

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

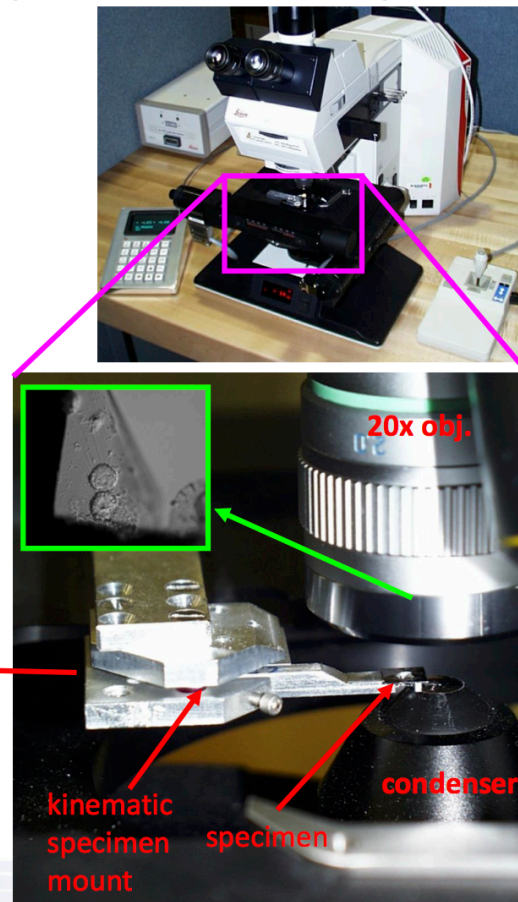


EPI Fluorescence Microscope to X-Ray Microprobe

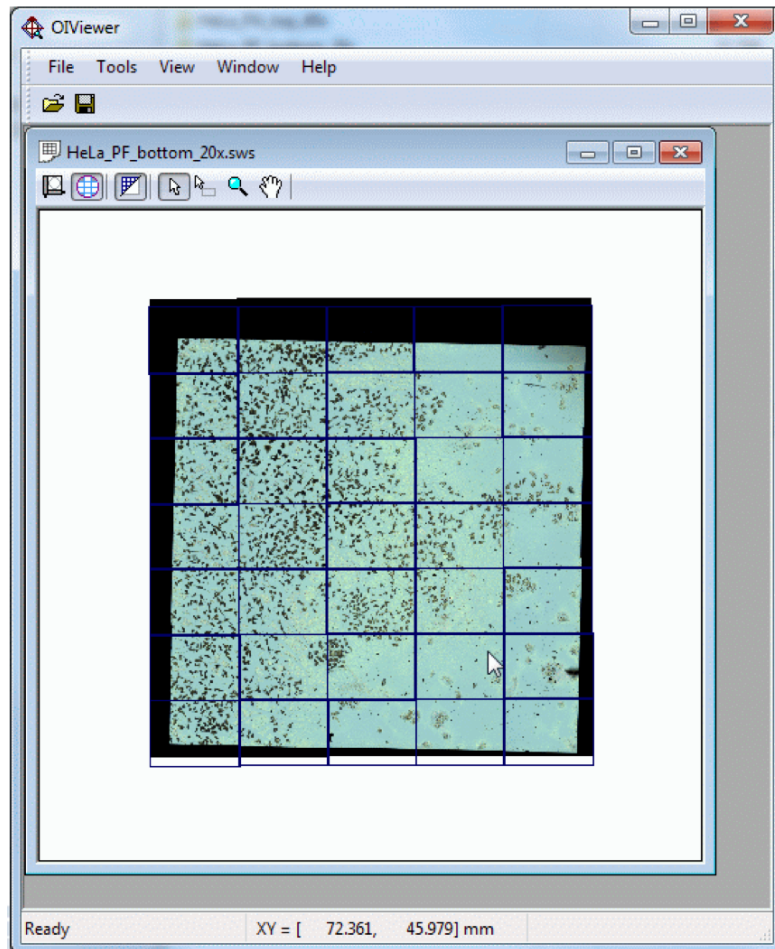
2-ID-E Hard X-ray Microprobe



Epi-Fluorescence Microscope



QI Viewer



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

Spread Sheet Transformation

Coordinate transformation

cosy_trafo

Normal Bad Good

Check Cell Explanatory ... Follow

M40 =IF(OR((I40+J40=0),(B40+C40=0)), "", (G40-J40)*1000)

BATS Oct 2013 CGrid 38																		
measured / input			predicted microprobe		measured microprobe						total error:							
from Leica (light microscope)			coordinates		coordinates						2xfm: x'		2xfm: y'					
[mm]	[mm]	[mm]	[mm]	[mm]	[mm]	[mm]	[mm]	[um]	[um]									
x	leica.y	leica.z	XRM: x'	XRM: y'	XRM x'	XRM y'	delta x'	delta y'										
Top sample (A-D)			surface 2b-up															
P (center)																		
A (3up, BL)																		
B (3down, BR)																		
C (3right, BL)																		
D (3left, AR)																		
reference pt	71.517	43.875		0.1132	1.6645	0.1132	1.6645	0.00	0.00									
b01	71.137	44.495		0.4891	1.0369													
b02	71.128	44.497		0.4982	1.0348													
b03																		
b04	71.297	43.994		0.3361	1.5410	0.3336	1.5420	2.51	-1.01									
b05	71.260	44.006		0.3738	1.5282	0.3690	1.5285	4.77	-0.28									
b06	72.055	43.764		-0.4359	1.7866	-0.4275	1.7814	-8.38	5.29									
b07	72.052	43.746		-0.4324	1.8046	-0.4244	1.7990	-8.01	5.57									
b08	72.032	43.745		-0.4119	1.8052	-0.4039	1.8107	-8.01	-5.57									
b09	71.201	43.673		0.4416	1.8598													
b10																		
b11	72.424	43.843		-0.8159	1.7154													
b12	70.743	43.915		0.9058	1.6084													
b13	70.733	43.965		0.9149	1.5582													

direct view, naked eye

Light microscope view

X-ray microprobe view

total error: 53.1248 37.94 15.18
2xfm: x' -64.1277 2xfm: y' -41.8937

1 0.0000 0.0000

1 2.5062 -1.0125

1 4.7720 -0.2761

1 -8.3833 5.2906

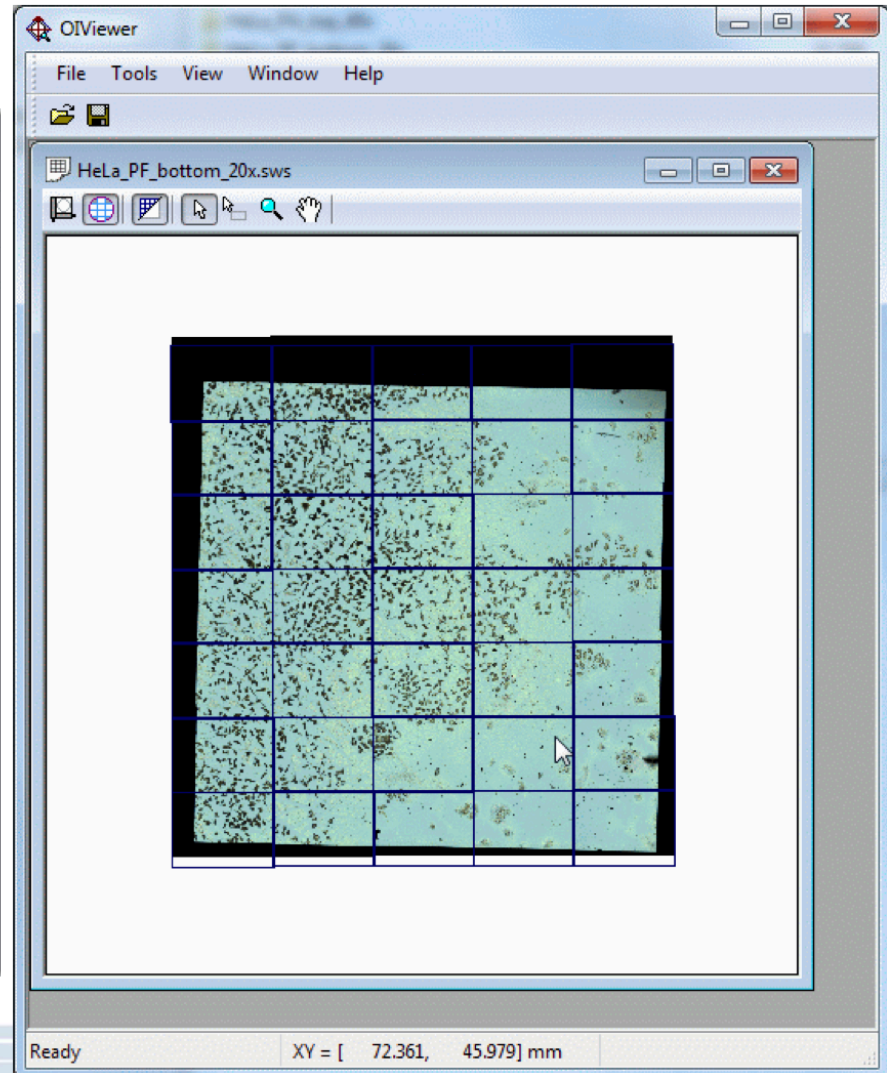
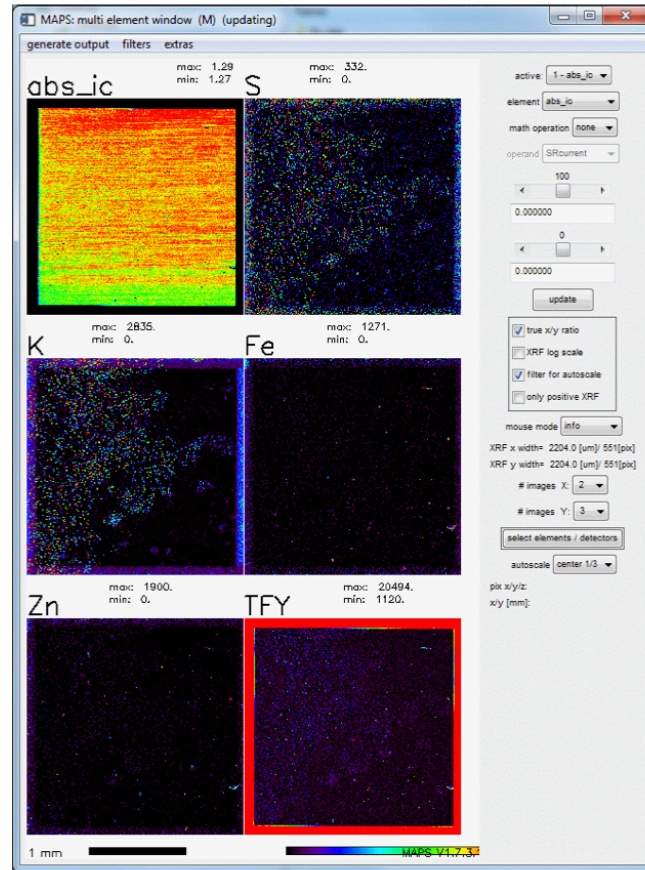
1 -8.0102 5.5676

1 -8.0102 -5.5676

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

3

Results



uProbeX Features

- Open SWS workspace generated by QI software light microscope
 - Parse coordinates
- Display stitched image with coordinates
- Allow calibration markers to be placed
 - 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

The screenshot shows the uProbeX software interface. The main window displays a calibration scene with a central crosshair and various patterns. Four calibration points are marked with colored crosses: A (red), B (blue), C (green), and D (cyan). The scene is overlaid with a grid and a coordinate system. The software title bar indicates the file path: uProbeX - [Y:/data/2ide/2014-2/comm1/vlm/cr_testpattern_bf_20x.sws]. The interface includes a menu bar (File, Help), a toolbar with navigation and zoom controls, and a status bar at the bottom showing coordinates: Light X: mm, Light Y: mm, Micro X: 0.0764146 mm, Micro Y: -1.63892 mm.

Calibration MicroProbe

2xfmX: 0 2xfmY: 0

Add Calibration Point Run Solver

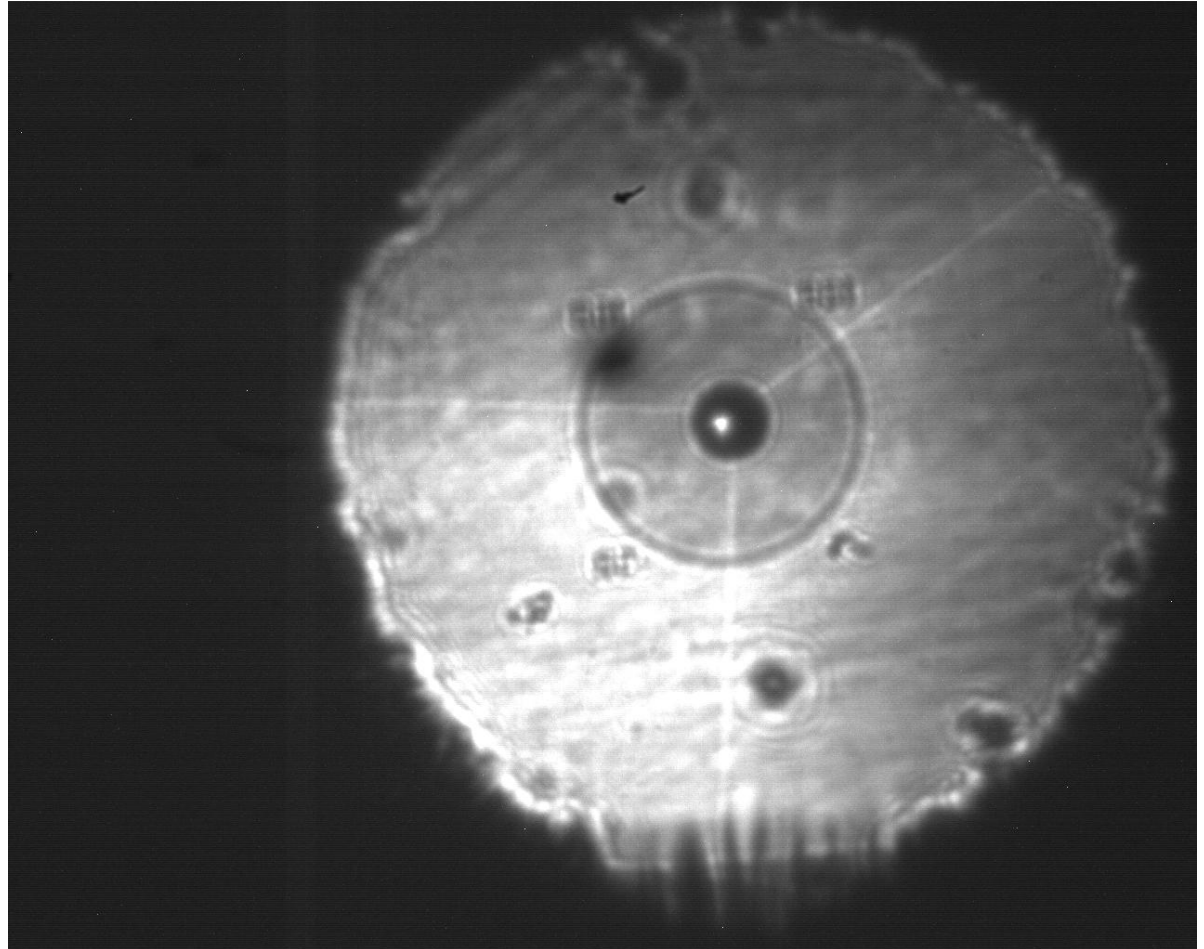
Calibration Point	Color	LX	LY	LZ	PX	PY	MX	MY
A	Red	94.6101	54.1666	0	-0.113087	-2.67454	-0.102	-2.680
B	Blue	93.6264	54.1602	0	0.900024	-2.68856	0.894	-2.69
C	Green	94.6101	53.1821	0	-0.0913214	-1.69035	-0.087	-1.690
D	Cyan	93.6315	53.1799	0	0.916432	-1.70843	0.907	-1.702

Light X: mm Light Y: mm
Micro X: 0.0764146 mm Micro Y: -1.63892 mm



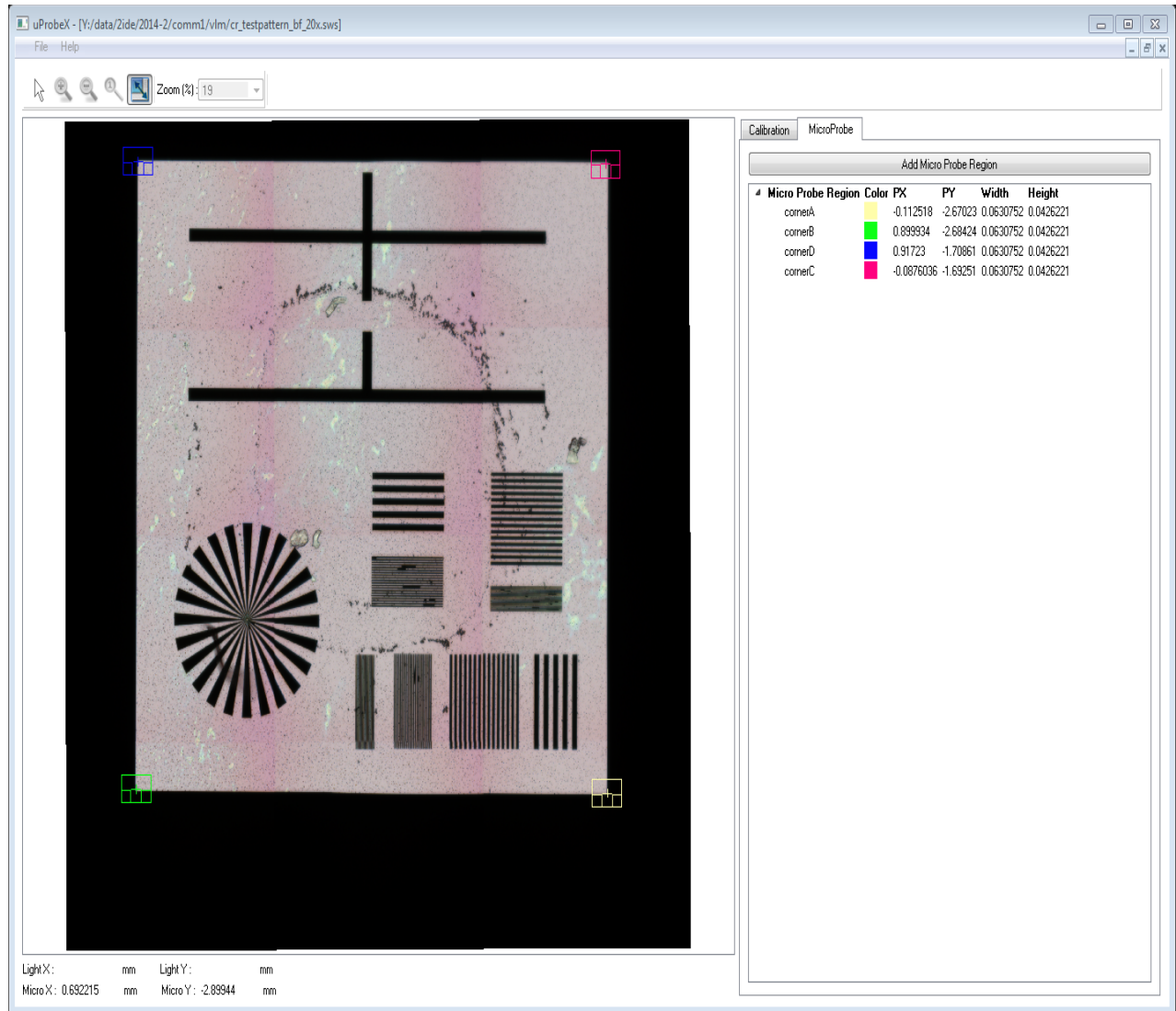
Find Microprobe coordinates for Reference Points

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



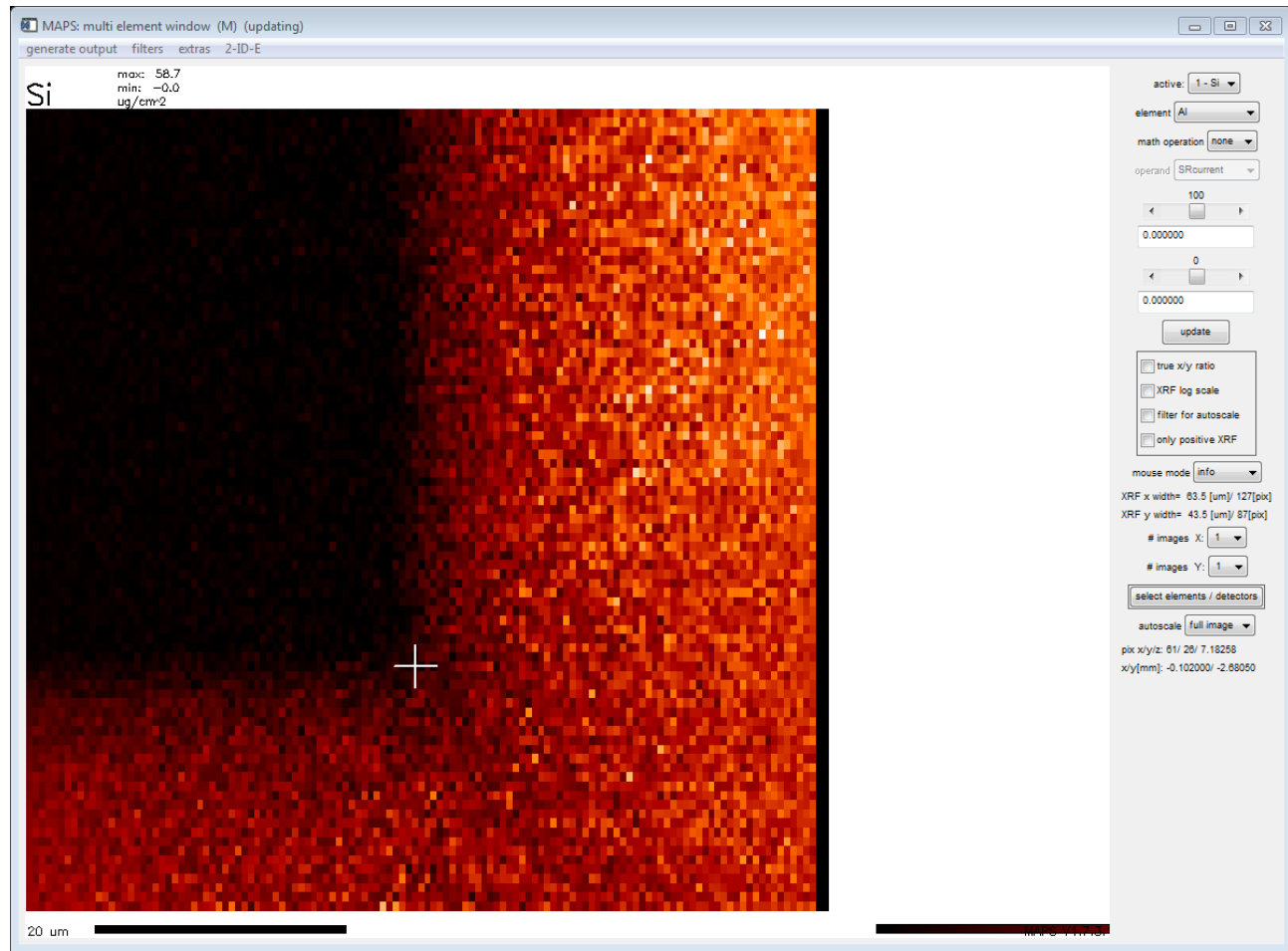
Create Regions

- 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

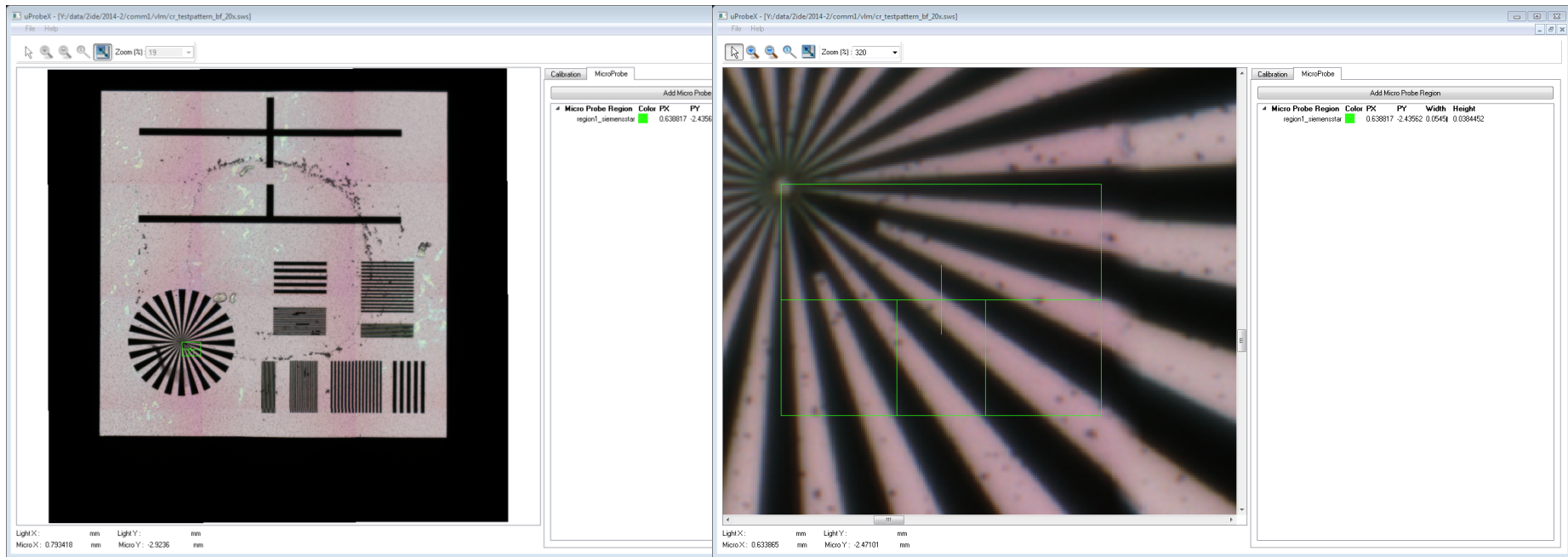


Scan of Reference Corner A

- X-Ray scan of region box A.
- Calibration looks good.



Select New Scan Region



New Scan Region

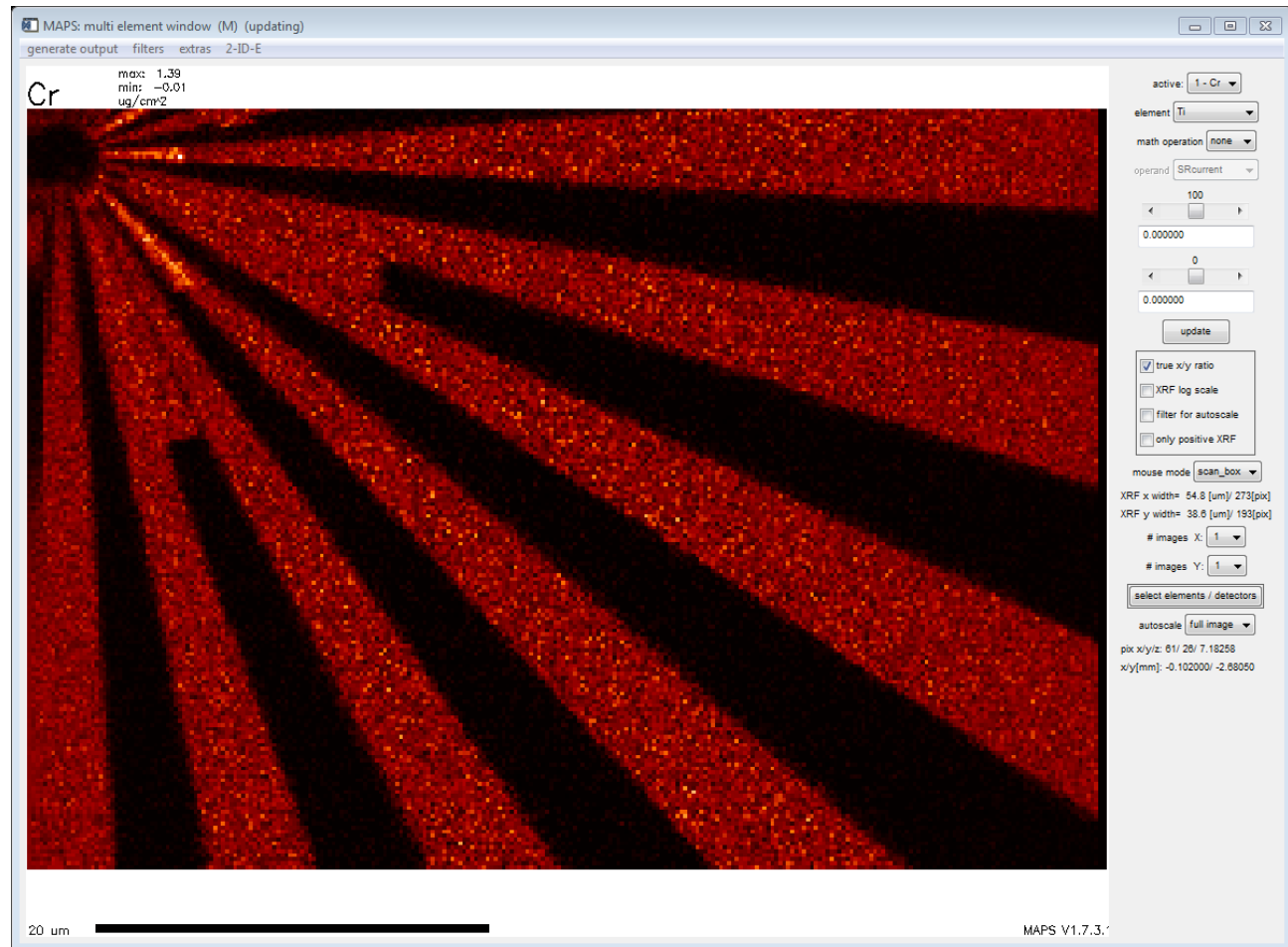
Zoomed in Scan Region

- New region box created
- Send the coordinates to x-ray microprobe to perform a scan



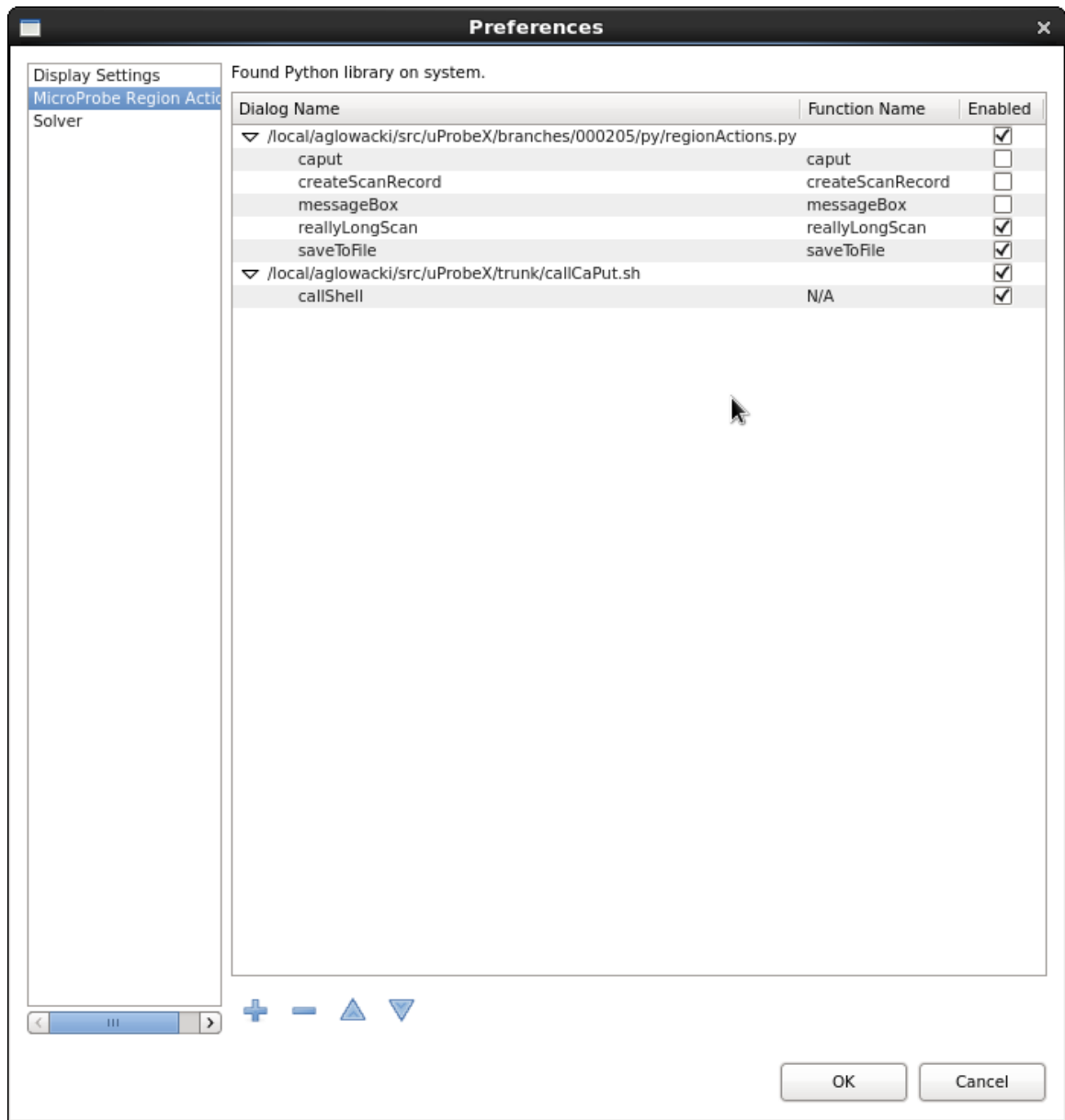
X-Ray Microprobe of Scan Region

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

The screenshot shows the 'Preferences' dialog box with the 'General Solver' tab selected. The 'Display Settings' sidebar on the left has 'Solver' selected. The 'General Solver' section is active, showing two tables: 'Coefficient' and 'Parameters'. The 'Python Solver' section is also visible, including a 'Python Script Path' field and a warning message.

General Solver

Coefficient

Name	Value	Min Enable
angle_alpha	1.19	<input type="checkbox"/>
m2xfm_x	-64...	<input type="checkbox"/>
m2xfm_y	-41...	<input type="checkbox"/>
offset_a	10.27	<input type="checkbox"/>
offset_b	2.13	<input type="checkbox"/>
offset_c	0	<input type="checkbox"/>
offset_d	0	<input type="checkbox"/>
omega	90	<input type="checkbox"/>
omega_prime	90.04	<input type="checkbox"/>
scaling_XF...	1.03	<input type="checkbox"/>

Parameters

Name	Value	Min Enable
Iterations	100...	<input checked="" type="checkbox"/>
StepSize	0.01	<input checked="" type="checkbox"/>

Python Solver

Python Script Path:

Open Python File...

The python solver must have solver and transform functions !

Coefficient

Name	Value	Min Enable
angle_alpha	1.19	<input type="checkbox"/>
m2xfm_x	-64...	<input checked="" type="checkbox"/>
m2xfm_y	-41...	<input checked="" type="checkbox"/>
offset_a	10.27	<input type="checkbox"/>
offset_b	2.13	<input type="checkbox"/>
offset_c	0	<input type="checkbox"/>
offset_d	0	<input type="checkbox"/>
omega	90	<input type="checkbox"/>
omega_prime	90.04	<input type="checkbox"/>
scaling_XF...	1.03	<input type="checkbox"/>

Parameters

Name	Value	Min Enable
op	1	<input checked="" type="checkbox"/>
name	0.0	<input type="checkbox"/>

Run Solver

OK Cancel

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

- <https://jenkins.aps.anl.gov/view/uProbeX>
- 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?

