Tutorial: How to import images into Acquiarium

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Last Updated Thursday, 25 October 2012

This tutorial will teach you, how to import images into Acquiarium 1.6. After importing you will be able to analyze the images in Acquiarium. If you have a problem or comments related to this tutorial, feel free to send an e-mail to .Â

Tutorial materials

Software: We have used Acquiarium 1.6 (built on June 30, 2009, with Bioformats support) to prepare this tutorial. However, the described
procedure should be valid for the other Acquiarium versions too.

Input data: Tutorial uses three sample images (Mitosis1.tif, Mitosis2.tif, and Mitosis3.tif) in Olympus TIF format. They are 3-D confocal images containing two channels (Chromatin and kinetochores). Mitosis1.tif and Mitosis3.tif have 55 z-sections and Mitosis2.tif has 71 z-sections.

Output: You will obtain three Volumes Of Interest ready for image analysis in Acquiarium.

Step 1: Create new depository

First, create a new empty depository. Acquiarium uses depositories for data storage.

- Run Acquiarium if it is not running yet.

- Select Depository->Open Depository... menu item, in the main application window. You can press 'CTRL-O', alternatively. Open new depository dialog will apear.

- Type or select new directory name (using [Browse...] button) in Directory field in the dialog. Acquiarium will be storing all images and computed results in this directory.

Step 2:Â Define channels

You can import many 2-D or 3-D multi-channel images in Acquairium. Channels typically correspond to fluorescent channels. In order to be able to refer to the channels you have to define their names. We import two-channel images in this tutorial. One channel contains nuclei and the other contains kinetochores. Therefore, we define two channels.

- Select Depository->Edit Channels menu item, in the main application window. You can press 'CTRL-E' or click on this icon in toolbar. Channels dialog will apear.Â

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Press 'Insert' or 'i' key on your keyboard to open the dialog for entering the name of one channel. Put the channel name of the first channel in Name: filed and press 'Enter'. The suitable name for the first channel in our tutorial is "Chromatin".

Remark: The version we are using in this tutorial does not have any fluorochrome database installed. Therefore, you cannot select fluorochrome and exitation/emission filter pair. These pieces of information are crucial for performing image acquisition using Acquiarium. You would need the up-to-date database describing your imaging system in this case.

- Press 'Insert' or 'i' key again and type the name of the second channel. We type "Kinetochores" and press 'Enter'.

- You will see this table with two channels in Channel dialog. OBR. Click on [OK] button to confirm the table.

Step 3:Â Start import plug-in

Now, we are ready to initiate the import of images.

- Start import plug-in by selecting Plugins->Import->Individual Images menu item in the main application window.

- Select the images you want to import. We select three images Mitosis1.tif, Mitosis2.tif, and Mitosis3.tif and confirm the dialog. Import parameters dialog appears.

Acquiarium goes through all the selected images and checks whether the selection contains grayscale and/or colour images. If there is at least one grayscale image in your list then the Gray channel section of the dialog is enabled. You can define in which Acquiarium channel (named in step 2) the grayscale images will be imported, here. If there is at least one RGB image in your list then the RGB channels section of the dialog is enabled. You can match the input colour channels and Acquiarium channel names defined in step 2, here. Optionally, you can rewrite information about camera binning (1x1, by default) and image resolution (taken from images metadata by default).

- The three images provided are recognized (incorrectly) as grayscale images. Select 'Chromatin' option from Gray channel choice list.

- Click on [OK] button. All three images are imported into one channel 'Chromatin' and each image is associated with one Volume Of Interest (VOI) in Acquiarium. VOIs are independent units, which are the subject of image analysis plug-ins.

- You can see how the images were imported. See Tutorial: How to browse images in Acquiarium.

Step 4:Â Reorganize VOIs

It can happen, that the images are imported incorrectly or the result does not meet your wishes. For example, two channels are mixed in one channel after import as in our case (Step 3). This is one of the reasons why Acquiarium provides VOI manipulation plug-ins. We will show how to split one channel VOIs into multi-channel VOIs.

- Open VOI browser.Â You can do it by selecting Browse->Browse VOIs menu item in the main application window or by pressing 'CTRL+V' key on the keyboard or by clicking on the magnifying glass icon in the main toolbar. VOI broweser will appear.

- If you open the VOI browser for the first time then the channel mapping will not be defined and you will see No data image in the first column of each VOI. You have to assign colours to your channels, first. It can be done in the right top part of the VOI browser. For example to map Chromatin channel to blue colour simply select 'Chromatin' item in Blue: choice list. Thumbnail image for each VOI (containing Chromatin channel) will appear in the first column.

- Start channel split plug-in. Select Plugins->VOI manipulation->Channel Split. Channel Split dialog will appear.

- Select the VOIs you want to reorganize. Individual items can be selected by left mouse clicking. Use Shift and CTRL keys with left mouse click to select more than one item.

- Increase the number of channels per output form 1 to 2. The channel of selected VOIs will be split in the middle of the image stack and every half will be stored in the channels you enter in choice lists. We enter 'Chromatin' in the upper choice list and 'Kinetochores' in the lower one.

- Click on [OK] button. New VOIs having 2 channels will be created.

- We recommend to map 'Kinetochores' channel into the red in the right part of the VOI browser. You will see the blue-red thumbnail image of the new VOIs in the first column of the VOI browser. The import is over.

var gaJsHost = (("https:" == document.location.protocol) ? "https://ssl." : "http://www.");
document.write(unescape("%3Cscript src='" + gaJsHost + "google-analytics.com/ga.js'
type='text/javascript'%3E%3C/script%3E"));

try {
var pageTracker = _gat._getTracker("UA-6322859-1");
pageTracker._trackPageview();
} catch(err) {}