

Microscopy Data Management

When you acquire an image and hit the “save” button, you face several seemingly inconsequential, but very important, questions. *What do I name it? Where do I save it?* Your answers to these and other questions will impact how easily you and others can access and use your data days, months, and years from now. Poorly managed data may be unusable by future-you, your lab, and other researchers—in other words, it may be useless. In contrast, well managed data may help you complete your project more easily, inform future research by your lab, and be a beneficial resource to the wider scientific community.

What are my data?

Your images... but also their metadata!

Assuming you took your **images** carefully and correctly, they are an accurate representation of what you observed through the microscope. To maintain their integrity, they need to be saved in the correct file format, stored in a safe location, and never directly modified. In addition, you will have **metadata** or “data about your data.” They are information about the instruments, parameters, and specimen preparation that were used to produce your data (images). They are critical for enabling someone (including future-you) to understand and reproduce your results.

In which file format should I save my image?

Use a file format that preserves your image exactly as you collected it.

There are many different image file formats and they differ in several important properties. There is a trade-off between files that preserve your images exactly as you acquired them and retain all the image’s metadata, and files that are small and easy to access. The following table shows the most common image formats you’ll encounter when imaging in our facility.

Format	Preserves original data?	Retains metadata?	File size	Compatible Software
.nd2, .lif, .czi	Yes	Yes	Large	Manufacturer’s program; FIJI/ImageJ
TIFF	Yes	Partial or no	Large	Manufacturer’s program; FIJI/ImageJ
JPEG-2000	Yes	Partial or no	Medium	Manufacturer’s program; FIJI/ImageJ
JPEG/JPG	No	No	Small	All common programs
PNG	It depends	No	Medium	All common programs
GIF	No	No	It depends	All common programs

If possible, save your images in the **microscope’s proprietary file format** (Nikon .nd2, Leica .lif, Zeiss .czi). These file formats contain the maximum amount of metadata meaning that you do not need to record as much by hand. Some systems do not have a proprietary format or your research advisor may have a reason why they don’t want to use them. In these cases, you should use TIFF (.tiff or .ome.tiff) or uncompressed/lossless JPEG-2000 (.jp2) files because these will save your data exactly as they were acquired.

The other file types (jpeg, png, gif) are easier to use because they are smaller, can be easily shared (email/text/chat), opened on most devices, and imported into other programs to make reports and presentations. However, in compressing the image, they lose information. *Therefore, you should never save original images in these “lossy” file*

types. A good strategy is to save your original images in one of the proprietary formats and then, when necessary, export a copy of the image in a different format. For example, you may save a copy as a PNG or JPEG to import into a PowerPoint presentation or as a TIFF to create a figure for a manuscript.

Where should I save it?

Save it somewhere safe, backed-up, and easily accessible to you and your lab.

Your lab likely already has a standard procedure for where to save data. Here are some additional tips:

- Do *not* keep your files on the facility computers. Data may be erased at any time and without warning.
- Store data on the biology server or in the cloud (e.g. OneDrive, OSF, Teams) because these locations are backed-up and easily accessible. Do *not* use photo-sharing sites or email them to yourself. Only use local physical drives (lab computer, USB drives, etc.) as a last resort and, if you do, be sure they are backed-up to a location in another building.
- Be sure that your research advisor (and the rest of your lab, if applicable) can access your files. Remember that, if you are conducting research in a lab, your data is ultimately the property of your lab and university.
- Do *not* modify your original files. These are your data. If you need to modify them, e.g. during analysis or to prepare a figure, make a copy and work on the copy instead.
- Do *not* delete your data.

What should I name my image file?

Names should be consistent, orderly, and informative.

Bad	Good
slide1b feb14 2016	slide1b_2016-02-14image1
Slide1B-feb14-2016 image2	slide1b_2016-02-14image2
1c 2-14-16 1 ...	slide1c_2016-02-14image1 ...

Organize images in folders to reduce the amount of information you need to put in image names. How you do it depends on what makes the most sense; just pick a scheme and stick with it. For example...

- Name folders by experiment/project (“exp10-protocolTest”)
- Name folders by date (“2016-01-01_more_cell_movies”)
- Make sub-folders (e.g. one folder for each slide in an experiment)
- If you are using the Leica confocal or DM6b, Leica Projects can serve the same purpose as folders

Do *not* use names to record notes about the experiment, e.g. “control2 re-do USE THIS ONE” or “treated3-weird-blob.” Write those notes in your lab notebook or create a “ReadMe” file. A “ReadMe” file is a simple text file (e.g. use Notepad) that you save with your images where you can keep notes about how your images were acquired and how they’re organized. This isn’t a replacement for your lab notebook; it’s a way for you and others to quickly identify and understand the images stored in a specific location.

Good naming conventions

No special characters (/, %, etc.)

No spaces. Use ‘_’, ‘-’, or useCamelText.

Format dates: 2016-01-31 so they’ll sort correctly. Don’t use slashes (/).

Pad numbered files with 0s (file01, file02...file10) so they’ll sort correctly.

Is there anything else I should save?

Be sure that important parameters about your imaging are recorded.

Some parameters (e.g. objective and fluorescence filter properties) can be found on the instrument specification sheets (<https://www.jmu.edu/microscopy/instrumentation/index.shtml>) as long as you know what objective you used. Others depend on how you acquired your images (e.g. exposure time). Luckily, if you save your image in one of the proprietary file formats (Nikon .nd2, Leica .lif, Zeiss .czi), these other parameters are saved in the image's metadata so you don't have to do anything! However, if you do not or cannot save your images in one of these formats, you must record the information by hand. You can generate a checklist of required parameters using this webapp: <https://kris-kubow.shinyapps.io/MicCheckJMU/>. Additional parameters may need to be reported if you are using an advanced technique such as Leica Lightning or FLIM.

When you present your research, you must also tell your audience how you performed your experiments. This information not only helps your audience understand your results, but also allows them to verify that your interpretations are correct and ultimately to reproduce or build upon your findings. The level of detail you report usually depends on how you're presenting your research.

Manuscript or other published format: Report *everything* needed to fully reproduce your imaging. This includes, at a minimum, the items in the checklist referenced above.

Poster presentation: At a minimum, report the objective specifications listed on the checklist, the microscope and component (e.g. camera) names, the microscope technique, any stains/fluorophores used, and include scalebars on your images.

Oral presentation: At a minimum, report the microscopy technique, any stains/fluorophores used, and include scalebars on your images.

