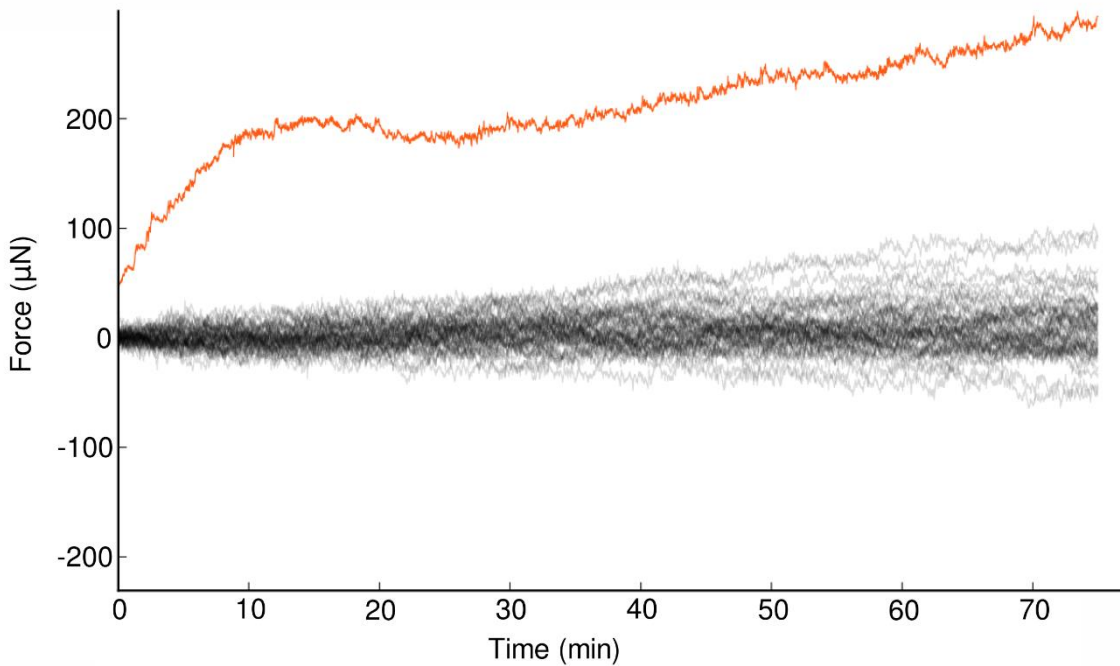
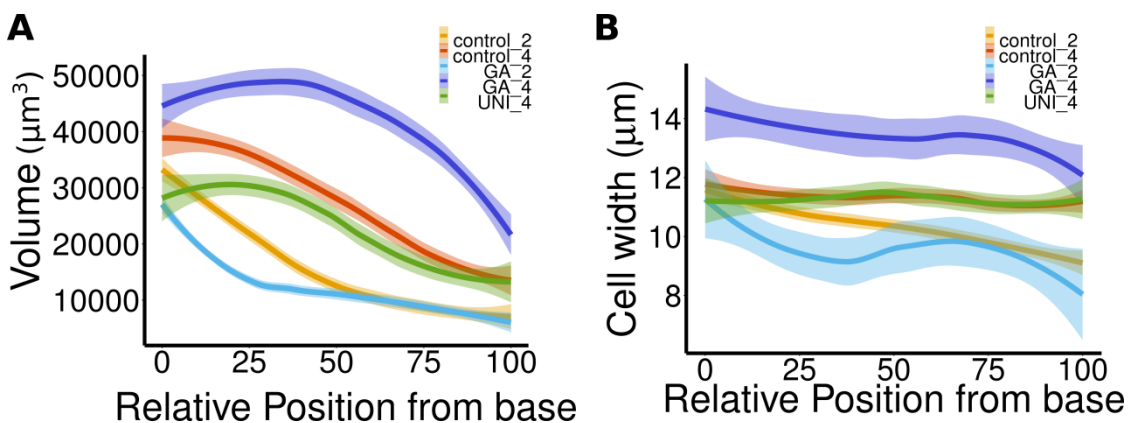


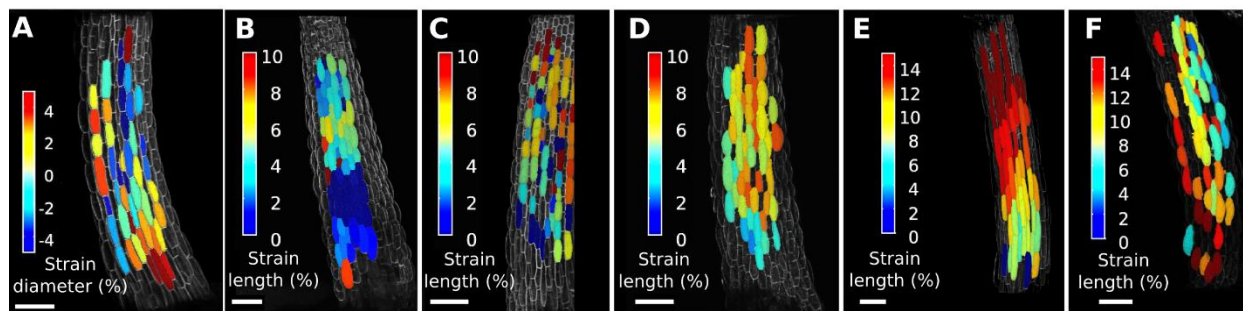
Supplemental Figure 1. ACME is easily assembled from a combination of custom and commercially available parts. Supporting Figure 1) **(A)** An image of the parts required to build ACME robot. **(B)** An exploded view of ACME robot to show how the parts are assembled (see ACME_AssemblyGuide.pdf for more information).



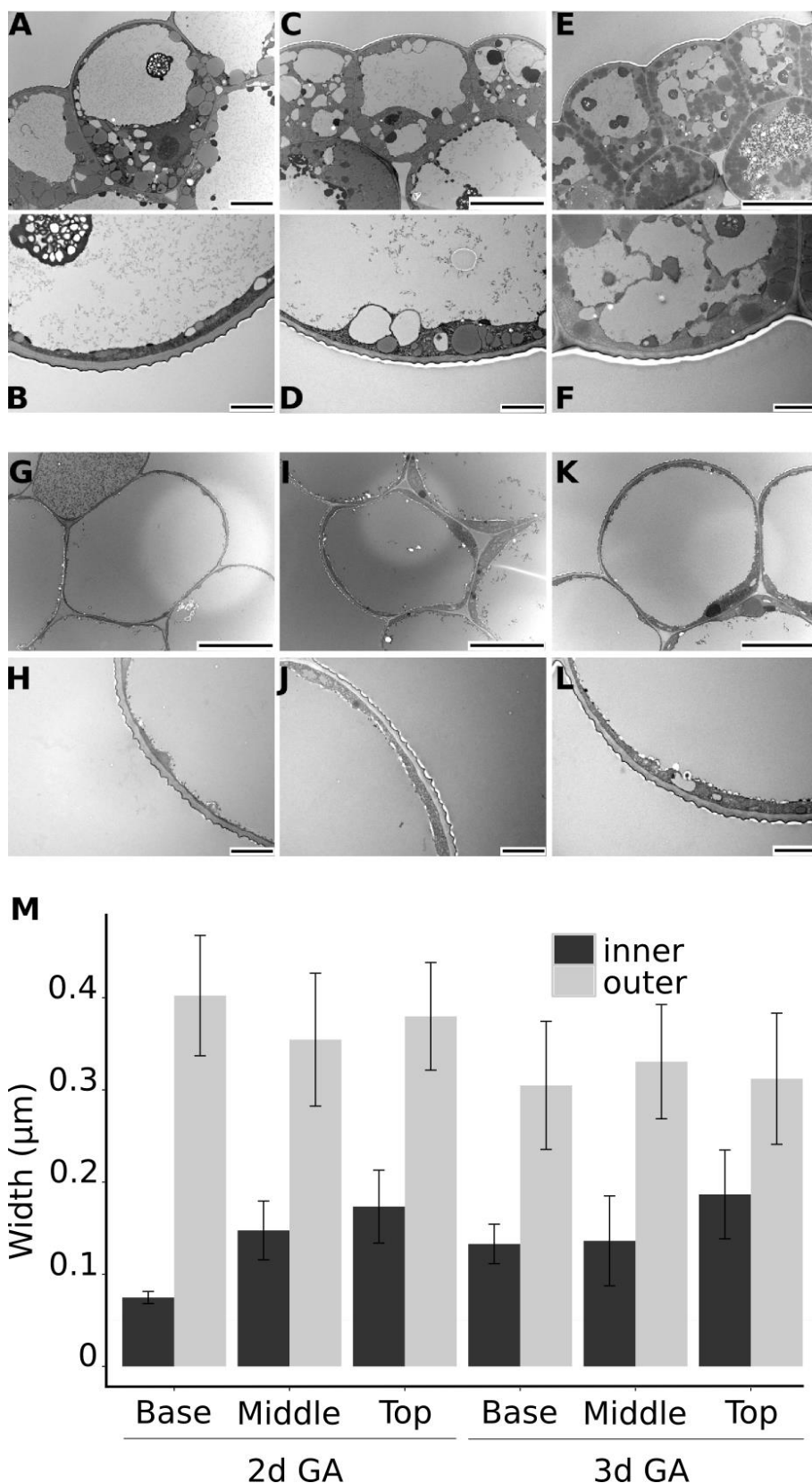
Supplemental Figure 2. Drift assessment of ACME. Drift was measured by holding the position of the plates and measuring the force. Supporting Figure 1). Fifty-one runs were performed sequentially. It was found that drift was highest in the first run (red) but after this (black) it dropped to below 100 μN per hour. This is 10% of the smallest force typically applied.



Supplemental Figure 3. Quantifying cell size and dimensions along the hypocotyl (Supporting Figure 3). **(A)** Cell volume and **(B)** cell width as a function of relative position along the hypocotyl for the same data as in Figure 3F. Means are shown plus 99% confidence interval.



Supplemental Figure 4. Using ACME to reveal spatial gradients in mechanical properties. Supporting Figure 5). **(A–E)** Creep tests were performed by applying 10 mN of force to three DAS live, turgid hypocotyl samples. Z-stacks were acquired and the images were segmented. Corresponding cells were co-labeled before and after stress was applied. Using PC analysis the length and diameter of the cells were computed and used to compute the percentage strain in length and diameter per cell, shown as a heatmap. **(A)** The change in diameter was small and often negative. The same hypocotyl as Figure 5G. **(B–E)** The GA treated samples showed spatial gradients in percentage strain in length in five out of six samples analysed, including the sample in Figure 5G. **(F)** One sample did not show the gradient as it was curved. This sample shows greater strain on the inside of the curve compared to the outside of the curve, as the hypocotyl straightens during the experiment. Scale bars = 100 μ m.



Supplemental Figure 5. Cell wall thickness measured from TEM images (see Methods) Supporting Figure 5). Representative images at the base (A, B, G, H), middle (C, D, I, J) and top (E, F, K, L) of two (2d; A–F) and three (3d; G–L) days after stratification GA treated seedlings. (B, D, F, H, J, L) are close ups of (A, C, E, G, I, K). (M) Cell wall thickness was measured in ImageJ in the inner and outer wall of the epidermal cells. Bars

Supplemental Data. Robinson et al. (2017). Plant Cell 10.1105/tpc.17.00753

show mean \pm SD. $n \geq 8$, from two seedlings. (**A, B, D, F, H, J, L**) scale bars = 2 μm , (**C, E, G, I, K**) scale bars = 10 μm .

Supplemental File 1. A complete guide to assembling and using ACME, and analyzing the resultant data.

Movies

Movie 1. Demonstration of sample mounting. Two tough tags (TTSW-1000, DiversifiedBiotech) are positioned on their backing paper with a gap between them equal to that of the ACME arms. The seedling is placed across the gap. Two strips of tough tag are put on top of the seedling parallel to the original ones. The tough tags are then used to stick the sample to ACME which is already on the confocal microscope and the backing paper is removed.

<http://www.botany.unibe.ch/deve/ACME/Movie1.avi>

Movie 2. Adding glue for high force measurements (> 5 mN). The sample is prepared as in Movie 1, except a small amount of cyanoacrylate glue is added to the small tough tag strips, this step is shown.

<http://www.botany.unibe.ch/deve/ACME/Movie2.avi>

Guide to ACME assembly

ACME_AssemblyGuide.pdf Detailed instructions on the ACME robot parts and how to assemble them.

http://www.botany.unibe.ch/deve/ACME/ACME_AssemblyGuide.pdf

LoadCellSignalAcquisitionSystemGuide.pdf Detailed instructions on how to connect the load cell, amplifier and PCI card.

<http://www.botany.unibe.ch/deve/ACME/LoadCellSignalAcquisitionSystemGuide.pdf>

ACMECalibrationGuide.pdf Instructions on how to calibrate the load cell before the first use.

<http://www.botany.unibe.ch/deve/ACME/ACMECalibrationGuide.pdf>

calibration_worksheet.ods Used to calibrate the load cell (for details see ACMECalibrationGuide.pdf)

http://www.botany.unibe.ch/deve/ACME/calibration_worksheet.ods

3D_printer_parts.zip Contains the files necessary to print the ACME parts.

http://www.botany.unibe.ch/deve/ACME/3D_printer_parts.zip

Download page for the ACME software Contains custom software the ACMEtracker, example protocols and parameter file

<https://github.com/ACME-Robinson/InstallPackage>

R scripts for data analysis

oscillations.R Used with ExtractOsc.R to compute strain from the csv file generated by the ACMEtracker.

<http://www.botany.unibe.ch/deve/ACME/oscillations.R>

ExtractOsc.R Used with oscillations.R to compute strain from the csv file generated by the ACMEtracker.

<http://www.botany.unibe.ch/deve/ACME/ExtractOsc.R>

plotACMEdata.R Generates graphs of force and position against time from the csv files generated by the ACME software.

<http://www.botany.unibe.ch/deve/ACME/plotACMEdata.R>