



BD™ Pathway Bioimager

Explore more. Discover more.

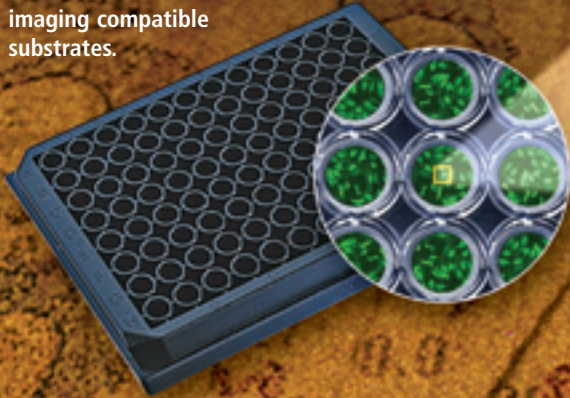


BD

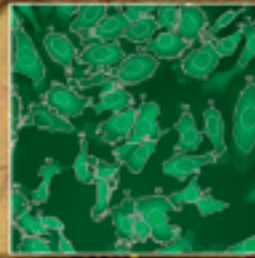
BD Biosciences

Biological Exploration with the BD™ Pathway Bioimager

1. Cells are plated into multiwell plates, chamber slides, or onto other imaging compatible substrates.



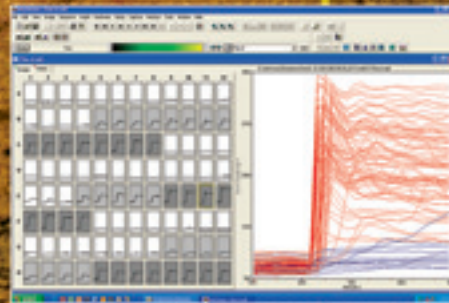
2. Images are captured in either confocal or widefield modes to provide the best possible images for analysis. Proprietary software algorithms then segment the image allowing measurements to be made within individual cells.



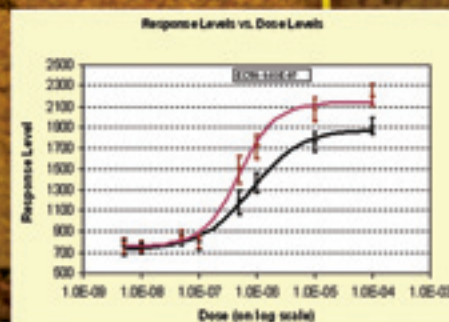
3. The cellular images may then be further divided into regions of interest (ROI) where fluorescence measurements are made. These measurements can include fluorescence intensity ratios, granularity, morphological features, fluorescence localization and more.



4. Individual cells may then be classified into different categories based on end-point or kinetic response profiles at the well or plate level.



5. Data may be visualized within the AttoVision software, or analyzed within the BD™ Image Data Explorer software – a custom Microsoft® Excel® based data analysis tool designed for single cell high content analysis. Data can also be exported for analysis within 3rd party software solutions.



Features and Benefits

Key Features

- True confocal real-time imaging produces outstanding images with reduced background fluorescence.
- Full-spectrum laser-free illumination gives you flexibility to work with almost any fluorescent dye.
- Ability to run kinetic and endpoint assays on one instrument with easy conversion of endpoint assays to kinetic applications.
- Integrated liquid handling with image-as-you-add capability for initial rate kinetic studies.
- Multiple slide and multiwell plate formats can easily be imaged.
- Environmentally controlled imaging chamber enables live cell experimentation.
- Flexible software allows easy data navigation and classification.
- Integrated binocular viewing – in both confocal and widefield modes.
- High precision linear x, y, and z motors.
- Motionless stage allows imaging of suspension or loosely adherent cells.

Features

Sub-second kinetics, time-lapse, or endpoint with image-as-you-add capabilities.

Full-spectrum, white light illumination.

True Confocal, Laser-free system.
Proven Nipkow system can be moved in or out of the lightpath.

Sophisticated image segmentation and novel boolean classification quickly and easily categorize cellular responses.

On-the-fly analysis.
See responses as they unfold.

Direct viewing of sample.
Integrated binocular viewing port allows direct sample evaluation.

Ultra-precise x,y,z positioning with motionless stage.
Objective moves with 100 nm precision in x and y (50 nm in Z).

Environmental control.
Temperature and CO₂ regulated environmental chamber.

Liquid handling. Single-channel pipettor allows image-as-you-add liquid addition and mixing at any position of the imaging plate.

Benefits

Select timepoints that best fit your assays.
Measure ion kinetics, translocations, or single-point assays on a single machine.

Work with a greater range of fluorescence dyes.
No laser maintenance and replacement cost.
Multiplex a broader range of dyes.

Develop 3D images and z-stacks quickly.
Run homogeneous assays without interfering fluorescence background.
Fast image acquisition time.

Configurable software enables easier integration of your assays.
Lower software costs.

No waiting for results – watch the data in real-time
Monitor assay performance without losing time or reagents

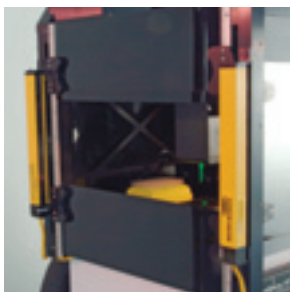
Use the system as a stand-alone confocal microscope for a “quick-look”.
Run a quality check on the data by visual inspection.
View samples in true color.

Revisit cells that have been imaged.
Capture montaged images of samples larger than the field of view.
Work with suspension or loosely adherent cells.

Work with living cells over long periods of time without needing to move plate from stage.

Allows detection of rapid events occurring immediately following compound addition without imaging lag.

The BD™ Pathway Bioimager at a glance



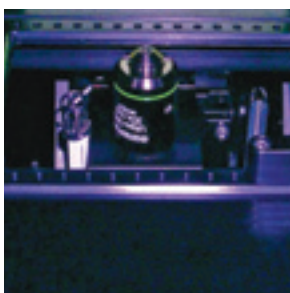
Robotic Automation

Full automation is enabled through the addition of a plate handling robot that places assay plates, compound plates, and pipet tips onto the imaging stage. The robotic scheduling software seamlessly integrates with the BD Pathway platform software.



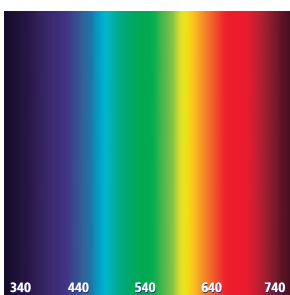
Liquid Handling

Using disposable tips, the Pathway Bioimager can add drugs or compounds for assay development, lead profiling and receptor kinetics studies. Multiple drug additions can be made during the imaging process without moving the sample.



Stationary Stage

The BD Pathway Bioimager features a proprietary motionless stage that ensures sample stability during the image acquisition process – even for non-adherent cells.



Full-spectrum Illumination

Two separate Mercury Arc lamps (100W each) illuminate the sample through one of up to 16 different excitation filters. The spectrum of the lamps spans the UV to the infrared allowing imaging of a wide variety of dyes (e.g. Fura-2 and Cy5™) in the same cell. Since virtually any filter combination is possible, true multi-color multiplexing is straight forward to set up and execute.





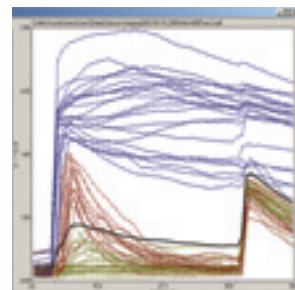
Environmental Control

The environmentally controlled chamber (temperature and CO₂) enables live cell imaging from fast-ion kinetic measurements to extended time-lapse imaging.



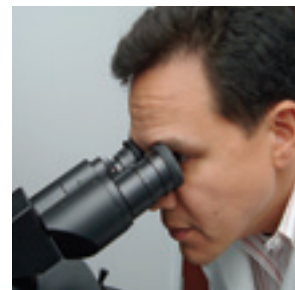
AttoVision Software

Flexible software for both kinetic and endpoint experimentation provides a broad range of acquisition and analysis tools. The intuitive and customizable software provides for both ready-to-run and fully customizable applications.



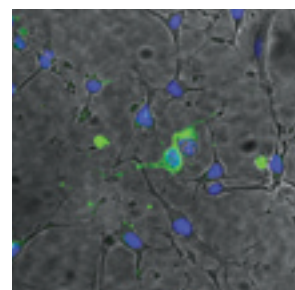
Binocular Eyepieces

The BD Pathway Bioimager is the only system in its class with eye-pieces for direct viewing of cells in both fluorescence and transmitted light modes. The unique spinning disk technology permits direct visualization of confocal images.



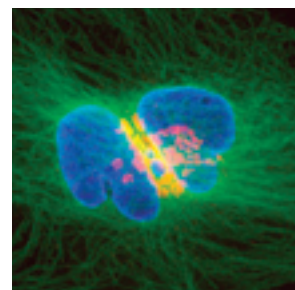
Transmitted Light

Transmitted light images can be superimposed onto fluorescence images to assist with localization of cellular events.



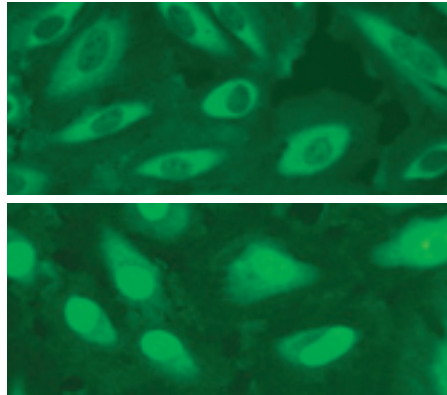
True Confocal Optics

The patented spinning disk confocal technology provides real-time imaging capabilities at virtually any wavelength. The confocal optics deliver high resolution images without background fluorescence often associated with standard wide-field imaging systems.

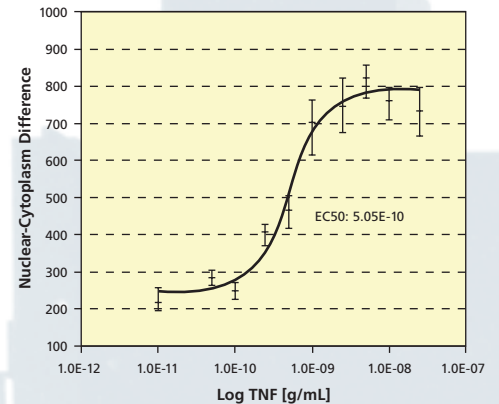


Applications

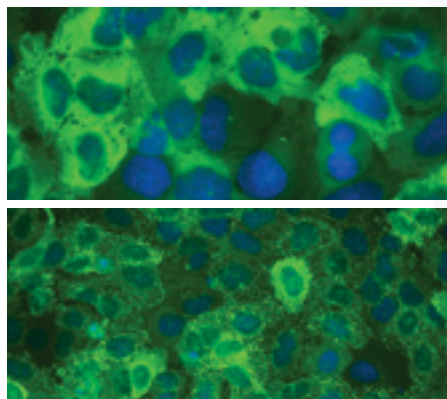
NFκB – Translocation of the transcription factor NFκB is indicative of receptor activation and is used to research inflammatory signaling pathways. In the example, HeLa cells were treated with TNF (50 ng/mL), fixed and antibody-stained. The proprietary image analysis algorithm isolates the cells from their background and quantifies the fluorescence intensity in the cytoplasm and nucleus. Either ratiometric or subtractive analysis between the nuclear and cytoplasmic intensities reveals the degree of protein translocation during drug stimulation. The flexibility of the algorithms ensures that multiple assay types can be analyzed with equal quality.



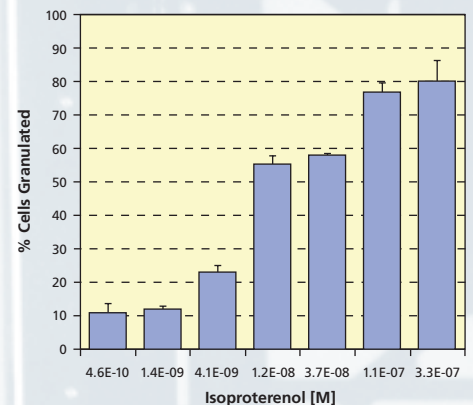
(Top image) NFκB-FITC-Cytoplasm-Control. (Bottom image) NFκB-FITC-Cytoplasm-TNF3



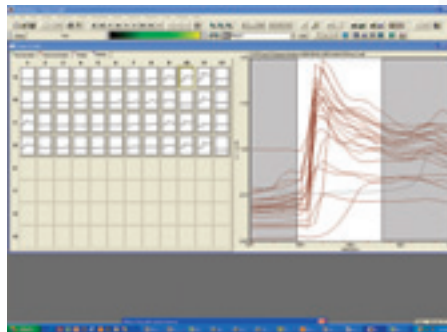
Transfluor® – The Transfluor® technology from Norak Pharmaceuticals™ is used widely to quantify GPCR activation by measuring aggregation and eventual internalization of GFP-labeled β-arrestin. The BD Pathway Bioimager uses a novel granularity algorithm which minimizes user input and works reliably under many different experimental conditions.



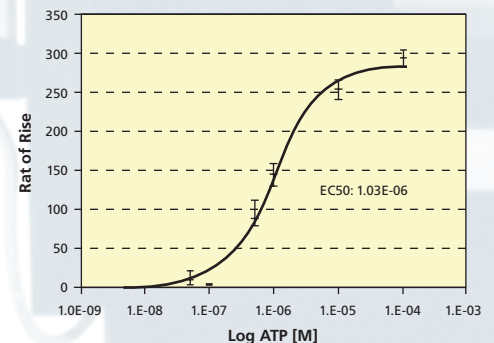
Following receptor activation, the evenly distributed β-arrestin-GFP construct (top image) rapidly accumulates in distinct vesicles prior internalization (bottom image).



[Ca²⁺]_c – Cytosolic Ca²⁺ concentration ([Ca²⁺]_c) can be measured efficiently using the ratiometric Fura-2™ probe. Alternate illumination with 340nm and 380nm by two separate mercury light sources ensures ultra rapid acquisition and analysis of [Ca²⁺]_c. The highly flexible and intuitive software tools enable the researcher to distill large kinetic datasets into concise information.



(Left image) Example of Fura 2 individual cell kinetic traces for multiple treatment zones with corresponding plate map. (Right image) Kinetic data is easily summarized as a dose response curve (average well Rate of Rise as a function of increasing concentration of ATP).



Additional Application Examples

These are representative applications that have been successfully run on the BD Pathway. Other applications are possible and are continually being developed.

Cytotoxicity

- Live vs. dead cell measurements
- Nile Red hepatotoxicity

Apoptosis

- Annexin V
- PARP
- Cleaved Caspase
- JC-1
- YO-PRO®

Mitotic Index

- Anti-phospho Histone H3

Translocations

- NFκB
- PKC-α
- Norak Transfluor®

Neurite Outgrowth

- CSIRO Neurite outgrowth algorithm

Kinetic

- Ca²⁺
- Na⁺
- cAMP (BD™ ACTOne)

Whole-organism and tissue imaging (confocal, 3D)

- *C. elegans*
- Zebrafish
- Arabidopsis
- Yeast

Software

Image Acquisition

The powerful and flexible image acquisition software of the BD Pathway Bioimager is designed to handle virtually any kinetic and end-point imaging situations. Pre-set applications are initiated quickly through the user interface. Designed for a multi-user environment, advanced users have full access to the object oriented macro builder enabling even the most demanding imaging-based experiments.

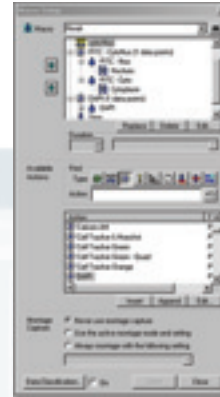
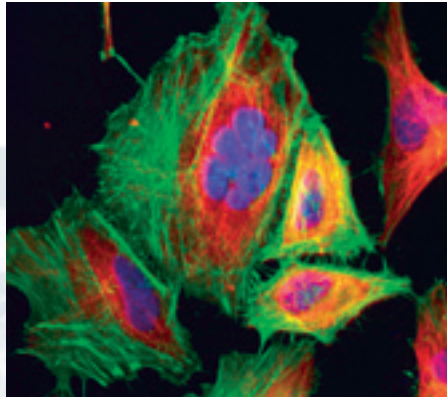
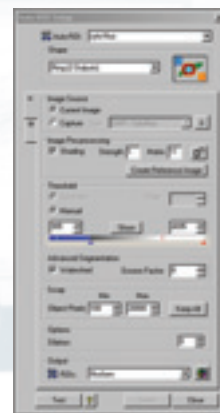
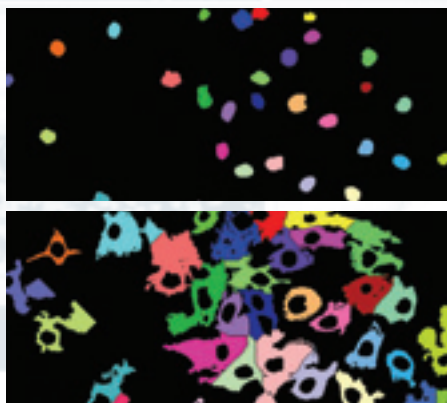


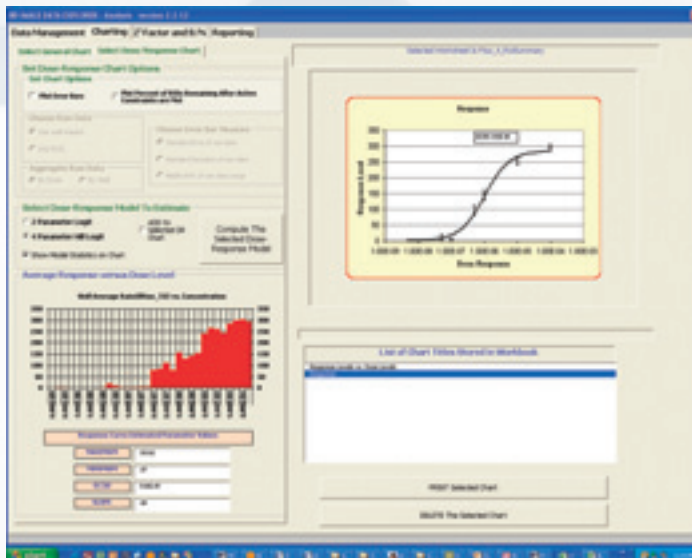
Image Analysis

The Pathway Bioimager comes “fully loaded” with image analysis routines included with the software at no extra cost. Measurements of protein localization, fluorescence intensity and granularity are achieved through the flexible AttoVision software. Image analysis occurs in real-time and the data is visualized throughout the experiment. Reducing complex kinetic datasets into manageable information is a key strength of the software.



Data Analysis

Designed for both endpoint and kinetic imaging, the BD Pathway Bioimager software is equipped with tools to analyze cellular responses with high spatial and temporal resolution. The BD Image Data Explorer is a unique Add-In for Microsoft® Excel® enabling complex pharmacological investigation in a truly cell-by-cell mode. Drawing from Microsoft® Excel®'s strengths, the BD Image Data Explorer generates Z', S/N, EC₅₀, dose-response curves, bar charts and more using an intuitive interface



Specifications

Imaging Platform

- Proprietary, fully integrated optical design
- Motionless stage handles 96 or 384 well plates, microscope slides, and chamber slides
- X, Y resolution: 100 nm (mechanical)
- Z resolution: 50 nm (mechanical)
- 5 images per second capture rate
- Works with Olympus 4X, 10X, 20X, 40X, 60X objectives
- Integrated temperature (ambient to 40° C) and CO₂ (ambient to 5%) control
- Montage capability for large field imaging
- Automation ready: see Robotics Integration Package

Confocal Unit

- Disk sampling rate: 100 fps
- Operation mode: Confocal fluorescence, widefield fluorescence, brightfield
- Confocal technique: Nipkow spinning disk
- Pinhole size: 70 µm
- Illumination: White light, laser free
- Excitation: 340 nm to near-IR
- Light source: Dual HBO 100W mercury arc lamps
- White-light source for brightfield microscopy

Filter configuration (user specified)

- Excitation: 16 filter positions
- Dichroic: 5 filter positions
- Emission: 8 filter positions
- All filter wheels are independent and automated to maximize filter combinations

Detection/Observation

- Internal high resolution Hamamatsu ORCA ER cooled CCD camera
- Binocular eyepiece for direct sample viewing in confocal or widefield mode

Liquid Handling

- On-stage pipet head with single-position syringe (2 – 100 µl)
- Mixing capability (titration)
- Disposable pipette tips

Software

- Automated focus and image acquisition
- Automated segmentation and region of interest (ROI) identification
- Interactive data and image navigation
- Concurrent multiple ratiometric dye kinetics (US Patent 5,332,905)
- Z sectioning and 3D rendering
- Novel hierarchical data classification algorithm
- Data analysis module for Microsoft® Excel® (Optional)
- Preconfigured and user configurable applications
- Supports kinetic and endpoint applications
- BMP, TIFF and TXT format data export to popular analysis packages

Computer (minimum specification)

- Pentium IV running Windows XP Professional
- 1 GB RAM
- 250 GB SATA Hard Drive
- DVD burner
- 23" wide aspect ratio LCD flat panel display

Robotics Integration Package – Optional

- Access door with safety light curtain
- Software interface to multiple scheduling systems

Physical Dimensions

- Height: 66" (168 cm)
- Width: 43" (109 cm) – 44" (112 cm) with optional robotics integration package
- Depth: 41" (104 cm) including keyboard shelf

Note: Specifications represented may include optional components. Specifications subject to change. For most updated specification, please contact BD Biosciences.

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All applications are either tested in-house or reported in the literature. See Technical Data Sheets for details.

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