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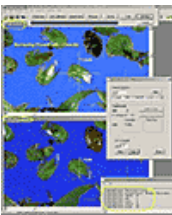


Image-Pro Plus used to automate insect classification

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Customer Spotlight

Image-Pro Plus Used to Develop Customized Root Cross-section Analysis Application

Georg von Arx, a plant ecologist of the Geobotanical Institute, Swiss Federal Institute of Technology ETH, in Zurich, Switzerland studies the root rings of perennial forbs. Annual rings in the roots of perennial forbs are an integrated record of a plant's life that are of great value to plant ecologists, population biologists and (eco-) physiologists. Discernible annual rings are characterized by wide earlywood vessels (water transporting structures of plant roots or shoots) formed in spring and narrow latewood vessels formed in late summer and autumn.

Up until now, analysis of these root rings involved manual measurements which were performed along three radii of root cross sections. Means of the three measurements were used as the width of the respective rings. If radial growth was asymmetric around the root, this approach could lead to large errors in the estimation of ring width and inconsistencies between different observers.



Georg von Arx

To extend and improve analysis of annual rings, von Arx used the programming language in Image-Pro Plus to co-develop a customized image analysis system called ROXAS[®] (Root Xylem Analysis System), which performs automated analysis of root cross-sections (von Arx & Dietz 2005 - Automated image analysis of annual rings in the roots of perennial forbs - International Journal of Plant Sciences: 166: 723-732). The Image-Pro Plus scripting language enabled von Arx to create processing and measurement commands and parameters specific to root cross-section analysis.

Von Arx captures root images with a digital camera (Nikon CoolPix 990, 3.34 Megapixels) through the receptacle tube of a dissecting microscope (Leica MZ 8; Fig. 1). Using ROXAS, the images are corrected for uneven illumination. Contrast of vessels is enhanced using several image and edge enhancement procedures. Segmentation is performed and vessels are extracted from the resulting image using several morphometric parameters (Fig. 2, 3). The local neighborhood of each vessel is analyzed in an anisotropic manner to search for vessels lying at the annual ring borders (Fig. 4). Afterwards, ring identification algorithms connect the obtained vessels and form continuous ring borders. Then, each vessel is attributed to the corresponding ring (Fig. 5).

From the core data (size and position of each vessel and/or trajectories of annual rings), many other anatomical parameters are automatically deduced and saved into a Microsoft Excel spreadsheet, such as mean ring width, ring area, number and size of vessels of each ring, vessel density, and the percentage of area occupied by vessels. Since the Image-Pro Plus scripting language is compatible with Microsoft[®] Visual Basic[®], the ROXAS application is able to easily export analysis data to Microsoft[®] Excel.

"With this automated procedure, not only the age and the width of perennial forbs can be determined, but also many anatomical features related to annual root rings and vessels," said von Arx. "With this additional data, new approaches to plant ecology are possible that were previously only accessible with laborious manual analysis."

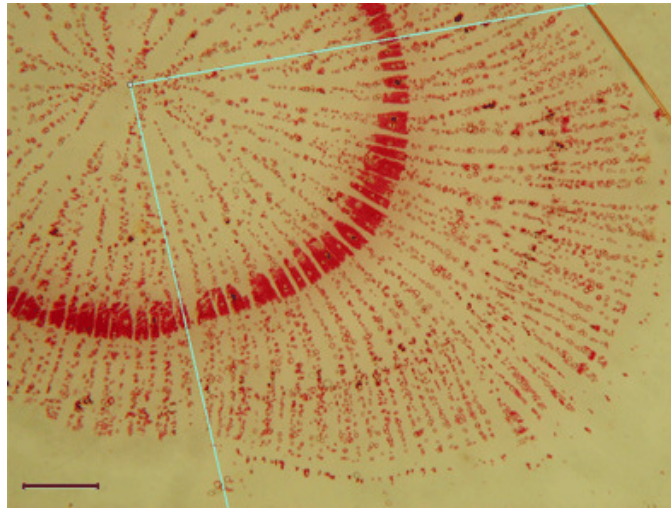


Fig. 1: Root cross-section of *Bunias orientalis*. The lignified (=woody) vessel walls are colored red by a specific stain (Phloroglucinol-HCl). Light-blue lines indicate the analyzed sector.

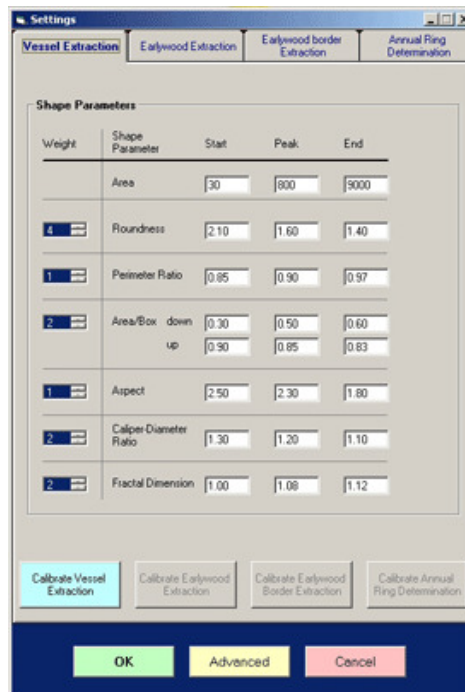


Fig. 2: Example of "ROXAS" dialog window, which includes several morphometric shape parameters used to distinguish vessels from image noise or artifacts.

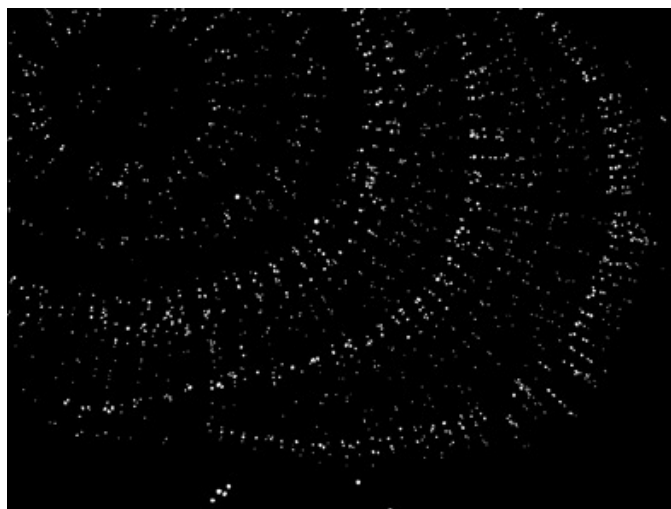


Fig. 3: Result of vessel extraction by ROXAS.

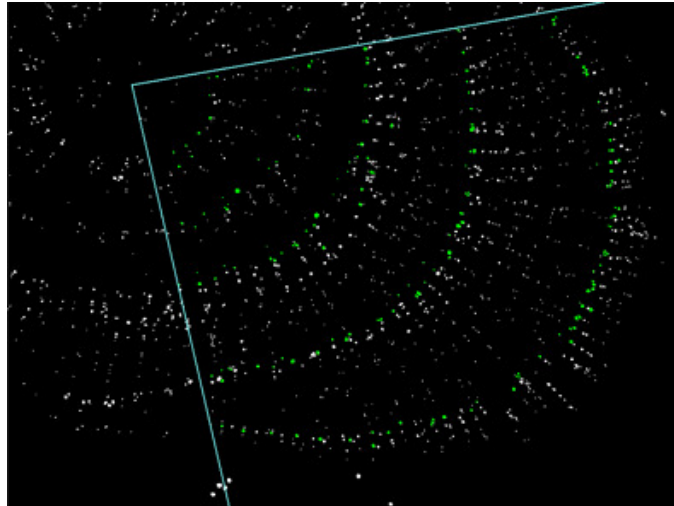


Fig. 4: Pattern recognition algorithms screen the local neighborhood of each vessel and extract the first (earlywood) vessels (green objects) of each annual ring within the analyzed sector indicated by the two radial lines.

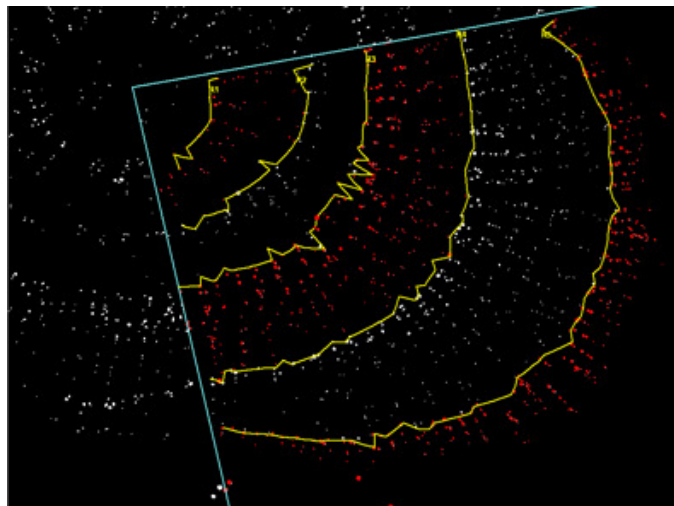


Fig. 5: Vessels of consecutive rings alternately colored white and red to improve visual contrast. The yellow line indicates the ring border as recognized by ROXAS.

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