
vipertools

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Welcome the documentation of VIPERTOOLS!

- **stitching_channel** – string indicating the channel name on which the stitching should be calculated. the positions for each tile calculated in this channel will be passed to the other channels.
- **crop** – dictionary of the form {'top':0, 'bottom':0, 'left':0, 'right':0} indicating how many pixels (based on a generated thumbnail, see `vipertools.stitch.generate_thumbnail`) should be cropped from the final image in each indicated dimension. Leave this set to default if no cropping should be performed.
- **plot_QC** – boolean value indicating if QC plots should be generated
- **filetype** – list containing any of [".tif", ".ome.zarr", ".ome.tif"] defining to which type of file the stiched results should be written. If more than one element all export types will be generated in the same output directory.

`vipertools.stitch.generate_thumbnail(input_dir, pattern, outdir, overlap, name, stitching_channel='DAPI', export_examples=False)`

Function to generate a scaled down thumbnail of stitched image. Can be used for example to get a low resolution overview of the scanned region to select areas for exporting high resolution stitched images.

Parameters

- **input_dir** – Path to the folder containing exported TIF files named with the following nameing convention: "Row{#}_Well{#}_{channel}_zstack{#}_r{#}_c{#}.tif". These images can be generated for example by running the `vipertools.parse.parse_phenix()` function.
- **pattern** – Regex string to identify the naming pattern of the TIFs that should be stitched together. For example: "Row1_Well2_{channel}_zstack3_r{row:03}_c{col:03}.tif". All values in { } indicate thos which are matched by regex to find all matching tifs.
- **outdir** – path indicating where the stitched images should be written out
- **overlap** – value between 0 and 1 indicating the degree of overlap that was used while recording data at the microscope.
- **name** – string indicating the slidename that is added to the stitched images generated
- **export_examples** – boolean value indicating if individual example tiles should be exported in addition to performing thumbnail generation.

TUTORIALS

2.1 Parsing and Stitching Data from Opera Phenix

First you need to export your data from harmony and rename the path to eliminate any spaces in the name. Then you can run the following script to parse and stitch your data.

Listing 1: example script for parsing and stitching phenix data

```
#import relevant libraries
import os
from vipertools.parse import parse_phenix, generate_stitched

#parse image data
path = "path to exported harmony project without any spaces"
parse_phenix(path, flatfield_exported = True, parallel = False)

#define important information for your slide that you want to stitch

# the code below needs to be run for each slide contained in the imaging experiment!
# Can be put into a loop for example to automate this or also can be subset to
↳seperate
# jobs to run on for example the hpc

input_dir = os.path.join(path, "parsed_images")
slidename = "Slide1"
outdir = os.path.join(path, "stitched", slidename)
overlap = 0.1 #adjust in case your data was aquired with another overlap

#define parameters to find correct slide in experiment folder
row = 1
well = 1
zstack_value = 1

#define on which channel should be stitched
stitching_channel = "Alexa647"
output_filetype = [".tif", ".ome.zarr"] #one of .tif, .ome.tif, .ome.zarr (can pass
↳several if you want to generate all filetypes)

#adjust cropping parameter
crop = {'top':0, 'bottom':0, 'left':0, 'right':0} #this does no cropping
#crop = {'top':72, 'bottom':52, 'left':48, 'right':58} #this is good default values for an
↳entire PPS slide with cell culture samples imaged with my protocol

#create output directory if it does not exist
if not os.path.exists(outdir):
```

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```
os.makedirs(outdir)

#define patter to recognize which slide should be stitched
#remember to adjust the zstack value if you aquired zstacks and want to stitch a
↪speciifc one in the parameters above

pattern = "Row"+ str(row) + "_" + "Well" + str(well) + "_{channel}_"+"zstack
↪"+str(zstack_value)+"_r{row:03}_c{col:03}.tif"
generate_stitched(input_dir,
                  slidename,
                  pattern,
                  outdir,
                  overlap,
                  crop = crop ,
                  stitching_channel = stitching_channel,
                  filetype = output_filetype)
```

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