

# Spark<sup>®</sup>

MULTIMODE MICROPLATE READER

TECAN.

SPARK.



# The fully flexible detection platform with smart automation.

Made for discovery, today and tomorrow.

The Spark multimode microplate reader platform offers solutions to suit virtually any life science research or drug discovery application. It allows researchers to freely configure the reader to their current needs, and is fully upgradable to access other techniques and features in the future.

Spark provides unparalleled wavelength accuracy, with a dedicated High Speed Monochromator for absorbance measurements. Together with a cuvette port and the patented NanoQuant Plate™, it provides an all-in-one solution for ELISAs, low volume DNA/protein quantification and fast spectral scanning.

At the heart of the instrument are its unique Fusion Optics for fluorescence, allowing any combination of filters or monochromators on both the excitation AND emission sides for every measurement; you no longer have to choose between sensitivity OR flexibility for your assays. This option features the latest generation QuadX Monochromators™ and dichroic mirrors, offering variable bandwidth selection and full wavelength flexibility to

provide exceptional measurement performance and speed.

Spark's multi-color luminescence module offers unparalleled flexibility for virtually any luminescence measurement, including flash, glow, BRET and laser-based Alpha Technology.

The instrument's bright field cell imaging capabilities, together with its incubator-like environmental control, enable long-term cell-based experiments and live monitoring of cell growth. The conditional workflow automation minimizes hands-on times and increases reproducibility, enabling long-term experiments with precious cell lines.

To ensure complete confidence in your data, Spark's Te-Cool™ module allows the user to set the reader temperature at or below ambient, offering complete environmental independence for more accurate and reliable results, regardless of the time of day or season.

## Spark capabilities

### Detection modes

- Absorbance – incl. UV/Vis spectra
- Fluorescence top & bottom – incl. spectra
- Time resolved fluorescence (TRF) – incl. spectra
- FRET
- TR-FRET
- Fluorescence Polarization (FP)
- Luminescence – glow, flash, multicolor, spectra
- AlphaScreen®, AlphaLISA® & AlphaPlex®
- Automated live cell imaging – cell counting and confluence

### Additional features

- Absorbance cuvette port
- NanoQuant Plate
- Temperature control (RT+3°C – 42°C)
- Liquid dispensers with reagent heater and stirrer
- CO<sub>2</sub> & O<sub>2</sub> control
- Evaporation protection (humidity cassette)
- Te-Cool™ (active temperature regulation from 18-42 °C)
- Integrated lid handling
- QC tools for IQOQ services
- Connect™ microplate stacker

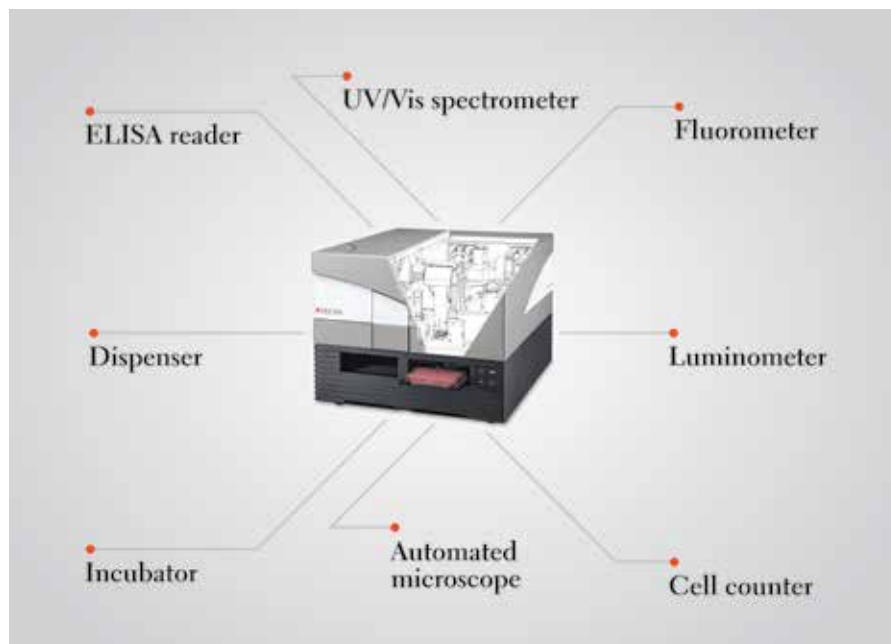
### Applications

- ELISA
- Low-volume DNA/RNA quantification
- Nucleic acid labeling efficiency
- Protein quantification
- Reportergene assays
- HTRF®
- Transcreeper®
- DLR®
- BRET – including NanoBRET®
- Cell counting and viability
- Confluence assessments
- Cell migration and wound healing



## Full modularity and upgradeability

Spark allows free combination of a broad range of modules, giving researchers the ability to configure the system to exactly match their needs. It is fully upgradeable, ensuring a future-proof investment with the flexibility to grow to meet future requirements.



## Absorbance UV/Vis Spectrometer

With its High Speed Monochromator, Spark provides unparalleled wavelength accuracy for DNA and protein analysis:

- Full absorbance spectrum data – from 200 to 1,000 nm – in less than five seconds
- Excellent accuracy in the deep UV range (230-260 nm) for improved DNA/RNA analysis
- An OD range from 0-4 ensures good linearity, requiring fewer dilutions and less manual pipetting

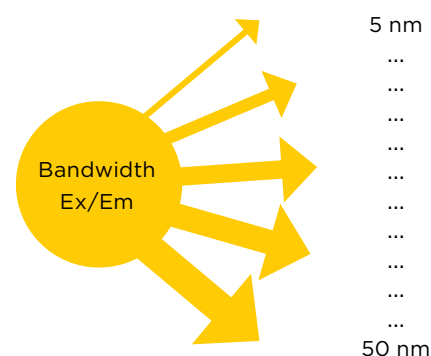


## Fluorescence

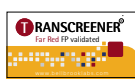
Spark's unique Fusion Optics offer a free choice of filters or monochromators, not just in the same instrument, but in the same measurement. This eliminates any compromise between sensitivity and flexibility, and is especially beneficial for assay development. All optical modules are available as standard or enhanced versions, with full upgradeability.

**High sensitivity across the spectral range** – The high performance of the PMT used for fluorescence detection delivers exceptional sensitivity over the complete spectral range, from green dyes to red dyes.

**Sensitivity** – Combining QuadX Monochromators with dichroic mirrors gives the Spark reader industry-leading sensitivity. These mirrors reduce unwanted noise, particularly for fluorophores exhibiting narrow excitation and emission spectra. Choose from three built-in dichroic mirrors, or even choose a user-selectable mirror.



Variable bandwidth improves sensitivity for highly demanding FRET assays by allowing excitation and emission bandwidths separately.





User-selectable deep blocking dichroic mirror increases sensitivity for fluorophores with narrow excitation and emission spectra.

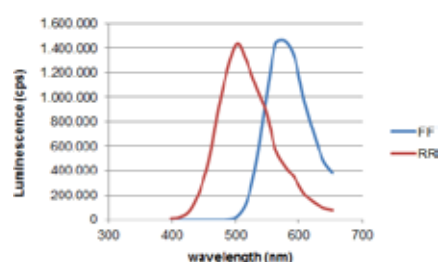
**Full wavelength flexibility** – The Premium QuadX Monochromators offer unparalleled wavelength accuracy and precision, as well as flexible bandwidth selection. In combination with the system's dichroic mirrors, this ensures enhanced flexibility and filter-like performance for assay development and screening.

### Automated cell-based experiments

Spark overcomes the typical loss in sensitivity associated with bottom reading of cell-based assays using a lens-based system which guides the light to a focal point on the cells. In combination with cell confluence measurements, it enables fully automated, parallel monitoring of cell growth and fluorescence signal intensities while the cells are incubated inside the measurement chamber.

### Fluorescence polarization

Spark's unique Fusion Optics allow flexible set-up of FP experiments using any combination of filter- or monochromator-based optics.



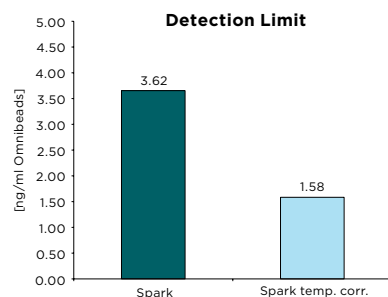
Emission spectral scan of Renilla (RR) and Firefly (FF) luciferases recorded with the Spark.

## Luminescence

Spark offers a choice of luminescence modules, allowing standard or multi-color measurements using 40 user-selectable filters. This enables monochromator-like scanning from 390-660 nm with the sensitivity of filter-based measurements. The system uses a dedicated PMT to offer single photon counting without compromising other detection modes, giving excellent sensitivity over a dynamic range of  $10^9$  using three OD attenuation filters. The use of dedicated fibers for each plate format (96 to 1,536 wells) minimizes crosstalk and gives luminometer-like performance. In combination with the instrument's injector module, this offers unprecedented freedom for luminescence applications, including glow, flash, multi-color, scanning and BRET.

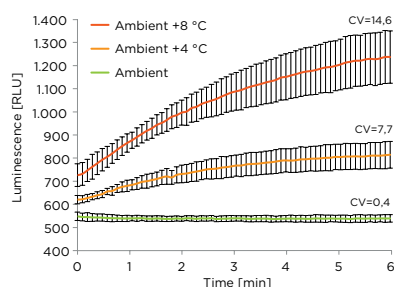
### Alpha Technology

Alpha Technology – including AlphaScreen®, AlphaLISA® and AlphaPlex® – is a luminescent bead-based assay technology designed for the measurement of biological interactions. Spark is equipped with a high performance laser excitation source and IR sensor for well-by-well temperature correction, ensuring better sensitivity, uniformity and linearity for Alpha Technology assays. SparkControl includes pre-determined filter settings for AlphaScreen, AlphaLISA and AlphaPlex, as well as user-selectable settings for future Alpha Technology applications.



## Improved temperature control with Te-Cool

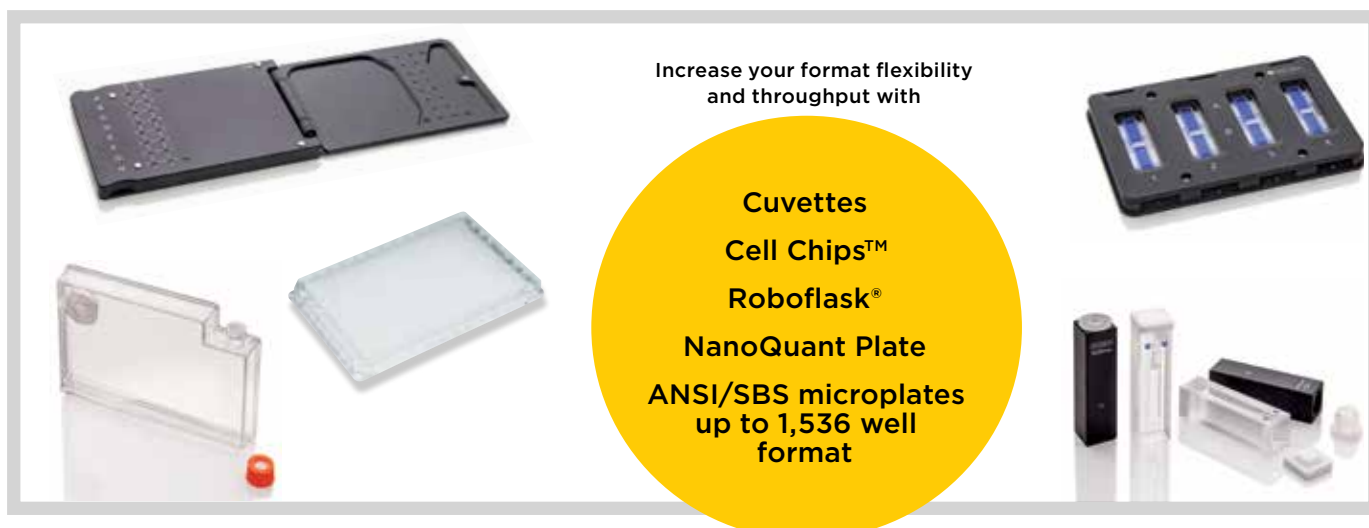
A stable temperature is a prerequisite for reliable results. For most readers, the specified minimum temperature inside the measurement chamber is defined as a few degrees above ambient. This can vary significantly depending on the time of year and location, and so will your results. Temperature gradients can also occur across a microplate with many readers, leading to poor precision and variability.



Enzymatic assays are temperature sensitive. A luminescence assay was measured kinetically over a time course of 10 min. Best results with lowest CV were obtained when setting Spark at a constant ambient temperature with Te-Cool.

Whether you are performing enzymatic reactions – such as luciferase assays – running temperature-dependent Alpha Technology assays, or studying assay kinetics in live organisms, you need precise, controllable cooling to ensure optimal performance. Spark's patent-pending Te-Cool module allows you to perform assays at, or even below, ambient temperatures, maintaining your specified temperature for long periods.





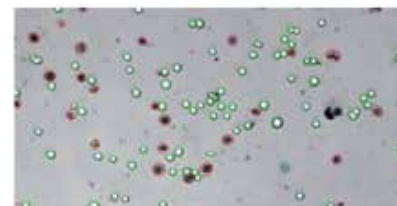
## Automated microscope

Spark's bright field imaging optics offers a 4x objective with laser-based autofocus. This system can perform image acquisition and confluence determinations in microplates, or cell counting applications using Tecan's disposable Cell Chip. In addition, a Live Viewer option in SparkControl offers microscope-like functionality.

### Label-free cell counting and fast viability analysis

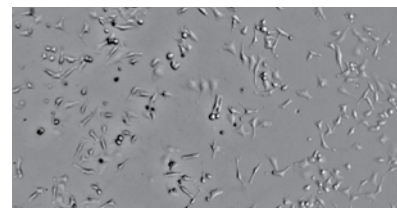
In combination with disposable Cell Chips, bright field imaging can be used for automated, label-free counting of a broad range of cell sizes and types. Trypan blue staining can be used to determine the number of live and dead cells.

- Accurate and reproducible cell counting, with flexible area selection for greater sensitivity
- Predefined, one-click applications for determination of cell number, size distribution or viability
- Easy export of cell images for visual confirmation
- Trypan blue-based staining for life/dead cell counts



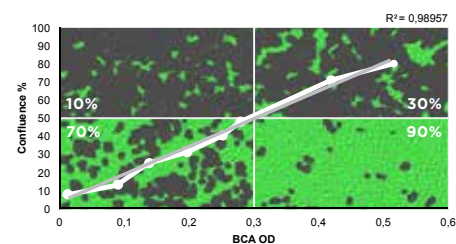
### Live viewer

Defined positions within a Cell Chip or microplate well can be viewed and saved with 4x magnification using Live Viewer. This allows microscope-like functionality, including user-definable focusing, for fast and easy quality control of your cells – take a last look before starting your assay.



### Automated cell imaging and confluence measurements

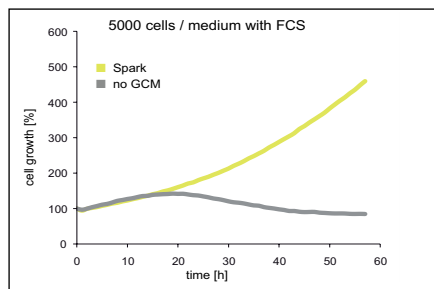
It is necessary to normalize results over several wells across a microplate to have confidence in your cell-based assay results. The problem is that you don't know how many cells have adhered to the bottom of the microplate, or even grown, over a certain period of time. With Spark, it is possible to automatically measure cell confluence and image cells in the well of a microplate. The system can then automatically inject a compound or measure a GFP signal once a user-defined cell confluence is reached, providing more reliable results.



The growth of HeLa cells at various confluence levels and the corresponding increase of protein content measured using a BCA assay protocol.







Comparison of cell proliferation in the Spark reader with integrated GCM and a standard microplate reader.



### Maintain stable culture conditions and improve cell growth

A patented integrated Gas Control Module (GCM™) features two independent gas inlets that allow automated control of CO<sub>2</sub> and O<sub>2</sub> partial pressures inside the reader chamber, offering:

- Stable long-term cell culture environments
- Improved cell viability and extended experimental times without adversely affecting results
- Optimization of gas levels and mixtures with independent regulation of CO<sub>2</sub> and O<sub>2</sub> partial pressures
- Software-controlled, automated adjustments for real-time modulation of gas partial pressures during a run

### Evaporation protection to enhance cell viability

Built-in evaporation protection enhances live cell kinetic assays for better reproducibility and more reliable data. A patented Humidity Cassette reduces evaporation in standard microplates, minimizing edge effects and enabling long-term live kinetic studies without the need to switch to dedicated and costly microplate types; simply use your current, validated plates.

### Minimize evaporation and contamination with automated microplate lid handling

The integrated patented microplate Lid Lifter™ allows automated lid handling within the reader, enabling incubation, measurements and injections without manual intervention. The Lid Lifter is compatible with the Humidity Cassette and any SBS-format plate lid which can be fitted with a magnetic pad. In combination with the GCM, advanced temperature control and the Humidity Cassette, Spark turns into a reader/incubator hybrid with flexible workflow automation capabilities, increasing reproducibility and reducing hands-on time. Additional benefits include:

- Reduced contamination risk for cell-based experiments
- Lower risk of user exposure when working with pathogens
- Luminescence measurements in lidded plates
- Reduced background noise for absorbance measurements in lidded plates

### Reagent dispenser with heating and stirring enhances application flexibility

Integrated injectors help to generate consistent data by eliminating precipitation and salt formation. Independent heating and stirring capabilities increase the reliability and reproducibility of compound handling, as well as enabling automated dispensing of viable cells.

### Connect microplate stacker for Spark

The Connect is a versatile microplate stacker designed to allow walkaway batch processing for up to 50 microplates per run. It interfaces directly with SparkControl Magellan software for easy operation, offering increased throughput for a wide range of applications in genomics, proteomics and drug discovery. Connect stacker highlights include:

- Dark cover to protect light-sensitive well contents
- Designed for reliable operation and ease of use
- Allows manual operation for single plates
- Barcode scanner option which can read either short side of the plate
- Choice of 30- or 50-plate cassettes to suit your throughput requirements



### Fluent® Laboratory Automation Solution for cell-based assays

Tecan's commitment to cell biology research goes beyond the capabilities of Spark. The revolutionary Fluent Laboratory Automation Solution for cell-based assays is designed to optimize cell-based workflows, automating everything from pipetting and reagent distribution to incubation and detection. For more information, please visit [www.tecan.com/fluent](http://www.tecan.com/fluent).



## SPARKCONTROL magellan

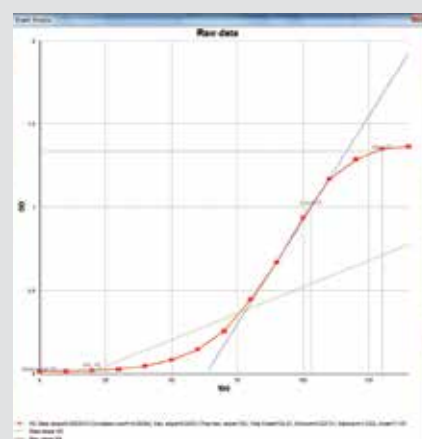
SparkControl Magellan is a data analysis package providing powerful data reduction tools for numerous detection modes.

**SparkControl Magellan offers users an array of tools designed to enhance functionality, simplicity and security.**

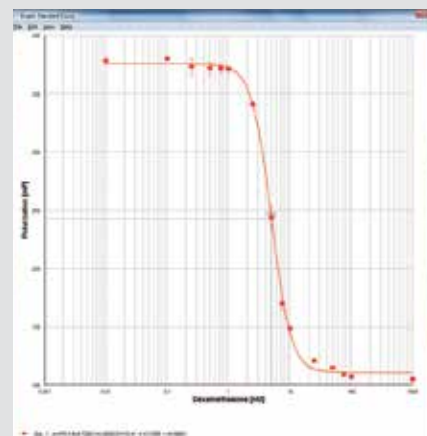
- Ideal for microplate-based applications such as ELISAs, end-point assays, kinetic assays, ratiometric measurements, multi-label measurements and 3D scanning
- Rapidly perform everything from data reduction and curve fitting to the calculation of kinetic parameters, such as Michaelis-Menten constants
- Video tutorials and example files simplify operation
- Plate definition editor allows creation of customized plate geometry files

**The software provides a suite of sophisticated functions including:**

- Full qualitative and quantitative EIA analysis
- All major curve fittings, including point-to-point, linear regression, non-linear regression, polynomial, cubic spline, Akima, logit-log, four- and five-parameter fits
- Convenient handling of dilution series and ICx calculations
- General data import and export options, as well as automated import of sample ID lists
- Kinetic data analysis with calculation of slopes, onsets and enzyme kinetics
- Spectral calculations to provide rapid background correction, curve smoothing, wavelength selection, peak identification and 3D scanning



SparkControl Magellan makes it easy to perform complete kinetic data analyses including the calculation of slopes, onsets and enzyme kinetics.



Designed to simplify the user experience, SparkControl Magellan conveniently handles all dilution series and ICx calculations.

**SparkControl Magellan Tracker** offers all the functionalities necessary for compliance with FDA regulation 21 CFR part 11 for electronic records and signatures, while still providing all the advantages of SparkControl Magellan Standard.



Fast, simple instrument operation is at your fingertips with SparkControl's touch-optimized, intuitive interface. Engineered to simplify your daily laboratory tasks, SparkControl offers:



**High definition well scans** provide a complete picture of the cell population in each well for more accurate signals, even with inhomogeneous cell layers. The software also provides a qualitative image of the cell distribution.



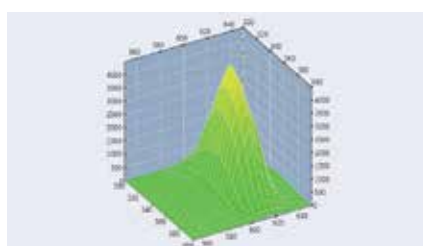
SparkControl makes it **easy to adjust** parameters during a run, including **environmental conditions** such as temperature (even below ambient) and the CO<sub>2</sub> and O<sub>2</sub> levels inside the reader.



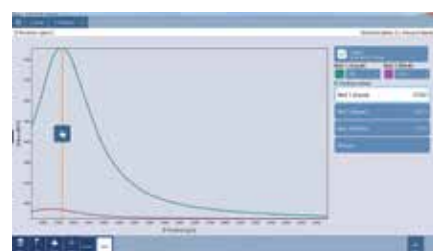
**Open kinetics** – Free-up your reader during long-term kinetic measurements. Pause/resume a kinetic run and allow your peers to access the reader in between. Increase productivity while running long-term cell-based assays.



**One-click applications** streamline your workflows, getting you from sample to results faster than ever before.

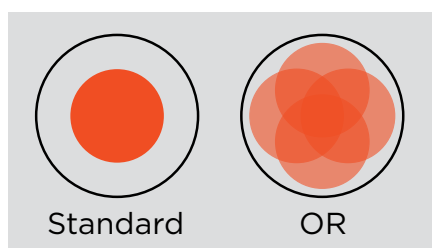


**3D scanning** accelerates assay development by providing simultaneous excitation and emission scans. This can help to identify changes in the spectral properties of fluorescent probes or characterize unknown fluorescent samples more quickly and easily.



**Automated z-focus** adjustment enhances the sensitivity of top-reading fluorescence intensity and fluorescence polarization modes, significantly improving the quality of results. No matter what your plate volume, sample volume or well shape, this unique feature makes it easy to set up your reader for optimum performance with varying assay parameters.





**Optimized fluorescence bottom-reading** with Tecan's unique Optimal Read (OR) function. OR ensures very low CVs by performing multiple measurements on spatially separated spots arrayed across each well.



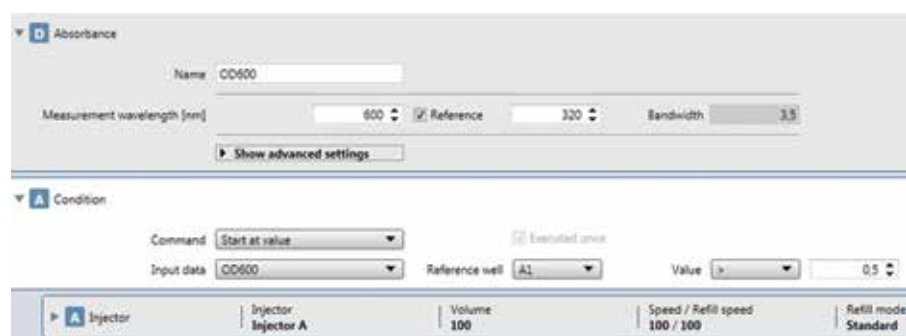
**Detect** even very low signals with the Spark **extended dynamic range**. This function automatically adjusts the gain settings during a measurement run, allowing the detection of very low signals without compromising on sensitivity. All results are automatically correlated and displayed within one single data set.



**Safeguard your kinetic assays** using automated gain regulation to avoid fluorescence measurements running into saturation. Measurements with different gain settings are then automatically correlated, allowing evaluation of the entire dataset.

## Smart automation

SparkControl excels in workflow automation. Combining an OD600 measurement with an injection at a specific absorbance value increases walkaway automation. Let SparkControl work for you, freeing up more time for important research.



## Typical performance values+

### Fluorescence – enhanced

Light source	High energy xenon flash lamp
Spectral range	Ex: 230–900 nm Em: 280–900 nm
Wavelength accuracy	Ex: < 0.5 nm; Em: < 0.5 nm
Wavelength reproducibility	< 0.5 nm
Bandwidth	Adjustable from 5–50 nm
Optical mirrors	50%, 510, 560, 625 nm built-in; 410, 430 nm user-selectable dichroics
Well scanning	Up to 100 x 100 data points

#### FI (Fluorescence intensity)

Limit of detection <sup>1</sup>	
Filter - top	≤ 8 amol/well (10 µl; 1,536 well) <sup>1</sup>
Fusion* - top	≤ 15 amol/well (10 µl; 1,536 well)
Mono - top	≤ 20 amol/well (10 µl; 1,536 well)
Filter - bottom	≤ 180 amol/well (10 µl; 1,536 well)
Fusion - bottom	≤ 200 amol/well (10 µl; 1,536 well)
Mono - bottom	≤ 220 amol/well (10 µl; 1,536 well)

#### FP (Fluorescence polarization)<sup>2</sup>

Spectral range	300 - 850 nm
Precision Filter	≤ 1.25 mP <sup>2</sup>
Precision Fusion	≤ 2.0 mP
Precision Mono	≤ 2.5 mP

#### TRF (time-resolved fluorescence)<sup>3</sup>

Limit of detection Filter	≤ 0.5 amol/well (20 µl; 384 sv well) <sup>3</sup>
Limit of detection Fusion	≤ 0.6 amol/well (20 µl; 384 sv well)
Limit of detection Mono	≤ 0.7 amol/well (20 µl; 384 sv well)

#### Fastest read time

384-well plate (FI)	≤ 22 sec
1,536-well plate (FI)	≤ 34 sec

### Fluorescence – standard

Light source	Dedicated xenon flash lamp
Spectral range	Ex: 230–900 nm Em: 280–900 nm
Wavelength accuracy	Ex: < 1 nm; Em: < 2 nm
Wavelength reproducibility	< 1 nm
Bandwidth	fix @ 20 nm
Optical mirrors	50%; 510 nm dichroic
Well scanning	Up to 100 x 100 data points

#### FI (Fluorescence intensity)

Limit of detection <sup>1</sup>	
Filter - top	≤ 25 amol/well (100 µl; 384 well) <sup>1</sup>
Fusion - top	≤ 35 amol/well (100 µl; 384 well)
Mono - top	≤ 50 amol/well (100 µl; 384 well)
Filter - bottom	≤ 500 amol/well (200 µl; 96 well)
Fusion - bottom	≤ 700 amol/well (200 µl; 96 well)
Mono - bottom	≤ 800 amol/well (200 µl; 96 well)

#### FP (Fluorescence polarization)<sup>2</sup>

Spectral range	300 - 850 nm
Precision Filter	≤ 1.5 mP <sup>2</sup>
Precision Fusion	≤ 2.5 mP
Precision Mono	≤ 3.0 mP

#### TRF (time-resolved fluorescence)

Limit of detection Filter	≤ 4.0 amol/well (100 µl; 384 well) <sup>3</sup>
Limit of detection Fusion	≤ 6.5 amol/well (100 µl; 384 well)
Limit of detection Mono	≤ 10 amol/well (100 µl; 384 well)

#### Fastest read time

96-well plate (FI)	≤ 13 sec
384-well plate (FI)	≤ 30 sec

### Absorbance (enhanced or standard)

Light source	Dedicated xenon flash lamp
Spectral range	200–1,000 nm
OD range	0–4 OD
Scan speed (200–1,000 nm)	≤ 5 sec
Wavelength accuracy	< 0.3 nm
Wavelength reproducibility	≤ 0.3 nm
Wavelength ratio accuracy (260/230)	< 0.08
Wavelength ratio accuracy (260/280)	< 0.07
Precision @ 260 nm	< 0.2%
Accuracy @ 260 nm	< 0.5%
Limit of detection (nucleic acids)	< 1 ng/µl

#### Plate formats for all read modes – enhanced

1 - 1,536 wells; NanoQuant Plate; Cell Chip;  
Cuvettes; Roboflask

#### Plate formats for all read modes – standard

1 - 384 wells; NanoQuant Plate; Cell Chip;  
Cuvettes; Roboflask

### Luminescence (enhanced or standard)

Spectral range	370–700 nm
Luminescence (glow) –	
Limit of detection <sup>4</sup>	≤ 225 amol/well (25 µl; 384 sv well) <sup>4</sup>
Luminescence (flash) –	
Limit of detection <sup>5</sup>	≤ 12 amol/well (55 µl; 384 well) <sup>5</sup>
Dynamic range	> 9 orders of magnitude
Multi-color luminescence	38 spectral filters; OD1, OD2, OD3 attenuation filters

#### AlphaScreen (enhanced or standard)

Limit of detection	< 100 amol/well bio-LCK-P <sup>6</sup> ; 20 µl < 2.5 ng/ml Omnibeads <sup>7</sup> ; 20 µl
Uniformity	≤ 3.0%
Z' value	> 0.9
Fastest read times <sup>8</sup>	≤ 2 min (384-well plate) ≤ 1 min (96-well plate)



**Cell counting**

Size range	4–90 $\mu\text{m}$
Counting accuracy	+/-10% (10–30 $\mu\text{m}$ )
Counting reproducibility	< 10% (10–30 $\mu\text{m}$ )
Cell concentration	$1 \times 10^4$ – $1 \times 10^7$ cells/ml
Imaging speed inc. data reduction	< 30 sec/sample
Number of samples/run	up to 8 samples

**Automated cell imaging**

Illumination	High power LED
Image	Bright-field
Objective	4x
Autofocus	Laser based
Optical resolution	1.3 $\mu\text{m}/\text{pixel}$
Read Speed	1 image/well (96-well plate); < 5min

**Gas Control Module**

Adjustable concentration range – $\text{CO}_2$	0.04–10% (vol.)
Adjustable concentration range – $\text{O}_2$	0.1–21% (vol.)
Concentration accuracy – $\text{CO}_2$	< 1% (vol.)
Concentration accuracy – $\text{O}_2$	< 0.5% (vol.)

**Reagent injectors**

Syringe sizes	0.5 ml; 1 ml; 2.5 ml
Pump speed	100–300 $\mu\text{l}/\text{sec}$
Injection volume	5–2,500 $\mu\text{l}$ ; step size: 1 $\mu\text{l}$
Dead volume	$\leq 100 \mu\text{l}$
Injection accuracy and precision	$\leq 0.5\%$ at 450 $\mu\text{l}$

**Temperature control**

	ambient +3 °C up to 42 °C
Uniformity	< 0.5 °C

**Te-Cool cooling module**

Temperature range	+18–42 °C
Cooling power	max 12 °C below ambient

**Shaking**

Linear, orbital, double-orbital;  
variable amplitudes and frequencies

\*Fusion Optics: a combination of filter and monochromator on the excitation and/or emission side

- 1) Detection limit for Fluorescein
- 2) FP detection limit @ 1 nM Fluorescein
- 3) Detection limit for Europium
- 4) Detection limit for ATP (144-041 ATP detection kit SL (BioThema))
- 5) Detection limit for ATP (ENLITEN® Kit)
- 6) (PE# 6760620; P-Tyr-100 assay kit)
- 7) (PE# 6760626D; Omnibeads)
- 8) Including temp. correction

**Spark multimode reader is for research use only.**

+ Specifications are subject to change. Performance values represent the average observed factory tested values.  
For product specifications refer to operators manual.



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