As part of its new mandated Data Sharing Policy, NIH is requiring that new grant submissions include a data sharing plan. Basic requirements for sharing include the use of persistent, unique identifiers for each published dataset, such as a DOI. For image data, files should be made available in a format that can be read by a variety of common image analysis software. Reproducibility of the experiment or reuse of the shared data for new analyses require inclusion of at least a minimal set of details about how the image was acquired (metadata). Often this information is included in the header of the image file when it is saved by the acquisition software. A number of public data repositories exist already for sharing data and many are listed on the NIH website. The information provided below is intended to assist those who rely on imaging data for their research but who have never thought about publicly sharing it. More information about the NIH policy can be found here:

Suggested Repositories for sharing image data:

Due to restrictions on file size and format, not all data repositories are amenable for sharing large imaging datasets, such as very large multidimensional images or multiple replicates representing a statistically significant sampling. Three public repositories that are often used by the imaging community are Zenodo (https://zenodo.org/), figshare (https://figshare.com/) and BioImage Archive (https://www.ebi.ac.uk/bioimage-archive/). Each of these repositories assigns unique persistent identifiers for each dataset that is published on their site. The online instructions for using these repositories are generally clear, and uploading the data is straightforward. Typically, single images or .zip files containing multiple images can be uploaded by drag-and-drop, and there is an ftp option for batch upload of multiple images or very large files. These repositories will only accept patient data if it has been de-identified. Zenodo and BioImage Archive require you to organize your dataset in hierarchical folders before uploading. figshare includes GUI tools for organizing the data online after uploading. Some important features of these three repositories are compared in the table below:

Repository	Cost	Maximum File Size	Total Allowable Quota	Data retention
Zenodo (CERN)	Free	50 GB	Unlimited , donation	indefinite
figshare	Free ≤ 20 GB	20 GB	Sliding cost scale > 20GB	≥ 10 years
BioImage Archive (EMBL)	Free	1 TB	Unlimited	As needed

Recommended Options for Image File Format:

Research image data always should be saved and preserved in its original format since this is the primary data. For compatibility with a wide variety of image analysis software, research image data also may be exported in a format that does not change the dimensions or intensity values of the original pixels. Two recommended options are:

Option 1 - Save as vendor proprietary format (.czi, .nd2, .obf, DICOM)
Option 2 - Export as ome-tiff (available option in most microscope vendor software and standard for home-built instruments)

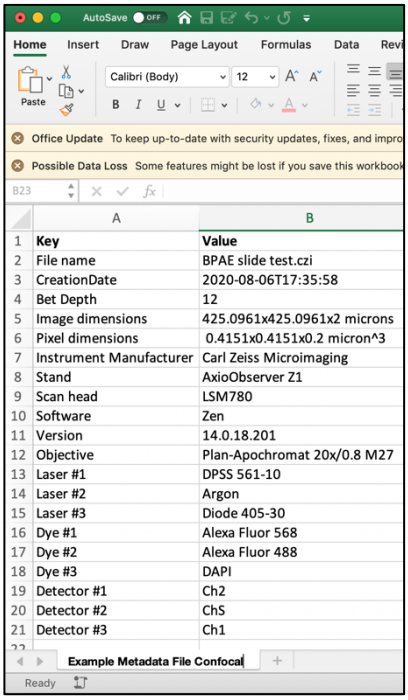
Most major microscope manufacturers' file formats, as well as OME-TIFF and DICOM, can be opened in the open source image analysis package ImageJ/Fiji using the BioFormats plugin. The OME-TIFF and DICOM formats can be opened also in a variety of other image analysis software, including MATLAB. Exporting as OME-TIFF format retains metadata written into the header of vendor proprietary image formats.

More about metadata:

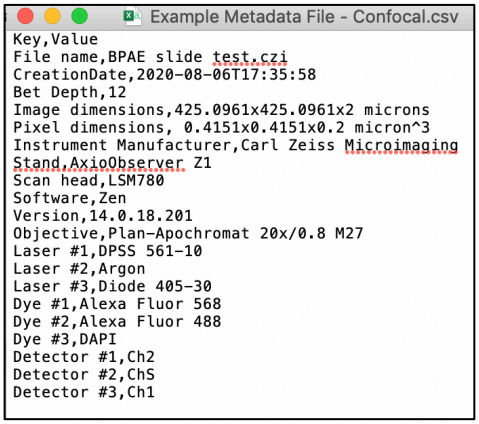
Metadata is additional information about the image, such as date and time of acquisition (timestamps for multiple images in a z stack or time sequence), image dimensions, pixel/voxel dimensions, and acquisition parameters. Many microscope vendors include extensive information in the metadata for each image when it is saved in their proprietary format. In principle, this metadata is retained when the image is exported into OME-TIFF format. The BioFormats plugin that ships with Fiji usually can read and display all or part of the metadata from the file header.

File formats such as standard .tiff and .jpg typically do not keep the metadata. Saving as .jpg is not recommended in any case because the .jpg file format discards some of the original pixel data to keep the file size small (called compression) and the image no longer represents the original data. If you have no other option than to save as .tiff or .jpg you will need to generate your own metadata file and upload it along with your image dataset.

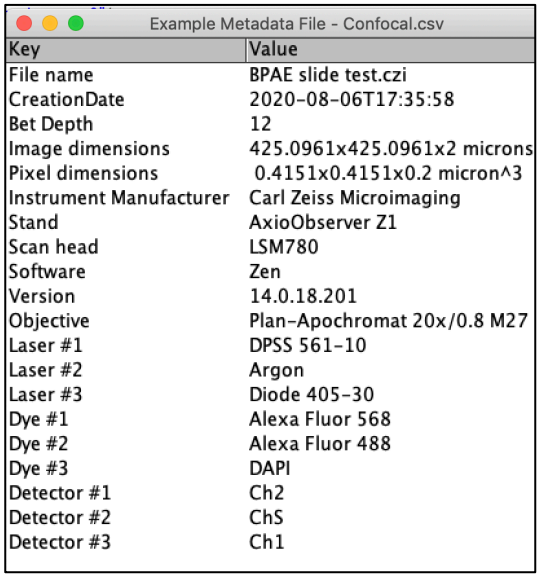
Metadata file types: Key/Value: The simplest type of metadata file is a Key/Value file in .csv format. This can be generated in Excel and saved as .csv so it can be read by ImageJ/Fiji. A Key/Value pair consists of two related data elements, where the Key is a constant that defines the pair, and the Value is a variable that specifies the details about the Key. For example: Channel/FITC or Microscope/Nikon Ti2. The Key/Value pairs are stored in a two column table. An example Key/Value file is shown as it looks in Excel, in a text editor and in Fiji are shown below:



	A	B
1	**Key**	**Value**
2	File name	BPAE slide test.czi
3	CreationDate	2020-08-06T17:35:58
4	Bet Depth	12
5	Image dimensions	425.0961x425.0961x2 microns
6	Pixel dimensions	0.4151x0.4151x0.2 micron^3
7	Instrument Manufacturer	Carl Zeiss Microimaging
8	Stand	AxioObserver Z1
9	Scan head	LSM780
10	Software	Zen
11	Version	14.0.18.201
12	Objective	Plan-Apochromat 20x/0.8 M27
13	Laser #1	DPSS 561-10
14	Laser #2	Argon
15	Laser #3	Diode 405-30
16	Dye #1	Alexa Fluor 568
17	Dye #2	Alexa Fluor 488
18	Dye #3	DAPI
19	Detector #1	Ch2
20	Detector #2	ChS
21	Detector #3	Ch1



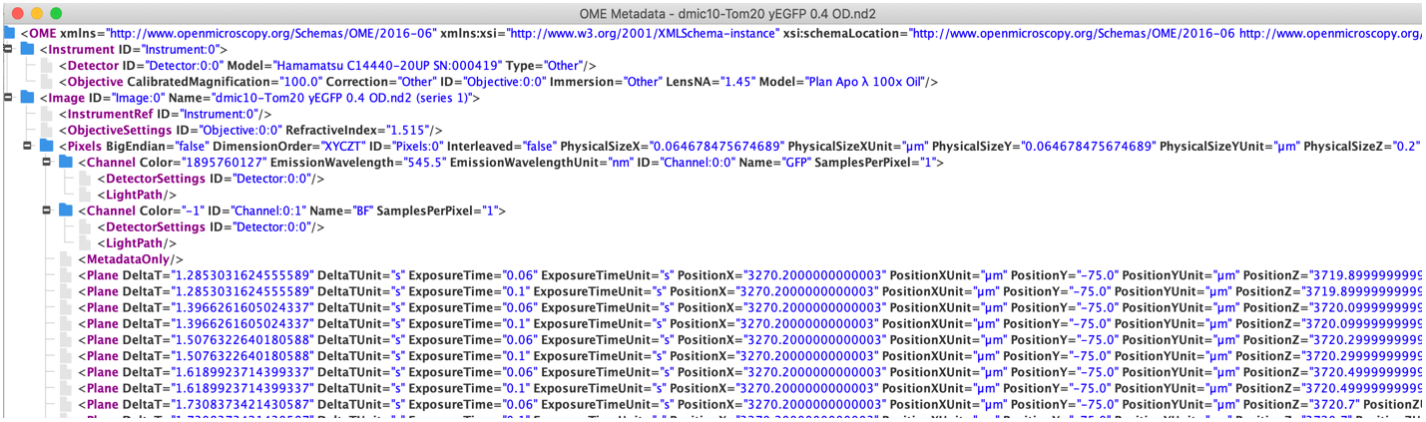
```
Key,Value
File name,BPAE slide test.czi
CreationDate,2020-08-06T17:35:58
Bet Depth,12
Image dimensions,425.0961x425.0961x2 microns
Pixel dimensions, 0.4151x0.4151x0.2 micron^3
Instrument Manufacturer,Carl Zeiss Microimaging
Stand,AxioObserver Z1
Scan head,LSM780
Software,Zen
Version,14.0.18.201
Objective,Plan-Apochromat 20x/0.8 M27
Laser #1,DPSS 561-10
Laser #2,Argon
Laser #3,Diode 405-30
Dye #1,Alexa Fluor 568
Dye #2,Alexa Fluor 488
Dye #3,DAPI
Detector #1,Ch2
Detector #2,ChS
Detector #3,Ch1
```



Key	Value
File name	BPAE slide test.czi
CreationDate	2020-08-06T17:35:58
Bet Depth	12
Image dimensions	425.0961x425.0961x2 microns
Pixel dimensions	0.4151x0.4151x0.2 micron^3
Instrument Manufacturer	Carl Zeiss Microimaging
Stand	AxioObserver Z1
Scan head	LSM780
Software	Zen
Version	14.0.18.201
Objective	Plan-Apochromat 20x/0.8 M27
Laser #1	DPSS 561-10
Laser #2	Argon
Laser #3	Diode 405-30
Dye #1	Alexa Fluor 568
Dye #2	Alexa Fluor 488
Dye #3	DAPI
Detector #1	Ch2
Detector #2	ChS
Detector #3	Ch1

XML: Metadata is sometimes saved as a separate file in XML format instead of written in the image file header. XML files are structured hierarchically as shown below. XML files can be read by the BioFormats plugin for ImageJ/Fiji and by many other image analysis software packages. ImageJ/Fiji has an option to display the metadata it reads from any image file header in XML format, but it is not possible to output the metadata in XML format, even by copy and paste from the display window. It is possible to generate an OME-XML file from the header of an image file using BioFormats Command line tools: https://downloads.openmicroscopy.org/bio-

formats/5.5.3/artifacts/bftools.zip). This file can then be edited in an XML editor such as Oxygenxml (https://www.oxygenxml.com/) or a text editor. Here is an example of metadata displayed as OME-XML in Fiji:



JSON: This file type currently is not standard output from most imaging acquisition software but has been proposed as a universal standard for microscopy as it can be read by many image analysis applications and is conveniently structured for describing microscope configurations and image acquisition parameters. An interactive graphical interface app (MicroMeta App) for generating JSON files to accompany microscope imaging data is under development (https://micrometaapp-docs.readthedocs.io/en/latest/) The current version can be downloaded here: https://github.com/WU-BIMAC/MicroMetaApp-Electron/releases/tag/1.7.25-b1

MicroMeta App assists you to generate a "microscope" file in JSON format that completely describes your instrument (only needed once unless the instrument is upgraded with new hardware or software). Using this microscope file, you can then generate an "acquisition settings" file that specifies what hardware components and settings were used to acquire the data you are sharing. You can then upload both files along with your dataset.

Beyond image metadata:

To be fully reproducible, image data must be accompanied by details about the experiment that likely are not included in a standard image metadata file, such as tissue type, species, experimental treatments, etc. If the image data is shared in the context of a published manuscript, presumably these details will be included in the Materials and Methods section of the accompanying publication. A pdf of the publication should be uploaded along with the images. The unique persistent identifier for the publication could be included as a key, value pair in the image metadata file. If the images you want to share are not associated with a publication, then completeness demands that a text file detailing the materials and methods of the experiment be uploaded together with the image data.

Licensing:

Unlike publishing a journal article, you retain ownership of research data that you share through a public repository. Publicly shared research data should be accompanied by some type of license specifying how the data can be used by a third party. If you do not choose or provide a license, the data you upload will be covered by the default license for the repository. There are three general categories of licensing:

- No rights reserved, public domain
- Attribution (reusers have to cite you, commercial use is permitted).
- Attribution – Non-Commercial (reusers have to cite you, no commercial use permitted)

Most public data repositories allow you to select from a list of OpenAccess Licenses. Of these, Creative Commons and OpenData Commons are the most widely used. Creative Commons has a tool for deciding which license is appropriate for your data: https://chooser-beta.creativecommons.org/ If you do not want to agree to one of these, or if the repository doesn't offer any of these, you may upload a custom license along with your data. A custom license suggested by the UT Southwestern Office of Technology Transfer is available here: [insert URL when available]. For a more detailed discussion of licensing see here:

https://www.dcc.ac.uk/guidance/how-guides/license-research-data#x1-8000doc