.18±0.11 d | 2.30±0.18 b | 6.86±0.10 c | 4.49±0.14 b | 4.64±0.19 e | 4.61±0.16 b | 11.50±0.13 f | 9.10±0.17 c |
| IT82E-18 | 4.15±0.16 b | 3.11±0.18 a | 6.65±0.12 c | 4.61±0.20 b | 4.74±0.18 e | 4.64±0.11 b | 11.39±0.15 f | 9.25±0.19 c |
| IT83D-442 | 5.19±0.18 a | 3.50±0.21 a | 7.26±0.17 b | 4.50±0.15 b | 6.44±0.14 b | 4.59±0.15 b | 13.70±0.22 c | 9.10±0.20 c |
| IT89KD-288 | 4.64±0.19 b | 3.22±0.11 a | 7.16±0.17 b | 4.35±0.21 b | 5.21±0.11 d | 4.16±0.16 bc | 12.37±0.16 e | 8.51±0.23 d |
| IT93K-93-10 | 4.18±0.17 b | 3.73±0.17 a | 8.69±0.12 a | 6.43±0.17 a | 6.43±0.18 b | 4.82±0.10 b | 15.11±0.17 b | 11.26±0.20 a |
| IT97K-556-6 | 3.61±0.13 c | 2.65±0.10 b | 8.69±0.13 a | 5.06±0.12 ab | 7.34±0.15 a | 5.29±0.15 ab | 16.03±0.12 a | 10.35±0.10 b |
| UC - CB46 | 2.34±0.13 e | 2.17±0.10 b | 5.77±0.24 d | 4.92±0.15 ab | 4.83±0.13 e | 3.76±0.18 c | 10.59±0.30 g | 8.68±0.18 d |
| IT84S-2049 | 3.19±0.15 d | 3.16±0.15 a | 3.42±0.15 e | 3.39±0.17 c | 3.51±0.19 g | 3.40±0.17 c | 6.93±0.16 i | 6.79±0.14 e |
| IT97K-207-15 | 2.70±0.17 e | 2.50±0.15 b | 7.44±0.14 b | 4.82±0.18 ab | 5.40±0.14 d | 4.55±0.13 b | 12.83±0.11 d | 9.37±0.12 c |
| IT97K-499-39 | 4.18±0.18 b | 2.76±0.18 b | 8.53±0.17 a | 4.66±0.18 b | 7.33±0.15 a | 5.79±0.12 a | 15.86±0.22 a | 10.44±0.20 b |
| IT98K-128-2 | 3.47±0.18 c | 2.78±0.16 b | 7.16±0.10 b | 4.36±0.15 b | 5.92±0.18 c | 3.85±0.10 c | 13.08±0.18 d | 8.22±0.12 d |
| IT99K-124-5 | 3.57±0.18 c | 2.62±0.16 b | 5.66±0.11 d | 4.49±0.16 b | 5.49±0.19 d | 3.82±0.13 c | 11.15±0.14 f | 8.32±0.19 d |
| KVx-61-1 | 2.87±0.19 e | 2.51±0.18 b | 6.61±0.14 c | 4.49±0.18 b | 4.72±0.11 e | 4.12±01.0 bc | 11.33±0.18 f | 8.62±0.22 d |
| Mouride | 3.71±0.15 c | 3.10±0.14 a | 5.79±0.12 d | 4.39±0.18 b | 3.89±0.17 f | 3.78±0.11 c | 9.57±0.17 h | 8.28±0.16 d |
| SuVita2 | 2.41±0.21 e | 2.39±0.16 b | 5.57±0.10 d | 4.60±0.19 b | 4.76±0.18 e | 4.70±0.16 b | 10.33±0.24 g | 9.30±0.17 c |
| ***nsitive Mean*** | ***3.90*** | ***2.96*** | ***7.30*** | ***4.91*** | ***5.66*** | ***4.55*** | ***12.96*** | ***9.46*** |
| ***lerant Mean*** | ***3.26*** | ***2.73*** | ***6.27*** | ***4.40*** | ***5.13*** | ***4.25*** | ***11.39*** | ***8.67*** |
| | **Root dw** | | **Stem dw** | | **Leaf dw** | | **Plt dw** | |
| | **F Value** | **Pr > F** | **F Value** | **Pr > F** | **F Value** | **Pr > F** | **F Value** | **Pr > F** |
| **GENOT** | 248.17 | < 0.0001 | 544.22 | < 0.0001 | 556.54 | < 0.0001 | 1165.44 | < 0.0001 |
| **TRT** | 1329.36 | < 0.0001 | 10213.80 | < 0.0001 | 2956.41 | < 0.0001 | 14515.10 | < 0.0001 |
| **GENOT X TRT** | 48.17 | < 0.0001 | 147.52 | < 0.0001 | 116.20 | < 0.0001 | 238.48 | < 0.0001 |

| ***Outdoor*** | **Root dw (g)** | | **Stem dw (g)** | | **Leaf dw (g)** | | **Plt dw (g)** | |
|---|---|---|---|---|---|---|---|---|
| **Genotypes** | WW | WS | WW | WS | WW | WS | WW | WS |
| Bambey 21 | 1.88±0.14 b | 1.66±0.16 b | 1.87±0.15 b | 1.33±0.16 ab | 1.96±0.13 c | 1.46±0.15 cd | 3.83±0.17 b | 2.79±0.17 b |
| IT82E-18 | 2.12±0.10 ab | 1.85±0.17 ab | 1.79±0.16 b | 1.50±0.18 ab | 2.19±0.14 b | 1.87±0.12 b | 3.98±0.18 b | 3.38±0.09 a |
| IT83D-442 | 2.63±0.16 a | 2.30±0.16 a | 1.61±0.18 c | 1.29±0.10 b | 2.11±0.16 b | 1.89±0.16 b | 3.72±0.11 c | 3.15±0.16 b |
| IT89KD-288 | 2.44±0.12 a | 1.78±0.14 b | 1.72±0.16 c | 1.60±0.17 a | 1.88±0.15 c | 1.79±0.17 b | 3.60±0.15 c | 3.39±0.13 a |
| IT93K-93-10 | 1.99±0.17 b | 1.72±0.15 b | 1.73±0.19 c | 1.14±0.15 bc | 2.21±0.15 b | 1.59±0.13 c | 3.94±0.14 b | 2.73±0.17 b |
| IT97K-556-6 | 1.80±0.14 b | 1.47±0.13 b | 1.51±0.16 c | 1.26±0.18 b | 1.88±0.15 c | 1.46±0.14 cd | 3.39±0.10 c | 2.72±0.10 b |
| UC - CB46 | 2.51±0.14 a | 1.91±0.17 ab | 2.22±0.17 a | 1.56±0.15 ab | 2.80±0.10 a | 2.31±0.14 a | 4.88±0.15 a | 3.89±0.14 a |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| IT84S-2049 | 1.54±0.12 b | 1.27±0.18 c | 1.17±0.13 d | 1.07±0.10 c | 1.53±0.13 d | 1.21±0.14 d | 2.71±0.12 d | 2.28±0.08 c |
| IT97K-207-15 | 1.78±0.13 b | 1.49±0.18 b | 1.54±0.13 c | 1.35±0.12 ab | 1.95±0.20 c | 1.61±0.14 c | 3.49±0.10 c | 2.96±0.19 b |
| IT97K-499-39 | 2.69±0.15 a | 2.14±0.13 a | 1.83±0.14 b | 1.56±0.12 ab | 2.68±0.17 a | 1.83±0.17 b | 4.52±0.14 ab | 3.39±0.16 a |
| IT98K-128-2 | 1.47±0.12 b | 1.24±0.14 c | 1.61±0.10 c | 1.32±0.11 b | 1.90±0.13 c | 1.53±0.13 c | 3.51±0.10 c | 2.85±0.11 b |
| IT99K-124-5 | 1.98±0.11 b | 1.76±0.13 b | 1.87±0.16 b | 1.45±0.15 ab | 2.28±0.15 b | 1.65±0.13 c | 4.15±0.11 b | 3.10±0.14 b |
| KVx-61-1 | 1.84±0.18 b | 1.31±0.18 bc | 1.66±0.12 c | 1.48±0.15 ab | 1.86±0.13 c | 1.64±0.16 c | 3.51±0.14 c | 3.12±0.14 b |
| Mouride | 1.66±0.13 b | 1.46±0.12 b | 1.95±0.16 b | 1.69±0.18 a | 2.19±0.16 b | 1.78±0.15 b | 4.13±0.19 b | 3.47±0.15 a |
| SuVita2 | 2.03±0.14 ab | 1.53±0.18 b | 1.90±0.13 b | 1.80±0.16 a | 2.09±0.18 b | 1.93±0.11 b | 3.99±0.17 b | 3.73±0.15 a |
| ***nsitive Mean*** | ***2.20*** | ***1.81*** | ***1.78*** | ***1.38*** | ***2.15*** | ***1.77*** | ***3.91*** | ***3.15*** |
| ***lerant Mean*** | ***1.87*** | ***1.52*** | ***1.69*** | ***1.47*** | ***2.06*** | ***1.65*** | ***3.75*** | ***3.11*** |

| | **Root dw** | | **Stem dw** | | **Leaf dw** | | **Plt dw** | |
|---|---|---|---|---|---|---|---|---|
| | **F Value** | **Pr > F** | **F Value** | **Pr > F** | **F Value** | **Pr > F** | **F Value** | **Pr > F** |
| **GENOT** | 308.35 | < 0.0001 | 51.11 | < 0.0001 | 251.84 | < 0.0001 | 298.91 | < 0.0001 |
| **TRT** | 1335.75 | < 0.0001 | 3997.19 | < 0.0001 | 1986.66 | < 0.0001 | 2645.69 | < 0.0001 |
| **GENOT X TRT** | 16.41 | < 0.0001 | 12.23 | < 0.0001 | 33.40 | < 0.0001 | 44.44 | < 0.0001 |

**Table 4.** Biomass increase (g per plant), total water transpired (g per plant), and transpiration efficiency (g biomass $kg^{-1}$ water transpired) of cowpea genotypes subjected to well watered (control) and progressive and controlled drought stress during the dry-down experiements in glasshouse (top) and outdoor (bottom) environements. Values shown with means are SE of five replicated plants for each genotype. Genotypes means followed with the same letter are not significantly different based on Tukey's test at significance level 0.05. Outputs from the analysis of genotype, environment, and genotype x environment interaction effects on the different growth parameters are presented at the bottom of the table.

| | ***Glasshouse*** | **Biomass increased (g)** | | **Total water transpired (g)** | | **Transpiration efficiency (g $kg^{-1}$)** | |
|---|---|---|---|---|---|---|---|
| | **Genotypes** | **Well watered** | **Water stress** | **Well watered** | **Water stress** | **Well watered** | **Water stress** |
| **Drought sensitive** | Bambey 21 | 3.59±0.11 d | 1.18±0.09 e | 1684±20.90 e | 843±14.39 b | 2.13±0.07 b | 1.40±0.11 e |
| | IT82E-18 | 3.68±0.31 d | 1.55±0.09 d | 2073±13.48 c | 647±21.28 d | 2.38±0.16 b | 1.80±0.17 d |
| | IT83D-442 | 6.50±0.23 b | 1.90±0.15 c | 2234±11.12 b | 749±27.89 c | 2.91±0.10 ab | 2.54±0.22 b |
| | IT89KD-288 | 7.55±0.24 a | 2.70±0.29 b | 2183±28.91 c | 967±25.34 ab | 3.46±0.12 a | 2.79±0.08 b |
| | IT93K-93-10 | 6.52±0.20 b | 2.67±0.22 b | 2166±14.54 c | 737±12.56 c | 3.01±0.11 ab | 2.62±0.32 b |
| | IT97K-556-6 | 7.39±0.31 a | 1.71±0.17 d | 2359±34.12 a | 713±23.45 c | 3.13±0.13 a | 2.40±0.27 c |
| | UC - CB46 | 3.35±0.21 d | 1.43±0.07 d | 1878±29.50 d | 751±15.25 c | 1.78±0.11 d | 1.91±0.08 d |
| **Drought tolerant** | IT84S-2049 | 3.57±0.28 d | 3.66±0.09 a | 1524±26.60 e | 1177±20.69 a | 2.34±0.19 b | 3.11±0.08 a |
| | IT97K-207-15 | 4.67±0.19 c | 1.21±0.18 e | 2363±27.83 a | 850±13.04 b | 1.98±0.08 c | 1.42±0.22 e |
| | IT97K-499-39 | 6.34±0.29 b | 0.93±0.07 e | 2210±23.87 b | 771±19.91 c | 2.87±0.13 ab | 1.20±0.09 e |
| | IT98K-128-2 | 6.77±0.21 b | 1.90±0.22 c | 2429±17.13 a | 730±15.03 c | 2.79±0.10 ab | 2.61±0.27 b |
| | IT99K-124-5 | 5.75±0.40 c | 2.91±0.22 b | 2126±28.50 c | 885±17.94 b | 2.70±0.18 ab | 3.30±0.29 a |
| | KVx-61-1 | 3.89±0.18 d | 1.18±0.24 e | 2051±25.37 c | 855±19.33 b | 1.90±0.09 c | 1.37±0.26 e |
| | Mouride | 4.73±0.11 c | 3.44±0.09 a | 1537±36.76 e | 1108±15.34 a | 3.08±0.08 a | 3.10±0.05 a |
| | SuVita2 | 5.00±0.23 c | 2.08±0.08 c | 1515±23.33 e | 1005±19.41 ab | 3.30±0.17 a | 2.07±0.04 c |
| | ***Sensitive Mean*** | ***5.51*** | ***1.88*** | ***2083*** | ***772*** | ***2.69*** | ***2.21*** |
| | ***Tolerant Mean*** | ***5.09*** | ***2.16*** | ***1969*** | ***923*** | ***2.62*** | ***2.27*** |
| | ***Outdoor*** | **Biomass increased (g)** | | **Total water transpired (g)** | | **Transpiration efficiency (g $kg^{-1}$)** | |
| | **Genotypes** | **Well watered** | **Water stress** | **Well watered** | **Water stress** | **Well watered** | **Water stress** |
| **Drought sensitive** | Bambey 21 | 1.63± 0.12 b | 0.59±0.07 c | 1516±17.48 c | 717±14.34 b | 1.08±0.08 bc | 0.82±0.01 d |
| | IT82E-18 | 1.46±0.06 b | 0.56±0.08 c | 1924±19.73 a | 707±15.18 b | 0.76±0.04 d | 0.79±0.12 d |
| | IT83D-442 | 1.12±0.07 c | 0.56±0.03 c | 1382±18.75 d | 630±16.26 c | 0.81±0.05 c | 0.88±0.05 d |
| | IT89KD-288 | 0.67±0.07 d | 0.45±0.04 d | 1449±14.16 d | 560±18.80 d | 0.46±0.04 f | 0.81±0.07 d |
| | IT93K-93-10 | 1.49±0.08 b | 0.27±0.04 f | 1485±25.47 c | 546±23.92 d | 1.00±0.05 bc | 0.50±0.06 g |
| | IT97K-556-6 | 1.11±0.05 c | 0.44±0.04 d | 1549±28.95 c | 603±15.79 c | 0.71±0.04 d | 0.73±0.04 e |
| | UC - CB46 | 1.45±0.06 b | 0.46±0.05 d | 1742±26.80 b | 644±15.85 c | 0.83±0.03 c | 0.71±0.06 e |
| **Drought tolerant** | IT84S-2049 | 1.28±0.08 c | 0.85±0.05 b | 1142±13.70 e | 894±13.35 a | 1.12±0.07 b | 1.55±0.03 ab |
| | IT97K-207-15 | 1.10±0.10 c | 0.57±0.03 c | 1367±19.78 d | 564±14.02 d | 0.81±0.07 c | 1.01±0.04 c |
| | IT97K-499-39 | 1.61±0.11 b | 0.48±0.05 d | 1338±16.91 d | 707±14.90 b | 1.20±0.10 b | 0.68±0.03 e |
| | IT98K-128-2 | 1.08±0.05 c | 0.43±0.05 d | 1363±11.33 d | 608±14.97 c | 0.80±0.04 c | 0.70±0.03 e |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| | IT99K-124-5 | 1.48±0.10 b | 0.43±0.03 d | 1189±12.18 e | 628±18.28 c | 1.24±0.08 b | 0.69±0.02 e |
| | KVx-61-1 | 0.73±0.05 d | 0.34±0.04 e | 1328±23.24 d | 556±15.77 d | 0.55±0.03 e | 0.61±0.04 f |
| | Mouride | 2.11±0.13 a | 1.45±0.06 a | 1013±17.99 f | 824±19.88 a | 2.08±0.11 a | 1.75±0.08 a |
| | SuVita2 | 1.05±0.10 c | 0.98±0.09 b | 1055±16.33 f | 811±17.12 a | 1.99±0.09 a | 1.21±0.10 b |
| | ***Sensitive Mean*** | ***1.28*** | ***0.48*** | ***1578*** | ***629*** | ***0.81*** | ***0.75*** |
| | ***Tolerant Mean*** | ***1.31*** | ***0.69*** | ***1224*** | ***699*** | ***1.10*** | ***0.95*** |
| | | **Biomass increased** | | **Total water transpired** | | **Transpiration efficiency** | |
| | | **Well watered** | **Water stress** | **Well watered** | **Water stress** | **Well watered** | **Water stress** |
| **Env** | **F Value** | 17759.30 | 5644.42 | 70707.80 | 7838.01 | 10031.50 | 4163.08 |
| | **Pr > F** | < 0.0001 | < 0.0001 | < 0.0001 | < 0.0001 | < 0.0001 | < 0.0001 |
| **Gen** | **F Value** | 150.32 | 204.50 | 2613.57 | 830.49 | 138.50 | 106.57 |
| | **Pr > F** | < 0.0001 | < 0.0001 | < 0.0001 | < 0.0001 | < 0.0001 | < 0.0001 |
| **G X E** | **F Value** | 198.43 | 134.25 | 1153.12 | 156.20 | 96.97 | 95.95 |
| | **Pr > F** | < 0.0001 | < 0.0001 | < 0.0001 | < 0.0001 | < 0.0001 | < 0.0001 |

**Table 5.** FTSW threshold values for the fifteen cowpea genotypes grown under progressive drying soil in glasshouse (A) and outdoor (B) conditions. FTSW thresholds were calculated using the two-segment plateau regression procedure with ± standard error (SE) and confidence interval (CI). Data are the means of five replicates plants for each genotype. FTSW Threshold values identified with the same letter are not statistically varied from each other based on Tukey's test at significance level 0.05.

| **(A)** | **Genotypes** | **FTSW Threshold** | **Approximate SE** | **95% CI** |
|---|---|---|---|---|
| **Drought sensitive** | Bambey21 | 0.6319 c | 0.0341 | 0.6036 - 0.6702 |
| | IT82E-18 | 0.6234 c | 0.0365 | 0.6062 - 0.6727 |
| | IT83D-442 | 0.6788 d | 0.0553 | 0.6458 - 0.716 |
| | IT89KD-288 | 0.6201 c | 0.0326 | 0.5048 - 0.6555 |
| | IT93K-93-10 | 0.6972 d | 0.0289 | 0.6492 - 0.7452 |
| | IT97K-556-6 | 0.6217 c | 0.0417 | 0.5780 - 0.6654 |
| | UC-CB46 | 0.6275 c | 0.0462 | 0.6047 - 0.6702 |
| **Drought tolerant** | IT84S-2049 | 0.4730 a | 0.0133 | 0.4466 - 0.4999 |
| | IT97K-207-15 | 0.5274 b | 0.0495 | 0.4882 - 0.5667 |
| | IT97K-499-39 | 0.5679 b | 0.0307 | 0.5362 - 0.6095 |
| | IT98K-128-2 | 0.5923 b | 0.0312 | 0.5797 - 0.6349 |
| | IT99K-124-5 | 0.5247 b | 0.0303 | 0.4840 - 0.5655 |
| | KVx-61-1 | 0.5904 b | 0.0355 | 0.5692 - 0.6215 |
| | Mouride | 0.4449 a | 0.0232 | 0.4186 - 0.4715 |
| | Suvita2 | 0.4765 a | 0.0240 | 0.4368 - 0.5032 |

| **(B)** | **Genotypes** | **FTSW Threshold** | **Approximate SE** | **95% CI** |
|---|---|---|---|---|
| **Drought sensitive** | Bambey21 | 0.6886 c | 0.0191 | 0.6503 - 0.7168 |
| | IT82E-18 | 0.7129 d | 0.0307 | 0.6813 - 0.7544 |
| | IT83D-442 | 0.6613 c | 0.0128 | 0.6358 - 0.6869 |
| | IT89KD-288 | 0.6650 c | 0.0102 | 0.6447 - 0.6853 |
| | IT93K-93-10 | 0.6864 c | 0.0128 | 0.6608 - 0.7121 |
| | IT97K-556-6 | 0.7227 d | 0.0140 | 0.6956 - 0.7518 |
| | UC-CB46 | 0.6724 c | 0.0160 | 0.6403 - 0.7045 |
| **Drought tolerant** | IT84S-2049 | 0.4920 a | 0.0125 | 0.4670 - 0.5271 |
| | IT97K-207-15 | 0.6596 c | 0.0145 | 0.6310 - 0.6890 |
| | IT97K-499-39 | 0.6092 b | 0.0148 | 0.5896 - 0.6388 |
| | IT98K-128-2 | 0.6180 b | 0.0113 | 0.5953 - 0.6307 |
| | IT99K-124-5 | 0.6353 b | 0.0071 | 0.6012 - 0.6694 |
| | KVx-61-1 | 0.5978 b | 0.0121 | 0.5736 - 0.6219 |
| | Mouride | 0.4821 a | 0.0056 | 0.4310 - 0.5033 |
| | Suvita2 | 0.4817 a | 0.0094 | 0.4630 - 0.5104 |

**Table 6.** Visual scores for stay green of contrasting cowpea genotypes rated under drought stress conditions outdoors and in the glasshouse. This rating was done at the end of the dry-down experiment [45 DAS]. Score values are the means (±SE) of five replicated plants per genotype. Average score of sensitive and tolerant lines are presented at the bottom of the table.

| **Genotypes** | **Glasshouse** | **Outdoor** |
|---|---|---|
| Bambey 21 | 2.0±0.0 b | 3.2±0.4 a |
| IT82E-18 | 3.6±0.5 a | 4.0±0.0 a |
| IT83D-442 | 1.6±0.5 b | 2.0±0.0 b |
| IT89KD-288 | 1.4±0.5 b | 3.2±0.5 a |
| IT93K-93-10 | 1.2±0.4 b | 1.4±0.5 c |
| IT97K-556-6 | 3.2±0.4 a | 4.4±0.5 a |
| UC - CB46 | 3.6±0.5 a | 3.6±0.5 a |
| IT84S-2049 | 1.8±0.4 b | 1.6±0.5 b |
| IT97K-207-15 | 1.8±0.4 b | 1.4±0.5 c |
| IT97K-499-39 | 1.2±0.4 b | 1.2±0.4 c |
| IT98K-128-2 | 1.0±0.0 c | 3.0±0.5 a |
| IT99K-124-5 | 1.4±0.5 b | 2.2±0.4 b |
| KVx-61-1 | 1.6±0.5 b | 1.8±0.4 b |
| Mouride | 1.0±0.0 c | 1.6±0.5 b |
| SuVita2 | 1.0±0.0 c | 1.4±0.5 c |
| ***Sensitive Mean*** | ***2.37*** | ***3.11*** |
| ***Tolerant Mean*** | ***1.35*** | ***1.78*** |

## FIGURES CAPTIONS

**Fig. 1.** Transpiration rate [TR, g $H_2O$ $cm^{-1}$ $2h^{-1}$] under well watered conditions of cowpea genotypes contrasting for terminal drought tolerance [tolerant lines: IT84S-2049, Mouride, Suvita2, solid lines] and [sensitive lines: Bambey 21, IT82E-18, UC-CB46, dotted lines] exposed to natural variation of atmospheric VPD cycle. Plants were grown in glasshouse (A) and outdoors (B) and tested at the vegetative stage over the course of an entire day. Values are the means (±SE) of five plants per genotype. The polynomial dotted line fitting with the dashed points represents the VPD variation during the course of the day of the experiments.

**Fig. 2.** Total water transpired [Tr, g $H_2O$ $Plant^{-1}$ $Day^{-1}$] of drought tolerant (black bars) and sensitive genotypes (grey bars) grown under non-limited water conditions. The amount of water loss was estimated on well watered plants over an entire day in glasshouse (A) and outdoors (B) conditions at vegetative stage. Values are the means (± SE) of five plants for each genotype.

**Fig. 3.** Normalized transpiration rate versus fraction of transpirable soil water of tolerant [Mouride and IT84S-2049, open symbols and solid lines] and sensitive [Bambey and IT82E-18, closed symbols and dotted lines] cowpea genotypes exposed to progressive drying soil under glasshouse (A and B) and outdoor (C and D) conditions. Values are transpiration data of five replicated plants for each genotype at each FTSW condition. The FTSW thresholds where transpiration initiated its decline were calculated with a plateau regression procedure from SAS. Then the regression lines of the relationships between NTR and FTSW were drawn by fitting NTR to FTSW data above and below the respective threshold for transpiration decline in each genotype with GraphPad Prism.

**Fig. 4.** Differences in canopy temperature depression among contrasting cowpea genotypes (Tolerant, black bars; sensitive, grey bars) grown under well watered and water stress conditions in glasshouse (A and B) and outdoor (C and D). Measurements of leaf temperature were done on the three most recent fully expanded leaves at the end of the dry-down between 8:00 and 9:00 am. Values are the means (±SE) of five replicates plants per treatment and genotype.

**Fig. 5.** Relationship between transpiration rate [TR, g $H_2O$ $cm^{-2}$ $h^{-1}$] and transpiration efficiency [TE, g biomass / kg water transpired] of fifteen tolerant and sensitive cowpea genotypes. TR was measured at the highest VPD under well watered [WW] conditions at the vegetative stage and TE was estimated under both WW and water stress [WS] conditions in the glasshouse (A and B) and outdoors (C and D). Data points are means of five replicated plants per genotype and treatment.

**Fig. 6.** Relationship between TR and the FTSW-threshold [glasshouse (A) and outdoors (B)] and between FTSW-threshold and transpiration efficiency [glasshouse (C) and outdoors (D)] of fifteen contrasting cowpea genotypes. Data points are means of five replicated plants per genotype.

**Fig. 7.** Relationship between canopy temperature depression and transpiration rate of fifteen contrasting cowpea genotypes under drought stress condition. CTD and TR were measured at the end of the dry-down experiment in glasshouse (A) and outdoors (B). Data points are means of five replicated plants per genotype.

**Suppl. Fig. 1**. Weather conditions during the experiments in ICRISAT. The data presented were daily minimum and maximum air temperature (A), relative humidity (B), and atmospheric VPD (C) recorded during the measurements period starting from 18$^{th}$ March 2010 (date of water treatments imposition) to 5$^{th}$ May 2010 (date of final harvest). The curves are in dotted lines for the glasshouse [Gh] and in solid lines for outdoors [Od].

**Suppl. Fig. 2.** Tolerant Suvita2 (A) and Mouride (B) and sensitive IT82-18 (C) and UC-CB46 (D) kept well watered (4 plants on the left) or exposed to water stress (4 plants on the right) conditions at the end of the dry-down experiment in the glasshouse. The tolerant genotypes had the lowest stay green scores [Suvita (1.0±0.0) and Mouride (1.0±0.0)] and the sensitive genotypes showed the highest scores [IT82E-18 (3.6±0.5) and UC-CB46 (3.6±0.5)].


# Lower Soil Moisture Threshold for Transpiration Decline under Water Deficit Correlates with Lower Canopy Conductance and Higher Transpiration Efficiency in Drought Tolerant Cowpea

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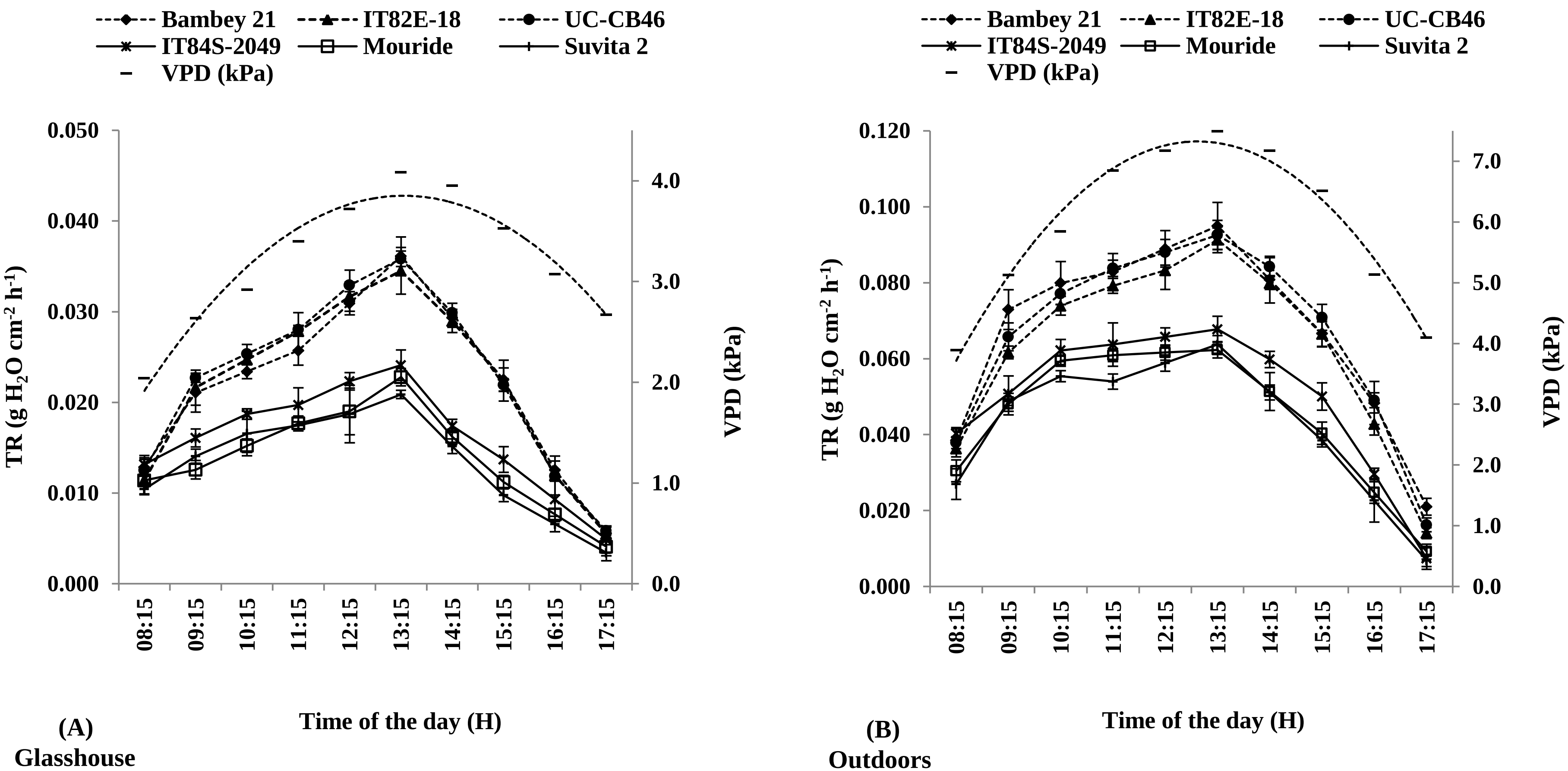
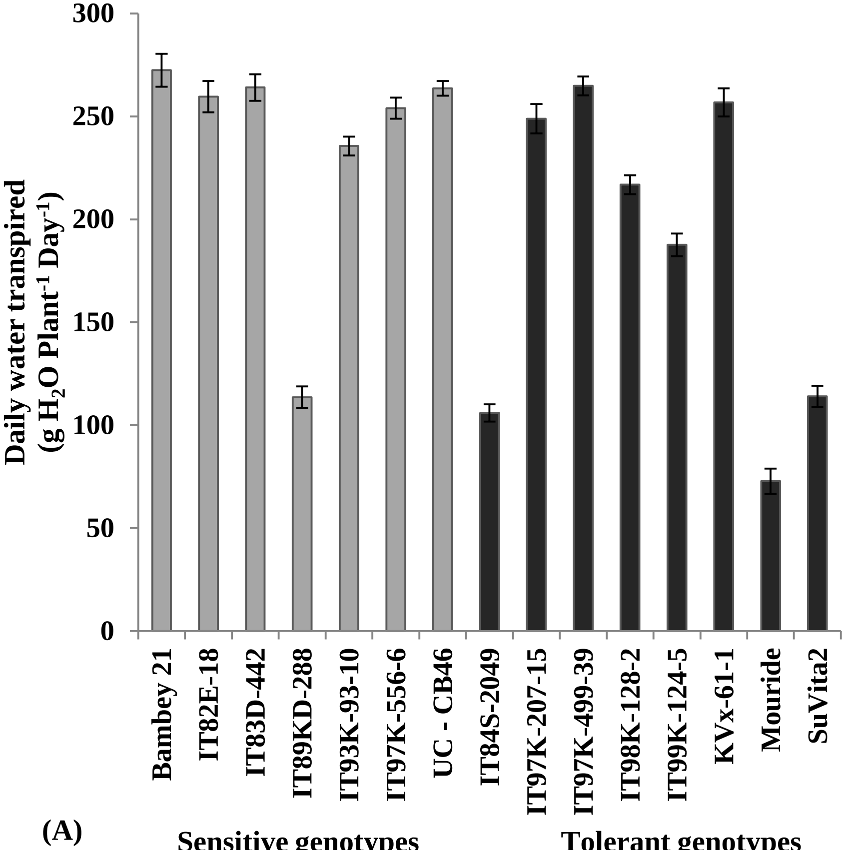
# Figure 1

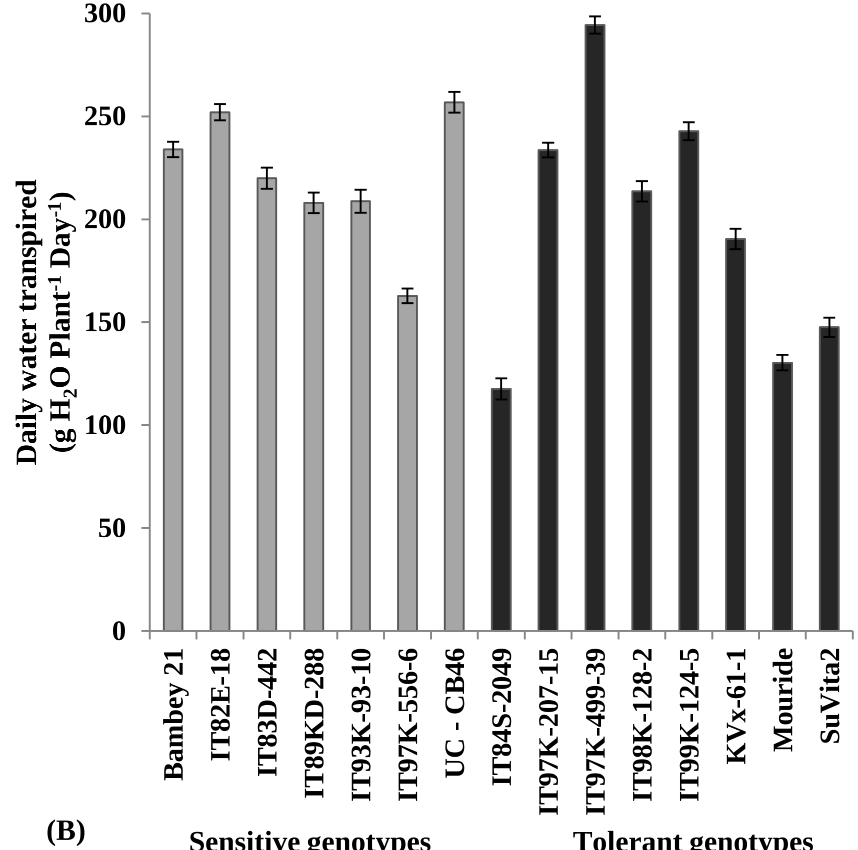


Bambey 21
IT82E-18
UC-CB46
IT84S-2049
Mouride
Suvita 2
VPD (kPa)
0.050
0.040
0.030
0.020
0.010
0.000
4.0
3.0
2.0
1.0
0.0
TR (g $H_2O$ cm$^{-2}$ h$^{-1}$)
VPD (kPa)
08:15
09:15
10:15
11:15
12:15
13:15
14:15
15:15
16:15
17:15
Time of the day (H)
(A)
Glasshouse
Bambey 21
IT82E-18
UC-CB46
IT84S-2049
Mouride
Suvita 2
VPD (kPa)
0.120
0.100
0.080
0.060
0.040
0.020
0.000
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
TR (g $H_2O$ cm$^{-2}$ h$^{-1}$)
VPD (kPa)
08:15
09:15
10:15
11:15
12:15
13:15
14:15
15:15
16:15
17:15
Time of the day (H)
(B)
Outdoors

# Figure 2

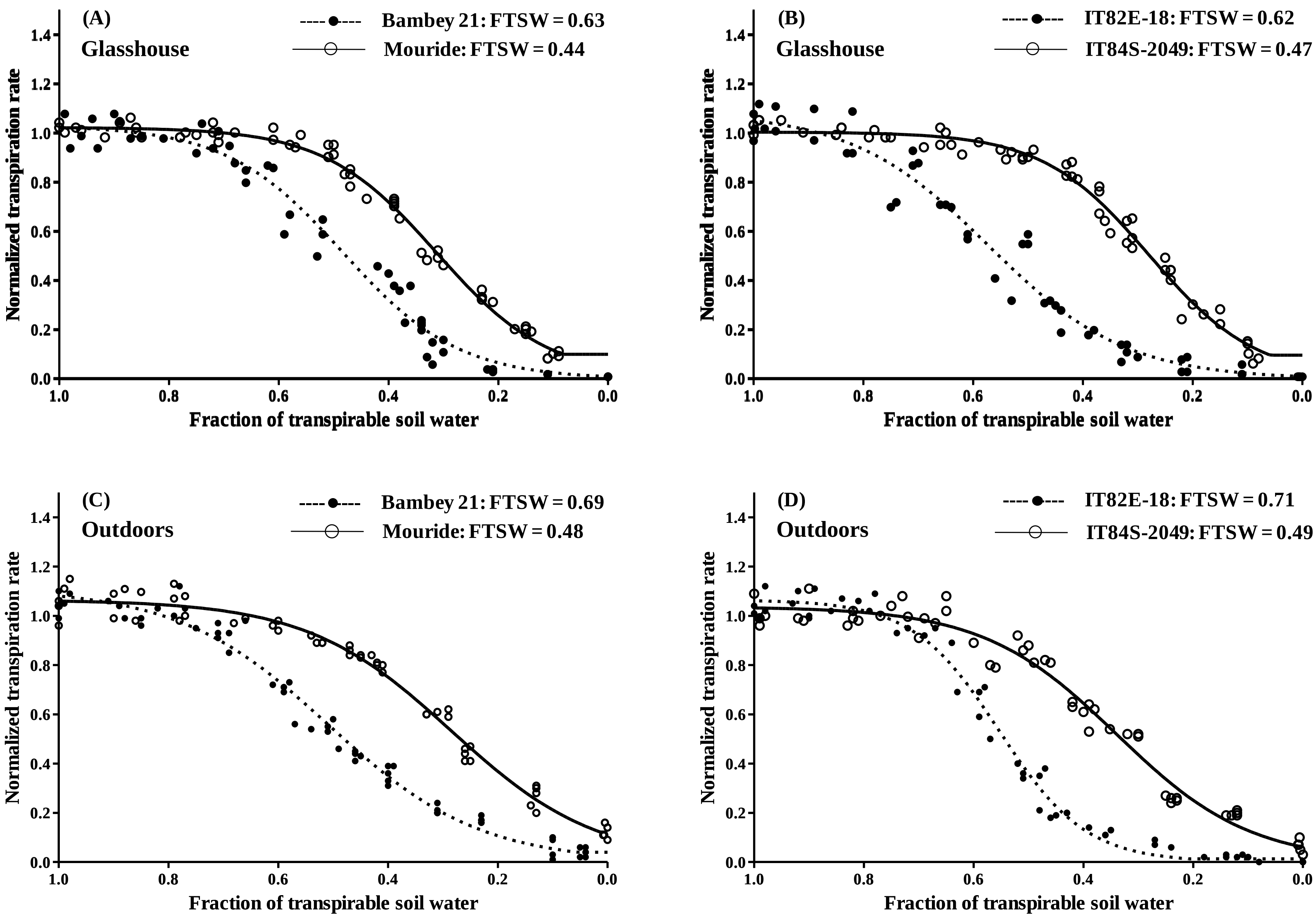



(A)
Glasshouse



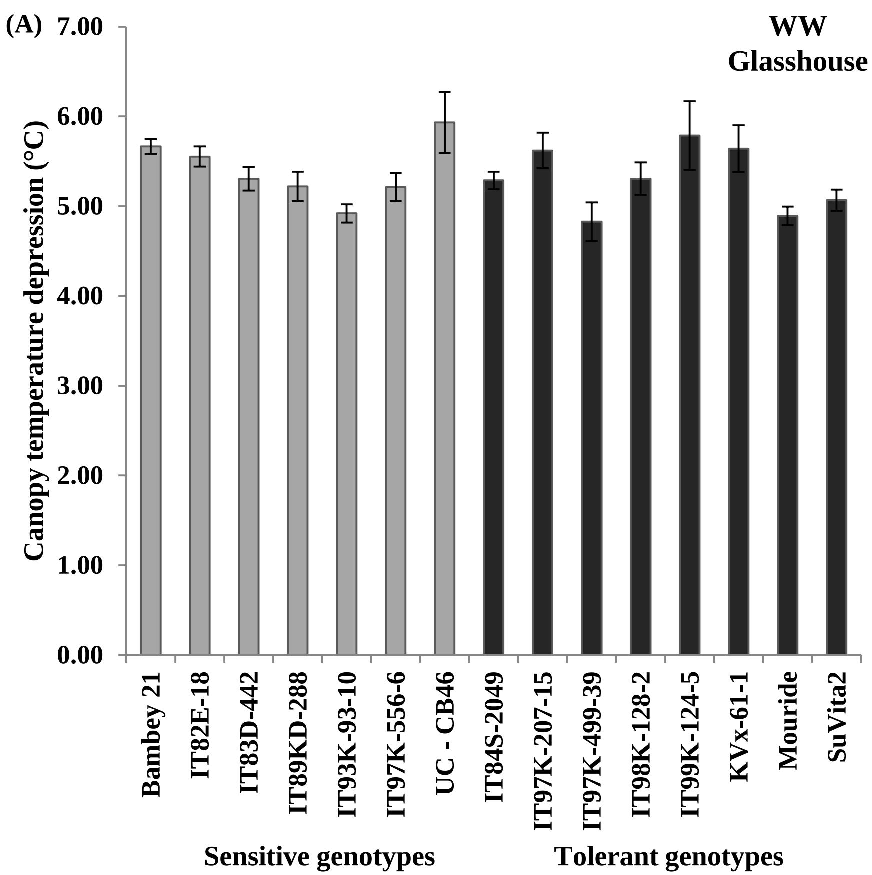

(B)
Outdoors

# Figure 3

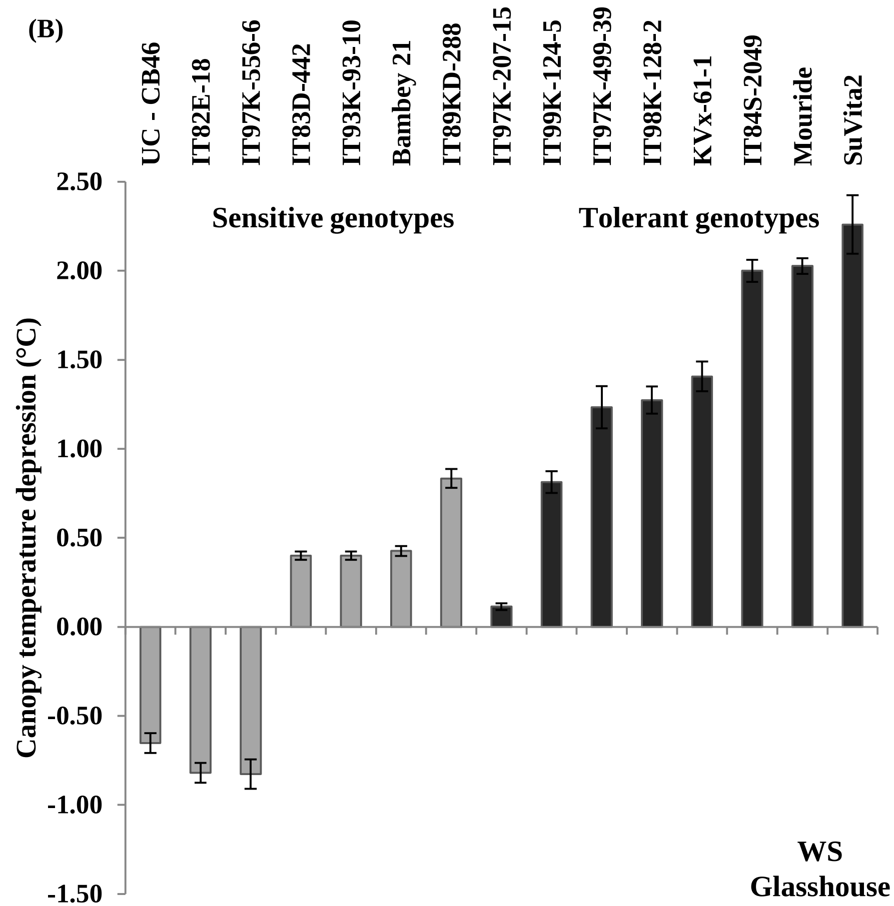


(A)
Glasshouse
Bambey 21: FTSW = 0.63
Mouride: FTSW = 0.44
(B)
Glasshouse
IT82E-18: FTSW = 0.62
IT84S-2049: FTSW = 0.47
(C)
Outdoors
Bambey 21: FTSW = 0.69
Mouride: FTSW = 0.48
(D)
Outdoors
IT82E-18: FTSW = 0.71
IT84S-2049: FTSW = 0.49
Normalized transpiration rate
Fraction of transpirable soil water

# Figure 4 A&B

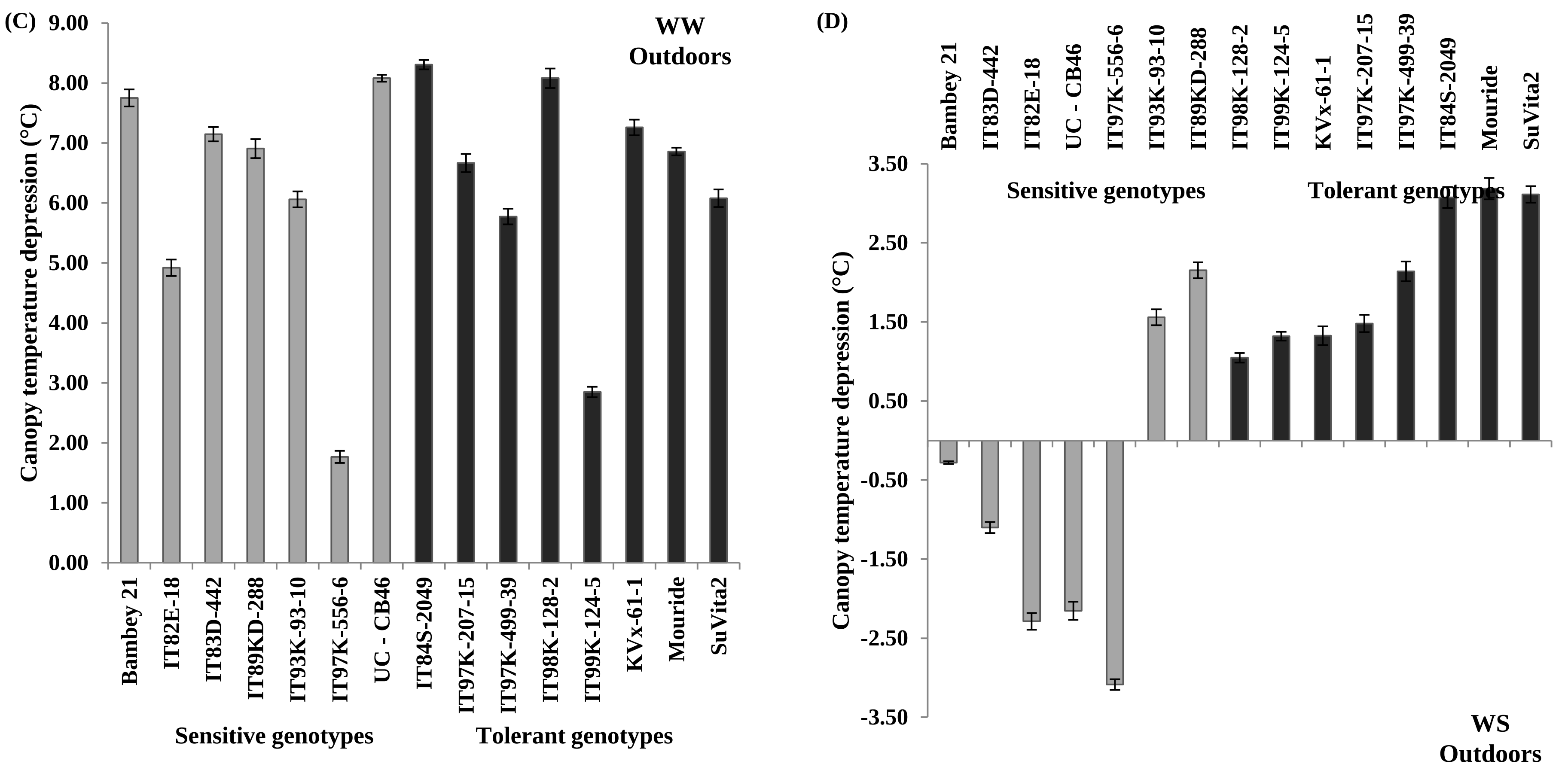


(A)
WW
Glasshouse
Canopy temperature depression (°C)
7.00
6.00
5.00
4.00
3.00
2.00
1.00
0.00
Bambey 21
IT82E-18
IT83D-442
IT89KD-288
IT93K-93-10
IT97K-556-6
UC - CB46
IT84S-2049
IT97K-207-15
IT97K-499-39
IT98K-128-2
IT99K-124-5
KVx-61-1
Mouride
SuVita2
Sensitive genotypes
Tolerant genotypes

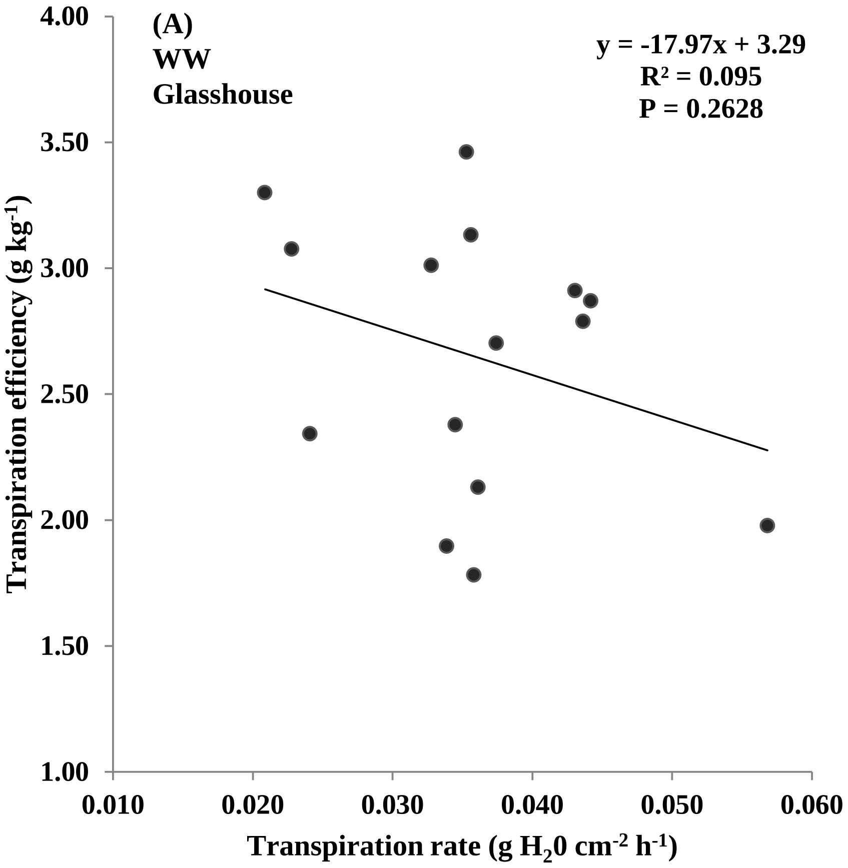



(B)
UC - CB46
IT82E-18
IT97K-556-6
IT83D-442
IT93K-93-10
Bambey 21
IT89KD-288
IT97K-207-15
IT99K-124-5
IT97K-499-39
IT98K-128-2
KVx-61-1
IT84S-2049
Mouride
SuVita2
Sensitive genotypes
Tolerant genotypes
Canopy temperature depression (°C)
2.50
2.00
1.50
1.00
0.50
0.00
-0.50
-1.00
-1.50
WS
Glasshouse

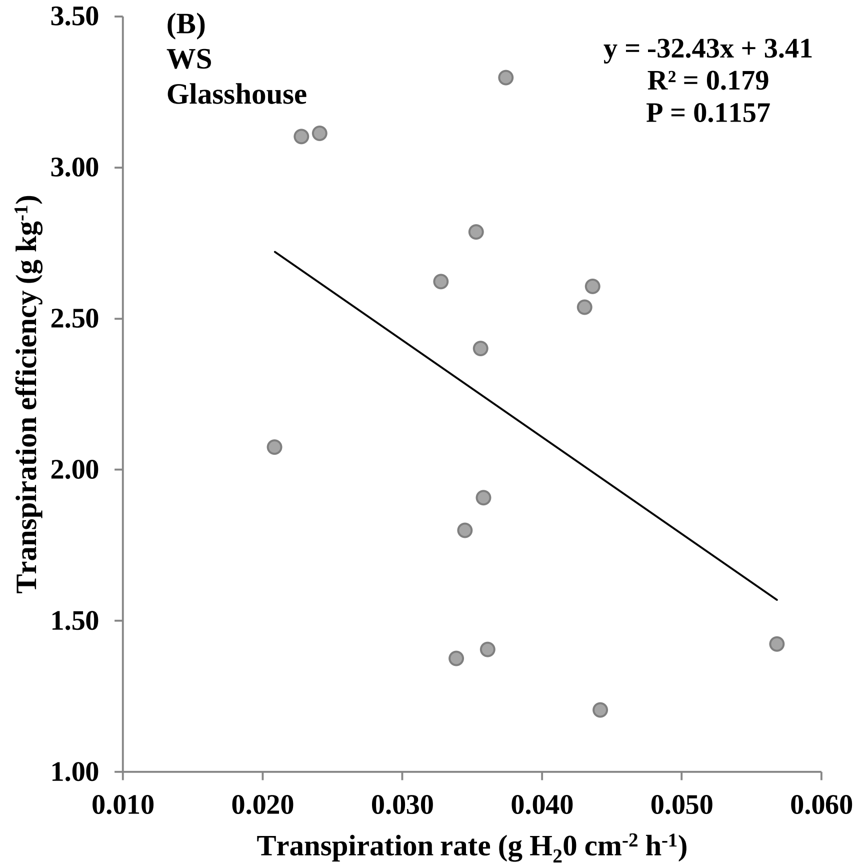
# Figure 4 C&D


(C)
WW
Outdoors
Canopy temperature depression (°C)
9.00
8.00
7.00
6.00
5.00
4.00
3.00
2.00
1.00
0.00
Bambey 21
IT82E-18
IT83D-442
IT89KD-288
IT93K-93-10
IT97K-556-6
UC - CB46
IT84S-2049
IT97K-207-15
IT97K-499-39
IT98K-128-2
IT99K-124-5
KVx-61-1
Mouride
SuVita2
Sensitive genotypes
Tolerant genotypes
(D)
Bambey 21
IT83D-442
IT82E-18
UC - CB46
IT97K-556-6
IT93K-93-10
IT89KD-288
IT98K-128-2
IT99K-124-5
KVx-61-1
IT97K-207-15
IT97K-499-39
IT84S-2049
Mouride
SuVita2
Sensitive genotypes
Tolerant genotypes
Canopy temperature depression (°C)
3.50
2.50
1.50
0.50
-0.50
-1.50
-2.50
-3.50
WS
Outdoors

# Figure 5 A&B

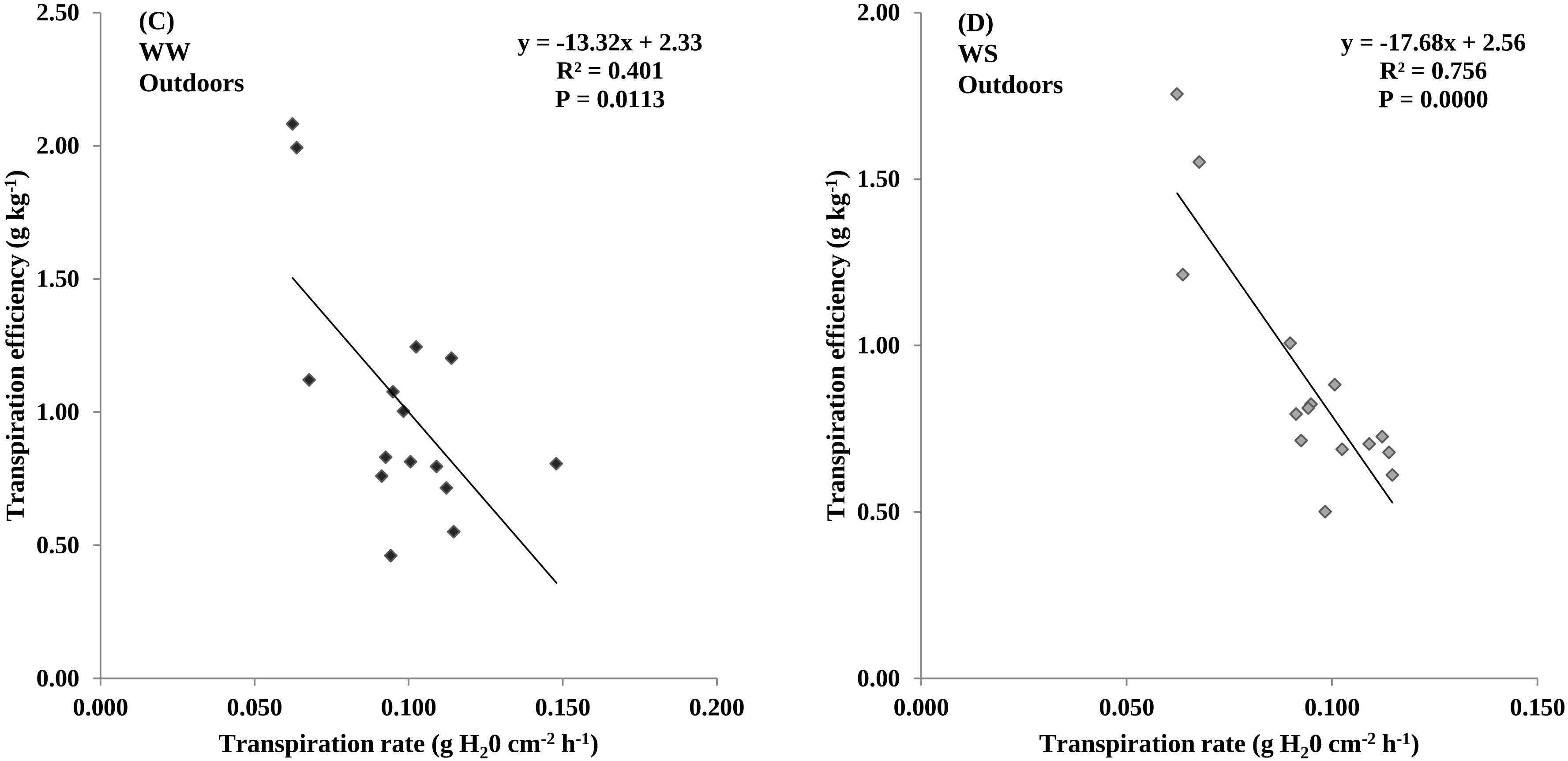


(A)
WW
Glasshouse
y = -17.97x + 3.29
R² = 0.095
P = 0.2628
Transpiration efficiency (g kg-1)
Transpiration rate (g H20 cm-2 h-1)
4.00
3.50
3.00
2.50
2.00
1.50
1.00
0.010
0.020
0.030
0.040
0.050
0.060

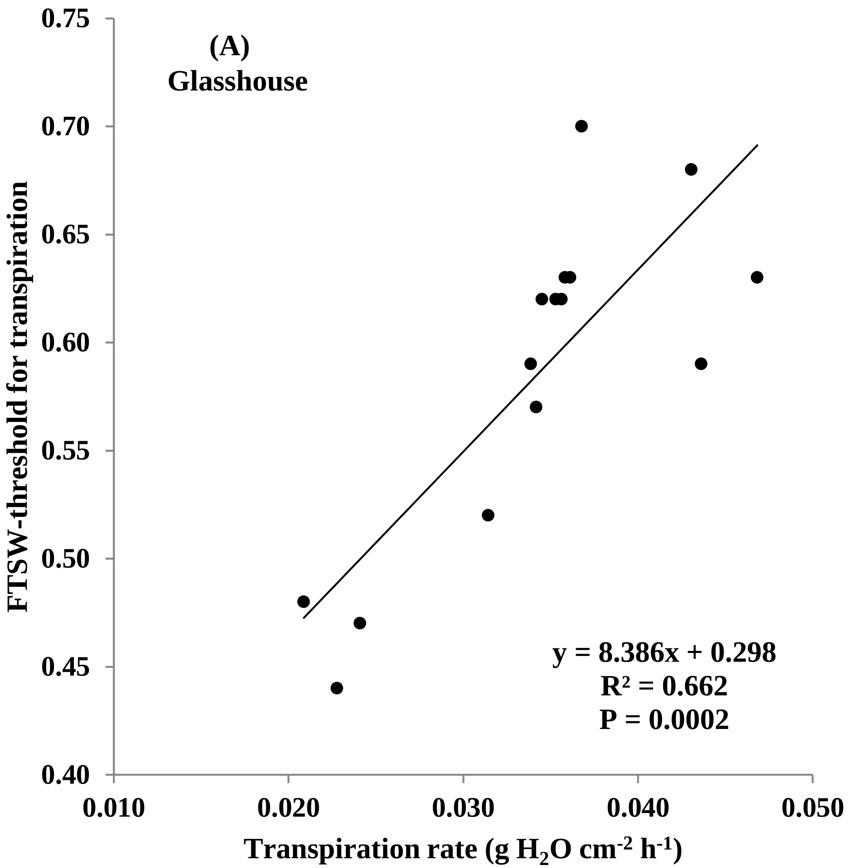
(B)
WS
Glasshouse
y = -32.43x + 3.41
R² = 0.179
P = 0.1157
Transpiration efficiency (g kg-1)
Transpiration rate (g H20 cm-2 h-1)
3.50
3.00
2.50
2.00
1.50
1.00
0.010
0.020
0.030
0.040
0.050
0.060

**Figure 5 C&D**

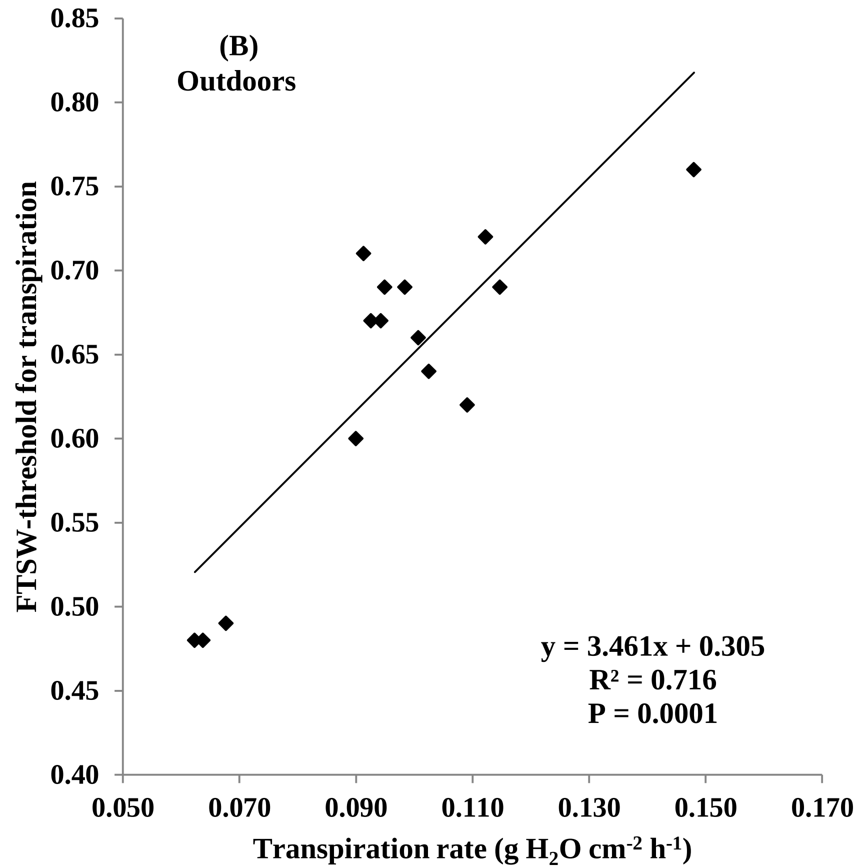


(C)
WW
Outdoors
y = -13.32x + 2.33
R² = 0.401
P = 0.0113
Transpiration efficiency (g kg-1)
Transpiration rate (g H20 cm-2 h-1)
2.50
2.00
1.50
1.00
0.50
0.00
0.000
0.050
0.100
0.150
0.200
(D)
WS
Outdoors
y = -17.68x + 2.56
R² = 0.756
P = 0.0000
Transpiration efficiency (g kg-1)
Transpiration rate (g H20 cm-2 h-1)
2.00
1.50
1.00
0.50
0.00
0.000
0.050
0.100
0.150

# Figure 6 A&B

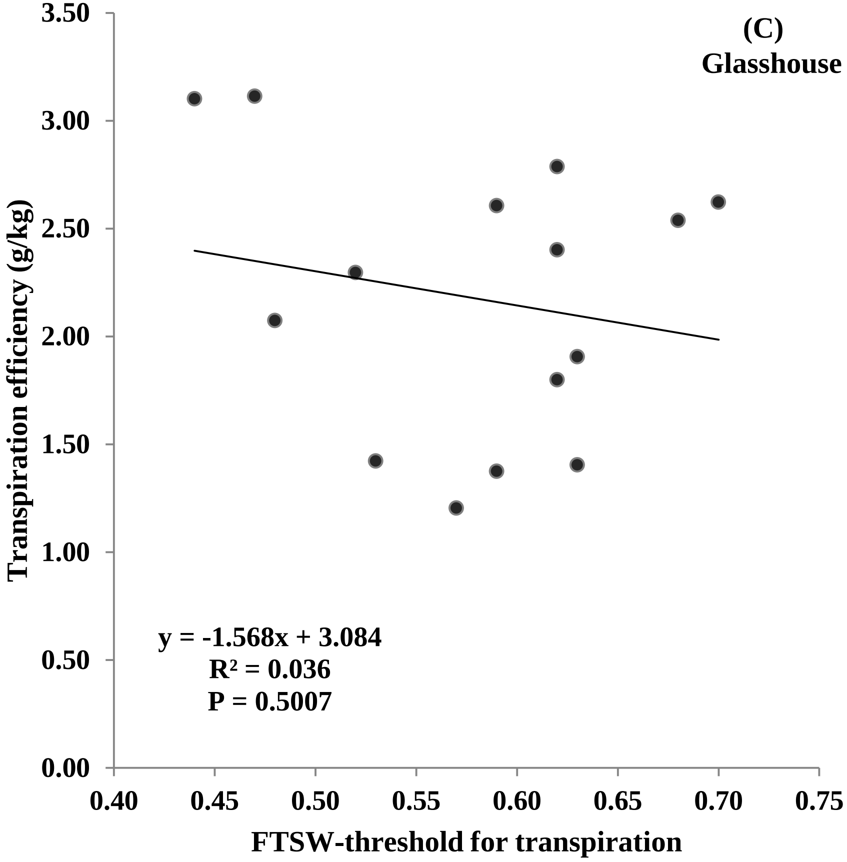


(A)
Glasshouse
y = 8.386x + 0.298
R² = 0.662
P = 0.0002
FTSW-threshold for transpiration
Transpiration rate (g H2O cm-2 h-1)
0.75
0.70
0.65
0.60
0.55
0.50
0.45
0.40
0.010
0.020
0.030
0.040
0.050

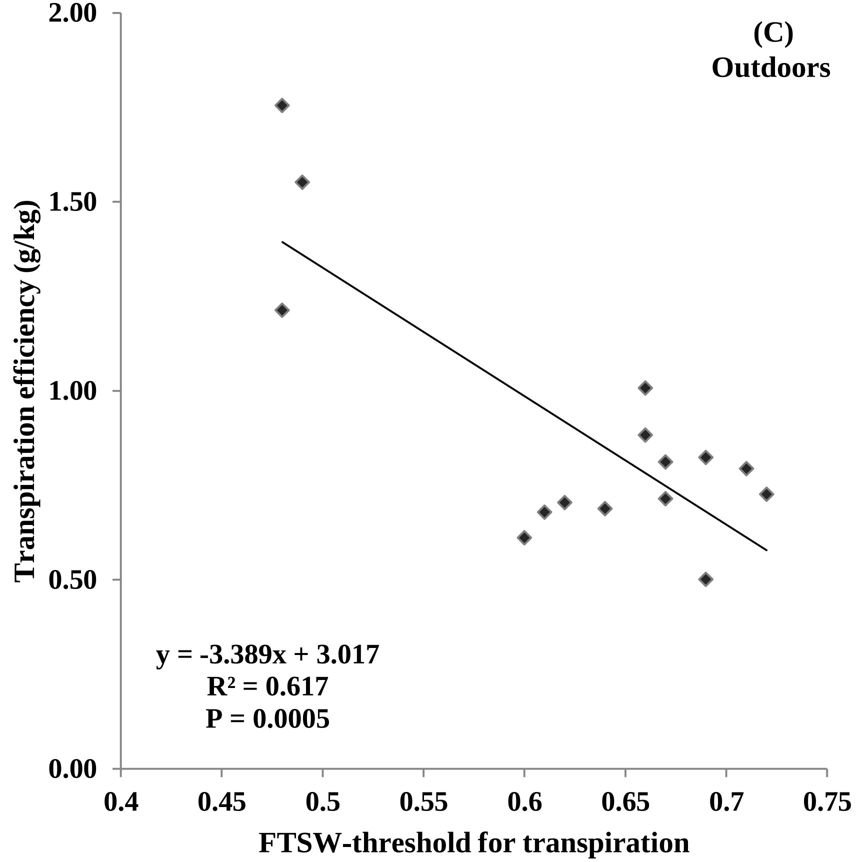



(B)
Outdoors
y = 3.461x + 0.305
R² = 0.716
P = 0.0001
FTSW-threshold for transpiration
Transpiration rate (g H2O cm-2 h-1)
0.85
0.80
0.75
0.70
0.65
0.60
0.55
0.50
0.45
0.40
0.050
0.070
0.090
0.110
0.130
0.150
0.170

# Figure 6 C&D

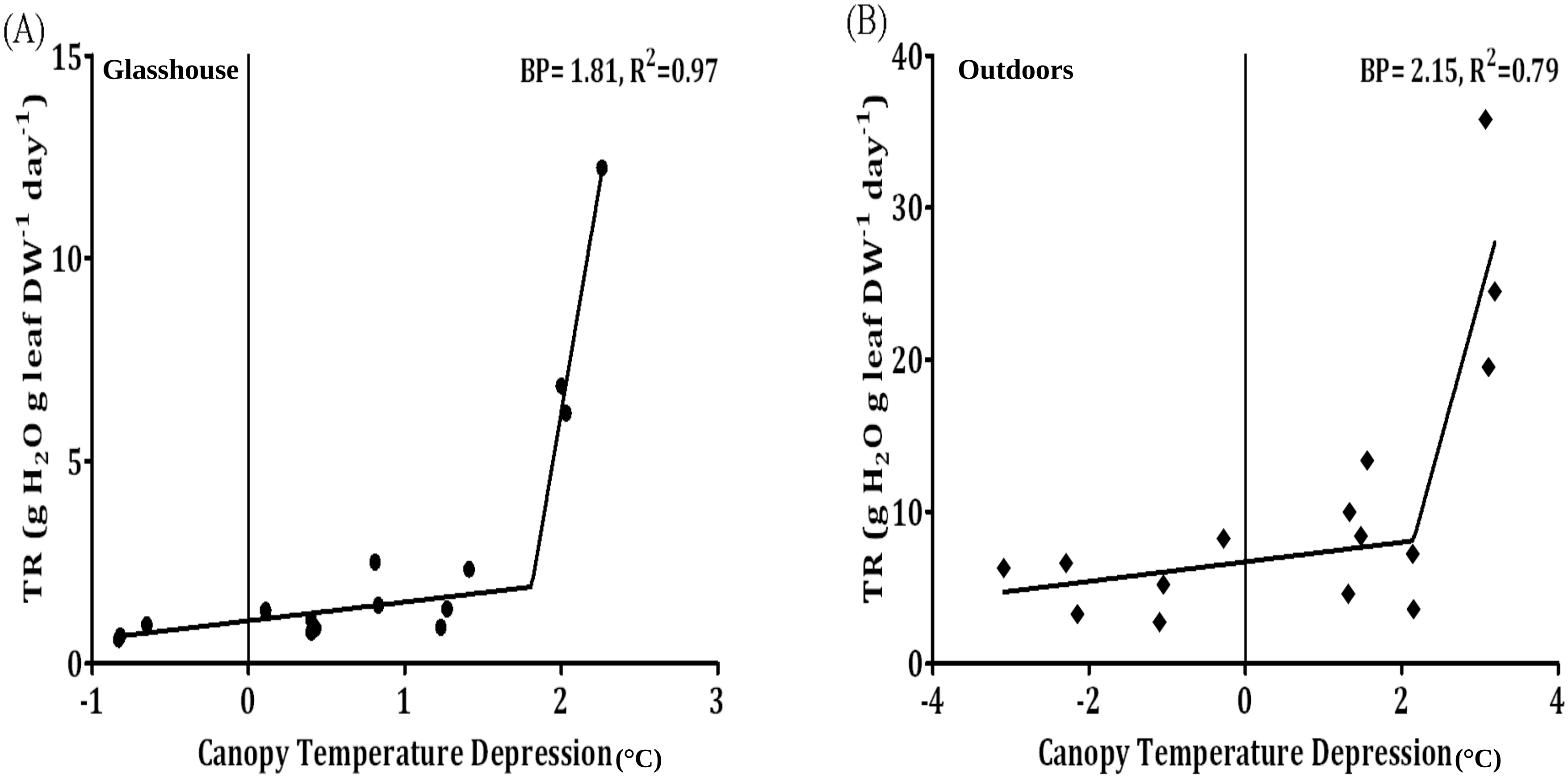


(C)
Glasshouse
Transpiration efficiency (g/kg)
3.50
3.00
2.50
2.00
1.50
1.00
0.50
0.00
0.40
0.45
0.50
0.55
0.60
0.65
0.70
0.75
FTSW-threshold for transpiration
y = -1.568x + 3.084
R² = 0.036
P = 0.5007

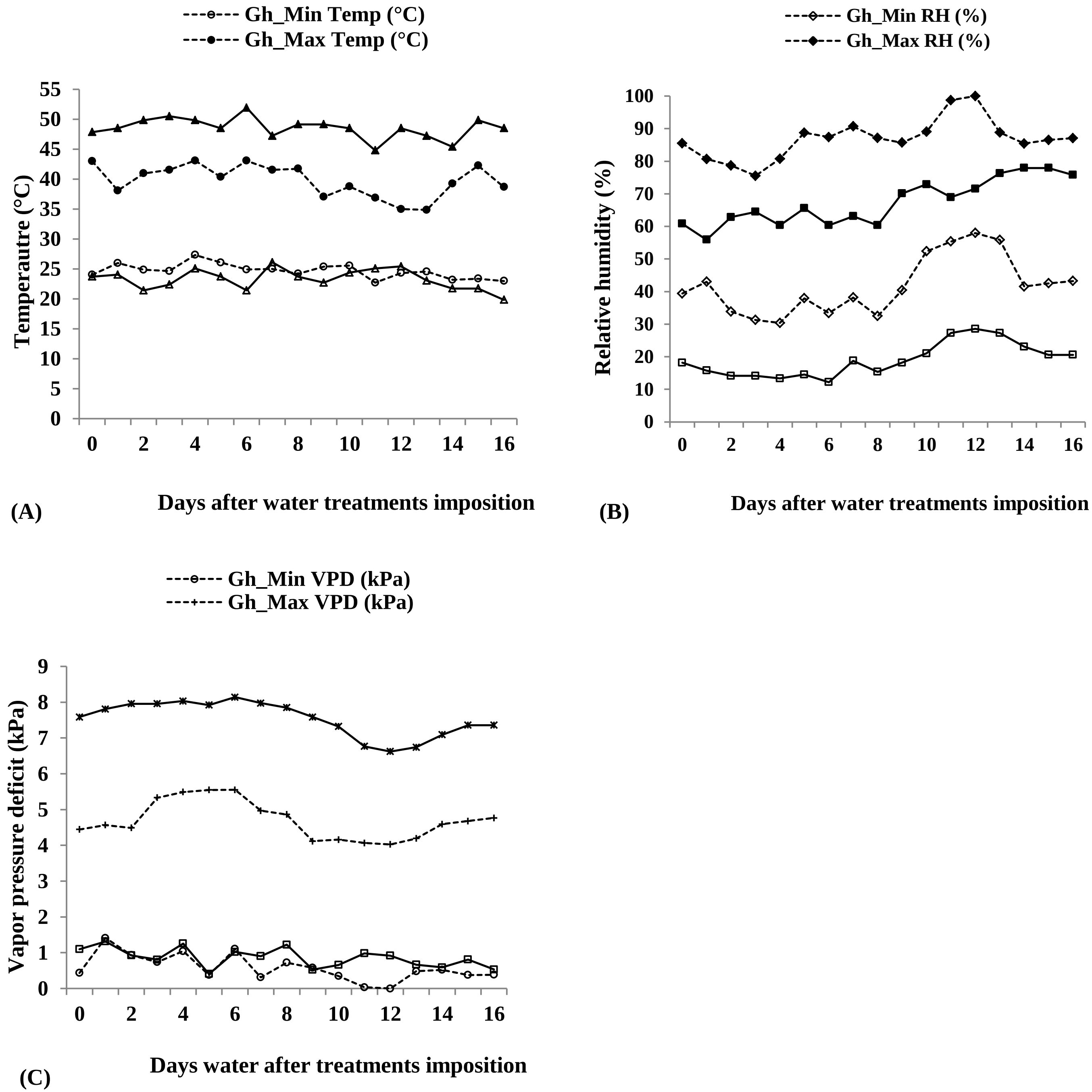
(C)
Outdoors
Transpiration efficiency (g/kg)
2.00
1.50
1.00
0.50
0.00
0.4
0.45
0.5
0.55
0.6
0.65
0.7
0.75
FTSW-threshold for transpiration
y = -3.389x + 3.017
R² = 0.617
P = 0.0005

**Figure 7**



(A)
Glasshouse
BP= 1.81, $R^2$=0.97
TR (g $H_2O$ g leaf $DW^{-1}$ $day^{-1}$)
15
10
5
0
-1
0
1
2
3
Canopy Temperature Depression (°C)
(B)
Outdoors
BP= 2.15, $R^2$=0.79
TR (g $H_2O$ g leaf $DW^{-1}$ $day^{-1}$)
40
30
20
10
0
-4
-2
0
2
4
Canopy Temperature Depression (°C)

**Suppl. Fig. 1**



Gh_Min Temp (°C)
Gh_Max Temp (°C)
Temperautre (°C)
55
50
45
40
35
30
25
20
15
10
5
0
0 2 4 6 8 10 12 14 16
(A)
Days after water treatments imposition
Gh_Min RH (%)
Gh_Max RH (%)
Relative humidity (%)
100
90
80
70
60
50
40
30
20
10
0
0 2 4 6 8 10 12 14 16
(B)
Days after water treatments imposition
Gh_Min VPD (kPa)
Gh_Max VPD (kPa)
Vapor pressure deficit (kPa)
9
8
7
6
5
4
3
2
1
0
0 2 4 6 8 10 12 14 16
Days water after treatments imposition
(C)

**Suppl. Fig. 2**