uProbeX TWG Talk

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Background

- Started working at APS in March 2013
- Worked on TXM software at sector 32ID
- Collaboration work with Diamond Light Source for HDF writer plugin in area detector
- uProbeX application
- QImaging area detector plugin
Typical Workflow

- **Visualize samples on EPI Fluorescence Microscope**
  - Verify sample quality
  - Localize area of interest
    - With coordinates to later find the area in the x-ray Microprobe
  - Acquire complimentary data / images
    - Brightfield, phase, fluorescence: DNA, Immunolabelling, GFP, ect
- **Kinetic mounts to move sample around**
  - Visible light microscope, 2IDB, 2IDD, 8BM, (21ID, 26ID)
- **Find fiducials (eg, corner of window, marker on EM grid) in x-ray microprobe**
- **Apply coordinate transform**
EPI Fluorescence Microscope to X-Ray Microprobe

2-ID-E Hard X-ray Microprobe

Epi-Fluorescence Microscope

Sample in sample chamber, purge with He

kinematic specimen mount
specimen
condenser

20x obj.
QI Viewer

- Used to control light microprobe
- Saves SWS workspace
  - Contain coordinate information
  - Tile Overlap
  - Can Export mosaic TIFF image
### Coordinate Transformation

#### Quick & dirty: 10-20 microns
Careful: <5 microns

<table>
<thead>
<tr>
<th></th>
<th>measured input</th>
<th>predicted microscope</th>
<th>measured micropro</th>
<th>x</th>
<th>y</th>
<th>leica x</th>
<th>leica y</th>
<th>XRM x</th>
<th>XRM y</th>
<th>delta x</th>
<th>delta y</th>
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<tbody>
<tr>
<td>P (center)</td>
<td>Top sample (A-D)</td>
<td>Surface 2b-up</td>
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<td>A (up, SL)</td>
<td>71.517</td>
<td>43.875</td>
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<td>B (3down, BR)</td>
<td>71.137</td>
<td>44.496</td>
<td>0.4801</td>
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<td>C (3right, BL)</td>
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<td>D (3left, AR)</td>
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<td>43.894</td>
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**Total error:** 53.1248 37.94 15.18
Results
uProbeX Features

- Open SWS workspace generated by QI software light microscope
  - Parse coordinates
- Display stitched image with coordinates
- Allow calibration markers to be places
  - Markers hold light microscope coordinates and allow entry of x-ray microprobe coordinates
- Ability to run minimization solver for transformation coefficients
  - Users mode allows basic X and Y offsets, Admin mode allow more control
- Create region boxes with transformed x-ray microprobe coordinated over existing light microscope image
- Export region box properties to outside applications
  - Can call python or shell scripts, allows channel access through scripts
- Ability for users to take software home and view data
uProbeX Specs

- Supported platforms
  - Windows 7 32 and 64 bit
  - Linux 64 bit
  - Mac OSX (coming soon)

- Available to download from our build server
  - Jenkins build server
  - SSG Updater application

- Python support
  - Linked to support python
  - Can still be used if python not found on system

- Developed by AES SSG
  - Arthur Glowacki: aglowacki@anl.gov
  - Ke Yue: kyue@aps.anl.gov
Test Sample

- Selected four calibration points
  - Named A, B, C, and D
- Different colors to easily differentiate between points.
- Mouse over Light and X-Ray coordinates available at bottom of the scene
- Able to quickly adjust X and Y offset coefficients in transformation equation
- Standard Zoom In/Out controls
Find Microprobe coordinates for Reference Points

- X-Ray alignment
- Fill in measured X and Y for each calibration point
- Run minimization solver to fine tune transformation
Create Regions

- Created four region boxes
  - Dynamically named
  - Saved/Restored when workspace is closed/opened
- Region boxes also have customizable colors
- Display predicted center X, Y, width, and height of the region box. (X-Ray coordinates)
- Programmable context menu (right click) options
- Can call python or shell script with region box properties as parameters
Scan of Reference Corner A

- X-Ray scan of region box A.
- Calibration looks good.
Select New Scan Region

- New region box created
- Send the coordinates to x-ray microprobe to perform a scan
X-Ray Microprobe of Scan Region

Off by 3um in X, 4.5um in Y
Expected from solver solution
Region Box Preferences

- Call user defined python scripts
- One way communication, send region box properties as parameters to python or shell functions
- Added python script is scanned for all functions and populated in table
- Ability to uncheck (hide) functions from users
- All callable functions appear as context menu items when right clicking a region box
Solver Preferences

General solver is implemented using Nelder-Mead cminpack implementation

General Transformer was supplied by Stefan Volg

Python Transformer
– Allows custom transformers
– Bidirectional, send calibration point, returns transformed point

Python Solver
– SciPy, NumPy
– User can make their own
– Add dynamic coefficients
Ideas for Expanding the Software

- **Ability to load HDF5 results and overlay on light microscope data**
  - Give the user the ability to blend, manage layers of the data
  - View multiple layers at once

- **Multi-point scan regions**
  - User defined multi point scan sections
  - Does not have to be rectangular
  - Properties can be passed to python or shell script like current region box
Documentation and Build

- Confluence
  - https://confluence.aps.anl.gov/display/CLMFS/Main
  - How to Install
  - FAQs

- Jenkins
  - Build Server
  - Supports Window 7 and Linux, Soon will have Mac OSX also
  - User does not have to compile the software
Summary

- Unify multiple application into one to simplify experiment process
- Boost beam line productivity by 10 – 30 %
- Sent scan properties through script to remove user error
- Integrate with Python to allow more dynamic user configurations
- For more information Email us
  - Arthur Glowacki : aglowacki@anl.gov
  - Ke Yue : kyue@aps.anl.gov
- Thanks to Charlotte Gleber and David Vine for testing and feedback of the software.
- Thanks to Stefan Vogt and Charlotte for presentations slides.
- Questions?