

# Absorbing roots areas and transpiring leaf areas at the tropical forest and savanna boundary in Brazil

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## Abstract

Plants capture essential resources for growth via absorbing surfaces on both roots and leaves. As a result, the allocation of assimilates to these resource exchange surface areas are of fundamental importance to plant growth and survival. Previous work on tropical forests and savanna vegetation has mainly focused on broad root:shoot biomass ratios. Yet, uptake of CO<sub>2</sub> (leaves), water and nutrients (roots) is a surface area phenomenon. In this study we compared the root:leaf ratio of the active absorbing area at the ecosystem scale, within eight structurally diverse stands, that were chosen to characterise the transition between the Amazonian forest (closed canopy) and Brazilian Cerrado (savanna). We use an earth impedance method to quantify the absorbing root area index ( $RAI_{\text{absorbing}}$ ) at each site, and compare these measurements to the more widely used fine root area index ( $RAI_{\text{fine}}$ ). Surprisingly, we found that  $RAI_{\text{absorbing}}$  and  $RAI_{\text{fine}}$  were not correlated, leading us to conclude that the two measurements are not direct substitutes. Additionally, we compared both measures of  $RAI$  with the leaf area index ( $LAI$ ) in these contrasting ecosystems. The resulting  $RAI_{\text{fine}}:LAI$  ratio ( $R^2=0.85$ ) was inversely proportional to basal area, with the highest values in the savanna vegetation. On the other hand, the  $RAI_{\text{absorbing}}:LAI$  ratio showed an opposite trend with basal area ( $R^2=0.83$ ), with highest values in the forest. We suggest this paradox may reflect different growth patterns by plants to access to adequate water and nutrient resources.

**Key Words:** root area index; leaf area index; earth impedance; fine root; stomatal area

## **Introduction**

Plants are capable of adjusting the relative size and distribution of organ systems, such as stems, leaves and roots, in response to changes in resource availability and disturbance regimes (De Castro & Kauffman 1998; Magnani et al. 1998). As resources are generally partitioned either aboveground (with respect to carbon dioxide and light) or belowground (with respect to the availability of water and nutrients), it is thought that plants adjust their biomass partitioning to counterbalance the relative abundance of aboveground and belowground resources (Robinson 1986; Johnson & Thornley 1987; Gedroc, McConnaughay, & Coleman 1996). These adjustments are a functional response, which determine the efficiency of future resource acquisition. This theory of optimal partitioning has been used to explain distinct root:shoot ratios between forests and woody savannas (Reynolds & Pacala 1993; Belcher, Keddy, & Twolan-Strutt 1995). For example root:shoot ratios in Brazilian woody savannas (Cerrado) range from 2.9-7.7 (De Castro & Kauffman 1998), while tropical dry forests have significantly lower ratios in the range 0.42-0.84 (Murphy & Lugo 1986; Castellanos, Maass, & Kummerow 1991). These distinct biomass allocations in turn result in contrasting vegetation structures which are thought to be associated with physiological adaptations to their contrasting environments.

Root:shoot ratios have often been interpreted as a differential investment of photosynthetically fixed carbon between above and belowground organs to maximise acquisition of limiting resources (Titlyanova et al. 1999). However, roots are known to perform a range of functions including structural support, storage, transport and absorption (Schulze, Schilling, & Nagarajah 1983). Acquisition of resources only occurs only through the absorbing surface area, which may not be strongly related to biomass (Butler et al. 2010). Therefore, the root:shoot biomass

ratio may only be a coarse indicator of physiological processes. In contrast, the partitioning between actively absorbing roots and transpiring leaves is functionally more informative, especially with respect to maintaining an efficient water balance (Shackleton, McKenzie, & Granger 1988; Litton et al. 2003; Butler et al. 2010).

Quantifying the belowground resource capturing ability of terrestrial vegetation remains a major challenge in ecosystem science. Conventionally, it is thought that fine roots represent the absorbing surfaces for both water and nutrient uptake (Hendrick & Pregitzer 1992; Pregitzer, Hendrick, & Fogel 1993; Gill & Jackson 2000). Accordingly, the belowground resource exchange surface area has previously been measured as the live fine root area index (RAI; surface area of fine roots per unit ground area). Resulting global estimates of RAI range from 4.6 in boreal forests to 42.5 in tropical grasslands and savannas (Jackson, Mooney, & Schulze 1997). However, in recent methodological advancements electrical impedance spectroscopy have been used to provide a direct estimate of the area of root surface which is in physiological contact with the soil (Aubrecht, Stanek, & Koller 2006; Čermák et al. 2006). Several field studies have demonstrated a correlation between electrical impedance at a single low frequency and several biometric parameters including root biomass. However, establishing a clear relationship between fine root surface area and electrical parameters in the field remains illusive. A laboratory study of individual willow saplings observed a decrease in electrical impedance as the contact surface area of the roots with a hydroponic solution increased. Accordingly, clearly characterizing the relationship between the plant root systems as measured by the electrical impedance method in the field and fine root surface area remains an important challenge.

The main objectives of this study was to compare the absorbing root area and leaf area of different vegetation types characterising the transition zone between the Amazonian forest and the Brazilian Cerrado. We quantified and compared RAI estimates using both the earth impedance method, and live fine root surface area. We then contrasted these estimates with the analogous

aboveground measure of leaf area index (LAI) across eight structurally contrasting stands. We evaluated the role of vegetation structure and density on ecophysiological allocation patterns of leaves and roots.

## **Materials and Methods**

### ***Study Site***

The study area was located in the northeast region of the state of Mato Grosso, Brazil, within the municipalities of Nova Xavantina and Ribeirão Cascaliera (Table 1). The region is classified as an area of ‘ecological tension’ in the transition zone between savanna (*cerrado*) and dry forest (*floresta estacional*) on the *Mapa de Vegetação do Brasil* (IBGE 1993). The climate is characterised by a seasonal rainfall pattern, with close to no precipitation for three consecutive months of the year (Fig. 1). We chose eight locations representing two main vegetation types, savanna and dry forest, growing on a variety of soil types (Table 1). All the plots had dominant woody vegetation of both trees and shrubs with very limited grass cover.

Seven one hectare (100 × 100 m) and one previously established 0.6 hectare plots were surveyed (Table 1). Four of the plots were located within the municipality of Nova Xavantina and four within the municipality of Ribeirão Cascaliera. A detailed inventory of the vegetation was carried out on all the trees larger than 10 cm diameter at breast height (*DBH*; 1.3 m). The species, *DBH* and crown diameter (two perpendicular measurements) were recorded for each tree in the plot. All measurements were taken in April 2007, at the end of the wet season when root biomass is expected to be at its maximum.

### ***Absorbing Root Surface Area***

Root absorbing area was quantified using an electrical impedance (EI) method (Aubrecht et al. 2006; Čermák et al. 2006; Butler et al. 2010). When a simple electrical circuit is established

between a tree and the soil, current passes across the tree-soil interface through the same channels used for water and nutrient uptake (Fig. 2). From the difference in conductivity of the water-conducting tissue of the tree and the soil, we can estimate the soil-root exchange surface area (Equation 1).

$$S = \rho l \frac{I}{U} \quad (1)$$

where  $S$  is the total root absorbing surface area ( $\text{m}^2$ ),  $\rho$  is the resistivity of the water conducting tissue ( $\Omega\text{m}$ ),  $l$  is the distance from the stem (m),  $I$  is the current (Amps) flowing between the tree and soil to auxiliary metal electrodes from an external power supply, and  $U$  is the potential difference between the stem boundary and a potential electrode in the soil (Volts).

Following established methods (Aubrecht et al. 2006; Čermák et al. 2006; Butler et al. 2010), we inserted between four and six (depending on the size of the tree) electrodes into the stem of the tree and ten soil electrodes in a  $60^\circ$  arc around the stem and connected them to an alternating current generator. The soil electrodes were 10 mm in diameter and inserted 20 cm into the soil to ensure a sufficient conducting surface. An auxiliary potential electrode was inserted at the base of the trunk and another in the soil at a defined distance,  $l$ , from the stem. The distance from the stem to both the current and potential soil electrodes was determined by the course of potential (voltage) characteristics. The amount of current flowing from the tree stem to the surrounding soil via the root segment decreases with increasing distance from the stem. This drop in voltage was mapped by progressively moving the soil potential electrode away from the stem in a radial direction. The point at which the drop in voltage reached a plateau is considered to be  $l$ , which corresponds to the mean distance of all the absorbing root segments of the tree.

The resistivity of the water conducting tissue ( $\rho_{\text{wood}}$ ) of the roots was calculated using the four point Wenner method, where  $\rho_{\text{wood}} = 2\pi a R$  (Aubrecht et al. 2006). The four (or six) stem electrodes were inserted into the sapwood at an equal distance,  $a$ , apart. The electrical impedance,

$R$ , was measured with the electrodes 2, 4, and 6 cm apart and the mean was taken. Current was generated and impedance measurements were made using a Earth/Ground Tester (Model 1623, Fluke, Utah, USA).

A minimum of 12 trees were measured within each plot using the electrical impedance method. The target trees were chosen at random with stratified diameter classes, and measurements were repeated for two 60° segments (Fig. 2) which were multiplied by three to estimate the whole tree root surface (360°). Two segments were measured in the opposite directions of North and South. For each of our target trees we recorded: species, *DBH*, height of the first branch, tree height and crown cover (based on longest and shortest crown diameters).

### ***Fine Root Area***

We measured fine root area by taking 15 intact soil cores in each of the 8 plots. Each core had a diameter of 8 cm and a depth of 30 cm. We carefully washed all the roots out of the soil and separated the live and dead roots based on visual distinction in colour and resilience. For each plot, a subsample of live fine roots ( $\leq 2$  mm in diameter) were scanned, dried and weighed. The images from the scans were then analysed using the WinRhizo software to calculate the surface area of fine roots ( $\text{cm}^2$ ). The resulting relationship with the sample weights were used to estimate the total surface area of the remaining samples. We calculated the fine root area index ( $RAI_{\text{fine}}$ ) as the surface area of fine roots per unit ground area.

### ***Leaf Area Index***

Images of the canopy were taken from the ground, at a height of 1m, with an upward-viewing digital camera and hemispherical lens (Nikon Coolpix 900, Nikon Corporation, Japan) at 25 locations within each plot (Breda 2003). Images were always taken in the late afternoon with the bottom of the camera facing north, in the absence of direct sunlight. The images were then analyzed with image analysis software (Gap Light Analyser, ring 5, GLA). For the forest plots we calculated

$LAI$  ( $m^2m^{-2}$ ) directly using the GLA software. However, the assumptions of this method are not consistent with the fragmented canopy structure found in savanna environments. Therefore, we used published values of leaf area per unit crown area within similar cerrado vegetation (Hoffmann et al. 2005). Canopy cover for the savanna plots was measured using the GLA software. Subsequently, to obtain an estimate of overall  $LAI$ , the leaf area per unit crown area was multiplied by the total canopy cover.

### ***Stomatal Area***

We measured the stomatal dimensions of the five most dominant species based on their importance value (IV), within all the plots located in the Nova Xavantina municipality. Species which presented difficulties in visualising the stomata were excluded from the study and the next most important was taken. Each species was represented by a sample of two sun leaves from five individuals. Stomatal density,  $D$ , and guard cell lengths,  $L$ , were determined using an optical microscope (Carl Zeiss MicroImaging GmbH 37081 Gottingen, GERMANY). Measurements were taken on the abaxial surface located centrally, midway between the midrib and margin. The lengths of five guard cells were averaged within each sample leaf, and a species average was compiled from the five sampled individuals. We estimated the area of each individual stoma as  $L$  multiplied by the guard cell width, which we approximated as half  $L$ . We then calculated stomatal pore area index ( $SPI$ ) which is a dimensionless index of the stomatal pore area per unit area of the leaf lamina (Sack, Tyree, & Holbrook 2005; England & Attiwill 2006).

### ***Data Analysis***

To explore general differences in absorbing root area between forest and savanna species we used an analysis of covariance to investigate vegetation specific correlations with various above ground biometric parameters. However, there was reason to believe that there may be variation associated with plot specific factors, such as vegetation structure, or soil type. Therefore the fixed effects

assumptions of the analysis of covariance were untenable, leading to systematic error when scaling to the stand level. Consequently, to scale our measure of absorbing root area to the stand level we estimated plot specific coefficients in a multiple comparisons random effect model using generalized least squares (Model 1):

$$S_i = aA_i^b + \alpha_i + u_i \quad \text{Model 1}$$

where  $i$  refers to the plot,  $S$  is root absorbing area ( $\text{m}^2$ ),  $A$  is basal area,  $a$  and  $b$  are coefficient vectors,  $\alpha$  is a coefficient for the random plot level effects, and  $u$  is the error term. We applied this model to the tree inventory data to estimate the total root absorbing area in each plot.

We investigated possible differences between the root absorbing area of evergreen and deciduous species within the savanna vegetation type using an analysis of covariance, as we felt any systematic differences would affect our results when scaling to the stand level. We pooled brevicedeciduous species as evergreen, as there is evidence to suggest they are physiologically similar (Jackson et al. 1999). Additionally, we evaluated differences between the stomatal size and density of forest and savanna species using the unpaired Wilcoxon rank sum test.

From our estimates of the total absorbing root area for each stand we estimated the absorbing root area index ( $RAI_{\text{absorbing}}$ ), which is a dimensionless index of the total absorbing root area per unit ground area. The correlations between RAI to LAI ratios (both  $RAI_{\text{absorbing}}:LAI$  and  $RAI_{\text{fine}}:LAI$ ) and other biometric parameters (including basal area, mean canopy height) were determined using ordinary least squares regression analysis. We used a weighted average of  $SPI$ , based on the importance value of the measured species, which was multiplied by  $LAI$  to estimate to the transpiring leaf area index ( $LAI_{\text{transpiring}}$ ). All statistical analyses were done using R (R Development Core Team, 2008).

## Results



### ***Vegetation Structure***

The study plots spanned a wide gradient of basal area and tree density. At the low end of the range was the open savanna or ‘*Cerrado aberto*’ (STM-03) with a basal area of  $3.2 \text{ m}^2\text{ha}^{-1}$  and tree density of 170 stems per hectare, while the high end was represented by the *Brosmium* mono-dominant forest (VCR-01) with a basal area of  $28.5 \text{ m}^2\text{ha}^{-1}$  and a tree density of 468 stems per hectare (Table 2). This structural gradient in the vegetation was also characterised by an increase in the mean tree height. Additionally, *LAI* increased rapidly with basal area among the savanna plots, saturating at basal areas represented by the forest stands (Table 2 & Fig. 4). In summary, the climates of all the study plots were very similar, yet they spanned a large structural gradient.

### ***Absorbing Root Surface Area***

We found the absorbing root area of individual trees, defined from the impedance method, to vary between  $0.25$  and  $5 \text{ m}^2$  across all tree sizes and vegetation types. We explored allometric relationships between absorbing root area ( $S$ ) and basal area ( $A_{bas}$ ), tree height ( $h$ ), crown area ( $A_{crown}$ ), and stem volume ( $V_{stem}$ ), for both forest and savanna species. Each of the aboveground biometric parameters showed a reasonably good correlation with absorbing root surface area ( $S$ ) with 39-69 % of the variation explained by each parameter ( $p \leq 0.05$ ; Fig. 3 & Table 3). There was considerably more unexplained variation within the savanna vegetation with  $R^2$  values ranging from 0.39-0.55, with crown projected area performing the best in terms of explained variation in  $S$ . Generally, savanna trees had significantly ( $p \leq 0.05$ ) less absorbing root area than their forest counterparts, with the difference increasing at larger tree sizes (Fig. 3). Absorbing root area showed a power function relationship with  $A_{bas}$ , crown projected area and stem volume, saturating at high values (Fig. 3:A, C, & D). In contrast, height showed a clearly linear correlation with absorbing root area (Fig. 3B). Additionally, we found no significant difference between evergreen and deciduous species ( $p > 0.05$ ).

### ***Stomatal Area***

We found that stomatal density ranged from 205-848 mm<sup>-2</sup> across all species, while guard-cell length ranged from 9.3-36.2 μm. We did not find any significant difference between the stomatal density of forest and savanna species ( $p=0.381$ ), however, forest species had significantly ( $p=0.01$ ) smaller stomata. Consequently, the stomatal cells of forest species occupied significantly ( $p=0.01$ ) less leaf surface area. The stomata of the forest species in this study were on average about 30% smaller, and the mean proportion of the leaf lamina occupied by stomatal cells was 10% for forest species and 17% for savanna species (Table 4).

### ***Root Area Index***

Estimates of  $RAI_{\text{fine}}$  from core samples ranged from 4.5 m<sup>2</sup>m<sup>-2</sup> in the Cerrado aberto (STM-03) to 7.4 m<sup>2</sup>m<sup>-2</sup> in both of the dense savanna (*'Cerrado sensu stricto'*) plots studied (STM-01 & NXV-01; Table 2 & Fig. 4). In contrast, to scale  $RAI_{\text{absorbing}}$  to the stand level we used a random effects model correlating absorbing root area with  $A_{\text{bas}}$  independently for each plot (AIC=98.11). We found plot level differences to be significant ( $p=0.02$ ). The resulting estimates of  $RAI_{\text{absorbing}}$  across all the plots ranged from 0.01 in the *Cerrado aberto* (STM-03), to 0.13 m<sup>2</sup> m<sup>-2</sup> in the *Brosimum* mono-dominant forest (VCR-01). We found no significant correlation between  $RAI_{\text{fine}}$  and  $RAI_{\text{absorbing}}$  ( $p=0.87$ ; Fig. 5A).

We found the  $RAI_{\text{fine}}:LAI$  ratio to be inversely proportional to basal area across all the study sites ( $p\leq 0.05$ ,  $R^2=0.85$ ; Fig. 5B), with savanna vegetation having more fine root area relative to leaf area. In contrast, the ratio of  $RAI_{\text{absorbing}}$  to  $LAI$  was directly proportional to both basal area ( $p\leq 0.05$ ,  $R^2=0.85$ ; Fig. 5C) and mean tree height ( $p\leq 0.05$ ,  $R^2=0.83$ ; Fig. 5D). Consequently, the forest plots with taller trees had a higher  $RAI_{\text{absorbing}}:LAI$  ratio than their savanna counterparts.

The stomatal pore area index ranged from 0.11 to 0.17, while the ratio of  $RAI_{\text{absorbing}}:LAI_{\text{transpiring}}$  ranged from 0.26 to 1.23 across the four plots (Fig. 6). The *Brosimum* forest (VCR-01) was the only vegetation type to have a ratio greater than one, indicating a higher surface

area at the root-soil interface. In contrast, only a quarter of the aboveground exchange surface area is mirrored belowground in the Cerrado (NXV-01) vegetation (Fig. 6).

## **Discussion**

### ***Absorbing Root Surface Area***

In general we would expect a given unit of absorbing root area to support a proportional amount of leaf area or water conducting tissue (Shinozaki et al. 1964; Sperry et al. 1998). Accordingly, the results from more than a hundred trees representing a high diversity of species show a consistent and reasonably strong relationship between absorbing root area and both basal area and crown projected area (Fig. 3). However, within these relationships there is a divergence between forest and savanna species, with savanna trees surprisingly showing less absorbing root surface area for a given basal area. We speculate that the difference between these relationships could be related to the attainment of maximum tree height. A large root absorbing area will reduce the resistance to water flow across the soil-root boundary, thereby offsetting any increase in hydraulic resistance associated with tall trees (Magnani, Mencuccini, & Grace 2000). This theory is additionally supported by the linear relationship between tree height and root absorbing area (Fig. 3B). Alternatively, savanna trees have a greater proportion of their fine roots deeper in the soil profile (Jackson et al. 1997). Therefore, if the EI method only measures the surface roots this will result in an underestimate of the absorbing root surfaces areas of savanna trees.

### ***Root Area Index***

Up to now, root area index has been estimated from the geometric surface area of fine roots (Jackson et al. 1997). *RAI* estimates from similar vegetation to that in our study fall in the range of 6.3 in tropical deciduous forests to 42.5 in tropical grasslands (Jackson et al. 1997). Our estimates of  $RAI_{\text{fine}}$  across all the study sites, all of which were dominated by trees, are most similar to those reported for tropical deciduous forests. However, there are complications with the use of fine root

area as a measure of the absorbing root surface. It has not been possible to clearly define which part of the root system is permeable to water and nutrients in field studies. This ambiguity has led to an inconsistency in the definition of fine roots between studies; ranging from <1mm to as much as <5mm in diameter (Persson et al., 1995; Pregitzer et al., 1998; Vanninen and Makela, 1999).

The attributes of the earth impedance method are assumed to be a measure of the active root surface area ( $RAI_{\text{absorbing}}$ ), which is a fundamental parameter for verification of many root water extraction paradigms. The rate of water and nutrient uptake by plant roots is based on the fluxes of these elements across active root surfaces. However, the area across which these fluxes take place has been difficult to clearly identify. Therefore, we may use our estimate of  $RAI_{\text{absorbing}}$  to identify the uncertainties that exist in quantifying the effective fraction of the total fine root surface area that is contributing to water and nutrient uptake. Our estimates of  $RAI_{\text{absorbing}}$  fell in the range of 0.01 to 0.13, and are more than an order of magnitude smaller than  $RAI_{\text{fine}}$  (Fig. 4), which suggests that only a very small fraction of the fine root surface area contributes to water and nutrient uptake. Furthermore, the lack of correlation between  $RAI_{\text{fine}}$  and  $RAI_{\text{absorbing}}$  suggest a potential discontinuity of the fraction of the total fine root surface area that is contributing to water and nutrient uptake. We acknowledge that there are discrepancies between these two estimates of  $RAI$ ; for example,  $RAI_{\text{absorbing}}$  only includes the larger trees ( $\geq 10$  cm  $DBH$ ), while  $RAI_{\text{fine}}$  encompasses the vegetation as a whole. However, these discrepancies would not account for the size of the differences between the two estimates.

### ***Root to Leaf Area Ratios***

A fundamental adaptation for maintaining an efficient water balance in arid climates is the absorbing root area to leaf area ratio (Shackleton et al. 1988; Sperry et al. 1998; Litton et al. 2003). In this study we have compared our estimates of  $RAI$  to the analogous aboveground parameter of  $LAI$ . The  $RAI_{\text{fine}}:LAI$  ratio varied widely across the eight study sites and was strongly correlated with basal area; with low basal areas corresponding to the highest root to leaf area ratios. As

expected, we observed that savanna ecosystems allocate more resources to fine roots relative to leaves, suggesting that they are limited by belowground resources. This result is consistent with the carbon fixation strategy of savanna ecosystems; where leaf life span (of deciduous species) is sacrificed for a higher rate of return on a given investment in leaf biomass. As a result, savanna species generally have a higher photosynthetic capacity (per unit leaf area) and a faster diffusion rate through the stomates. There is also mounting empirical evidence for the Co-ordination between the plant hydraulic system and photosynthetic carbon assimilation rates (Brodribb and Field 2000; Brodribb et al. 2002). It is thought that to capitalize on higher potential rate of carbon fixation per unit leaf area it is essential to maintain a continuous supplied of water to the leaves thus requiring a higher fine root to leaf area ratio.

In contrast to the  $RAI_{\text{fine}}:LAI$  ratio, we found that the  $RAI_{\text{absorbing}}:LAI$  ratio showed the opposite trend with basal area, with forest vegetation types having the highest values. This result appears to indicate that the low biomass savanna plots are not limited by belowground resources. This interpretation is in discord with evidence showing that net primary productivity in the Brazilian cerrado limited by soil nutrients (Arens 1963; Furley & Ratter 1988). Accordingly, several authors have found a correlation between basal area and soil fertility in the cerrado and dry forests of central Brazil (Goodland & Pollard 1973; Lopes & Cox 1977) (Ruggiero et al. 2002). One explanation for this discrepancy could lie in differences in water uptake efficiency. The volume of water moving across a given surface area (whether it is the active root surface area or the transpiring leaf area) is dependent on the volume flux density ( $\text{m}^3 \cdot \text{m}^{-2} \cdot \text{s}^{-1}$ ). Therefore any differences in the hydraulic resistances of water conducting organs between these contrasting ecosystems will complicate any comparisons. Molecular studies of Aquaporins indicate they may be important for bulk water flow through absorbing roots (Luu and Maurel, 2005). Aquaporins are membrane water-channel proteins that facilitate water movement along a passive gradient in water potential. For example, the hydraulic conductivity of root cortex cells, as measured with a cell pressure probe, was reduced by upto 30% by altering the expression of an aquaporin gene (Javot et

al., 2003), elsewhere over expression of aquaporins in rice plants was shown to increase the radial hydraulic conductivity by 140% which resulted in an increase in the mass ratio of shoot to root of 150% (Katsuhara et al., 2003). There is also evidence to suggest that aquaporins interact with mycorrhizal fungi to regulate root hydraulic properties (Aroca et al., 2007).

### ***Stomatal Area***

Canopy conductance is influenced by the combination of the total leaf area and the proportion of that area which is occupied by stomata. At the individual leaf level it has been shown that *SPI* correlates well with leaf laminar conductance (Sack et al. 2005). Overall we found that the *SPI* was significantly less for forest species than for savanna trees, which corresponds with lower stomatal conductance (Sack et al. 2003; Sack et al. 2005; England & Attiwill 2006). High stomatal conductance tends to be positively correlated with maximum photosynthetic rates both of which decrease with increasing leaf life span (Reich et al. 1997; Ackerly and Reich 1999; Reich et al. 1999). This difference in *SPI* is driven by a reduction in the size of the stomatal aperture in forest leaves rather than a change in stomatal density. Smaller stomata have been shown to enhance water use efficiency, both by responding faster to reduced soil water supply, and also by having a shorter diffusion distance that would allow for more efficient gas exchange (Aasamaa & Sober 2001; Franks & Farquhar 2007). Consequently, strategies for balancing carbon gain with water loss seem to diverge between forests and savannas and are governed by a series of trade-offs.

We scaled the porous leaf-atmosphere interface represented by stomata to the stand level which is directly analogous to the porous soil-root interface measured by the EI method. We found that the differences in leaf area between the plots were counterbalanced by differences in stomatal pore area, resulting in largely similar values of  $LAI_{\text{transpiring}}$  across the different vegetation types. Therefore the increase in higher root to leaf area ratios found in forests is driven by higher  $RAI_{\text{absorbing}}$  values. In steady state conditions the flow of water across each component (roots stem or leaves) should be constant as nearly all the water taken up by the roots is lost through transpiration.

## Conclusions

The results from this study have demonstrated that measures of fine root area and absorbing root area (using the EI method) are not direct substitutes. Allocation patterns between leaves and fine roots are associated with a gradient of aboveground biomass, with savanna vegetation having the highest fine root to leaf area ratio. However, this measure of fine roots is not proportional to the functionally absorbing area measured using the EI method. This finding highlights the need to develop an alternative approach for estimating water absorption in plant roots in the field. Further research is needed to investigate the relationship between volume flux density of fine roots and their surface area in the field.

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## References

- Aasamaa, K. & Sober, A. (2001) Hydraulic conductance and stomatal sensitivity to changes of leaf water status in six deciduous tree species. *Biologia Plantarum*, **44**, 65–73.
- Arens, K. (1963) As plantas lenhosas dos campos cerrados como vegetação adaptada às deficiências minerais do solo. *III Simpósio Sobre o Cerrado*. pp. 13–115. Edgard Blucher /EDUSP, São Paulo.
- Aubrecht, L., Stanek, Z. & Koller, J. (2006) Electrical measurement of the absorption surfaces of tree roots by the earth impedance method: 1. Theory. *Tree Physiology*, **26**, 1105-1112.
- Belcher, J.W., Keddy, P.A. & Twolan-Strutt, L. (1995) Root and shoot competition intensity along a soil depth gradient. *Ecology*, **83**, 673-682.
- Breda, N.J.J. (2003) Ground-based measurements of leaf area index: a review of methods,

instruments and current controversies. *Journal of Experimental Botany*, **54**, 2403-2417.

- Butler, A., Barbier, N., Cermak, J., Koller, J., Thornily, C., McEvoy, C., Nicoll, B., Perks, M., Grace, J. & Meir, P. (2010) Estimates and relationships between aboveground and belowground resource exchange surface areas in a Sitka spruce managed forest. *Tree Physiol*, doi:10.1093/treephys/tpq022.
- Castellanos, J., Maass, M. & Kummerow, J. (1991) Root biomass of a dry deciduous tropical forest in Mexico. *Plant and Soil*, **131**, 225-228.
- Čermák, J., Ulrich, R., Stanek, Z., Koller, J. & Aubrecht, L. (2006) Electrical measurement of tree root absorbing surfaces by the earth impedance method: 2. Verification based on allometric relationships and root severing experiments. *Tree Physiology*, **26**, 1113-1121.
- De Castro, E.A. & Kauffman, J.B. (1998) Ecosystem structure in the Brazilian Cerrado: a vegetation gradient of aboveground biomass, root mass and consumption by fire. *Journal of Tropical Ecology*, **14**, 263-283.
- England, J.R. & Attiwill, P.M. (2006) Changes in leaf morphology and anatomy with tree age and height in the broadleaved evergreen species, *Eucalyptus regnans* F. Muell. *Trees-Structure and Function*, **20**, 79-90.
- Franks, P.J. & Farquhar, G.D. (2007) The mechanical diversity of stomata and its significance in gas exchange control. *Plant Physiology*, **143**, 78–87.
- Furley, P.A. & Ratter, J.A. (1988) Soil resources and plant communities of the central Brazilian cerrado and their development. *Journal of Biogeography*, **15**, 97-108.
- Gedroc, J.J., McConnaughay, K.D.M. & Coleman, J.S. (1996) Plasticity in Root/Shoot Partitioning: Optimal, Ontogenetic, or Both? *Functional Ecology*, **10**, 44-50.
- Gill, R.A. & Jackson, R.B. (2000) Global patterns of root turnover for terrestrial ecosystems. *New Phytologist*, **147**, 13-31.
- Goodland, R. & Pollard, R. (1973) The Brazilian Cerrado Vegetation: A Fertility Gradient. *Journal of Ecology*, **61**, 219-224.
- Hendrick, R.L. & Pregitzer, K.S. (1992) The demography of fine roots in a northern hardwood forest. *Ecology*, **73**, 1094-1104.
- Hoffmann, W.A., da Silva, E.R., Machado, G.C., Bucci, S.J., Scholz, F.G., Goldstein, G. & Meinzer, F.C. (2005) Seasonal leaf dynamics across a tree density gradient in a Brazilian savanna. *Oecologia*, **145**, 306-315.
- IBGE. (1993) *Mapa de Vegetação do Brasil*. Fundação Instituto Brasileiro de Geographia e Estatística.
- Jackson, P.C., Meinzer, F.C., Bustamante, M., Goldstein, G., Franco, A., Rundel, P.W., Caldas, L., Iglar, E. & Causin, F. (1999) Partitioning of soil water among tree species in a Brazilian Cerrado ecosystem. *Tree Physiology*, **19**, 717.
- Jackson, R.B., Mooney, H.A. & Schulze, E.D. (1997) A global budget for fine root biomass,



surface area, and nutrient contents. *Proceedings of the National Academy of Sciences of the United States of America*, **94**, 7362-7366.

- Johnson, I.R. & Thornley, J.H.M. (1987) A Model of Shoot: Root Partitioning with Optimal Growth. *Ann Bot*, **60**, 133-142.
- Litton, C., Ryan, M., Tinker, D. & Knight, D. (2003) Belowground and aboveground biomass in young postfire lodgepole pine forests of contrasting tree density. *Canadian Journal of Forest Research*, **33**, 351-363.
- Lopes, A.S. & Cox, F.R. (1977) Cerrado Vegetation in Brazil: An Edaphic Gradient. *Agron J*, **69**, 828-831.
- Magnani, F., Leonardi, S., Tognetti, R., Grace, J. & Borghetti, M. (1998) Modelling the surface conductance of a broad-leaf canopy: effects of partial decoupling from the atmosphere. *Plant, Cell & Environment*, **21**, 867-879.
- Magnani, F., Mencuccini, M. & Grace, J. (2000) Age-related decline in stand productivity: the role of structural acclimation under hydraulic constraints. *Plant, Cell & Environment*, **23**, 251-263.
- McDowell, N. McDowell, Barnard, H. Barnard, Bond, B. Bond, Hinckley, T. Hinckley, Hubbard, R. Hubbard, Ishii, H. Ishii, Köstner, B. Köstner, Magnani, F. Magnani, Marshall, J. Marshall, Meinzer, F. Meinzer, Phillips, N. Phillips, Ryan, M. Ryan, Whitehead & D. Whitehead. (2002) The relationship between tree height and leaf area: sapwood area ratio. *Oecologia*, **132**, 12-20.
- Murphy, P.G. & Lugo, A.E. (1986) Ecology of Tropical Dry Forest. *Annual Review of Ecology and Systematics*, **17**, 67-88.
- Pregitzer, K.S., Hendrick, R.L. & Fogel, R. (1993) The Demography of Fine Roots in Response to Patches of Water and Nitrogen. *New Phytologist*, **125**, 575-580.
- Reynolds, H.L. & Pacala, S.W. (1993) An Analytical Treatment of Root-to-Shoot Ratio and Plant Competition for Soil Nutrient and Light. *American Naturalist*, **141**, 51.
- Robinson, D. (1986) Compensatory Changes in the Partitioning of Dry Matter in Relation to Nitrogen Uptake and Optimal Variations in Growth. *Ann Bot*, **58**, 841-848.
- Ruggiero, P.G.C., Batalha, M.A., Pivello, V.R. & Meirelles, S.T. (2002) Soil-Vegetation Relationships in Cerrado (Brazilian savanna) and Semideciduous Forest, Southeastern Brazil. *Plant Ecology*, **160**, 1-16.
- Ryan, M.G. & Yoder, B.J. (1997) Hydraulic limits to tree height and tree growth. *Bioscience*, **47**, 235-242.
- Sack, L., Cowan, P.D., Jaikumar, N. & Holbrook, N.M. (2003) The hydrology of leaves: coordination of structure and function in temperate woody species. *Plant, Cell & Environment*, **26**, 1343-1356.
- Sack, L., Tyree, M.T. & Holbrook, N.M. (2005) Leaf hydraulic architecture correlates with regeneration irradiance in tropical rainforest trees. *New Phytologist*, **167**, 403-413.

- Schulze, E.D., Schilling, K. & Nagarajah, S. (1983) Carbohydrate partitioning in relation to whole plant production and water use of *Vigna unguiculata* (L.) Walp. *Oecologia*, **58**, 169-177.
- Shackleton, C.M., McKenzie, B. & Granger, J.E. (1988) Seasonal changes in root biomass, root/shoot ratios and turnover in two coastal grassland communities in Transkei. *South African Journal of Botany*, **54**, 465-471.
- Shinozaki, K., Yoda, K., Hozumi, K. & Kira, T. (1964) A quantitative analysis of plant form - the pipe model theory I. Basic analysis. *Japanese Journal of Ecology*, **14**, 97-105.
- Sperry, J.S., Adler, F.R., Campbell, G.S. & Comstock, J.P. (1998) Limitation of plant water use by rhizosphere and xylem conductance: results from a model. *Plant, Cell & Environment*, **21**, 347-359.
- Titlyanova, A.A., Romanova, I.P., Kosykh, N.P. & Mironycheva-Tokareva, N.P. (1999) Pattern and Process in Above-Ground and Below-Ground Components of Grassland Ecosystems. *Journal of Vegetation Science*, **10**, 307-320.

## Fig. and Table Captions

**Fig. 1** Geographical location of the study areas and mean monthly maximum temperature (circles) and precipitation (bars) (data obtained from WorldClim).

**Table 1** Plot location and meteorological information.

**Fig. 2** Diagrammatic representation of the earth impedance method.  $G$  is an alternating current generator,  $mA$  is a milliammeter,  $V$  is a voltmeter.

**Table 2** Stand level biometric data, including root area index ( $RAI$ ) estimates based on both fine root and earth impedance measurements (standard errors are reported where appropriate).

**Fig. 3** Absorbing root area derived from the EI method of forest and savanna trees. Correlation with, (A) basal area, (B) height, (C) crown area, and (D) stem volume. Equations for numbered regression lines are reported in Table 3 (all  $p$ -values are  $\leq 0.05$ ).

**Table 3** Estimated parameters and statistics for relationships between absorbing root surface area and aboveground biometric parameters (corresponding to regression function in Fig. 3).

**Fig. 4** Ecosystem level resource exchange surface area indices across a gradient of basal area. Indices include leaf area index ( $LAI$ ) and root area index ( $RAI$ ) based on the earth impedance method ( $RAI_{\text{absorbing}}$ ) and fine root surface area ( $RAI_{\text{fine}}$ ), across eight one hectare plots representing tropical forest and savanna vegetation types (error bars represent standard errors were applicable).

**Fig. 5** Comparison of root area index ( $RAI$ ) based on the earth impedance method ( $RAI_{\text{absorbing}}$ ) and fine root surface area ( $RAI_{\text{fine}}$ ), across eight one hectare plots representing tropical forest and savanna vegetation types. (A) relationship between  $RAI_{\text{absorbing}}$  and  $RAI_{\text{fine}}$ . (B) Relationship between  $RAI_{\text{fine}}:LAI$  ratio and stand basal area. (C) Relationship between  $RAI_{\text{absorbing}}:LAI$  ratio and stand basal area. (D) Relationship between  $RAI_{\text{absorbing}}:LAI$  ratio and mean tree height.  $RAI_{\text{absorbing}}:LAI$  ratios are multiplied by a factor of 100.

**Table 4** Stomatal dimensions of species represented in four plots in the Nova Xavantina municipality.

**Fig. 6** Comparison of absorbing root surface area index with transpiring leaf area index (stomatal pore area per unit ground area) of two forest and two savanna vegetation types. Ratios of  $RAI_{\text{absorbing}}:LAI_{\text{transpiring}}$  are presented above each bar.

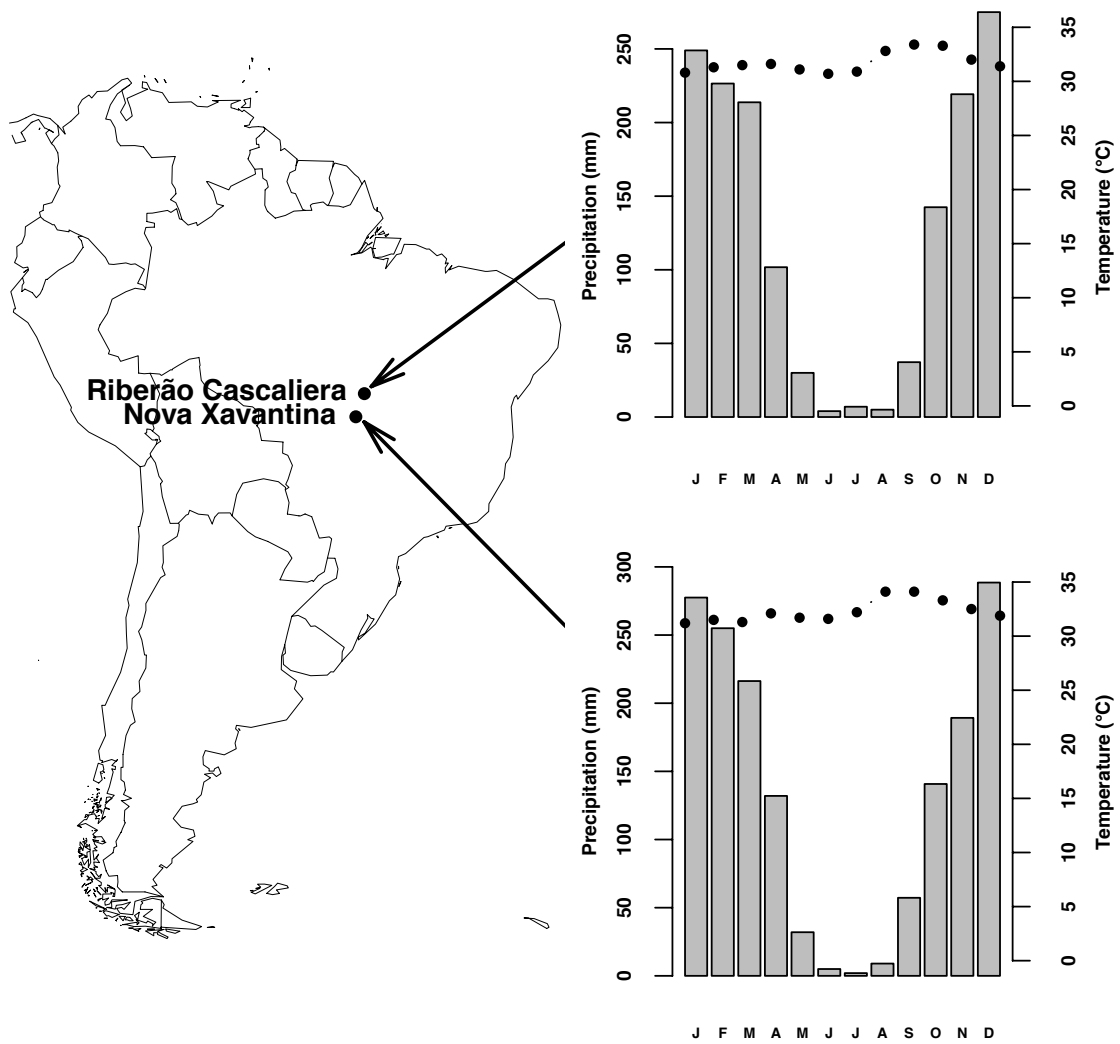


Figure 1

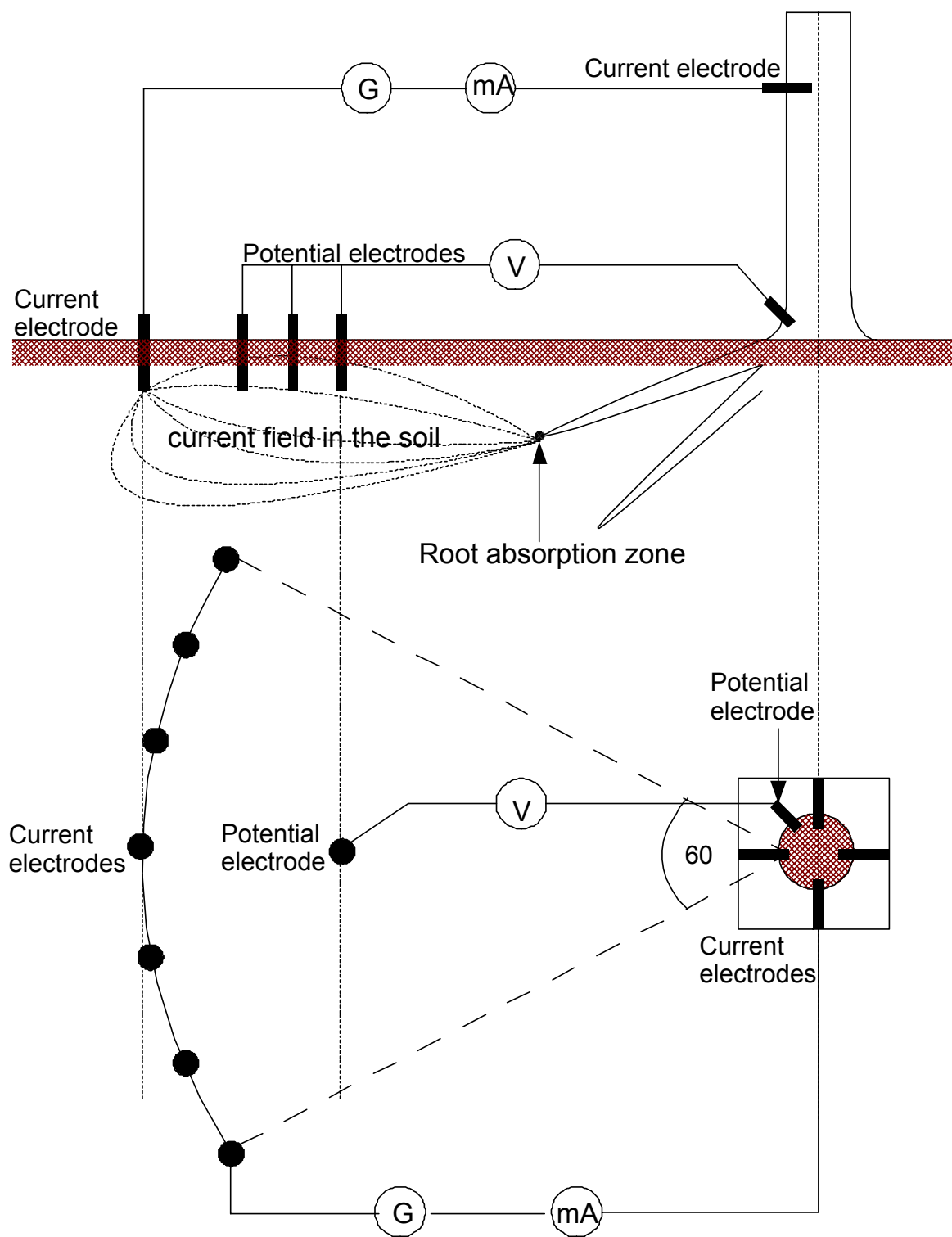


Figure 2

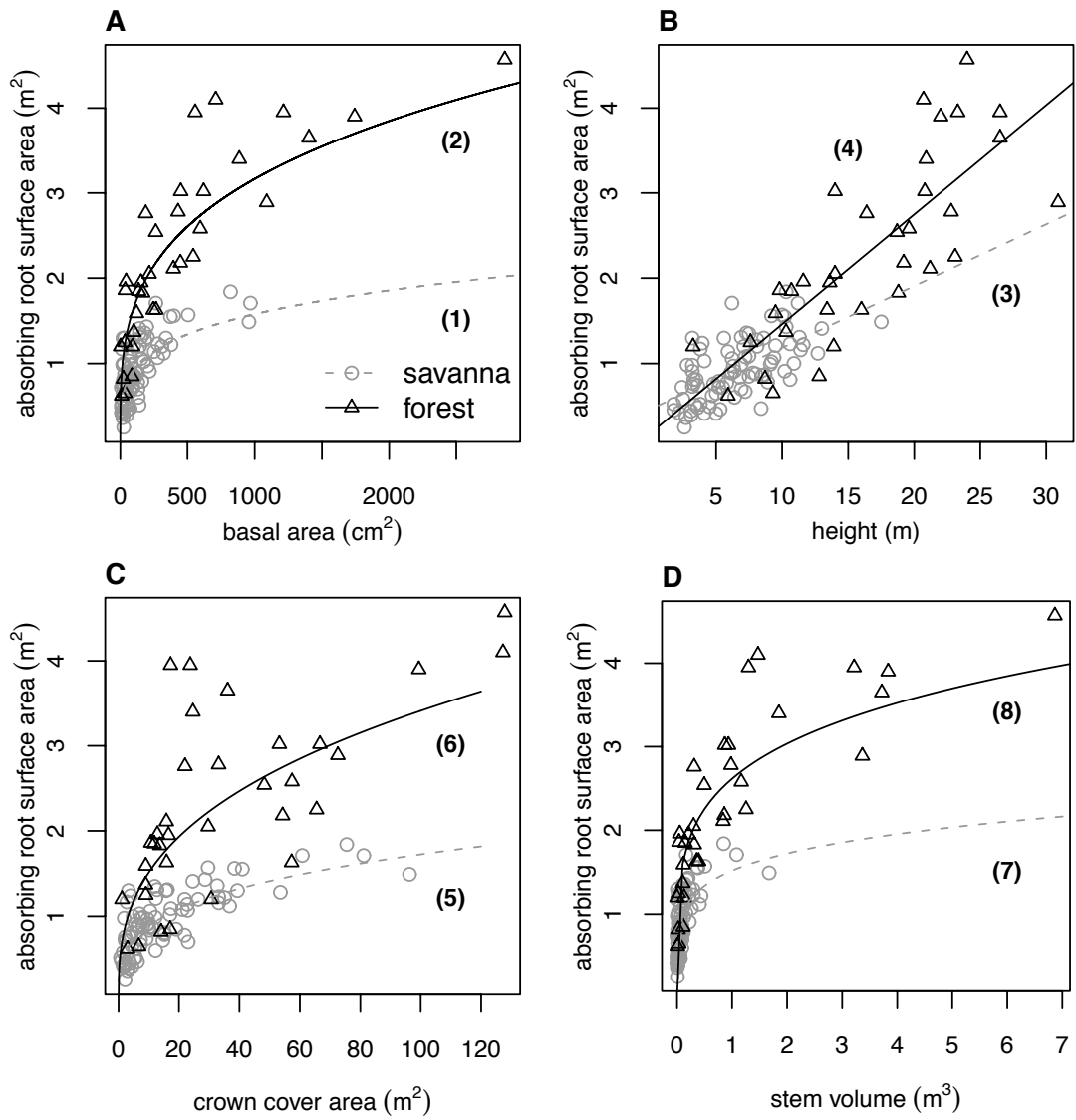


Figure 3

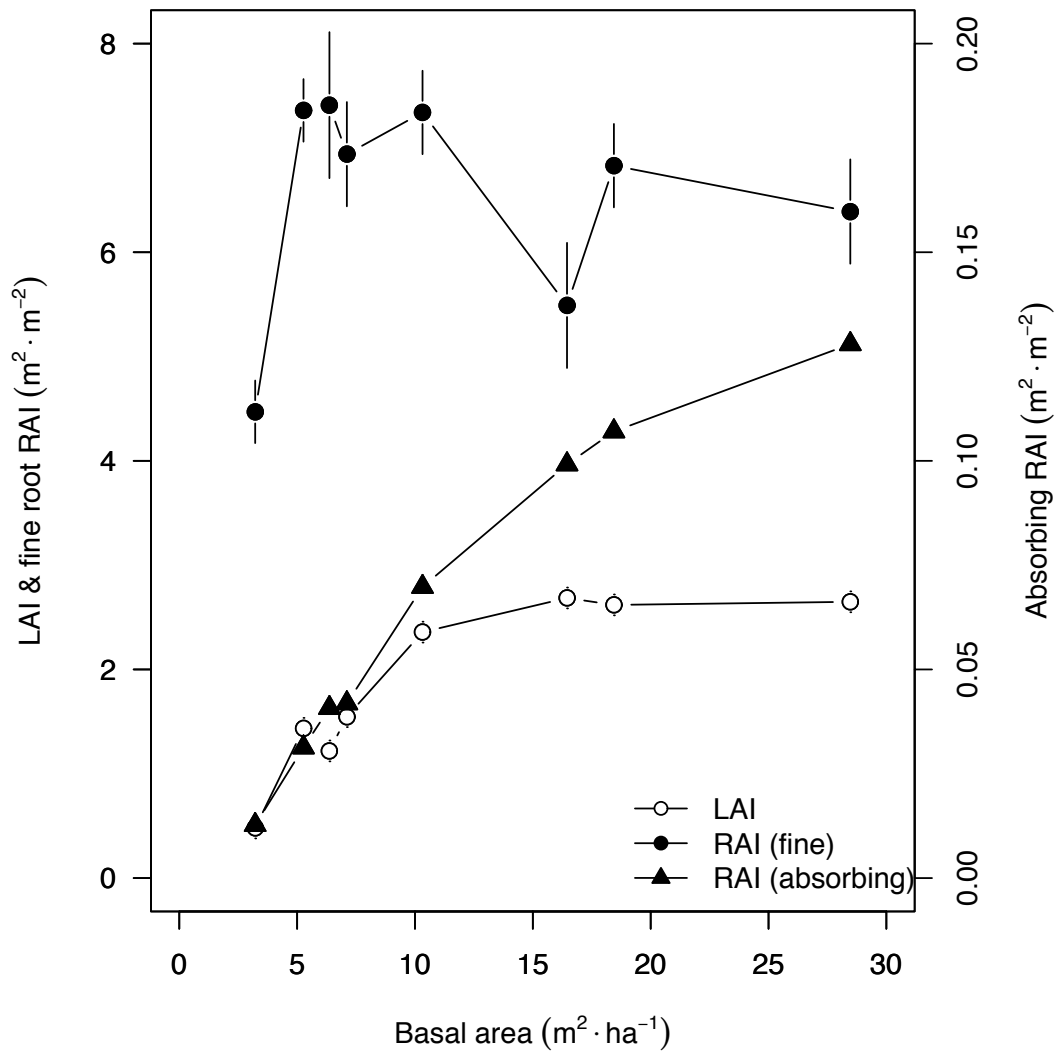


Figure 4

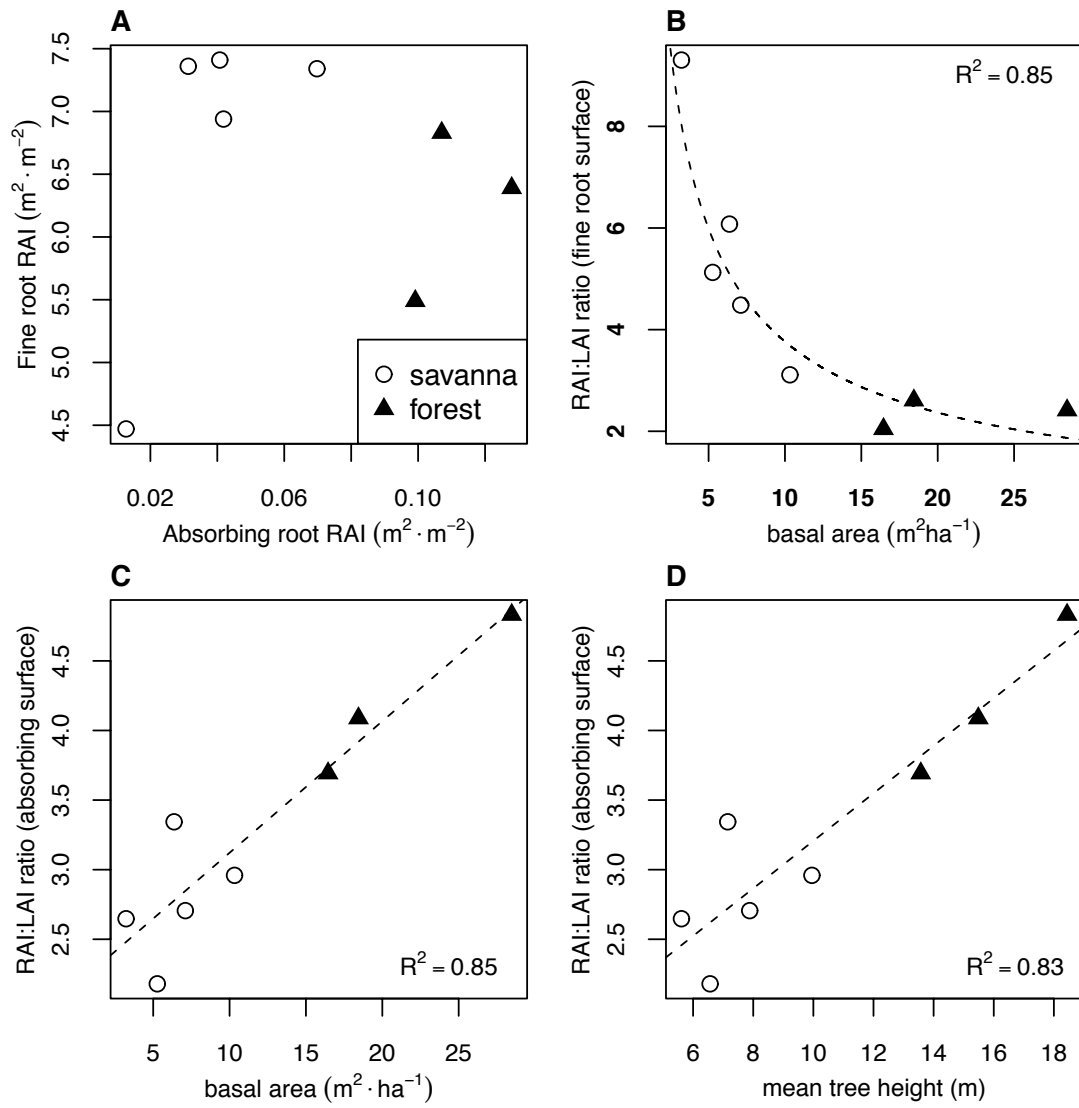


Figure 5



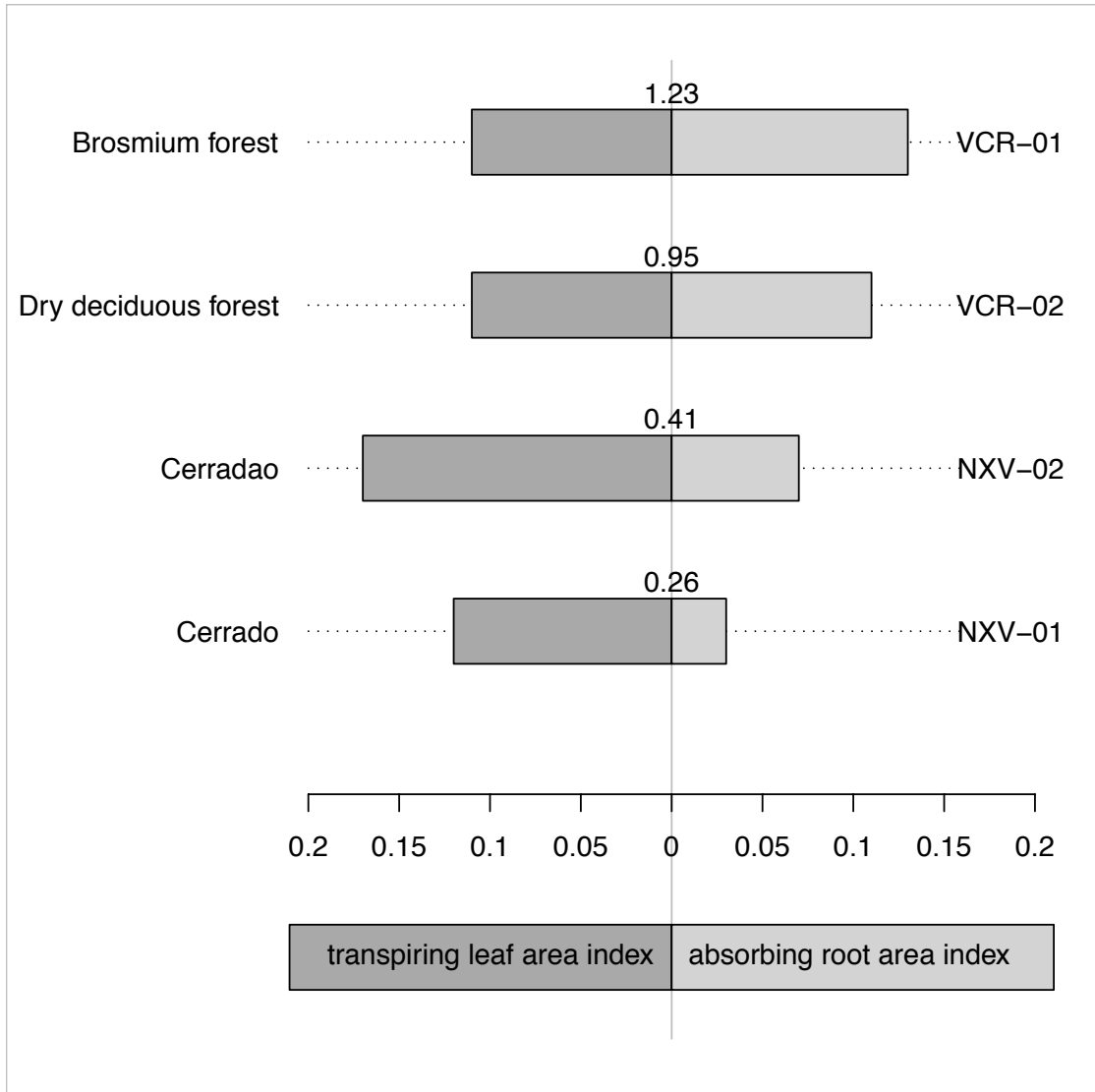


Figure 6

Table 1

Plot	Vegetation Type (Classification)	Location (nearest town)	Latitude	Longitude	Soil Type	Mean Annual Precipitation (mm)	Mean Precipitation of the Wettest Quarter (mm)	Mean Precipitation of the Driest Quarter (mm)
1	Cerrado <i>sensu stricto</i> (savanna)	Reserva Biológica, Municipal Mário Viana (Nova Xavantina)	14°71'S	52°35'W	Latosol	1508	747	16
2	Cerradão (savanna)	Reserva Biológica, Municipal Mário Viana (Nova Xavantina)	14°70'S	52°35'W	Latosol	1508	747	16
3	Evergreen mono-dominant ( <i>Brosimum</i> ) forest	Fazenda Vera Cruz (Nova Xavantina)	14°83'S	52°16'W	Ferrosol	1516	754	16
4	Dry deciduous forest	Fazenda Vera Cruz (Nova Xavantina)	14°83'S	52°17'W	Ferrosol	1512	754	16
5	Cerrado <i>sensu stricto</i> (savanna)	Fazenda Santa Marta (Ribeirão Casaliera)	12°82'S	51°77'W	Latosol	1603	820	16
6	Cerradão (savanna)	Fazenda Santa Marta (Ribeirão Casaliera)	12°82'S	51°77'W	Latosol	1603	820	16
7	Cerrado aberto (savanna)	Fazenda Santa Marta (Ribeirão Casaliera)	12°83'S	51°77'W	Ferrosol & Plinthosol	1599	818	16
8	Dry deciduous forest	Fazenda Floresta (Ribeirão Casaliera)	12°81'S	51°85'W	Oxisol	1613	826	16

Table 2

Plot	Vegetation	Tree Density (stems ha <sup>-1</sup> )	Basal Area (m <sup>2</sup> ha <sup>-1</sup> )	Mean Tree Height (m)	Leaf Area Index (m <sup>2</sup> m <sup>-2</sup> )	Fine Root Area Index (m <sup>2</sup> m <sup>-2</sup> )	Absorbing Root Area Index (m <sup>2</sup> m <sup>-2</sup> )
STM-03	Cerrado aberto (savanna)	170	3.22	5.61	0.48	4.5 (±0.3)	0.01
NXV-01	Cerrado <i>sensu stricto</i> (savanna)	341	5.27	6.56	1.55	7.4 (±0.3)	0.03
STM-01	Cerrado <i>sensu stricto</i> (savanna)	364	6.37	7.15	1.44	7.4 (±0.7)	0.04
STM-02	Cerradão (savanna)	411	7.11	7.88	1.22	6.9 (±0.5)	0.04
NXV-02	Cerradão (savanna)	564	10.27	9.95	2.36	7.3 (±0.4)	0.07
VCR-02	Dry deciduous forest	459	16.45	13.57	2.69	5.5 (±0.6)	0.11
FLO-01	Dry deciduous forest	604	18.44	15.49	2.62	6.8 (±0.4)	0.11
VCR-01	Evergreen mono- dominant ( <i>Brosimum</i> ) forest	468	28.47	18.44	2.65	6.4 (±0.5)	0.13

Table 3

Regression Line	Equations	Vegetation	Estimated parameters	
			<i>A</i>	<i>B</i>
1	$S = \alpha \cdot A_{bas}^\beta$	Forest	0.45	0.28
2		Savanna	0.31	0.23
3	$S = \alpha \cdot h + \beta$	Forest	0.13	0.18
4		Savanna	0.07	0.46
5	$S = \alpha \cdot A_{crown}^\beta$	Forest	0.67	0.35
6		Savanna	0.46	0.29
7	$S = \alpha \cdot V_{stem}^\beta$	Forest	2.62	0.21
8		Savanna	1.52	0.18

Table 4

VEGETATION/ Species	Importance value (IV)	Stomatal density (mm <sup>-2</sup> )	Guard cell length (µm)	Percent epidermal area occupied by stomata (%)
<i>CERRADO sensu stricto</i>				
<i>Qualea parviflora</i>	1 <sup>st</sup> 27.9	499.2 (±23.5)	17.4 (±0.7)	7.6 (±0.5)
<i>Davila elliptica</i>	2 <sup>nd</sup> 19.9	492.8 (±23.5)	17.6 (±0.4)	7.7 (±0.5)
<i>Roupala montana</i>	3 <sup>rd</sup> 13.8	326.4 (±8.9)	24.9 (±0.8)	10.1 (±0.7)
<i>Vochysia rufa</i>	7 <sup>th</sup> 8.2	620.8 (±10.3)	20.6 (±0.7)	13.1 (±0.6)
<i>Kielmeyera rubriflora</i>	8 <sup>th</sup> 8.2	281.6 (±19.8)	20.7 (±0.9)	5.9 (±0.3)
<i>Byrsonima pachyphylla</i>	10 <sup>th</sup> 8.0	552.0 (±17.8)	17.8 (±0.3)	8.7 (±0.1)
<i>CERRADÃO</i>				
<i>Hirtella glandulosa</i>	1 <sup>st</sup> 41.7	233.6 (±17.2)	18.2 (±0.2)	3.8 (±0.2)
<i>Sclerolobium paniculatum</i>	2 <sup>nd</sup> 24.1	435.2 (±16.7)	19.7 (±0.7)	8.5 (±0.7)
<i>Xylopia aromatica</i>	3 <sup>rd</sup> 20.8	633.6 (±23.5)	17.3 (±0.3)	9.4 (±0.3)
<i>Eriotheca gracilipes</i>	4 <sup>th</sup> 11.5	206.4 (±11.1)	36.2 (±0.5)	13.4 (±0.6)
<i>Guapira graciliflora</i>	6 <sup>th</sup> 11.3	361.6 (±15.9)	20.5 (±0.5)	7.5 (±0.3)
<i>Roupala montana</i>	7 <sup>th</sup> 11.2	307.2 (±29.4)	20.7 (±0.8)	6.4 (±0.4)
<i>DRY SEMI-DECIDUOUS FOREST</i>				
<i>Cheilochlinium cognatum</i>	1 <sup>st</sup> 36.0	478.4 (±17.2)	11.5 (±0.7)	3.2 (±0.4)
<i>Amaioua guianensis</i>	2 <sup>nd</sup> 29.3	204.8 (±16.7)	17.9 (±0.7)	3.2 (±0.04)
<i>Tetragastris altissima</i>	3 <sup>rd</sup> 23.6	643.2 (±29.0)	9.6 (±0.6)	2.9 (±0.2)
<i>Himenaëa courbaril</i>	4 <sup>th</sup> 17.0	528.0 (±21.9)	18.2 (±0.6)	8.7 (±0.6)
<i>Mabea fistulifera</i>	7 <sup>th</sup> 13.4	521.6 (±13.5)	14.4 (±0.5)	5.4 (±0.2)
<i>BROSIMUM MONO-DOMINANT EVERGREEN FOREST</i>				
<i>Brosimum rubescens</i>	1 <sup>st</sup> 131.9	243.2 (±16.9)	18.3 (±0.8)	4.1 (±0.4)
<i>Amaioua guianensis</i>	2 <sup>nd</sup> 29.9	275.2 (±13.0)	19.2 (±0.4)	5.1 (±0.04)
<i>Cheilochlinium cognatum</i>	3 <sup>rd</sup> 29.6	536.0 (±12.6)	9.9 (±0.2)	2.6 (±0.2)
<i>Tetragastris altissima</i>	4 <sup>th</sup> 18.3	848.0 (±20.4)	9.3 (±0.2)	3.6 (±0.2)
<i>Ephedranthus parviflorus</i>	10 <sup>th</sup> 4.2	684.8 (±13.5)	18.1 (±0.1)	11.2 (±0.3)