Light Field Imaging Plugin for napari

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**Project Purpose: (limited to one sentence)**

To port and adapt existing python code into napari to acquire, process, and display light field images, and to further enhance the code for processing light field images using deep learning approaches.

**Project/Work Plan: (limited to 750 words, currently 762 words)**

This project builds on existing python code for processing light field images, typically recorded on a microscope. The code, called lfanalyze, was developed in the laboratory of Marc Levoy at Stanford University between 2010 and 2013 and handles light field images of fluorescently labeled, transparent specimens or similar objects whose optical properties are characterized by a single parameter associated with volume elements (voxels) in object space (e.g. fluorescence density or absorption coefficient). We have the permission of the original code developer, Michael Broxton, and of Marc Levoy to port their code from python 2.7 to python 3.9 and adapt it as a plugin to napari, providing functions that generate optical sections of an object and projections of it along different viewing directions.

In addition to the port, we will enhance the code by including functions for processing light field images using deep learning and other neural network approaches, such as LFMNet. LFMNet is an existing python framework for "Learning to Reconstruct Confocal Microscope Stacks from Single Light Field Images" [GitHub, refs]. LFMNet was developed by Josue Page, who is now at the Technical University in Munich and who agreed to support the inclusion of the LFMNet framework in napari plugins for light field imaging. As a further benefit of this inclusion, we will refine the interface between napari and the HDF5 data file format.

A light field image represents a four-dimensional raster array of light intensities recorded in the image plane of a camera or microscope. Two dimensions of this array represent the positions {X,Y} in the image plane and an additional two dimensions represent the direction angles {Theta, Phi} in such a way that the value of each array element represents the intensity of light rays that are impinging at a position {X,Y} and from a direction {Theta,Phi} at that position. A conventional optical image averages over all direction information, so a light field image encodes substantially more information. As has been shown by Marc Levoy and collaborators, a light field image of a 3-dimensional object, recorded as a single snapshot, can be converted into either an array of projections of the object along different viewing directions, or a sequence of optical sections, akin to focus stacks of the object [refs].

From the start, napari was designed to handle multi-dimensional image arrays such as light field images and is therefore uniquely suited as a platform for this up-and-coming imaging mode. Currently, light field imaging is used where fast, volumetric imaging is required, such as recording nerve activity in brain tissues [refs]. Research laboratories, which are developing and applying light field imaging, typically develop their own software or are using basic light field processing functions that are available on commercial platforms such as MATLAB [refs]. A well-designed series of napari plugins for light-field imaging, available in the public domain, will be a catalyst for new uses of this promising new experimental tool.

Our long-term goal, beyond the scope of the project described here, is combining polarization analysis with light field imaging. Over the last decade, we have combined light field imaging and polarization analysis on microscope stations in our lab at MBL. By combining polarization analysis available with the OpenPolScope components [openpolscope.org] and a light field camera, we can already capture the required images to fully characterize the birefringence, dichroism, and polarized fluorescence of specimens, including the 3D orientation of the optic axis (alignment axis) in every resolved specimen point. What is still missing, however, are reliable and fast processing algorithms to solve the inverse problem for this imaging mode. While the forward problem, inferring the image from a known object, has a straightforward physical solution, the inverse problem, i.e. inferring an unknown object from its recorded light field image, has resisted a unified, analytical solution, in part because it is an ill-posed and non-linear mathematical problem.

For the next 3 years the PI and Co-PI have received NIH funding to crack this nut, and they have been joined by the graduate student Geneva Schlafly, whose initial work will be porting python code to napari plugins, thereby familiarizing herself with light field imaging. For much of her time, Geneva will be located at the University of Chicago, but funds from the CZI grant, if awarded, are earmarked for her travel and room and board while working at MBL with the PI, where she will acquire experimental skills and refine the napari plugins for experimental work. We ask for additional support for Grant Harris, who is a consultant on the project and mainly focused on moving the LFMNet code onto the napari platform.

**Milestones and Deliverables: (limited to 500 words, currently 288 words)**

We plan to build functionality around light field imaging for napari by achieving the following milestones and deliverables:

1. Port the existing lfanalyze code, which runs under python 2.7, to python 3.9.

2. Create one or more Jupyter Notebooks as an initial user interface for reading and writing light field data in the HDF5 file format and for enlisting upgraded lfanalyze methods for the processing of identified light field images. napari will be integrated into this initial user interface for displaying and analyzing raw light field images and their processed results.

3. A parallel effort will create a Notebook-based or equivalent user interface for training a neural network (LFMNet) using existing experimental data of light field images and simultaneous confocal image stacks of fluorescently labeled brain tissue. Again, napari will be integrated for the display of image data before and after processing.

4. We will then create plugins for napari that implement a graphical user interface for some or all functionality encapsulated in the Jupyter notebooks or equivalent python scripts. We plan to design the plugins with the guidance and advice of napari developers, including Talley Lambert of Harvard Medical School, who has already given us some helpful advice.

5. Create GitHub repository (expand)

6. In addition to existing data sets, we will generate new data using our existing light field microscope setup and also computational approaches that simulate light field images of fluorescent and birefringent objects based on ray tracing methods. We are starting to generate data pairs, consisting of simulated objects with known optical properties on a 3-dimensional voxel grid and their light field images. The experimental and simulated data together will be publicly available for training and testing neural network approaches to solving the inverse problem in light field imaging.