Odyssey® CLx Near-Infrared Imaging System + Data Integrity™ Bundle

The Odyssey CLx Imager comes with the Data Integrity Bundle. The Data Integrity Bundle includes resources like training, protocols, and software to help you meet publisher requirements for Western blotting, including validation experiments, normalization, and analysis of replicates. With the Odyssey CLx and the Data Integrity Bundle, you will be confident that your results are reliable, impactful, and publishable.

Antibody Validation

- Antibody validation training on the Lambda U™ training portal
- Workflow for antibody validation in Empiria Studio Software

Linear Range Determination

- The Odyssey CLx provides 6 logs of linear dynamic range, so you can accurately capture strong and faint bands in the same image
- Protocol for determining the linear range for Quantitative Western blot detection
- Training for determining the linear range for Quantitative Western blot detection on the Lambda U training portal
- Workflow for analyzing linear range determination experiments in Empiria Studio Software to find the correct amount of sample to load for an accurate experiment

Housekeeping Protein Validation

- Protocol for housekeeping protein validation
- Housekeeping protein validation training
- Workflow for analyzing housekeeping protein validation experiments in Empiria Studio Software

Replicate Analysis

- Protocol for Quantitative Western blot analysis with replicate samples
- Replicate analysis training on the Lambda U training portal
- Workflow for analysis of replicate samples and replicate blots in Empiria Studio Software

Training Resources

- Quantitative Western blot training, including theory, mechanics, and best practices
- Empiria Studio Software training
- Advanced training

Total Protein Stain Normalization

- Multiplex fluorescent detection with the Odyssey CLx
- Protocol for total protein stain normalization
- Total protein stain normalization training on the Lambda U training portal
- Guided workflow for total protein stain normalization in Empiria Studio Software

Post-translational Modification Normalization

- Multiplex fluorescent detection with the Odyssey CLx
- Protocol for post-translational modification normalization
- Post-translational protein modification normalization training on the Lambda U training portal
- Workflow for post-translational modification normalization in Empiria Studio Software

Housekeeping Protein Normalization

- Multiplex fluorescent detection with the Odyssey CLx
- Protocol for housekeeping protein normalization
- Housekeeping protein normalization training on the Lambda U training portal
- Workflow for housekeeping protein normalization in Empiria Studio Software

Share Results

- Empiria Studio Software makes it easy to share quantification data, statistical results, and images with colleagues and reviewers in the formats that publishers require
- Image Studio Software makes it easy to share images with colleagues and reviewers in formats that publishers require

Additional Options

REVERT™ Total Protein Stain enables detection of all the protein in a lane for accurate and reliable normalization. High quality primary antibodies, validated on Odyssey Imaging Systems, enable reliable detection of housekeeping proteins.

Odyssey CLx Imager

Although a new imager is only part of the solution for reliable quantitative Western blot results, the Odyssey CLx is the standard for quantitative Western blot imaging. Odyssey instruments have been cited in over 10,000 scientific publications, because of their unique technology and capabilities.

Unlike traditional detection methods, the Odyssey CLx uses a patented direct-detection system featuring two spectrally separate solid state lasers to detect infrared dye-labeled conjugates and near-infrared (NIR) stains. With this unique detection system, you will be able to see all the detail and complexity of your Western blot, plate-based assay, or tissue section in a single image. You can be confident that your findings will be meaningful and that your data meet the emerging rigor of journal requirements.

Note:

LI-COR Biosciences is the sole manufacturer, vendor, and service provider for the Odyssey® CLx Near-Infrared Imaging System. The system is sold exclusively in the United States and Canada by LI-COR Biosciences, Lincoln, NE. This product is not represented or sold in the United States or Canada by any distributors or other third parties.

Wide Linear Dynamic Range

The Odyssey CLx provides a 6-log linear dynamic range, allowing you to capture strong and weak bands from the same blot in a single digital image. Because all the signal from a blot is recorded in a single image, you can accurately compare bands without the error prone process of capturing and comparing multiple exposures. A detection system with a wide linear dynamic range provides the basis for accurate quantification and helps you meet increasingly stringent publisher requirements.

For example, publishers are beginning to require normalization to an internal loading control (ILC). Normalization corrects for lane-to-lane variation in loading and transfer. For accurate normalization, you must load an amount of sample that allows your target and ILC to be detected within the same linear range (if a combined linear range exists). The wide linear dynamic range of the Odyssey CLx captures the full range of target and ILC signal and will help you determine if there is a combined linear range for your target and ILC, which will enable accurate normalization that will pass an editor's scrutiny.

Two-Color Detection and Quantification

The Odyssey CLx can detect multiple protein targets in each sample lane (called multiplex detection), with great sensitivity in both fluorescence channels. Multiplex detection accelerates throughput and unlocks accurate normalization techniques that comply with publication requirements for Western blot data.

Normalization corrects for lane-to-lane variation in loading and transfer by comparing your target to an internal loading control (ILC). By using a second color to detect the ILC, you can normalize without variation caused by stripping and re-probing the blot. Multiplex detection on the Odyssey CLx is compatible with normalization against ILCs that publishers recommend for normalization.

High Sensitivity

Even subtle shifts in protein expression can be a meaningful discovery, so the Odyssey CLx was designed for high sensitivity near-infrared detection. Near-infrared detection allows you to see your targets with low background interference from autofluorescence and light scatter.

Low-background detection expands the range of signals that can be quantified, compared to traditional methods. A wider range of quantifiable signals makes it easier to determine if your target and internal loading control (used for normalization) can be detected in the same linear range. Determining this linear range is a crucial step in the type of normalization procedures publishers are beginning to require.

Direct Detection

No film, darkroom, or messy substrates are required when working in the near-infrared channels. Near-infrared dyes are stable indefinitely on membranes, as long as membranes are properly stored.

Hardware Specifications

- Simultaneous, two-channel detection must be possible for multiplex sample analysis and/or normalization.
- For each channel, a dedicated and optimized laser and detection system must be provided.
- Two infrared, solid-state laser diodes emitting at 685 nm ±5 nm and 785 nm ±5 nm, respectively, must be the means of excitation of the fluorescent labels.
- Detection system must be based on single point detection by cooled Avalanche Photodiodes (APDs), which are optimal for ensuring high quantum efficiency of the detected signal in the NIR range.
- Laser lifetime must exceed 40,000 hours of operation to keep maintenance costs low.
- Detection of the fluorescent dyes must be in the 710 nm - 730 nm and 810 nm - 830 nm ranges, for the 700 nm and 800 nm channels respectively, in order to obtain a high signal-to-noise ratio. No excitation/ detection thresholds below 700 nm will be accepted.
- Automatic image capture that offers a dynamic range of greater than 6 logs and optimal image acquisition in a single capture.
- Laser/Microscope must be integrated into a precision scanning mechanism capable of scanning at resolutions of 21 - 337 µm.
- Data output must be an individual 16-bit TIFF file format for each channel of detection.
- The system must have a minimal footprint to save lab space.
- Imager should be compatible with a Verification Plate that allows you to test the consistency of the imager over time to ensure that performance does not vary.
- The system must utilize a sealed, flat-bed scanning surface to accommodate varying sample media (membranes, multi-well plates, gels, small animals, slides, etc.).

Acquisition Software

Image Studio™ Software must be able to control and capture images on the Odyssey CLx. Software is available for PC and Mac.

- Multiple display modes will allow you to view strong and faint bands captured in the same image, without altering underlying data. A minimum of three display modes must be available: colored overlay, colored single channel, and single channel grayscale.
- Images must be exportable as high resolution TIFF files that conform to common journal requirements, PNG and JPEG formats that can be easily shared in slide presentations and in other digital media, and in a format compatible with Empiria Studio software.
- Data must be exportable to widely used formats, including text, spreadsheets (compatible with common spreadsheet software such as Excel), and PDFs.
- Images and data must be easy to organize and store for later retrieval, such as if raw data are requested during a journal's review process.

Analysis Software

Empiria Studio™ Software must be available for analysis of images captured on the Odyssey CLx. Software is available for PC and Mac.

- For analysis that meets publisher requirements, quantification must be performed on raw images and must not be affected by image display.
- For accurate and complete analysis that meets publisher recommendations for reproducible results, analysis software must provide workflows for Antibody Validation, Housekeeping Protein Validation, Combined Linear Range Detection, Normalization (HKP, total protein stain, post-translational modification), and Replicate Analysis.
- Features should be available in a single software package.
- Features should be available in straightforward workflows to minimize user-to-user variability and check for missed steps.
- Results, images, and all supporting data should be able to be exported in a single file that can be imported into Empiria Studio by other users anywhere the software is installed.

Open Platform for Multiple Applications

The Odyssey CLx has been used extensively (and published in many peer-reviewed journals) for applications including, but not limited to, Western blots, EMSA, protein arrays, In-Cell Western™ Assay, On-Cell Western Assays, *in vivo* imaging, Coomassie gel documentation, DNA gel documentation, and tissue section analysis.

- Two-color quantitative and highly sensitive membrane-based Western blot analysis with signals being simultaneously detected at 700 nm and 800 nm for ultimate sensitivity.
- Two-color quantitative detection of In-Gel Western Assays for the immediate detection of proteins prior to blotting.
- Simultaneous 2-channel detection of infrared-labeled or NIR-stained nucleic acids should be possible using either agarose or PAGE gels.
- Quantitative detection of target molecules (typically proteins) in cell culture grown in 6- to 384-well microtiter plates. Quantitative analysis must be possible for all target molecules or of surface-exposed target molecules. Normalization of the target molecule by the cell number must be possible by simultaneous and quantitative detection of a second target protein or quantitative detection of nucleic acids.
- Two-color quantitative and highly sensitive detection of protein arrays with a spot size >150 µm on nitrocellulose coated glass slides must be possible.
- EMSA detection and quantitative signal analysis using NIR-labeled DNA oligos must be possible in two channels simultaneously without the time-consuming need to disassemble the gel / plate sandwich.
- Two-color tissue section analysis must be possible at a resolution of 21 µm at 700 nm and 800 nm to minimize autofluorescence of the tissue by near- infrared fluorescence imaging.
- Near-infrared *in vivo* detection of fluorescently probed targets in small animals like mouse or rat must be possible to provide a common platform for *in vitro* and *in vivo* experiments.
- Highly sensitive and quantitative detection of Coomassie stained protein gels must be possible.
- Compatible with MousePOD® Imaging Accessory for small animal imaging.
- A maximum sample area of 25 x 25 cm must be available.

Reagents

- Available reagents must include fluorescent IRDye infrared dyes for labeling antibodies, antibody fragments, peptides, proteins, oligonucleotides and DNA fragments for emissions higher than 700 nm.
- Reagent offerings should include a NIR solution for the detection of HRP-conjugated antibodies.
- IRDye infrared dyes must be available for direct infrared detection of samples on membranes, gels, glass, and plastic support, or within small animals and organs.
- The system must be able to scan for two different IRDye infrared dyes with emissions separated by at least 100 nm to eliminate spectral overlaps.
- No enzyme substrates, films, and darkrooms will be used to detect the signals.
- Reagents must be available for protocols that can assess if housekeeping protein (HKP) expression is affected by experimental treatments. According to journal requirements, HKPs should not be used for normalization unless evidence can be provided that HKP expression is unaffected by experimental conditions
- A total protein membrane stain (TPS) must be available to enable normalization to total protein loading, which is the preferred normalization method according to publisher requirements. The TPS must wash off to enable normalization, along with accurate two target detection.
- IRDye fluorophores must be very stable over several months and should provide the option to be rescanned with equivalent results.

Service and Support

Knowledgeable professionals will be readily available to provide training and to answer questions.

- **Installation, an on-site training session, and a one-year warranty for parts and labor will be included with purchase.** Training will cover proper use of the instrument and guidance on how to gather data in a way that meets current publication standards. Additional support contract options are available for purchase within one year of purchasing the instrument.
- **Technical Support** will be available to service the instrument, as well as to answer questions related to hardware, software, and applications. Professionals will be available for on-site visits or can be reached by phone, email, or through chat on LI-COR's website.

Resources

- **Publications** that reference the Odyssey CLx, as well as other LI-COR products, will be readily findable in a publications database on LI-COR's website.
- **Technical Resources**, such as comprehensive product instructions and application guides, will be readily available for reference to educate team members on best practices for gathering accurate, publishable data.
- **Online Western Blot Training Courses** from Lambda U™ learning portal must be available to ensure all members of a lab are trained consistently to obtain high quality Western blot results that meet publisher requirements.

Patents

Odyssey CLx Near-Infrared Imaging System

U.S. Patent 6,495,812, Apparatus and method for intersecting a light beam and a focal point of a detector at an object of interest. Issued August 2, 2000.

IRDye Infrared Dye Reagents

1. U.S. Patent 6,027,709, Fluorescent Cyanine Dyes. Issued Feb. 22, 2000.
2. U.S. Patent 6,995,274, Cyanine Dyes. Issued Feb. 7, 2006.
3. U.S. Patent 7,005,518, Phthalocyanine Dyes. Issued Feb. 28, 2006.
4. U.S. Patent application Serial #11/267643, Cyanine Dyes. Filed Nov. 4, 2005.
5. U.S. Patent application Serial #11/419457, Optical Fluorescent Imaging. Filed May 19, 2006.

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