IT'S LIGHTING THE WAY TO A NEW LEVEL OF UNDERSTANDING

Operetta CLS™
High-Content Analysis System

PerkinElmer®
For the Better

HepG2 microtissue stained with Nile Red to study hepatosteatosis, 20x W objective. Cells provided by InSphero AG.

Human cardiomyocytes labeled with the hypertrophy marker proBNP/488, Rhodamine-Phalloidin, Hoechst and CellMaskBlue, 20x W objective. Cor.4U cardiomyocytes provided by Axiogenesis AG.
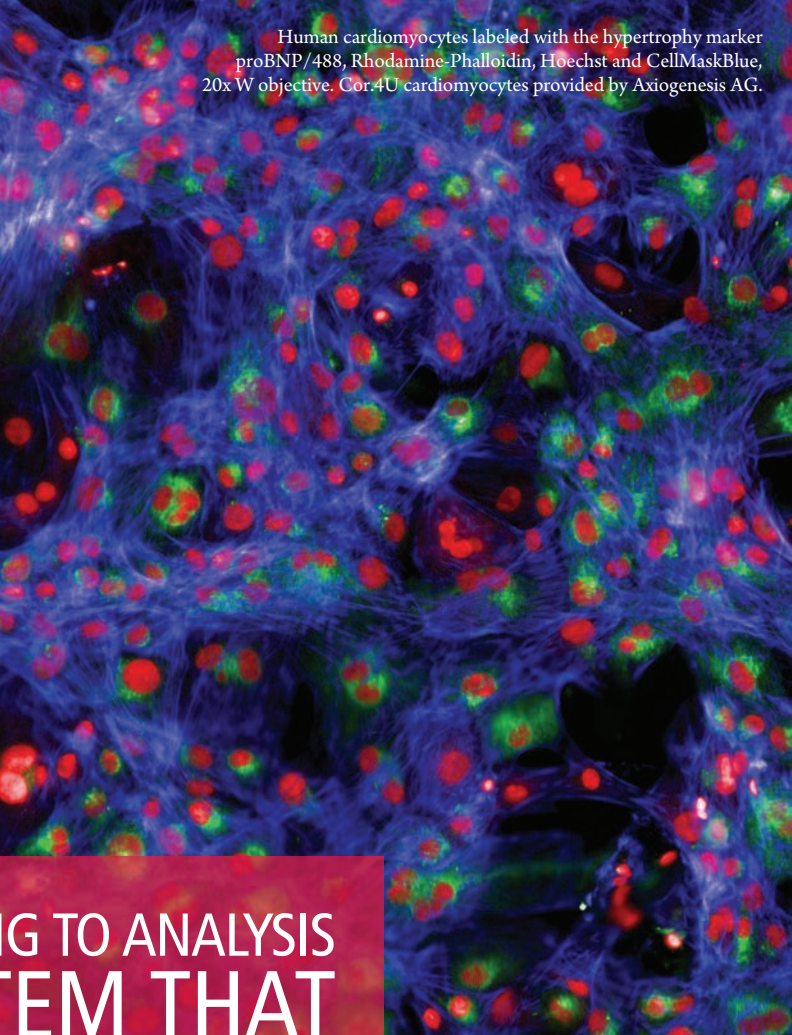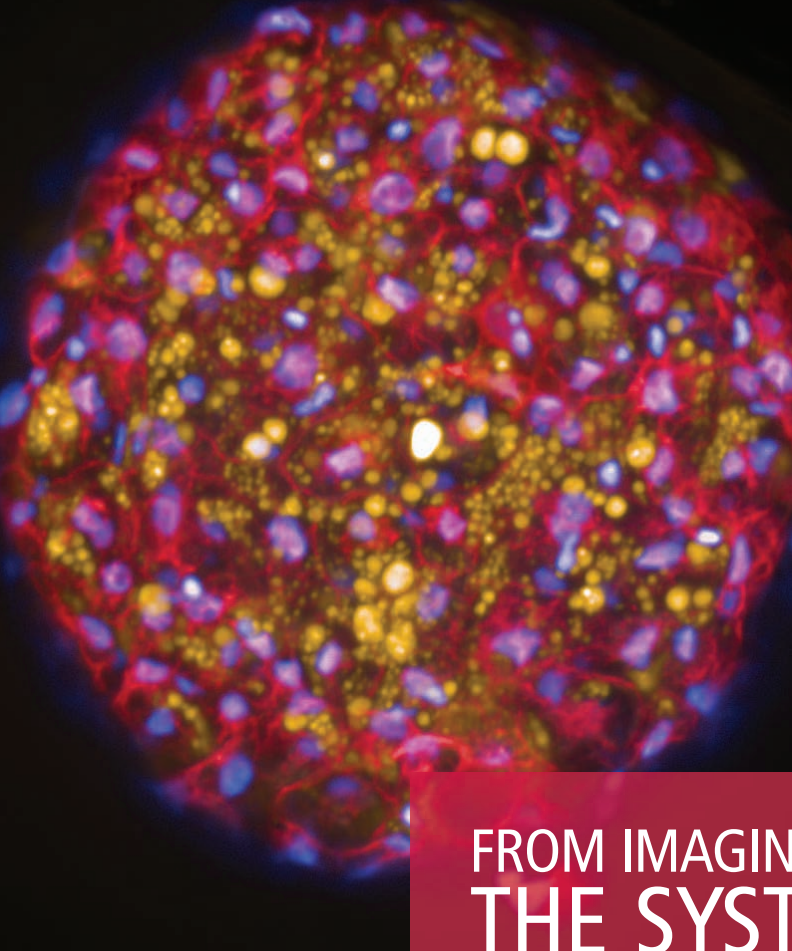
FROM IMAGING TO ANALYSIS
THE SYSTEM THAT
MAKES THE EVERYDAY
EXTRAORDINARY
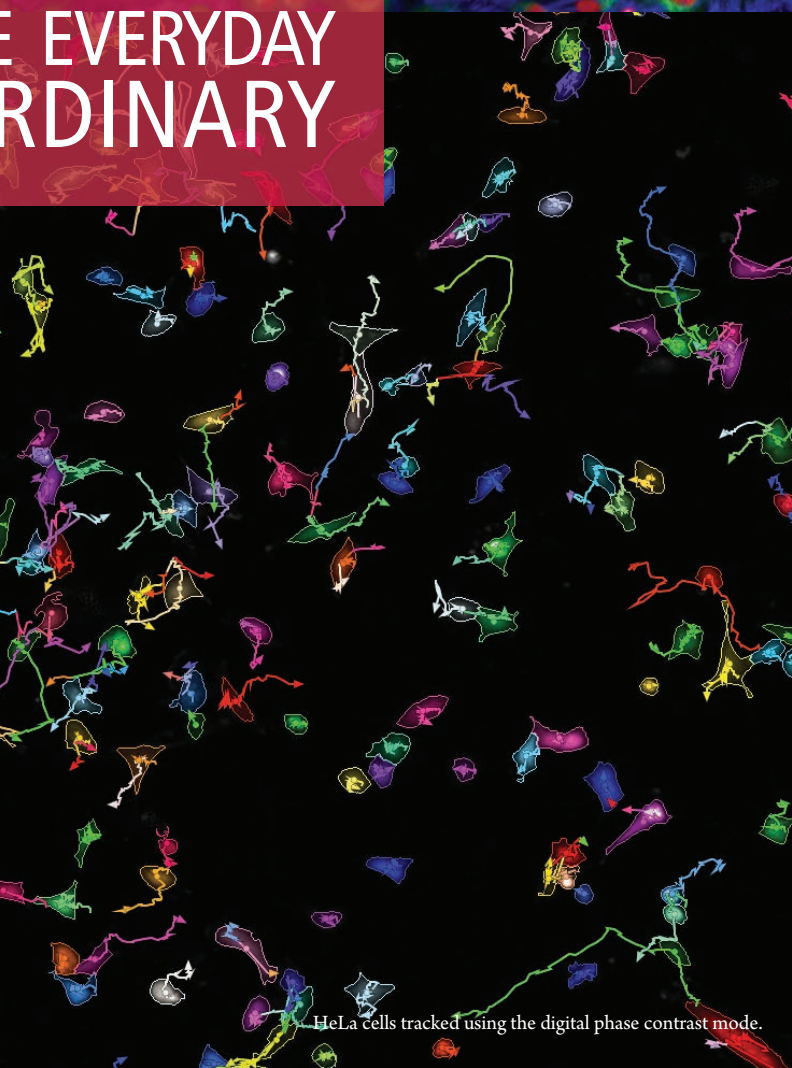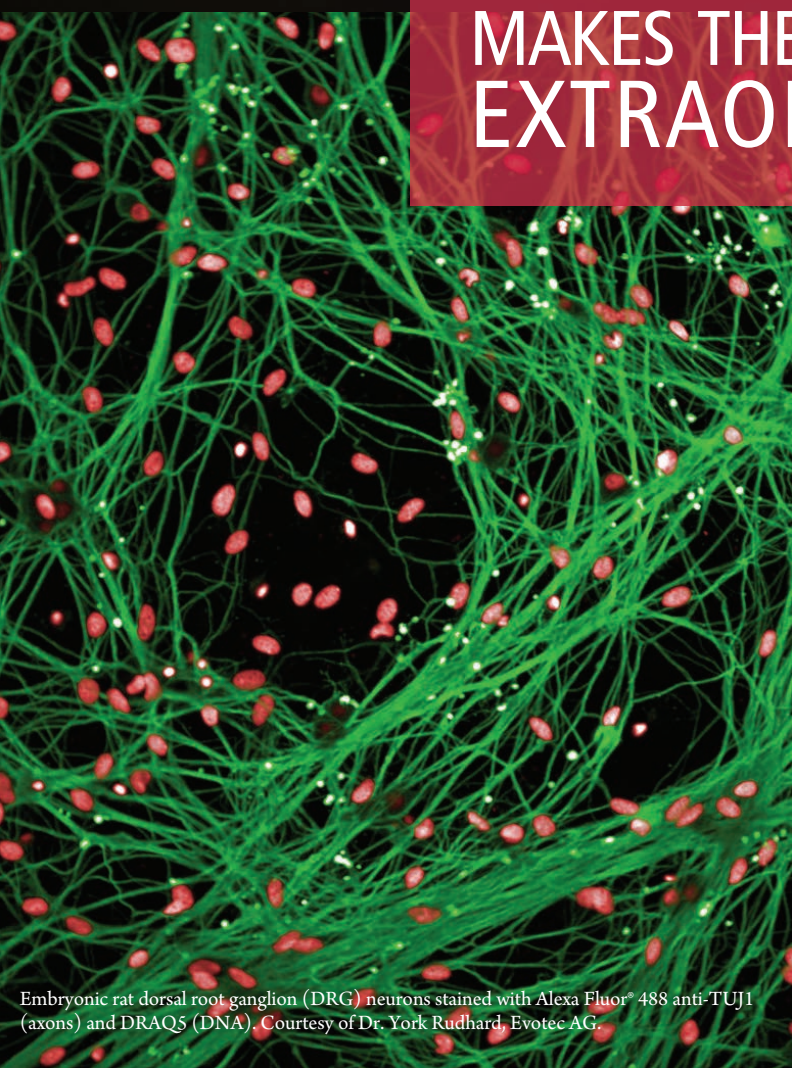
Embryonic rat dorsal root ganglion (DRG) neurons stained with Alexa Fluor® 488 anti-TUJ1 (axons) and DRAQ5 (DNA). Courtesy of Dr. York Rudhard, Evotec AG.

HeLa cells tracked using the digital phase contrast mode.

Everything you've come to rely on (and so much more)

Across nearly every therapeutic area and application in basic research and assay development – whether it's systems biology or drug discovery – there's a growing demand for greater physiological relevance.

Scientists want to study more complex models such as primary cells, live cells, or cells cultured in 3D, and generate detailed phenotypic fingerprints for deeper biological insights. But to gain these insights, you need the throughput to assess varied conditions, the resolution to capture high-quality image data – and the tools to turn that data into knowledge.

Now there's a system that combines speed and sensitivity with the powerful and intuitive data analysis you've come to trust from the Operetta platform: It all comes together in the Operetta CLS system.

The Operetta CLS delivers everything you need from high-content analysis – and more:

- Water-immersion objectives that capture more light than air objectives

- Stable LED light source with up to eight excitation wavelengths for flexible labeling and better results with less background noise

- Confocal and widefield imaging

- A highly sensitive sCMOS camera for a large-field of view and high-resolution capture

- Easy-to-learn-and-use Harmony high-content imaging and analysis software to empower scientists doing their own analysis

What's more, the Operetta CLS system is part of our comprehensive HCS workflow – everything from HCS systems and microplates to automation and informatics for every application. All from one knowledgeable, trusted vendor.

The Operetta CLS system: Everything about it says *extraordinary*.

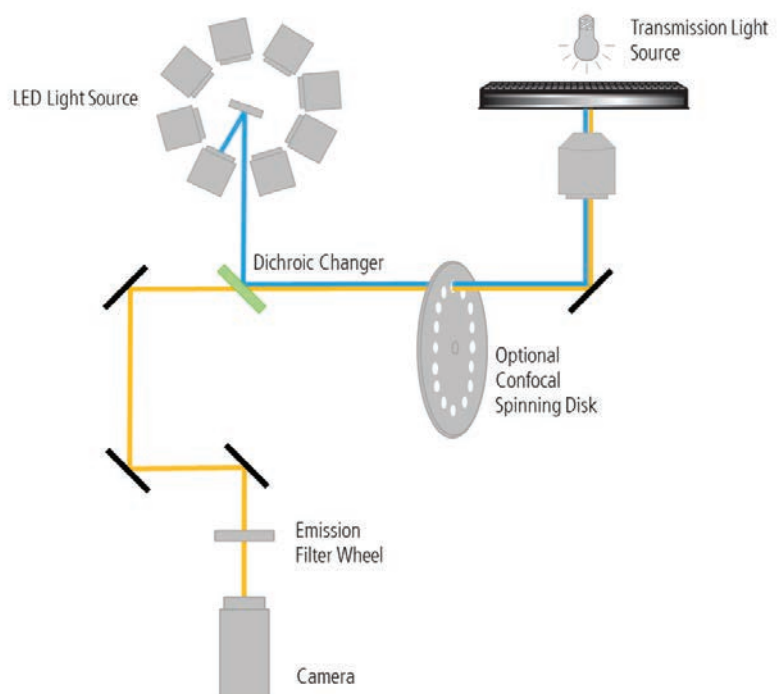# WHEN GREAT TECHNOLOGIES COMBINE THE RESULT IS ILLUMINATING

The product of more than a decade of experience in high-content screening, the Operetta CLS delivers the speed and sensitivity for everyday assays, and even complex challenges – live cells, phenotyping, rare events, and much more. All in an easy-to-use system that everyone in your lab can get started with right away.

Top corner: HeLa cells stained with Hoechst (DNA), Alexa Fluor™ 488 labeled anti-tubulin and TRITC-Phalloidin (actin) imaged with a high NA 40x water immersion objective (NA 1.1). Bottom corner: Same sample imaged with a 40x air objective NA 0.75 with the same exposure settings resulting in a dimmer image as the objective captures fewer photons.

Brighten things up

At the core of the Operetta CLS system is a whole new way to "see" your sample, with a new light path that ensures efficient excitation of your samples and careful collection of emitted signals. The compact 8x LED light source lets you choose the optimal wavelength for your fluorophore and delivers the power directly to your cellular samples. User-accessible emission filters let you further optimize detection.

With the stable LED light source and optional temperature and CO_2 control, you can run your live-cell assays reliably, assured in the knowledge that the changes you're seeing in the sample are due to the biology, *not* the technology. Once your assay parameters are optimized, you know you can trust the results you're seeing, time after time.

Optical path diagram labels:
LED Light Source
Transmission Light Source
Dichroic Changer
Optional Confocal Spinning Disk
Emission Filter Wheel
Camera

More light on the subject

Proprietary automated water-immersion objectives with very high numerical aperture deliver and capture more photons and provide a higher resolution in XYZ than conventional objectives – in fact, they capture up to four times more light than high numerical aperture air objectives can. You can benefit in two ways: Delicate live-cell samples can be excited with less light to protect them from photodamage or you can significantly increase the throughput of applications such as 3D stack acquisition.

The perfect way to image

The Operetta CLS system's proven spinning disk technology minimizes photobleaching and phototoxicity, and provides several advantages over conventional confocal microscopy techniques: Since multiple points are collected simultaneously rather than by scanning a single point at a time, the imaging process is much faster and gentler, enabling efficient background rejection, live-cell experiments, and 3D imaging. What's more, the large-format sCMOS camera delivers low noise, wide dynamic range, and high resolution – perfect for sensitive and quantitative measurements at short exposure times.
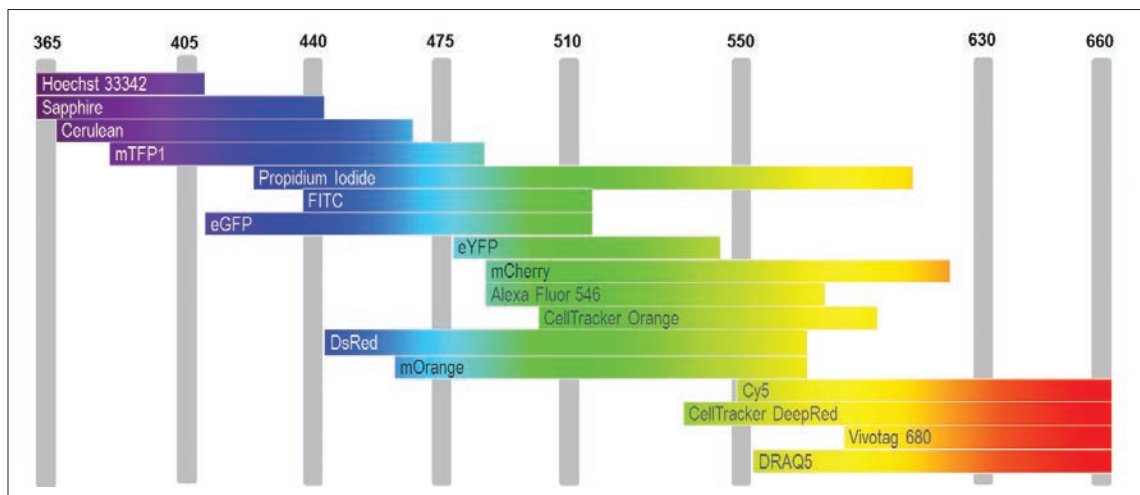
Up to eight LEDs excite a wide range of fluorophores, from UV to NIR, including DNA stains, live-cell stains, fluorescent proteins, and typical covalent labels. Some popular dyes are shown here.
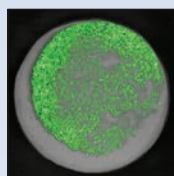
Your Applications, *Your* Way
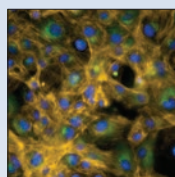
Fixed-cell assays

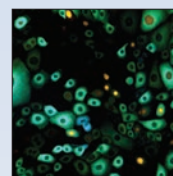To develop robust, content-rich assays, you may need a wide range of fluorescent stains and labels. With the Operetta CLS, you can choose from fluorescent widefield and confocal spinning-disk imaging with up to eight high-power excitation sources and user-accessible emission filters for maximum flexibility – so you can optimize your assays quickly.

3D cell culture models

Achieving good image quality from 3D samples such as spheroids, organoids, cysts, or zebrafish requires good confocal quality and sensitivity. Combined with spinning disk confocality, the water immersion objectives on the Operetta CLS collect up to 4 times more light from samples and increase the X, Y, and Z resolutions, enabling high-quality images from your 3D samples. With Harmony software, you have one software platform to acquire, visualize, and analyze your 3D samples – for more physiologically relevant results.

Live-cell assays

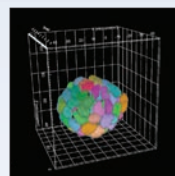Meaningful live-cell assays depend on stable excitation and minimal photodamage, spinning disk confocal optics, and synchronized LED illumination. For live-cell analysis without fluorescent labels, you can also choose brightfield or digital-phase contrast imaging modes.

Complex cellular models

Complex models, such as co-culture systems used in stem-cell research, drug discovery, and similar applications, pose unique challenges for imaging and analysis. The Operetta CLS combines a large-format sCMOS camera with water immersion objectives for sensitivity and high resolution, while machine-learning-based software (PhenoLOGIC) helps you to distinguish and characterize cell types based on phenotypic features such as morphology, fluorescence intensity, fluorescence distribution, and texture.

Advanced assays

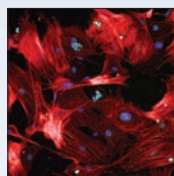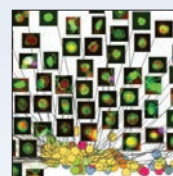FRET is a powerful tool for investigating conformational changes and protein-protein interactions. With a good FRET sensor, it's possible to track activity of kinases such as ERK in live cells and much more. And with sensitive imaging and dedicated analysis tools for ratiometric imaging, robust results are easy to obtain with Operetta CLS.

Phenotypic fingerprinting

At the core of successful phenotypic assays is the ability to create robust fingerprints of subtle differences. The Operetta CLS system combines high-resolution imaging with advanced software tools such as STAR morphology and machine learning for true multiparametric hit selection.

EVERYTHING COMES
TOGETHER
IN HARMONY

With Harmony® high-content imaging and analysis software, you can simply and easily turn the data you glean from your assays into deep, relevant knowledge. Harmony software enables you to handle all the data your experiments generate, analyze it quickly, discriminate phenotypes, and much more. You concentrate on your science – and Harmony does the rest.

Your entire lab is on the same page

Simple, powerful Harmony high-content imaging and analysis software unleashes the full potential of your Operetta CLS system. Harmony provides a complete solution, enabling you to set up assays and automate high-content imaging experiments, acquire images and analyze data, and store, retrieve, and present those results in meaningful ways. Its workflow-based interface makes the whole process simple and straightforward, even for new users with little microscopy or programming knowledge.

- Easily set up acquisition channels and parameters
- Utilize more than 30 ready-made solutions for common assays
- Use image analysis building blocks to create, configure, and customize your own protocols
- Get detailed cellular phenotype descriptions for more robust, reproducible classification with advanced analysis features
- Automatically store metadata such as assay layout, instrument settings, analysis results, and user-defined keywords and annotations
- Visualize and analyze your samples in 3D for greater depth of information and insights in a more physiologically relevant context

Harmony software also powers the Opera Phenix™ high-content screening system so you can transfer your Operetta CLS assays to higher throughput with ease. And with regular upgrades, it's a flexible solution for gathering unbiased data from your cell samples, keeping you ahead of your growing research needs.
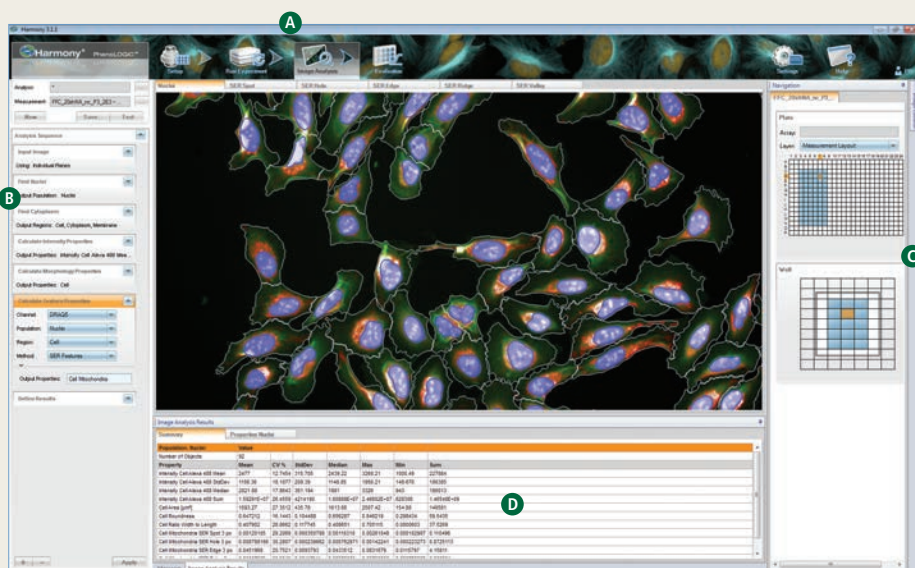
A. Workflow-based interface with easy-to-read icons

B. Analysis building blocks for easy protocol design

C. Clear plate navigation and wizard for easy setup of new plate types

D. Results summary with immediate numerical output for faster insights

Easily quantify cellular phenotypes – even in complex 3D models

Everything you need for 3D image acquisition, visualization, and analysis is right here in Harmony software. You can speed up 3D image acquisition through targeted imaging independent of culture method e.g. plates or hydrogels, and better understand your cell models by exploring them in a 3D viewer and an XYZ viewer. Plus, you can measure morphology, volume, and texture in 3D; count nuclei within spheroids; and calculate XYZ positional properties. You can also quickly analyze your z-stack as maximum intensity projection, with 3D information preserved, using PlaneMap technology.
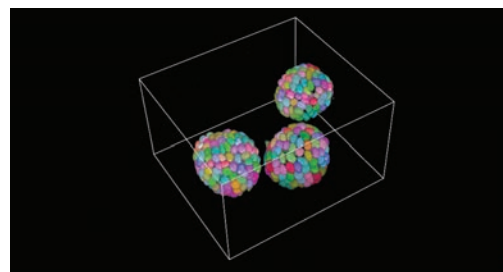

3D view of MDCK cysts with 3D segmentation of nuclei.

Accurately target your objects of interest for greater efficiency

High-content screening can capture fine subcellular detail with very high resolution images but high resolution can slow down acquisition times and create large amounts of data. So, ideally, you want to acquire high-resolution data only from your regions or objects of interest.

The **PreciScan** plug-in for Harmony software enables intelligent image acquisition to let you accurately target your objects of interest in x, y and z dimensions, such as spheroids, stem cell colonies, or rare cell phenotypes. The software delivers a fully automated, integrated workflow of low-magnification prescans, image analysis, and higher magnification rescans to reduce acquisition times and data volume and ultimately speed up analysis significantly.
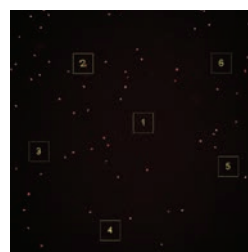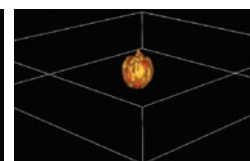

5x magnification pre-scan.
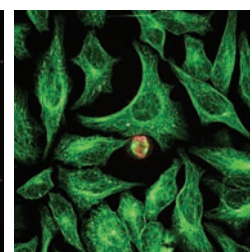

63x magnification re-scan.
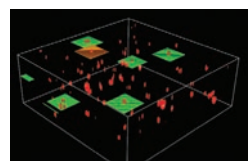
10x pre-scan in x, y, and z dimensions.

63x magnification re-scan.

Become an image analysis expert with machine learning

With the **PhenoLOGIC™** software plug-in, you can recognize different cell populations and regions by example – making it easy to create optimized algorithms. Just click on a few cells of each type to show the software what you're looking for. PhenoLOGIC software then selects the most meaningful combination of parameters to robustly discriminate phenotypes or to segment images based on texture features.
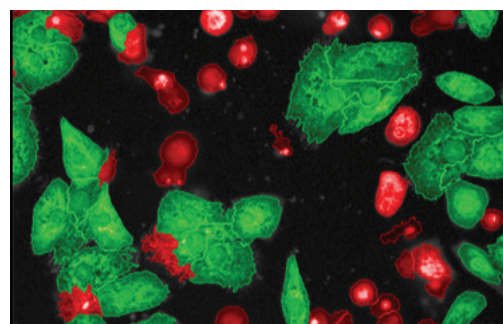
PhenoLOGIC classification of primary hepatocytes (green = healthy, red = dying).

Make better decisions sooner

Export your results automatically into the **Columbus™** image data storage and analysis system, so you can access, reanalyze, store, and share image data from Opera Phenix and other HCS systems across your organization. You can also use **High-Content Profiler™**, powered by TIBCO Spotfire® software, to aggregate your data with data from multiple sources, perform data QC and normalization, perform true multiparametric phenotypic analysis with its machine learning tools, and benefit from its interactive visual dashboards.

A solution configured to suit every need

Whatever your application, there's an Operetta CLS system configured to meet your requirements. And the system is modular, so it can change with your research demands. Configuration options include:

Operetta CLS Quattro

With four LEDs and widefield fluorescence, the basic configuration of the Operetta CLS system is ideal for common applications that need sensitivity and resolution, with the capacity to grow if the need arises.

Operetta CLS FLEX

With eight LEDs, and confocal and widefield fluorescence, the FLEX configuration offers flexibility in excitation and imaging modes for many challenging applications – and it can be upgraded to even higher performance.

Operetta CLS LIVE

With all the features of the FLEX configuration, plus gas and temperature control and water-immersion objectives, this system is ideal for gentle yet highly sensitive live-cell imaging.

It pays to automate

Higher throughput, improved productivity, reduced variability, savings on reagent costs – these benefits and more come from automating your Opera Phenix system. Integrate with the **plate::handler™** system for automated plate loading to enable overnight runs, or automate entire high-content screening workflows with **cell::explorer™** automated workstations.

The perfect plate for the work you do

For best results, choose the microplates designed specifically for high-end imaging systems. We've developed and validated a range of microplates for HCS applications, including **CellCarrier™** plates with a unique patented design for high-content imaging readers.

Count on our support

Your application needs are as individual as you are. So we take a team-based, consultative approach to every engagement with you – one that addresses your unique set of requirements. Our expert, global service and support teams, comprised of dedicated lab- and field-based applications specialists, can work with you in partnership to overcome the unique challenges your application brings.

For more information, visit www.perkinelmer.com/OperettaCLS

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