

First Steps in ZEN Blue

Skill Builder Series: Image Processing



Exporting Images and Movies in ZEN Blue

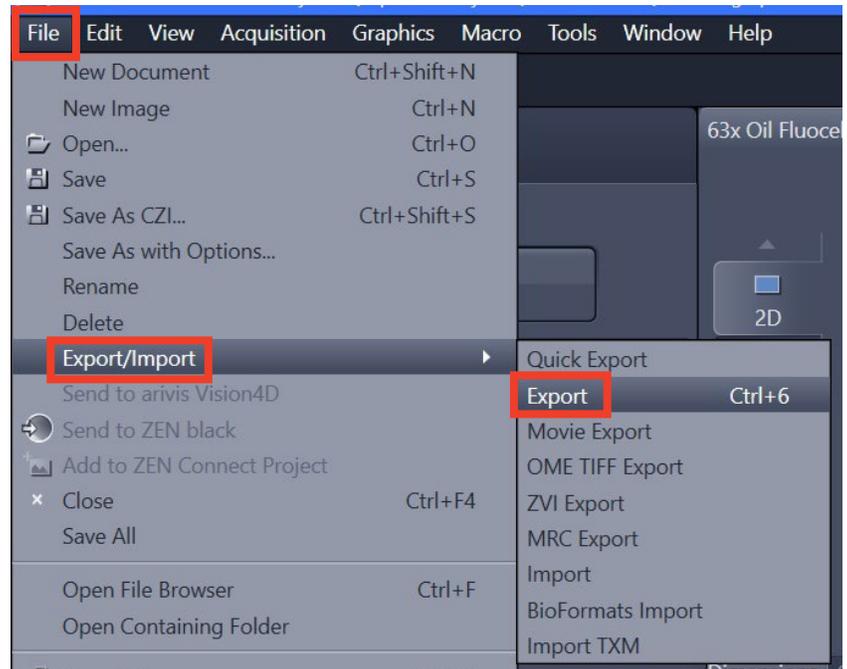


Seeing beyond

The ZEISS ZEN Blue software has an expanded set of tools for adjusting, annotating, and performing measurements on images – but how can these tweaks be finalized into formats ready for collaborative use? The following guide details the steps required for exporting image sets and standalone movies, all while providing starting point recommendations for the parameters of each.

Single Image Export

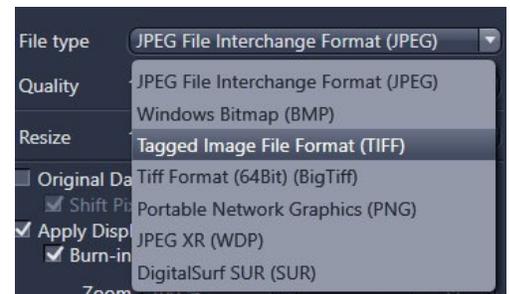
Exporting an image or images from a single .CZI file input is possible regardless of the dimensions involved (e.g. – multichannel, Z-stack, time series, multiple positions). However, all of these options first require that the image file is first open within ZEN. Once the image is active, the Image Export tool can be easily accessed via the menu item File → Export/Import → Export, or via the CTRL+6 keyboard shortcut. Both options will take the user to the Processing tab automatically.



Once there, expand the Input window at the bottom. This controls which image in the workspace will be used for exporting. If the Set Input Automatically checkbox is ticked, the currently-displayed image will be used.

Above the Input window is the main Parameters window. This window contains all options for controlling the output of the image export process. Make sure the Show All toggle in the upper right is active to reveal expanded details, if needed. These full options include:

1. File type – determines the output format; options include JPEG, TIFF, BigTiff, PNG, and more
2. Quality/Compression – permits reduction of the exported file size; options visible will differ depending on file type
3. Resize – changes the final image dimensions (pixel count) by the given percentage value

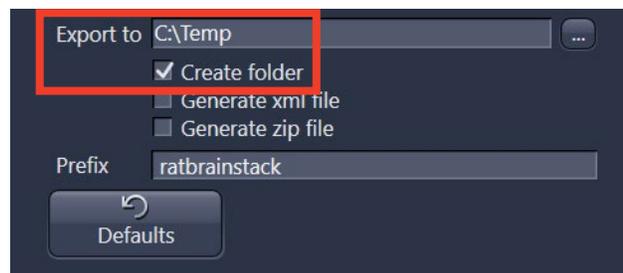
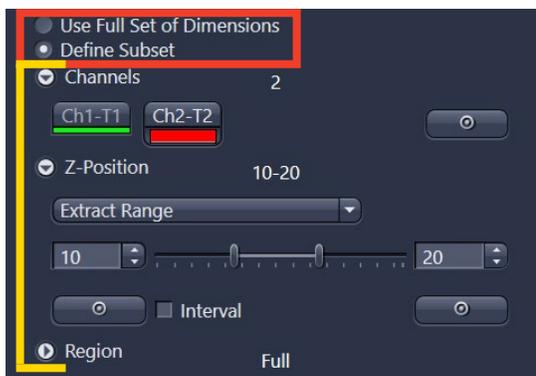
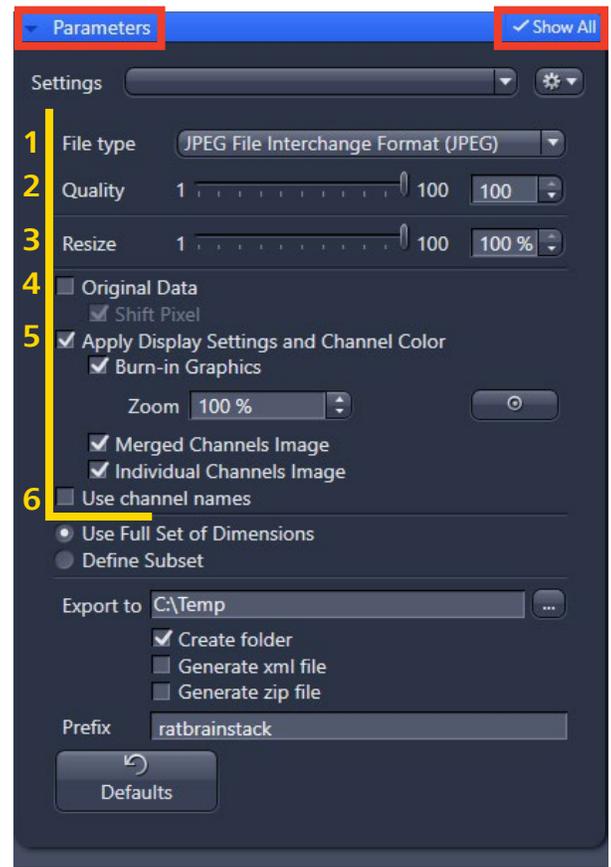


4. Original Data – only the raw data will be exported with no pseudocoloring, display adjustments, or graphic overlays applied
 - a. Shift Pixel – converts image pixels to 16-bits before further downsampling
5. Apply Display Settings and Channel Color – exports the image with pseudocolor and display adjustments applied as seen on the screen; this is the default setting
 - a. Burn-In Graphics – scale bars, time stamps, or other annotations will be kept on the exported image; the zoom of these elements can also be adjusted
 - b. Merged Channel Images – exports a merge of all existing channels
 - c. Individual Channel Images – exports each channel as a separate image; independent of the Merged Channel option above
6. Use Channel Names – uses any existing channel names from ZEN and applies it in the resulting exported image file name

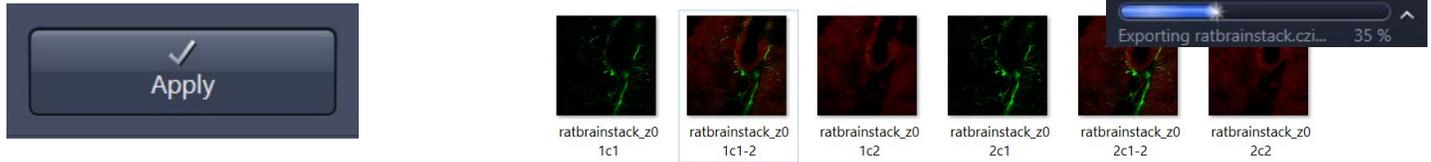
The Use Full Set of Dimensions option will perform the exporting functions on all multidimensional aspects of the image set. This means that, for example, a multichannel Z-stack will yield individual images at each Z plane within the original file.

The alternative option, Define Subset, permits extraction of more limited portions of such an image set. When selected, the fields shown here will update based on the current active image. Channels, Z-Position, Time, Scenes, Region (ROI), or even Tiles can be independently restricted. This can minimize generation of unnecessary data.

The bottom of the Parameters window contains a directory path for determining where files will be exported. Note that the Create folder checkbox is ticked by default, which will place all resulting images in a self-contained folder. Additional options for creating an .XML metadata file or a complete .ZIP file of all data are also available. Finally, a common prefix for all exported images can be assigned.

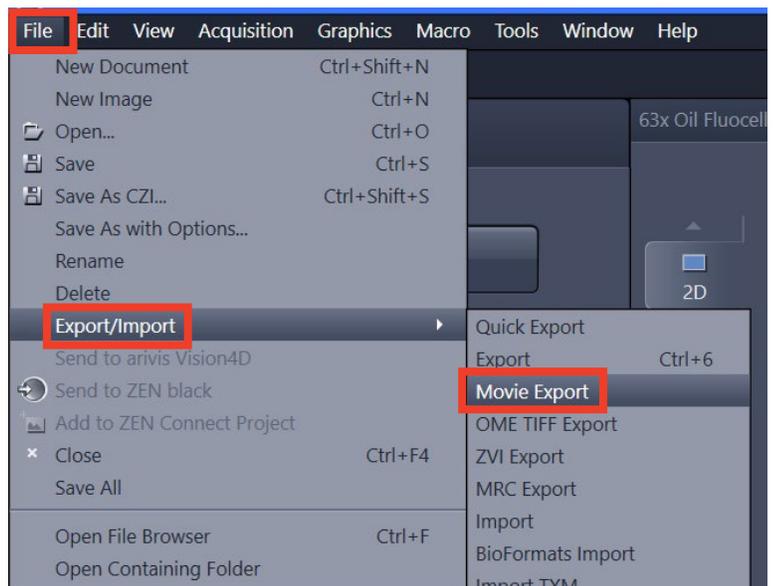


Once the **Parameters** are set, the export action can be initialized with the large **Apply** button above the window. This process is relatively fast and proportional to the input file size, but can be monitored with a progress bar that appears at the bottom of ZEN. Resulting images can be retrieved immediately and will appear as they are generated.



Movie Export

A standalone movie file can be exported from most multidimensional datasets (multichannel, Z-stack, time series, multiple positions) with control over the playback sequence. To access the Movie Export toolkit, click the menu item File → Export/Import → Movie Export when a suitable image is active.



Use the **Input** window to define the target image; the Set Input Automatically checkbox can speed up this step for selecting an image.

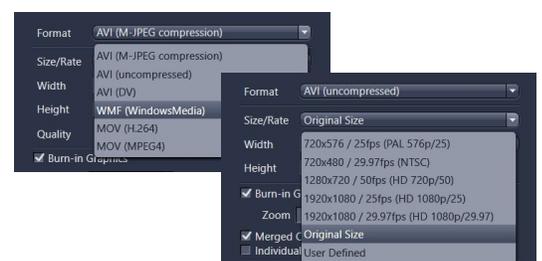
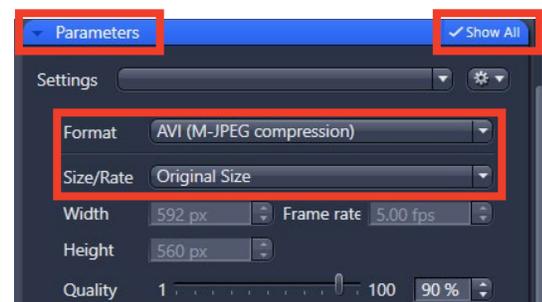
The **Parameters** window contains all available options for controlling the movie output. The Format and Size/Rate sections are linked, providing options for .AVI, .WMV, and .MOV and the relative video size/quality. The Original Size selection will maintain the raw image pixel counts and should be considered as a starting point.

Note that due to licensing restrictions, the .MOV format requires a simple installation of additional encoding files. The Windows 64-bit build should be downloaded from the link below, and the ffmpeg.exe file extracted to the given directory:

<https://ffmpeg.zeranoe.com/builds/>
C:\Program Files\Carl Zeiss\ZEN 2\ZEN 2 (blue edition)



Seeing beyond



Once a file format is determined, additional settings include:

1. Burn-in Graphics – scale bars, time stamps, or any drawn annotations will appear on the exported image; the relative zoom of these elements can be adjusted (from 1% and up)
2. Merged/Individual Channels Image – one or both options can be checked; if multiple options are active, the exported movie cycles through all channel images
3. Fitting – if a specific set of video pixel dimensions is selected (in the Size/Rate section above), this option allows control over how the result is stretched or cropped to fit the format
4. Sequence – the order can be determined if cycling between multiple dimensions during the final movie playback
5. Mapping – options include 1 Frame per Image and Fixed Duration; the latter permits manually typing a frames/sec value to control the Final Movie Length field

The Use Full Set of Dimensions option will export a movie using all multidimensional aspects of the image set. This means that, for example, a Z-stack + time series input will potentially result in a movie that cycles through all Z planes before advancing the time point

The Define Subset option is therefore useful to help restrict a time series output to a single focal plane (for Z-stacks) or a shorter time range when sample dynamics are most interesting.

The Export To field controls the destination folder for the generated movie file. The addition of a Prefix to the file name is also possible.

With the **Parameters** window set, the movie export can be initialized with the **Apply** button at the top near the main **Processing** tab. The overall movie encoding time may vary significantly depending on the compression settings utilized.

