

# Day-night root dynamics change through the growing season

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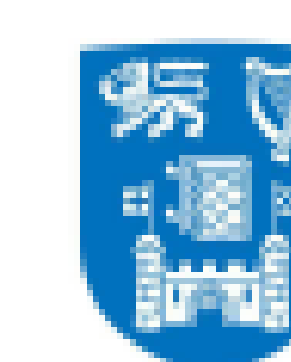
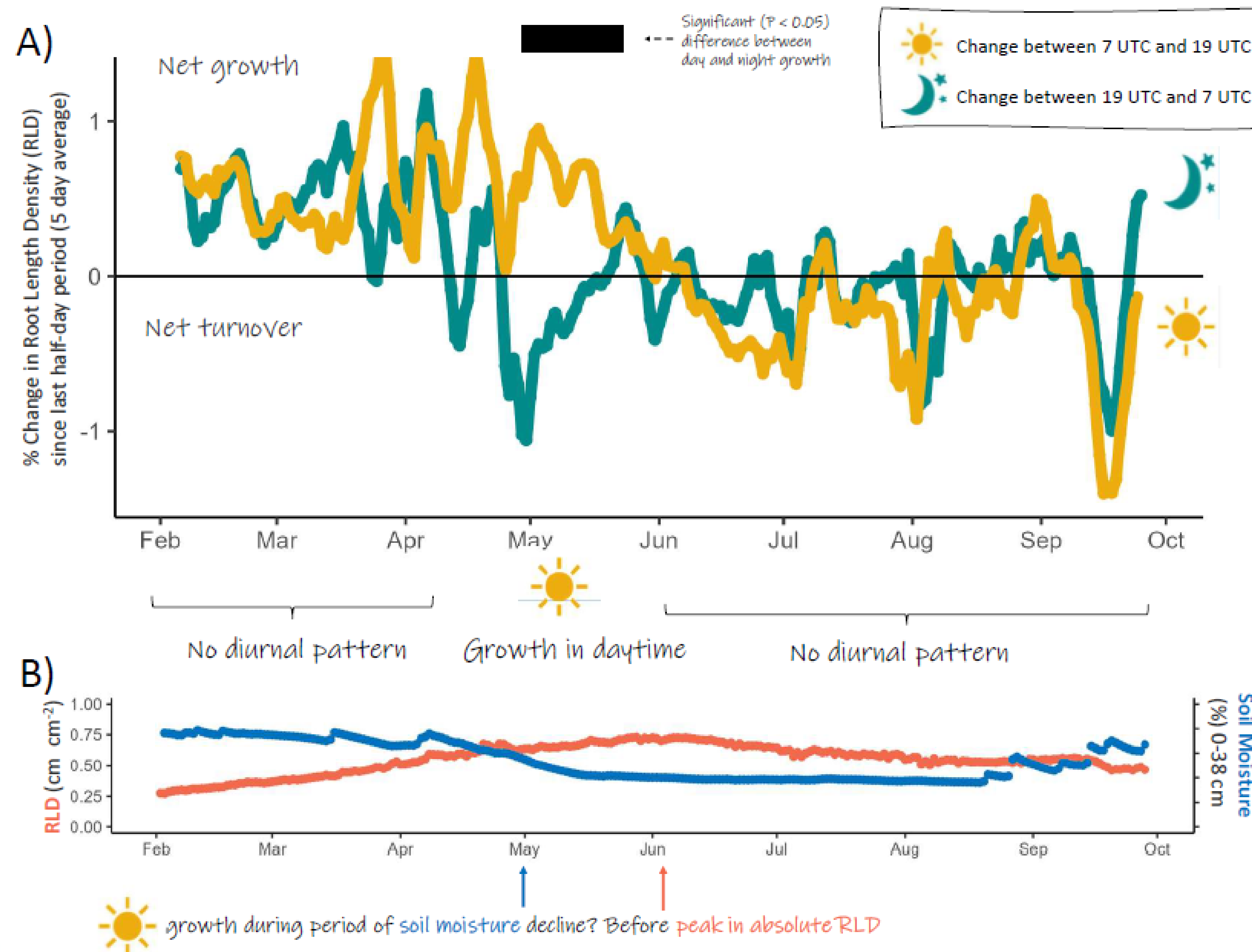
Are roots growing **during the day** ☀ or are roots growing **at night** 🌙?

We observed sub-daily root growth with **automated minirhizotrons** and **machine learning** in a temperate grassland, analysing sequential change between ~ 270 000 high frequency root images.

Plant growth is influenced by **source and sink limits**. Carbon transport in herbaceous plants is often rapid. Shifts in diurnal root growth (A) in early summer despite ongoing RLD increase (B) suggest a changing control mechanism, possibly due to fresh photosynthate supply or cell turgor limits<sup>4</sup>.

Growth timing matters for **understanding ecosystem processes and dynamics**. Soil respiration may have different contributions by roots depending on when they are made. Night-time EC partitioning splits daytime fluxes using night-time respiration temperature sensitivity. If growth timing shifts diurnally, night-time is not representative of day-time processes.

<sup>4</sup> Carbohydrate supply or transpiration-driven turgor limits? GPP and T are tightly linked. There is evidence that leaf growth is more driven by hydraulics than C – but is it the same at the other end of the vascular pathway?



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# Online Supplementary Information



# Methods

We used [homemade automated minirhizotrons](#) and sampled six times a day in a floodplain grassland in Jena, Germany for 268,000 individual images over 9 months.

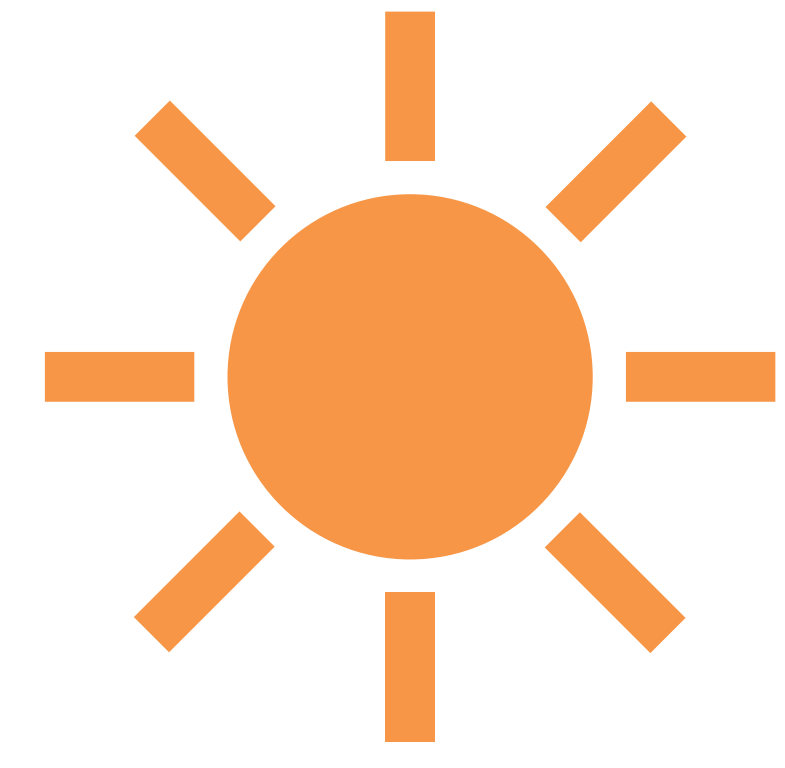
We processed all the images in a using the [rootpainter CNN](#) and custom scripts to extract root traits - including the RLD shown here. We use RLD as this was stable to extract. In a grass dominated ecosystem this correlates well to root surface area.

We controlled for diurnal variation in root diameter and selected two time periods used in this study. **Night time** was growth between **7pm and 7pm** and **Day time** between **7pm and 7am**.

We show a 7 day rolling average of A) change in RLD, B) total RLD and (without rolling average) soil moisture from sensors at 0-37 cm. Sign tests A) are between independent day-night pairs in each 7 day periods.



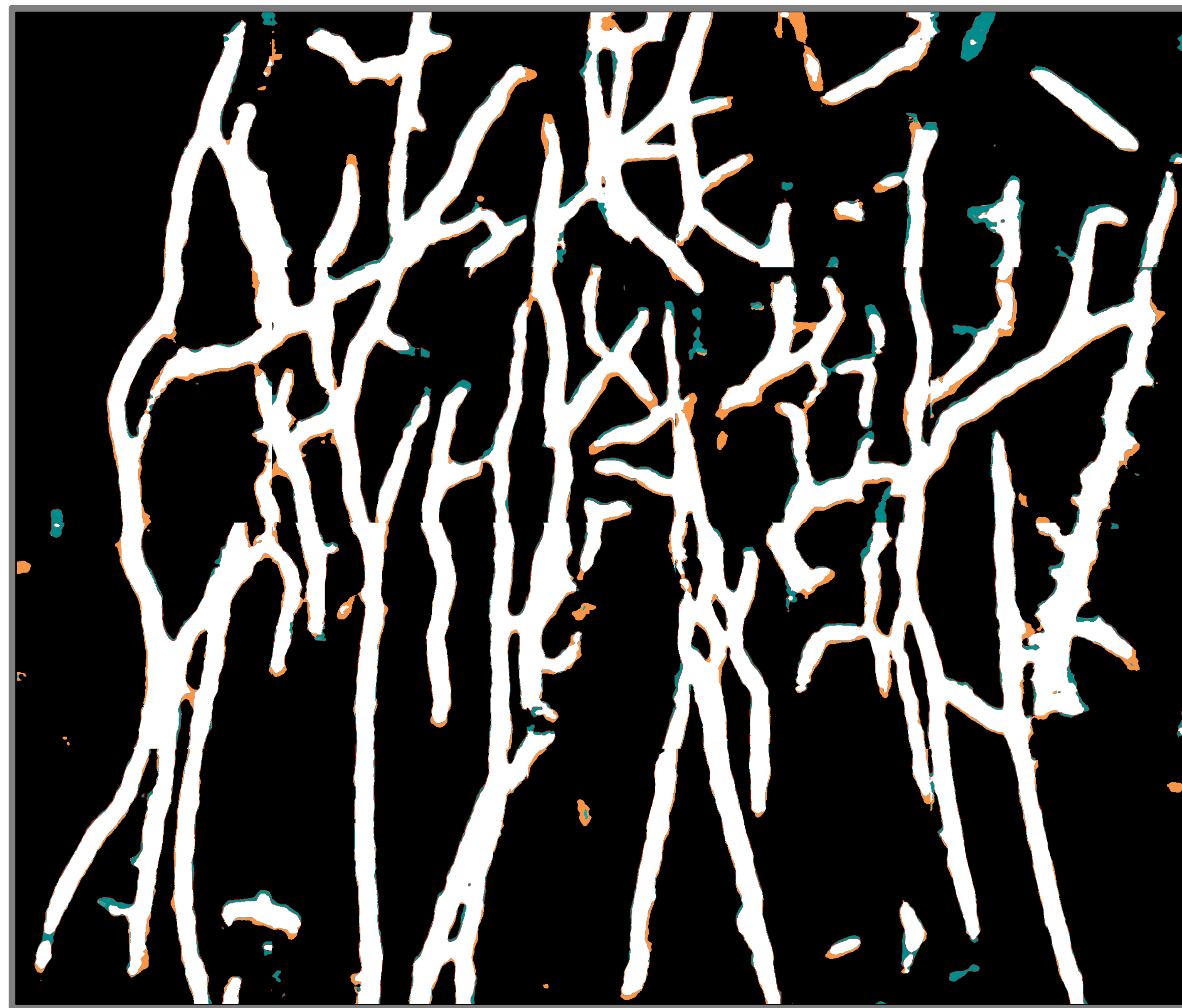




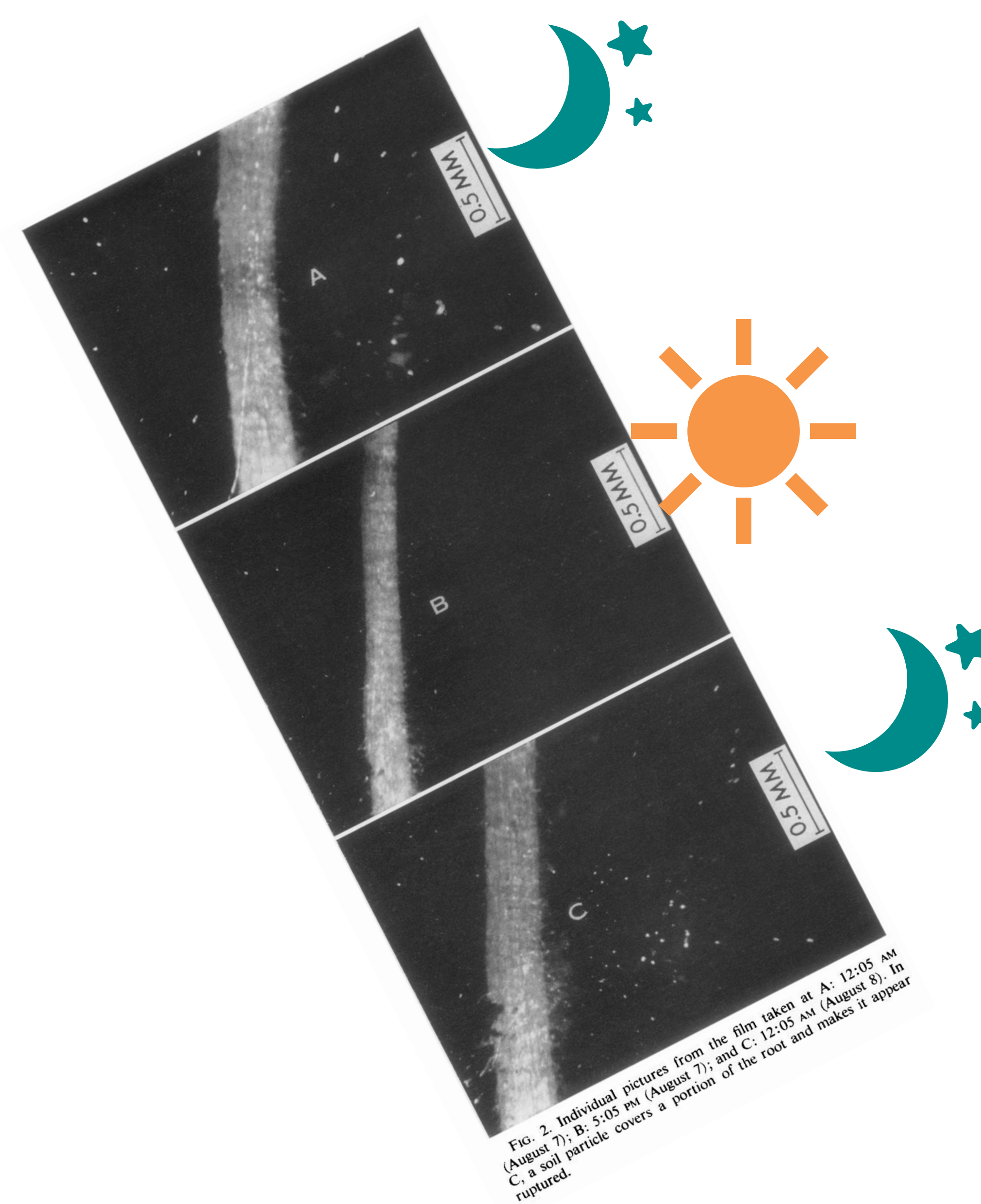
# Eliminating diurnal variation



Despite efforts to eliminate segmentation bias in our minirhizotron observations, we found a weak diurnal cycle that we attribute to root cell turgor matching  $\Psi$  differences in leaves. Daytime transpiration drains water from roots, making them appear smaller later in the day. To overcome this bias, we used the two time points closest in size, around dawn and dusk (7am and 7pm)



A composite of two segmentations from **day** and night observations shows **areas only segmented as roots during the day** and **areas only segmented as roots at night**. Roots segmented at both times are white.



Real variations in fine root diameter driven by ecohydrology ([Huck et al. 1970](#))

