phenoVein - A software tool for leaf vein segmentation and analysis

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phenoVein is a software tool dedicated to automated segmenting and analyzing images of leaf veins. It includes comfortable manual correction features. Advanced image filtering automatically emphasizes veins from background and compensates for local brightness inhomogeneities. Phenotypical leaf vein traits being calculated are total vein density, vein lengths and widths and skeleton graph statistics. For determination of vein widths, a model based vein edge estimation approach has been implemented. We demonstrate the capabilities of phenoVein on a set of vein structure mutants of Arabidopsis thaliana.
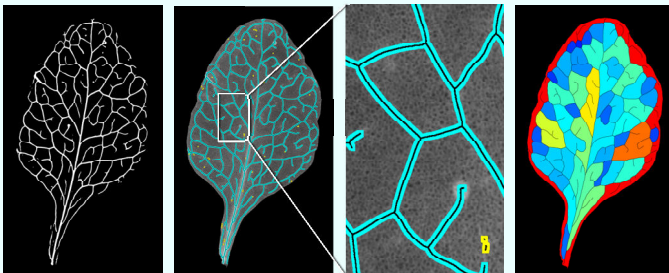
Automated vein segmentation
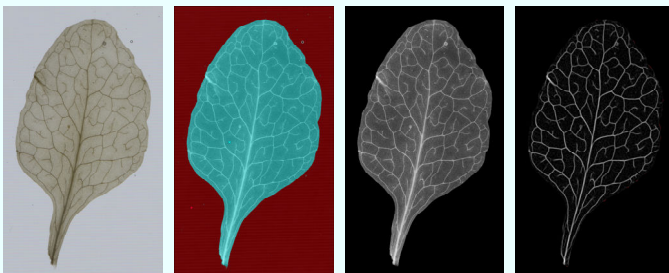
Original image of a cleared leaf of Arabidopsis thaliana (*hve2*, leaf 6, 30 DAS)

Leaf / background segmentation, determination of leaf area

Conversion from color to gray value image, e.g. green channel from RGB

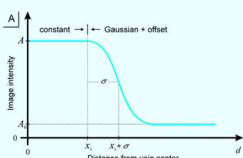Vesselness filtering of vein structures and compensation of irregular lighting

Threshold based vein mask
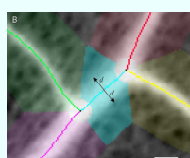
Skeletonization of the vein mask

Magnification of skeleton subsection

Color encoded areole sizes

Vein width estimation

Sketch and parameters of vein edge model. The vein is assumed to have a constant brightness. The edge and the areole are modeled by a smooth half-Gaussian

Inverted subsection of leaf image. Vein piece of interest and surrounding area (next closes pixels) are highlighted in cyan.
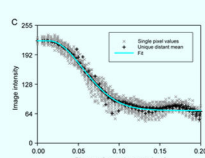
Pixel intensities vs shortest distance to skeleton (gray). The vein edge model (cyan) is least squares fitted to the pixel intensities.

Vein edge function:
$$v_e(x) = \begin{cases} A & x < x_s \\ A_0 + (A - A_0)\exp\left(-(x - x_s)^2/(2\sigma^2)\right) & x \ge x_s \end{cases}$$

Mean vein width:
$$w_i = 2\left(x_s + \sqrt{2\ln(2)}\sigma\right)$$

Leaf vein traits

Important vein traits

Total vein length
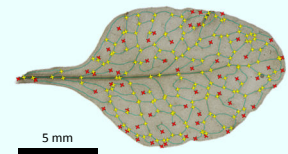Projected leaf area
Leaf vein density
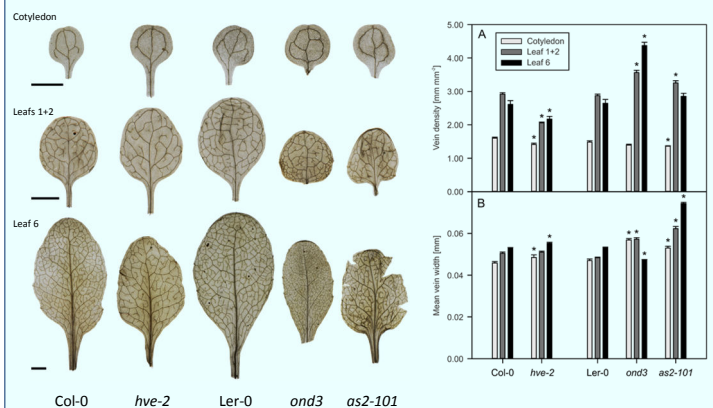Mean vein width
Vein graph statistics
 - Branching points
 - Free ending points
 - Areoles

5 mm

Total skeleton length	242.7	mm
Leaf area	136.4	mm^2
Vein density	1.78	mm mm^{-2}
Mean vein width	50.3	μm
Single skeleton pieces	273	
Free ending points	63	
Branching points	161	
Areoles	52	

Analysis of venation mutants

Cotyledon

Leafs 1+2

Leaf 6

Col-0   *hve-2*   Ler-0   *ond3*   *as2-101*

Photographs of cleared leafs of Arabidopsis thaliana mutants. Cotyledons (14 DAS), pooled leafs 1+2 (14 DAS) and leaf 6 (30 DAS). Scale bar = 2.5mm

Main results of vein analysis. Vein density and mean vein width of different genotypes. * indicate statistically significant differences ($p < 0.05$).

Software

phenoVein is a plug-in for MeVisLab, the source code is freely available as open source software.

phenoVein can be downloaded from:
www.phenovein.de

Publication of phenoVein:
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