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# Automated imaging and high-content analysis



# Automated imaging and high-content analysis

## A complete portfolio of imaging platforms, reagents, and software tools for high-content analysis

High-content analysis requires high-quality microscope images and multiple channels of data. When you incorporate automation into data collection and analysis, you increase the number of samples you can process and the amount of biological data you can acquire. Automated microscopy and analysis provide the foundation for the entire field of highcontent analysis, while capabilities like environmental control and integrated fluidics expand the range of questions you can ask. See how EVOS<sup>™</sup> imaging systems and Thermo Scientific<sup>™</sup> high-content analysis platforms can help you take your research to the next level with automated imaging and analysis to increase your throughput and data content.

Take advantage of the entire spectrum of Molecular Probes<sup>™</sup> fluorescent reagents matched to the channels on your imaging system. See how functional probes and labeling strategies can be multiplexed to create high-content assays that answer your research questions (page 11).

|                        | EVOS <sup>™</sup> FL<br>system | EVOS <sup>™</sup> FL Auto<br>system | CellInsight <sup>™</sup> CX5<br>platform | CellInsight <sup>™</sup> CX7<br>platform | ArrayScan <sup>™</sup><br>module |
|------------------------|--------------------------------|-------------------------------------|--|--|----------------------------------|
| LED light cubes        | Х                              | X                                   |  |  |                                  |
| LED light engine       |                                |                                     | Х  | 7-channel                                | 7-channel                        |
| Monochrome camera      | Х                              | Х                                   | Х  | Х  | Х                                |
| Color camera           | Optional                       | Х                                   |  |  |                                  |
| Widefield fluorescence | 4-channel                      | 4-channel                           | 5-channel                                | 7-channel                                | 7-channel                        |
| Brightfield            | Х                              | Х                                   | Х  | Х  | Х                                |
| Phase contrast         | Х                              | X                                   |  |  |                                  |
| Confocal               |                                |                                     |  | 7-channel                                | 6-channel                        |
| Objective turret       | 5-position                     | 5-position                          | 1-position                               | 3-position                               | 4-position                       |
| Autofocus              |                                | Software                            | Software                                 | Laser                                    | Laser                            |
| Live-cell chamber      |                                | Optional                            |  |  | Optional                         |
| Automated scanning     |                                | Х                                   | Х  | Х  | Х                                |
| Robotics compatible    |                                |                                     | Х  | Х  | Х                                |
| Automated fluidics     |                                |                                     |  |  | Optional                         |
| Software               | EVOS FL                        | EVOS FL Auto                        | HCS Studio                               | HCS Studio                               | HCS Studio                       |
| Page                   | 3                              | 4                                   | 5  | 6  | 7                                |

## EVOS FL Imaging System

Form, function, and flexibility for fluorescence imaging

Manual imaging for multichannel fluorescence, brightfield, and phase-contrast microscopy

- Exceptional ease of use
- Wide range of sample types
- Multichannel overlay
- Automatic cell counting and time-lapse imaging



| Hardware     |   |
|--------------|---|
| Illumination | Select from 14 adjustable-intensity LED light cubes             |
| Camera       | High-sensitivity CCD (choice of monochrome or color)            |
| Widefield    | 4-channel fluorescent imaging (wide choice of wavelengths)      |
| Brightfield  | White light brightfield and phase-contrast imaging              |
| Objectives   | 5-position objective turret (1.25x–100x range, low and high NA) |
| Stage        | Wide range of slide, plate, and vessel holders available        |
| Software     | Manual cell counting, time-lapse, and transfection measurement  |

## EVOS FL Auto Imaging System

## An intuitive, affordable fully automated system

Multidimensional and time-lapse imaging, tile stitching and Z-stack automation

- Exceptional ease of use
- Touch screen software control
- Automated X/Y scanning stage, autofocus, and flat-focus Z-stack
- Precise environmental control for live-cell imaging



| Hardware          |   |
|-------------------|---|
| Illumination      | Select from 14 adjustable-intensity LED light cubes                       |
| Cameras           | Dual monochrome (CCD) and color (high-sensitivity CMOS) camera            |
| Widefield         | Automated 4-channel fluorescent imaging                                   |
| Brightfield       | Automated white light brightfield and phase-contrast imaging              |
| Objectives        | Automated 5-position objective turret (1.25x-100x range, low and high NA) |
| Focus             | Software-based autofocus  |
| Live-cell chamber | Environmental control with EVOS <sup>™</sup> Onstage Incubator            |
| Automation        | Automated multi-well plate scanning                                       |
| Software          | Automated cell counting, time-lapse, image tiling and stitching           |

## CellInsight CX5 High Content Screening Platform

Compact, affordable screening system for everyday use

Fast scanning and quantitative analysis for high-content screening

- Quantitative fluorescence and brightfield imaging
- Full automation of plate handling and scanning for high-throughput screening
- Compact footprint for lab bench or robotic line
- Thermo Scientific<sup>™</sup> HCS Studio<sup>™</sup> software provides a comprehensive suite of analysis tools



| Hardware     |   |
|--------------|---|
| Illumination | 5-channel solid-state LED light engine                          |
| Camera       | Photometrics X <sup>™</sup> camera                              |
| Widefield    | 5-channel fluorescent imaging                                   |
| Brightfield  | White light brightfield imaging                                 |
| Objectives   | Single-position objective (2x-20x range, low and high NA)       |
| Focus        | Software-based autofocus  |
| Automation   | Configured for fully automated plate handling and scanning      |
| Software     | HCS Studio software for integrated data collection and analysis |

## CellInsight CX7 High Content Platform

### Integrated system for all-around performance

More wavelengths and integrated confocal optics to expand application areas

- Fully integrated confocal imaging for higher resolution of thick samples
- 7-channel fluorescence and 5-channel brightfield imaging
- Full automation and laser autofocus for high throughput
- HCS Studio software provides a comprehensive suite of analysis tools



| Hardware     |   |
|--------------|---|
| Illumination | 7-channel solid-state LED light engine                          |
| Camera       | Photometrics X1 camera  |
| Widefield    | 7-channel fluorescent imaging                                   |
| Brightfield  | 5-channel brightfield imaging                                   |
| Confocal     | 7-channel confocal imaging                                      |
| Objectives   | 3-position objective turret (2x-40x range, low and high NA)     |
| Focus        | Software- and laser-based autofocus for consistent scan times   |
| Automation   | Configured for fully automated plate handling and scanning      |
| Software     | HCS Studio software for integrated data collection and analysis |

## ArrayScan High Content Platform

## Modular system for specialized applications

Modular design allows addition of confocal scanning, brightfield, and live-cell options

- Add confocal imaging for higher resolution of thick samples
- Add environmental control for long-term kinetic measurement of live cells
- Configured for full automation and high throughput
- HCS Studio software provides a comprehensive suite of analysis tools



| Hardware          |   |
|-------------------|---|
| Illumination      | 7-channel solid-state LED light engine                          |
| Camera            | Photometrics X1 camera  |
| Wide field        | 7-channel fluorescent imaging                                   |
| Bright field      | White light brightfield imaging option                          |
| Confocal          | 6-channel confocal imaging option                               |
| Objectives        | 4-position objective turret (1.25x–63x range, low and high NA)  |
| Focus             | Software and laser-based autofocus for reduced phototoxicity    |
| Live-cell chamber | Live-cell imaging chamber option                                |
| Liquid handling   | Optional 1-channel aspirate and dispense, read-through imaging  |
| Automation        | Configured for fully automated plate handling and scanning      |
| Software          | HCS Studio software for integrated data collection and analysis |

## HCS Studio cell analysis software

## Intuitive interface and intelligent design

Intelligent data acquisition analyzes just enough cells for statistical relevance

- Icon-based guidance, accessible to novice users
- Fully customizable for experienced users
- Thermo Scientific<sup>™</sup> BioApplications software tools for assay development and screening
- Scalable to many thousands of images





Manager



Plate maps to manage your experimental design.



Icon-based guidance, accessible to novice users.

#### Analysis or screening

Whether you are analyzing a few slides to answer basic research questions or screening thousands of samples in a systems biology study, we can offer you the platform of choice to meet your application.

Go from image collection to tabulated results and population statistics—and back again, because the data always come back to cells.

- Data are seamlessly linked to both image and protocol
- Move from tabulated data to view cells, wells, or fields
- Make decisions grounded in biology



Multiple imaging modes for cells and tissues.



Confocal image acquired with the CellInsight CX7 platform, of spheroid labeled with Hoechst<sup>™</sup> 33342 stain, calcein AM, and ethidium homodimer.







#### **Assay performance**

Tool

Cell

entation

n Red

18 2 22 24 26 28 ect.Shape(WR.Ch]

Label

Detect

Measure

Analyze

Automated

With HCS Studio Cell Analysis Software you can be confident of robust assay performance. Rank your assay parameters based on Z-prime before starting a screen, and then adjust your stopping criteria to collect only the data you need for statistical significance.







## Multiplexing reagents for assay optimization

#### **Recommended reagents**

Drawing on decades of experience in fluorescence imaging, Molecular Probes HCS products are developed using Thermo Scientific high-content platforms with special considerations for the high-throughput workflow and automated imaging:

- Alexa Fluor<sup>™</sup> secondary antibodies for brightness and stablity
- Cell and nuclear masks for automated demarcation
- Robust functional probes for cell health interrogation
- Validated on multiple imaging platforms

Take advantage of the entire fluorescent spectrum to multiplex your assay—and maximize your instrument performance. Use the table below to select reagents for each platform and channel.

| EVOS light cubes                         | DAPI Light Cube                                      | CFP Light Cube              |       |
|--|--|-----------------------------|-------|
| CellInsight CX5 channels                 | Blue   |                             |       |
| CellInsight CX7 channels                 | Blue   | Cyan                        |       |
| ArrayScan channels                       | Blue   | Cyan                        |       |
|  |  |                             |       |
| Secondary antibodies                     |  |                             |       |
| Goat anti-mouse                          | Alexa Fluor 350 (A11045)                             | Alexa Fluor 405 (A31553)    |       |
| Goat anti-rabbit                         | Alexa Fluor 350 (A11046)                             | Alexa Fluor 405 (A31556)    |       |
| Cell segmentation                        |  |                             |       |
| Whole cell segmentation                  | HCS CellMask Blue (H32720)                           |                             |       |
| Nuclear segmentation                     | HCS NuclearMask Blue (H10325)                        |                             | 9     |
| Cell tracking and tracing                | CellTracker Blue (C12881)                            | CellTracker Violet (C10094) |       |
|  |  |                             |       |
| Cell strucuture                          |  |                             |       |
| Cytoskeleton (actin)                     | Alexa Fluor 350 Phalloidin (A22281)                  |                             |       |
|  |  |                             |       |
| Lysosomes                                |  |                             |       |
|  |  |                             |       |
| Mitochondria                             |  |                             |       |
| Cell function probes                     |  |                             |       |
| Cell viability                           |  |                             | In    |
| Cell proliferation                       |  |                             | Click |
| Apoptosis (caspase activity)             |  |                             |       |
| To see more reagents validated for use i | in high-content analysis, go to thermofisher com/hcs |                             |       |



Image acquired with the Cellnsight<sup>™</sup> CX5 platform, of cells labeled with Hoechst 33342 stain and Alexa Fluor<sup>™</sup> 680 phalloidin.



Image acquired with the ArrayScan platform, of cells labeled with DAPI, Alexa Fluor<sup>™</sup> 488 goat anti–mouse IgG, and Alexa Fluor<sup>™</sup> 647 goat anti–rabbit IgG.

| GFP Light Cube | YFP Light Cube | RFP Light Cube |       |
|----------------|----------------|----------------|-------|
| Green          |                | Orang          | e/red |
| Green/yellow   |                | Orange         |       |
| Green/yellow   |                | Orange         |       |

| Alexa Fluor 488 (A11001)                    | Alexa Fluor 532 (A11002) | Alexa Fluor 555 (A21422)                        |             |
|---|--------------------------|---|-------------|
| Alexa Fluor 488 (A11008)                    | Alexa Fluor 532 (A11009) | Alexa Fluor 555 (A21428)                        |             |
|   |                          |   |             |
| HCS CellMask Green (H32714)                 |                          | HCS CellMask Orange (H32713)                    |             |
| YTO 9 Green Nucleic Acid Stain (S34854)     |                          | SYTO 82 Orange Nucleic Acid Stain (S11363)      | Н           |
| CellTracker Green (C7025)                   |                          | CellTracker Orange (C34551)                     |             |
|   |                          |   |             |
| Alexa Fluor 488 Phalloidin (A12379)         |                          | Alexa Fluor 555 Phalloidin (A34055)             | Ale         |
|   |                          |   | Ly          |
| MitoTracker Green FM (M7514)                |                          |   |             |
|   |                          |   |             |
| nage-iT DEAD Green Viability Stain (I10291) |                          |   |             |
| -iT EdU Alexa Fluor 488 HCS Assay (C10351)  |                          | Click-iT EdU Alexa Fluor 555 HCS Assay (C10353) | Click-iT Ec |
| CellEvent Caspase-3/7 Green (C10423)        |                          |   |             |
|   |                          |   |             |



Image acquired with the EVOS FL Auto platform, of cells labeled with Qdot<sup>™</sup> 655 secondary antibody, NucBlue<sup>™</sup> Live ReadyProbes<sup>™</sup> reagent, and ActinGreen<sup>™</sup> 488 ReadyProbes<sup>™</sup> reagent.



Image acquired with the CellInsight CX7 platform, of cells labeled with DAPI, Alexa Fluor<sup>™</sup> 488 wheat germ agglutinin, and Alexa Fluor<sup>™</sup> 568 phalloidin.

| Texas Red <sup>™</sup> Light Cube    | Cy°5 Light Cube                                 | Cy®7 Light Cube          |
|--------------------------------------|---|--------------------------|
|                                      | Deep red  | Near-IR                  |
| Red                                  | Deep red  | Near-IR                  |
| Red                                  | Deep red  | Near-IR                  |
|                                      |   |                          |
|                                      |   |                          |
| Alexa Fluor 594 (A11005)             | Alexa Fluor 647 (A21235)                        | Alexa Fluor 750 (A21037) |
| Alexa Fluor 594 (A11012)             | Alexa Fluor 647 (A21244)                        | Alexa Fluor 750 (A21039) |
|                                      |   |                          |
| HCS CellMask Red (H32712)            | HCS CellMask Deep Red (H32721)                  |                          |
| CS NuclearMask Red (H10326)          | HCS NuclearMask Deep Red (H10294)               |                          |
| CellTracker Red (C34552)             | CellTracker Deep Red (C34565)                   |                          |
|                                      |   |                          |
| xa Fluor 594 Phalloidin (A12381)     | Alexa Fluor 647 Phalloidin (A22287)             |                          |
| soTracker Red DND-99 (L7528)         | LysoTracker Deep Red (L12492)                   |                          |
| MitoTracker Red (M7513)              | MitoTracker Deep Red FM (M22426)                |                          |
|                                      |   |                          |
| Propidium Iodide (P1304MP)           |   |                          |
| U Alexa Fluor 594 HCS Assay (C10355) | Click-iT EdU Alexa Fluor 647 HCS Assay (C10357) |                          |
|                                      |   |                          |
|                                      | ·   | ·                        |



#### Find out more at thermofisher.com/hcs

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