ONLINE FEATURE TRACKING FOR TIME LAPSE IMAGING IN LIGHT SHEET MICROSCOPY

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For biological time lapse imaging it is important to keep the area of interest in the observation field of view over the complete time course. Due to the nature of a developing organism and depending on the length of the time course, the location and shape will change and move out of the observation area. Furthermore, it is often difficult to predict where a particular area of interest will move to in order to continue imaging it. One solution is to image much larger area than necessary to make sure the area of interest stays in the observation volume all the time. However, since this would mean higher light doses as well as longer acquisition times this is not always an option. We developed a way to overcome this limitation and automatically follow a feature of interest without increasing light exposure or extending acquisition time during a time course of several hours where the feature of interest would have been out of the field of view within an hour.

The method has been implemented on an upright scanning light sheet microscope allowing for fast and gentle imaging and could be incorporated in various microscope types where the imaging volume can be moved in three dimensions either by moving the sample or the optics. To determine and follow the feature of interest different algorithms have been implemented and the functionality has been used for time lapse imaging of multiple samples. Due to the fast imaging possibilities of light sheet microscopy the image data has to be downscaled in order to for the automatic tracking to be completed in time to not interfere with the next image acquisition.

The implemented automated feature tracking allows for multi-day tracking of physically varying samples with minimal intervention by the user.