



MetaXpress[®] 6 Software Guide

Using Modify Objects Tools in CME

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Chapter Purpose

The purpose of this chapter is to describe the **Modify Objects** tools available in the **Custom Module Editor (CME)** plugin.

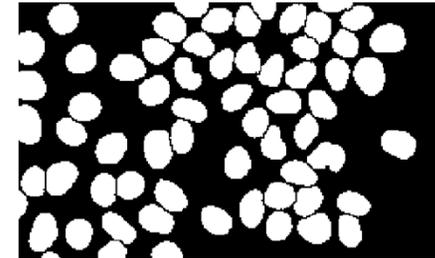
Modify Objects tools are used to modify an existing segmentation (1-Bit binary) mask. The original segmentation mask is generated from a **Find Objects** or **Application Modules Objects** step. The final segmentation mask can be used to make measurements.



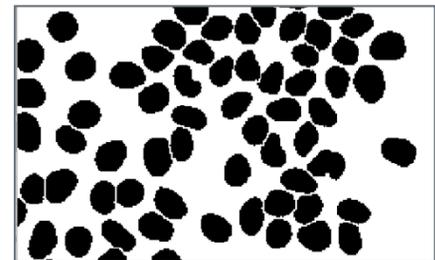
Modify Objects Tools: Overview



1^o Segmentation Mask



2^o Segmentation Mask



- **Modify Objects** tools can be used to modify existing segmentation masks generated from a **Find Objects** or **Application Module Objects** step
- Source image must be a segmentation mask (1-Bit binary)
- Result image is a segmentation mask that can be used to make measurements with or can be modified further with additional **Modify Objects** tools



Ribbon: Modify Objects Tools

The screenshot displays the software's ribbon interface with the 'Modify Objects' tab selected. A red box highlights the 'Modify Objects' group on the ribbon, with an arrow pointing to a drop-down menu. Another red box highlights the 'Invert Objects' icon, with an arrow pointing to a preview window showing the inverted image. A third red box highlights the 'Invert Objects' tool panel on the left, with an arrow pointing to the 'Apply' button. The tool panel includes a 'Source' dropdown set to 'Auto Find Blobs', a 'Result' dropdown set to 'Invert Objects', and a description: 'Inverts the image histogram so dark pixels become bright and bright pixels become dark.' Below the tool panel is a 'Custom Module Step Images' panel showing a sequence of four images: 1. Original image, 2. Binary image, 3. Inverted binary image, and 4. Final processed image.

Click here for a drop-down menu of **Modify Objects** tools

Click on the appropriate icon

Clicking on a **Modify Objects** tool icon will add a step card on the panel to the left



Modify Object Tools Basic Descriptions



Fill Holes

- Fills holes in objects, which allows the filled area to be included in measurement data



Remove Border Objects

- Excludes all objects that touch the edge of the mask image



Keep Marked Objects

- Compares the objects in two masks. If any part of an object overlaps in both masks, the object from the source mask is kept



Grow Objects Without Touching

- Expands objects by the number of pixels that you specify. Does not allow objects to touch.



Logical Operations

- Applies a Boolean operator (AND, OR, ANDNOT, XOR) between two masks to determine which pixels will be displayed in a new result mask



Watershed

- Creates boundaries between objects based on intensity peaks and valleys in the image source



Remove Marked Objects

- Compares the objects in two mask. If any part of an object overlaps in both masks, the object from the source image is removed



Filter Mask

- Remove objects from the mask based on measurement values of the objects



Invert Objects

- Inverses the mask histogram so dark pixels become bright and bright pixels become dark



Grow Objects

- Expands objects by the number of pixels that is user-specified. Allows objects to touch.

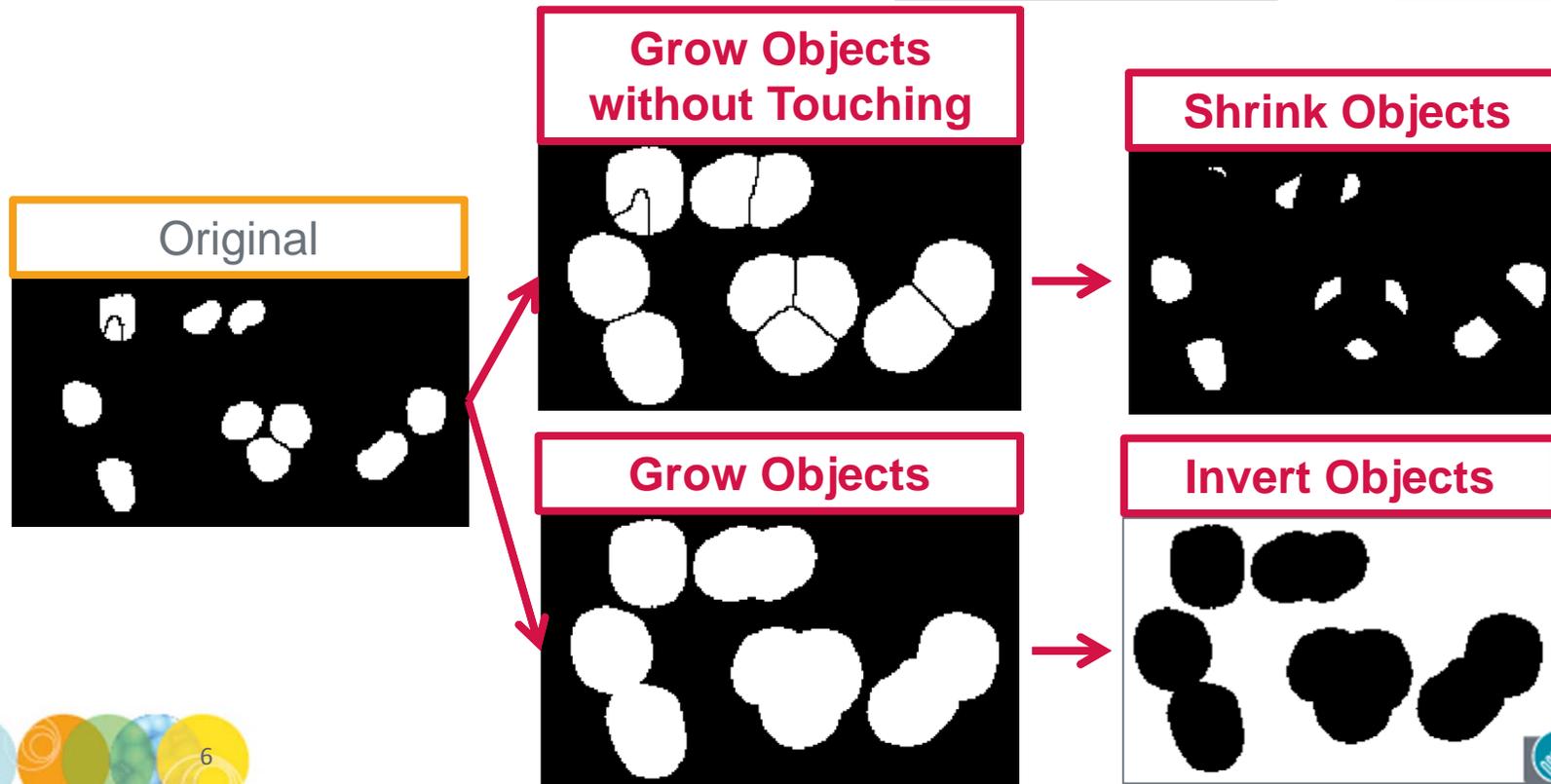
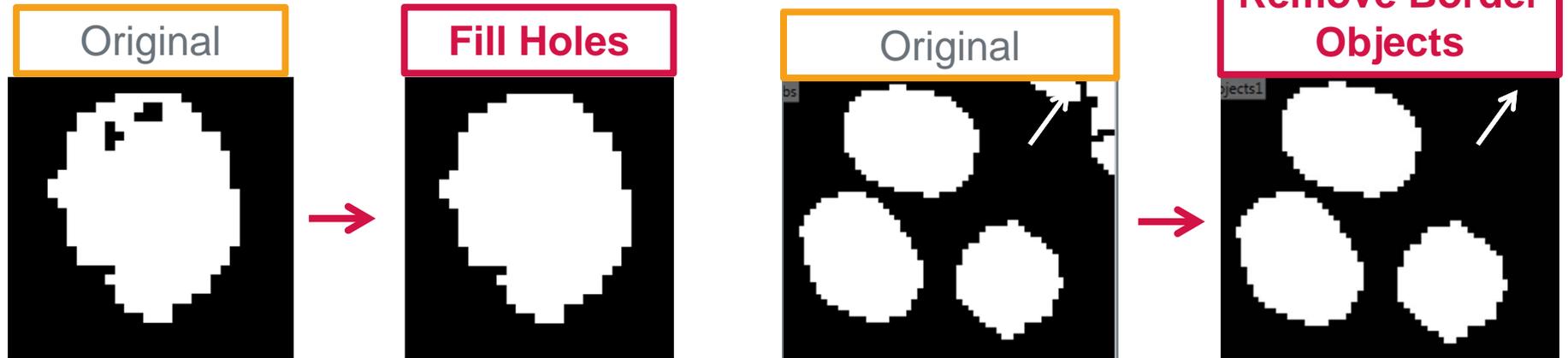


Shrink Objects

- Shrinks objects by the number of pixels that is user-specified

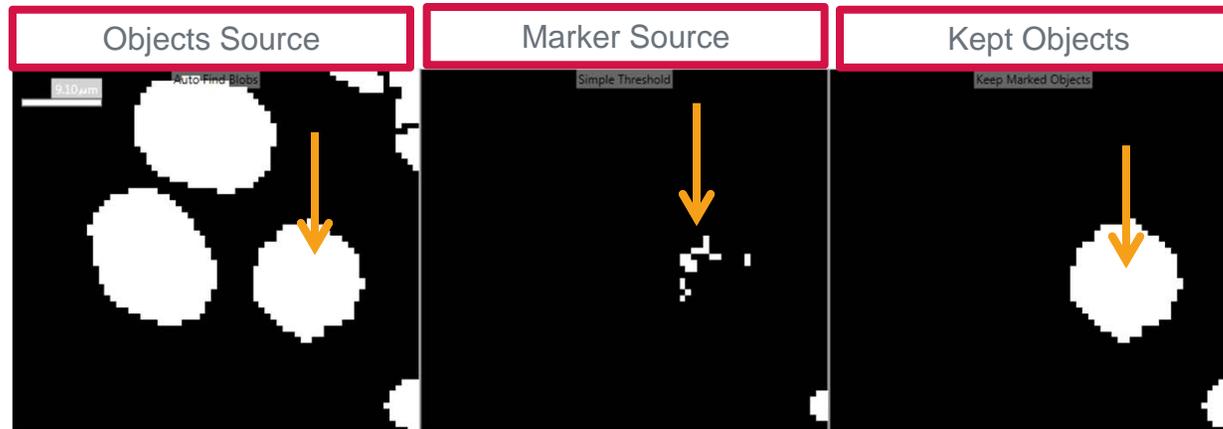


Modify Objects Tools Examples

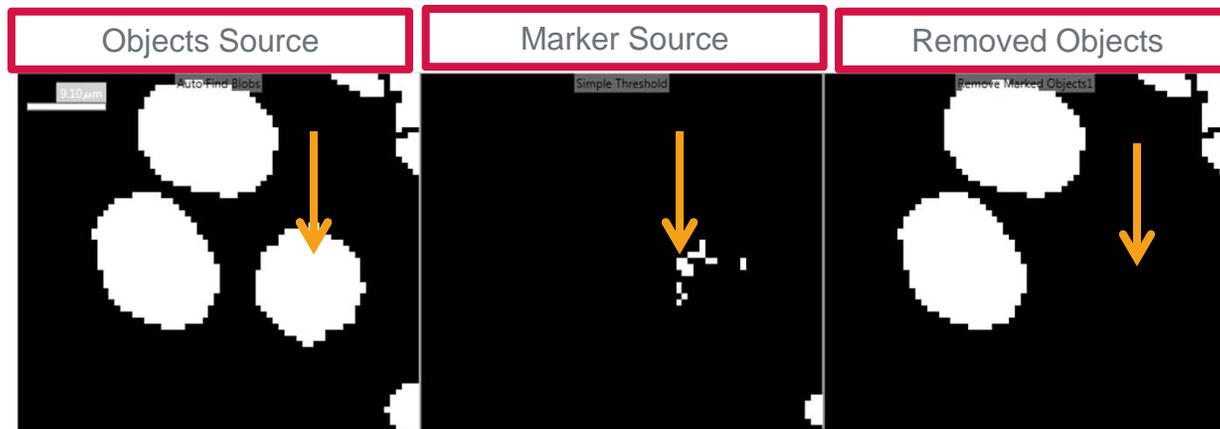


Modify Objects Tools Examples

Keep Marked Objects: objects in the **Objects Source** image that touch or overlap in the **Marker Source** image are kept

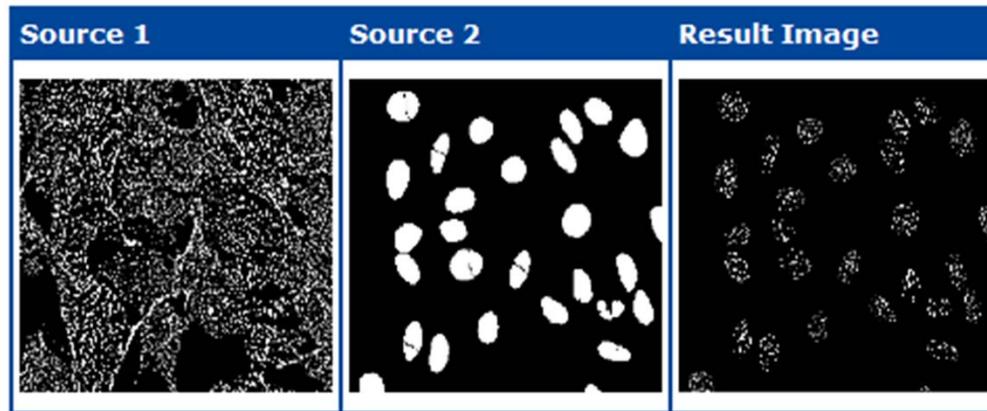


Remove Marked Objects: objects in the **Object Source** image that touch or overlap in the **Marker Source** image are removed



Modify Objects: Logical Operations (AND & OR)

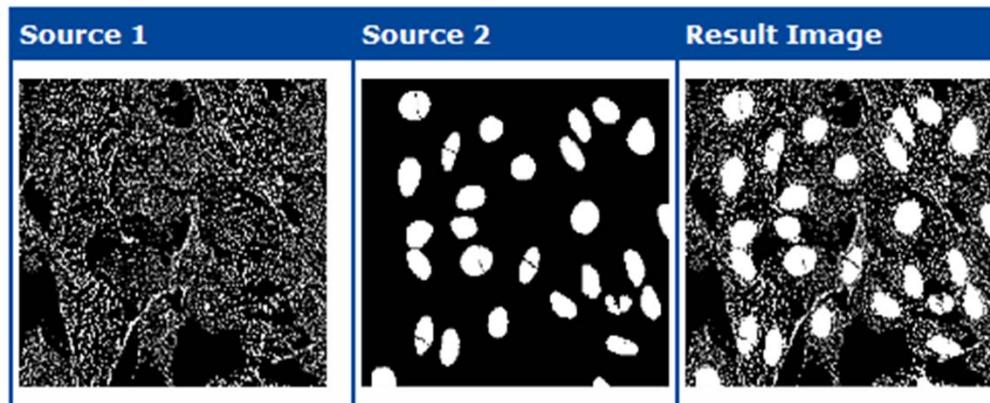
The **AND** operator compares two source images and creates a result image that contains only those pixels that appear in the same location in both of the source images



Pixel by pixel
definition

Source 1	Source 2	Result Image
Black (0)	Black (0)	Black (0)
Black (0)	White (1)	Black (0)
White (1)	Black (0)	Black (0)
White (1)	White (1)	White (1)

The **OR** operator compares two source images and creates a result image that contains only those pixels that appear in either or both of the source images



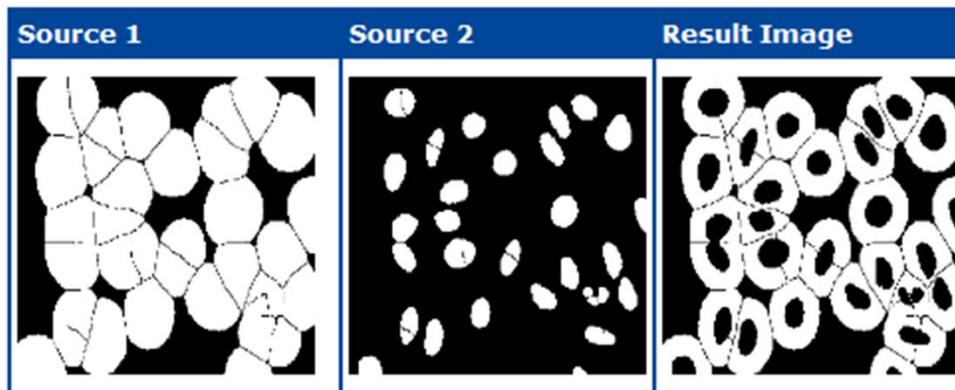
Pixel by pixel
definition

Source 1	Source 2	Result Image
Black (0)	Black (0)	Black (0)
Black (0)	White (1)	White (1)
White (1)	Black (0)	White (1)
White (1)	White (1)	White (1)



Modify Objects: Logical Operations (ANDNOT & XOR)

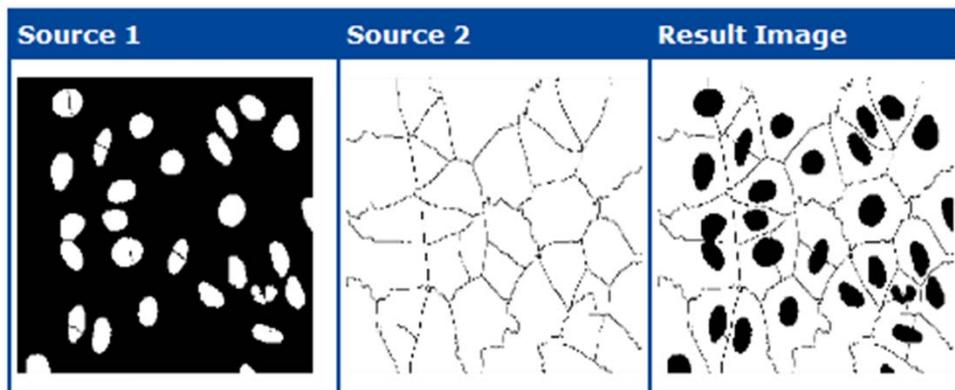
The **ANDNOT** operator compares two source images and creates a result image that contains pixels that appear in the first source image and not in the second source image. Essentially, you subtract the second image from the first:



Pixel by pixel
definition

Source 1	Source 2	Result Image
Black (0)	Black (0)	White (0)
Black (0)	White (1)	White (0)
White (1)	Black (0)	White (1)
White (1)	White (1)	Black (0)

The **XOR** operator compares two source images and creates a result image that contains pixels that are in either source image but that are not in both source images.



Pixel by pixel
definition

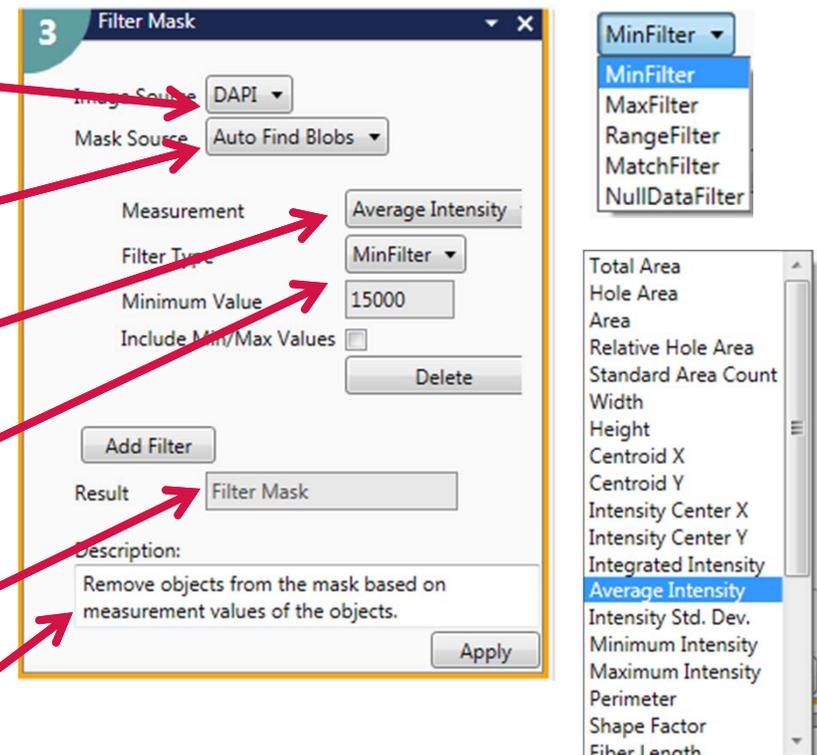
Source 1	Source 2	Result Image
Black (0)	Black (0)	Black (0)
Black (0)	White (1)	White (1)
White (1)	Black (0)	White (1)
White (1)	White (1)	Black (0)



Modify Objects: Filter Mask

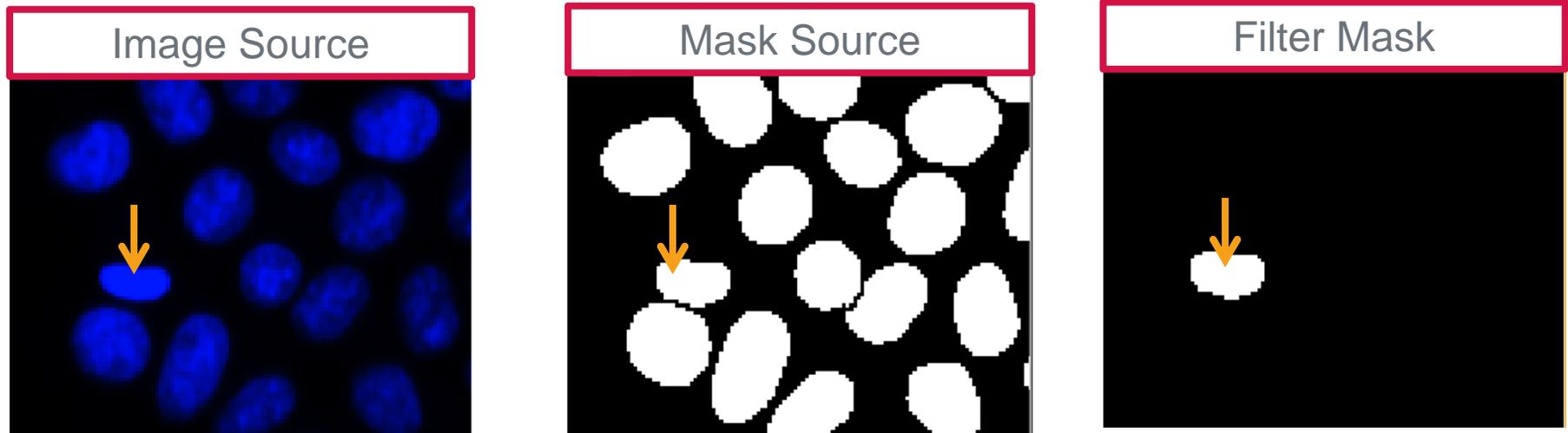
The **Filter Mask** tool can be used to filter the objects based on the measurements available from the drop down menu. This step requires a gray scale image (**Image Source**) and a binary mask (**Mask Source**). Additional filtering steps can be added using the **Add Filter** button. Filtering will be done in a hierarchal manner.

- Select image to measure
- Select previously created binary mask to make measurements with
- Choose measurement
- Choose filter type and enter appropriate values
- Enter a name for the new result
- If desired, enter a note. Default text is a description of the text.



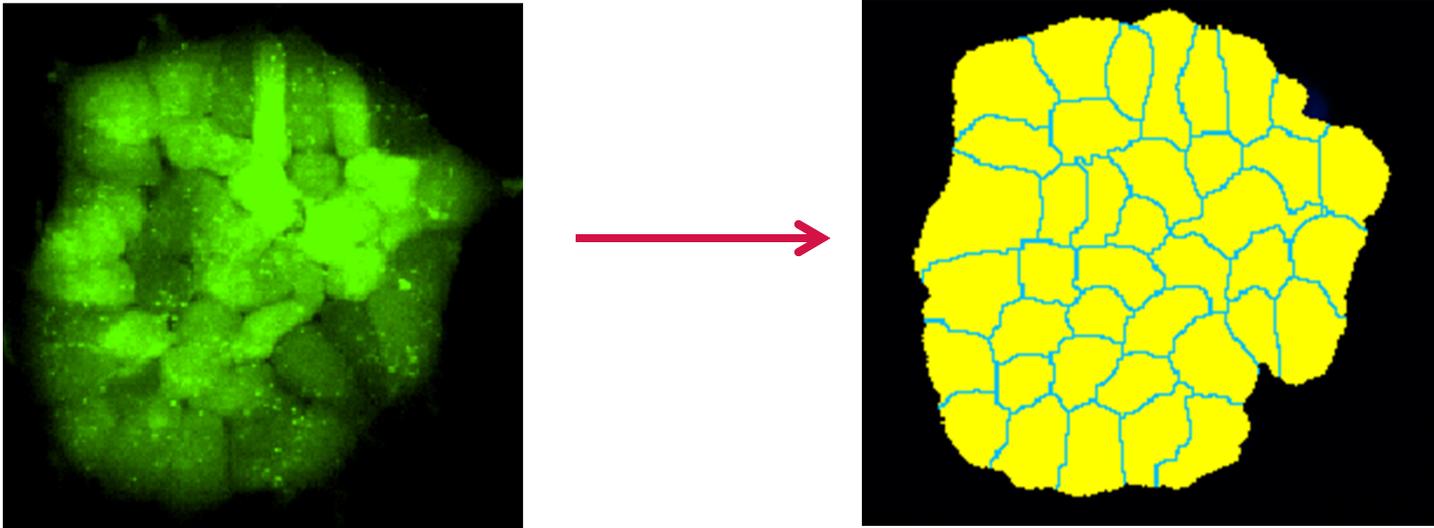
Filter Mask Example

In the example below, the MinFilter is used to filter out objects whose integrated intensity is less than 15000 in the grey scale (DAPI) image



Modify Objects: Watershed

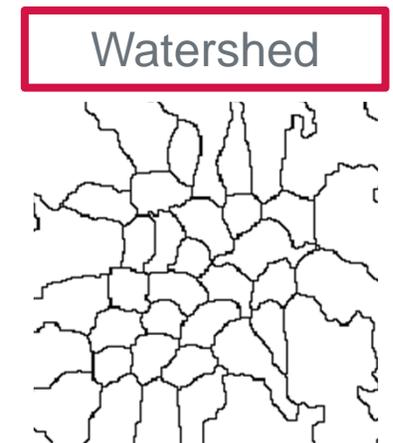
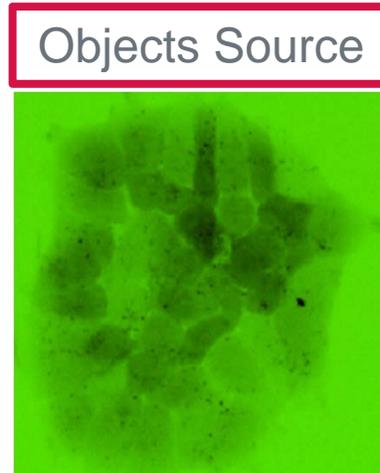
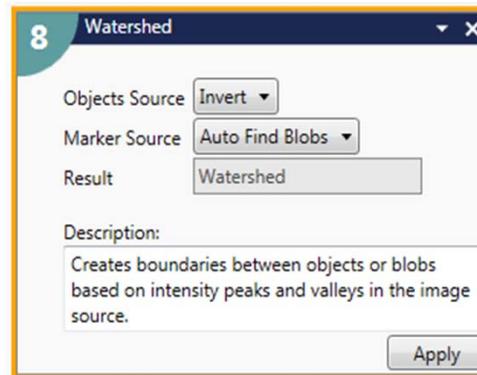
The **Watershed** tool can be used to create cell boundaries in an image set that does not contain a marker for the plasma membrane.



Modify Objects: Watershed

Two images are required for watershed tool:

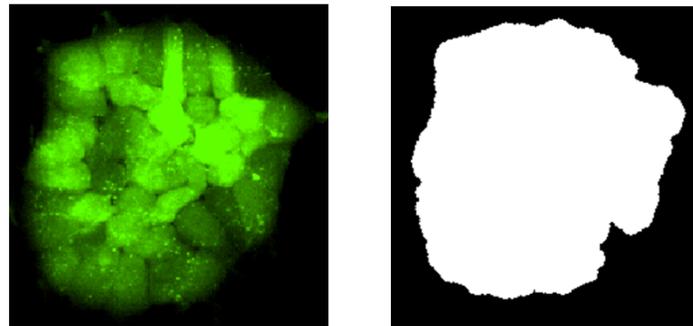
- **Objects Source:** Grayscale image that represents the object boundaries as an intensity peak (invert the image using **Invert** under the **Modify Image** tools)
- **Marker Source:** A segmentation mask that identifies each object (i.e. nuclear mask)



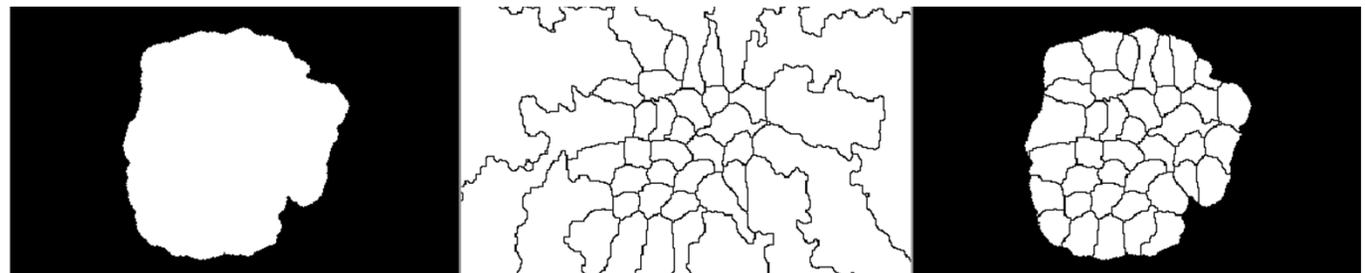
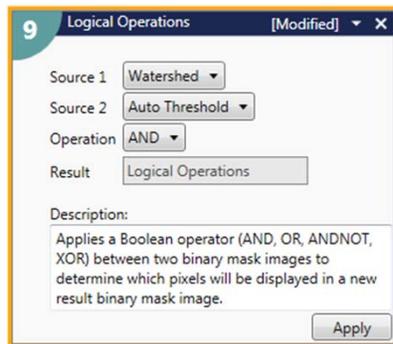
Modify Objects: Watershed

After the **Watershed** result image has been generated, the next step is to apply this to a mask of the original image

- Use one of the **Find Objects** or **Application Module Objects** tools to create a mask of the original image



- Use **Logical Operations- AND** step with the mask of the original image (see above) and **Watershed** (see previous section) mask



Support Resources

- F1 / HELP within MetaXpress® Software
- Support and Knowledge Base: <http://mdc.custhelp.com/>
- User Forum: <http://metamorph.moleculardevices.com/forum/>
- Request Support: <http://mdc.custhelp.com/app/ask>
- Technical Support can also be reached by telephone:
 - 1 (800) 635-5577
 - Select options for Tech Support → Cellular Imaging Products → ImageXpress Instruments





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