

Zeiss Efficient Navigation Standard Operation Protocol

Basic Application

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Install appropriate ZEN version in Windows
ZEN 2009/2010 for Windows XP/Vista
ZEN 2011 (Black version) for Windows 7 (64 bits)

Launch the **ZEN** software by double clicking the shortcut icon.



Load image in LSM format by clicking **File > Open**

Analyze the Image Data in different View

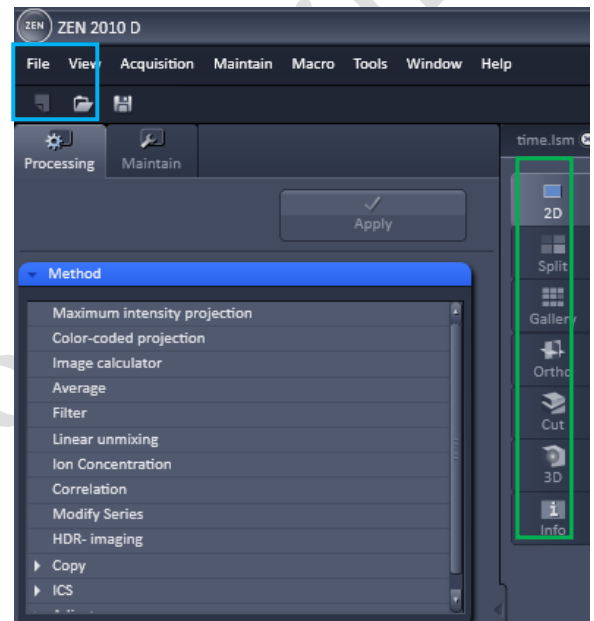
2D display single frame channel-superimposed image

Split display the individual channels of a multi channel image as well as the superimposed image

Gallery display images (Z-Stack, time series, combination of both) side by side in a tiled fashion

3D show the Z-Stack images displayed in three dimensions with various rendering

Info show summary information of all relevant image acquisition parameters

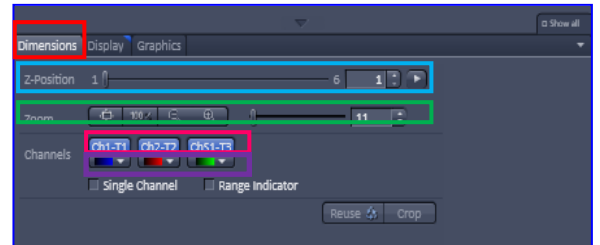


Display Control under the Image Windows

Dimensions

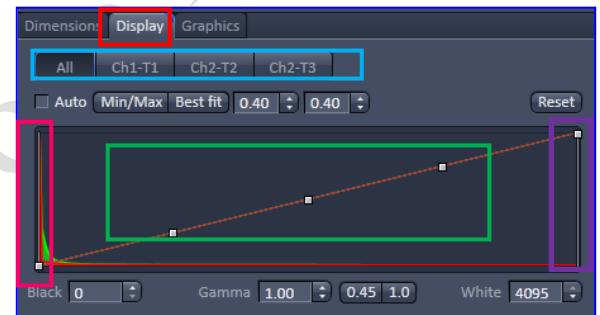
In **Dimensions**, **Slide bar** can be used to control the showing frame in Time or Z series while Image can be **Zoomed in or out** using various tools.

Acquired channels can be **Toggled ON / OFF** in the channels control boxes. **Pseudocolor** of each channel can also be changed.



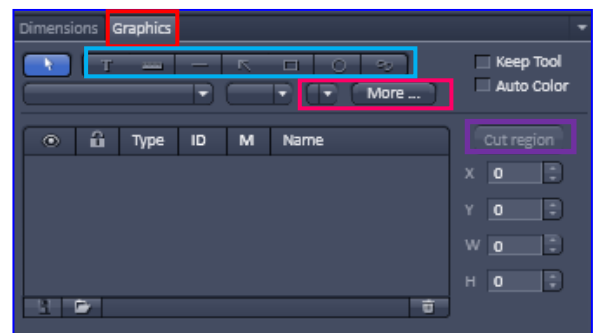
Display

In **Display** tap, brightness, contrast, upper and lower threshold for **each or all channels** display can be adjusted. While the **Left** and **Right** represent the **Lower** and **Upper** display threshold respectively, the **Curve** between controls how actual intensity (x-axis) of the image will be displayed (y-axis).



Overlay Graphics

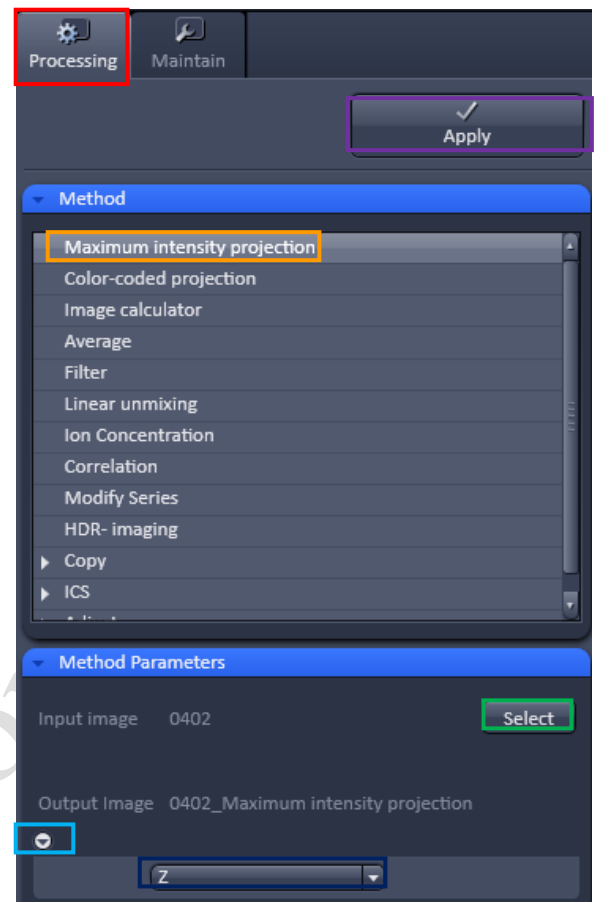
To add various overlay objects in the image, go to **Graphics** tap below the Image window (also click “show all” in the left top corner if necessary). You can add ① **Text box**; ② **scale bar**; ③ **measurement line or arrow**; ④ **Region**; on the image. **Color and Font** can be modified in the color pull-down box and “More...” button. Selected Region can be cropped out by clicking “**Cut region**” button.



Processing tap

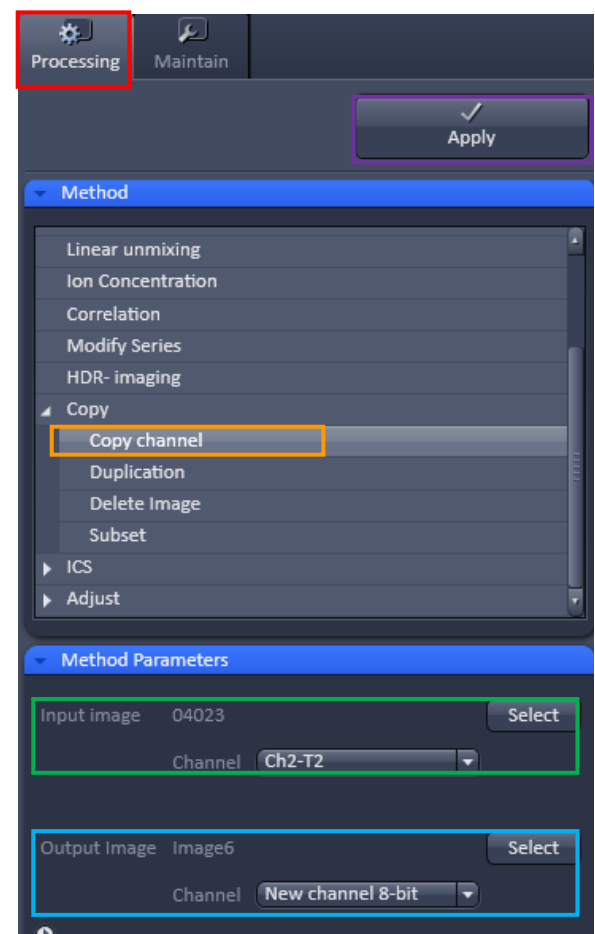
Maximum Intensity Projection

Maximum projection is useful for displaying three-dimensional structure in one two- dimensions image. A two-dimensional image with the data from the highest intensity pixel along the projection axis will be generated. Click **Maximum intensity projection** in the **Processing tab**, highlight the three-dimension image to be processed in the image window and click “**Select**”. Expand the Method Parameters tap by click the **arrow** in left bottom. Choose “**Z**” to collect maximum intensity data across different z- frame. Then click “**Apply**” on the top of the processing tap to generate the new two-dimension image.



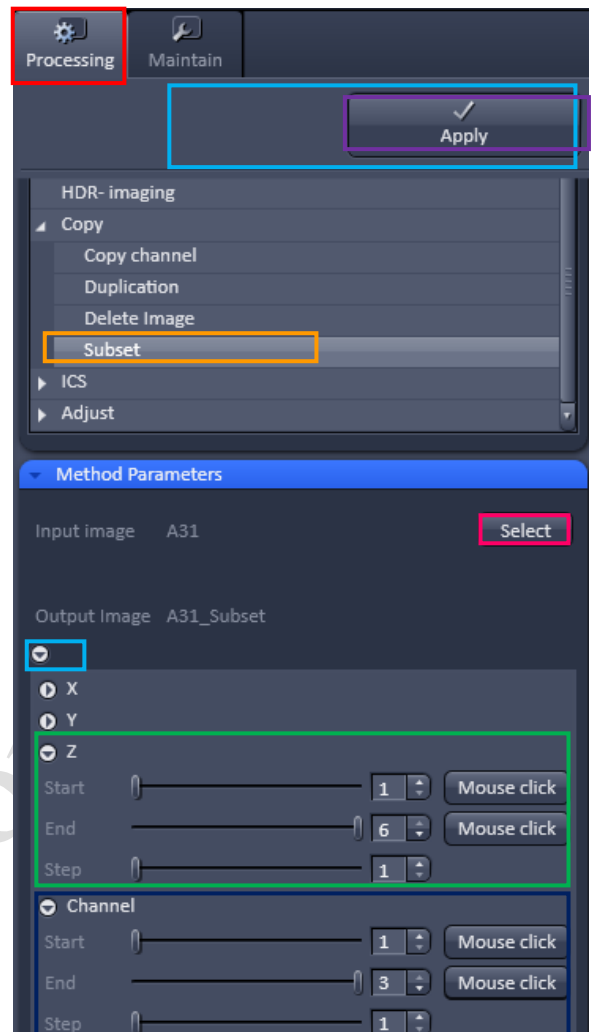
Copy Channel

To Copy a channel from an image or insert a copy channel into another image, click **Copy> Copy Channel** in the **Processing tab**, highlight the image to be copy in the image window and click “**Select**”. Select the **required channel or all** channels in the **Channel** pull-down box. In Output image, choose “**New Image 8/12/16-bit**” for creating a new LSM image file in different bit depth; choose “**New channel 8/12/16-bit**” for inserting the copy channel to the **Selected** image file “**Output Image**”; or replacing an **existing channel** in selected image file “**Output Image**” by choose the corresponding channel in the pull-down box. Then click “**Apply**” on the top of the processing tap.



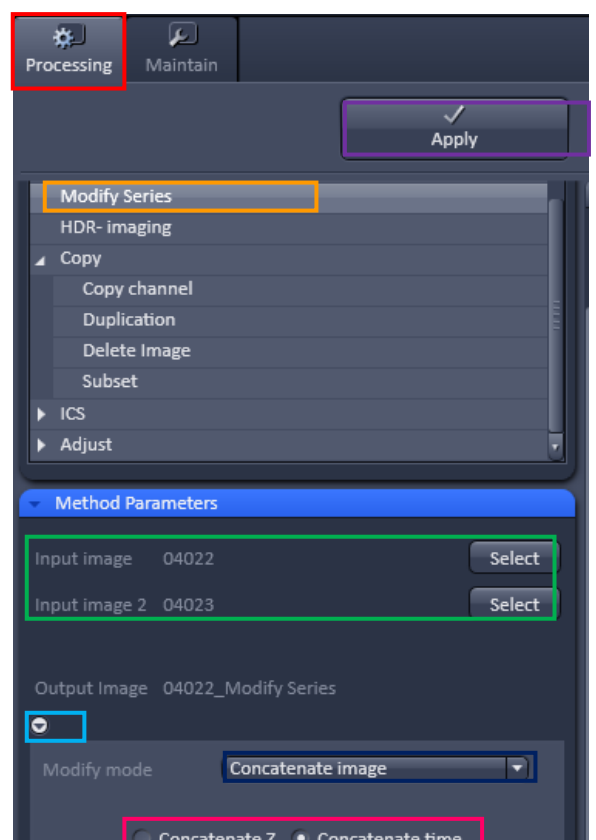
Copy Subset

To Copy Time / Z-position / Channel subset of images from an original images set, Click **Copy>Subset** in the **Processing tab**, highlight the image to be processed in the image window and click “**Select**”. Expand the Method Parameters tap by click the **arrow** in left bottom. Select the required subset by adjusting start and end frame position for **Time / Z-position / Channel** setting. Then click “**Apply**” on the top of the processing tap.



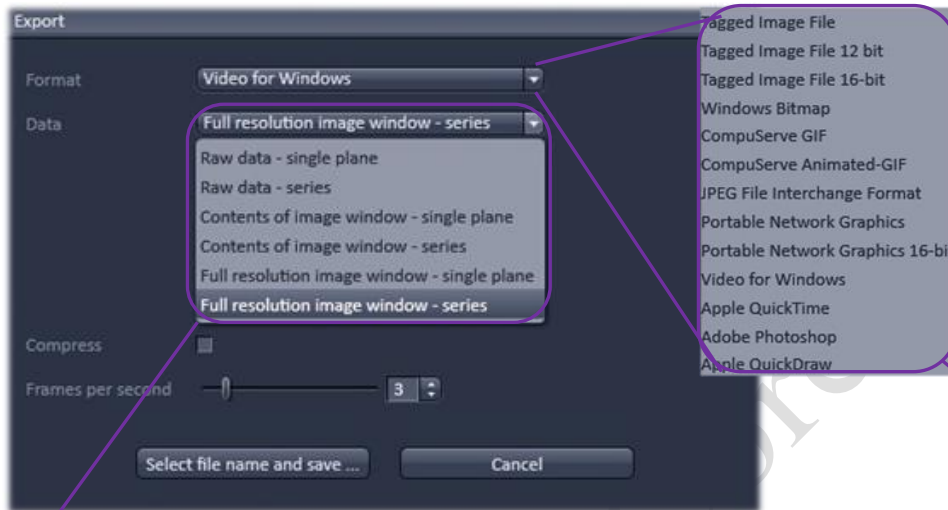
Merging Series

To merge 2 set of images in time-series or z-series, go to **Modify Series** in the **Processing Tab**, highlight the image to be in the top (in z-series) or first (in time series) part of the new series and click “**Select**” next to **Input Image** and then highlight the image to be in the bottom (in z-series) or second (in time series) part of the new series and click “**Select**” next to **Input Image 2**. Expand the Method Parameters tap by click the **arrow** in left bottom, choose “**Concatenate image**” in Modify mode and choose **Concatenate Z or Concatenate time**. Click **Apply** to generate the new series Image.



Exporting Images

Image can be export as various Image format and series images can be export as video. Go to File > Export, and choose the **Format and Data** as followed. For series images, adjust Frames per seconds if necessary. Click **Select file name and save** and save in your own folder.



Single Frame	Format	Tagged Image File JPEG File Interchange Format Portable Network Graphics
	Date	Raw data – single plane (without any overlay graphics) Contents of image window – single plane (with overlay; as displayed as in image container; compressed) Full resolution image window – single plane (with overlay; as displayed as in image container; uncompressed)
Series (Gallery of images; Time series movie or 3D animation)	Format	Video of Window Apple Quick Time
	Date	Raw data – series (without any overlay graphics) Contents of image window – series (with overlay; as displayed as in image container; compressed) Full resolution image window – series (with overlay; as displayed as in image container; uncompressed)