

Quantitative analysis of microtubule dynamics in *Arabidopsis thaliana* aerial organs using Light Sheet Fluorescence Microscopy

Matthieu Cortes, Claire Lionnet, Igor Lyuboshenko, Olivier Hamant, Christophe Tréhin

Laboratoire de reproduction et développement des plantes
ENS de Lyon
46, allée d'Italie, 69364 LYON Cedex 07 France
E-mail : matthieu.cortes@ens-lyon.fr

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LSFM is increasingly used in live imaging, as a good compromise between resolution, time of acquisition, photobleaching and phototoxicity. Indeed, only the focal plane of the LSFM samples are illuminated by a sheet of light, making the acquisition much faster and less phototoxic. Limitations of the techniques include the size of the sample (only one sample at a time) and embedding of the sample in agarose. For these reason, in plants, LSFM has been mainly used to image roots so far (1). Here we adapted a LSFM (Alpha3, Phaseview) to image aerial organs and were able to image up to 7 plant shoots with the same set-up. As a proof of concept, we focus our work on microtubule dynamics during tissue morphogenesis and cell division. We also combined this LSFM approach with quantitative image analysis using software like MorphographX (2) or Fibriltool (3). Altogether, this provides a competitive alternative to confocal microscopy for aerial organs.

1 – B. Berchet , A. Maizel « Light sheet microscopy and live imaging of plants » *Journal of microscopy* , (28 March 2016).

2 – Pierre Barbier de Reuille, Anne-Lise Routier-Kierzkowska, Daniel Kierzkowski, George W Bassel, Thierry Schüpbach, Gerardo Tauriello, Namrata Bajpai, Sören Strauss, Alain Weber, Annamaria Kiss, Agata Burian, Hugo Hofhuis, Aleksandra Sapala, Marcin Lipowczan, Maria B Heimlicher, Sarah Robinson, Emmanuelle M Bayer, Konrad Basler, Petros Koumoutsakos, Adrienne HK Roeder, Tinri Aegerter-Wilmsen, Naomi Nakayama, Miltos Tsiantis, Angela Hay, Dorota Kwiatkowska, Ioannis Xenarios, Cris Kuhlemeier, Richard S Smith « MorphoGraphX: A platform for quantifying morphogenesis in 4D », *eLife* , 2015

3 – Boudaoud, A., Burian, A., Borowska-Wykręć, D. *et al.* FibrilTool, an ImageJ plug-in to quantify fibrillar structures in raw microscopy images. *Nat Protoc* **9**, 457–463 (2014)
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