How to run the simflux command line interface

This readme shows how to process a TIFF file using SIMFLUX

Requirements

Hardware
A GPU with CUDA compute capability >= 3.0
This means most GPUs at least as new as the GTX 640 or GTX 740. The code has been developed on a mobile GTX 1050 GPU. CUDA compute capabilities per videocard can be found here:
https://developer.nvidia.com/cuda-gpus

Data
Some measurement parameters need to be known in advance:

- The width of the point spread function (the sigma of the 2D Gaussian PSF)
- The modulation pitch needs to be known within a reasonable range.
- Modulation patterns are assumed to be mostly in X or Y, with small angle variation. The order of X,Y is estimated automatically.
- Pixel size
- Camera gain/offset

The steps below assume the data zip has been extracted in a data directory that should be in the same path as "simflux.py". The TIFF file path is then "data/sim4_1_MMStack_Pos0_merge.ome.tif"

Installation steps
Step 1. Download and install Anaconda for Windows 64 bit
https://www.anaconda.com/distribution/
Direct link: https://repo.anaconda.com/archive/Anaconda3-2018.12-Windows-x86_64.exe

Step 2. Install CUDA Toolkit 10.0
https://developer.nvidia.com/cuda-downloads.

Step 3. Make sure to download and install the latest drivers specific to your GPU.
The drivers included in the CUDA Tookit are not sufficient.
https://www.nvidia.co.uk/Download/index.aspx?lang=en-uk

Step 4. Disable the automatic windows driver reset (if the graphics driver takes more than a few seconds windows will reset it), by using Disable_CUDA_Driver_Timeout.reg

Step 5.
Create a virtual environment for simflux:
You should have an "Anaconda Prompt" somewhere in the windows apps now.
Open this anaconda prompt and run the following to create an anaconda environment named simflux:
```
conda create -n simflux anaconda
conda activate simflux
```

Step 6.

Using the open anaconda prompt, install python and anaconda packages:
```
conda install -c anaconda cudatoolkit
pip install scipy matplotlib numpy tifffile tqdm
```

Step 6.
Using the anaconda prompt, go to the SIMFLUX directory where simflux.py is located.

Step 7.
Run SIMFLUX. Passing no arguments will show the help:
```
python simflux.py
```

The example dataset can be processed with the following (if copied to a directory named data):
```
python simflux.py data/sim4_1_MMStack_Pos0_merge.ome.tif --sigma
1.83 --gain 2.2 --offset 100 --threshold 10
```

Step 8.
The results will be stored in {input directory}/results/{tif name}/*.
The HDF5 files contain localizations that can be opened in Picasso Render
(https://github.com/jungmannlab/picasso)

Each result directory contains:
simflux.hdf5 - SIMFLUX localizations
g2d.hdf5 - Conventional SMLM localizations

Command line arguments

--patterns: Set number of modulation patterns. Default is 6
--sigma: Set PSF width in pixels (sigma of 2D Gaussian), default is 2 pixels.
--gain: Set gain, default is 1. The photon count is computed as follows:
      Photons = (PixelValue – offset) / gain
--offset: Set camera offset.
--roi: Set size of fitting window, default is 10.
--threshold: Set detection threshold. Default is 10.
--pixelsize: Set pixelsize in nm. Default is 65nm.
--minpitch: Set minimum pitch in nm. Default 200nm.
--maxpitch: Set maximum pitch in nm. Default 250nm.
--minfilter: Set SIMFLUX on/off transition filter [photons], defaults to 30
--fixdepth: Fix modulation depth instead of estimating, for example: 0.95. Disabled by default
--fixshift: Fix phase shifts instead of estimating, in [degrees]. Disabled by default.
--numframes: Number of frames to process. Default is all frames.