CHEMICAL IMAGING OF COTTON FIBERS USING AN INFRARED MICROSCOPE AND A FOCAL-PLANE ARRAY DETECTOR

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Abstract

Infrared spectroscopy can provide information on the structure and quality of cotton fibers. Recent advances have combined the informational capability of infrared spectroscopy with the spatial ability of microscopic techniques to allow for the fast chemical imaging of cotton samples. Herein, we explored use of a high resolution, Fourier-transform infrared (FTIR) microscope to examine cotton fibers. Imaging of cotton fibers was performed with a Focal-Plane Array (FPA) detector. FPA detectors present significant advantages over conventional detectors. Notably, FPA detectors allowed for simultaneous spectral acquisition at hundreds of sampling points. Chemical distribution maps and principal component analysis (PCA) plots visually depict the composition of the cotton samples. Results suggest the FTIR microscopy can potentially be utilized as a tool to monitor and assess important cotton properties during fiber development.

Introduction

Market demands call for the development of fast, reliable analytical methods for monitoring fiber development and measuring cotton fiber properties. Mid-infrared (Mid-IR) spectroscopy has been previously used to estimate important fiber properties and to monitor cell wall development (Abidi et al., 2010; Liu et al., 2011). In addition, recent advances in vibrational microspectroscopy allow for the examination of samples in a manner that combines the informational capability of infrared spectroscopy with the spatial ability of microscopic techniques (Lewis et al., 1995). This combination allows for fast chemical imaging of cotton samples.

Chemical imaging is a process by which analytical data is used to visually depict the chemical distribution of a sample. Chemical distribution plots can then be used to visually depict changes in vibrational bands associated with cotton fiber properties such as fiber maturity (Liu et al., 2011) or moisture content (Montalvo and VonHoven, 2004). Herein we present the chemical imaging of developing cotton fibers with an FTIR microscope equipped with a Focal-Plane Array (FPA) detector. FPA detectors present significant advantages over the more traditional MCT detectors. Notably, FPA detectors allows for simultaneous testing of hundreds of sampling points. This capability allows for the analysis of sample areas in a reduced amount of time and with high spatial resolution. For this initial study, FTIR multi-point analysis of developing cotton fibers was performed. In addition, principal component analysis (PCA) scatter plots and high resolution chemical distribution maps were calculated to visually differentiate between stages of cotton fiber development. Cotton fibers used were harvested beginning at 18 days post-anthesis (DPA) until the fibers where mature (fully opened cotton bolls) a time period with high secondary cell wall development.

Material and Methods

The cotton fiber samples used were grown in 2009 under standard field conditions in New Orleans, LA. The development of the line used, MD 90ne, was previously described (Hinchliffe et al., 2010). Samples were harvested at different developmental stages between 18 and 40 DPA. Mature samples were also collected. Cotton fibers were examined with a Hyperion 3000 FTIR microscope (Bruker Optics, Billerica, MA) equipped with a focal-plane array (FPA) detector and video camera. Fiber samples from each developmental stage were individually mounted on a metal plate with a small opening. FTIR microscope data was collected in the transmission mode. Typical sample scans were between 64 and 96 scans. Samples were manually flattened with a knife roller before mounting. Scans were measured with a resolution of 8 cm^{-1} (3800 – 900 cm^{-1}). Band assignments were taken from the Marechal and Chanzy FTIR study on cellulose I_β crystalline samples (2000).

Results and Discussion

Multi-point FTIR microspectroscopy

Spectra of multiple points were collected simultaneously utilizing an FPA Mid-IR detector. Figure 1 shows the video image (20x objective) of mature cotton fibers (left) and the FTIR spectra of multiple sampling points along these fibers (right). The spectra show absorption bands with positions that closely match major reported bands of cotton fibers examined with a conventional FTIR spectrometer (Abidi N et al., 2010). Notably, the relative intensity and shape of peaks in the C-O region (1185-985 cm^{-1}) closely compares to those observed in the FTIR ATR spectrum of a small cotton bundle. While the O-H stretching bands appear symmetrical, they exhibit some noise. Better resolution of the O-H bands might require further fiber flattening with commercial tools or deuteration. However, close examination of the changes in the O-H - hydrogen bonding network of developing cotton fibers is beyond the scope of this study.

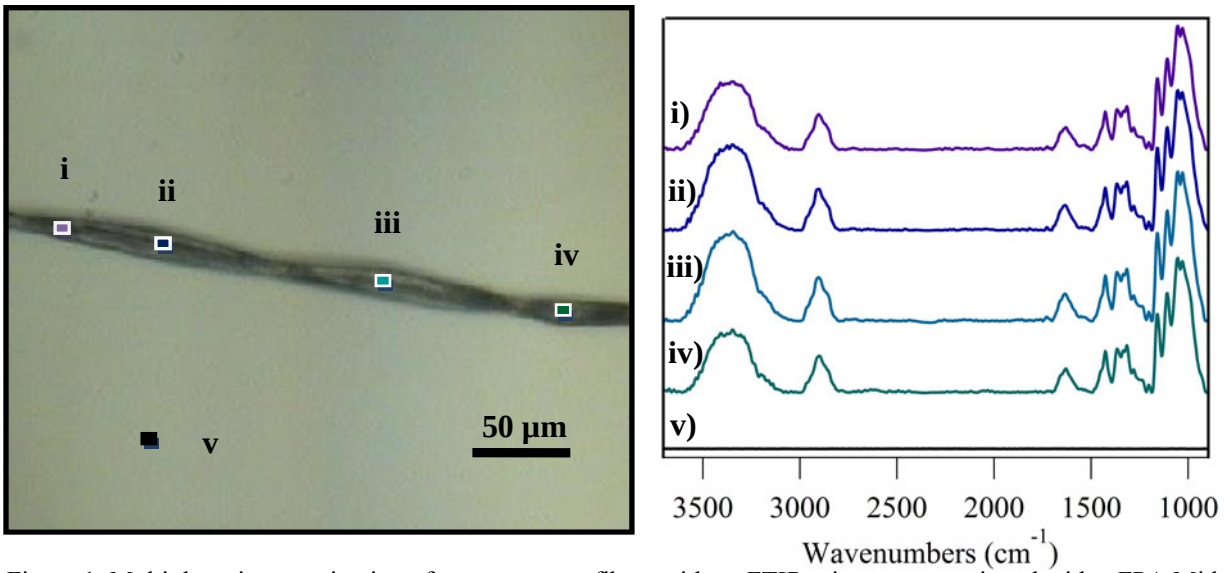

Figure 1. Multiple-point examination of mature cotton fibers with an FTIR microscope equipped with a FPA Mid-IR detector. Video image (20x objective) of two mature MD90 cotton fibers (left) and FTIR spectra of various sampling points (right).

PCA of FTIR microspectroscopy data

PCA allows for the simple comparison of complex data groups through a process of transformative reduction. A two-component depiction of cotton fiber first derivative spectra following PCA is plotted in Figure 2. Good separation of the first derivative spectra is observed, with each of the four groupings observed corresponding to one of the fiber developmental time points examined (18, 24, 32 and 40 DPA). Most of this separation is observed along the principal component 1 axis (PC1; left to right), which suggests that this principal component is greatly influenced by the developmental progress of the samples. The first two principal components in the first derivative model account for 82.7% of the variation observed in Figure 2. This PCA plot suggests that FTIR microspectroscopy spectra can be used to differentiate between developmental time points of cotton fibers.

FTIR chemical distribution maps

Chemical distribution maps of selected spectral regions were calculated to visually depict spectroscopic changes observed in developing cotton fibers (Figure 3). Three developmental time points were examined; 18, 24 and 40 DPA. Integration of the dominant C-O bending peak near 1054 cm^{-1} resulted in maps with high intensity regions (red tones) that evenly covered space associated with cotton fiber samples (Section ii maps). Section iii maps show the integration of the C-O bending region near 1016 cm^{-1}, a shoulder band that significantly increases in intensity as the cotton fibers develop secondary cell wall. Section iii maps were normalized to the maximum intensity of the corresponding 1054 cm^{-1} peak (ii). As the fiber develops SCW, the integration of the 1016 cm^{-1} region (iii) transitions from showing medium intensities (green & yellow tones for the 18 DPA sample and yellow/orange tones for the 24 DPA sample; Figure 3a & b) to strong integrations (mostly red and pink areas for the 40 DPA sample; Figure 3c). This observation mimics the increase in relative intensity of the C-O bending region near 1016 cm^{-1}

previously reported (Abidi et al., 2010). Taken together, our observations suggest that chemical distribution maps can also be used to visually represent general changes observed during cotton cell wall development.

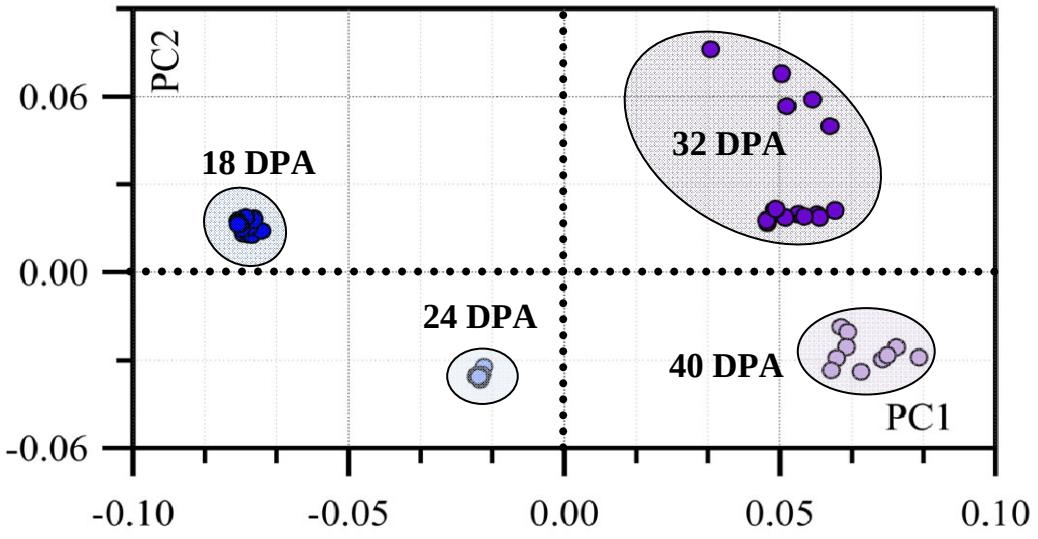

Figure 2. PCA scatter plots of first-derivative FTIR spectra for cotton fibers and harvested at the indicated developmental time points.

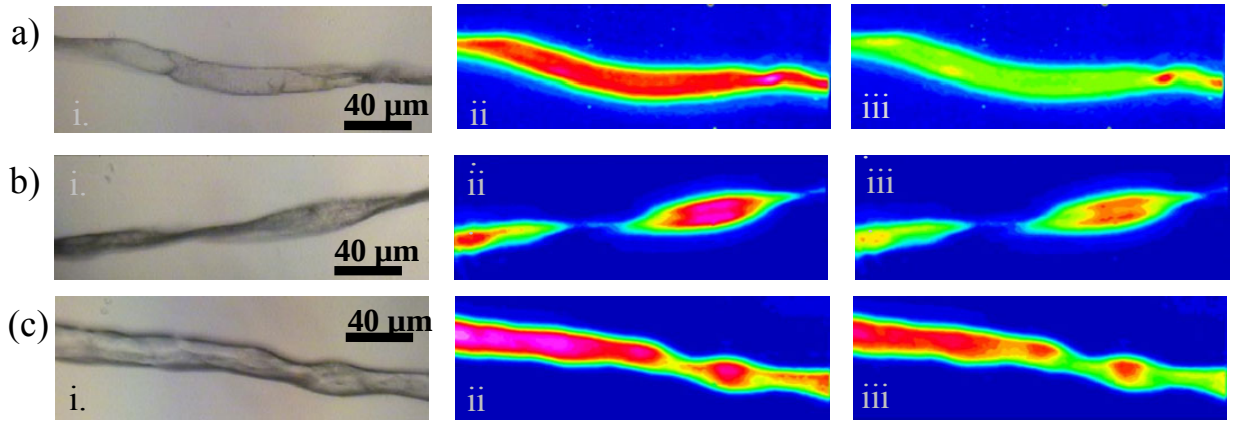

Figure 3. Chemical distribution maps for cotton fibers as determined with a FTIR microscope; developmental points shown are for (a) 18 DPA, (b) 24 DPA, and (c) 40 DPA fiber samples. For each developmental point an optical image (i) and Intensity maps correspond to the integration of a primary (ii) and a less prominent (iii) C-O peak (~1053 cm^{-1} and ~1016 cm^{-1}, respectively).

Summary

FTIR microspectroscopy can be used to assess general infrared spectral changes of developing cotton fibers Measurements are fast and require little sample preparation. PCA scatter plots of first derivative FTIR spectra allows for the visual distinction of some of the developmental time points. Also, microspectroscopy with the FPA detector also allows for the chemical imaging of cotton fibers. These chemical distributions maps can visually depict general spectral changes observed for developing cotton fibers. Our results suggest that FTIR microspectroscopy with a FPA detector could be used to examine cotton fiber properties and cell wall development.

Disclaimer

The use of a company or product name is solely for the purpose of providing specific information and does not imply approval or recommendation by the United States Department of Agriculture to the exclusion of others.

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