Hydraulic traits methodology (from Nobby, need to reword—these are part of BCI paper)

*LMA, PLA and WD*

We weighed and scanned three re-hydrated leaves from each individual. We were choosing a small, middle-sized and a large leaf per individual. Petioles were removed before weighing and scanning. Afterwards the leaves were dried in an oven at 60 ̊ C for at least two days. After the drying process we weighed and scanned the leaves again. Leaf mass per area (LMA, kg m-2) was calculated as the ratio of leaf dry mass and the fresh leaf area. Percentage loss of area (PLA, %) was derived as the ratio of the dry leaf area and the dry leaf area. Wood density (WD, g cm-3) was measured on 2-3cm long segments of the re-hydrated branches. We removed the bark and pith and used Archimedes' principle to estimate the volume of the wood. The wood samples were dried in an oven at 60 ̊ C for at least two days and weighed.

*Osmotic determination of the turgor loss point (TLP)*

We used the rapid determination method to estimate the turgor loss point (πtlp) (Bartlett et al., 2012) based on measurements of the sap osmotic potential of leaves (Ball and Oosterhuis, 2005). The osmotic potential was measured with a vapour pressure osmometer (VAPRO 5520, Wescor, Logan, UT, USA). Per tree individual, we collected one sun-exposed branch. Directly after cutting the branches from the trees we placed the branches in humid and opaque plastic bags. The samples were transported to the laboratory as fast as possible and recut underwater at least two nodes distal to the original cut. The recut samples were places in buckets with water, covered with plastic bags again and rehydrated overnight. On the following day, we removed two mature, fully expanded leaf per branch. Two sample discs were cut from the leaf centrally between the midrib and margin with a 4‐mm‐diameter cork borer. Discs were tightly wrapped in aluminum foil and submerged in liquid nitrogen (LN2) for at least 2 minutes (Bartlett et al., 2012). We used the standard 10 μL chamber well of the osmometer. Immediately before putting the discs into the chamber of the osmometer, the discs were punctured with a dissection needle for about 10 to 15 times to improve evaporation through the cuticle and to reduce equilibration time (Kikuta and Richter, 1992). The osmometer was set in the auto repeat mode and measurements were recorded until the equilibrium was indicated by an increase between measurements of < 0.01 MPa (approximately 5 osmometer readings). The turgor loss point (πtlp) was calculated from the osmotic potential given by the osmometer (πosm) using the calibration equation established by Bartlett et al. (2012):

(eq. 1)

Ball, R.A., Oosterhuis, D.M. (2005) Measurement of root and leaf osmotic potential using the vapor-pressure osmometer. Environmental and Experimental Botany 53, 77-84.

Bartlett, M.K., Scoffoni, C., Ardy, R., Zhang, Y., Sun, S., Cao, K., Sack, L. (2012) Rapid determination of comparative drought tolerance traits: using an osmometer to predict turgor loss point. Methods in Ecology and Evolution 3, 880-888.

Kikuta, S.B., Richter, H. (1992) A simplified pressure-volume method for the estimation of osmotic adjustement with the pressure chamber. Bodenkultur 43, 307–318.