

Phenomenal  
fluorescence.



revvity

PhenoVue™ cellular imaging reagents



## PhenoVue reagents

# Achieve your moment of clarity

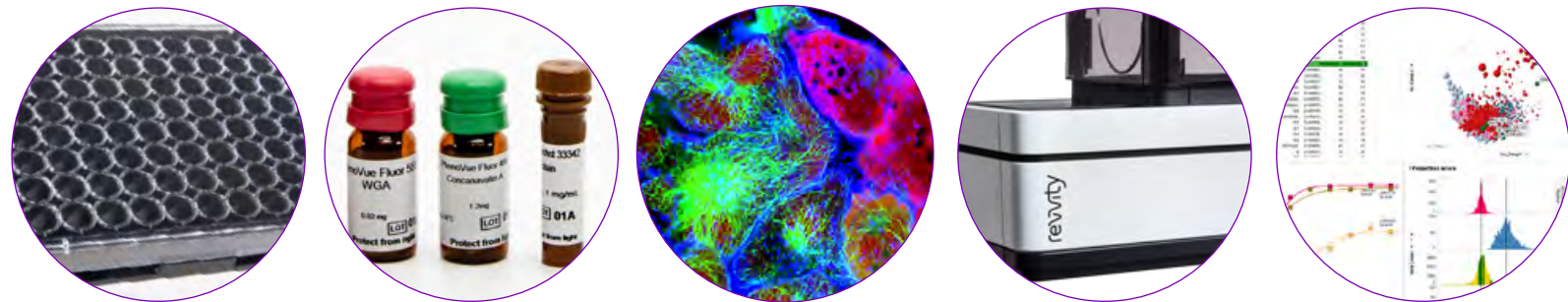
Faster results, simple reagents, easy-to-operate instrumentation, and seamless workflows – these benefits are not only nice-to-haves, but they’re necessary to keep up with scientific advances in studying cell biology and discovering novel drug treatments.

Cellular imaging allows you to detect and analyze numerous cellular components, from macromolecules to organelles and cell compartments. Scientists like you are studying more complex models such as 3D cell cultures with the goal of generating detailed phenotypic fingerprints for deeper insights. But pressures to do so with ease, speed, and reliability are mounting.

It’s time to discover PhenoVue™, our suite of cellular imaging reagents – including cell painting kits, organelle and cell compartment stains, fluorescent-labeled secondary antibodies, and cell function reagents – that complement our proven high-content screening instruments and image analysis software.

Working together to deliver reliable, accurate, physiologically relevant results, our portfolio of cellular imaging reagents, microplates, imaging instruments, image analysis software, and automation solutions is designed to streamline your high-content workflows and lead you to breakthroughs faster.

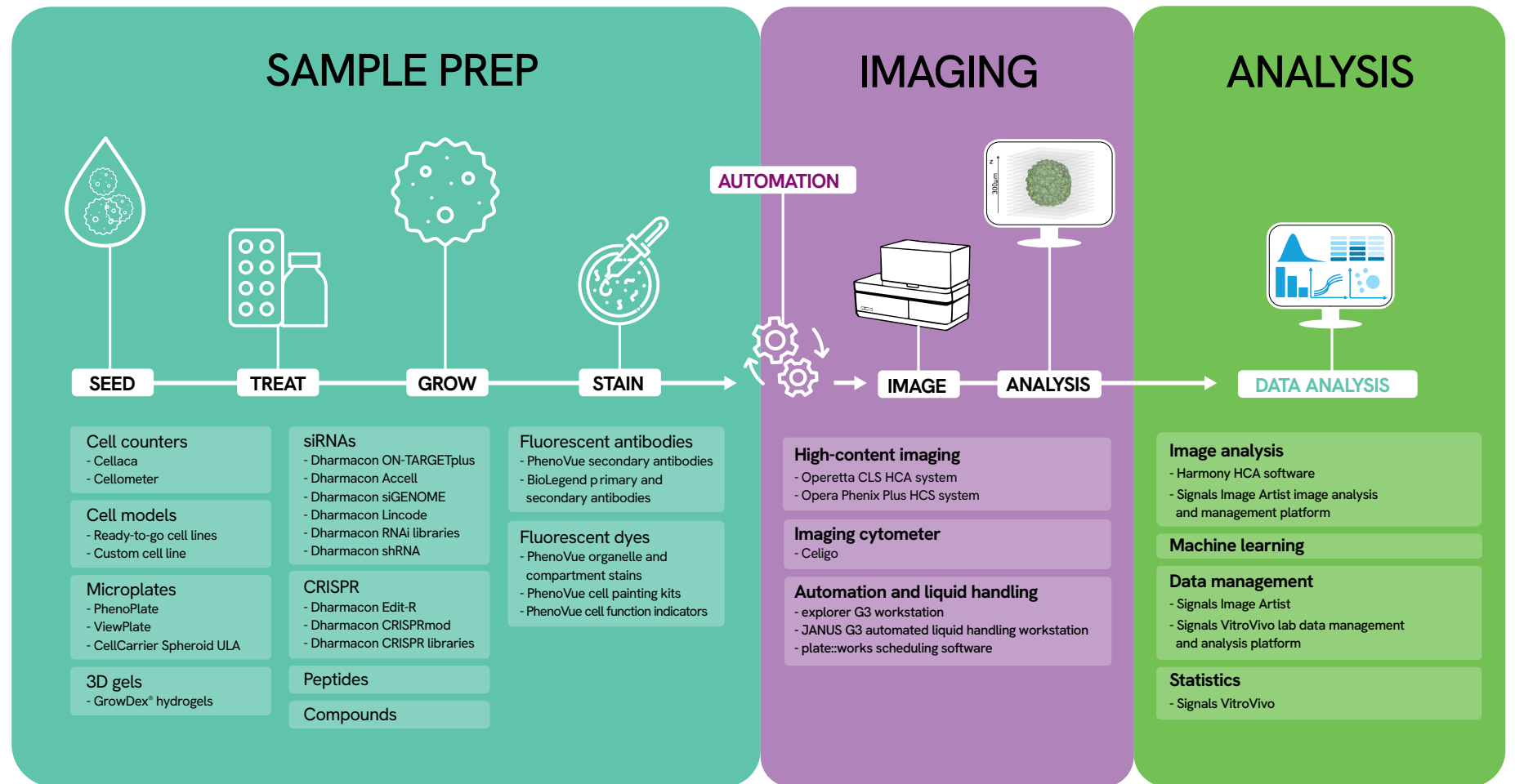
**Get to know our PhenoVue portfolio of reagents – and get to know better science.**





# Cellular imaging workflow

PhenoVue reagents





# Paint a picture of your analysis

Cell painting is a powerful phenotypic high-content screening approach that combines cell and computational biology to unravel cells' responses when subjected to perturbations.

In this process, cells are “painted” by labeling various cellular compartments with different fluorescent bioprobes to quantitatively profile multiple phenotypic parameters to better understand the effects of chemical compounds, drugs, genes, or other test articles.

Cell compartments and organelles are simultaneously tagged with six fluorescent probes, followed by acquisition and analysis of images. The six probes target specific cell compartments to determine protein expression or signaling pathways, identify organelles and their function, or identify whole-cell morphology.

Two versions of the PhenoVue cell painting kit are available, each in three different pack sizes. Both kits comprise validated, pre-optimized fluorescent bioprobes. The PhenoVue cell painting kit reflects the protocol described in Bray et al.<sup>1</sup> The PhenoVue cell painting JUMP kit reflects the JUMP-CP consortium protocol v3.<sup>2</sup>

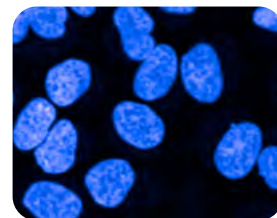
1. Bray, M.A., et al., *Cell Painting, a high-content image-based assay for morphological profiling using multiplexed fluorescent dyes*. *Nat Protoc*, 2016. 11(9): p. 1757-74.
2. [https://github.com/carpenterlab/2016\\_bray\\_natprot/wiki#updates-to-the-cell-painting-protocol](https://github.com/carpenterlab/2016_bray_natprot/wiki#updates-to-the-cell-painting-protocol) (Accessed June 2021).

## PhenoVue cell painting kits

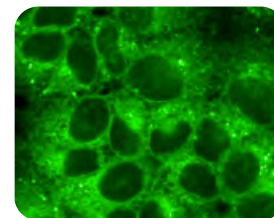
- PhenoVue cell painting kits for 1 x, 10 x, or 100 x 384-well plates
- PhenoVue cell painting JUMP kits for 1 x, 10 x, or 100 x 384-well plates

## PhenoVue complementary reagents

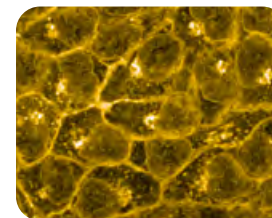
- PhenoVue dye diluent A (5X)
- PhenoVue paraformaldehyde 4% methanol-free solution
- PhenoVue permeabilization 0.5% Triton X-100 solution
- PhenoVue pluronic F-127, 20% solution
- PhenoVue probenecid ready to use solution



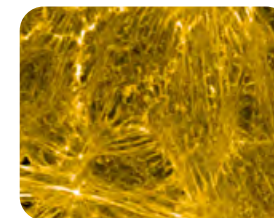
PhenoVue Hoechst 33342  
nuclear stain (nucleus)



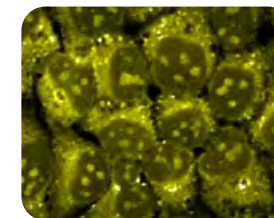
PhenoVue Fluor 488 -  
Concanavalin A  
(Endoplasmic Reticulum)



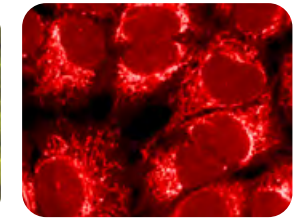
PhenoVue Fluor 555 -  
WGA (plasma membrane  
+ Golgi apparatus)



PhenoVue Fluor 568 -  
Phalloidin (actin)



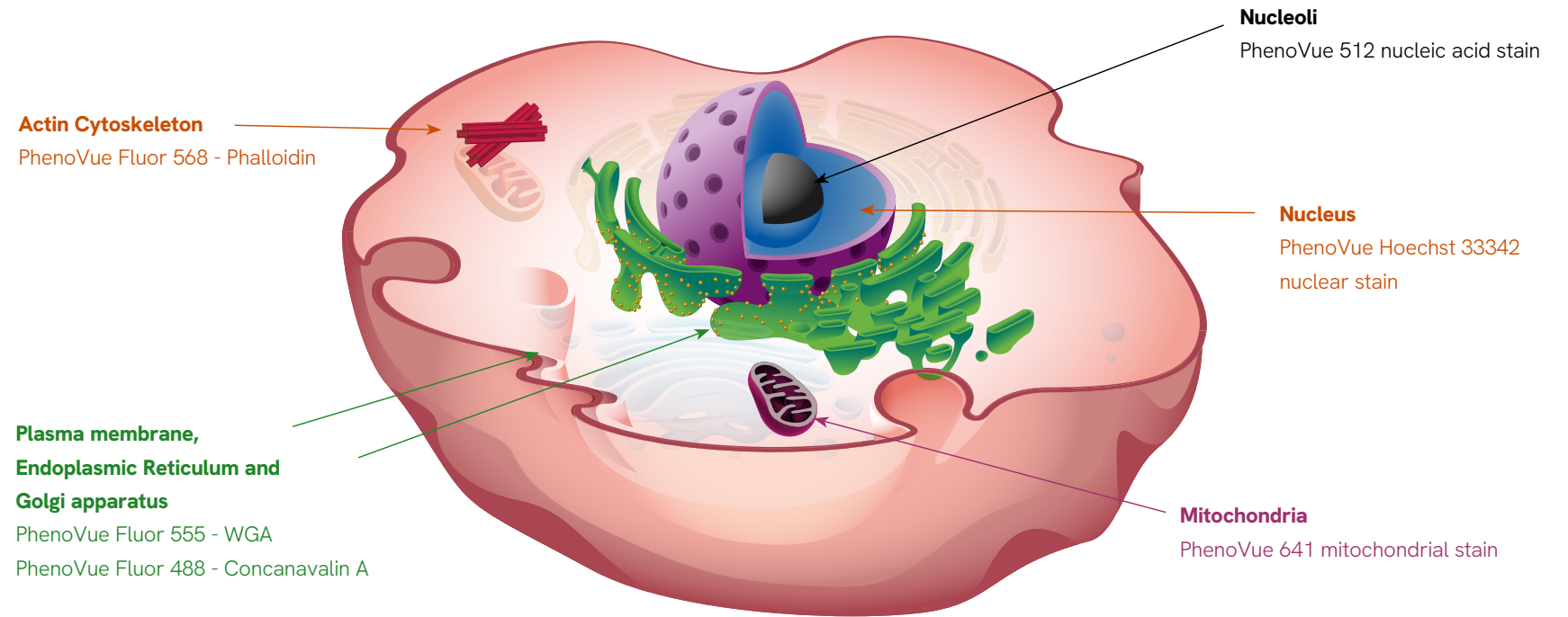
PhenoVue 512  
nucleic acid stain  
(nucleoli - RNA)



PhenoVue 641  
mitochondrial stain  
(mitochondria)

PhenoVue reagents

Cell painting kit components:



PhenoVue cell painting kits for 1 x 384-well plates



PhenoVue cell painting kits for 10 x or 100 x 384-well plates



# A simplified approach to studying biology, compounds, and diseases

## PhenoVue reagents

Mitochondria, lysosomes, lipid droplets, and actin play crucial roles in various cellular processes and interact with each other in intricate ways to maintain cell homeostasis.

Our PhenoVue multi organelle staining kit comprises five ready-to-use fluorescent dyes or antibodies to visualize mitochondria, lysosomes, lipid droplets, actin cytoskeleton, and nuclei in a 5-plex experiment.

Each component has been extensively validated and carefully optimized, with no spectral overlap, making this kit a straightforward solution to unravel fundamental biological processes, study compounds' mechanisms of action, and examine disease phenotypes.

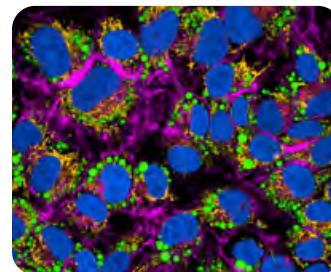
Applied to paraformaldehyde-fixed cellular models, the PhenoVue multi organelle staining kit provides an alternative to the established cell painting assay, avoiding using living cells, increasing your throughput, and providing a convenient method for phenotypic screening.

### PhenoVue multi organelle staining kits

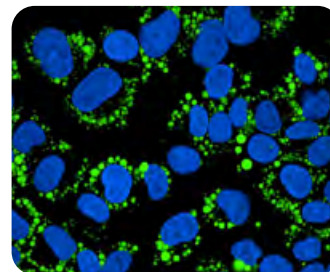
- PhenoVue multi organelle staining kit 1x384

### PhenoVue complementary reagents

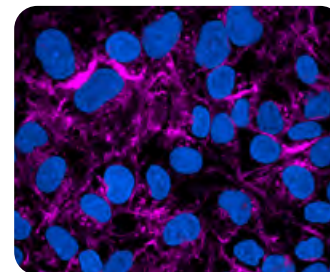
- PhenoVue dye diluent A (5X)
- PhenoVue paraformaldehyde 4% methanol-free solution
- PhenoVue permeabilization 0.5% Triton X-100 solution
- PhenoVue anti-LAMP1 antibody 100X
- PhenoVue anti-HSP60 antibody 100X



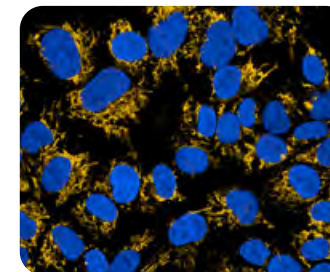
Merged



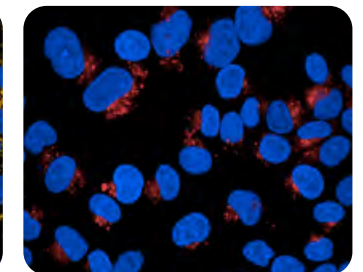
Lipid droplets



Actin



Mitochondria



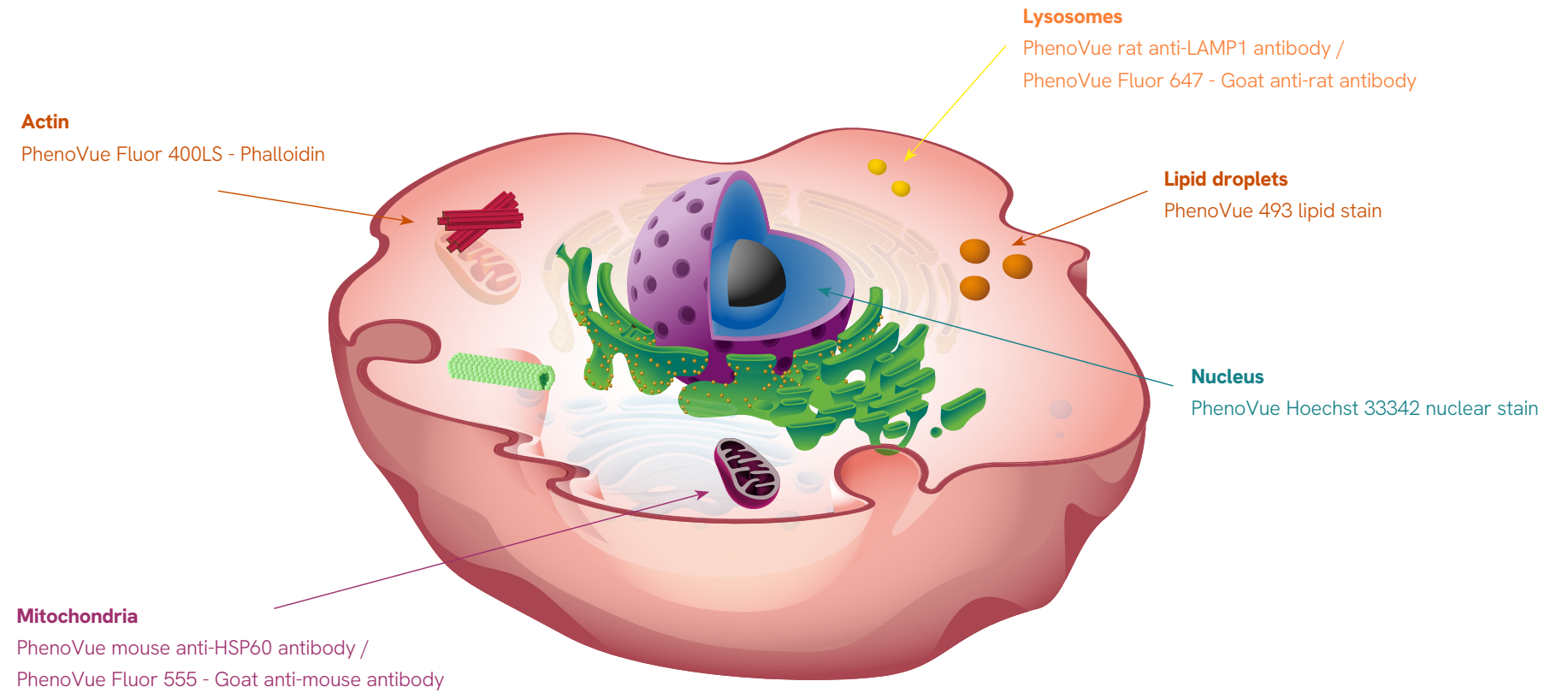
Lysosomes

HepG2 cells stained with PhenoVue multi organelle staining kit after oleic acid treatment, imaged on the Opera Phenix Plus high-content screening system.



PhenoVue reagents

Multi organelle staining kit components:





## PhenoVue reagents

# Qualify neuronal differentiation with ease

In neuroscience, more physiologically relevant cell models are essential to better recapitulate pathophysiological states underlying disease onset. Accurate qualification of neuronal cell models is critical, especially when long and complex differentiation processes are required — for example, iPSC-derived cells.

Our PhenoVue neuronal differentiation staining kit provides an easy solution to visualize mature neurons in brain cell derived co-cultures, as well as monitor the neuronal differentiation process from progenitors.

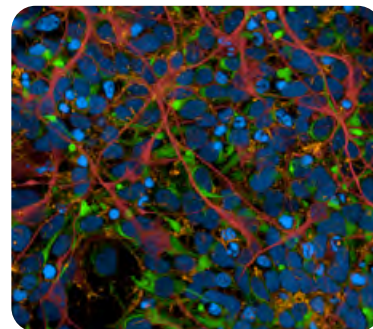
The kit comprises four validated, optimized, and ready-to-use fluorescent probes to visualize nuclei, actin, B3 tubulin, and nestin proteins in a 4-plex experiment with maximum spectral separation and no spectral overlap. We also provide a step-by-step protocol to facilitate your assay.

### PhenoVue neuronal differentiation staining kit

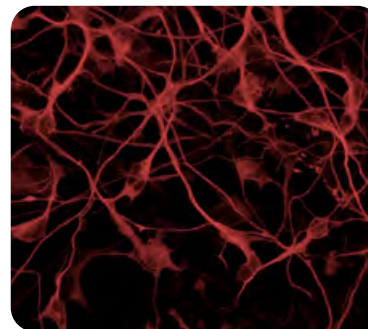
- PhenoVue neuronal differentiation staining kit 1x384

### PhenoVue complementary reagents

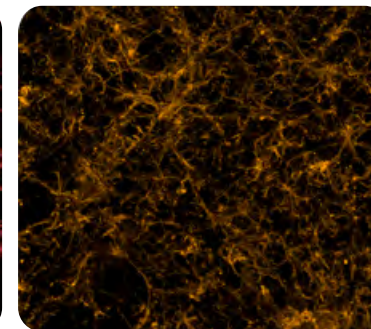
- PhenoVue dye diluent (5X)
- PhenoVue paraformaldehyde 4% methanol-free solution
- PhenoVue permeabilization 0.5% Triton X-100 solution
- PhenoVue anti-B3 tubulin antibody 100X
- PhenoVue anti-nestin antibody 100X
- PhenoVue anti-MAP2 antibody 100X



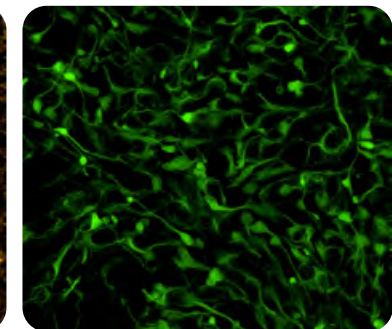
Merged



B3-tubulin



Actin



Nestin

HepG2 cells stained with PhenoVue multi organelle staining kit after oleic acid treatment, imaged on the Opera Phenix Plus high-content screening system.





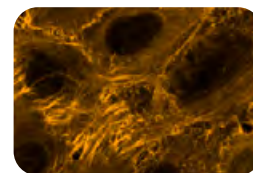
## PhenoVue reagents

# Gain deeper insights from your analysis

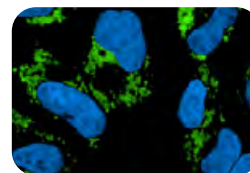
Cellular imaging techniques, such as high-content analysis, rely on the ability to detect and distinguish between specific cellular compartments and organelles. High-quality data depends on high-quality images, which all depends on bright fluorescent dyes.

Building on our extensive expertise in imaging instrumentation, fluorescent dye chemistry, and assay development, our PhenoVue organelle-specific stains detect various organelles and cellular compartments and are optimized for high-content screening. Features include:

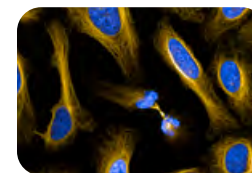
- Range of stains and fluors for commonly studied organelles and compartments
- Variety of fluorescent colors to enable multiplexing while avoiding spectral overlap
- Validation in high-content screening applications
- Bright fluorophores to enable high-quality images
- Photostable and sensitive live cell stains, with low cytotoxicity to maintain cell viability



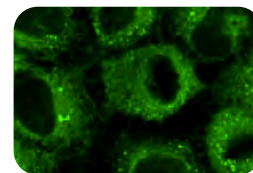
Fluor 568 - Phalloidin



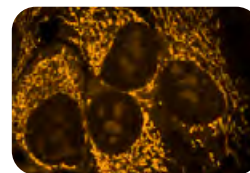
493 lipid stain



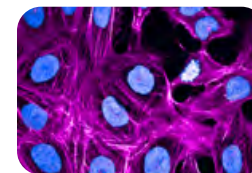
Fluor 555 live cell tubulin stain



Fluor 488 - Concanavalin A



551 mitochondrial stain



Fluor 400LS - Phalloidin

## PhenoVue cell compartment and organelle stains

Compartment or organelle	PhenoVue Fluor conjugates	Fluor 400 LS*	Fluor 488	Fluor 554	Fluor 555	Fluor 568	Fluor 594	Fluor 647
Plasma membrane, ER, Golgi	WGA	●	●	●		●	●	●
	Concanavalin-A	●	●	●		●	●	●
Actin cytoskeleton	Phalloidin	●	●	●		●	●	●
	Jasplakinolide derivative **							●
Microtubule network	Taxane derivative **				●			●

\*Long Stokes shift dye. \*\*Live cell imaging stains.

Compartment or organelle	PhenoVue stains
Mitochondria	PhenoVue 551 mitochondrial stain
	PhenoVue 578 mitochondrial stain
	PhenoVue 641 mitochondrial stain
Lysosome	PhenoVue 503 lysosomal stain
	PhenoVue 577 lysosomal stain
	PhenoVue 633 lysosomal stain
Lipids	PhenoVue Nile Red lipid stain
	PhenoVue 493 lipid stain
Nucleus	PhenoVue 512 nucleic acid stain
	PhenoVue Hoechst 33342 nuclear stain
	PhenoVue DAPI nuclear stain
	PhenoVue DRAQ5™ total cell nuclear stain
	PhenoVue DRAQ7™ dead cell nuclear stain

\*DRAQ5™ and DRAQ7™ are trademarks of BioStatus Limited.



PhenoVue reagents

PhenoVue cell compartment and organelle stains:

**Actin cytoskeleton**

- PhenoVue Fluor 400LS - Phalloidin
- PhenoVue Fluor 488 - Phalloidin
- PhenoVue Fluor 555 - Phalloidin
- PhenoVue Fluor 568 - Phalloidin
- PhenoVue Fluor 594 - Phalloidin
- PhenoVue Fluor 647 - Phalloidin

**Lysosome**

- PhenoVue 503 lysosomal stain
- PhenoVue 577 lysosomal stain

**Lipid droplets**

- PhenoVue Nile-Red lipid stain
- PhenoVue 493 lipid stain

**Nucleoli**

- PhenoVue 512 nucleic acid stain

**Nucleus**

- PhenoVue Hoechst 33342 nuclear stain
- PhenoVue DAPI nuclear stain
- PhenoVue DRAQ5™ total cell nuclear stain
- PhenoVue DRAQ7™ dead cell nuclear stain

**Mitochondria**

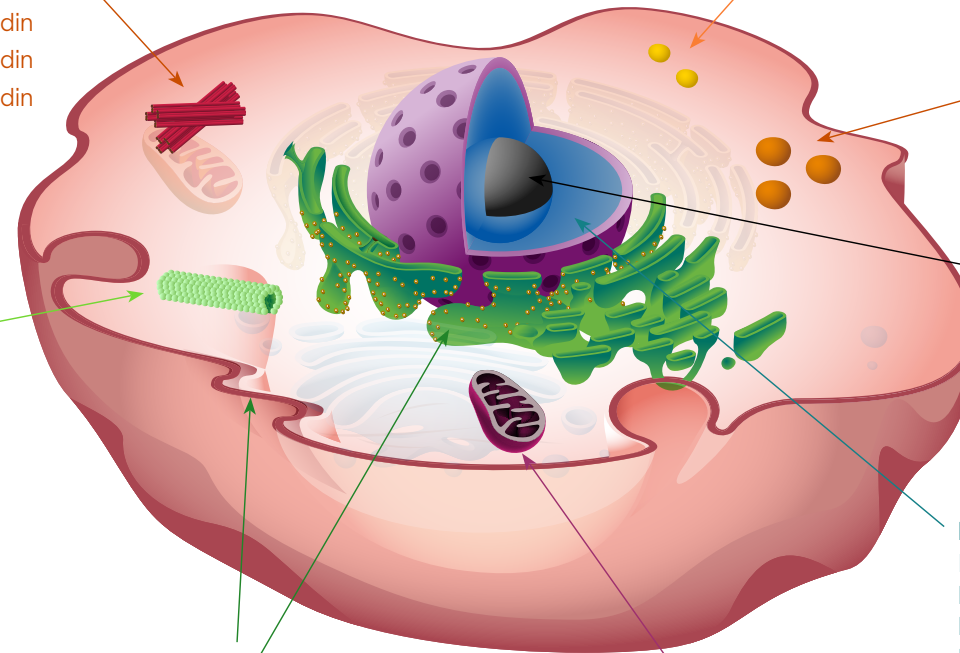
- PhenoVue 551 mitochondrial stain
- PhenoVue 578 mitochondrial stain
- PhenoVue 641 mitochondrial stain

**Microtubule network**

- PhenoVue Fluor 555 - Live cell tubulin stain
- PhenoVue Fluor 647 - Live cell tubulin stain

**Plasma membrane, endoplasmic reticulum, and Golgi apparatus**

- PhenoVue Fluor 488 - WGA
- PhenoVue Fluor 555 - WGA
- PhenoVue Fluor 568 - WGA
- PhenoVue Fluor 594 - WGA
- PhenoVue Fluor 647 - WGA
- PhenoVue Fluor 488 - Concanavalin A
- PhenoVue Fluor 555 - Concanavalin A
- PhenoVue Fluor 568 - Concanavalin A
- PhenoVue Fluor 594 - Concanavalin A
- PhenoVue Fluor 647 - Concanavalin A



# Imagine the possibilities

Fluorescent secondary antibodies are everyday tools for researchers performing high-content analysis, cellular imaging, flow cytometry, and western blotting. The method of indirect immunofluorescence detection relies on an unconjugated antigen-specific primary antibody coupled with a fluorescent secondary antibody.

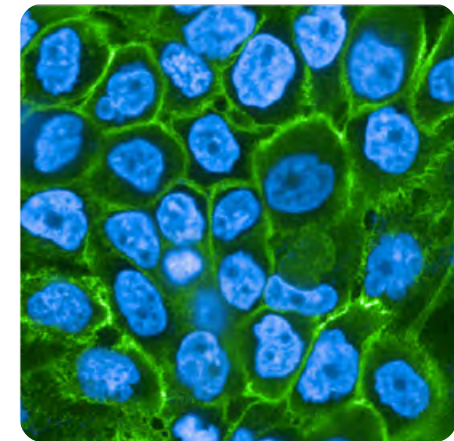
Secondary fluorescent antibodies offer enhanced sensitivity - binding to the primary antibody and then amplifying the fluorescence signal. In addition, they provide greater flexibility since they can be easily combined for multiplexing experiments. Get to know our extensively validated PhenoVue Fluor-labeled secondary antibodies. Features include:

- Bright fluorophores for high-quality images
- Highly cross-adsorbed antibodies to minimize cross-reactivity in multiplexing applications
- Anti-rabbit, anti-mouse and anti-rat antibodies for your common applications
- Validated in high-content screening applications

## PhenoVue fluorescent secondary antibodies

		Fluor 488	Fluor 555	Fluor 568	Fluor 594	Fluor 647
Goat anti-rabbit	Cross-adsorbed	●	●	●	●	●
	Highly cross-adsorbed	●	●	●	●	●
Goat anti-mouse	Cross-adsorbed	●	●	●	●	●
	Highly cross-adsorbed	●	●	●	●	●
Donkey anti-rabbit	Cross-adsorbed	●	●	●	●	●
	Highly cross-adsorbed	●	●	●	●	●
Donkey anti-mouse	Cross-adsorbed	●	●	●	●	●
	Highly cross-adsorbed	●	●	●	●	●
Goat anti-rat	Highly cross-adsorbed	●	●	●	●	●
Rat anti-mouse (isotype specific)	Highly cross-adsorbed	●*				●**

\* IgG1 \*\* IgG2a



A431 cells stained with mouse anti-EGFR antibody + PhenoVue Fluor 488 - Goat anti-mouse antibody, highly cross-adsorbed, and PhenoVue Hoechst 33342 nuclear stain.

## PhenoVue complementary reagents

- PhenoVue dye diluent A (5X)
- PhenoVue paraformaldehyde 4% methanol-free solution
- PhenoVue permeabilization 0.5% Triton X-100 solution

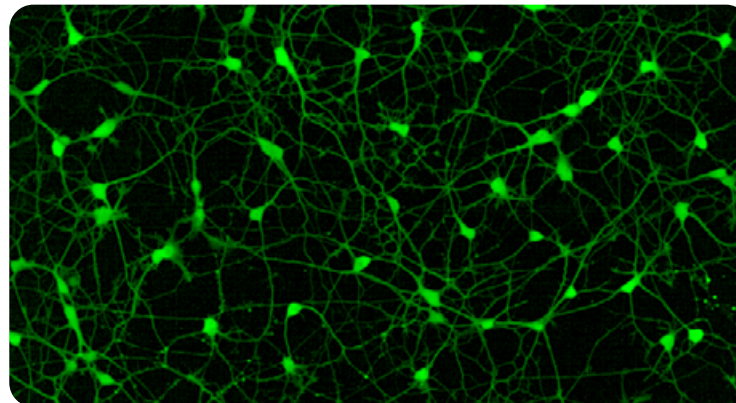


## PhenoVue reagents

# Visualize cell functionality

Understanding cell function is essential for cellular research. We offer a complete suite of cell function reagents and kits for visualizing key cellular events including viability, apoptosis, mitochondrial membrane potential, as well as indicators for calcium, reactive oxygen species (ROS), or hypoxia. Validated for use in imaging microscopy and high content screening applications:

- ROS total oxidative stress indicators enable a myriad of applications as well as multiplexing capability to unravel ROS related biological effects.
- Fluorescent calcium indicators enable a variety of applications as well as multiplexing capability to unravel intracellular calcium mediated functions.
- HypoxiTRAK™ is a permeable hypoxia indicator, which exhibits far-red fluorescence. Its spectral characteristics are shared with DRAQ5™.
- Live/dead cell viability assay kit enables the simultaneous determination of live and dead cells using two different dyes.
- Live cell caspase-3/7 activity stain is a no-wash and cell permeable fluorogenic dye for visualizing apoptotic cells.



| iPSC-derived neurons (Nexel) stained with PhenoVue Cal-520 AM.

## PhenoVue cell function reagents and kits

	PhenoVue reagents and kits
Total oxidative stress indicators	PhenoVue ROS-490
	PhenoVue ROS-570
	PhenoVue ROS-670
Calcium indicators	PhenoVue Cal-520 AM
	PhenoVue Cal-590 AM
	PhenoVue Cal-520 AM Bright
	PhenoVue Cal-590 AM Bright
	PhenoVue Fluo-4 AM
Hypoxia indicators	PhenoVue Fura-2 AM
	PhenoVue HypoxiTRAK™
Cell viability	PhenoVue live/dead cell viability assay kit
Apoptosis	PhenoVue 505 live cell caspase-3/7 activity stain

## PhenoVue complementary reagents

- PhenoVue dye diluent A (5X)
- PhenoVue paraformaldehyde 4% methanol-free solution
- PhenoVue permeabilization 0.5% Triton X-100 solution
- PhenoVue pluronic F-127, 20% solution
- PhenoVue probenecid, ready to use solution



# Imaging microplates for precision analysis

See what others can't when you use our imaging microplates designed for cell-based assays, confocal microscopy, and high-content phenotypic imaging and analysis. And to facilitate 3D cell cultures for imaging applications, we offer our specialized CellCarrier™ Spheroid ULA microplates.

## PhenoVue reagents

### PhenoPlates



### PhenoPlate™ microplates

Achieve accurate results with our PhenoPlates, engineered to deliver superior images and maximum quality data for all high-content applications. Our PhenoPlates feature an exceptionally flat bottom, enabling fast autofocusing for optimal clarity. Plus, you'll benefit from superior images thanks to their 188  $\mu\text{m}$  thickness and the high-optical glass-like quality of the cyclic olefin foil imaging surface. Cyclic olefin also offers better transparency in the near UV range when compared to polystyrene, giving you a clearer image for better results. Additional features include:

- Ultralow plate bottom provides better well access when using water immersion and high-numerical aperture (NA) objectives
- Low-profile polystyrene lid results in reduced evaporation
- Unique corner spacers minimize the risks of damaging the imaging surface when stacking
- Various coatings to choose from to suit your application
- 96-, 384-, and 1536-well formats



# Imaging microplates for precision analysis

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PhenoVue reagents

## CellCarrier Spheroid ULA microplates



### CellCarrier Spheroid ULA microplates

A unique ultralow attachment (ULA)-coated surface in round well plates enables the formation of consistently round spheroids from numerous cellular models. This microplate coating also helps eliminate satellite spheroid growth, which allows for easier data acquisition and analysis. Additional features include:

- Unique design made specifically for 3D spheroids
- Automation compatibility for quick, hassle-free analysis
- Compatible with high-content screening systems such as Operetta CLS™ and Opera Phenix™ Plus

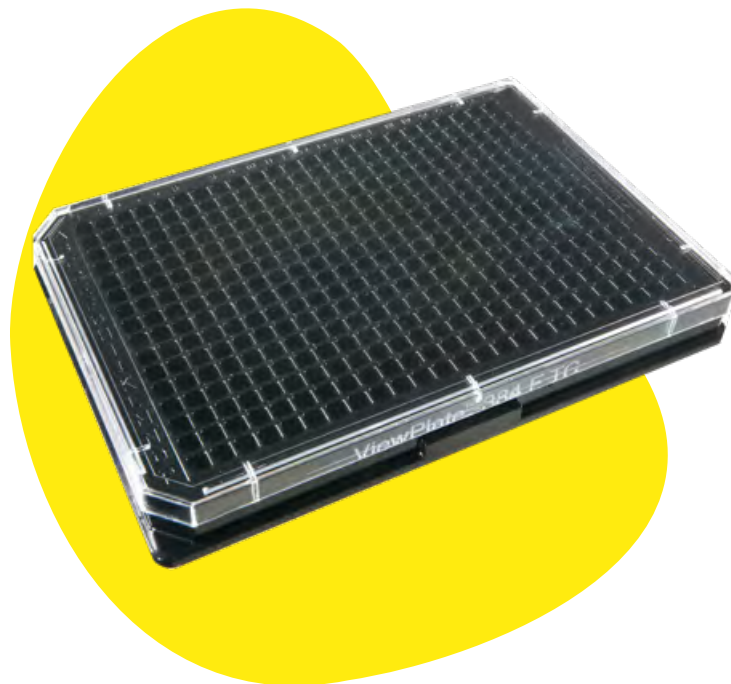


## PhenoVue reagents

# Imaging microplates for precision analysis

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## ViewPlates



### ViewPlate™ microplates

These poly-D-lysine (PDL)-coated and collagen-coated microplates feature a clear-bottom plastic base with an opaque black or white frame and are specifically designed for microscopy-based HCS applications. In addition, they are ideal for multimodal analyses on microplate readers that also have well imaging, such as the EnSight™ multimode plate reader. Varieties include:

- 96-, 384-, and 1536-well formats
- White ½-area 96-well plates
- Untreated for biochemical assays
- Tissue culture (TC)-treated, collagen-coated, or PDL-coated for cellular assays

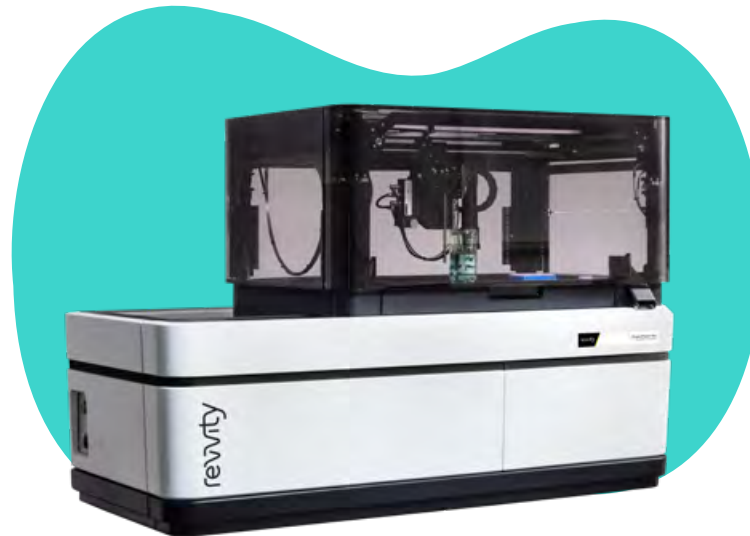


# High-content analysis and screening systems

## PhenoVue reagents

From basic research to assay development and screening, our high-content analysis (HCA) and high-content screening (HCS) systems produce the highest possible image quality so you can take your research further - in less time than ever before - especially when combined with our easy-to-use workflow-based Harmony™ software, robotic systems, and advanced data analytics.

### Opera Phenix Plus high-content screening system



### Opera Phenix™ Plus high-content screening system

For your most demanding high-content applications. Benefits include:

- Modular design adapts to your changing application needs
- Enhanced speed using a dual- or four-camera configuration with simultaneous imaging
- Synchrony Optics™ combines a microlens-enhanced Nipkow spinning disk with a pinhole distance optimized for thick and 3D samples
- Dual-view excitation of neighboring spectral channels minimizes crosstalk
- Custom-designed high-NA water immersion objectives capture more photons and provide high-image resolution even in thick samples
- Fast imaging frame rate of up to 100 fps and optional pipettor module captures fast cellular responses

**Best for disease research and screening labs needing fast throughput and high performance.**





# High-content analysis and screening systems

## PhenoVue reagents

From basic research to assay development and screening, our high-content analysis (HCA) and high-content screening (HCS) systems produce the highest possible image quality so you can take your research further - in less time than ever before - especially when combined with our easy-to-use workflow-based Harmony™ software, robotic systems, and advanced data analytics.

### Operetta CLS high-content analysis system



### Operetta CLS™ high-content analysis system

For routine 2D high-content assays and complex assays. Benefits include:

- Confocal spinning-disk technology provides a gentle imaging process (especially for live-cell experiments) for efficient background rejection
- Flexible excitation with a powerful and stable 8x LED light source
- Proprietary automated water-immersion objectives with very high numerical aperture, giving faster read times and reduced photodamage

**Best for basic research and assay development labs with multiple users and intermediate throughput requirements.**



# Accelerate your science through automation

Get the most out of your analysis with the right software. Harmony, Signals Image Artist™, and Signals VitroVivo™ software are all compatible with our imaging instruments and PhenoVue cellular imaging reagents. Our software makes it easy for you to generate and analyze image data from cellular samples and *in vivo* models.

## PhenoVue reagents



From acquisition to analysis, Harmony high-content imaging software empowers you to easily quantify more of what you see in your images. With everything you need to analyze even the most complex cellular models in 3D, discriminate phenotypes confidently, and turn your data into discovery, Harmony software enables you to control every aspect of your analysis through a single, easy-to-use workflow-based interface.



For universal high-volume image data storage and analysis, Signals Image Artist software provides a central location to quickly process, analyze, share, and store the vast volumes of data generated by high-content screening and cellular imaging, including 3D imaging, phenotypic screening, and cell painting. Compatible with all major HCS and cell imaging systems, Signals Image Artist is a multiuser platform that can support your entire lab and is scalable to expand with your lab's data storage needs over time. It integrates seamlessly with the Operetta CLS and Opera Phenix Plus high-content screening systems, as well as the Signals VitroVivo software platform.



Signals VitroVivo software platform provides lab data management and analysis for all modalities in one platform. It unites assay development, low throughput to ultra-high throughput production assays, high-content screening, and *in vivo* studies so you can search across all assay and screening data in a single platform. With Signals VitroVivo, you can perform screening data analysis and validation, QC analyses, calculate reliable normalization, multivariate hit stratification, dose response curves, drug response profiling, and more.



PhenoVue reagents

# Integrated. Automated. Uncomplicated.

Higher throughput, improved efficiency, and reduced variability might sound like a dream. But these benefits are a reality when you automate your lab. Our experts have extensive experience developing integrated workstations for automating cell-based assays. Whatever your screening needs, we have a solution that's right for your lab.

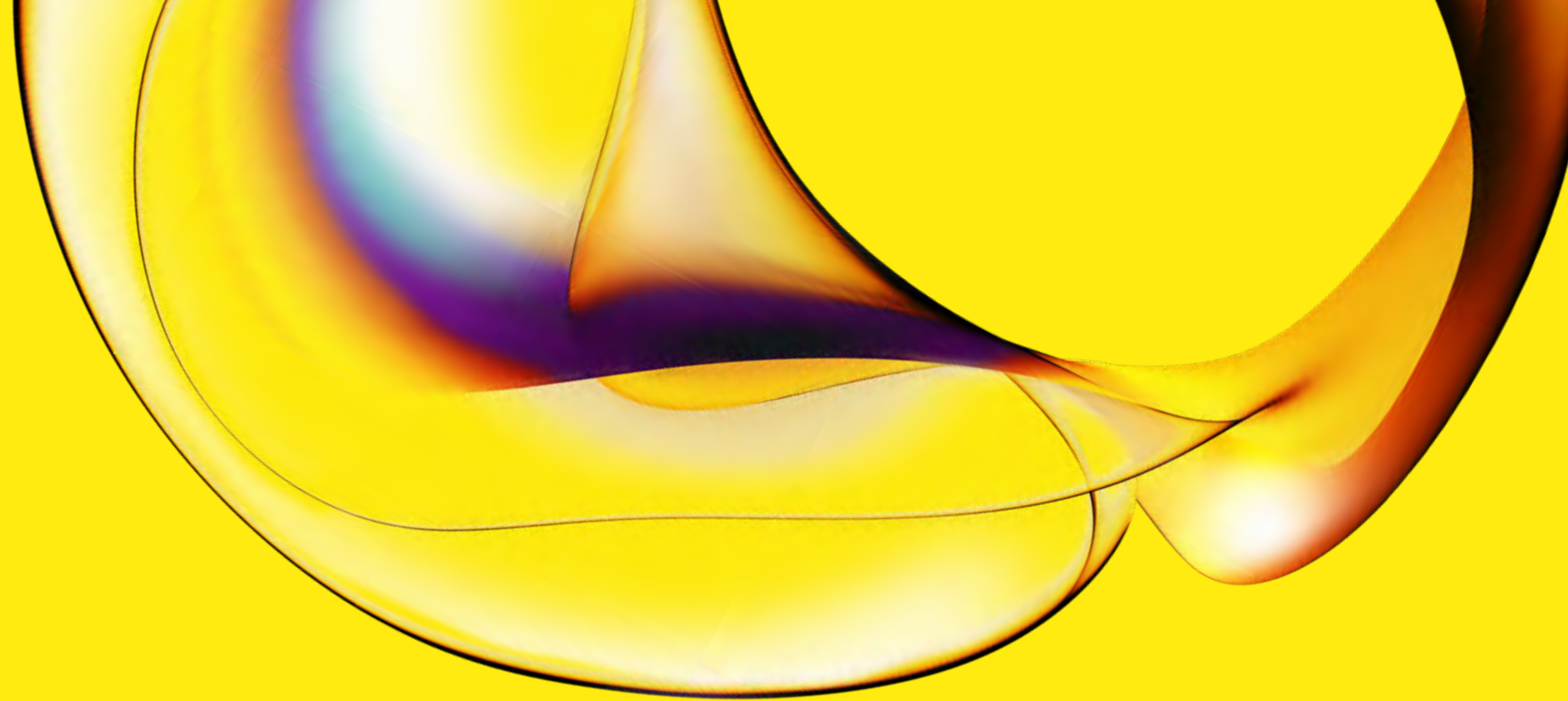
Whether you're integrating with our plate::handler™ FLEX system for automated plate loading, or integrating your entire high-content screening workflows with our explorer™ G3 automated workstations, the difference in your analysis is clear – increased productivity, less risk of human error, safer working conditions, and the ability to maximize your data for better results.



plate::handler FLEX system



explorer™ G3 workstation



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