Image Informatics for Multidimensional Visualization

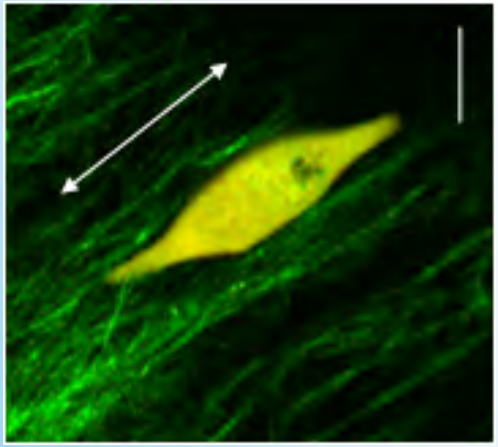
Fiji

OME

ImageJ2

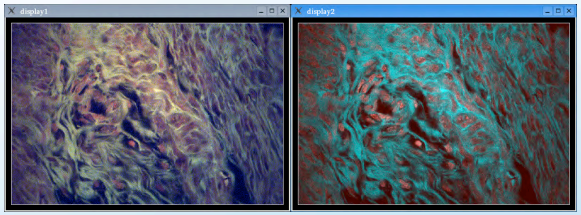
Kevin Eliceiri
Laboratory for Optical and Computational Instrumentation
www.loci.wisc.edu
eliceiri@wisc.edu

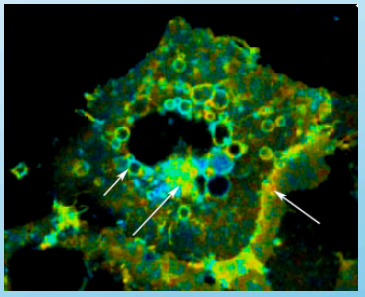
- New optical instrumentation to facilitate studies of the dynamics of living specimens.
- Better software for capture and visualization of dynamic, 3-D biological events
- Been OME Development partner since 2002
- Image Informatics for multidimensional data
  - spatial and temporal
  - spectral and lifetime dimensions
  - large 3D and xy montages
  - Light Sheet/SPIM

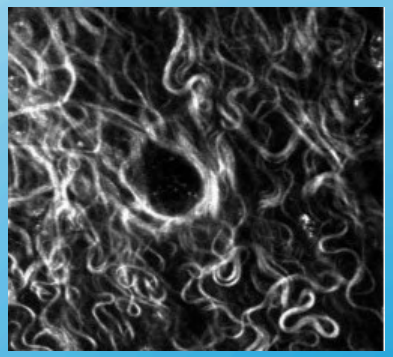


Mission of LOCI

Our data:







Fluorescence,
Spectra, Lifetime
(λ,τ)

Time-Lapse
(t)

Dynamics

Chemistry

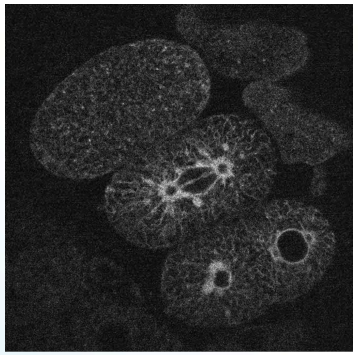
Optical Microscopy

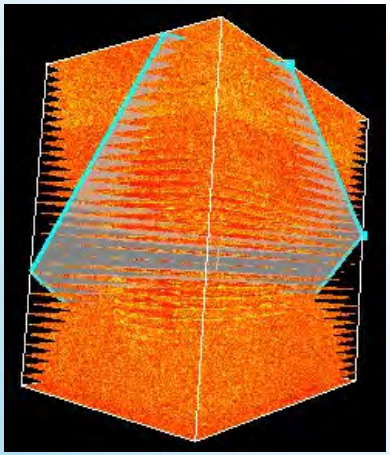
Physics

Structure

Phase, Polarization,
Scatter, Harmonics
(ϕ, θ,…)

Space
(x, y, z)



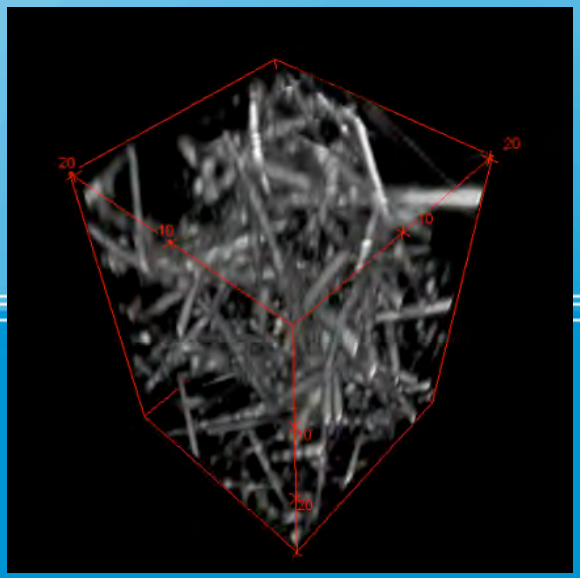


Supporting Technologies

Hardware
Acquisition software
Analysis and Visualization
Data Management

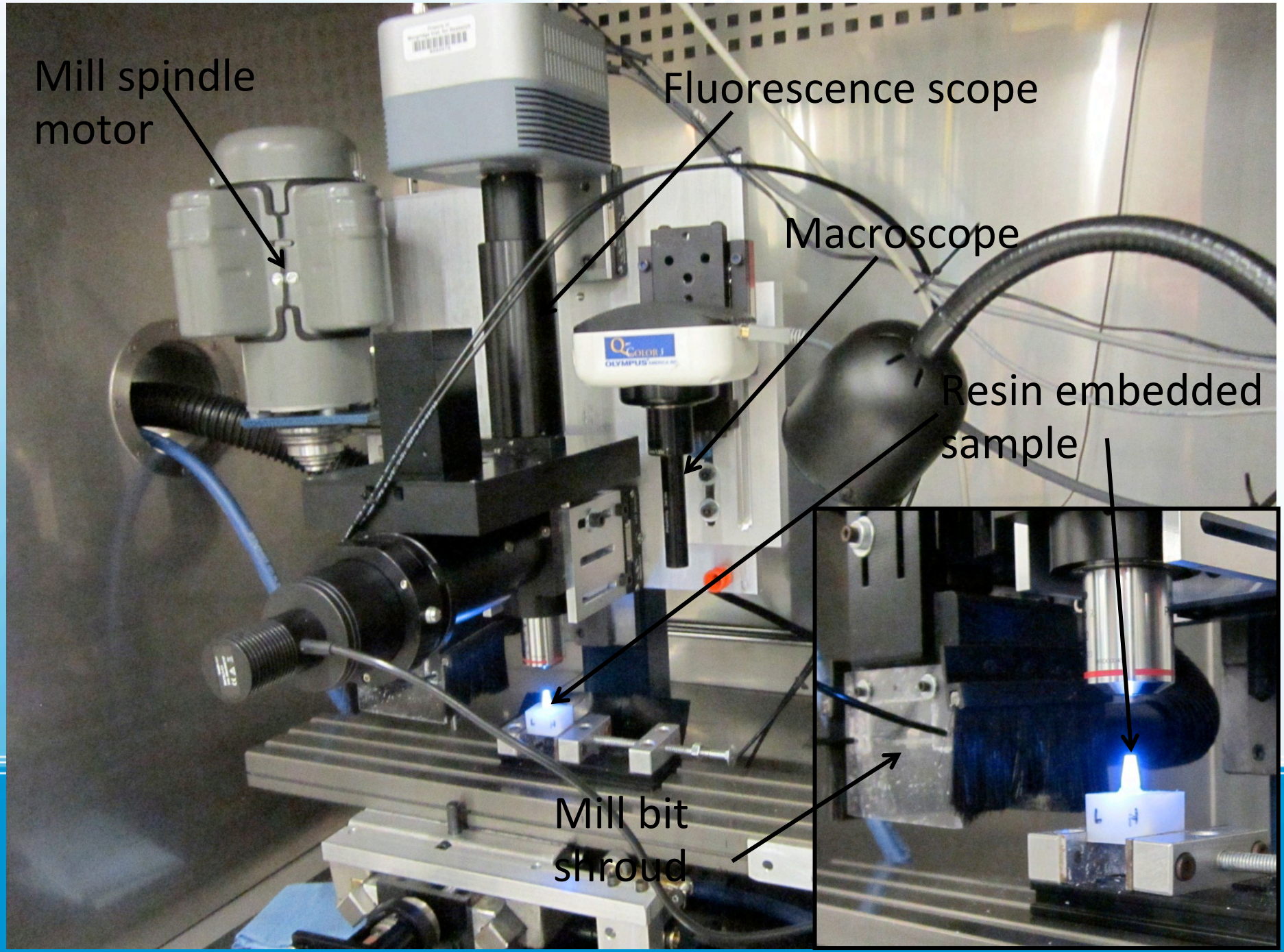
LOCI

Compact Automated Multiphoton Microscope (CAMM)



- Compact fiber laser source
- Open hardware platform
- Open software platform
- Completely motorized
- Large field of view capture
- Compact/optional portability

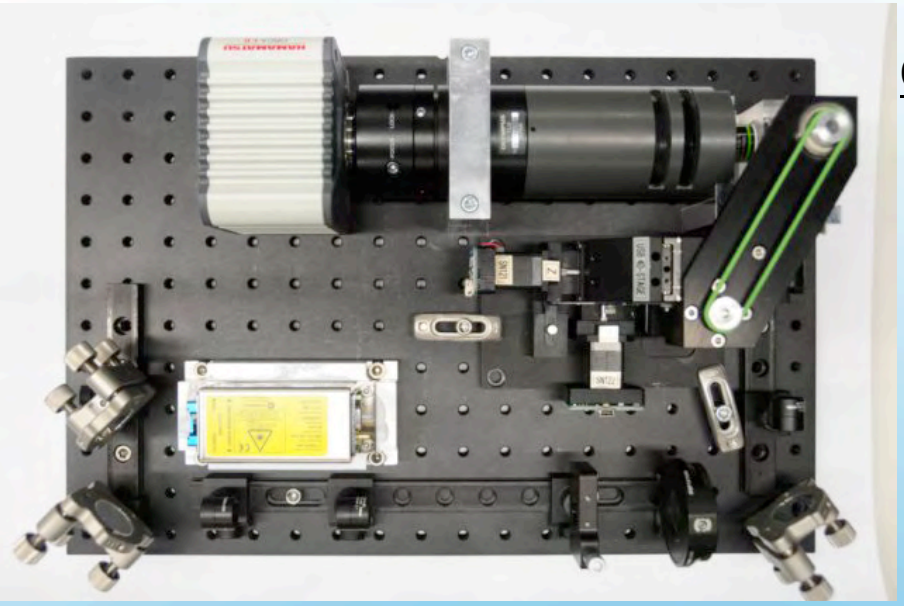
Sequential Erosive Tissue Imaging (SETI)

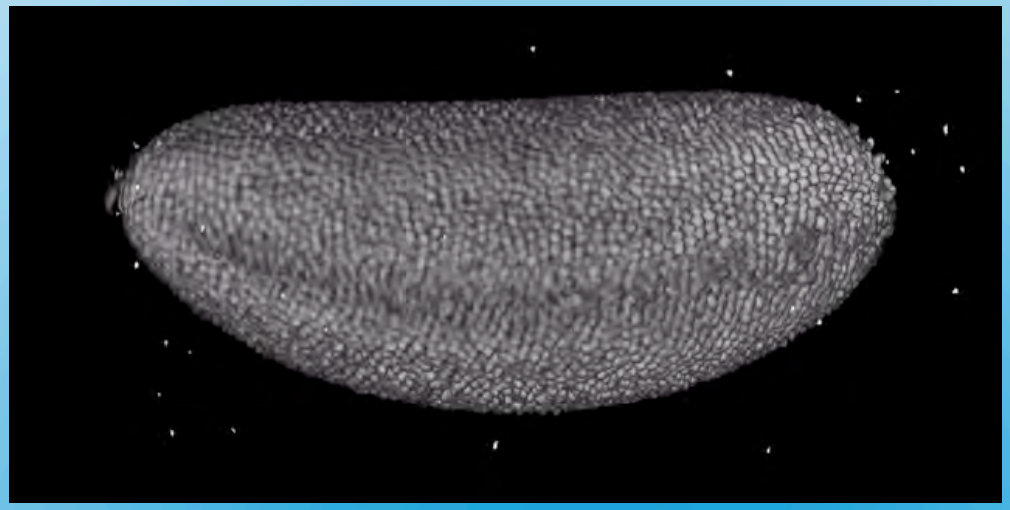


SETI Features

- 3D Image Capture of Biological Tissue Samples
- High resolution
  - Up to 1.5 micron optical resolution
- Large field of view
  - 1-2 cm^3
- Fully automated image acquisition
- Oblique reflectance or fluorescence contrast
- Frozen or Plastic Embedded Samples

Axolotl Limb Development





Open Access Light Sheet Microscope

- Selective Plane Illumination Microscopy
  Fully open hardware and software
- OME-TIFF Saving
- FIJI and MicroManager Based
- image big samples
- fast processes
- record entire embryos
- multiple angles across long periods of time.

OpenSPIM (www.openspim.org)

Overview of our OME efforts:

- **Specific OMERO linked applications**
  - BK Cho in Murphy lab on Omero.searcher
  - Forward Project for data dissemination

- **Originated OME-TIFF in 2004**
  - Now fully integrated into OMERO
  - Used by many open and commercial tools
  - Our current focus on robust tools to read and write OME-TIFF

- **Originated Bio-Formats in 2005**
  - partnership with OME and Glencoe
  - Over 120 formats
  - Over 30,000 installations
  - Recent focus on SCIFIO and native program interfacing

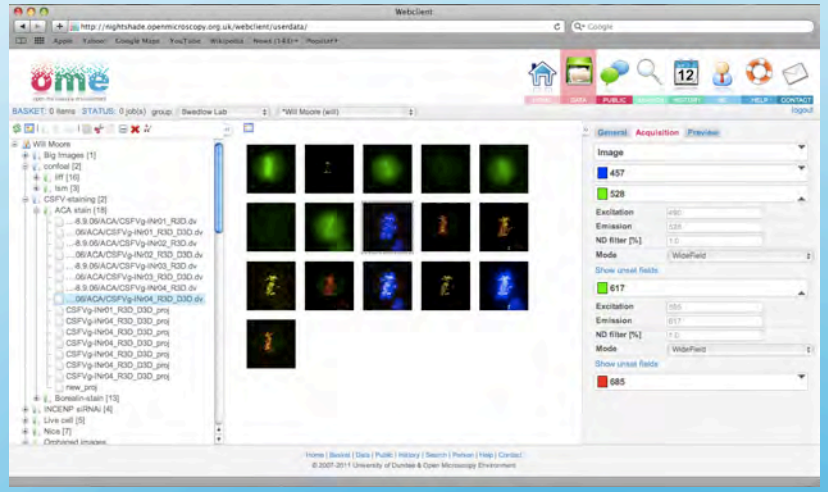
- **XML Strategies for Acquisition and Analysis**
  - Our WiscScan software and now MicroManager
  - Plans to extend to others that want richer “OME-TIFF”

- Interoperability between OME and other tools (FarSight, CellProfiler, FIJI, ImageJ

- ImageJ 2.0 (developer.imagej.net) as OMERO Client

Bio-Formats: the tool for interoperability

BIO-FORMATS



CellProfiler™
cell image analysis software

GLENCOE SOFTWARE

icy

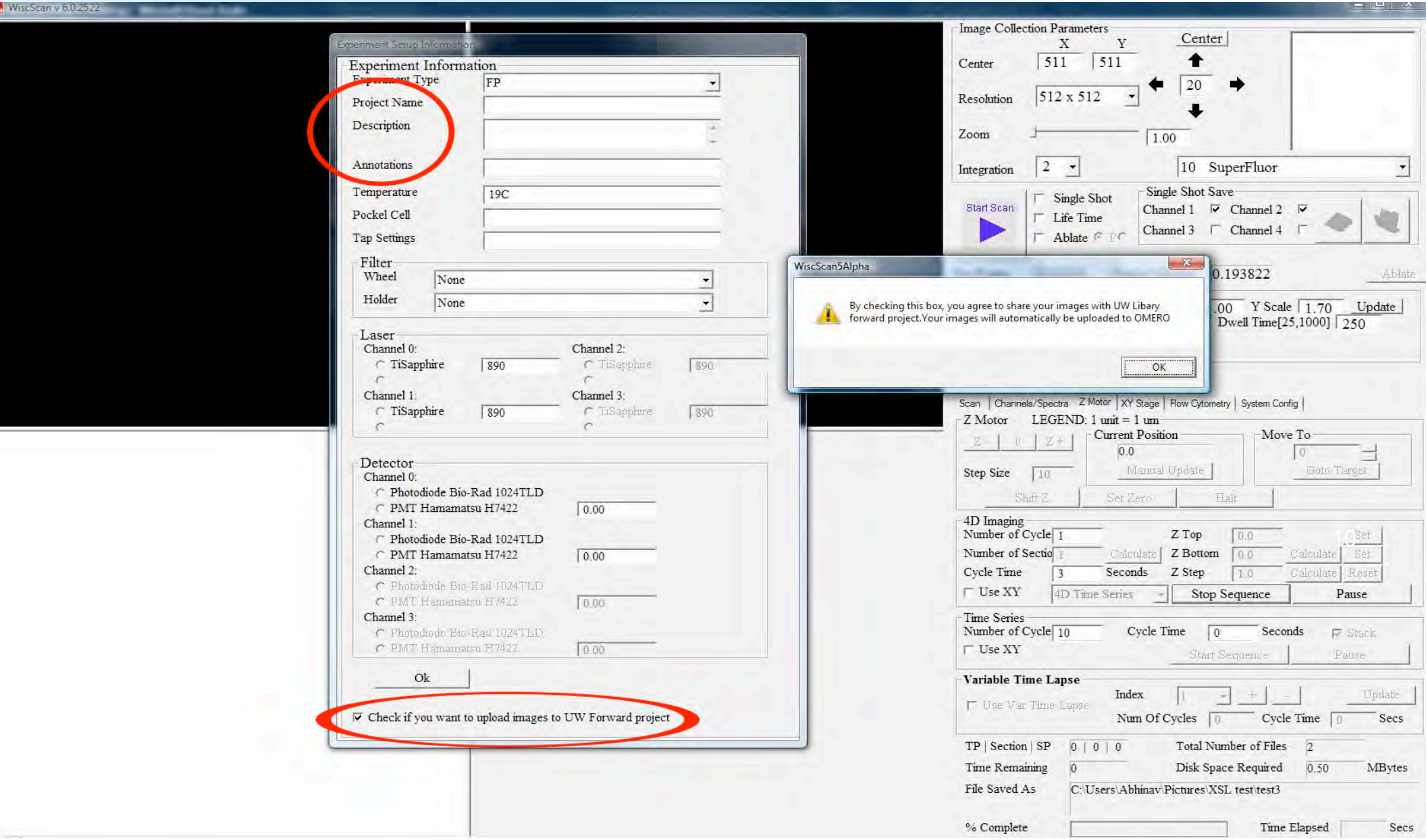
ImageJ
Image Processing and Analysis in Java

JCB Data Viewer

vtk

itk

Bio-Formats: ease of integration, even from other platforms



Stitching with OME-XML and FIJI

81 individual images of stitched together showing the entire layout of collagen within a tumor.
This is a z-projection of 100 individual stacks taken at 3um increments to show the full collagen projection

- SCientific Image Format Input & Output
- SCIFIO is simply the core of Bio-Formats
- Isolates core from PFFs and OME data model
- Convert other scientific imaging data models
- Provided under Simplified BSD License
- Bundled with ImageJ2, ITK, VisAD…
- Bio-Formats becomes a “SCIFIO plugin”
- Core Project of OME
- Recently funded by NSF

Reqs for Image Informatics?

Interoperability

- ✓ Metadata
- ✓ Interfaces
  - ✓ Bio-Formats
  - ImageJ2
  - ImgLib

ImageJ2 + OME

- OMERO is powerful server-side imaging software
- ImageJ is powerful client-side imaging software
- Both paradigms are valuable to scientists
- ImageJ as client to OMERO
- ImageJ on server side
- Many operations are common to client & server
  - E.g.: Bio-Formats is used for both
- Goal is to identify more areas for code sharing:
  - Big images (e.g., tiling with mipmaps)
  - Image rendering and thumbnails
  - Many others

- Support the next generation of image data
- Interoperate and collaborate with other projects
- Broaden the ImageJ community
- Reuse each others' work wherever practical
- Provide a central online resource for ImageJ
  - Program downloads, a plugin repository, developer resources and more

Why ImageJ2?

Universität Konstanz

OME

Max Planck Institute of Molecular Cell Biology and Genetics Dresden

MARINE BIOLOGICAL LABORATORY · WOODS HOLE · 1888

HHMI janelia farm research campus

BROAD INSTITUTE

And anyone who wants to contribute!

LOCI

Who Develops ImageJ2?

NATIONAL INSTITUTES OF HEALTH

- Preserve backwards compatibility
- Maintain good performance
- Support N-dimensional imaging
- Use common input and output for data
- Minimize complexity
- Employ open source software dev. practices

Guiding Principles

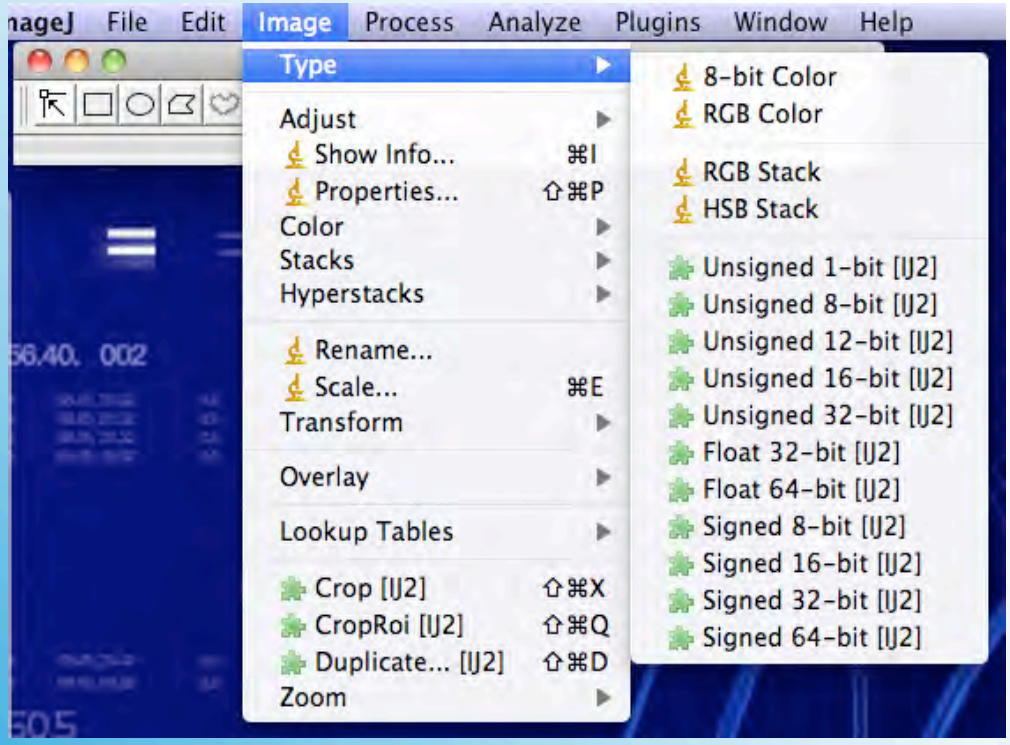
- Supports n-dimensional data (beyond ImageJ 1.x's X,Y,Z,C,T)
- Support image data from sources beyond disk
- Support additional pixel types
- More flexible color look up tables (16 bit palettes)
- No limit on # of channels in composite image
- Designed to support the cloud (i.e. execution without a graphical desktop

Break Limitations of ImageJ 1.x

- Supports most ImageJ1 plugins and macros
- Many new pixel types
- Multidimensional data beyond 5D
- Import and export of many file formats
- Improved region of interest (ROI) tools
- Truly headless
- Automatic updates
- Easily install additional plugins (e.g., Fiji >350 plugins!)
- >200 new/reimplemented core ImageJ2 plugins so far

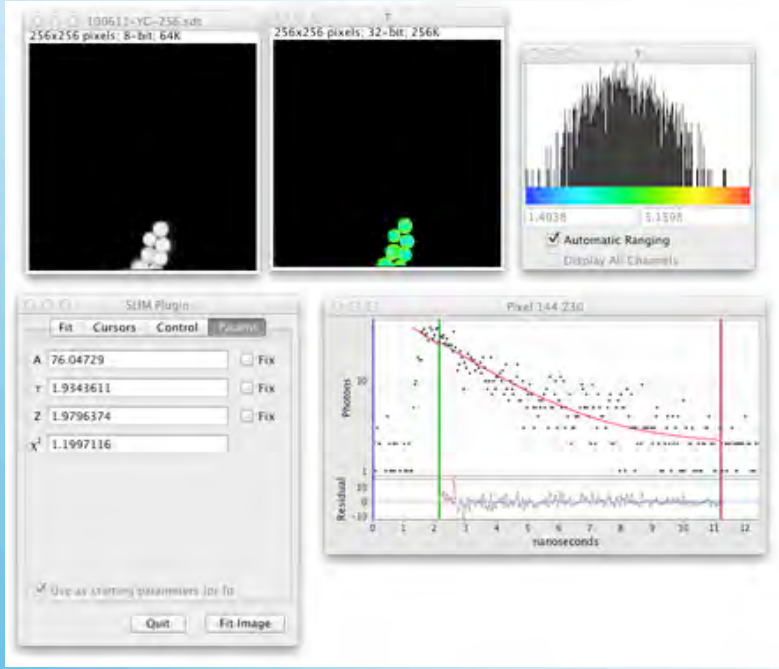
ImageJ 2-beta

- Based on ImgLib2 library
- Any data source (files, URLs, DBs...)
- N-dimensional images
- Unlimited pixel types
- Write algorithms once



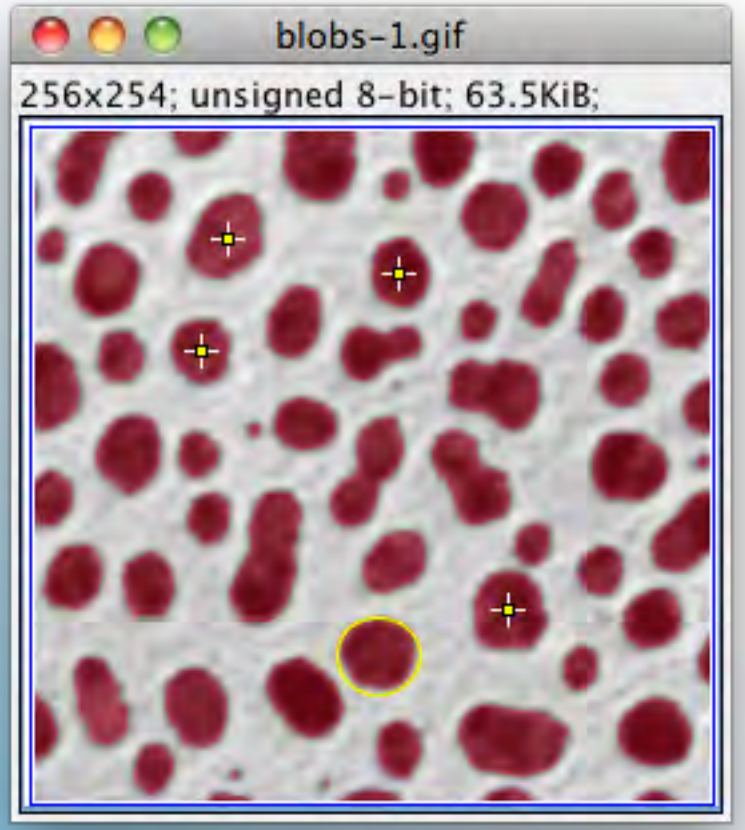
Data Model

- The user interface provides views of the data
- Show multiple datasets in one window
- Or multiple windows showing the same dataset
- Composite any # of channels
- Fully pluggable



Displays and Visualization

- ROIs are functions that identify samples upon which to operate
- Overlays are visuals superimposed over a dataset, often (but not always) linked to ROIs

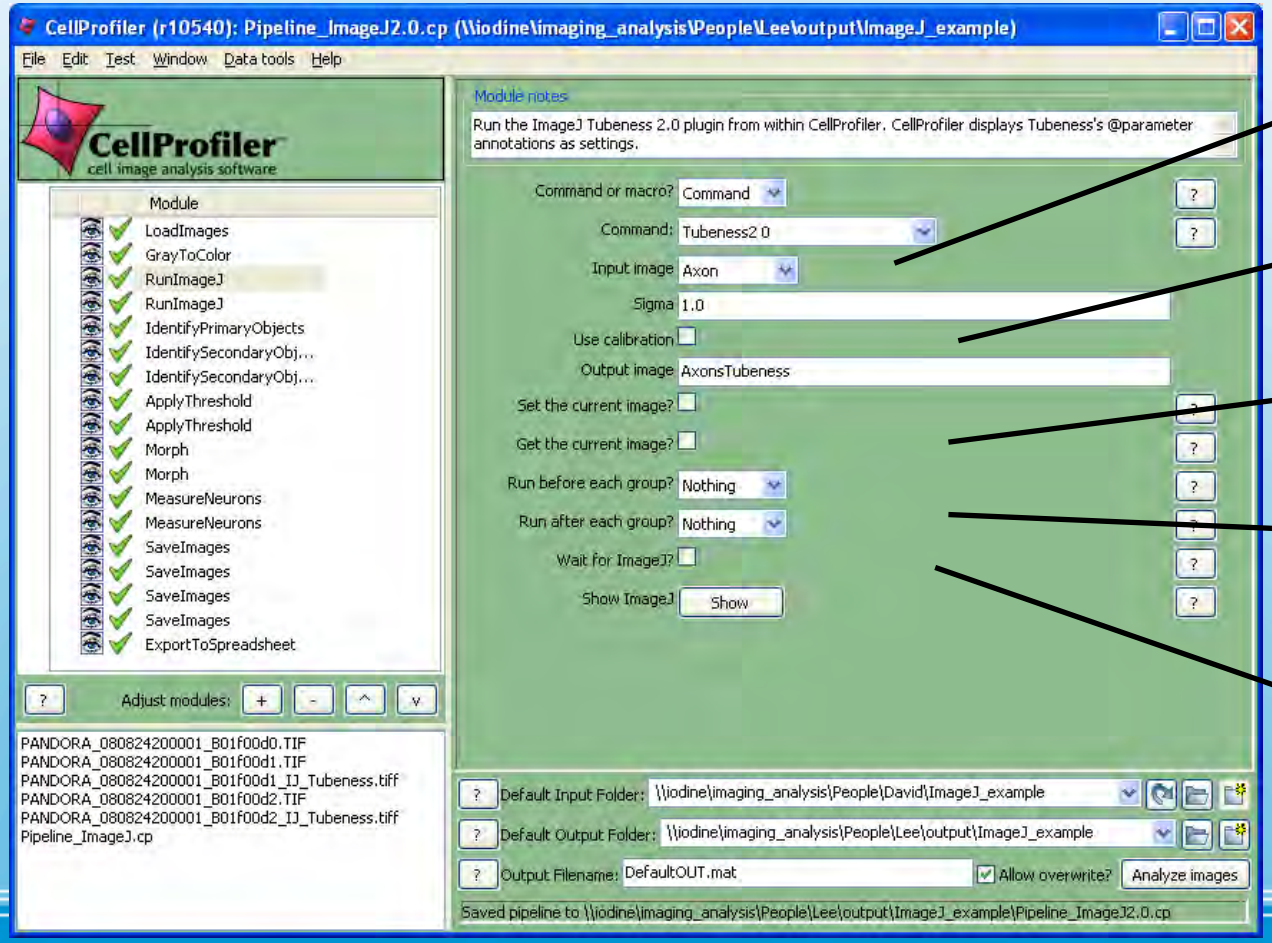


Regions of Interest

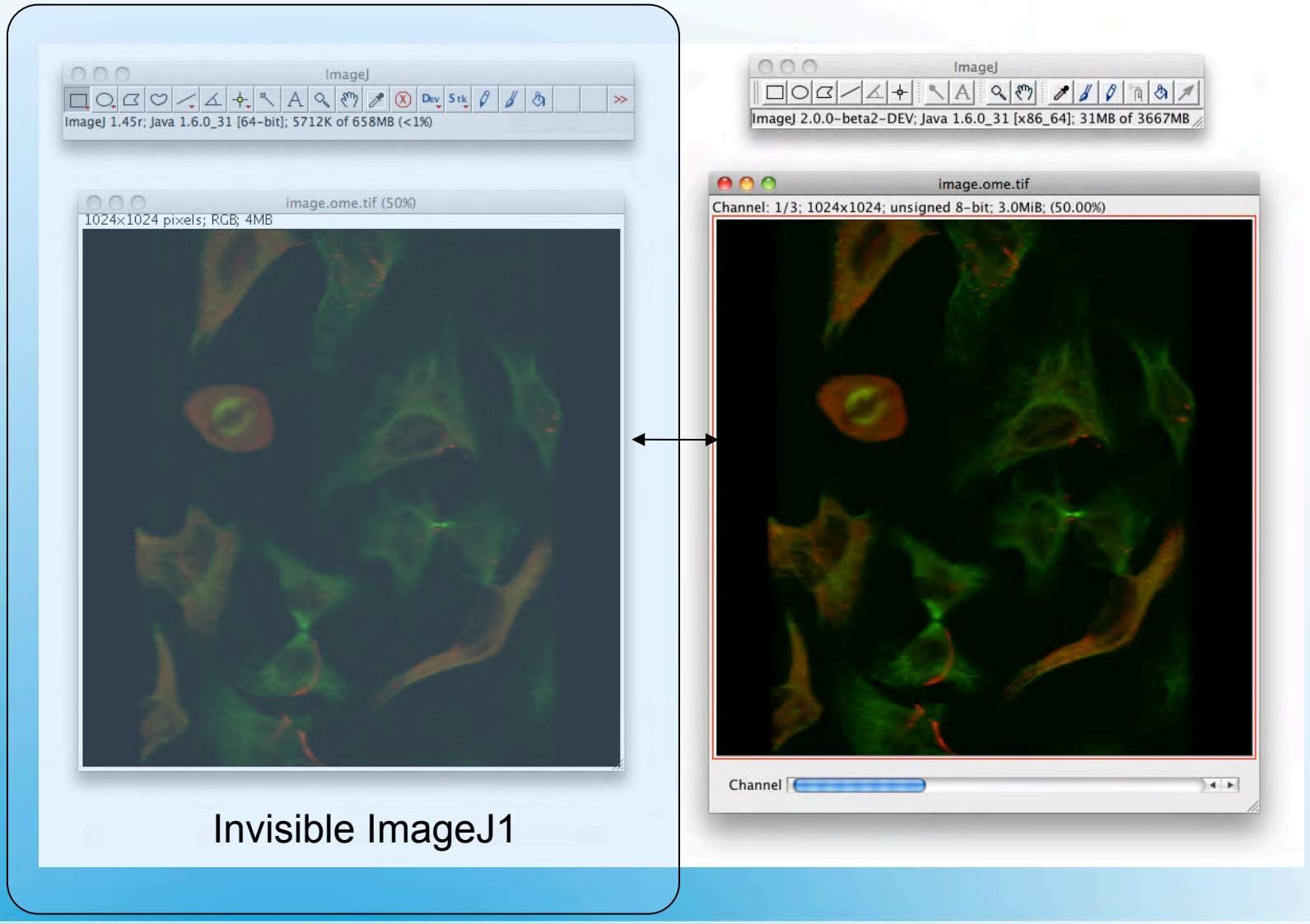
- Provides widgets for several UI styles:
  - Swing
  - “Pure” AWT
  - Eclipse SWT
  - Apache Pivot
- Custom UIs possible
- Can run headless
- Use ImageJ2 as a library to execute plugins

User Interface

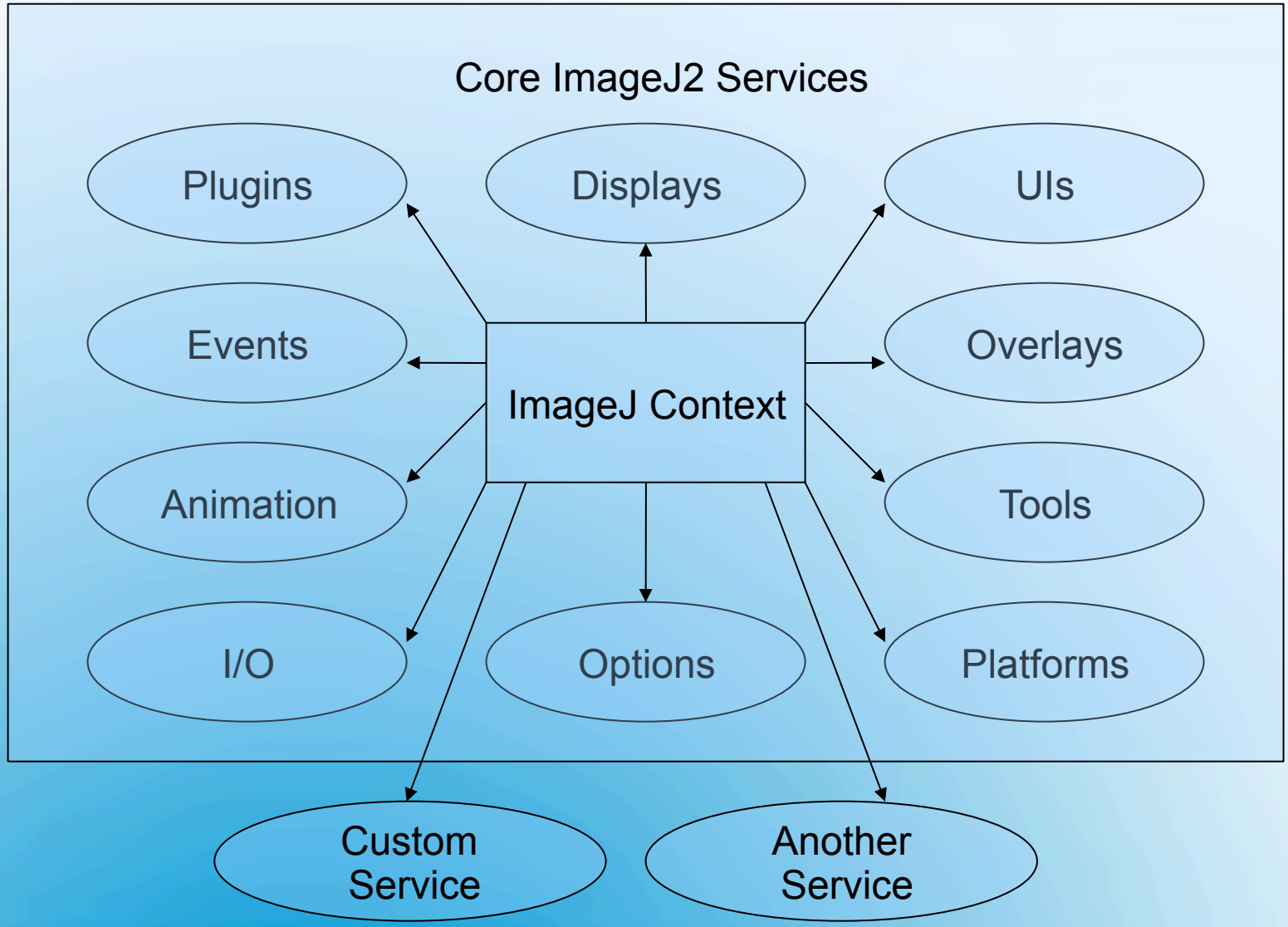
CellProfiler can run ImageJ 2.0 plugins




```
public class Tubeness2_0
extends AbstractPlugIn
implements PlugIn {
    @Parameter(label="Input
image")
    public ImagePlus
original = null;


@Parameter(label="Sigma")
    public double sigma =
1.0;

    @Parameter(label="Use
calibration")
    public boolean
useCalibration = false;

    @Parameter(label="Output
image", output=true)
    public ImagePlus result
= null;
```

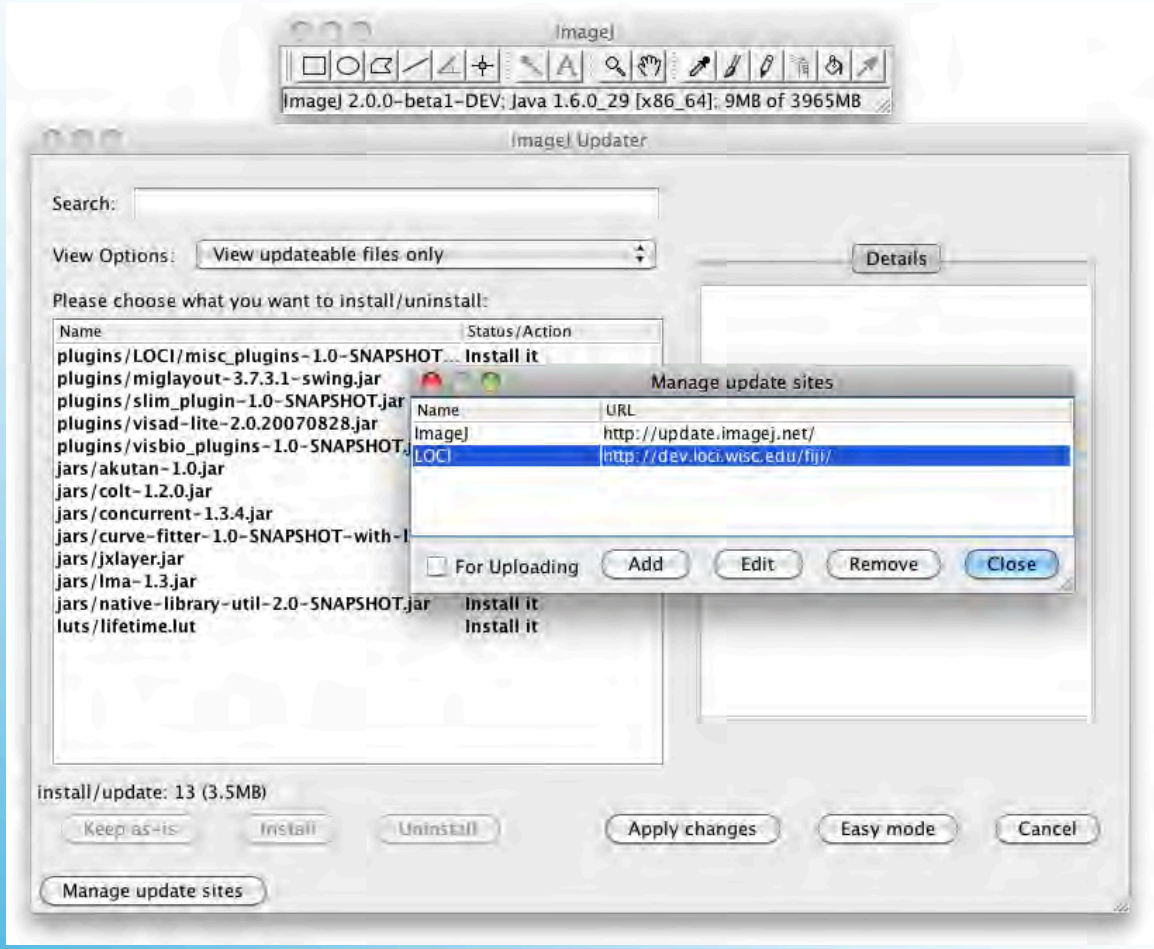


Compatibility



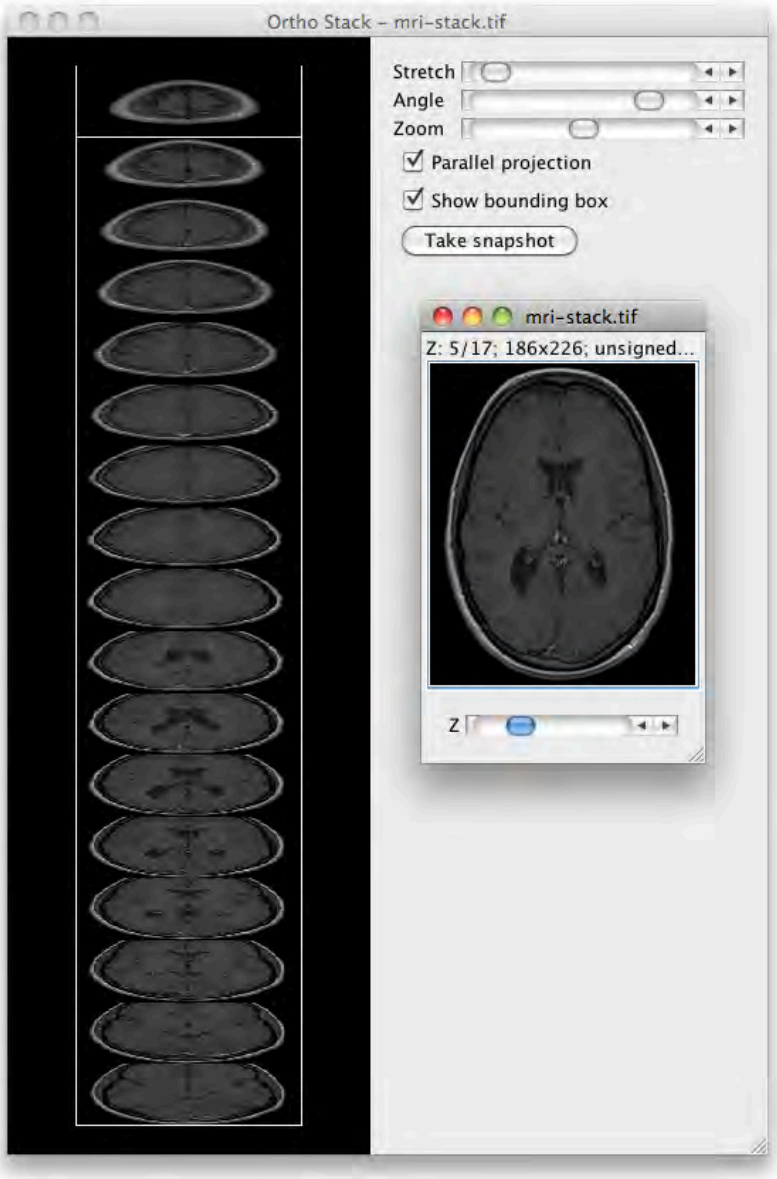
Service Architecture

- Keeps ImageJ2 plugins up-to-date
- Installs new plugins
- Anyone can create an update site and upload their own plugins to it
- Compatible with existing Fiji update sites (including fiji.sc)



ImageJ Updater

- Can work with third party update sites too
- On right, VisBio Ortho Stack plugin from LOCI update site

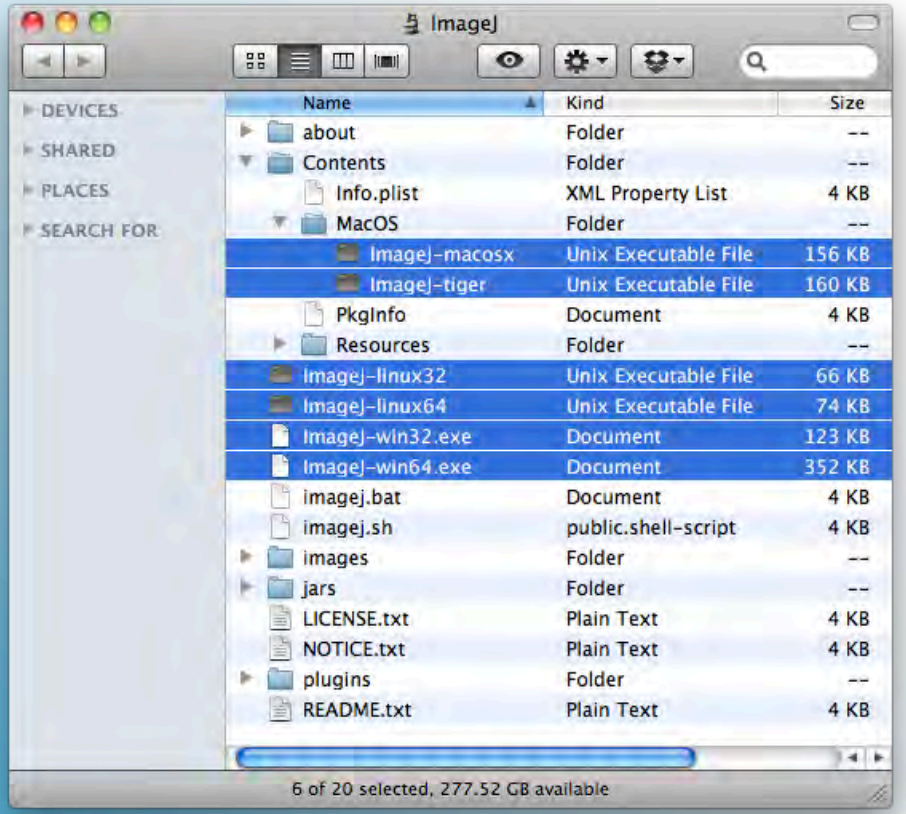


ImageJ2: Updater

- Uses SCIFIO (SCientific Image Format Input & Output) library for reading and writing data
- New formats can be added as SCIFIO plugins

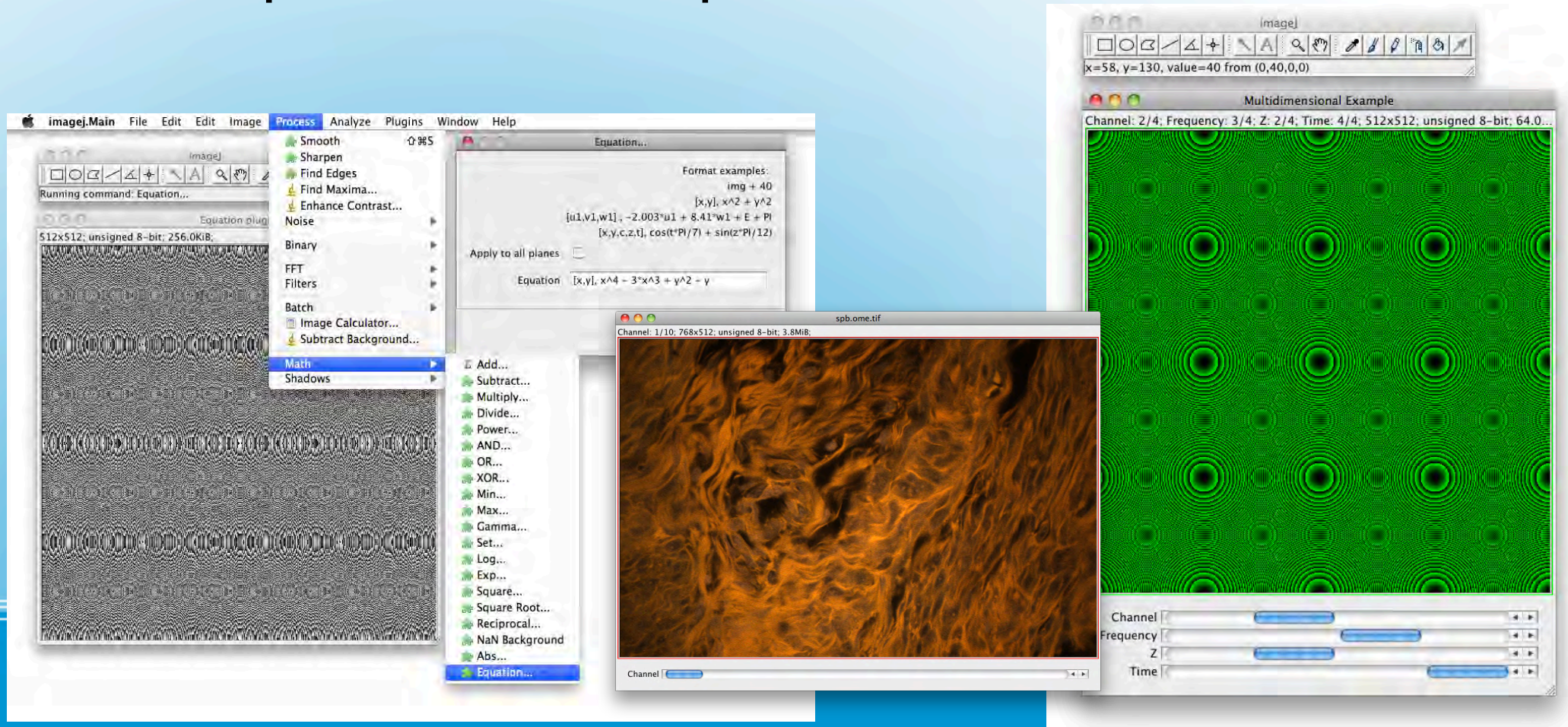
Data I/O

- Many options for launching ImageJ2
- Run scripts in batch mode
- Multiple platforms
- Run headless



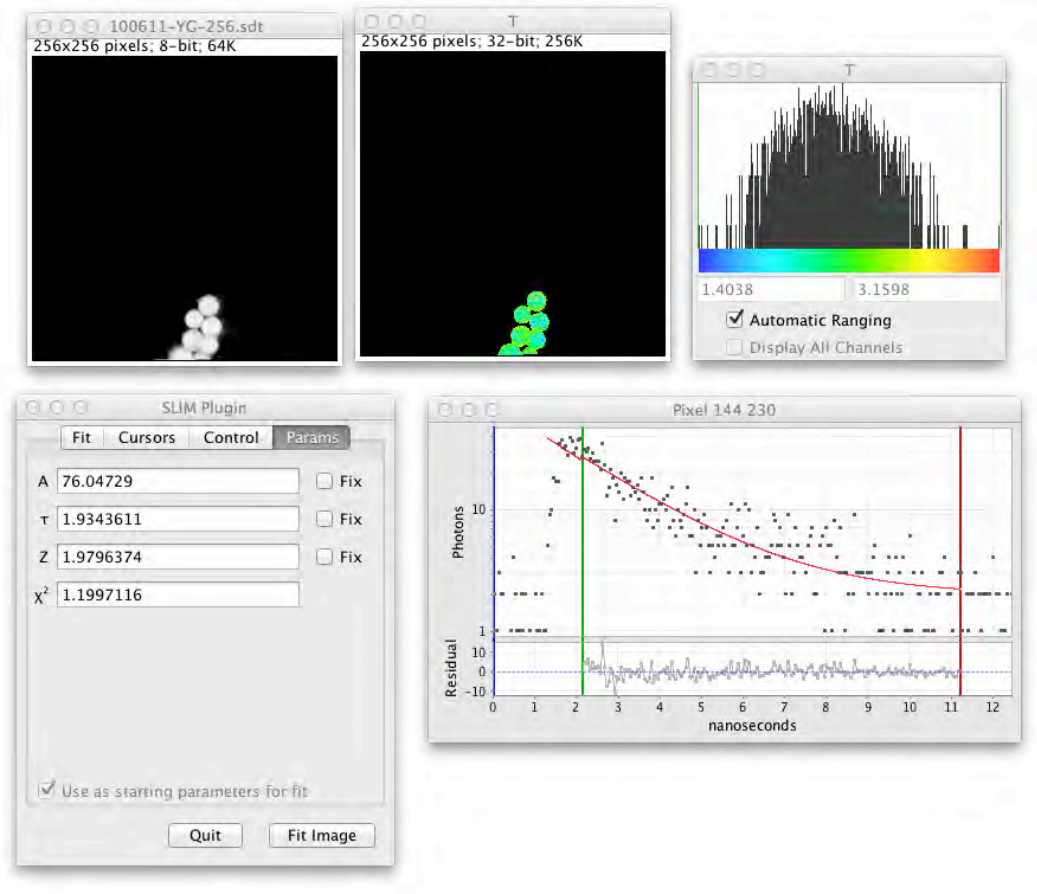
Launcher

- Support for more than five dimensions
- Composite more than seven channels
- Improved math equation editor



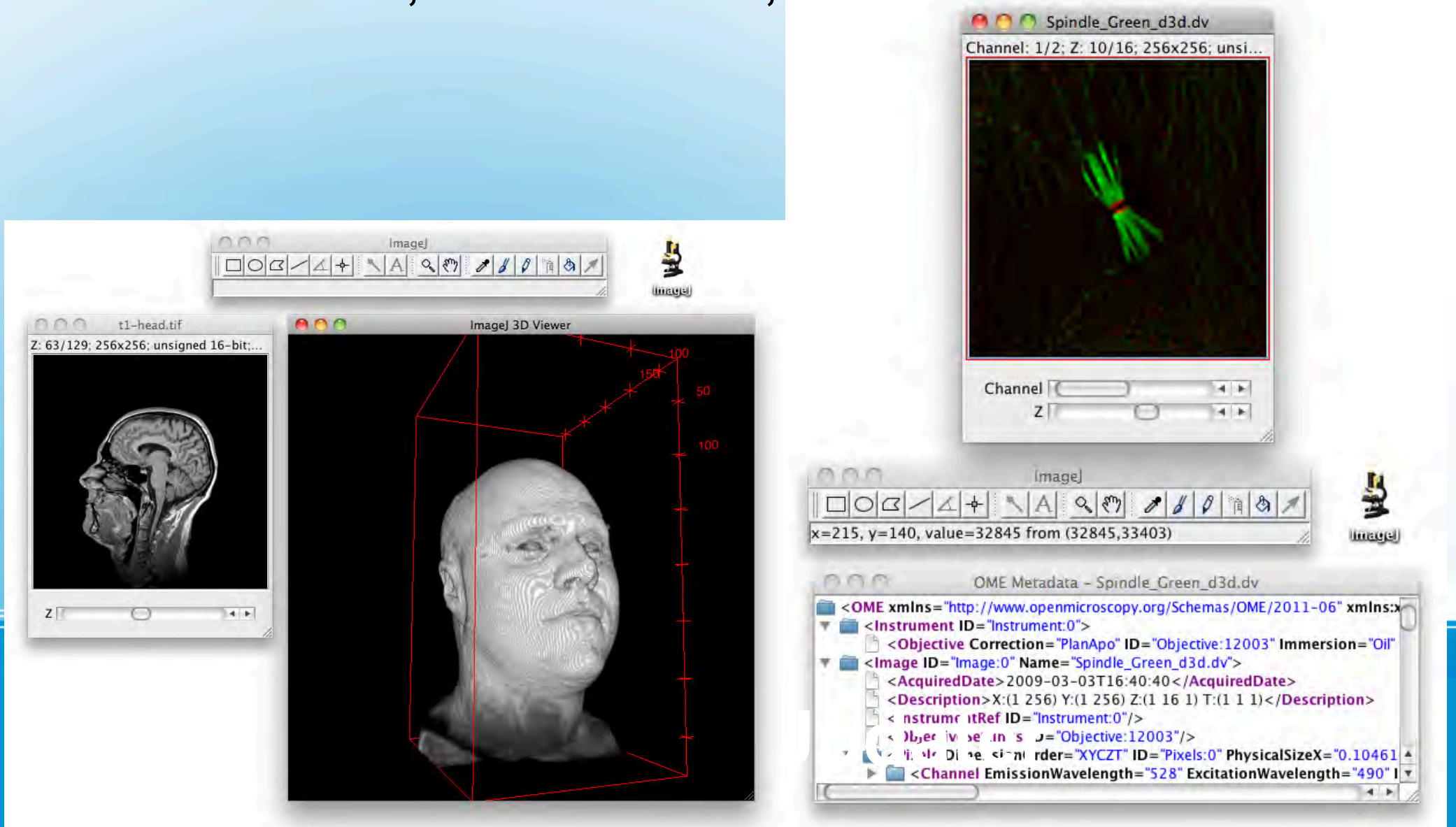
ImageJ2: N-Dimensional

- SLIM Plugin for visualizing and analyzing combined spectral lifetime image data
- Works with data in time domain
- Has open-source curvefitting library
- Available from LOCI update site

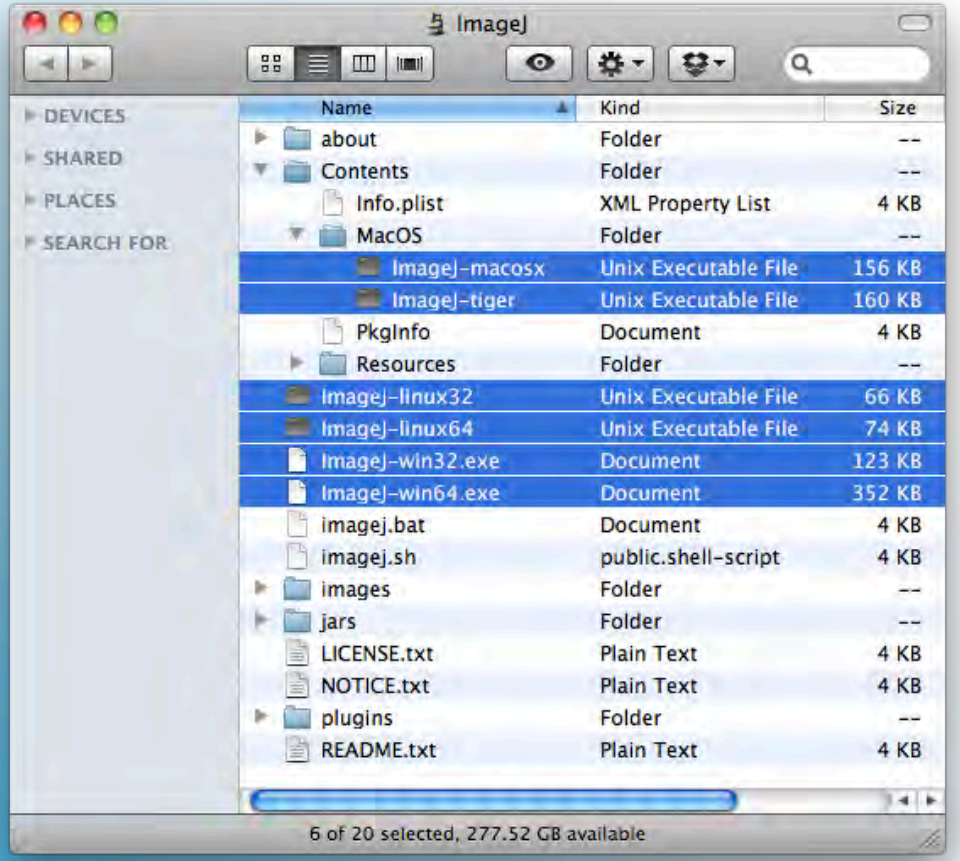


ImageJ2: Spectral Lifetime Analysis

- Install and use Fiji plugins
- 3D Viewer, Bio-Formats, more



- New launcher with many more features
- Based on Fiji's launcher
- Single, multi-platform distribution of ImageJ



ImageJ2: Launcher

- Separate data model from user interface
- Plugin framework works completely ***headless***
- Enables integration with other tools:
  - CellProfiler
  - KNIME Image Processing
  - OMERO servers and clients

ImageJ2: Interoperability

- Modular design allows applications to cherry-pick what parts of ImageJ2 they want to reuse

- Open source community development process (GitHub, Trac, Maven, update sites, mailing lists)

Professional Practice

Benefits of ImageJ2

- What Will ImageJ 2.0 Do for Me?
  - Work with existing plugins and macros
  - Work with new, exciting plugins and scripts
  - Handle larger, more complex datasets
  - Multidimensional visualization tools
  - Easier to link with other software
  - Easier plugin management

ImageJ2 and OME

- Need feedback on features community wants in a ImageJ client interface for OMERO
- What types of ImageJ analysis do users want to run on OMERO potentially?
- Specific OMERO-ImageJ use cases?
- Please give feedback to Dundee-LOCI developers collaborating on this

Acknowledgements

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  - Lee Kamentsky (CellProfiler), Melissa Linkert (Bio-Formats)
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