

Using Scan Files

The Vectra Polaris generates qptIFF files. Metadata is retained within this file for fluorescence scans. qptIFF files are compatible with a variety of image viewers that incorporate the BioFormats extension. Older version of some programs may require addition of BioFormats as a plug-in.

TMPSR will maintain backups of scan files for a limited period of time. However, you will be responsible for long-term storage of your data.

Slide Viewing and Analysis Software Options:

InForm

InForm software performs spectral unmixing to provide maximum resolution between fluorescence signals in Polaris scans. This software is also capable of several analyses, including quantitation and phenotyping, on selected fields of your slide. Because of the complexity of this approach and the logistical considerations associated with the limited number of workstations that have this software, discussion with a Core director is necessary to confirm that this analysis can be used for your project. If you are interested in using InForm software, several additional steps will be required:

- 1) If you are not using stains performed by the TMP Shared Resource, please submit additional slides needed to generate a spectral library.
 - a. Unstained slide for autofluorescence
 - b. For each marker in the panel, provide one slide stained with only that single marker (if you have multiple pieces of tissue on the slide, do not apply a different marker to each tissue piece)
- 2) We recommend using OPAL approach for immunofluorescence staining
- 3) For some projects (especially if an OPAL staining approach was not used), a 2nd round of scanning may be required
- 4) Although we currently do not charge for InForm per se, please be aware that you may be charged at the usual rate for the extra scans required for this analysis

Akoya Phenochart—Freeware for viewing fluorescence and brightfield slides. Recommended for viewing fluorescence slides. (For Mac users, QuPath is another freeware option.)

<https://akoyabio.helpdocs.com/phenoptics-software-updates/phenochart-version-1011>

Aperio Imagescope—Freeware for viewing brightfield slides.

<https://www.leicabiosystems.com/digital-pathology/manage/aperio-imagescope/>

QuPATH—Visualization and analysis.

Note: If pixels showing instead of microns, go to Edit>Preferences>Bioformats, then enter “qptiff” into the “Always use Bio-Formats for specified image extensions”

imageJ/FIJ—Visualization and analysis

Requires Bioformats extension/plugin.