

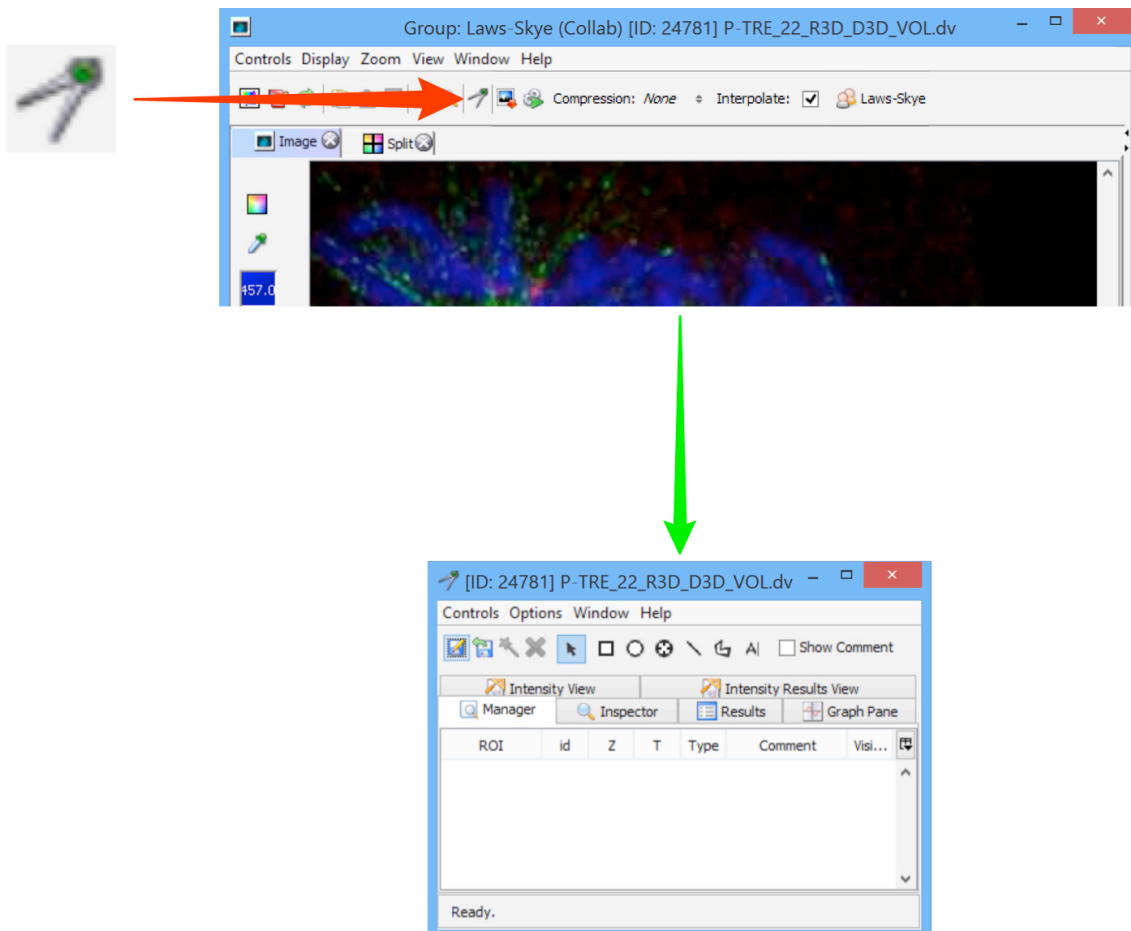
Using ROIs and the Measurement Tool

The measurement tool in OMERO.insight is used to draw **Regions of Interest** (ROIs) and perform some basic analysis.

OMERO.web allows you to view existing ROIs, but not edit them or create new ones.

Drawing ROIs

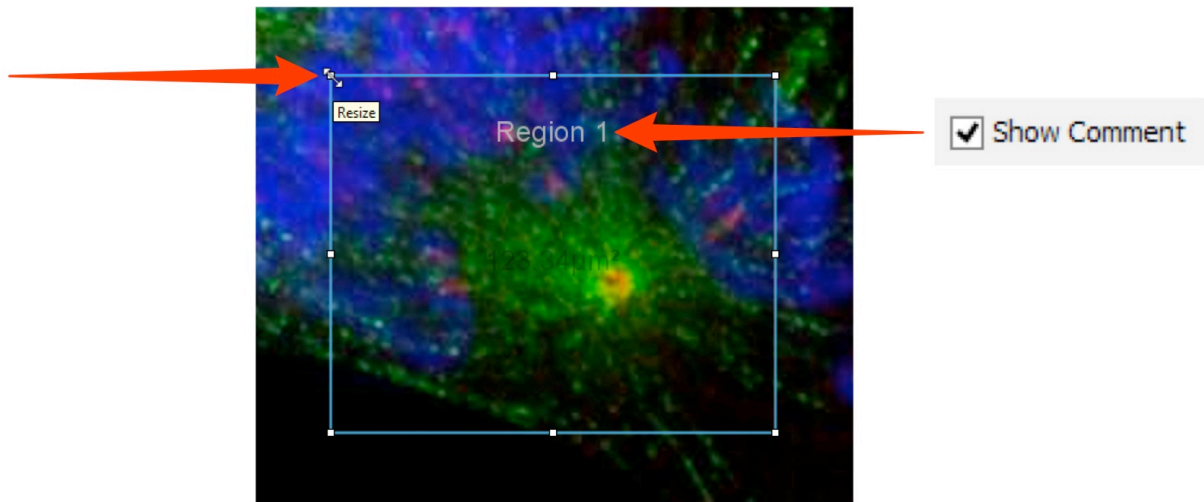
- 1 Open the image in the **Image Viewer** and click on the **Measuring Tool** icon to open the tool palette.



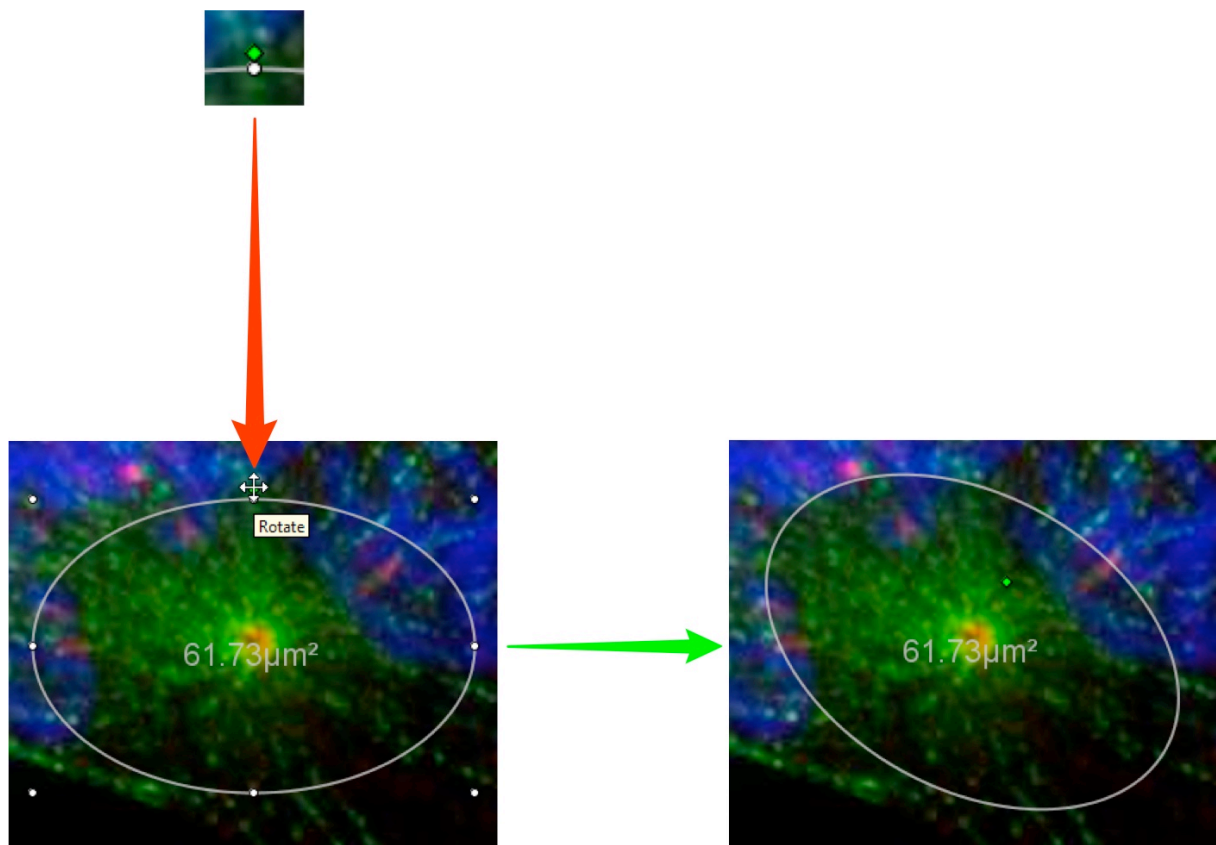
- 2 Use the **ROI selector** icon to select ROIs for analysis, resizing, reshaping or moving on image.
Select the appropriate tool to draw rectangular, ellipse, point, line or polygon ROIs.
Text ROIs can be added with the text tool.



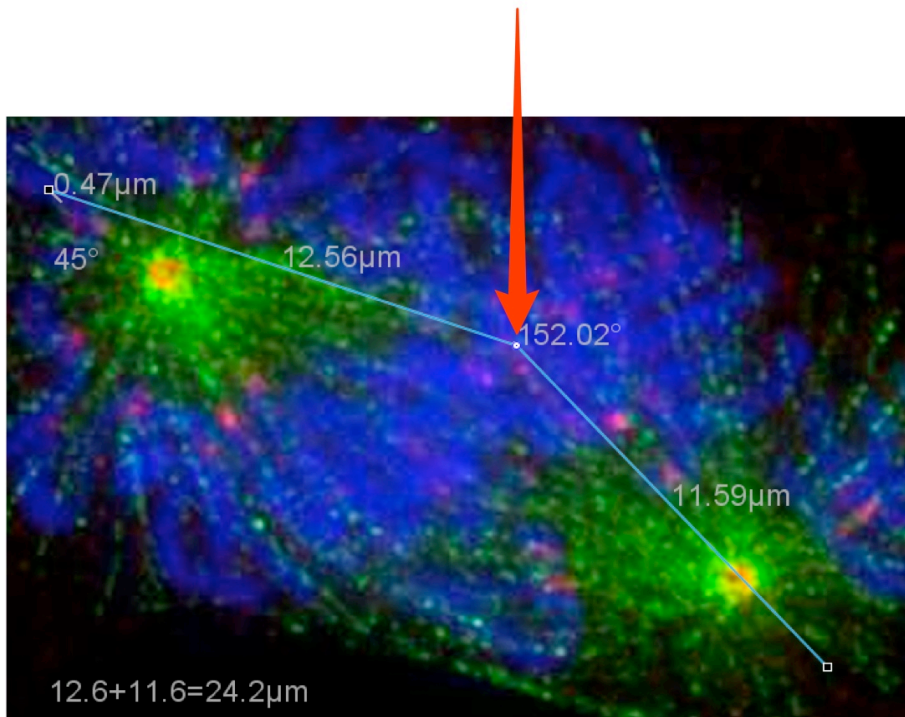
- 3** Click on the **Rectangle** or **Ellipse** icons in the tool bar to draw a rectangular or elliptical ROI.
Use the corners or edges to resize the ROI.
Use the selection tool to move ROI on image.
Double-click on the ROI to add a label to the ROI.
Use the **Show Comment** checkbox in the toolbar to toggle labels on and off.



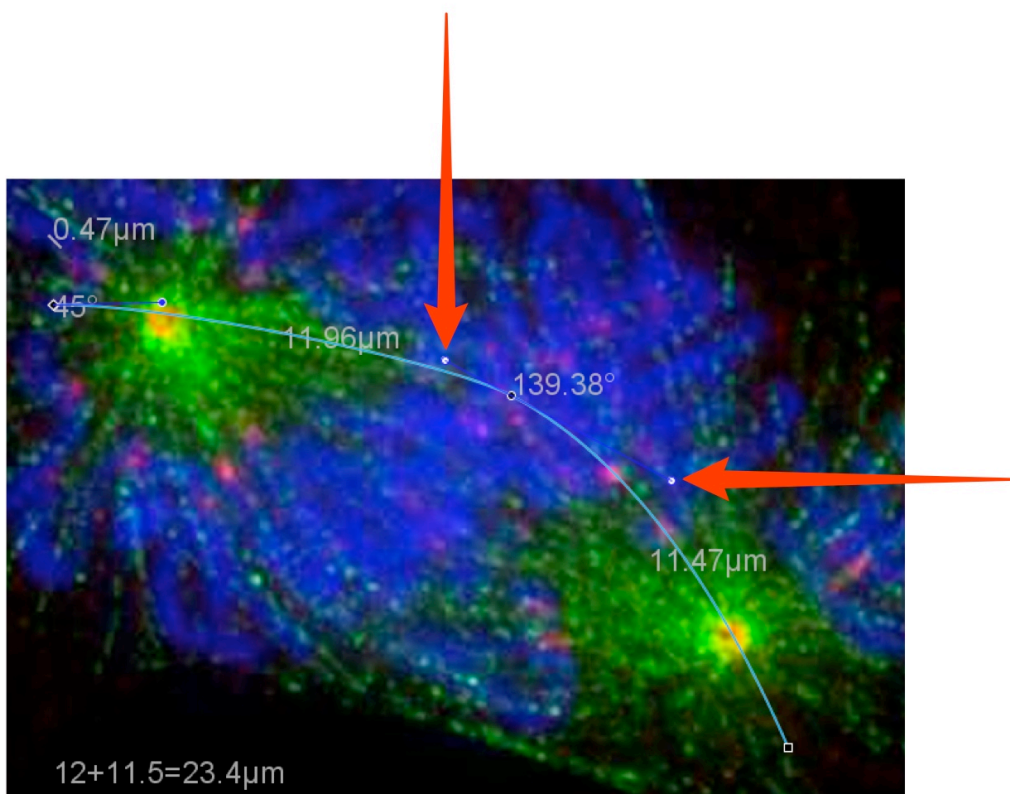
- 4** On elliptical ROIs click on and drag the green dot at the top to rotate the ellipse.



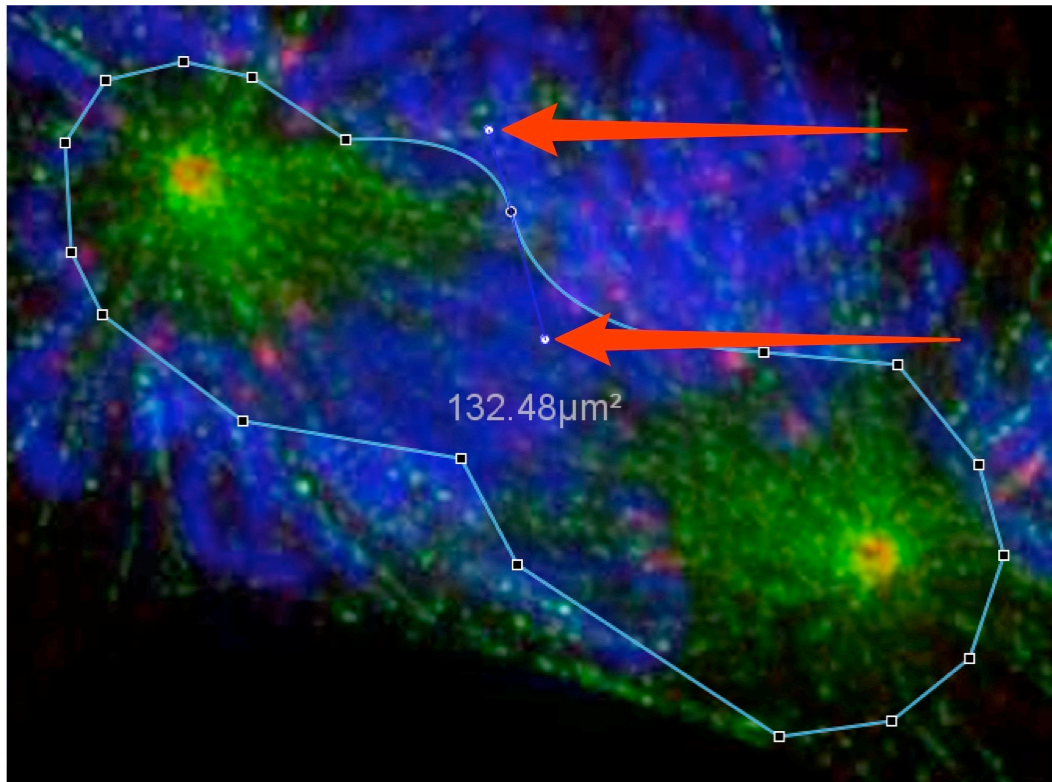
- 5** Double-click on a line ROI to insert an additional point into the line.



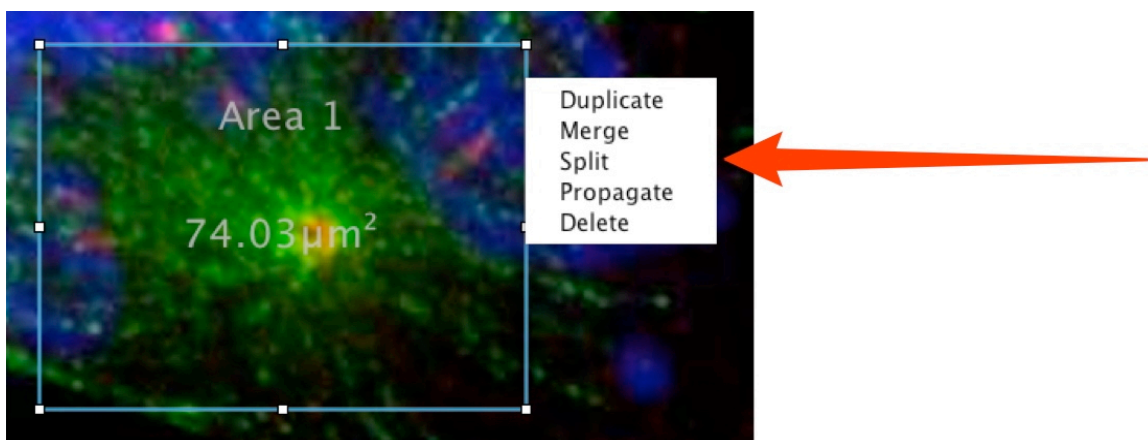
- 6** **Ctrl-click** (Windows) or **Cmd-click** (Mac) changes the type of a point. The default type is an edge point, with collinear control point, unconstrained control point and quadratic curve point the other options. Point types other than the default edge point, have Bezier controls that allow the line adjacent to the point to be curved as needed e.g. to pass through specific structures.



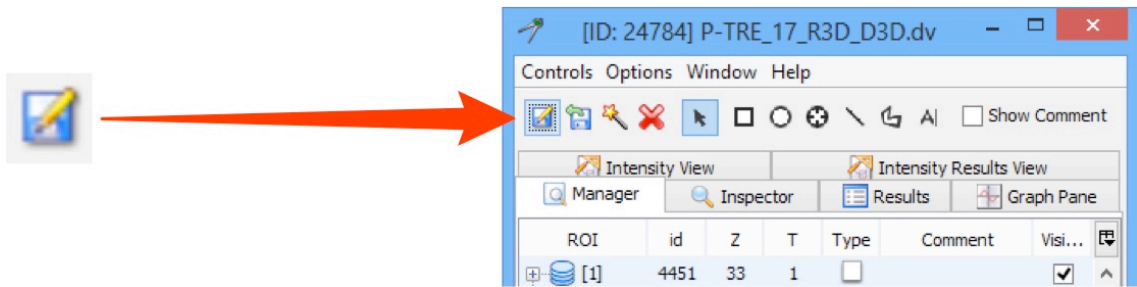
- 7** Click the **Polygon** icon, click at the starting point on the image, and then click again to start drawing a polygon ROI. Each subsequent click will add another point. Double-click to complete the polygon. Double-click with the ROI selector tool to add additional points to an existing polygon. The type of any point on a polygon ROI can be changed by **Ctrl-click** or **Cmd-click**, and the line curved using the Bezier controls.



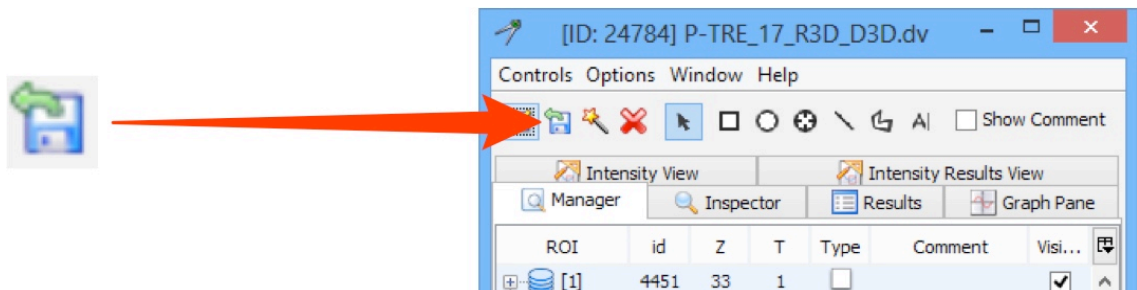
- 8** Right-click on any ROI to use the contextual menu to perform the operations available.



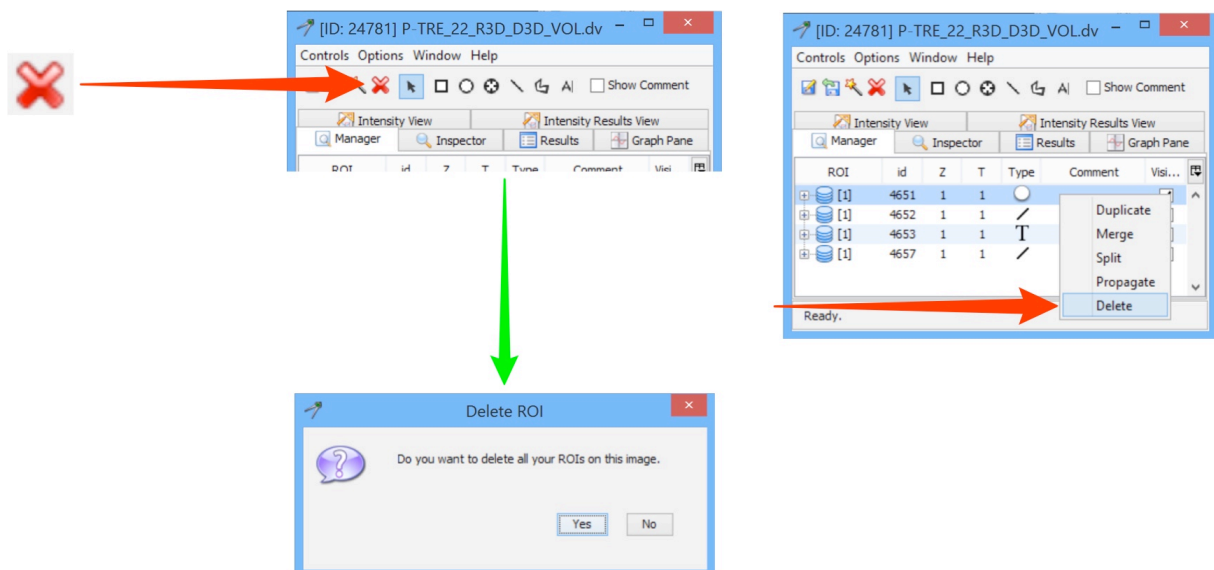
- 9 Click on the **Save** icon to save ROIs.



- 10 Click on the **Upload** icon to upload ROIs from an external file.



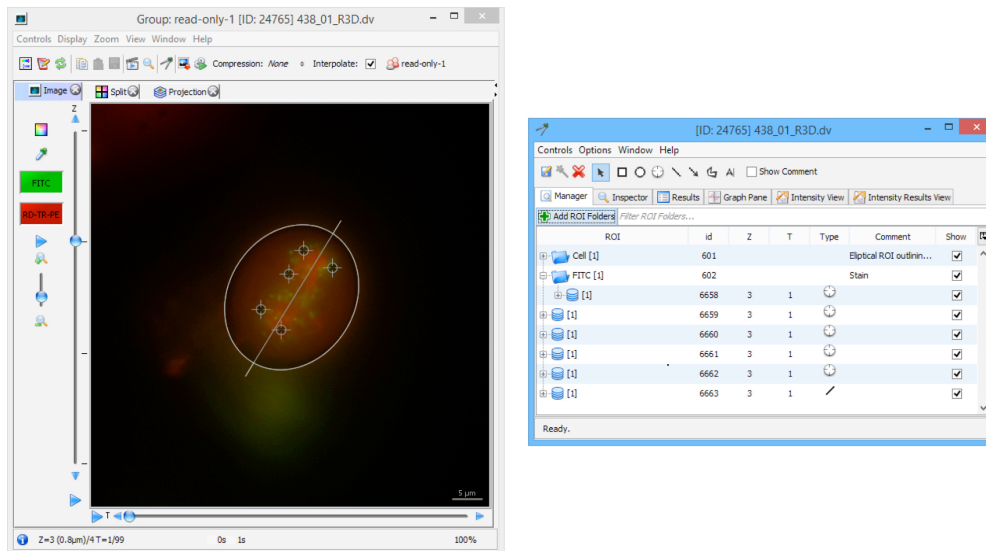
- 11 Click the **Delete** button to delete all ROIs on an image.
Right-click on an individual ROI and select **Delete** to delete only a single ROI.
Select multiple ROI's, right-click and select **Delete** to delete those ROIs.



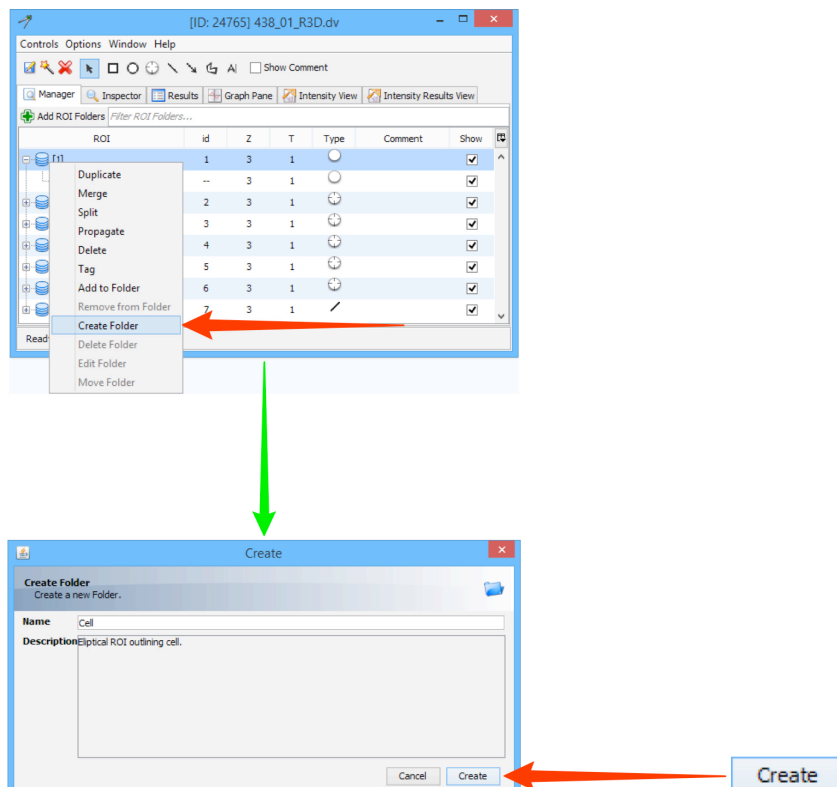
ROI Folders

ROI Folders provide a way of grouping ROIs on an image or across images for the purposes of organisation, analysis or visualisation. ROI Folders support hierarchies, so can contain both other folders and ROIs.

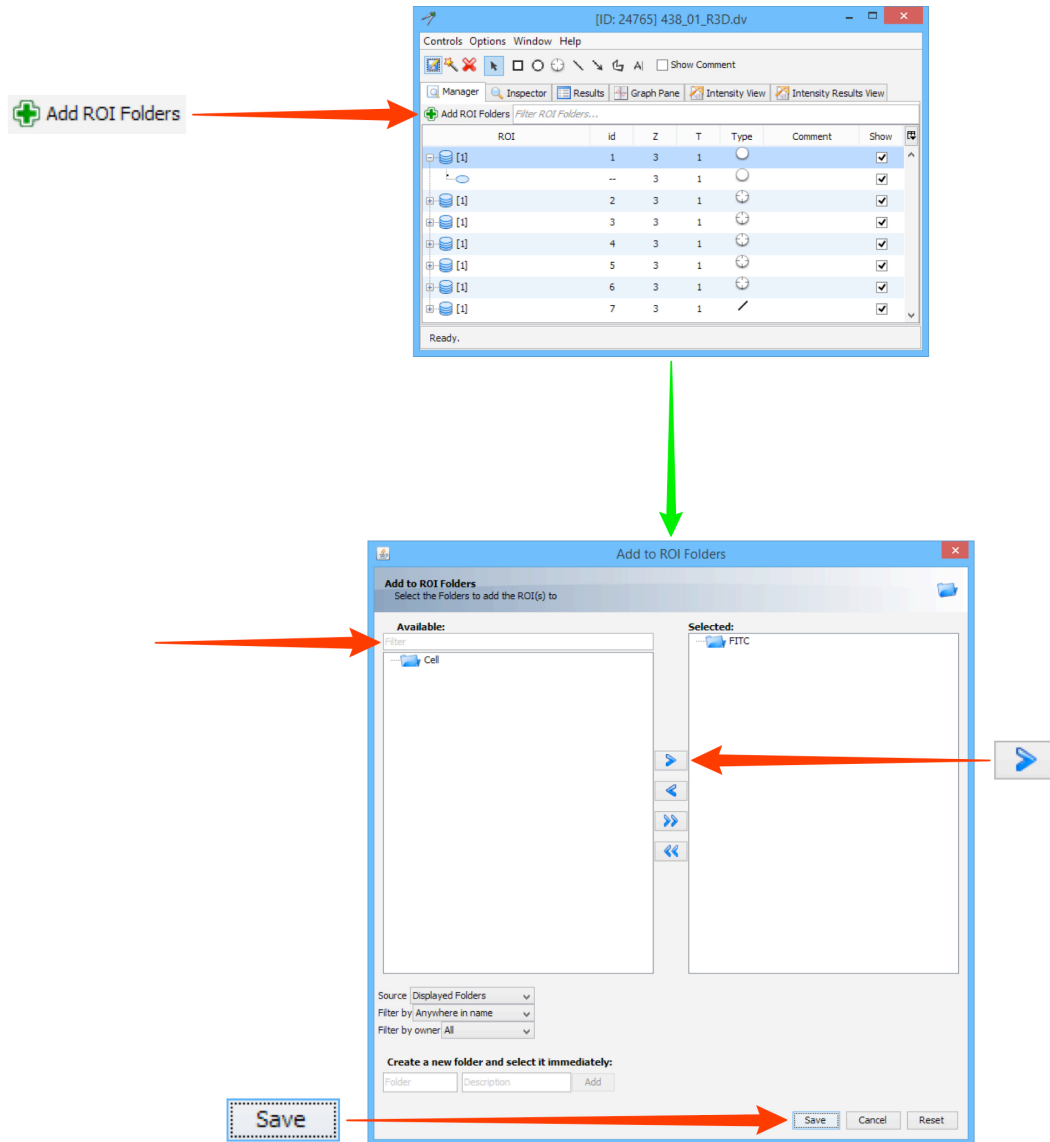
- 1 ROIs and folders are shown in the **Manager** tab of the Measurement Tool. By default only folders containing ROIs on the image being viewed are shown.



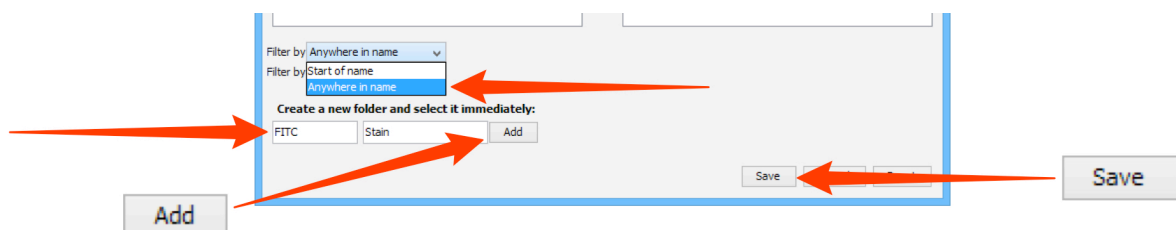
- 2 Select one or more ROIs to add to a folder. Right-click on the selection and choose **Create Folder** from the menu. Type the name and any description into the text boxes. Click **Create**.



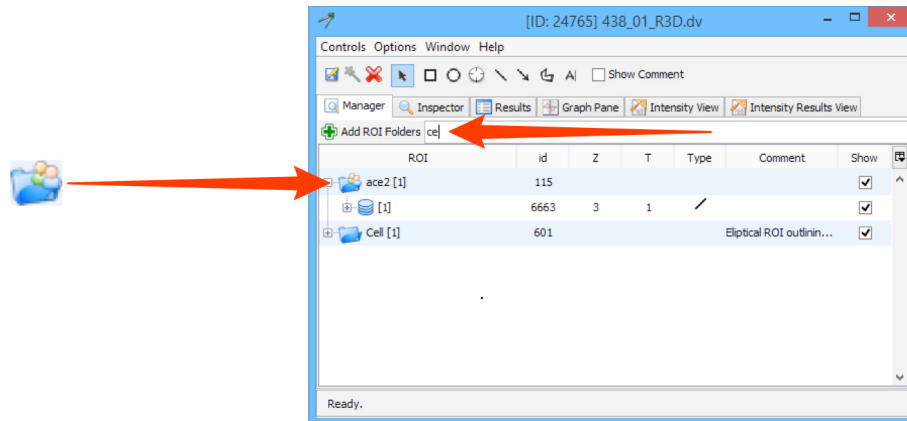
- 3** Click the **Add ROI Folders** button to add an existing folder to the Manager display. In the Add ROI Folders to Display dialog, select available folders in the left pane. Type in the text box above the Available pane to filter the ROI Folders shown. Click the **blue arrow** to add them to the Selected pane on the right. Click **Save** to add the folders to the Manager display.



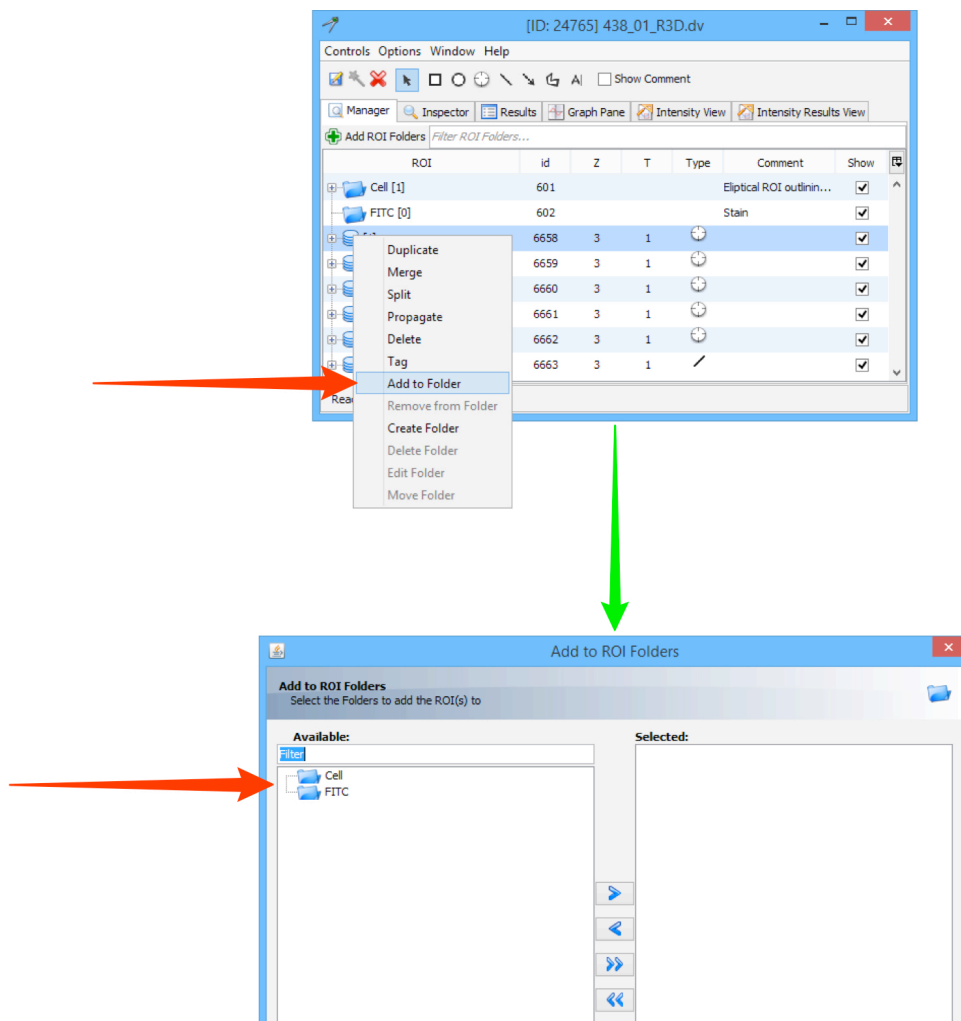
- 4** Use the drop-down menus below the pane, to refine the search parameters by selecting **Anywhere in the name** or **Start of the name**, **All**, **Owned by me**, or **Owned by others** from the menus. Create a new folder by typing the name in the text box at the bottom of the window, add any description, and clicking **Add**. Click **Save** to add the folders to the Manager display.



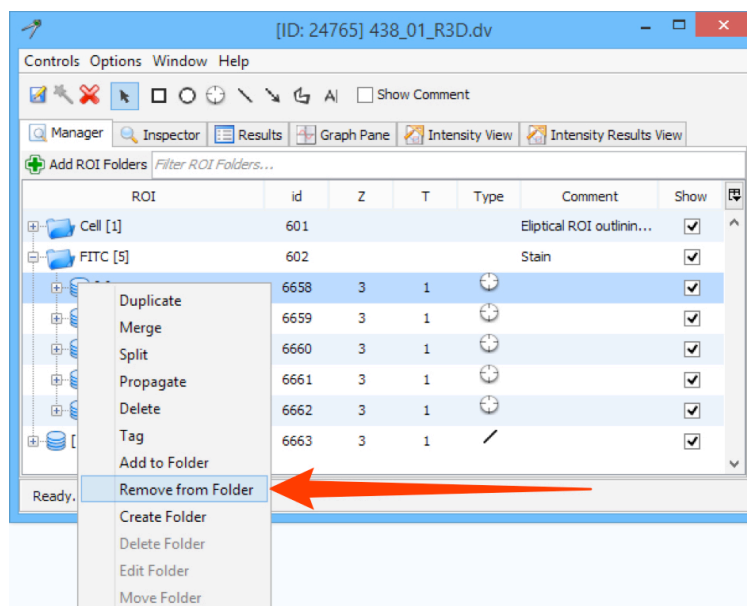
- 5 In the **Manager** tab of the Measurement Tool, use the **Filter ROI Folders** text box to filter the folders displayed. Folders owned by other users have the shared flag on them.



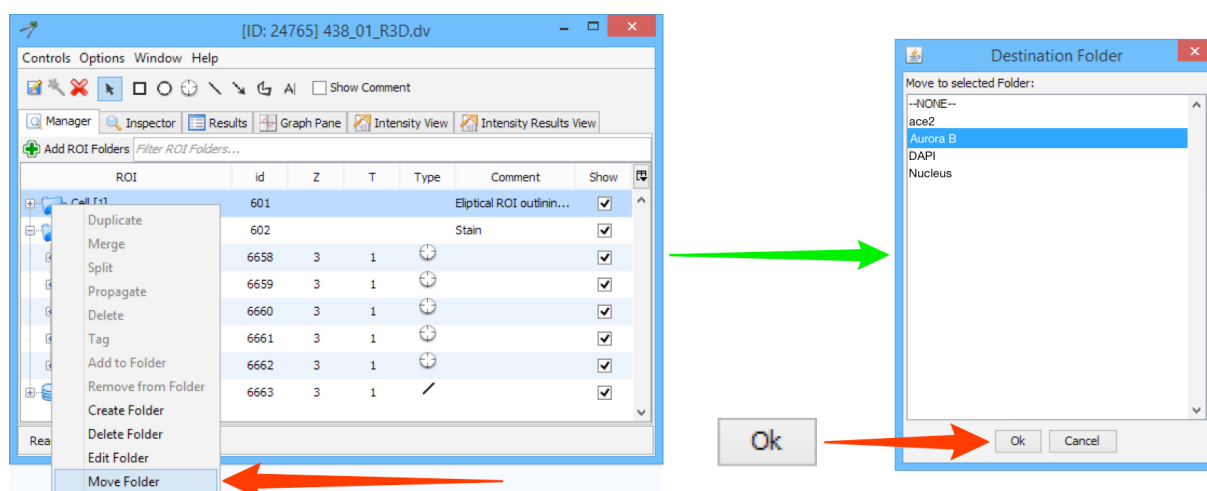
- 6 Right-click on a ROI and select **Add to Folder** to add the ROI to an existing or new folder. On opening, the **Add to Folders** dialog will only show folders currently displayed in the Management Tab. Use the **Filter ROI Folders** text box to filter by folder name. Click on the **Source** drop-down and select the **All Folders** option to display all the folders in the system.



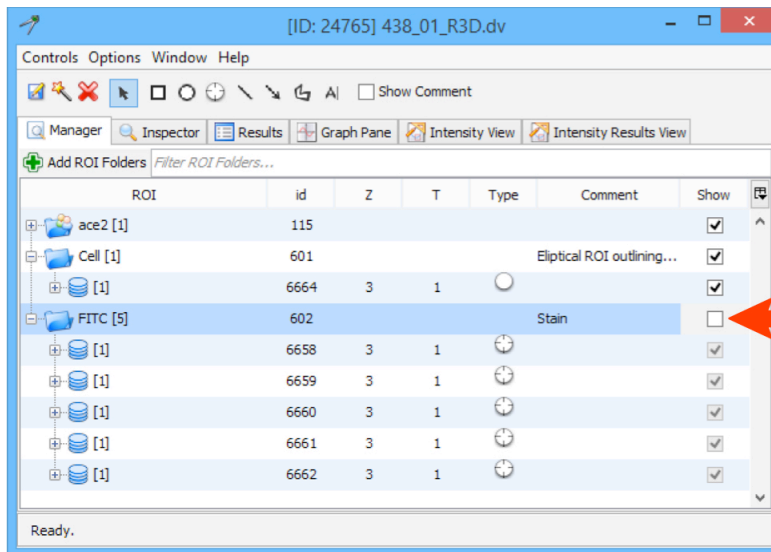
- 7 Drag and drop a ROI onto a folder to add it, or out of a folder to remove it. Similarly folders can be dragged and dropped in and out of other folders. When dragging a ROI out of a folder to “orphan” it, it needs to be dragged clear of the last ROI at the bottom of the list.
- 8 Right-click on a ROI within a folder and select **Remove from Folder** to “orphan” a ROI.



- 9 Right-click on a folder within another folder and choose **Move Folder** from the menu to move a folder and its contents to another folder or to “orphan” it. Select a folder or **None** in the Destination Folder window. Click **OK**.



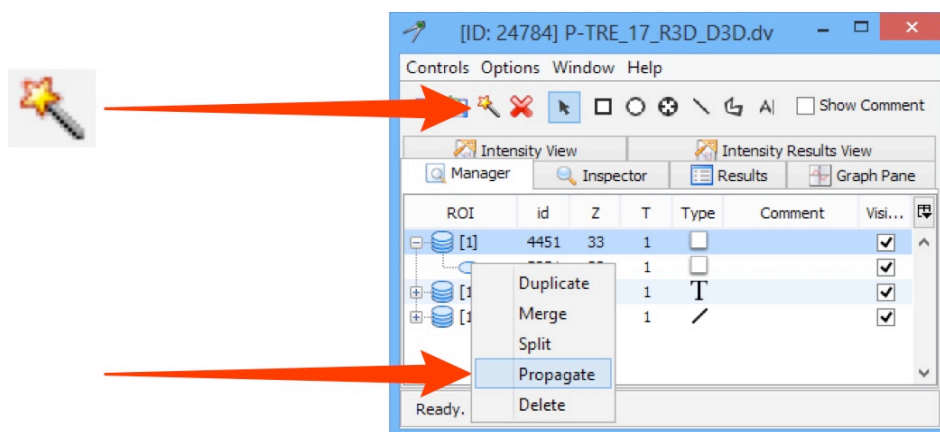
- 10** Deselect or select the **Show** checkbox to hide or show folders or ROIs. A change in the state of the Show checkbox for a folder is also applied to the contents of the folder.



Note: All these operations can be performed on multi-selected objects.

Propagating ROIs

- 1** Any ROI drawn will be on the currently viewed Z section and time point. Click on the **ROI Assistant** icon to propagate an ROI across Z sections or time points.



2

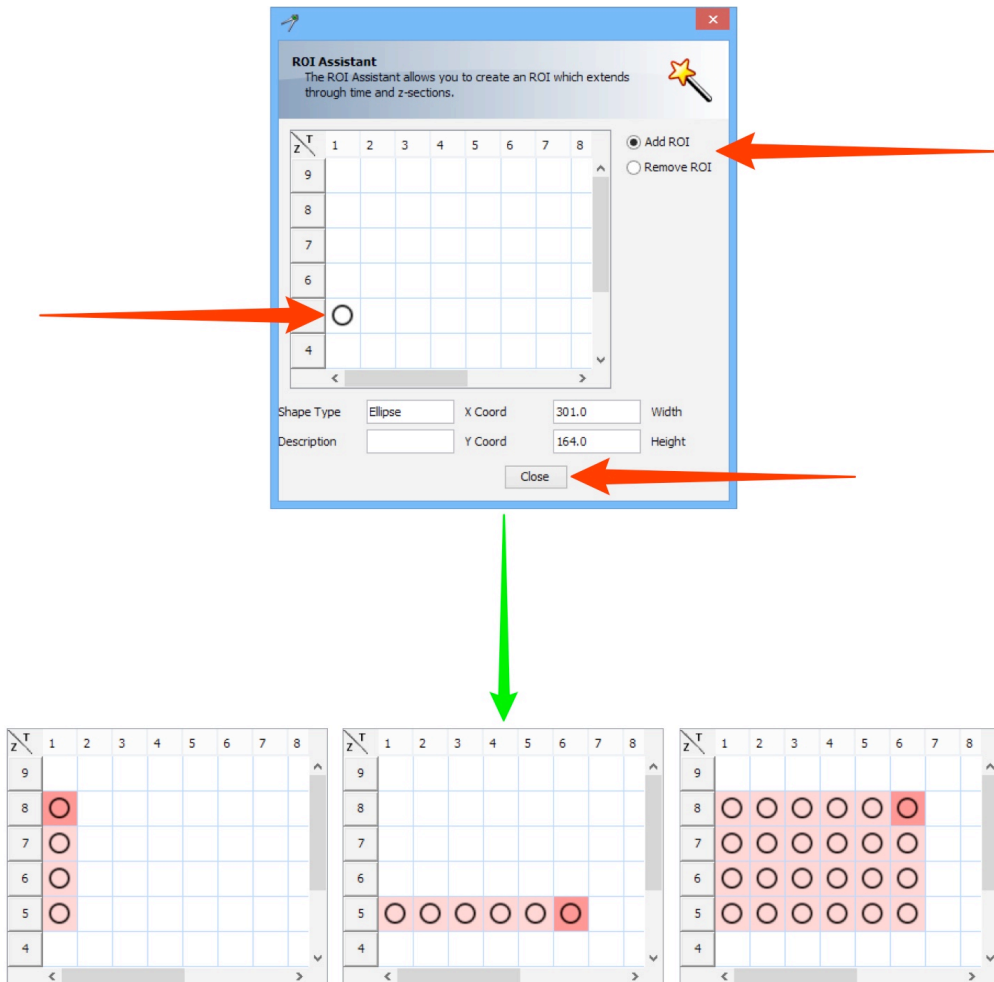
In the **ROI Assistant** window, click on the ROI and drag it.

Up or down propagates through the Z sections.

Drag left or right to propagate through the time points (T).

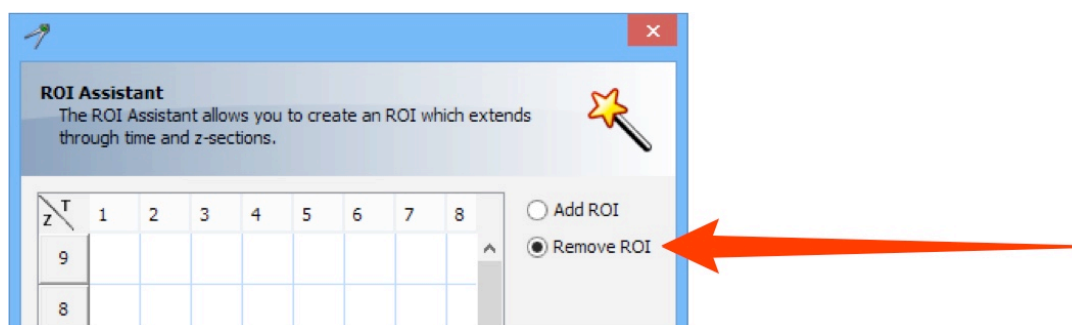
Dragging diagonally will propagate the ROI through both Z sections and time points.

Click **Close** to return to the ROI palette.



3

Click the **Remove ROI** radio button and click and drag on ROIs to remove them from Z sections or time points.



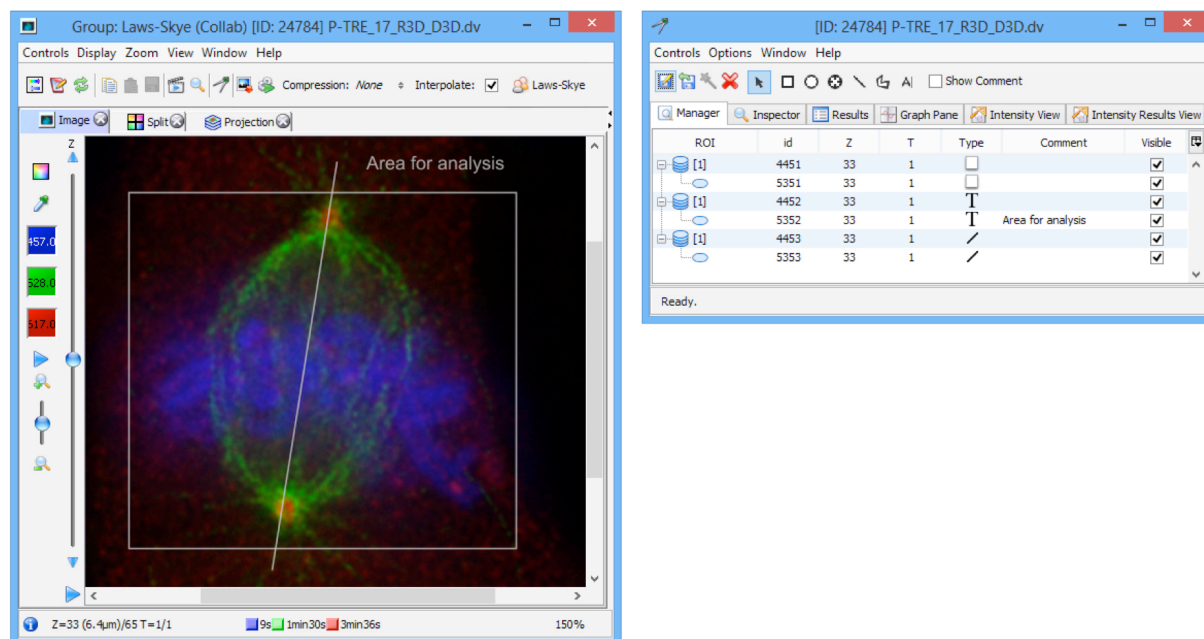
Managing and analysis

1

Details of **ROIs** are viewed in the **Manager** tab of the tool palette.

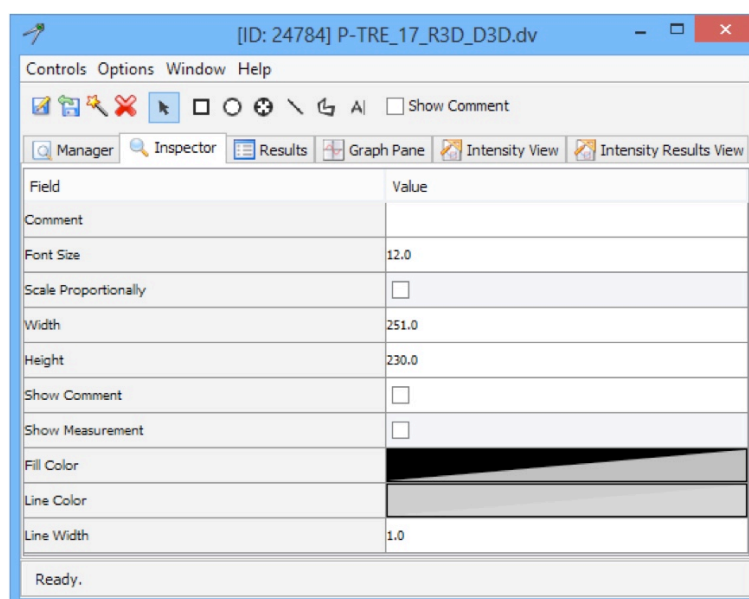
The columns show:

- ID of the ROI
- Z sections and time points the ROI is drawn on
- ROI type
- text associated with the ROI
- check box to toggle the visibility of the ROI.

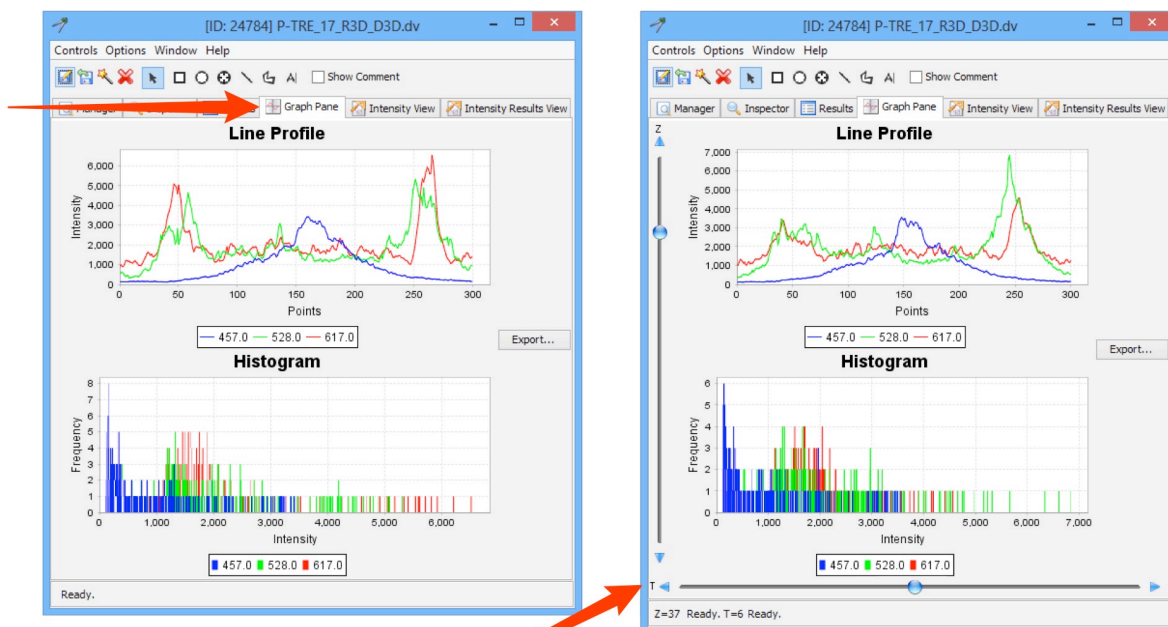


2

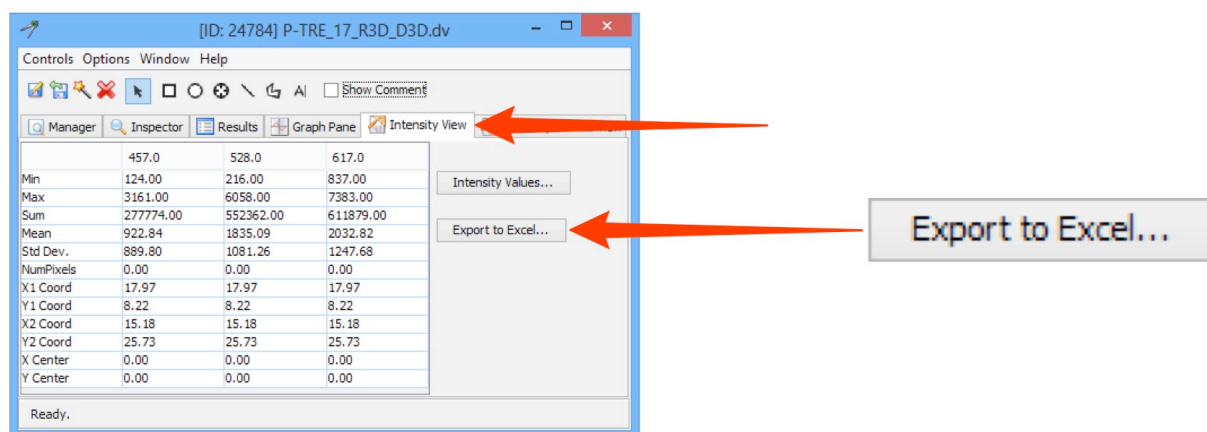
Select an ROI and click on the **Inspector** tab to view and edit the text label, dimensions, fill and line colour, and toggle the visibility of the text and measurements on and off.



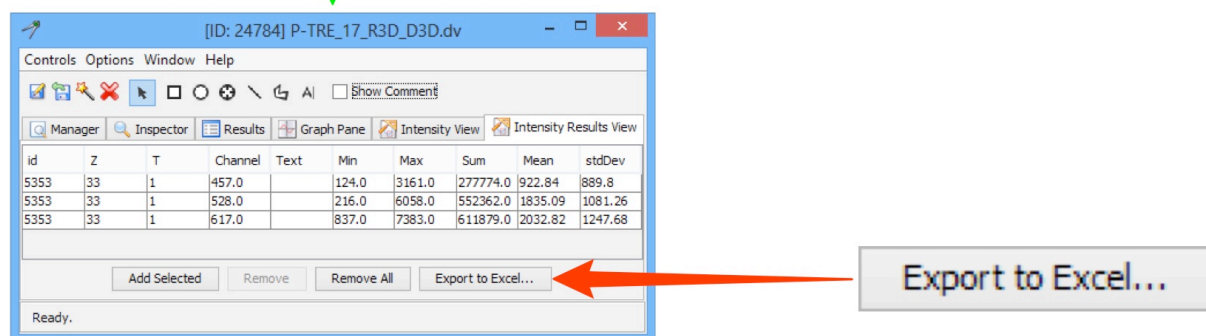
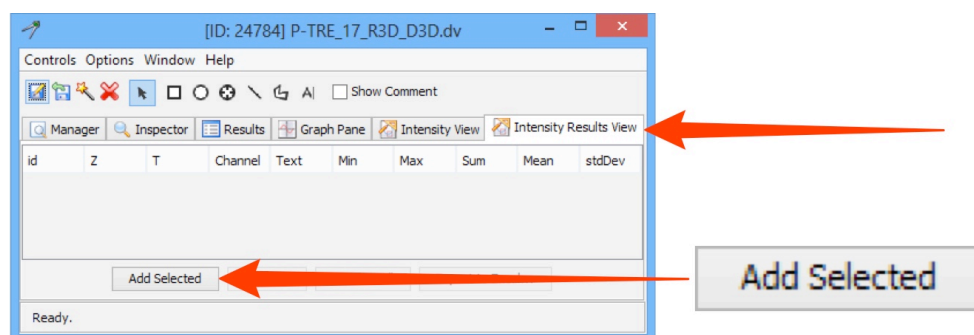
- 3 Click on the **Graph Pane** tab to view a plot of the channel intensity values for an ROI. A line ROI has a profile of intensity values along the line and a histogram of intensity vs. frequency (other types of ROI show only the histogram). If an ROI is propagated across Z sections or time points, the sliders to the left of the histogram allow stepping through the sequences.



- 4 Click on the **Intensity View** tab to see tables of intensity values for the ROI. These values can be exported in Excel format for further analysis.



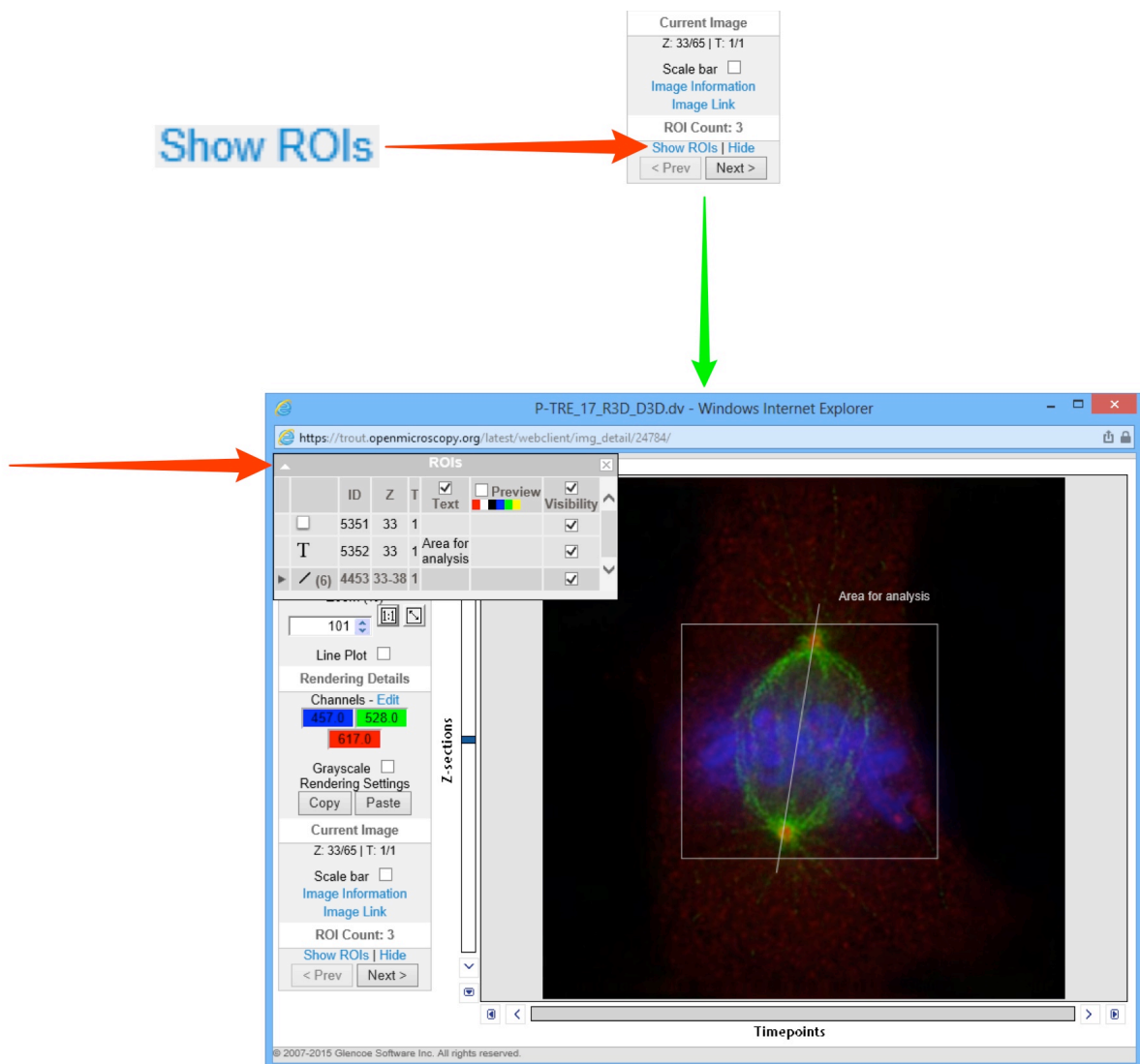
- 5 Click on the **Intensity Results View** tab to see tables of intensity values for the ROI. Click **Add Selected** to show results for the selected ROI. These values can be exported in Excel format for further analysis.



Viewing ROIs in OMERO.web

ROIs cannot be drawn in OMERO.web, but can be viewed, along with basic line plot of channel intensity values.

- 1 Open the image in the **Full Viewer**.
Click the **Show ROIs** link at the bottom of the toolbar.
The ROIs palette opens in the top left corner.
Toggle visibility on and off using the checkboxes.



The screenshot shows the OMERO.web interface. At the top right, a toolbar contains the following elements: 'Current Image', 'Z: 33/65 | T: 1/1', 'Scale bar' (checkbox), 'Image Information', 'Image Link', 'ROI Count: 3', 'Show ROIs | Hide', and '< Prev' / 'Next >' buttons. A red arrow points from the 'Show ROIs' link in the toolbar to the 'Show ROIs' link in the main toolbar at the bottom of the page.

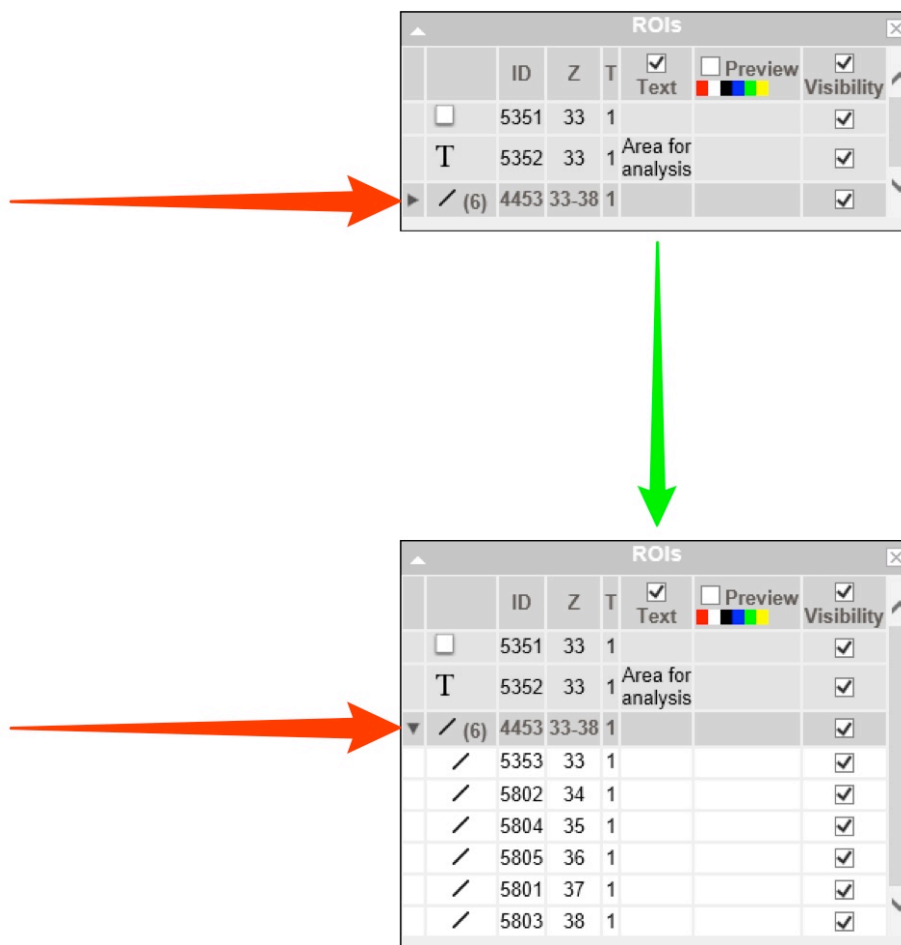
The main toolbar at the bottom of the page includes: 'Line Plot' (checkbox), 'Rendering Details', 'Channels - Edit', '457.0', '528.0', '617.0', 'Grayscale' (checkbox), 'Rendering Settings', 'Copy', 'Paste', 'Current Image', 'Z: 33/65 | T: 1/1', 'Scale bar' (checkbox), 'Image Information', 'Image Link', 'ROI Count: 3', 'Show ROIs | Hide', and '< Prev' / 'Next >' buttons.

The 'ROIs' palette is open in the top left corner, displaying a table of ROIs:

ID	Z	T	Text	Preview	Visibility
5351	33	1		<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>
5352	33	1	Area for analysis	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>
(6) 4453	33-38	1		<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>

The main image viewer displays a fluorescence image with a green rectangular region of interest (ROI) labeled 'Area for analysis'. The image is rendered in a 3D-like view with a 'Z-sections' slider on the left and a 'Timepoints' slider at the bottom.

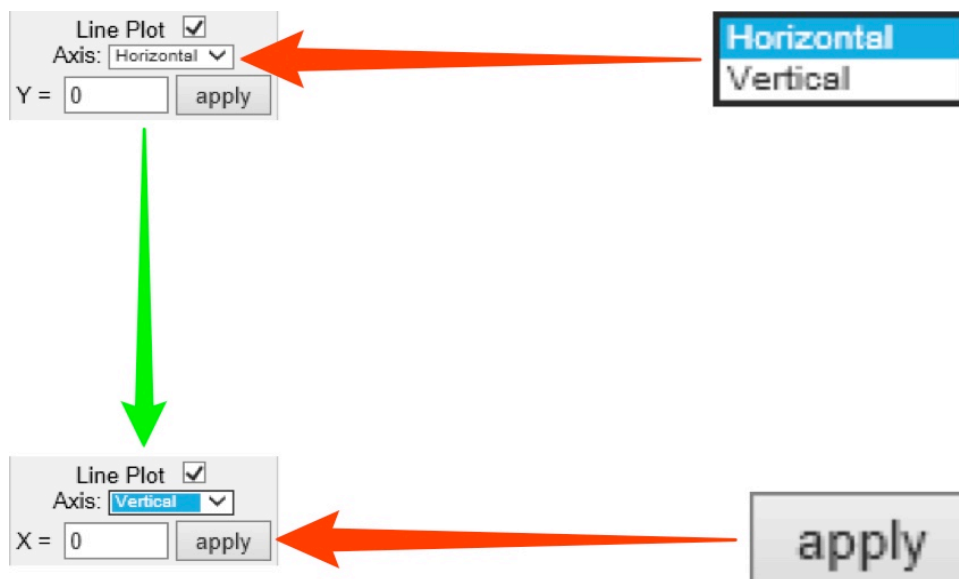
- 2 Click on the **Expansion Arrow** on propagated ROIs to show the shapes on Z sections or time points.



- 3 Click the **Line Plot** check box to show a line plot on the image.



- 4 Select orientation from the **Axis** drop-down.
Click **Apply**.



- 5 Enter the desired **X** or **Y** coordinate in the text box and click **Apply**, or click on a point in the image to set the line plot position.

