

Plant plumbing: How do leaves design their vein networks?

1) Introduction

Plant leaves 'sweat' during the day; losing water through the pores which they open to 'breathe in' CO₂. In warmer climates, some species have evolved CAM (Crassulacean Acid Metabolism), an adaptation allowing them to conserve water by 'breathing' during the cooler night (Figure 1) (Borland et al. 2015). The water conserved using CAM varies between species, depending on the extent to which they use it to 'breathe' at night. The water conserving properties of CAM could have important implications for vein networks, the leaf plumbing system that replaces lost water, yet this is something that has never been studied before.

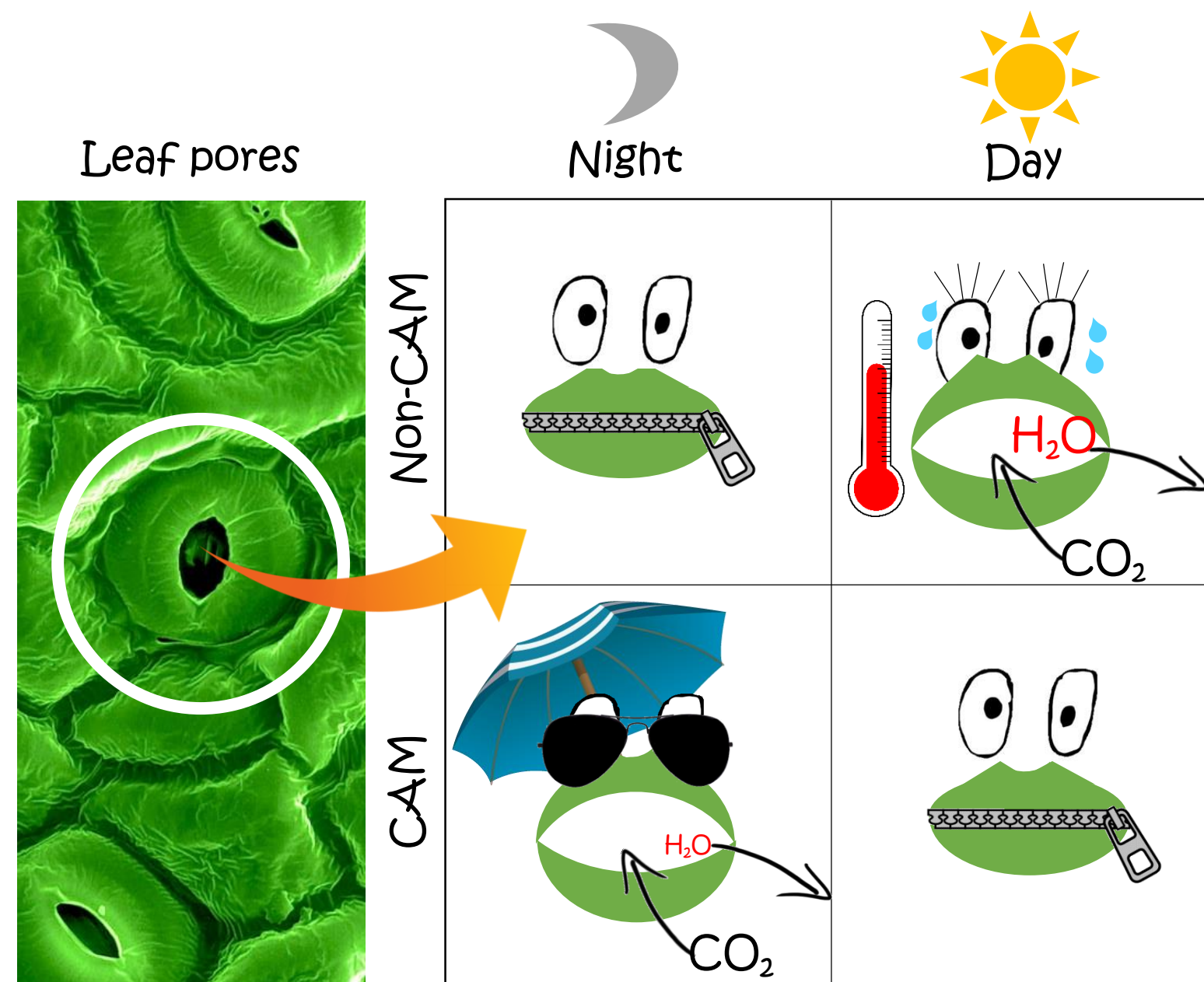


Figure 1- The pattern of opening and closing the pores in leaves of CAM and non-CAM plants.

2) Aims

- To test the hypothesis that vein density would be significantly lower in the leaves of species doing the most night-time CO₂ uptake (i.e. using the most CAM).
- To use this knowledge to contribute to work done as part of the CAMBioDesign project at Newcastle University. This project aims to better understand the properties of CAM plants and exploit these for engineering plants more tolerant of drought, which poses a future challenge to global agriculture.

3) Methods

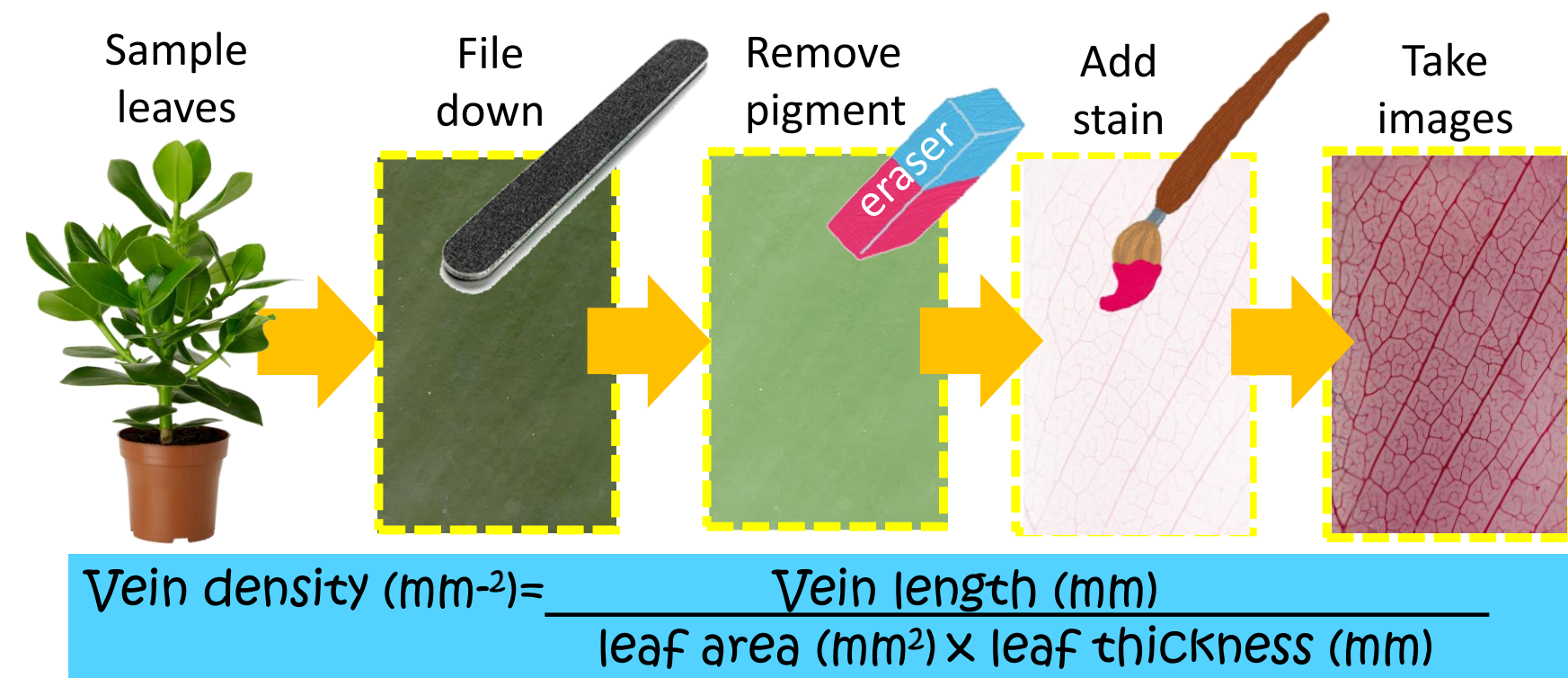


Figure 2- The steps taken to visualise the vein network (Scoffoni and Sack 2013) and calculate the vein density in samples of *Clusia* leaves.

- Using Newcastle University's collection of *Clusia* species, which is the largest in Europe, leaf vein density was measured for 10 species including non-CAM, intermediate-CAM and full-CAM species of *Clusia* tree.
- As shown in Figure 2, samples from the leaves of each species were filed, cleared of green pigment and then dyed with a pink stain, which bound preferentially to the veins to reveal the network.
- Using scanner and microscope images, the vein length was measured manually and vein density was calculated using the equation above (Figure 2).

4) Results and Discussion

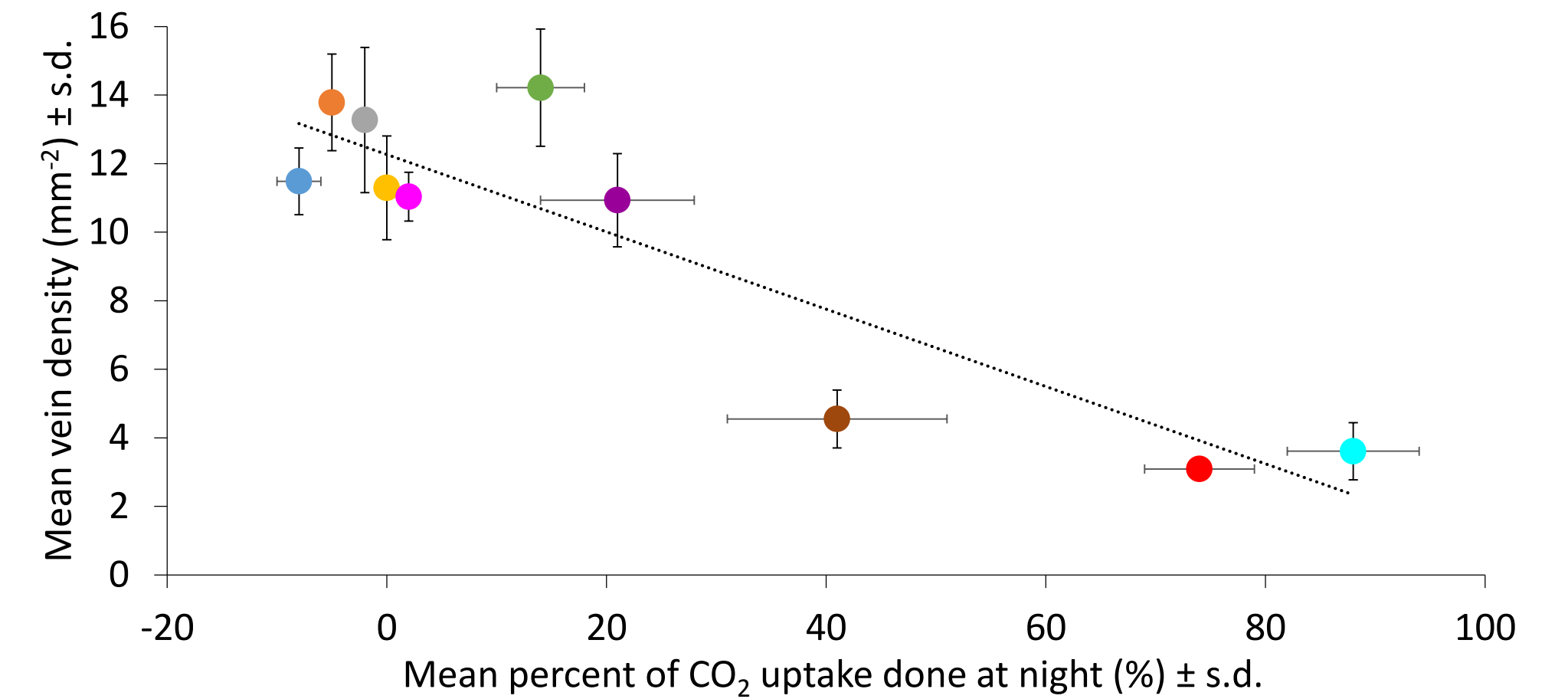


Figure 3- The change in leaf vein density (mm²) with increase in the percentage of CO₂ uptake at night (%) across 10 different species (n=9) of *Clusia* tree. Error bars represent ±1 standard deviation from the mean.

The species which used CAM to 'breathe' the most during the night had leaves with a significantly lower vein density. Since CAM plants lose less water than non-CAM species, fewer veins would be needed to replace this water, resulting in lower vein density. This supported the initial hypothesis and showed how influential CAM was to the design of the vein network.

5) Visual Summary of Results

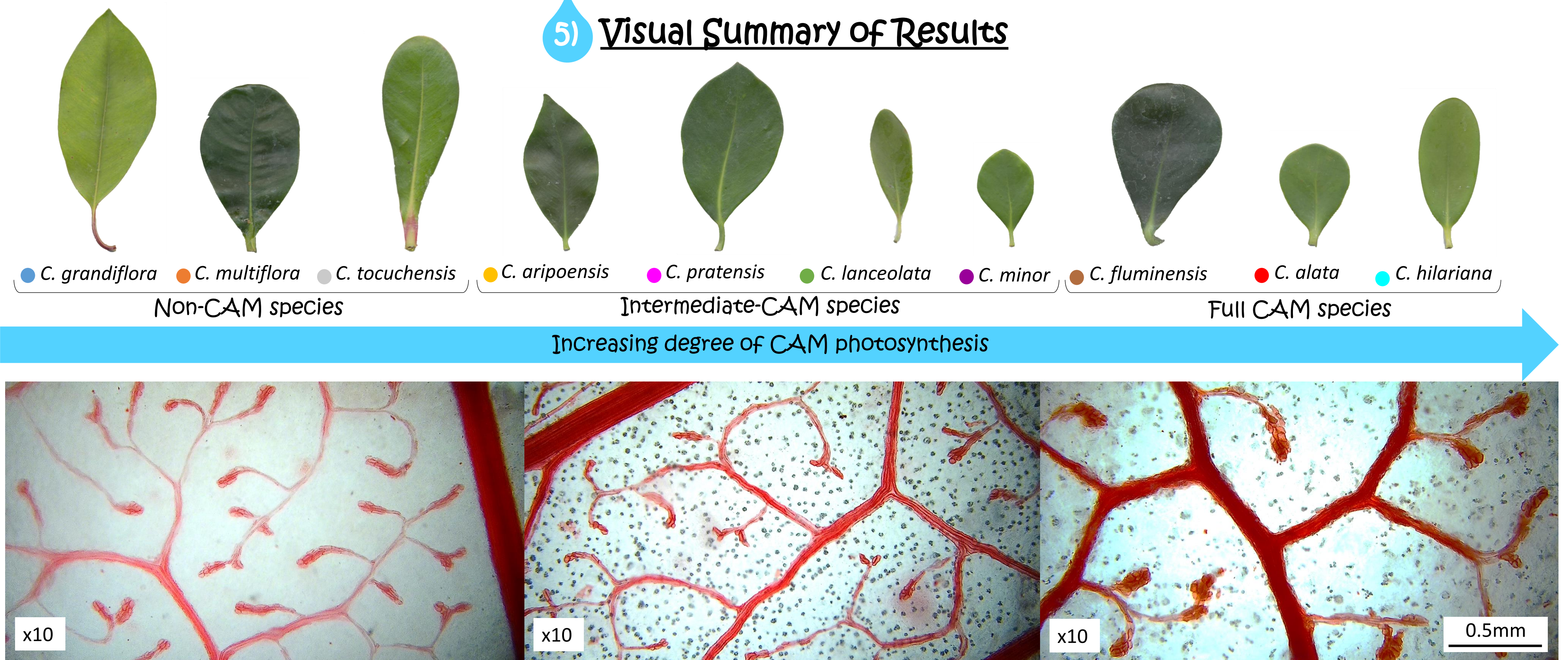


Figure 4- The decrease in vein density (mm²) with increasing degree of CAM photosynthesis (or increasing night time CO₂ uptake) across the 10 different species of *Clusia* studied. Vein density images were all taken at x10 magnification and the characteristic leaves of each species are shown, labelled with the coloured dots that correspond to the data represented in Figure 3.

6) Future work

With changing rainfall patterns expected in future, this work has shown that plant species which have leaves with a lower vein density will be most suitable for hosting CAM, which researchers are endeavouring to engineer into traditional crops to improve their drought tolerance. CAM engineering will make agriculture more sustainable in future and these results were presented at the International Xylem Meeting in Bordeaux in September, to inform further work within the field of plant hydraulics.