To: File

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Re: PerkinElmer image format

**Background**

This describes a TIFF format that PerkinElmer uses for its tissue images. The imagery may be a simple RGB image, a set of unmixed multispectral image (MSI) components, or a whole-slide scan. In the latter case, it may be a brightfield (BF) color RGB image or a multiband fluorescence (FL) image.

The goal is to use the same syntax and metadata for all these kinds of images, and minimize the semantic distinctions where possible. Specifically, the unmixed MSI images can be considered to be an idealized multiband FL image where signal corresponds to the presence of a stain or fluorescent dye in the sample.

**Data format**

The files will be TIFF or BigTIFF images, depending on image size, with multiple images per file.

For images larger than about 2K x 2K pixels, tiled format is used, and the image is provided in several resolutions (pyramidal tiled images). Tile size is 512 x 512 pixels.

The finest resolution is shown first, and for each resolution there are N images where N depends on the contents. For BF images, N=1 and each image is an RGB image. For FL images or unmixed component images, N = number of bands, which is usually > 1, and each image is a grayscale image.

A thumbnail RGB image is provided, and this is a good image to use as an icon in graphical image lists. This comes after the baseline images, meaning it is the second image in BF (RGB) images, and the (N+1)st image for FL images or unmixed component images.

Next come the reduced-resolution images (if present). The pyramid contains enough levels that the image size is no larger than 2K x 2K at the coarsest resolution.

For whole-slide scans, there are two more non-tiled images after these: an RGB image of the label, and a macro (low-resolution) RGB image of the whole slide.

Overall, the arrangement is:

**Table 1. Images**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Description** | **RGB / mono** | **Tile / Strip** | **Resolution** | **Notes** |
| Baseline image | Varies | Varies | Full | Tiled if > 2K x 2K  RGB for BF, else mono |
| More full-resolution images | Mono | Varies | Full | If N > 1 |
| Thumbnail | RGB | Stripped | ~500 x 500 |  |
| Half-resolution images | Varies | Tiled | Half | Only if baseline is tiled |
| Quarter, eighth, etc. | Varies | Tiled | Quarter, eighth, etc. | Continues until 2K x 2K or smaller |
| Macro (overview) image of whole slide | RGB | Stripped | ~2000 x 4000 | Required for whole-slide scans  Optional for simple RGB and MSI components |
| Label image | RGB | Stripped | ~500 x 500 | Optional, whole-slide scans |

**Detection**

Readers can recognize PerkinElmer tissue images via the contents of the “Software” TIFF tag (see below). The file suffix is .qptiff.

**Metadata**

Metadata is contained in two locations: within standard TIFF tags as listed in the table below, and within the ImageDescription string, using a set of XML tags described below. These are provided for each image (IFD) in the file, and describe that image rather than the baseline image or the scan as a whole. The ScanProfile tag is only provided on the first, baseline image as it may be large.

**Table 2. TIFF tags**

|  |  |  |
| --- | --- | --- |
| **TIFF Tag** | **Optional** | **Description of contents** |
| Software |  | Starts with “PerkinElmer-QPI” |
| ImageDescription |  | Further metadata in XML format (see next section) |
| ImageWidth |  | Width of the image in pixels |
| ImageLength |  | Height of the image in pixels |
| ResolutionUnit | Y | Unit used for resolution and position (see below) |
| XResolution | Y | Pixel X resolution (see below) |
| YResolution | Y | Pixel Y resolution (see below) |
| XPosition | Y | Sample X location in ResolutionUnits. This is ULHC location except for Macro image which reports its image center. |
| YPosition | Y | Sample Y location in ResolutionUnits. This is ULHC location except for Macro image which reports its image center. |
| SampleFormat |  | Integer (1) for BF, FL; or float (3) for unmixed MSI images |
| SMinSampleValue |  | Minimum signal value in the image |
| SMaxSampleValue |  | Maximum signal value in the image |
| BitsPerSample |  | 8 (FL); 8, 8, 8 (RGB); or 32 (unmixed component) |
| SamplesPerPixel |  | 1 (FL or unmixed component) or 3 (RGB) |
| NewSubfileType |  | 0 for full-resolution images, 1 for reduced res images |
| TileWidth | Y | Tile width (512) if tiled format is used |
| TileLength | Y | Tile height (512) if tiled format is used |
| TileOffsets | Y | List of tile offsets, if tiled format is used |
| TileByteCounts | Y | Size of each (compressed) tile, if tiled format is used |
| StripOffsets | Y | List of strip offsets, if tiled format is **not** used |
| RowsPerStrip | Y | Number of rows per strip, if tiled format is **not** used |
| StripByteCounts | Y | Size of each (compressed) strip, if tiled format is **not** used |
| PlanarConfiguration |  | 1 (chunky) for RGB images, 2 (planar) otherwise |
| PhotometricInterpretation |  | 2 (RGB) for RGB images, 1 (BlackIsZero) otherwise |
| DateTime |  | Acquisition time |
| Compression |  | May be None, CCITT Group 3, PackBits, LZW, or JPEG |
| JPEG fields | Y | JPEG fields are defined when JPEG compression is used |

ResolutionUnit, XResolution and YResolution are required fields in a valid TIFF file. When the true resolution of the image is known, ResolutionUnit will be 3 (cm) and XResolution and YResolution will be pixels/cm. When the true resolution is not known, ResolutionUnit will be 2 (inch) and XResolution and YResolution will be 96 (pixels/inch). PKI TIFF pixels are always square so XResolution and YResolution will always have the same value.

The TIFF spec is not explicit about the data type and value for SMinSampleValue and SMaxSampleValue; the PerkinElmer writer uses the same data type as the image pixels (byte or float).

**Image Description contents**

The ImageDescription tag contains a string in XML format. The string contains a top-level < PerkinElmer-QPI-ImageDescription> element. Nested within this element are child elements with the tag names and values as listed in the table below. Elements appear in the order listed. Values are stored as text content of the element. Elements are required unless otherwise specified. See the example below.

**Table 3. Image Description tags**

|  |  |  |
| --- | --- | --- |
| Tag | Optional | Contents |
| DescriptionVersion |  | Version of the image description field itself, a single number. This document describes version 2 of the field. |
| AcquisitionSoftware |  | Software used to acquire the image |
| Identifier |  | GUID in string format. This is an identifier for the image file itself. |
| SlideID | Y | ID of the slide that this image was taken from. |
| Barcode | Y | Barcode text of the slide this image was taken from. |
| ComputerName | Y | Name of the computer on which the slide was scanned. |
| ImageType |  | A string identifying the type of image within the file (Table 1), with the following values:   * FullResolution * ReducedResolution * Thumbnail * Overview * Label |
| IsUnmixedComponent |  | “True” for unmixed multispectral images, otherwise “False”. |
| ExposureTime |  | Exposure time as an integer number of microseconds.  For unmixed images, this is the exposure time for the dominant wavelength band for the component (FL); or the brightest wavelength in the cube (BF). |
| SignalUnits |  | A byte *wwww tttt* where the *tttt* nibble indicates the signal unit type from the following:   1. – raw counts 2. – normalized (counts/second/gain/full-scale/binning) 3. – OD (optical density) 4. – dark-corrected counts   and the *wwww* nibble indicates how the signal is weighted across the spectral bands (or colors):   1. – average across all bands 2. – total summed signal across all bands 3. – peak signal in highest-valued band   Thus, for example, a value of 68 (hex 44) encodes OD units with peak-signal weighting. |
| Name | Y | Band (component) name for FL or unmixed MSI images, not present for RGB images |
| Color | Y | Color to use when rendering this band, as decimal r,g,b byte triplet, r FL or unmixed MSI images. Not present for RGB images. |
| Objective | Y | Objective name, if known, otherwise not present |
| ScanProfile | Y | Element containing scan and/or and unmix parameters. It is valid XML whose contents are opaque to most readers. It is only provided on the first (baseline) image, and is omitted from all other IFDs. |
| ValidationCode |  | Used for internal data integrity checks – readers can ignore this. |

For whole slide images (BF and FL), SignalUnits will be 64 (hex 40) (raw counts, peak signal). For unmixed images, SignalUnits will reflect the unmix settings.

Sample ImageDescription for the DAPI band of a FL image:

<?xml version="1.0" encoding="utf-8"?>

< PerkinElmer-QPI-ImageDescription >

    <DescriptionVersion>1</DescriptionVersion>

    <AcquisitionSoftware>VectraScan 1.0.0</AcquisitionSoftware>

<ImageType>FullResolution</ImageType>

    <Identifier>AABED946-BB58-44FB-95B3-48E177E3BB83</Identifier>

    <IsUnmixedComponent>False</IsUnmixedComponent>

    <ExposureTime>50</ExposureTime>

    <SignalUnits>64</SignalUnits>

    <Name>DAPI</Name>

    <Color>0,0,255</Color>

    <Objective>4x</Objective>

    <ScanProfile><!-- this will be a serialized scan protocol. It is valid XML but otherwise opaque -->

</ScanProfile>

    <ValidationCode>4281ff86778db65892c05151d5de738d</ValidationCode>

</ PerkinElmer-QPI-ImageDescription >