

High Quality Luminescence

Instrumentation and Reagents



Reporter Gene Assays, GPCR, Kinase,
Cytotoxicity, and Cell Proliferation research

streamline your research and discovery

luminescence assay systems

Why luminescence?

Luminescence is an extremely attractive detection platform in some applications for the following reasons:

- Wide dynamic range
- More sensitive than fluorescence technologies
- Less interference
- Cost effective
- Easy to automate for high throughput applications
- Homogenous assay set up



G-Protein Coupled Receptors

- Aequorin parental cell lines
- AequoScreen™
- AequoZen™ FroZen™ cells
- reporter gene assays
- steadylite plus™
- britelite™ plus

Cytotoxicity and Cell Proliferation

- ATPlite
- ATPlite 1step

Kinases

- easylite-Kinase

Luminescence Detection Instruments

- Cellular Workstation
- EnVision™
- LumiLux™
- MicroBeta®
- TopCount®
- VICTOR³™
- VICTOR™ Light
- ViewLux™

LUMINESCENCE ASSAY SYSTEMS — Simpler. Brighter. Smarter.

	AequoScreen™ AequoZen	britelite plus	steadylite plus	ATPlite 1step	ATPlite	easylite-Kinase
Application	GPCR/ Calcium Flux	Reporter Gene Assay	Reporter Gene Assay	Cytotoxicity/ Cell Proliferation Assay	Cytotoxicity/ Cell Proliferation	Kinase Activity Assay
Half-life (hours)	Flash	0.5	4 to 5	0.5	5	5
Sensitivity	Ultra High	High	Moderate 5 cells/well	As little as 5 cells/well	As little as 5 cells/well	High
Plate Format (wells)	96 384 1536	96 384 1536	96 384 1536	96 384 1536	96 384 1536	96 384 1536
Instrumentation	TopCount MicroBeta Jet EnVision — VICTOR Light ViewLux LumiLux	TopCount MicroBeta EnVision VICTOR ³ VICTOR Light ViewLux LumiLux				

calcium flux assays for GPCR analysis

AequoScreen and AequoZen

Platform for analysis and screening of GPCRs

Aequorin is a photoprotein isolated from the jellyfish *Aequoria victoria*. Upon calcium binding, aequorin oxidizes coelenterazine into coelenteramide with the production of CO₂ and emission of light. AequoScreen is PerkinElmer's aequorin product range providing a generic GPCR technology which can be used with G_s, G_o and G_i coupled receptors and calcium coupled ion channels. It offers an ideal replacement technology for the standard fluorescent dyes enabling both cost and productivity advantages. The product portfolio includes cell lines as well as frozen cells:

- **AequoScreen parental cell lines:** expressing mitochondrially targeted apoaequorin with or without the promiscuous G protein G_{α16}
- **AequoScreen double transfected cell lines:** expressing mitochondrially targeted apoaequorin and GPCR with or without the promiscuous G protein G_{α16}
- **AequoZen FroZen cells:** our most popular double transfected cell lines irradiated and available frozen

The advantage of AequoScreen

- Fully automatable and miniaturizable to 1536 format
- Enables reduction in reagents and consumable costs, including the number of cells required for an assay

- High signal-to-background ratio enables detection of agonists, antagonists and allosteric modulators
- Robust assay (Z', CV values, solvent resistance)
- Reduces the number of false positives from auto-fluorescent compounds in screening libraries
- Option to work with cells in suspension to increase throughput
- Throughput: up to 100,000 data points per day (depending on assay and microplate format)

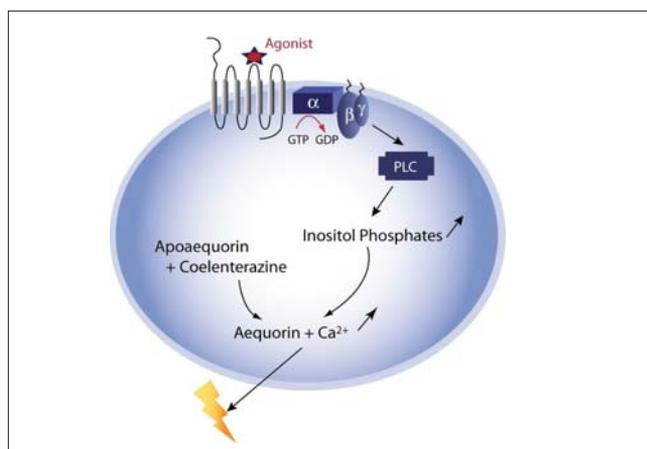


Figure 1. Principle of aequorin: Following GPCR stimulation, increases in intracellular calcium enable measurement of the resulting flash luminescence signal.

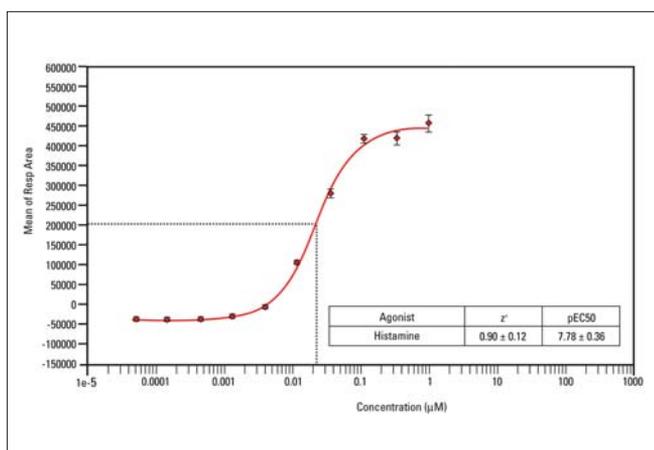


Figure 2. LumiLux CHO aequorin H1 agonist assay — 384-well suspension assay, 6000 cells/well (mean +/- SD from 8 wells).

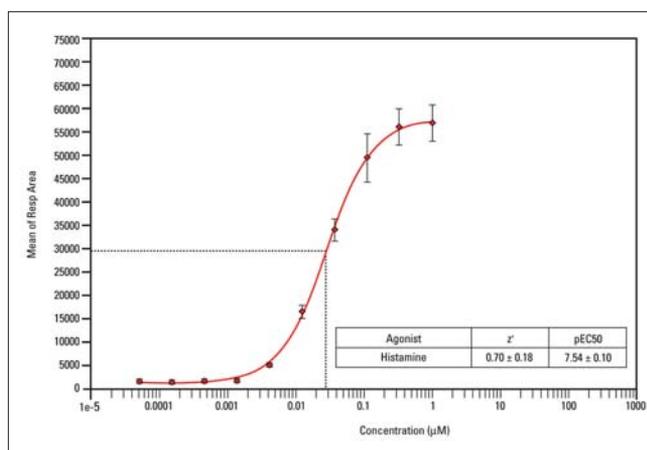


Figure 3. LumiLux CHO aequorin H1 agonist assay — 1536-well suspension assay, 1500 cells/well (mean +/- SD from 32 wells).

ready-to-use cells for calcium detection

AequoZen takes the stress out of functional testing

PerkinElmer can transfer any amount of validated cells for culture-free functional testing of GPCRs. The AequoZen FroZen cells are fully validated for aequorin calcium testing.

We offer

- Pre-validated batches of ready-to-use cells
- Step-by-step protocols for culture-free functional testing on our validated cells
- Affordable access to small aliquots of cells (minimum size: 3×10^6)
- Ready-to-use cells for profiling, pharmacology or screening applications
- Irradiated cells to minimize biological variation
- Up-scaling and customized developments

We remove the lengthy process of cell culture from your functional testing. That means that we will culture the cells, freeze them with an optimized protocol, and irradiate them with gamma rays to stop all replication. These cells are then stored in liquid nitrogen or simply in a -80°C freezer, thawed and used directly in a functional, cellular GPCR test with previously validated performance.

Validation

For each receptor in our FroZen cell list, we have tested the optimal freezing conditions, stability at -80°C for safe shipment of cells anywhere in the world and validated assay

protocol performance. Once you receive your AequoZen cells, you will also receive a complete, step-by-step protocol guiding you each step of the way to get great results. Validated batches give results meeting our QC criteria (both on EC_{50} and window) so that you can get the same reliable results with every purchase, every time.

Flexibility

All our FroZen cells are available in off-the-shelf aliquots of 1 to 10 million cells per vial that you can receive ready for testing within a week from your order. We also scale-up to provide you with validated cells for full screening campaigns. Whether it is for screening, lead optimization or profiling, you can now perform cellular GPCR tests immediately by using our batches of FroZen cells on multiple receptors at a time. We validate defined families of GPCRs that will be available off-the-shelf to let you perform selectivity studies rapidly and cost-effectively as needed.

Convenience

All our FroZen cells are priced on a per unit basis so that you pay for the cells that you need. Since our FroZen cells preparations are growth arrested by irradiation, you can better standardize cell numbers following overnight incubation in plates (thus avoiding plate artifacts). The irradiation procedure does not leave any traces of hazardous chemicals in the cell media, and has been titrated to preserve functional response.

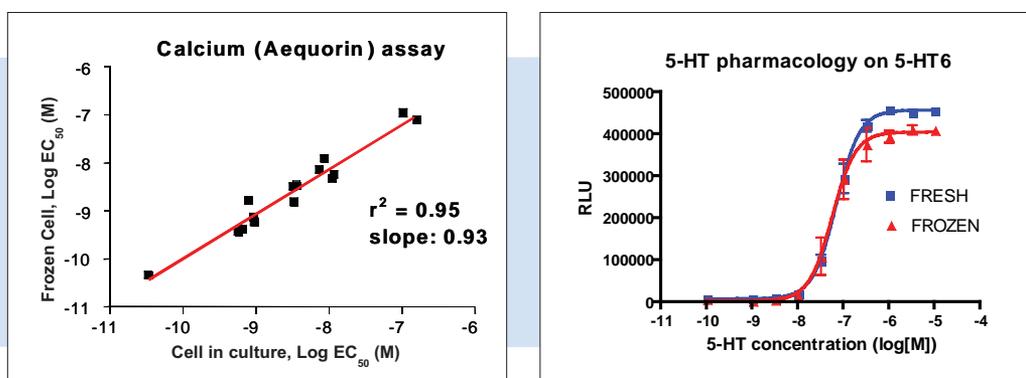


Figure 4 and 5. All batches are tested for quality of assay window (left graph, cAMP on MC4 receptor) and consistency of EC_{50} values (right graph, correlation of EC_{50} values between fresh AequoScreen: 5HT6 (G_s) and G α_{16} and equivalent FroZen cells).

the most convenient luciferase detection systems available

reporter gene assays **britelite plus**

No more DTT and no need for -20 °C storage – safer, more stable

The **NEW** PerkinElmer luciferase detection kits bring the simplicity of reporter gene assays to a whole new level. We have significantly improved the ease of storage and handling, at the same time maintaining the superior performance of our original products.

- **Convenience:** The new enhanced britelite plus and steadylite plus can be stored at 2-8 °C
- **Safety:** Unlike conventional luciferase detection systems, we no longer use DTT (dithiothreitol) as a reagent component. This removes the repugnant odor and hazards associated with the use of DTT*
- **Ease:** No washing or removal of medium – all in one well culture and assay; removal of DTT eliminates the need for hood work

- **Performance:** Superior S/B allow for excellent Z' values in antagonist and agonist studies

Reporter genes can be attached to a gene of interest to monitor gene expression in cell culture. steadylite plus and britelite plus contain the necessary reagents to detect the expression of the reporter gene luciferase of the North American firefly *Photinus pyralis*.

Luciferase catalyzes the bioluminescent oxidation of the substrate luciferin in the presence of ATP, magnesium and oxygen. Luciferase assays are highly sensitive, have a wide dynamic range, low cost and do not interfere with the biology of a cell. In addition, luciferase is not naturally present in most cell types.

* *patent pending*

Stability comparison of traditional luciferase detection kits and the new plus products

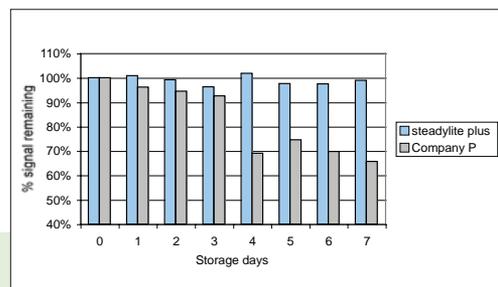
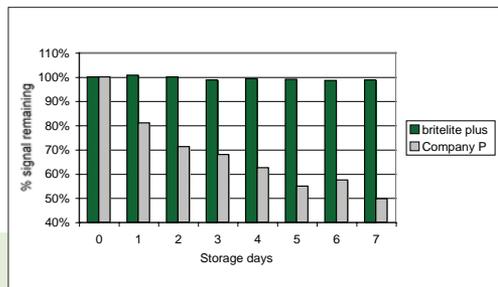


Figure 6 and 7. Stability comparison of lyophilized substrates at 22 °C for an old technology luciferase detection kits from Company P and the new plus products.

Light output and signal decay (steadylite plus and britelite plus)

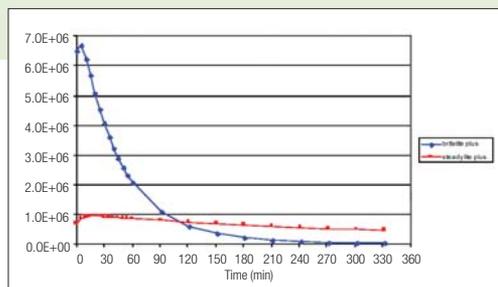


Figure 8. The difference in light output and signal decay is illustrated for 1×10^{-10} gram luciferase.

Sensitivity of steadylite plus compared to britelite plus

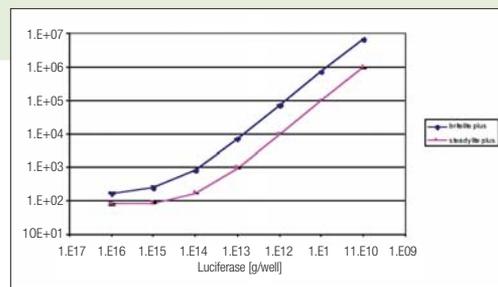


Figure 9. A dilution series of luciferase enzyme where britelite plus has a sub-femtomole detection limit for luciferase compared to steadylite plus.

and steadylite plus



briteLite plus has an extremely strong signal and a half life of 30 minutes and is ideal when additional sensitivity is required and for continuous processing mode (within 30 minutes).

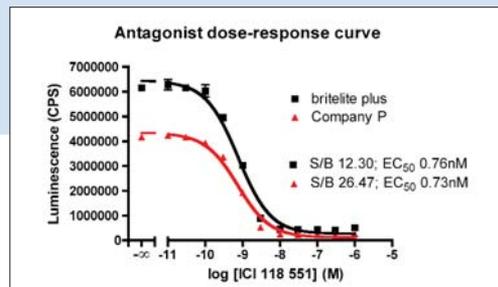
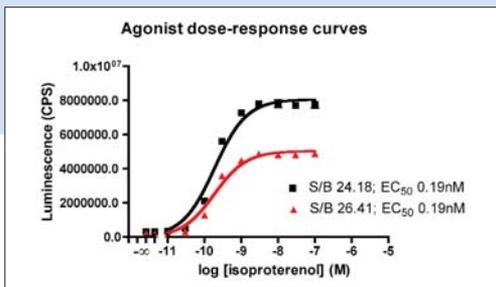
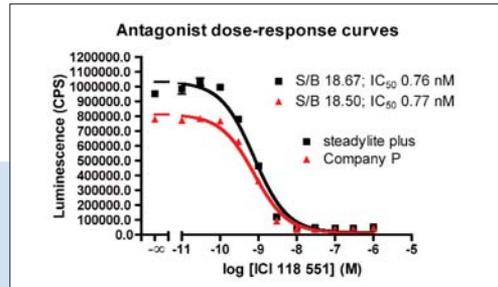
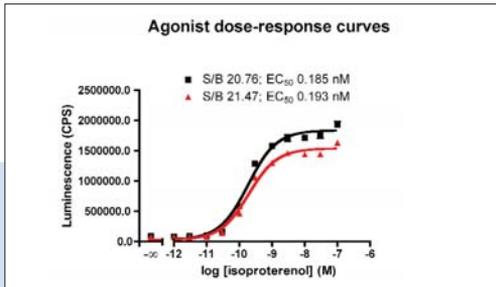


steadyLite plus has an extended half life of up to 5 hours, which allows for batch processing which is particularly useful in high throughput applications.

Part Number		Volume (mL)	Data Points* in 96-, 384- and 1536-well microplates
briteLite plus	steadyLite plus		
6016766	6016756	10	100, 400, 3300
6016761	6016751	100	1000, 4000, 33000
6016767	6016757	500	5000, 20000, 165000
6016769	6016759	1000	10000, 40000, 330000

* The recommended assay volumes are: 100 μ L for 96-well microplates, 25 μ L for 384-well microplates and 3 μ L for 1536-well microplates.

CHO β 2AR



Agonist and antagonist dose-response curves for both steadylite plus (top) and briteLite plus (bottom) compared to Company P products.

cytotoxicity and cell proliferation
 mix-and-measure or time course studies

ATPlite 1step and ATPlite

ATPlite 1step & ATPlite

Fast, reliable detection of ATP for cell quantification

The ATPlite 1step and ATPlite are patented* innovative technologies that measure cell proliferation and cytotoxicity in mammalian cells based on the detection of ATP using firefly luciferase. Light production caused by the reaction of ATP with added luciferase and D-luciferin is proportional to the ATP concentration.

ATP is a marker for cell viability because it is present in all metabolically active cells. Because ATP concentration declines rapidly when cells undergo necrosis or apoptosis, monitoring ATP is a good indicator of cytotoxic, cytostatic and proliferation effects.



ATPlite 1step

ATPlite 1step provides a continuous processing system for the highest sensitivity ATP detection.

- True “mix-and-measure”
- Ultra high sensitivity, 3 times more light output than ATPlite
- Short equilibrium time for faster processing
- Signal half life of 30 minutes

ATPlite

ATPlite is a two-step non separation assay with a prolonged signal half life of 5 hours which allows batch for batch processing. The two-step format allows you to treat your cells, lyse them and then freeze them to be read at different times.

- Two-step format for time course studies: lysate can be stored frozen and tested later
- Quantitate from < 5 cells per well
- Prolonged equilibrium time for batch processing
- Signal half life of 5 hours

Description	Part Number	Volume (mL)	Data Points* in 96-, 384- and 1536-well microplates
ATPlite 1step 	6016736	10	100, 400, 3300
	6016731	100	1000, 4000, 33000
	6016739	1000	10000, 40000, 330000
ATPlite 	6016943	30	300, 1200, 9900
	6016941	100	1000, 4000, 33000
	6016947	500	5000, 20000, 16500
	6016949	1000	10000, 40000, 330000

* The recommended assay volumes are 100 µL for 96-well microplates, 25 µL for 384-well microplates and 3 µL for 1536-well microplates.

* U.S. Pat. 6503723; EP Pat. 117825 (CH, DK, GB, DE 69924127); NL Pat. 1010224; Canada Patent Appl. No. 2345721; Australian Patent Appl. No. 754602; other patents pending.

a high throughput assay for kinase activity

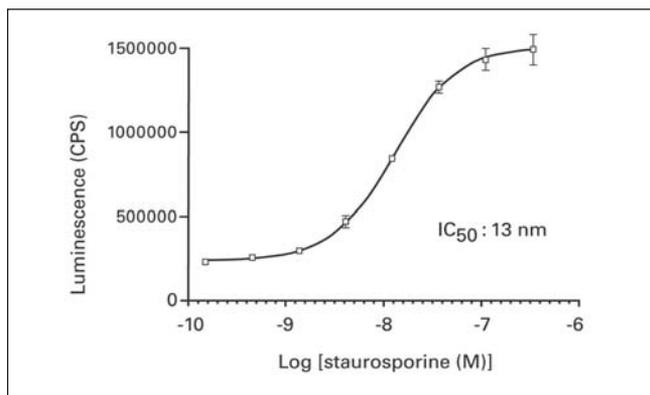
easylite-Kinase

easylite-Kinase is a sensitive ATP monitoring, single-step addition assay system based on firefly luciferase for the evaluation of kinase activity. The system relies on the detection of ATP consumption during a kinase reaction without the need for labeled substrates, labeled enzymes or phospho-specific antibodies.

Due to its cost effectiveness and applicability to uHTS, easylite-Kinase is often used as a primary screening tool followed by a more specific phosphosubstrate measurement technology. It can be read on any luminescence reader, making it a technology that is accessible to almost all laboratories.

Characteristics of easylite-Kinase Assays

- **“Mix-and-measure” homogeneous assay:** simplify the screening process with only one reagent addition step, no separation steps and no luminescence signal stabilization time.
- **High sensitivity and wide dynamic range:** measures a broad range of samples.
- **Ultra-high throughput:** easy to miniaturize from 96- to 1536-well format.
- **Long-lived luminescent signal allows batch processing:** saves valuable time.



The graph above shows the dose response curve (IC₅₀) for staurosporine, a known inhibitor, as determined in a white OptiPlate-384 using easylite-Kinase.

Description	Part Number	Volume (mL)	Data Points* in 96-, 384- & 1536-well microplates
	6016746	10	100, 400, 3300
	6016741	100	1000, 4000, 33000
	6016749	1000	10000, 40000, 330000

* The recommended assay volumes are 100 μ L for 96-well microplates, 25 μ L for 384-well microplates and 3 μ L for 1536-well microplates.

high quality, application-focused

microplates for luminescence assays

Featured microplates for luminescence assays

OptiPlate™: white polystyrene OptiPlate microplates provide excellent light reflection and the highest efficiency with low background for luminescence. Available in 24-well to 1536-well formats.

ProxiPlate™: a shallow well design brings the reagent into closer proximity to the reader's detectors and increases signal. Available in 96-well and 384-well formats.

CulturPlate™: optimized for work with cell-based applications, providing a sterile, tissue culture treated environment with an opaque well bottom. Available in 24-well to 384-well formats with lids.

ViewPlate™: optimized for work with cell-based applications providing a sterile, tissue culture treated environment with a clear well bottom for microscopic visualization or bottom reading. Available in 24-well to 384-well formats.

Microplate	Color (Frame/Bottom)	Applications	Format (Wells)	Product No.	Packaging (case of)
OptiPlate	white/white	Opaque plates	96	6005290	50
		for general luminescence assays	384	6007290	50
		and suspension cell-based assays	1536	6005228	20
ProxiPlate	white/white grey/grey	Opaque shallow-well	96	6006290	50
		for assay miniaturization	384	6008280	50
			384	6008270	50
CulturPlate	white/white	Opaque, tissue culture treated,	24	6005168	50
		sterile for cell-based assays	96	6005680	50
		and luminescence applications	384	6007680	50
ViewPlate	white/clear	Clear bottomed, tissue culture	24	1450-604	56
		treated, sterile for cell-based	96	6005181	50
		luminescence applications	384	6005262	30
		where microscopic visualization			
		or bottom reading is required			

Black versions of the above microplates are also available and are recommended only when the signal is very high (resulting in unacceptable crosstalk). These are suggested for use with the LumiLux. For the ViewLux, high sensitivity grey OptiPlate-384 HS microplates are recommended. Microplates are available in different package configurations.

Our microplates meet the ANSI/SBS-1 (2004) standards.



For more information and a complete listing of our microplate product offering – go to: www.perkinelmer.com/microplates

luminescence detection instrumentation and automation

PerkinElmer offers a wide range of luminescence microplate readers to suit all of your needs, from the VICTOR Light, which is ideal for basic research and assay development, to the uHTS LumiLux Cellular Screening platform for drug discovery screening campaigns. In addition, we supply a wide range of liquid handling solutions to enable you to automate all luminescence applications.



VICTOR Light

VICTOR Light

VICTOR Light is a luminescence counter for microplate applications, including cell-based assays, toxicology screens and more. The capability of the system can be extended through a wide range of optional accessories to meet specific needs. As a small benchtop unit, VICTOR Light can operate as a stand-alone instrument or can be integrated into a robotic system. It provides easy access to waste and syringes, and features a dead volume less than 500 uL. Plus, VICTOR Light is available with a newly designed dispenser.

EnVision Multilabel Reader

The EnVision Multilabel Plate Readers are fast, sensitive and versatile benchtop readers that deliver optimized performance in every application and for every label. Their unique design features modular label-specific optical mirror modules, high energy flash lamps, and high speed detectors.

The plate conveyor and stackers have been optimized to achieve maximum possible speed, with parallel functionality as well as fast physical movements. The EnVision instruments are easily integrated into robotic systems and are designed to provide the greatest configuration flexibility including accepting microplates from 1- to 3456-wells. The EnVision also features dedicated LASER excitation modules for optimal LANCE™ (TRF) and AlphaScreen® technology performance.



EnVision

EnVision meets today's needs of cell-based assays

When combining the speed of EnVision with the high precision dispenser unit and temperature control, the versatile EnVision can perform fast kinetic measurements, enzyme assays and numerous other cell-based drug discovery assays.

high quality luminescence

instrumentation and reagents

TopCount and MicroBeta

The TopCount and MicroBeta are the most popular scintillation and luminescence counters in the world for low and medium throughput applications.

Super Sensitive Luminescence Measurements

Thanks to high speed photon counting circuitry, MicroBeta and TopCount have best-in-class glow-type luminescence performance with linear responses in excess of 20 million CPS and backgrounds below 100 CPS. Temperature control features guarantee the optimum counting conditions for luminescence assays.

If your applications require flash luminescence, choose MicroBeta JET. It uses one or more reagent injectors for measuring prompt (or 'flash') reactions. MicroBeta JET can be used for flash luminescence applications such as aequorin/ Ca^{2+} measurement and dual label reporter gene assays.



ViewLux Screening Platform

The ViewLux is an ultra-high throughput microplate imager for high sensitivity and fast measurement of light from fluorescence polarization (FP), fluorescence intensity, time-resolved fluorescence (TRF), luminescence and absorbance assays.



Because the instrument reads entire plates in one exposure, throughput is not affected by plate density. ViewLux provides today's leading option for 1536-well plates, and supports even higher density plates, as they become available. Using 1536-well plates and, for example, fluorescence intensity, ViewLux can allow throughputs in excess of 200,000 samples per hour under continuous operation.

The instrument supports both robot loading and batch mode operation.

Up to 64 plates can be loaded for unattended operation. Users can alternate between batch and robot loading according to needs.



LumiLux Cellular Screening Platform

The new LumiLux Cellular Screening Platform is an ultra high throughput cellular imaging platform for both kinetic flash and glow luminescence assays with advanced liquid handling functionality. An instrument with walk-away capability, it provides sensitive detection based on patented fiber-optic imaging technology, state-of-the-art liquid handling, and sophisticated data analysis tools— all integrated into one advanced solution. Plus, LumiLux introduces a breakthrough advance—a unique cell stirrer that permits the use of suspension cells in uHTS screens.

- Performs both flash and glow luminescence assays in 96-well, 384-well and 1536-well formats



- Works with suspension cells as well as adherent cells
- Superior sensitivity and dynamic range reducing the number of cells required per well and the option to work with either transients or stably transfected cell lines
- Fully automated, walk-away platform enabling >100,000 data points per day (aequorin screening)



The JANUS Automated Workstation

As your workload grows, PerkinElmer also offers a range of automated pipetting solutions with our modular JANUS® platform. Powered by Packard® innovation, the JANUS Automated Workstation delivers precision liquid handling and flexible pipetting capabilities for pre-dilutions and serial dilutions. Our decades of experience mean we have the technical expertise to completely automate your applications to maximize your efficiency and results quality.

As your applications demand becomes even larger, choose a complete integrated solution, the JANUS Cellular Workstation. The Cellular Workstation combines JANUS liquid handling and automation technology with labware movement and dynamic scheduling, the EnVision multilabel detection plus pre-defined lite application templates for a single integrated system solution to run your PerkinElmer cellular assays. You'll get total assay automation—hands-off, efficient, and precise.

luminescence instrument selection guide

Detection Instrument			VICTOR Light	MicroBeta Jet
Features		Glow	Up to 50,000 samples per day	Up to 50,000 samples per day
	Throughput	Flash (Aequorin)	Up to 1,000 samples per day (96-well)	Up to 6,500 samples per day (96-well)
	Additional/Optional Read Modes		Dedicated Luminescence	Radiometric Luminescence
	Temperature Control		+2 °C to 45 °C	PMT cooling option; no temp control on sample
	Automation		ActiveX/COM interfaces.	Robot loading
	Injectors		1-4	1-2/PMT channel; no injector for 12 detector model available
	Injection volumes		5–350 µL in 1 µL	10-250 uL; 1 uL increments
	Shaker		Linear, Orbital & Double Orbital	NA
Microplate Format		1-384 well plates	24, 96, 384	
Luminescence Applications	Flash		✓	✓
	Glow		✓	✓
	Dual		✓	✓
	Dual emission (such as BRET)		✓	
	Aequorin		✓	✓
	Kinetics		✓	✓
	Bottom Read			✓
Luminescence Specifications	Detector		low noise photo multiplier tube	1, 2 or 6 parallel PMTs (12 PMTs without dispensers available for glow)
	Dynamic Range		6 logs	5.5 logs
	Sensitivity		ATP typically < 20 pM in glow assays	ATP typically < 10 pM in glow assays luciferase < 1fg/well in glow assays; 96-well
	Crosstalk		< 0.02% for 96-well B&W Isoplates; <0.2% for 384	< 0.01% (96-well)



TopCount	EnVision	ViewLux	LumiLux
Up to 50,000 samples per day	100,000 samples per day	200,000 samples per day	>200,000 samples per day
N/A	Up to 1,200 samples per day (96-well)	N/A	384 head: 384-well agonist assay – 120,000 cpds/8hr day; antagonist assay – 80,000 cpds/day; 1536 head: 1536-well agonist assay – 450,000 cpds/8hr day; antagonist assay – 300,000 cpds/day
Radiometric Luminescence	Ultrasensitive Luminescence option; TRF, FI, FP, Absorbance AlphaScreen	TRF, FI, FP, Absorbance	Radiometric (red and blue) Luminescence
Cooling only (PMT and Sample)	+2 °C to 45 °C	+0 °C to 37 °C	No
Robot loading	Robot loading	Robot loading	ActiveX /COM interfaces
NA	1-2	NA	384 or 1536 dispense head
NA	2–475 µL in 1 µL	NA	0.5-10ul (P10 tips); 0.5-30uL (P30 tips)
NA	Linear, Orbital & Double Orbital	NA	No, mixing is achieved through multiple aspiration and dispense steps
24, 96, 384	1-3456 well plates	1-3456 well plates	96, 384 and 1536
	✓		✓
✓	✓	✓	✓
	✓		2 reporters with different end points can be read
	✓		
	✓		✓
	✓	✓	✓
	✓	✓	✓
2, 6 or 12 parallel PMTs	Dedicated low noise PMT for Luminescence, 1-2 parallel PMTs for other technologies	-100 °C cooled CCD	-80 °C cooled CCD with fiber-optic taper
5.5 logs	6.5 logs	3.5 logs	3.5-4 logs for single measurement; 4.5-5 logs for two measurements
ATP typically < 10 pM in glow assays; luciferase < 1fg/well in glow assays; 96-well	ATP (384-well plate, 50 µL) < 10 pM	ATP (384-well plate, 50 µL < 10 pM)	Flash Luminescence (Aqualite): 384-well (400mS) = 0.3pg/well (1.4 x 10e-17 moles/well), 1536 (400mS) = 0.5pg/well (2.3 x 10e-17 moles/well)
<0.1% (96-well)	< 0.02% (384-well)		< 0.1% (384-well) and <1% (1536-well when one hot well, at 50% of the dynamic range is surrounded by 100 times less active wells)

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For a complete listing of our global offices, visit www.perkinelmer.com/lasoffices

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