# Measuring and Visualizing Clonal Development in Live Cell and Tissue Microscopy

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Time-lapse microscopy of live cell and tissue captures "5-D" movies of living cells and subcellular organelles. These 5-D image sequences consist of 2 or 3 spatial dimensions plus time and multiple imaging channels captured under different conditions of disease and development. Optical imaging allows the long term observation of complex biological events including the development of clones (family trees) of stem or cancer cells. This data captures cellular patterns of motion, morphology, and appearance and clonal, or lineage, information such as cell cycle times and progeny fate. There is also the opportunity to visualize the dynamics of sub-cellular organelles, all in the intact tissue microenvironment. This rich and complex view of the dynamic processes of disease and development requires computational tools that enable rigorous quantification and also allow human exploration and interaction with the data[1].

We have developed computational image analysis approaches for "summarizing" 5-D image sequence data using denoising, segmentation, tracking and lineaging algorithms. The LEVER (Lineage Editing and Validation) software tools are an open-source NIH funded application suite designed to enable efficient segmentation, tracking and lineaging of 5-D time-lapse microscopy image sequences [2-4]. LEVER is able to process hundreds of movies with thousands of cells and millions of segmentations and works with both phase and fluorescence microscopy. While there is still a need for customized segmentation algorithms, LEVER includes general-purpose approaches to tracking and lineaging [5], together with inference-based learning approaches for incorporating human knowledge. Any errors in the automated processing are easily identified and quickly corrected. LEVER extracts clonal (lineage tree) properties of the data including cell cycle timing and fate commitment from the image data and also from immunohistochemistry or other sources. LEVER also extracts cellular properties including the patterns of motion, morphology, texture, and co-localization from the image data. These cell and clone features can be analyzed using new techniques in algorithmic information theory [6]. For 3-D images, LEVER includes GPU accelerated visualization and analysis directly from MATLAB.

Visualization plays a key role in the analysis, allowing the user to explore the full range of image data interactively with the analysis. A new tool called CloneView, built on the HTML 5 framework, makes *all* of the image and summarization results available on any modern computing device – game changing advances for publishing and reproducibility. CloneView uses the analysis results to guide multi-resolution visualization of the image data. Image data and results from 2-D and 3-D time-lapse microscopy with multiple fluorescence channels has been processed and visualized from a wide variety of applications involving human and mouse stem and cancer cells.

One of the key aspects of this work is making available not only the source code, but also providing visualization tools to show the image data together with the analysis results. An example of this can be found at the CloneView archive from a recent paper: http://n2t.net/ark:/87918/d91591. This website shows all of the data from the paper, all in an interactively exploreable format.

The web link above for the CloneView website is a special archival link intended to be embedded in a publication. This link is referred to as an 'ark', and is managed by the Univ. of California libraries. Similar to a DOI link, these ark links are intended as permanent object identifiers so that the link integrity can be maintained permanently. If the data needs to be moved or renamed (e.g. due to server configuration changes) the link destination can be modified.

The source code for the LEVER and CloneView are available free and open-source under the GPL version 3 license. This allows all users the freedom to use the software for any purpose, the freedom to change the software to suit their needs, and the freedom to redistribute the software, including any changes. LEVER uses the MATLAB environment for some functionality, and can be run directly from within MATLAB if that software is available. In addition to the source code, pre-built stand-alone executables for the Windows 10 operating system are also available. All software is available at https://git-bioimage.coe.drexel.edu.

References:

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