

P-017: Spatiotemporal chemical cartography of plant cell wall dynamics during growth and after gravitropic stress

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The plant cell wall is composed of a number of different polymers such as cellulose, hemicelluloses and lignin. Its composition, as well as architecture, are dependent on the stage of development, the tissue considered, and culture conditions. Abiotic stress can also greatly modify cell walls. Flax is an excellent model for studying cell wall biology as it produces both cellulose-rich and lignified secondary cell walls. The use of vibrational spectroscopies to study cell walls were reported in flax literature on isolated, scutched or woven elementary fibers [1-5], but rarely on stem transversal sections [6].

Here, we present the use of FT-IR spectroscopy to analyze cell walls in flax stem transversal sections during optimal growth conditions or after a gravitropic stress. A three-step method was used: **A**) spectra acquisition, **B**) PCA analyses, and **C**) chemical cartography through FPA (Focal Plan Array).

A bank of spectrotypes was collected during optimal growth (1.5, 2, 3 months) for 6-cell types with secondary cell walls: bast fibers (BF), bast fiber junction, young xylem ray cells and vessels, mature xylem ray cells and vessels). Analyses of data indicated that average spectra depend on cell wall type and on their level of differentiation (ontogenic status) as expected, but also, on the chronological age of the plant. PCA analyses of FT-IR spectra allowed us to determine five significant windows of bands that discriminated cellulosic- (cluster A) *versus* lignified- (cluster B) secondary cell walls. Within a given cluster, cell types could also be clearly distinguished according to plant age. The spatiotemporal distribution of these significant bands were then imaged through FPA allowing us to monitor bast fiber (BF) and xylem differentiation during optimal growth conditions (Fig. 1).

The effect of a gravitropic stress was also investigated (45° bending during 6 weeks). Important morphological alterations of BF phenotypes were induced and the occurrence of a G-layer in xylem was noticed as previously reported [7]. PCA analyses based on such morphometric parameters established that tension pole data were clearly in a separate cluster whereas control and opposite pole data were closer together. FPA chemical cartography was performed using the 5 previously established windows. The results obtained provide new information on the modifications in cell wall metabolism underlying the observed phenotypic modifications.

Overall our results confirm that vibrational spectroscopies, combined with statistical analyses and chemical cartography, are a powerful tool to decipher changes in cell wall metabolism during development and in response to the environment.

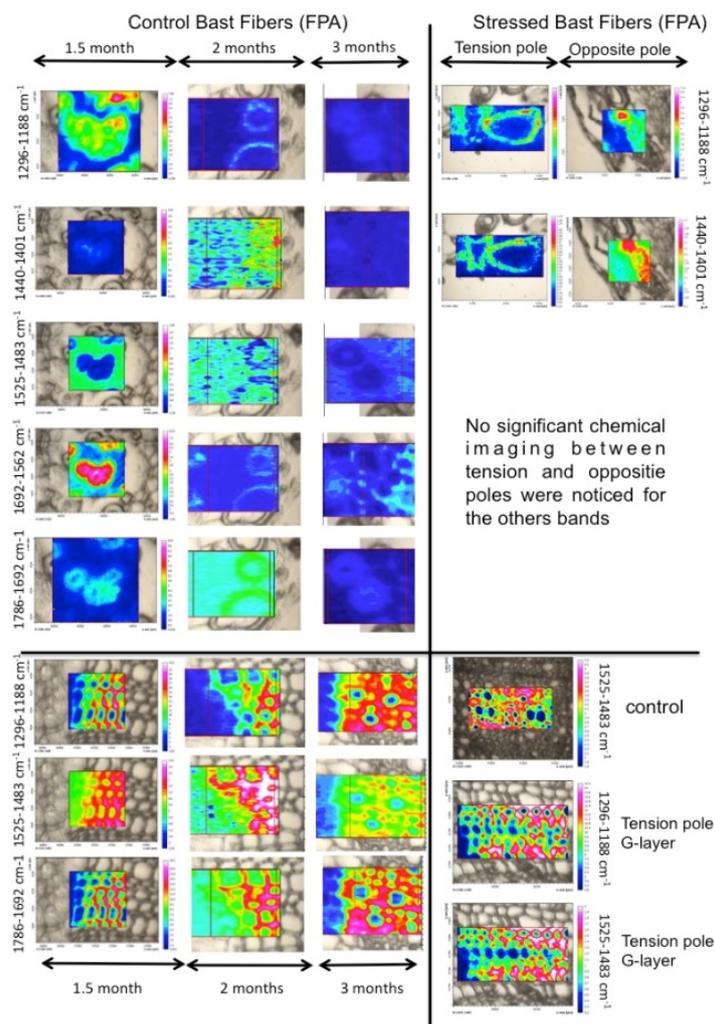


Fig. 1 FPA-chemical cartography of the 5 significant windows (determined through PCA analyses) on flax. Control BF corresponds to optimal growth conditions (1.5, 2, 3 months), whereas Stressed BF corresponds to phenotypically altered BF provoked by the gravitropic stress (45° bending for 6 weeks, total duration of culture 3 months). Images of xylem cell walls (bottom) were provided in order to compare with bands discriminating vessel G-layer. Color scale is similar for all images. Thirty micron transversal stem sections were used. PCA significant windows: [1,296-1,188 cm^{-1} : XG, Xyl] [1,440-1,401 cm^{-1} : C, Pect] [1,525-1,483 cm^{-1} : G-unit L] [1,692-1,562 cm^{-1} : G-unit L, Xyl, Pect] [1,786-1,692 cm^{-1} : Xyl, Pect]. PCA were performed on the pool of all FT-IR spectra acquired on the 6-cell wall types (for 1.5, 2, 3 months) and, independently, on 7-cell wall types for gravitropic stress. The two PCA clusters (A, B, see text) were determined in both culture conditions.

References

- [1] BONIZZONI, L., et al., 2016: *Microchem. J.*, **125**, 69. doi:10.1016/j.microc.2015.11.011.
- [2] FANTI, G., et al., 2013: *Vib. Spectrosc.*, **67**, 61. doi:10.1016/j.vibspec.2013.04.001.
- [3] HIMMELSBACH, D.S., and D.E. AKIN, 1998: *J. Agric. Food Chem.*, **46**, 991. doi:10.1021/jf970656k.
- [4] KAVKLER, K., and A. DEMŠAR, 2011: *Spectrochim. Acta - Part A Mol. Biomol. Spectrosc.*, **78**, 740. doi:10.1016/j.saa.2010.12.006.
- [5] WRÓBEL-KWIATKOWSKA, M., et al., 2009: *Spectrochim. Acta - Part A Mol. Biomol. Spectrosc.*, **73**, 286. doi:10.1016/j.saa.2009.02.034.
- [6] HIMMELSBACH, D.S., et al., 2002: *J. Sci. Food Agric.*, **82**, 685. doi:10.1002/jsfa.1090.
- [7] IBRAGIMOVA, N.N., et al., 2017: *Protoplasma*, **254**, 749. doi:10.1007/s00709-016-0985-8.