

## Fiji-OMERO Workflows

### Introduction

This is a step-by-step guide to the workflows used in OMERO training sessions and the videos produced to illustrate the basic Fiji-OMERO image analysis workflows. Links to the videos are in the **Other resources** section at the end of the document.

Instructions on downloading and installing the OMERO.insight-ij plugin for Fiji and ImageJ are in the <u>Using ImageJ with OMERO</u> section. (http://help.openmicroscopy.org/imagej.html)

### Manual workflow

1 Open the Fiji application. Select **Plugins > OMERO > Connect to OMERO**.

A version of OMERO.insight will open and can be used as normal.

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As ImageJ needs to load the plugin software, it can take a minute or two for the OMERO login screen to appear, and then another minute for the progress bar to start moving and complete.

If you have not previously connected to OMERO, enter the server address. Enter your OMERO **Username** and **Password**. Click **Login**.

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4.14-6655-6000 OMERO is distributed under the terms of the GNU GPL. For more information, visit openmicroscopy.org	44.4-ex5-6000 OMERO is distributed under the terms of the GNU GPL. For more information, visit openmicroscopy.org	
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3 In the OMERO.insight window, select the image you wish to work with.



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4 Right-click on the image and select **View in ImageJ...**. In the Bio-Formats Import Options dialog, click **OK**.



5 In Fiji, select Image > Adjust > Threshold.

Click on the **Auto** button, which should bring up the default for the image. If necessary use the sliders to adjust the threshold values to obtain the desired thresholding.

Select the **Dark Background** checkbox. Click **Apply** and close the dialog.





## 6 Select Analyze > Analyze Particles.

In the Analyze Particles dialog, check the values are as shown below, ensuring the **Display Results**, **Clear Results** and **Add to Manager** checkboxes are selected. Click **OK**.



The following dialogs should be visible:

- the image with ROIs highlighted
- Results table
- ROI Manager



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### 8 In Fiji, select Plugins > OMERO > Save Results to OMERO.

Note: If you are not logged into the OMERO server you will be prompted to log in.



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In the **Save ImageJ Results** dialog select the **Image from Current Window** radio button. Check the **ROI** and **Measurements** checkboxes. Enter a file name for the **Measurements File** in the text box. Click **Save**.

Click Yes in the Import Images dialog.







10 In the Import Location dialog, select the Project and/or Dataset from the drop-downs to save the results to. Click Add to the Queue.

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**11** Once the import is completed, the OMERO server takes a little time to generate the ROIs in OMERO form.

Once the ROI generation process is complete, select the imported image to show the ROIs attached.

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🖬 🛱 Fiji Workflow [1]	Image ID: 2394 Owner: ome-user-30 ome-user-30			
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🗄 - 🧊 Orphaned Images	Add Description			
	Import Date: 2016-09-05 13:06:57			
	Dimensions (XY): 2688 x 2048 Pixel Type: uint16			
	Pixels Size (XY) (µm): 338.67x338.67 Z-sections/Timepoints: 1 x 1			
	Channels: 0			
	ROI Count: 1508			
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### **Scripting workflow**

### Additional setup

**1** Download the OMERO.matlab plugin from:

http://downloads.openmicroscopy.org/latest/omero5

OMERO 5.2.5 Downloads			
Clients   Plugins   Additional   Servers	API   Python	Java   Code   Components   P	revious versio
<ul> <li>Information on this release of OMERO is in</li> </ul>	n the release annot	uncement	
<ul> <li>Full documentation is available as <u>web doc</u> on our <u>Help website</u></li> </ul>	umentation or PD	F documentation and there are user gu	ides for the clie
<ul> <li>A standard OMERO user just needs to dow server e.g., 5.0.0 clients will connect to 5.0</li> <li>If you do not have an institutional server, y</li> <li>OMERO client downloads</li> </ul>	vou can apply for a	ickage with the same major version as to 5.1.0 servers. n <u>account on our Demo server</u>	their institution
Client	Size	File Name	Checksum
<b>Windows</b>	83.48 MB	OMERO.insight-5.2.5-ice35-b28- win.zip	b4e00aa7 ( <u>SHA1</u> )
Mac OS X	83.26 MB	OMERO.insight-5.2.5-ice35-b28- mac.zip	26aaa275 ( <u>SHA1</u> )
👌 Linux 🗍	83.13 MB	OMERO.insight-5.2.5-ice35-b28- linux.zip	8303e6eb ( <u>SHA1</u> )
<ul> <li>OMERO.web is part of the server package</li> <li>Full instructions for installing the client are</li> </ul> OMERO plugin downloads	, so individual user e on the Help webs	rs do not need to install it locally ite: <u>Getting Started with OMERO.ins</u>	ight Version 5.2
Plugin	Size	File Name	Checksum
🛓 ImageJ / Fiji	76.57 MB	OMERO.insight-ij-5.2.5-ice35- b28.zip	2b3e5901 ( <u>SHA1</u> )

## 2 Unzip the archive. Drag-and-drop or copy the libs > omero\_client.jar file into the jars folder of Fiji.

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commons-collections	29/07/2016 09:26	JAR File	\mu win32	05/09/2016 10:35	File fold
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🔄 log4j	29/07/2016 09:26	JAR File	🔚 ant-junit	02/09/2016 16:03	JAR File
🔄 omero_client	20/07/2016 00:26	JAR File	🔄 ant-launcher	02/09/2016 16:03	JAR File
🔄 slf4j-api	29/07/2016 09:26	JAR File	ant-nodeps	02/09/2016 16:03	JAR File
🔄 slf4j-log4j12	29/07/2016 09:26	JAR File	🔤 args4j-2.0.25	02/09/2016 16:03	JAR File





# **3** Download the example script and related files from:

https://github.com/bramalingam/Omero-Imagej-Scripts

Links to individual files:

bg\_subtract.ijm master/credentialsTemplate.txt omero\_batch\_analysis.py

(https://github.com/bramalingam/Omero-Imagej-Scripts/blob/master/bg\_subtract.ijm) (https://github.com/bramalingam/Omero-Imagej-Scripts/blob/master/credentialsTemplate.txt) (https://github.com/bramalingam/Omero-Imagej-Scripts/blob/master/omero\_batch\_analysis.py)

Editing scripts

## 1

Open Fiji.

Use **File > Open...** to select and open the <code>omero\_batch\_analysis.py</code> file. The jython script will open in the script window.

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omero_batch_analysis.py							
1 import os		<b></b>					
2 from os import path							
4 from java.lang import Long							
5 from java lang import String							
7 from java util import Array list	6 from Java.lang.Long import longValue						
8 from jacrav import array							
9 from java, lang, reflect import Array							
10 import java							
11							
12 # Omero Dependencies							
13 from omero.gateway import Gateway							
14 from omero.gateway import LoginCredentials							
15 from omero.gateway import SecurityContext							
16 from omero.gateway.exception import DSAccessException							
17 from omero.gateway.exception import DSOutOfServiceException							
18 from omero.gateway.facility import BrowseFacility							
19 from omero.gateway.tacility import DataManagerFacility							
20 trom omero.gateway.model import DatasetData							
21 from omero.gateway.model import Experimenterbata							
23 from omero log import logger							
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```
2 Edit the following:
```

```
Line 131:
From: HOST = "omero-latest-analysis.docker.openmicroscopy.org"
To: HOST = "your.omero.server.url"
```

### Line 133:

```
From: datasetId = "1"
To: datasetId = "x"
```

Where "x" is an OMERO Dataset ID of your choice.

### Line 139:

To: CREDENTIALS = "PATH\_TO/credentials.txt"

### Line 142:

```
From: macroFilePath =
    "/Users/bramalingam/Desktop/FijiDemonstration/bg_subtract.ijm"
To: macroFilePath = "PATH_TO/bg_subtract.ijm"
```

### Line 145:

```
From: paths = "Users/bramalingam/Desktop/FijiDemonstration"
To: paths = "PATH TO"
```

Where "PATH\_TO" is the file path on your system to the files you downloaded previously.

- **3** Use **File > Save** to save the script file.
- 4 Use File > Open... to select and open the credentialsTemplate.txt file.
- 5 Enter the following using the OMERO login credentials you have been given:

Line 1: From: USERNAME = <OMERO USERNAME> To: USERNAME = <your\_username>

Line 2: From: PASSWORD = <OMERO PASSWORD> To: PASSWORD = <your password>

6 Use File > Save As > Text... to save the credentials file as credentials.txt in the same directory.



7 Click the **Run** button to run the script.



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- 8 Once the script is completed, use OMERO to check the processed images, which should be found in the dataset with the ID used in Step 2, Line 133. Repeat Steps 5 7.
- To run the same macro on a different set of Images, in Step 2 Line 133, enter a different Dataset ID number.
   Repeat Steps 5 7.
- 10 To run a different macro on the same set of Images, in Step 2 Line 142, enter a different .ijm file name. Repeat Steps 5 - 7.

### Other resources

Videos of these workflows:

fiji\_omero\_manual\_workflow.mov

fiji\_scripting\_demo.mov

(http://downloads.openmicroscopy.org/movies/omero-5-2/mov/fiji\_omero\_manual\_workflow.mov) (http://downloads.openmicroscopy.org/movies/omero-5-2/mov/fiji\_scripting\_demo.mov)

Fiji Scripting Tutorials:

http://imagej.net/Jython\_Scripting

https://www.ini.uzh.ch/~acardona/fiji-tutorial/

