LOCI, Fiji & ImageJ2

Philosophy of optical research

- *In vivo* developmental biology
- Capture everything possible about a sample
- Multiple dimensions
  - Emission spectra
  - Lifetime
  - Cell polarization
- Greater resolution and range
  - Adaptive optics (peer deeper into the soup)
  - Longer time series

Philosophy of software development

- Interoperability
  - Leverage existing tools
  - “Glue code”
  - Implement missing features
- Open source (federally funded—ethical obligation)
- Of benefit to LOCI's research
- Of benefit to the broader scientific community

Computational tools

- Acquisition software
  - WiscScan
  - Micro-Manager
- Visualization & analysis
  - ImageJ, Fiji
  - VisBio
  - Slim Plotter, TRI2
  - FARSIGHT, ITK
  - CellProfiler
- Data management
  - Open Microscopy Environment

A major goal of ImageJ2 is to integrate these tools into a common framework

LOCI's vision of scientific workflow

- ***Acquire*** data however you like (e.g., µManager)
  - If possible, record data in a standard format (OME-TIFF)
  - If not, convert from proprietary format (Bio-Formats)
- ***Store and organize*** data centrally (OMERO server)
- ***Analyze*** data through various client & desktop software
  - Smart clients can access data from the server (Fiji via plugins, OMERO.webclient, OMERO.insight)
  - Other software can work with data on disk (Fiji, etc.)
- ***Record changes*** back in the database, with versioning
- ***Share*** data over the Internet with colleagues
  - Both informally and as part of publication

What is Fiji?

- An easy way to develop novel algorithms
  - Plugins, macros or scripts
- A distribution platform for those algorithms
- A suite of tools to facilitate these goals
  - Script Editor (currently supports 7 languages)
  - Tutorial Maker
  - Fiji Updater
    - Access latest updates easily
    - Submit code directly to Fiji repository

Fiji vs. ImageJ

- Fiji is a *distribution* of ImageJ bundled with useful scientific image processing routines

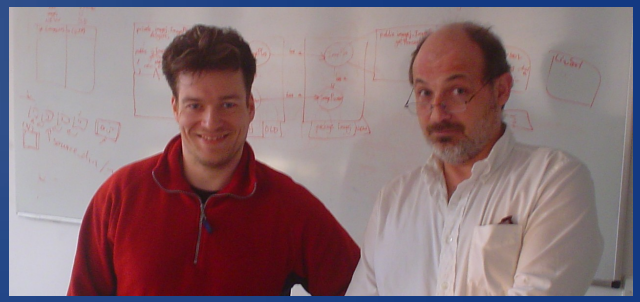
	ImageJ	Fiji
Size	~3-5 MB	~45 MB
Focus	Various image processing	Segmentation & registration
License	Public domain	GPL (various plugin licenses)
Tools	Minimal	Rich

- Fiji is the GNU/Linux distribution to ImageJ's Linux kernel
- "**F**iji **I**s **J**ust **I**mageJ—batteries included"
- "Some assembly required"

Origins of Fiji

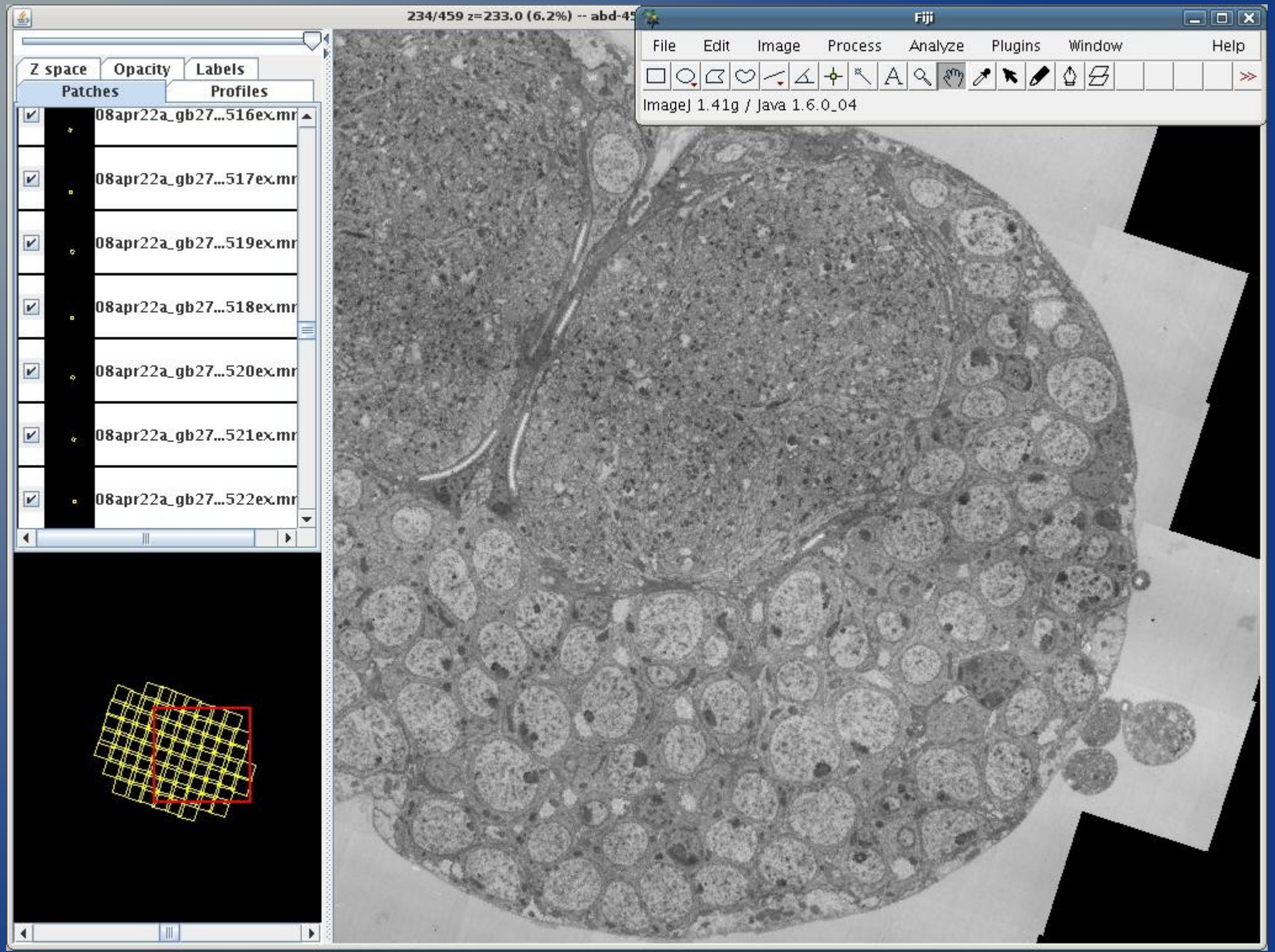
- Albert Cardona (“Father” of Fiji)

  - Institute of Neuroinformatics, Uni/ETH Zurich
  - Albert is a biologist who wrote TrakEM2
  - Began as a way to easily distribute TrakEM2
- Johannes Schindelin (“Mother” of Fiji)
  - Max Planck Institute, Cell Biology Division (MPI-CBG) in Dresden
  - Johannes is the one who did the hard work
  - Maintains the repository, build system, etc.



- Pavel Tomancak

  - Principal Investigator at MPI-CBG
  - Working to secure additional funding for Fiji

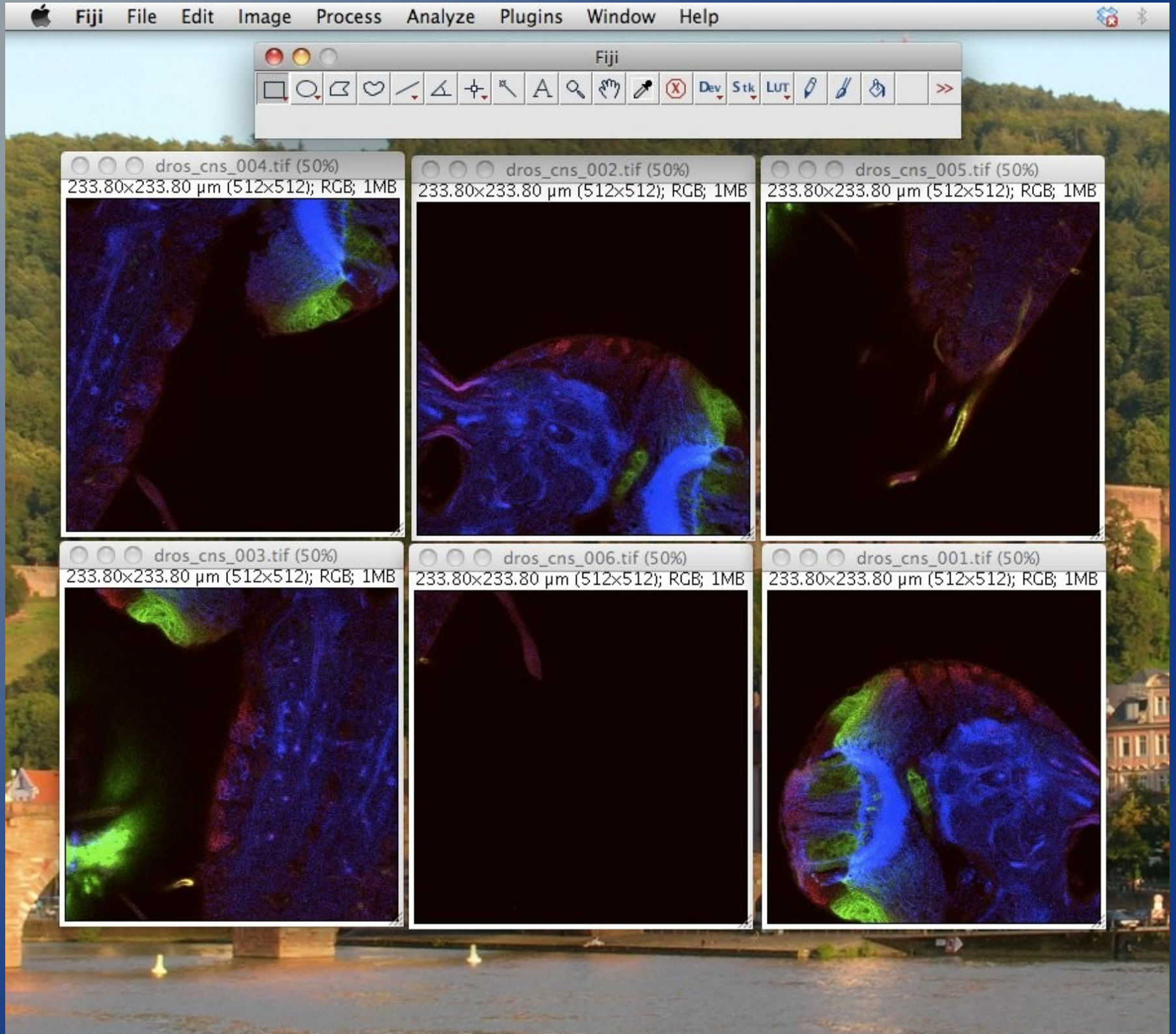

234/459 z=233.0 (6.2%) -- abd-45
Fiji
File Edit Image Process Analyze Plugins Window Help
ImageJ 1.41g / Java 1.6.0_04
Z space
Opacity
Labels
Patches
Profiles
08apr22a_gb27...516ex.mr
08apr22a_gb27...517ex.mr
08apr22a_gb27...519ex.mr
08apr22a_gb27...518ex.mr
08apr22a_gb27...520ex.mr
08apr22a_gb27...521ex.mr
08apr22a_gb27...522ex.mr

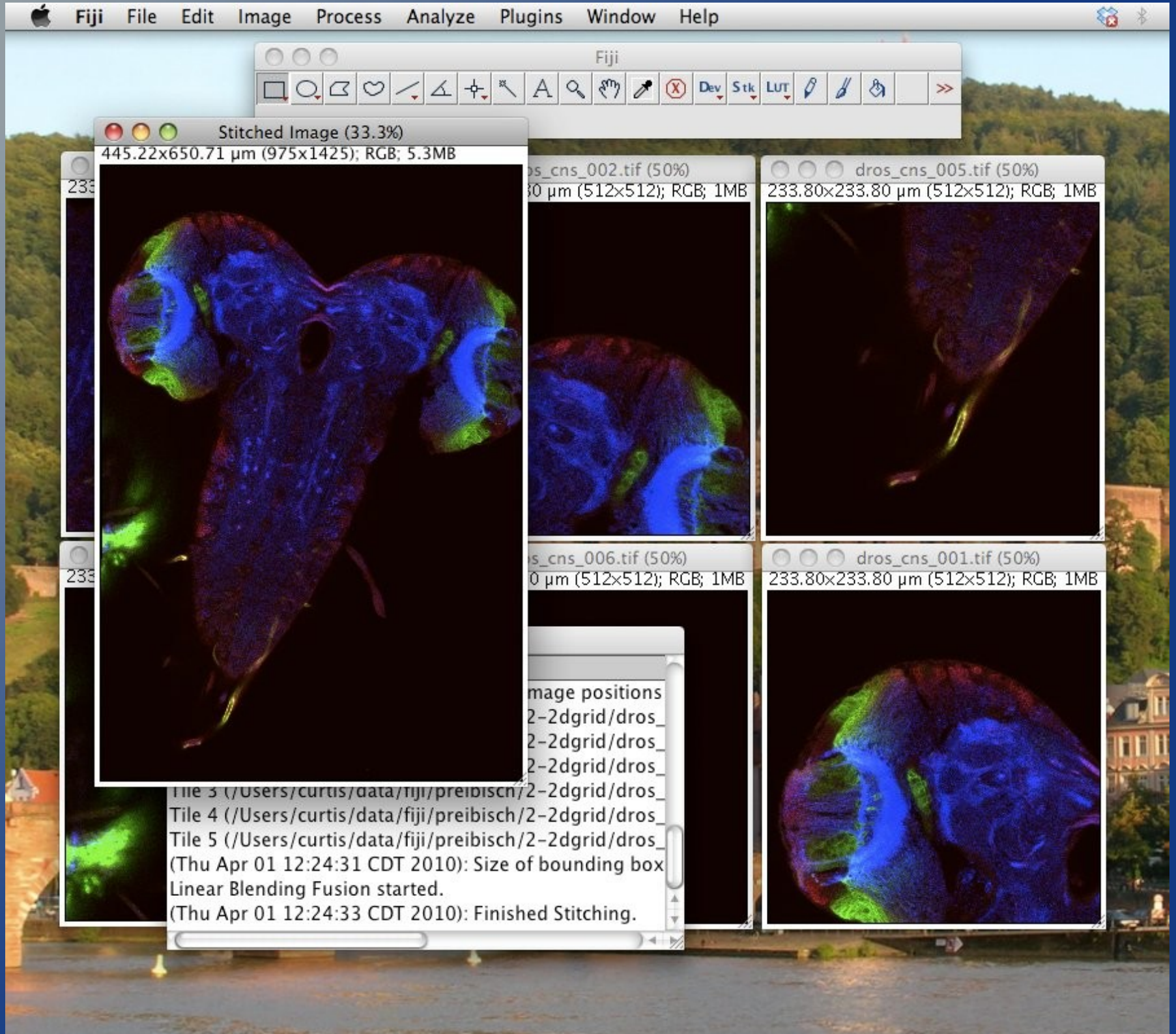
Other hackathon participants

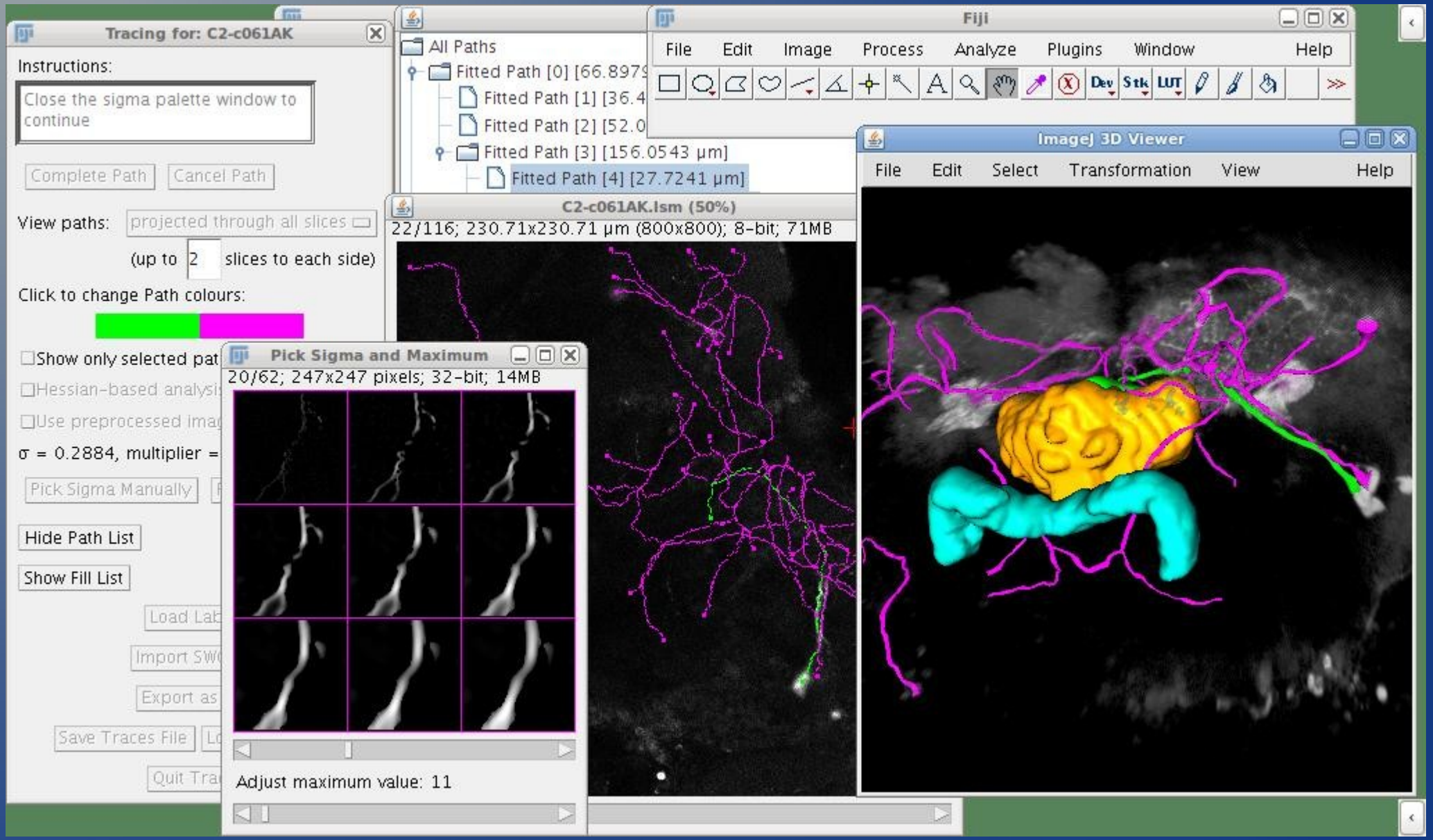
- Stephan Saalfeld & Stephan Preibisch

  - PhD students at MPI-CBG
  - Created excellent imglib library
  - Saalfeld developed much of TrakEM2's registration code
  - Preibisch developed Fiji's 2D & 3D Stitcher plugins
- Mark Longair

  - Just finished his PhD in neurobiology
  - Studies drosophila, specifically neuropils
  - Developed Fiji's Simple Neurite Tracer plugin
  - Moving from U. Edinburgh to Albert's group in Zurich


Fiji File Edit Image Process Analyze Plugins Window Help
Fiji
dros_cns_004.tif (50%)
233.80×233.80 µm (512×512); RGB; 1MB
dros_cns_002.tif (50%)
233.80×233.80 µm (512×512); RGB; 1MB
dros_cns_005.tif (50%)
233.80×233.80 µm (512×512); RGB; 1MB
dros_cns_003.tif (50%)
233.80×233.80 µm (512×512); RGB; 1MB
dros_cns_006.tif (50%)
233.80×233.80 µm (512×512); RGB; 1MB
dros_cns_001.tif (50%)
233.80×233.80 µm (512×512); RGB; 1MB

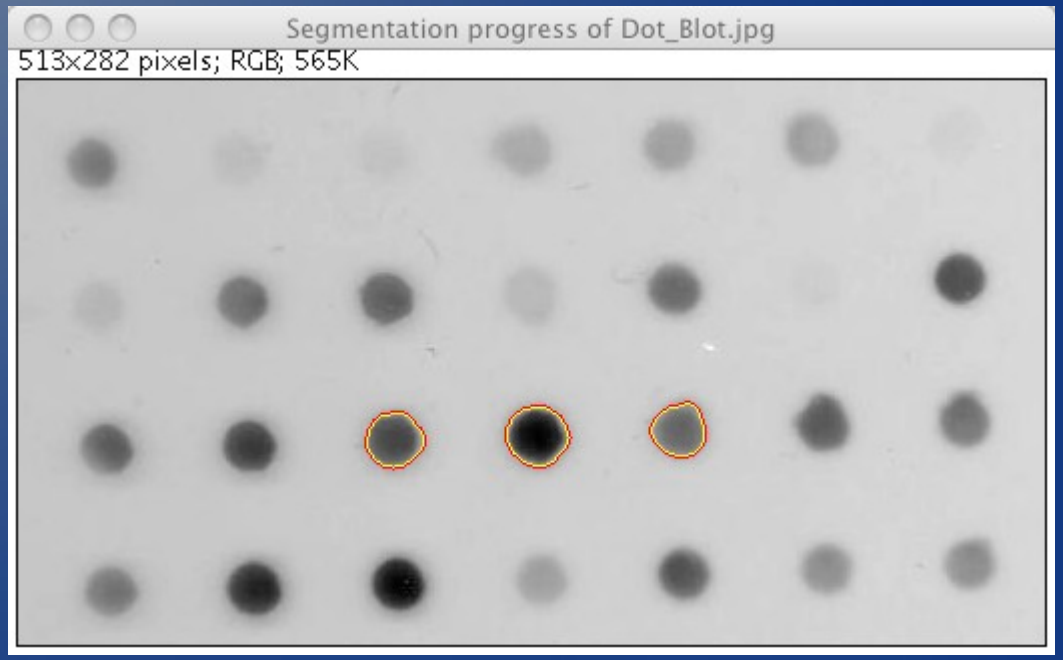

Fiji File Edit Image Process Analyze Plugins Window Help
Fiji
Stitched Image (33.3%)
445.22x650.71 µm (975x1425); RGB; 5.3MB
dros_cns_005.tif (50%)
233.80x233.80 µm (512x512); RGB; 1MB
dros_cns_001.tif (50%)
233.80x233.80 µm (512x512); RGB; 1MB
Tile 4 (/Users/curtis/data/fiji/preibisch/2-2dgrid/dros_
Tile 5 (/Users/curtis/data/fiji/preibisch/2-2dgrid/dros_
(Thu Apr 01 12:24:31 CDT 2010): Size of bounding box
Linear Blending Fusion started.
(Thu Apr 01 12:24:33 CDT 2010): Finished Stitching.


Tracing for: C2-c061AK
Instructions:
Close the sigma palette window to continue
Complete Path
Cancel Path
View paths:
projected through all slices
(up to 2 slices to each side)
Click to change Path colours:
Show only selected pat
Hessian-based analysis
Use preprocessed imag
σ = 0.2884, multiplier =
Pick Sigma Manually
Hide Path List
Show Fill List
Load Lab
Import SW
Export as
Save Traces File
Quit Tra
All Paths
Fitted Path [0] [66.897
Fitted Path [1] [36.4
Fitted Path [2] [52.0
Fitted Path [3] [156.0543 µm]
Fitted Path [4] [27.7241 µm]
C2-c061AK.lsm (50%)
22/116; 230.71x230.71 µm (800x800); 8-bit; 71MB
Pick Sigma and Maximum
20/62; 247x247 pixels; 32-bit; 14MB
Adjust maximum value: 11
Fiji
File Edit Image Process Analyze Plugins Window Help
ImageJ 3D Viewer
File Edit Select Transformation View Help

Other hackathon participants

- Bene Schmid
  - Developed Fiji's 3D Viewer (uses Java3D)
- Erwin Frise
  - Developed level sets plugin for image segmentation
- Larry Lindsey

  - U. Austin
  - Charged with maintaining a package called “Reconstruct”
  - Saw TrakEM2/Fiji could already do most of the same things
  - Spent his time learning about imglib

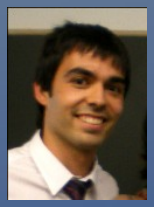

Segmentation progress of Dot_Blot.jpg
513x282 pixels; RGB; 565K

Other hackathon participants

- Verena Kaynig

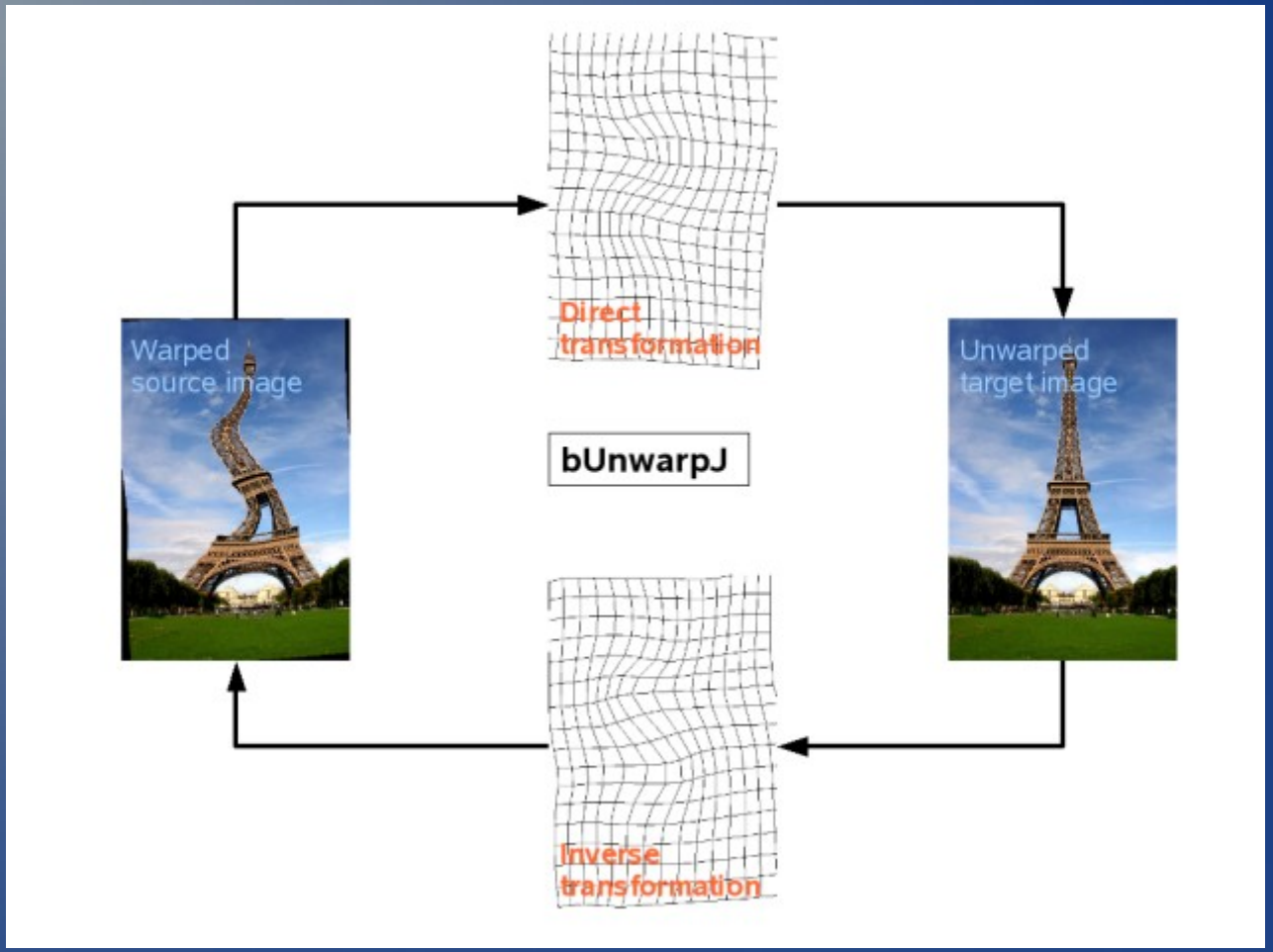
  - Computer scientist interested in machine learning
  - Wrote a “trainable segmentation” plugin

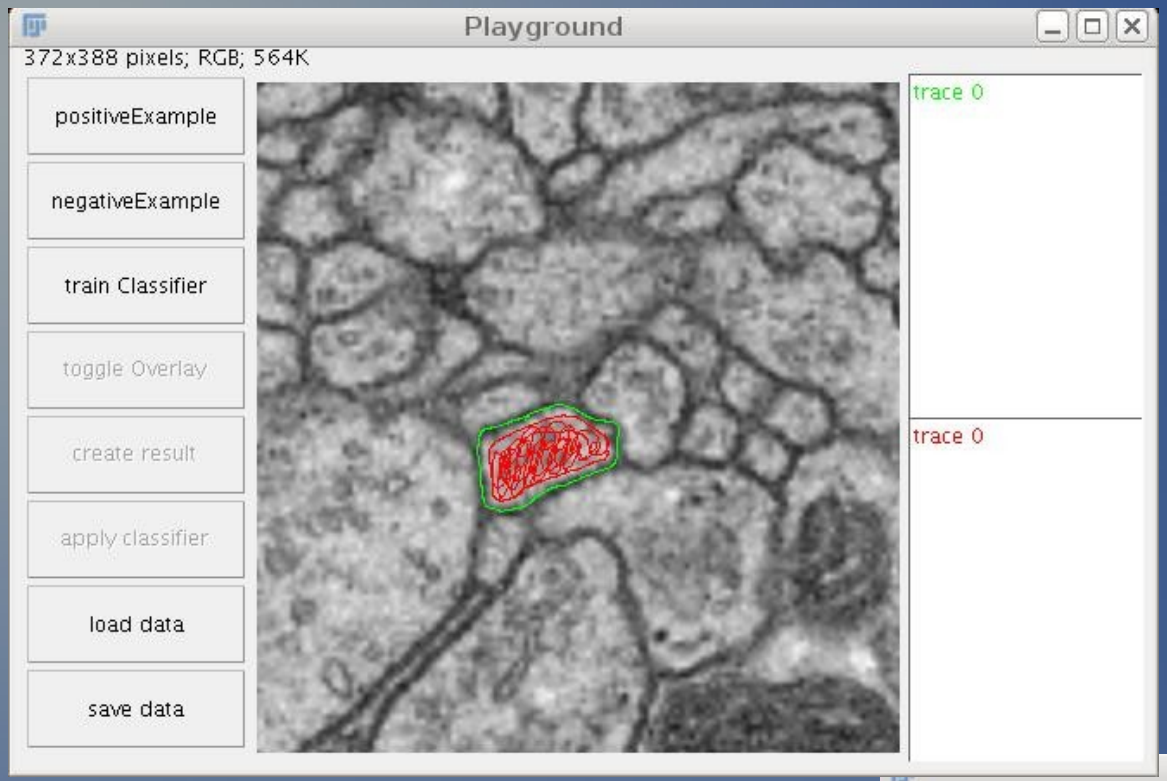
- Ignacio Arganda

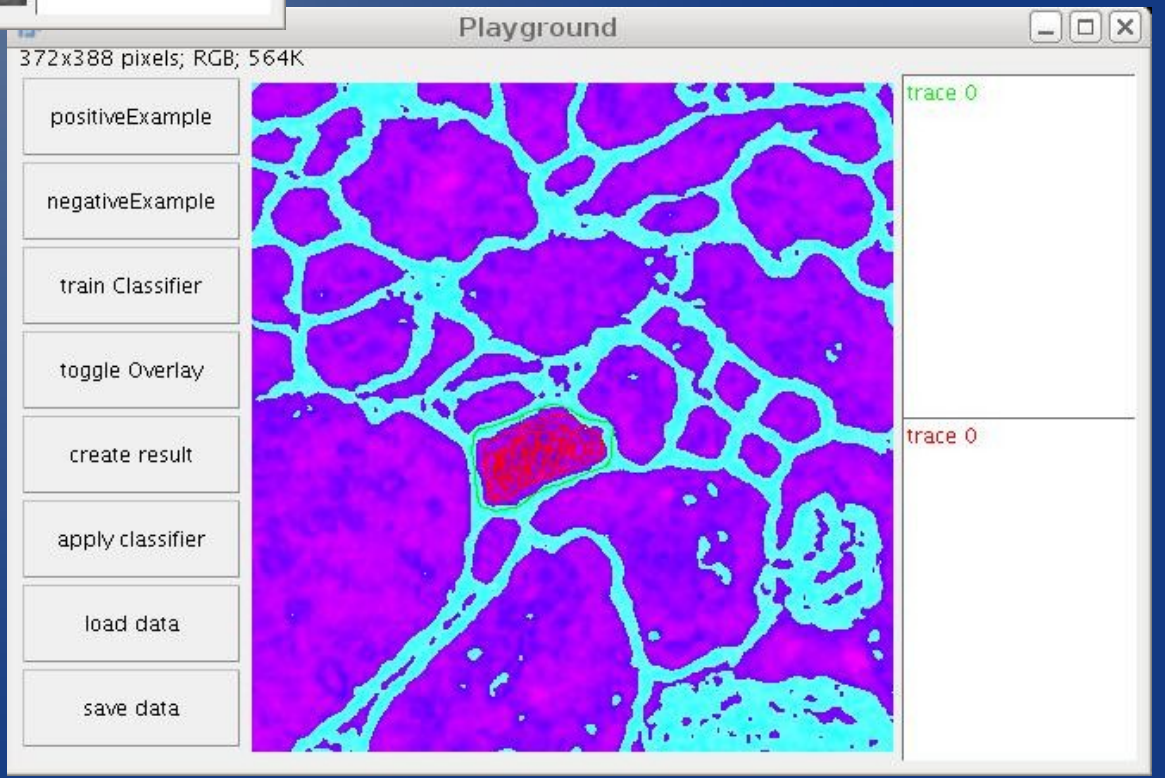


  - Developed Fiji's bUnwarpJ plugin for image registration
  - Improved Verena's Trainable Segmentation plugin after she left

- Rubén Muñoz
  - EMBL programmer working with Olympus ScanR systems
  - Discussed Bio-Formats and OME
  - Developed his own database with web interface
  - Uses OME-TIFF


Warped
source image
Direct
transformation
bUnwarpJ
Unwarped
target image
Inverse
transformation


Playground
372x388 pixels; RGB; 564K
positiveExample
negativeExample
train Classifier
toggle Overlay
create result
apply classifier
load data
save data
trace 0
trace 0

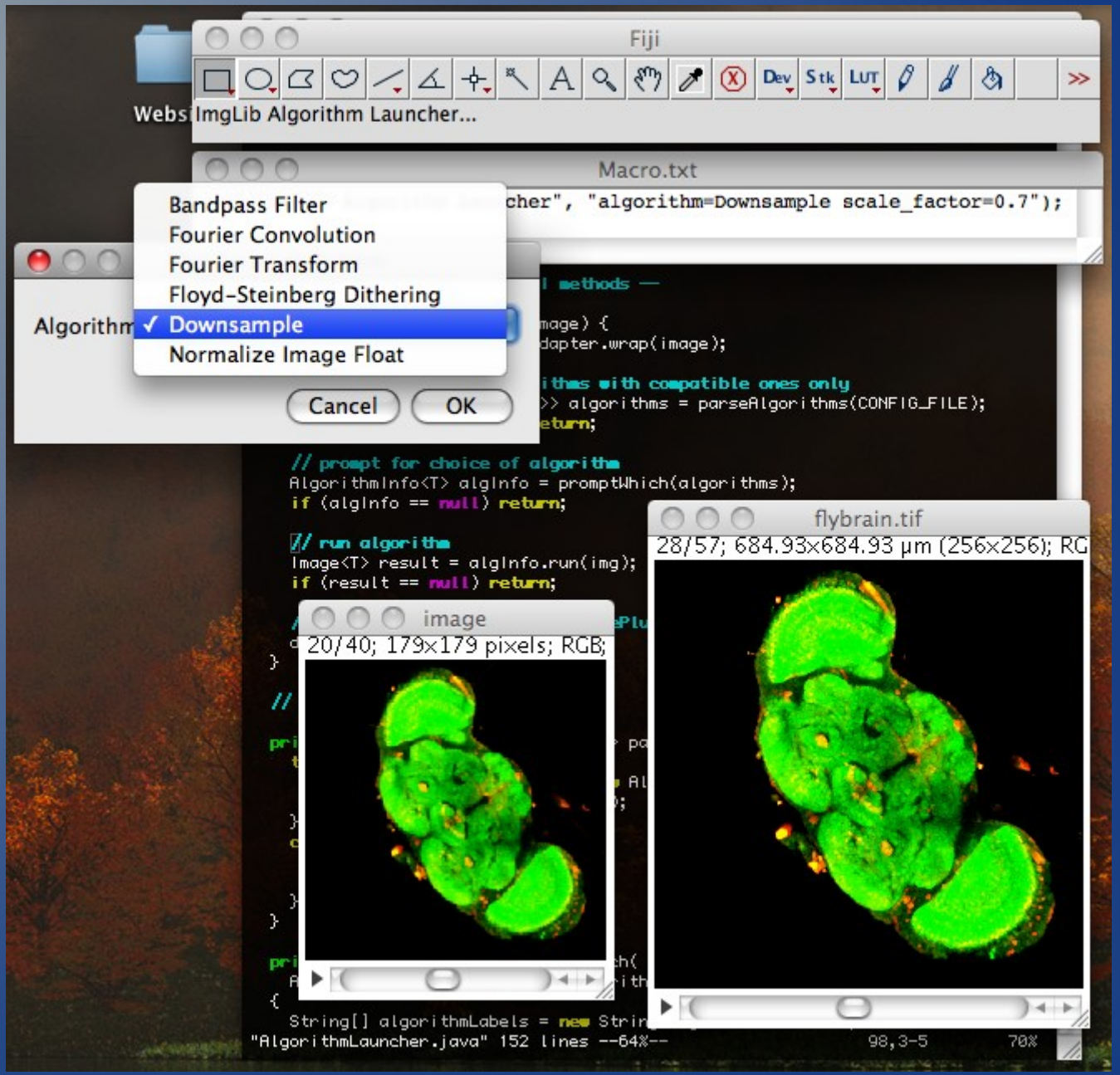

Playground
372x388 pixels; RGB; 564K
positiveExample
negativeExample
train Classifier
toggle Overlay
create result
apply classifier
load data
save data
trace 0
trace 0

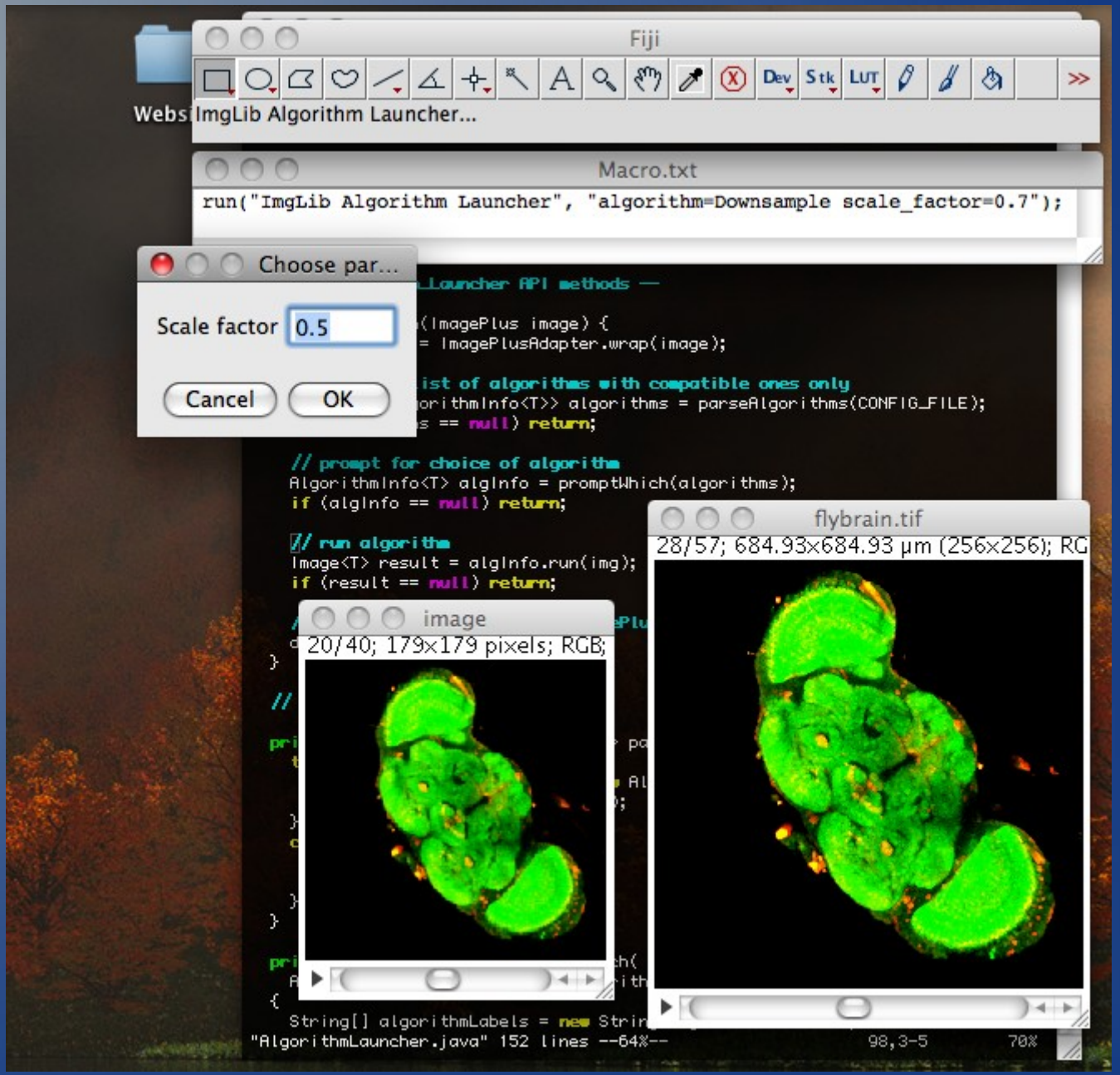
Other Fiji developers

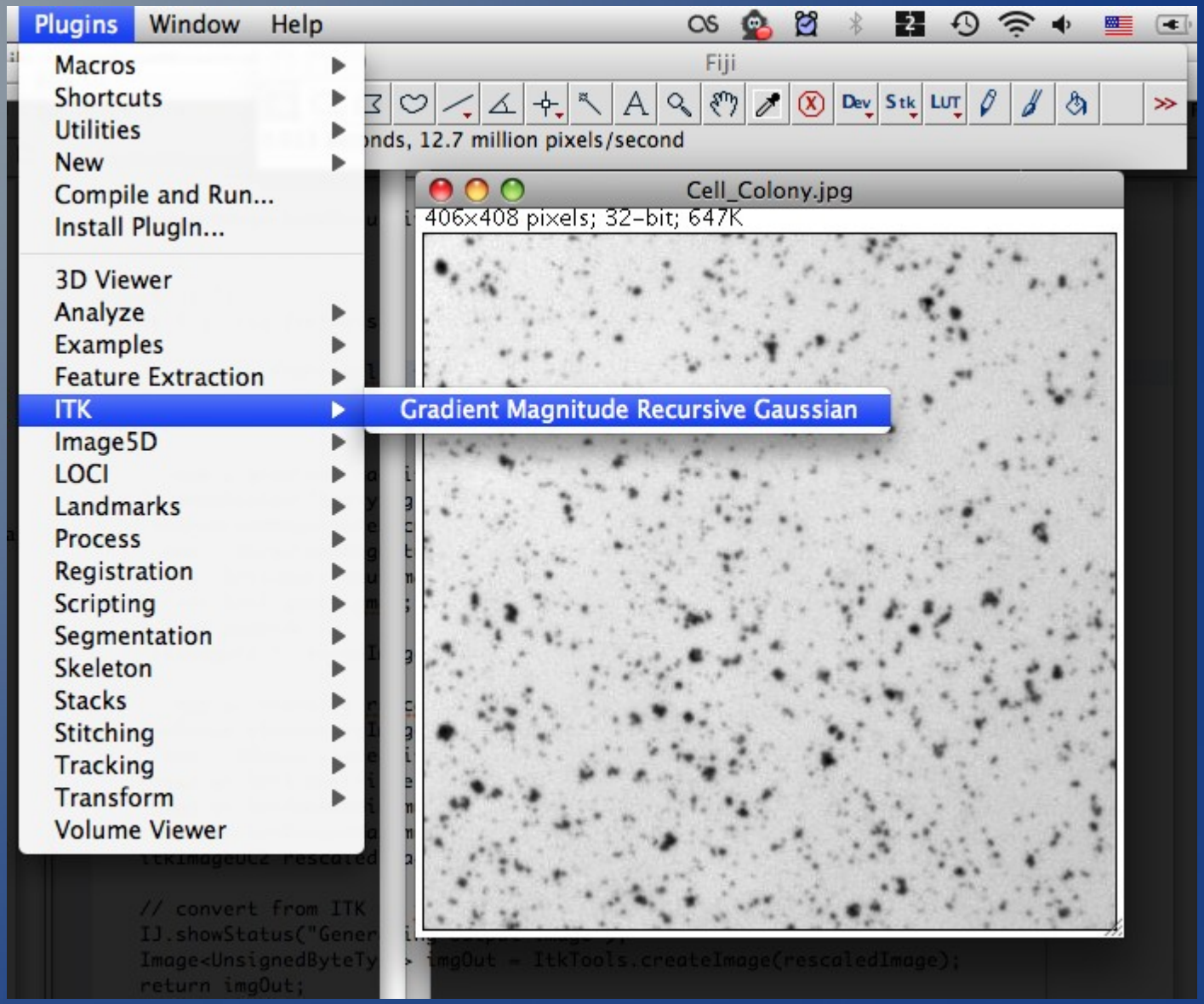
- Jean-Yves Tinevez
  - Institut Pasteur
- Gabriel Landini
  - Prof. Of Analytical Pathology
  - School of Dentistry, U. Birmingham, England
- Us!
- Probably others I'm forgetting

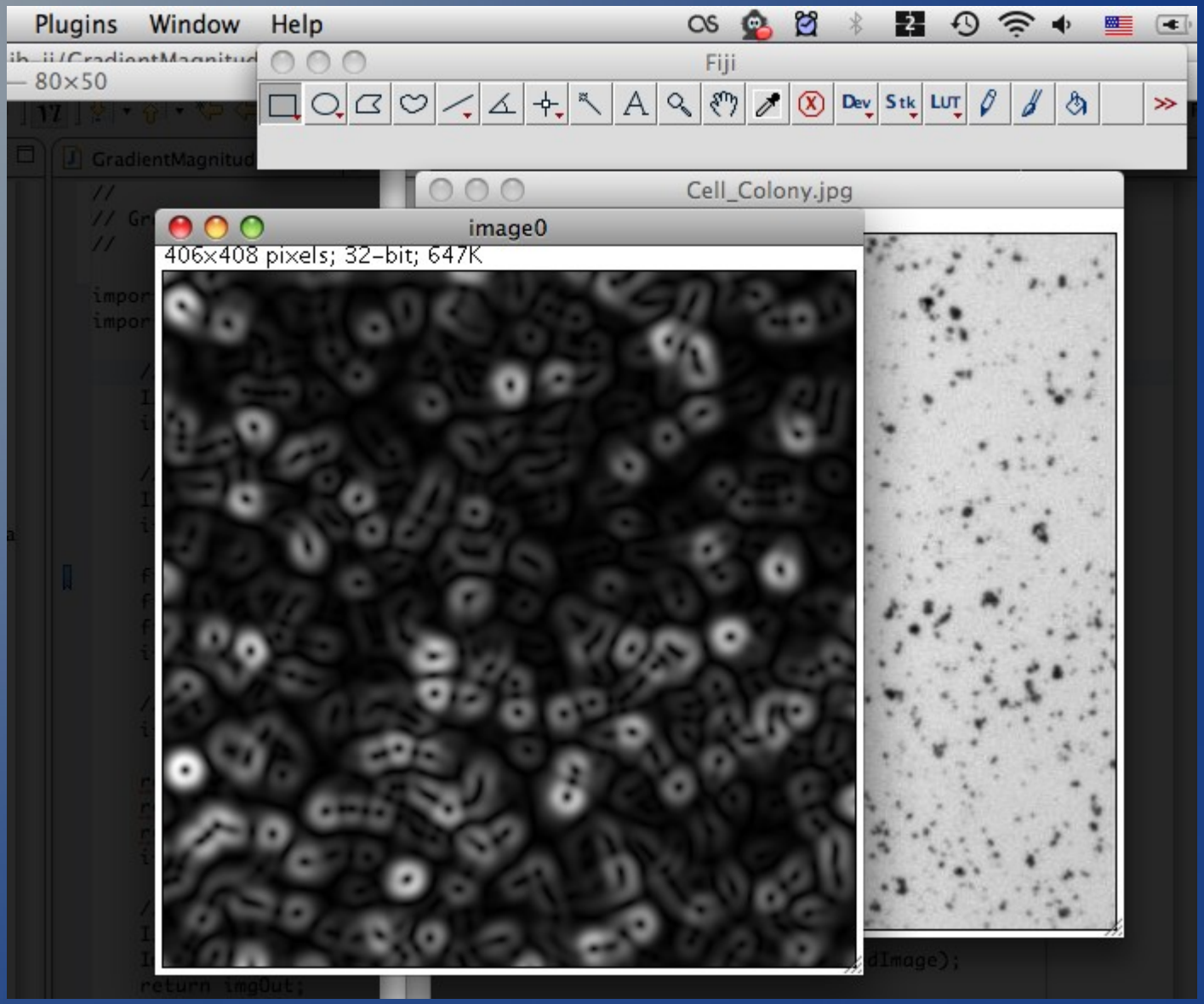
What I worked on

- Learned Fiji development workflow
- Algorithm launcher for imglib
  - Execute any imglib OutputAlgorithm in Fiji
- Autogenerated an ImageJ delegation layer
  - Attempt failed; now using a simpler approach
- Fiji/ITK integration
  - Proof of concept; call ITK image filters from Fiji
- Some work on Bio-Formats
  - Same dependency injection pains as us


Fiji
ImgLib Algorithm Launcher...
Macro.txt
cher", "algorithm=Downsample scale_factor=0.7");
Bandpass Filter
Fourier Convolution
Fourier Transform
Floyd-Steinberg Dithering
✓ Downsample
Normalize Image Float
Algorithm
Cancel
OK
image
20/40; 179x179 pixels; RGB;
flybrain.tif
28/57; 684.93x684.93 µm (256x256); RG
"AlgorithmLauncher.java" 152 lines --64%--
98,3-5
70%


Fiji
ImgLib Algorithm Launcher...
Macro.txt
run("ImgLib Algorithm Launcher", "algorithm=Downsample scale_factor=0.7");
Choose par...
Scale factor
0.5
Cancel
OK
// prompt for choice of algorithm
AlgorithmInfo<T> algInfo = promptWhich(algorithms);
if (algInfo == null) return;
// run algorithm
Image<T> result = algInfo.run(img);
if (result == null) return;
image
20/40; 179x179 pixels; RGB;
flybrain.tif
28/57; 684.93x684.93 µm (256x256); RG
"AlgorithmLauncher.java" 152 lines --64%--
98,3-5
70%


Plugins
Window
Help
Macros
Shortcuts
Utilities
New
Compile and Run...
Install PlugIn...
3D Viewer
Analyze
Examples
Feature Extraction
ITK
Image5D
LOCI
Landmarks
Process
Registration
Scripting
Segmentation
Skeleton
Stacks
Stitching
Tracking
Transform
Volume Viewer
Gradient Magnitude Recursive Gaussian
Fiji
Cell_Colony.jpg
406x408 pixels; 32-bit; 647K


Plugins
Window
Help
Fiji
Dev
Stk
LUT
>>
Cell_Colony.jpg
image0
406x408 pixels; 32-bit; 647K
80×50

Reminder: Vision of Fiji

- An easy way to develop novel algorithms
  - Plugins, macros or scripts
- A distribution platform for those algorithms
- A suite of tools to facilitate these goals
  - Script Editor (currently supports 7 languages)
  - Tutorial Maker
  - Fiji Updater
    - Access latest updates easily
    - Submit code directly to Fiji repository

Vision of ImageJ2

- Core image processing library for Fiji
- Major goals:
  - Improve the technical design
  - Integration and interoperability
  - Grow the ImageJ community

Vision of ImageJ2

- Improve the technical design
  - A solid base on which to build scientific tools
  - Flexible enough to interface with a variety of existing software
  - Able to represent scientific image data in a variety of contexts (e.g., N-dimensional)
  - Buzzwords like “modular” and “extensible”

Vision of ImageJ2

- Integration and interoperability
  - Adapt Slim Plotter & VisBio features into ImageJ2
  - Glue code between ImageJ2 and CellProfiler
  - Glue code between ImageJ2, ITK and FARSIGHT

Vision of ImageJ2

- Grow the ImageJ community
  - Backwards compatible with existing plugins as much as possible (at least initially)
  - The more users of ImageJ, the easier it will be to share analysis routines and results
  - Fiji certainly shares this goal

Vision: Fiji vs. ImageJ2

ImageJ2	Fiji
Software development infrastructure	Image processing algorithm development and deployment
General image processing	Scientific image processing, particularly life sciences (microscopy, neuroscience, but also computer vision)
Boring baseline stuff	Fun domain-specific applications

- Where ImageJ2 ends and Fiji begins is fluid
- We may develop analysis routines useful to LOCI that end up in Fiji rather than ImageJ2

Components of ImageJ2

- Major components of ImageJ2
  1) **Data model** – imglib library
  2) **Display** – Java AWT, JAI, Swing
  3) **Input/output** – Bio-Formats architecture
  4) **Regions of interest (ROIs)** – Java AWT, JHotDraw
  5) **Scripting & plugins** – Java 6 Scripting Framework
- All of these areas have significant limitations and will benefit from enhancements and refactoring

Components of ImageJ2

- To start, each of us focus on one?
  1) **Data model** – Aivar?
  2) **Display** – Rick?
  3) **Input/output** – Barry?
  4) **Regions of interest (ROIs)** – Brian?
  5) **Scripting & plugins** – Grant?

An old MIDIAS diagram, just for fun... :-)