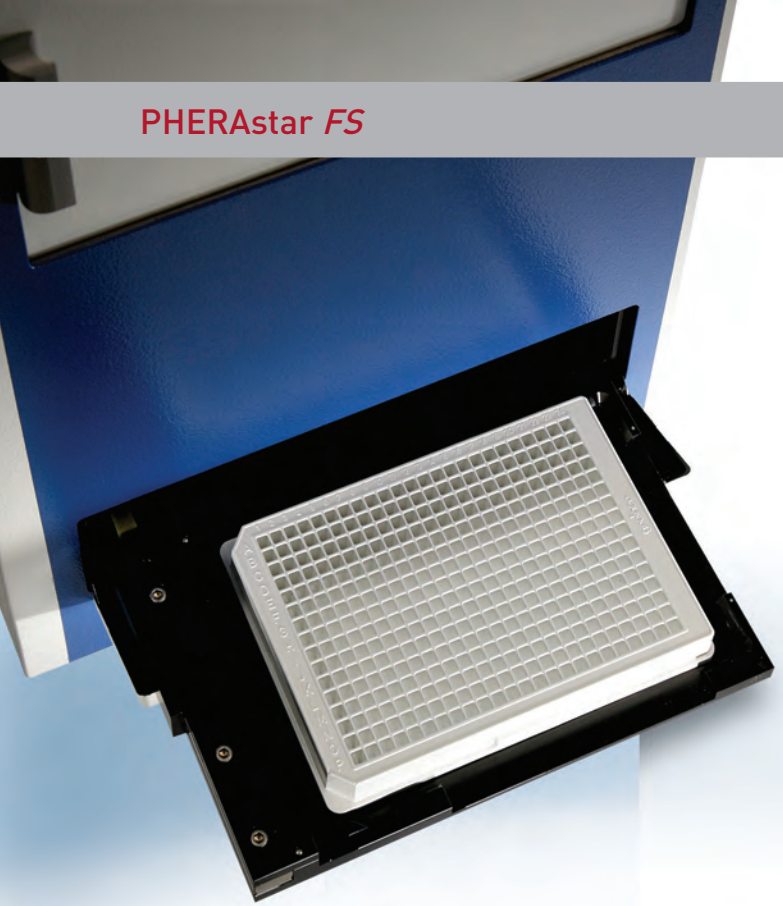
PHERAstar® *FS*

The Gold Standard for High-Throughput Screening

BMG LABTECH

The Microplate Reader Company



The **Gold Standard** for High-Throughput Screening

Whether you need speed, sensitivity or flexibility, the PHERAstar *FS* is the ultimate microplate reader for your research and HTS applications.

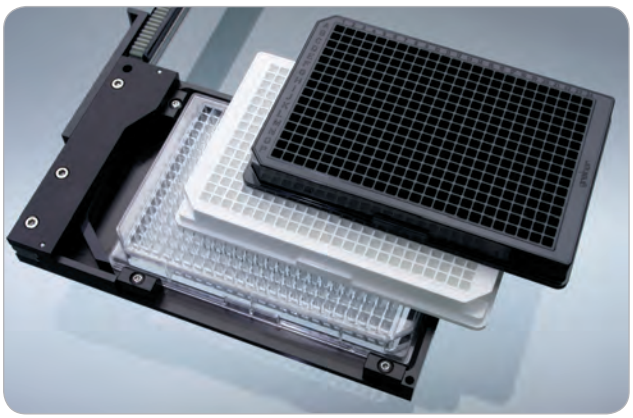
Best Performance for all HTS Assays

The PHERAstar® *FS* is the next generation microplate reader for High-Throughput Screening, specifically conceived by BMG LABTECH for highest sensitivity and speed.
Its new and unique features make it superior to any other microplate reader currently on the market. German engineering with the latest technology makes the PHERAstar *FS* the Gold Standard for HTS. It performs all the leading non-isotopic detection technologies such as:

- Ultra-fast UV/Vis Absorbance Spectra
- Fluorescence Intensity, including FRET
- Fluorescence Polarization/Anisotropy
- Time-Resolved Fluorescence, including TR-FRET
- High-end AlphaScreen®/AlphaLISA®
- Luminescence (flash and glow), including BRET

Sequential Dual Excitation, Simultaneous Dual Emission and ratiometric calculations are just some of the key features of the PHERAstar *FS* for multichromatic applications such as FRET, TR-FRET, BRET and FP.

Application specific Optic Modules can be activated by a simple mouse click, and all the necessary components are selected automatically. Assay flexibility is enhanced by top/bottom reading, onboard reagent injectors, precise temperature control and multi-mode shaking capabilities. The PHERAstar *FS* provides uncompromised sensitivity, speed and dynamic range in plate formats up to 3456.



Any plate format up to 3456.

Tandem Technology

The PHERAstar *FS* incorporates BMG LABTECH's unique Tandem Technology, which is based on two technological concepts: an ultra-fast UV/Vis spectrometer for absorbance and high-performance optical filters for all other detection modes.

Innovative Optical Design

The outstanding sensitivity of the PHERAstar *FS* is based on a new, innovative lens-based optical design which is composed of three independent light sources, Simultaneous Dual Emission detection, and software-controlled top/bottom reading. Depending on the application, users can choose one of the following light sources:

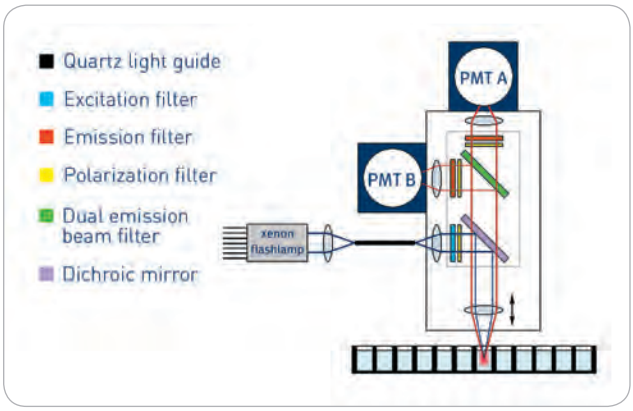
- High energy xenon flashlamp
- Nitrogen laser for TRF/TR-FRET
- Solid-state laser for AlphaScreen®/AlphaLISA®

Two matched pairs of photomultiplier tubes (PMTs) are used in the PHERAstar *FS*. The first pair of PMTs is for simultaneous luminescence and fluorescence detection, and the second pair of PMTs detects time-resolved fluorescence based assays.

Distinct advantages of the nitrogen laser include increased assay window and the "flying mode" measurement for TR-FRET based assays. A single laser flash excites the donor molecules sufficiently so that measurement in a well takes place without stopping plate movement. Thus, reading time for an entire plate is significantly decreased.

Simultaneous Dual Emission

BMG LABTECH pioneered the technique of Simultaneous Dual Emission detection for microplate readers. Because numerous assays require detection of two wavelengths, Simultaneous Dual Emission offers a significant advantage by cutting read times in half. It corrects flash-to-flash variations, assay effects such as photobleaching, decaying kinetic signals, or fluctuating conditions like temperature, pH, and evaporation. Simultaneous Dual Emission detection can be used in any assay that measures two wavelengths or polarization vectors, including FP, FRET, and HTRF®.



Schematic layout of the Simultaneous Dual Emission (SDE) optical pathway incorporated in the PHERAstar FS.

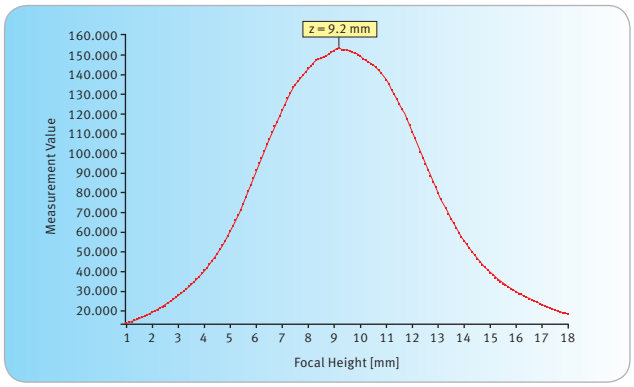
Optic Modules

The assay specific Optic Modules are the core elements of the PHERAstar *FS*. They contain all the necessary optical components including excitation and emission filters, dichroic mirrors, beam splitters and polarization filters. The PHERAstar *FS* can accommodate up to six Optic Modules and the user can easily add or replace them within seconds. All Optic Modules are barcoded, and the PHERAstar *FS* automatically selects the proper module for your assay.

Assay specific Optic Modules, all inclusive, installed in seconds.

Focal Height Adjustment

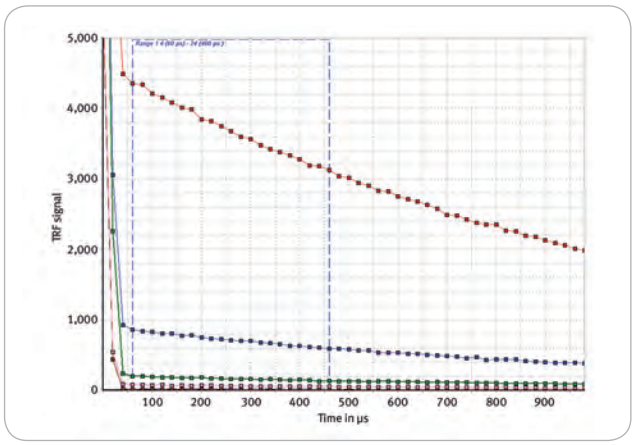
The PHERAstar *FS* incorporates automated focal height adjustment for both top and bottom reading at a resolution of 0.1 mm. The optical system directs the excitation light to a small focal point in the center of the well, giving excellent sensitivity in all plate formats up to 3456. This feature eliminates the influence of microplate formats, sample volumes, surface tension and evaporation. The automated focal height adjustment ensures the best signal-to-noise ratio for every plate, application and volume.



The automated focal height adjustment ensures the best signal-to-noise ratio in all detection modes and all plate formats.

Decay Curve Monitoring

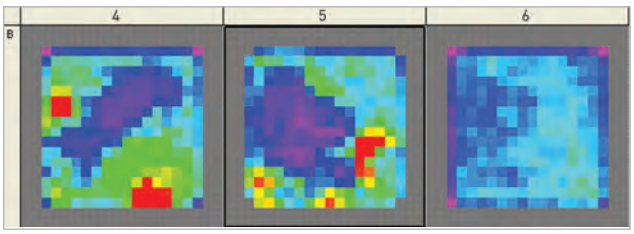
The Decay Curve Monitoring feature of the PHERAstar *FS* enables users to visualize the time-resolved emission curve and to optimize timing parameters for AlphaTechnologies and TRF modes.



Unique TRF Decay Curve Monitoring feature for assay optimization.

Advanced Cell Layer Scanning

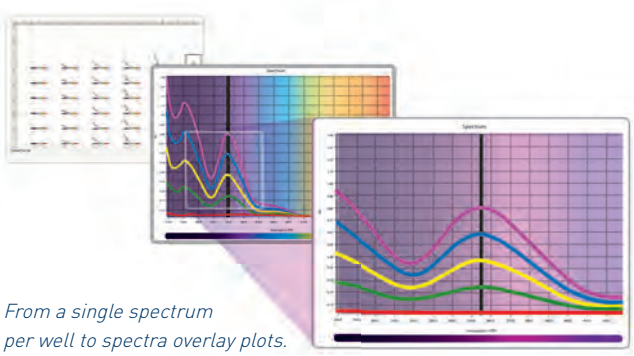
Unmatched top and bottom reading performance is offered in the PHERAstar *FS*. Switching between these modes is automatically carried out via software. Bottom reading for high resolution cell based scanning is achieved through innovative engineering using a series of lenses and mirrors. In Well Scanning mode, the PHERAstar *FS* can take multiple measurements in each well with up to 30 x 30 data points. The software then displays each scan point graphically and creates a map for each well. BMG LABTECH's unique Orbital Averaging can be used to measure nonhomogeneous well content. Using this mode, the PHERAstar *FS* takes several measurements on a defined orbit, collects the data, and calculates an average for each well.



Well Scanning for advanced cell based assays.

Ultra-fast Absorbance Spectrometer

The PHERAstar *FS* is the first multidetection HTS microplate reader to integrate a spectrometer for absorbance measurements. This technology can capture a full UV/Vis absorbance spectrum (220 to 1000 nm) at selectable resolutions from 1 to 10 nm. A full absorbance spectrum can be measured as fast as one second per well, which is significantly faster than any conventional method. Furthermore, users can capture up to eight discrete wavelengths simultaneously in a single measurement with no wavelength switching.



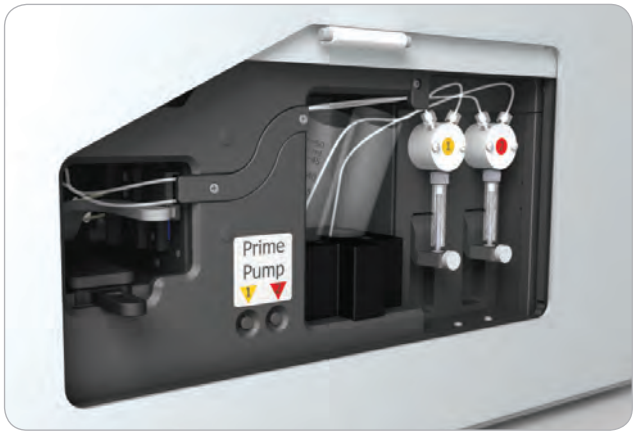
From a single spectrum per well to spectra overlay plots.

Sensitivity and Speed

The PHERAstar *FS* combines fast read times necessary for High-Throughput Screening and the sensitivity to read small volumes. The user can always find the best combination of sensitivity and speed by choosing the number of flashes. In single flash mode, the PHERAstar *FS* can read a 1536-well plate in 27 seconds, making the PHERAstar *FS* among the fastest in the microplate reader industry.
The unsurpassed sensitivity of the PHERAstar *FS* detection system provides outstanding S/N, %CV, and Z' values, even at low concentrations and small assay volumes.

Smart Reagent Injection

Many popular assays require the ability to monitor a signal before, during and after the addition of a reagent to a well. The two onboard Reagent Injectors are ideally positioned so reagents can be added to the well currently being read. Thus, concurrent reading and injection ensures that even the initial part of fast kinetic reactions can be monitored. The injectors are readily accessible and are housed within the instrument to safeguard any light sensitive reagents.



Onboard Reagent Injectors for simultaneous injection and reading.

An exceptionally small dead volume and back flushing ensure that precious reagents are conserved. Users can tune all parameters, such as plate shaking, injection speed, timing, and the number of injections per well. Delivery volumes are adjustable for each well, allowing users to automatically produce dilution schemes and concentration gradients across the microplate.

Automation

Automation is a key feature in High-Throughput Screening. BMG LABTECH understands this and has designed its microplate readers to be automated. All BMG LABTECH microplate readers have similar x-y dimensions and plate in/out locations. This minimizes the cost of automation solutions for customers. BMG LABTECH has cultivated collaborations with various robotics companies over the years and thus many proven robotic integrations are available.

The small standardized reader footprint and robotic software interface makes it easy to integrate the PHERAstar *FS* into all leading robotic platforms. In order to provide simple plate management, the PHERAstar *FS* comes with three integrated microplate barcode readers capable of reading the east, west and south sides of a microplate.

Microplate Stacker

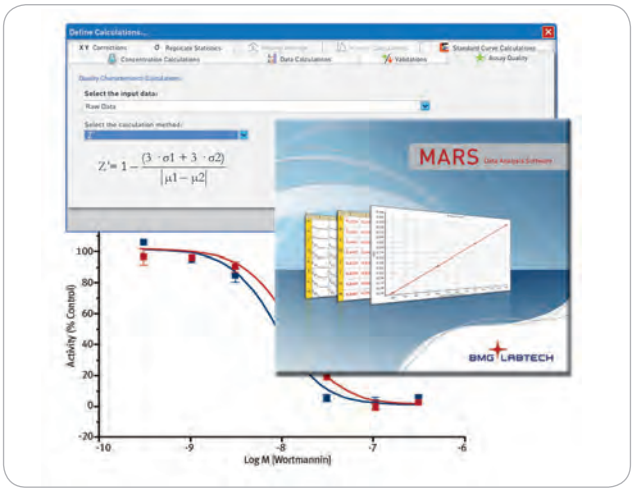
For medium level throughput, BMG LABTECH offers a Stacker that can be used with the PHERAstar *FS*.
The Stacker is an ideal solution for mid-throughput labs that wish to have the small footprint of an automated plate feeder along with the simplicity and reliability the Stacker offers. It provides loading, unloading, restacking and a continuous load feature of up to 50 microplates.

Automated plate handling with Stacker.

The Control Software of the PHERAstar *FS* operates the Stacker and a script mode gives the user unlimited flexibility to run diverse assays. This function can be used to choose different test definitions for different plates in one batch run or to perform more than one measurement on one plate.

Control and MARS Data Analysis Software

The PHERAstar *FS* software package provides an extensive range of possibilities for both test protocol definitions and data analysis. It is fully compliant with FDA regulation 21 CFR Part 11. The Control Software allows users to define instrument parameters and test protocols. For assay development, kinetic measurements can be interrupted at any time in order to change reaction conditions.



MARS Data Analysis Software for automated data reduction.

The Data Analysis Software MARS allows the user to display and process data using predefined templates. One example is the automatic calculation of enzyme kinetic parameters (V_{max} and K_m) with a variety of fits based on Michaelis-Menten or Lineweaver-Burk equations.
The software is also capable of creating standard curves based on the following curve fitting algorithms to calculate values such as EC_{50}, IC_{50} and r^2:

- Linear regression fit
- 4 parameter fit
- Segmental regression fit
- Cubic spline fit
- 2nd and 3rd polynomial fit
- Point-to-point fit
- Enzyme kinetic (e.g. Michaelis-Menten; Lineweaver-Burk)

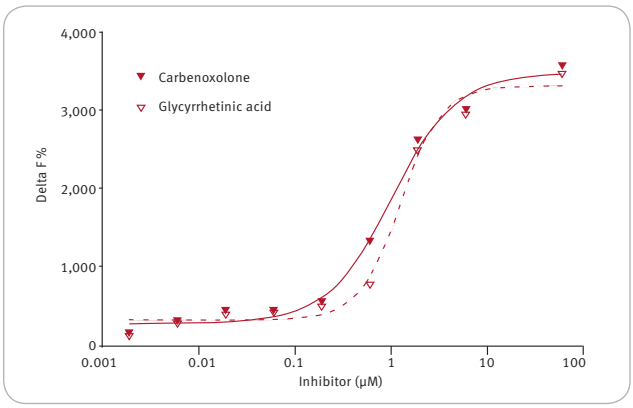
The MARS standard curve wizard creates a step-by-step calculation of a standard curve and important parameters such as S/N, Delta F % and Z' are easily obtained.

Applications Center

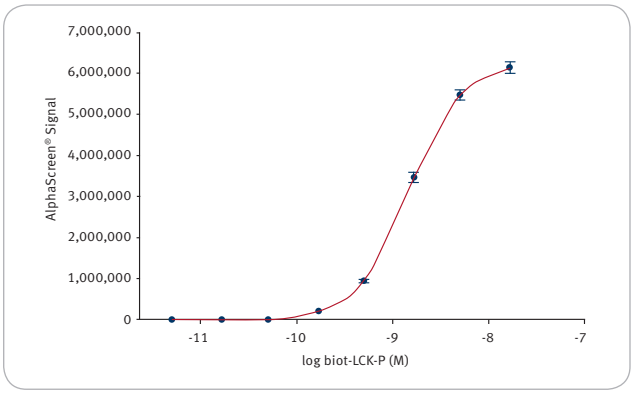
A perfectly engineered microplate reader is only part of the solution. The reader's ability to effectively perform all of the leading applications is the final piece of the puzzle. With the PHERAstar *FS*, BMG LABTECH offers a unique combination of features to support all major existing applications as well as future needs. Applications include:

- Protein-protein interactions
- Genotyping
- Molecular binding assays
- Receptor-ligand binding
- DNA, RNA, and protein quantification
- Enzyme activity and kinetic assays
- Cell based assays
- Reporter gene assays

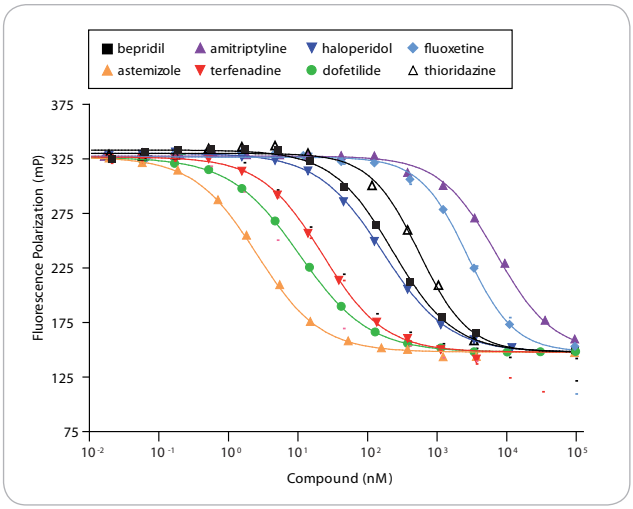
The PHERAstar *FS* provides excellent performance in all HTS applications, including HTRF®, AlphaScreen® and FP assays, illustrated by the following examples:



HTRF® assay showing response curves of cortisol inhibitors[1].



Biot-LCK-P titration curve measured in AlphaScreen® mode[2].



Competitive binding of known hERG blockers using the Predictor™ hERG Fluorescence Polarization (FP) assay[3].

BMG LABTECH continuously works with all the leading reagent companies to optimize instrument settings for their existing assays and their newest chemistries.

Visit BMG LABTECH's Applications Center online to find references to all applications, listed as:

- Application notes
- Scientific posters
- Peer-reviewed papers

BMG LABTECH's comprehensive searchable applications database reflects more than 20 years of expertise and innovations in microplate reading technology. Over 3,800 references exemplify the flexibility and versatility of our readers, as well as their use in the chemical and biological sciences.

Support and Training

BMG LABTECH operates globally through an extensive network of subsidiaries and trained distributors. Customers can rely on qualified support and assistance with regard to software, assay development, or general enquiries related to the PHERAstar *FS* and all other BMG LABTECH microplate reading solutions.

[1,2,3] The graphs were taken from BMG LABTECH's Application Notes AN 154, AN 153 and AN 173.

Transcreener®
DNA / RNA quantifications
Protein quantifications
HTRF®
Ca2+ assays
Reporter gene assays
Kinase activity
AlphaScreen®/AlphaLISA®
DLR™
Protease activity
LanthaScreen®
ORAC
HTS
Immunoprecipitation
Dual luciferase assays
BRET assays
Apoptosis
ROS detection
Enzyme kinetics
PCR product quantifications
NADH / NADPH assays
ELISA
Cell Viability
ATP and ADP detection
LANCE®
Binding studies
Enzyme activity
Gene expression
Solubility tests
DELFIA®
SNP Genotyping
FRET assays

PHERAstar *FS* - Technical Specifications

Due to the modularity of BMG LABTECH's instruments, all or combinations of the features below can be installed at purchase or upgraded at any time. Please contact your local representative for more details or a quote.

Detection Modes	Fluorescence Intensity - including FRET Fluorescence Polarization/Anisotropy High-End AlphaScreen®/AlphaLISA® Luminescence (flash and glow) - including BRET Time-Resolved Fluorescence - including TR-FRET UV/Vis Absorbance Spectra	
Measurement Modes	Top and bottom reading Endpoint and Kinetic measurements Sequential Multi Excitation measurements Sequential Multi Emission measurements Simultaneous Dual Emission measurements Real-time ratiometric measurements Well Scanning	
Microplate Formats	Up to 3456-well plates, user-definable	
Light Sources	High energy xenon flashlamp Nitrogen Laser for TRF and TR-FRET Solid state Laser for AlphaScreen®/AlphaLISA®	
Detectors	Four photomultiplier tubes, optimized for different detection modes	
Optic Module Capacity	Up to six application-specific and barcoded Optic Modules built in	
Z-Adjustment	Automatic focal height adjustment (0.1 mm resolution)	
Spectral Range	230 - 750 nm or 230 - 900 nm for FI, FP 230 - 750 nm for LUM 230 - 900 nm for TRF 220 - 1000 nm for ABS	
Sensitivity	FI (top)	0.4 pmol/L sodium fluorescein (8 amol/well) (384) 2.0 pmol/L sodium fluorescein (16 amol/well) (1536)
	FI (bottom)	1.0 pmol/L sodium fluorescein (<50 amol/well) (384)
	FP	1.0 mP SD at 1 nmol/L sodium fluorescein (384) 3.0 mP SD at 1 nmol/L sodium fluorescein (1536)
	TRF	25 fmol/L europium (0.5 amol/well) (384) 100 fmol/L europium (0.8 amol/well) (1536)
	HTRF®	Reader Control Kit (Eu) after 18h (384) Delta F > 1100 % (High Calibrator) Delta F > 25 % (Low Calibrator)
	LUM	10 amol/well ATP (384)
	AlphaScreen®	< 100 amol/well* (384)
	ABS with spectrometer	Spectral range: 220 - 1000 nm Full spectrum captured in < 1 s/well Selectable spectral resolution: 1 nm, 2 nm, 5 nm, 10 nm OD range: 0 - 4 OD Accuracy: < 1 % at 2 OD Precision: < 0.5 % at 1 OD and < 0.8 % at 2 OD
Read Times	1 flash	14 s (384), 27 s (1536)
	10 flashes	38 s (384), 1 min 52 s (1536)
	50 flashes	1 min 29 s (384), 5 min 18 s (1536)
Reagent Injection	Up to 2 onboard reagent injectors Injection at measurement position (6 to 384-well) Individual injection volumes for each well (3 to 350 µL) Variable injection speed up to 420 µL/s Up to four injection events per well Reagent back flushing	
Shaking	Linear, orbital, and double-orbital with user-definable time and speed	
Barcode Reader	Up to three integrated microplate barcode readers	
Incubation	+5 °C above ambient to 45 °C	
Software	Multi-user software package including Reader Control and MARS Data Analysis Software, FDA 21 CFR Part 11 compliant	
Dimensions	Width: 45 cm, depth: 51 cm, height: 47 cm; weight: 49 kg	
	Accessories	
Stacker	Magazines for up to 50 plates - continuous loading feature	
THERMOstar	Microplate Incubator and Shaker	
Optic Modules	Available for all applications	
Upgrades	Upgrades to include options such as additional detection modes, reagent injectors, etc. are available. Please contact your local representative for more information.	

* Limit of detection: < 100 amol of biotinylated and phosphorylated polypeptide (P-Tyr-100 assay kit, PerkinElmer, #6760620C), measured in white 384 small volume microplates (17 µL / well)

Limit of detection (sensitivity) was calculated according to the IUPAC standard: $3x(SD_{blank})$ / slope

Specifications are subject to change without notice.

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