UltraMicroscope II: Image Handling Considerations

AMCF UltraMicroscope II: DRC I Room 1063

Schedule through AMCF core staff (advancedmicroscopy@unmc.edu)

Working with LSFM Images

The UltraMicroscope software (ImSpector) can only perform *basic visualization* of samples post-capture (only single field of view). The default file format set by Miltenyi is ome tiff, a universal image type (open microscopy environment tiff). The default save/storage setting can alternatively be set to Imaris (*.ims). This is dependent upon how individual researchers choose to work with their data (FIJI/ImageJ or Imaris). For researchers planning on using the data analysis workroom, set the file type to *.ims (or plan on converting *.ome tiff to *.ims file, see below).

a. Imaris file converter software has been placed on the LSFM acquisition computer and the Imaris 1 workstation in the data analysis room (DRC I Rm 1036). Once converted from *.ome tiff to *.ims (if not originally saved as *.ims), images can be viewed using the full Imaris software (data analysis room only) or the free version of the Imaris software (very limited functions, viewing options).
   i. Imaris Viewer - Imaris - Oxford Instruments (oxinst.com)
   ii. ImSpector software (data acquisition workstation)
      1. *does not* stitch files together.
      2. Exporting ome tiff images to *.ims using ImSpector, is slow and not recommended.
      3. Imaris file converter software is available on the LSFM workstation and IMARIS 1 workstation (data analysis workroom).
b. Imaris stitcher software is part of the core facilities package and is therefore only available on the IMARIS 1 workstation in the data analysis workroom.
   i. Set the acquisition parameters to save as*. ims or convert individual images to *.ims files prior to stitching, see below.
   ii. Mosaic/tiled images *must be converted to *.ims then stitched* into a composite image before viewing, see below.
c. Alternative Option: There is a 'big stitcher' plugin for FIJI that can be used to fuse and visualize *.ome tiff files on any computer with sufficient memory to do so (https://imagej.net/plugins/bigstitcher/).

LSFM/volumetric images are Large (10's to 100's of GBs)

Please ensure adequate storage space and upload/transfer times. The AMCF has installed 10 Gb ethernet connections between the new instrumentation (Axioscan whole slide imager, Light Sheet Fluorescence Microscope) and the Data analysis room. Many locations on campus are 10 Gb 'ready,' not actively installed/configured. Researchers should verify individual transfer capabilities in their location/building and plan accordingly (table from LSFM/UltraMicroscope II user guide).

- Data write *speeds* under good conditions are:
  - 10 Gb Ethernet: 500 Mb/s
  - 1 Gb Ethernet: 50-100 Mb/s
  - USB3: 50 Mb/s
- Data write *times* are shown in Table 11.1.

| | Transfer method | | |
Amount of data	10 Gb Ethernet	1 Gb Ethernet	USB 3
1 Gb	2s	10-20s	20s
10 Gb	20s	1-3 min	3 min
100 Gb	3 min	15-30 min	30 min
1000 Gb	30 min	2.5-5 hours	5 hours

Table 11.1 Data write times for different amounts of data and transfer methods. Note that these are best-case values which are not always achieved in practice.

Image Analysis Workstation (IMARIS)

These systems are currently connected to the network. Data may be securely transferred to individual researchers/research groups using the AMCF OMERO server or researcher provided external drives. Data may reside on the workstation for *up to 3 months*. After this time (or data not stored on the designated data drive) will be removed from the acquisition workstations *without prior notification*. Data temporarily stored/transferred using BOX may remain there, it is the responsibility of the researcher to establish long-term data storage for these files. The AMCF recommends researchers move images collected prior to 2024 to the AMCF OMERO server.

Data Analysis Workroom

Located in the Data Analysis Room (DRC I RM 1036).

Contact the AMCF team to request permission to independently reserve/use workstations (RSS) and obtain card access for this area.

You can find more information about the available modules and associated training resources by accessing the AMCF website.