SeBaLIT™ Multiplexing Technology

Abstract

Multiplex staining and imaging in fluorescence microscopy empower researchers to observe multiple cell structures and processes concurrently. Laxco's SeBaLIT™ multiplexing technology enhances insights into cellular localization, protein-protein interactions, and dynamic changes within cells by addressing limitations inherent in traditional fluorescence imaging methods.

Impact on Live Cell Fluorescence Imaging

The Laxco SeBaLIT Multiplex Imaging System represents a substantial leap forward in fluorescence imaging. By enabling live full-color, multi-channel imaging in real-time, it overcomes limitations associated with traditional methods, such as pseudo-coloring and the need for stacking monochrome images.

This real-time, full-color imaging approach greatly enhances both clarity and inter-pretability, making it an ideal solution for complex workflows across various fields, including cell biology, pathology, and neuroscience. Moreover, the system's capacity for dynamic, in vivo observation supports more effective studies of cellular processes and interactions, offering researchers deeper insights with unparalleled immediacy.

SeBaLIT™ Technology Innovation

Laxco's patented fluorescence modules integrate wavelength-specific excitation LEDs with a single multi-bandpass filter, significantly reducing illumination degradation commonly associated with traditional mercury light sources. This innovative design allows for each LED to be individually controlled with the push of a button to activate any combination of channels required by the user.

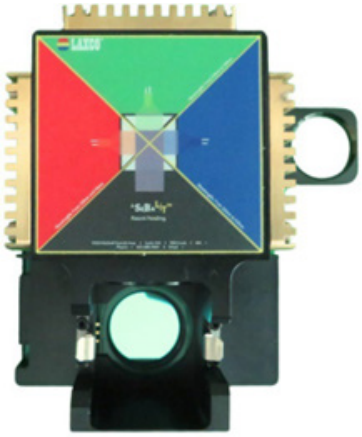

Figure 1: SeBaLIT™ Fluorescence Module Top View

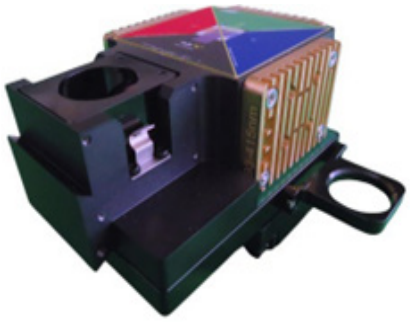

Figure 5: SeBaLIT™ Fluorescence Module Side View

SeBaLIT™ Technology Innovation (cont'd)

- **Standard SLi3PRO Module (M1)**: This M1 fluorescence light module supports the most commonly used fluorochromes like DAPI, FITC, and TRITC; with a spectral range of 395 nm to 640 nm and compatible with over 70 different dyes.

- **Advanced SLi6PRO Modules (M2 and M3):** These fluorescence light modules are equipped with wavelength-specific emission filters to optimize single color detection and enhance image clarity across a broader spectrum of fluorochromes; including those in the far-red, enabling compatibility with over 110 different dyes.

    - The M2 Module supports the standard dyes (DAPI, FITC, and TRITC) includng extended detection further into the red spectrum with dyes like M-Cherry, Mito-Tracker Red, Texas Red, and CY3; with a spectral range of 390 nm to 647 nm.

    - The M3 Module is optimized to enhance specialized dyes providing detection into the cyan, yellow and far-red spectrum allowing dyes like CFP, YFP, Lucifer Yellow RFP, Alexa Fluor 594, and CAL Fluor 590/610; with a spectral range of 418 nm to 662 nm.

- **User-Friendly Design:** These plug-and-play modules are designed for easy swapping in the field, allowing users to replace or upgrade without needing assistance from a service technician.

- **Crosstalk Reduction:** To achieve publication-quality fluorescence imaging across three channels, crosstalk reduction is critical. This is accomplished through our specialized filter systems tailored to minimize overlap between fluorescence emissions.

- **Digital Crosstalk Reduction Techniques:** Implemented to enhance real-time imaging capabilities and improve image clarity.

Advantages of Live Multiplex Imaging

- **Real-time Multi-channel Imaging:** This allows for accurate representation of fast biological events, eliminating delays from sequential imaging. This capability captures rapid cellular events, offering insights into dynamics that were previously difficult to observe.

- **Live Cell Video and Time-Lapse:** The system enables simultaneous video recording and time-lapse imaging on a single screen, providing precise temporal parameters tailored to experimental needs.

- **Enhanced Accuracy:** By reducing artifacts associated with time delays and vibrations, simultaneous imaging enhances precision in comparing fluorescent signals and determining cell component orientations.

- **Improved Workflow Efficiency:** The elimination of filter changes and repetitive imaging streamlines workflows, allowing for high-throughput experiments and more productive lab sessions.

Advantages of Live Multiplex Imaging (cont'd)

- **Easy to Use:** User-friendly hardware with push-button controls and fully integrated intuitive software simplify microscope operation for seamless multiplex fluorescence imaging.
- **Publication-Quality Images:** The SeBaLIT™ system produces high-quality images suitable for publication (See Figures 3, 4, and 5).

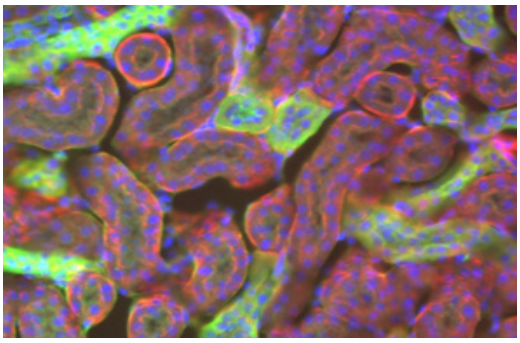

Figure 3: SliPRO Series Microscope Multiplex Image of Mouse Kidney, cells stained with MitoTracker Red CMXRos (Mitochondria), 40x magnification

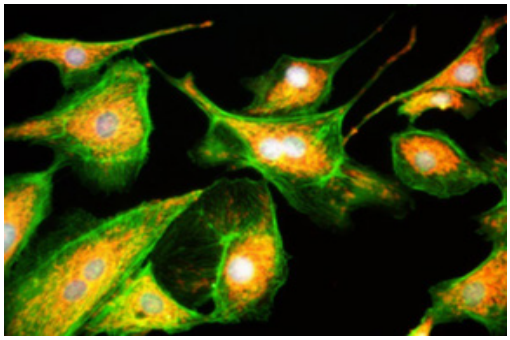

Figure 4: SliPRO Series Microscope Multiplex Image of BPAE cells stained with MitoTracker Red CMXRos (Mitochondria), AlexaFluor 488 phalloidin (F-Actin), DAPI (nuclei), 40x magnification

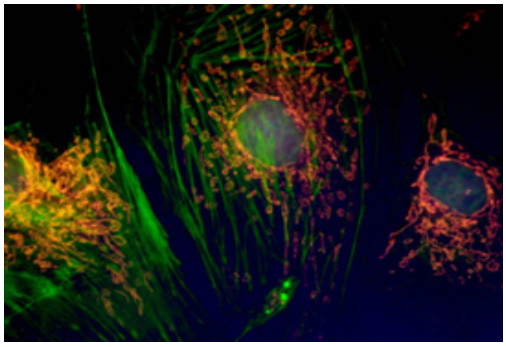

Figure 5: SliPRO Series Microscope Multiplex Image of BPAE cells stained with MitoTracker Red CMXRos (Mitochondria), AlexaFluor 488 phalloidin (F-Actin), DAPI (nuclei), 100x magnification (oil)

Conclusion

Laxco's SeBaLIT™ Live Multiplex system represents a major leap forward in multiplex fluorescent imaging. Its capability for simultaneous imaging across multiple channels not only boosts temporal resolution, while also significantly enhances accuracy and efficiency. These advancements are crucial for researchers investigating live specimens, allowing for the capture of rapid biological processes with remarkable clarity. As technology progresses, innovations like SeBaLIT™ will further broaden the horizons of fluorescence imaging in biological research, paving the way for new discoveries and insights.