

Supervised Feedback Microscopy with the Acquirer IM and Plate-Viewer

Modern high-content screening microscopes allow rapid automated imaging of entire microtiter plates by imaging fixed positions within each well. This is ideal for in-vitro cell culture-based readouts or other assays with evenly distributed phenotypes. However, it imposes limitations when large specimens or rare events are studied because the limited field of view (FOV) of high-magnification objectives may not encompass the full region of interest. Therefore, users are often limited to lower magnification acquisition, leading to low-resolution data, or omitting features of interest in many wells. This technical note discusses how Bruker's Acquirer Imaging Machine (IM) overcomes these challenges with Plate-Viewer software, and offers a semi-automated approach for advanced supervised feedback microscopy experiments.

Automated Experimental Approaches

For tissue-specific imaging in large specimens (e.g., zebrafish) an approach is needed that can automatically identify and zoom in on the tissue or organ of interest. Fully automated tissue detection and imaging often demand the development of sophisticated image processing routines. Therefore, each project needs careful balancing between software development time and project size. Semi-automated approaches offer an ideal compromise, as they only require minimal user interaction and no custom algorithm development. Even complex or variable structures that would require extensive development of image detection routines can be readily handled via this simpler alternative. This approach is beneficial for screening projects as they start instantly, saving time and resources.

Plate-Viewer Software

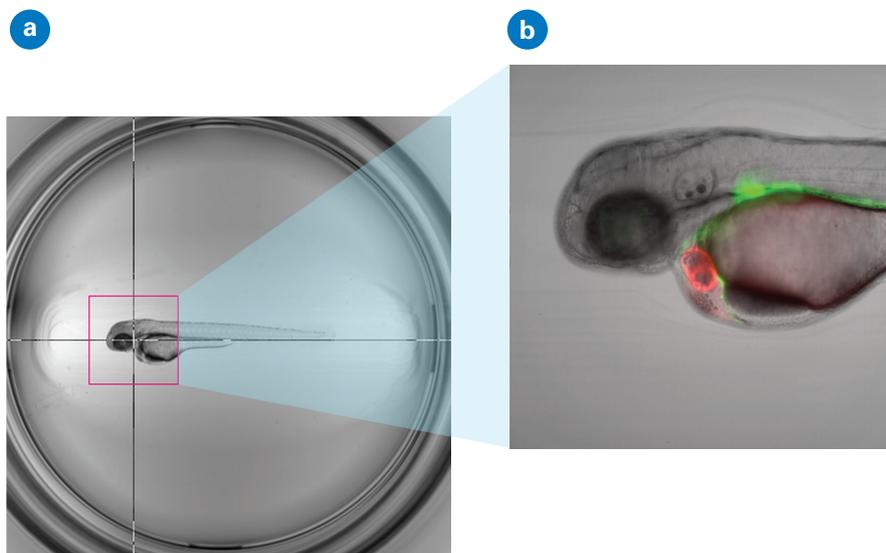


FIGURE 1. Illustration of Click-Tool functionality. (a) Zebrafish embryo imaged with a 2x objective and visualized in Plate-Viewer. The crosshair is centered on the heart region in a three-day-old embryo of the epi:GFP;myl7mR transgenic line. The red bounding box indicates the field of view of a 10x objective used for subsequent high-resolution imaging. (b) Single Z-plane of a high-resolution dataset is automatically acquired on Acquirer IM.

Plate-Viewer software offers a semi-automated approach for supervised feedback microscopy. Low-magnification pre-screen data of a full microtiter plate is visualized following the plate layout for a quick and intuitive overview. The integrated “click-tool” functionality allows assay experts to select regions of interest (ROIs) for each well. Moreover, built-in and generic “template matching” algorithms enable robust automatic localization of various target structures. These can be complex reporter expression patterns, morphological features, or rare events in each well. Acquirer IM automatically acquires data at high resolution from selected regions according to predefined settings and acquired high-resolution data is then readily and intuitively visualized in Plate-Viewer.

Plate-Viewer has several unique features that enable researchers to:

- Select ROIs to be automatically imaged by a simple mouse click or template matching;
- Preview FOVs of higher magnification objectives using an adjustable bounding box;
- Intuitively visualize and browse even large-scale screening datasets;
- Readily navigate through complex multidimensional datasets;
- Apply LUTs and overlay channels for improved manual inspection;
- Adjust basic image parameters to enhance visualization of details; and
- Plug-in interface for integration of external image processing tools.

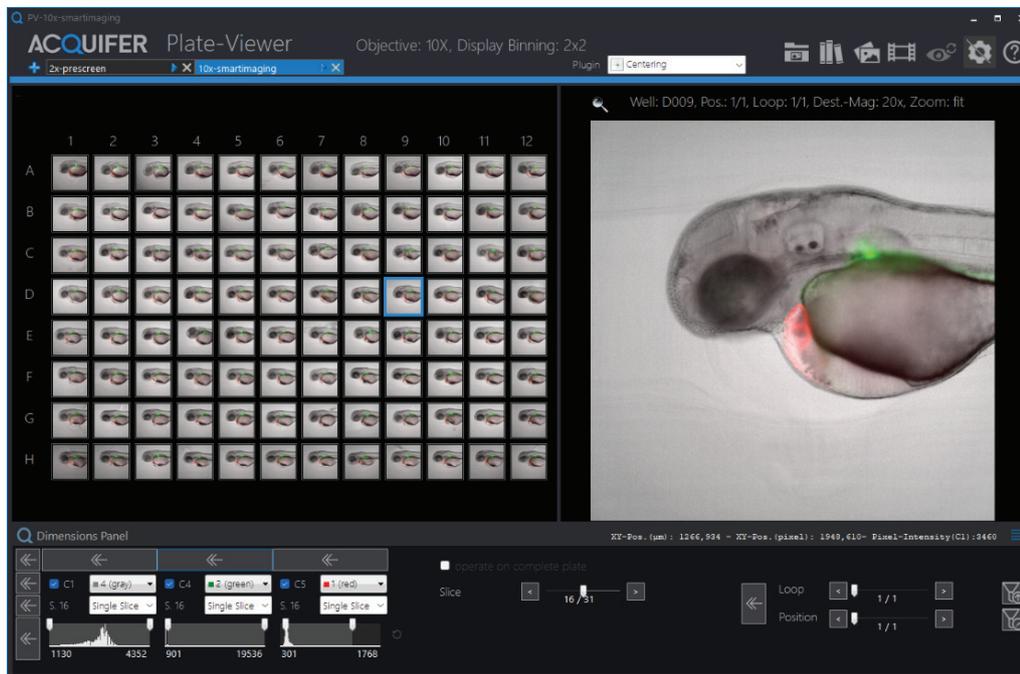


FIGURE 2. Overview of the Plate-Viewer functionalities. Visualization of identical planes of all available wells within a microtiter plate (left). Preview of the currently selected well coordinate (right). Control panels in the top right corner enable the generation of colored overlay images and the display of selected channels. Control panels in the lower left corner allow channel-specific adjustment of the histogram and the choice of navigation mode through multidimensional datasets (e.g., slice, loops, or subpositions).

Intuitive Solution for High-Content Screening

Acquifer IM is a complete solution for supervised feedback microscopy experiments. Its Plate-Viewer software utilizes a semi-automated approach to greatly simplify complex high-content screening experiments. This innovative tool enables researchers to more easily and reliably perform tissue-specific imaging of complex or variable structures in large specimens.

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