Article: Single vessel air injection estimates of xylem resistance to cavitation are impacted by vessel network characteristics and sample length. *Journal:* Tree Physiology. *Authors:* Venturas MD, Rodriguez-Zaccaro FD, Percolla MI, Crous CJ, Jacobsen AL, Pratt RB. *Corresponding author:* Martin Venturas, Forest Genetics and Ecophysiology Research Group (GENFOR), School of Forest Engineering, Technical University of Madrid, 28040 Madrid, Spain, martin.venturas@gmail.com

SUPPLEMENTAL MATERIAL



Figure S1. Air volume flow rate delivered by the pulled microcapillaries when they are connected to the single vessel air injection system. The volume rate was calculated by measuring the time it took for the air injected through the pulled microcapillaries to displace the water from a 4.5 mL glass pipette. The equations correspond to the linear fits for each of the capillaries. The mean slope of the flow rate for the pulled capillaries was $41.5 \pm 13.9 \text{ mL MPa}^{-1} \text{ min}^{-1}$ (\pm SE; n=4). The mean flow rate at 0.05 MPa (the minimum pressure at which vessels were injected) was $2.0 \pm 0.9 \text{ mL min}^{-1}$ (\pm SE; n=4).

The capillaries, even the narrowest ones, were capable of injecting enough gas to fill even the largest vessels within samples within only a few seconds or less. Thus, it is not likely that the gas delivery rate impacted measures or that there was a long delay between injection and equilibration of an injected vessel with the injected pressure.

Once the injected vessel was filled, vessels were capable of holding the gas at pressure until the air-seeding threshold was reached. We tested this by injecting a vessel and bringing it up to a low pressure that was below the air-seeding point and holding at that pressure for >10 min. During that time, no gas emerged from vessels at the opposite end of the injected stem. Only once the pressure was increased to the point of air-seeding did gas emerge (data not shown).



Figure S2. Relationship between sample size and median seeding pressure (P_m) for three species. Upper and lower confidence intervals (CI) estimates for each sample size were established by bootstrapping. We obtained 10,000 subsamples of each size by repeated random sampling with replacement from the measured pool of seeding pressures. Then we calculated P_m of each subsample and established the 95% CI for each sample size as the 2.5 and 97.5 percentiles of P_m of the 10,000 subsamples. The maximum bootstrapping sample size for each species and segment length was equal to the total number of observed measurements.