The zcorrectorgui For 3D ToF-SIMS Cell Analysis

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Statement of Purpose: ToF-SIMS imaging is a powerful technique for obtaining chemically specific maps of the surface chemistry of a wide range of samples including polymers, metals, tissues, and cells. When combined with the sputtering capabilities of modern ToF-SIMS instruments, one can obtain chemically and biologically specific 3D depth profiles, as well as track chemical signatures throughout a sample volume. This is done by taking an image of the surface, sputtering away a given amount of the material and then taking a new image of the freshly exposed area. This process is repeated until the object of interest is gone or the desired depth is reached.

When working with surfaces with significant topography such as the surface of a cell, the z axis of the resulting data cube is not correct. This is due to the fact that each image slice of the surface is displayed as a 2-D image taken from a 3-D surface. This results in a type of inverted topography of the 3D structure within the ToF-SIMS image volume. To correct for this, NESAC/BIO (Seattle, WA) has developed a Matlab (Mathworks, Natick MA) toolbox to correct the z-axis of cell depth profiles and display the data properly. The corrected depth profiles have been compared to AFM images and shown to reproduce accurate cell shapes.

Methods: The zcorrectorgui is a toolbox developed in Matlab. It can import a series of IONTOF (Muenster, Germany) *.bif files that contain the data from sequential slices through a cell surface. The zcorrectorgui determines the contour line of the cell by thresholding the total ion image of an X-Z or Y-Z slice through the image stack and finding the interface between the cell and the background. This contour line is set as the new z = 0 line and the data for all pixels in the given plane are shifted accordingly. The corrected data is then displayed. This correction assumes a constant sputter rate throughout the depth profile. The zcorrectorgui enables the user to produce the corrected profiles for any X-Z or Y-Z slice, create a movie slicing through the image matrix in any direction, and display the corrected data as a 3D volume.

Results: AFM and ToF-SIMS images were acquired sequentially on a NIH 3T3 cell. The AFM data was acquired and processed using an Icon system (Bruker, Santa Barbara CA). Peaks corresponding with known fragments from lipids and proteins were selected and the peak area images were exported into *.bif images using the IONTOF software (Muenster, Germany). The *.bif images were then imported into the zcorrectorgui. The resulting data matrix was z-corrected. Figure 1 shows images of the process steps for finding the z = 0 line and correcting the data. Figure 2 shows a 3D rendering of the corrected SIMS data and the AFM data. As seen in Figure 2 the corrected ToF-SIMS data shows the same shape and structure as the AFM data.

The similarity between the images from AFM and the corrected ToF-SIMS data suggests that the assumption of a constant sputter rate is valid, at least for the cells used in this study.
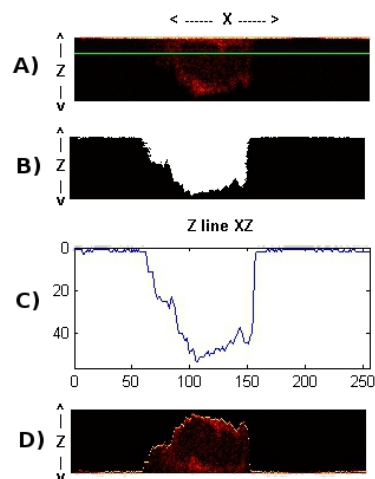

Figure 1. Processing steps in the zcorrectorgui. A). Original X-Z slice. B). Thresholded X-Z slice. C) Contour line used for new z = 0. D). Corrected X-Z slice.
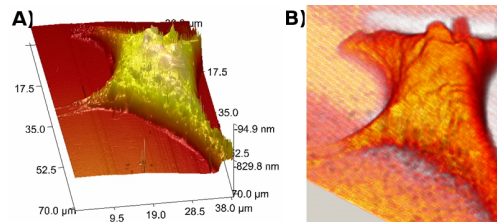

Figure 2. AFM and ToF-SIMS 3D Images A). AFM 3D height map image. B). Z-Corrected ToF-SIMS 3D rendered image.

Conclusions: The zcorrectorgui is a Matlab toolbox developed by NESAC/BIO that enables the user to correct the z-axis of a 3D depth profile. The current version is focused on 3D depth profiles of cells, but should work for any set of data that has a constant sputter rate throughout the depth profile. Data processed using the zcorrectorgui was shown to produce a very similar cell shape as generated from AFM imaging of the same cell.

The zcorrectorgui is available on the NESAC/BIO MVSA website (http://mvsa.nb.uw.edu).