

Mithras² LB 943

Monochromator & Filter
Multimode Microplate Reader*

detect and identify



Mithras² LB 943

Monochromator & Filter

Multimode Microplate Reader*

Berthold Technologies Mithras² multimode reader for microplates is a unique system offering the benefits of both, monochromator and filter technologies for wavelength selection. The instrument is a powerful unit with ergonomic design for all non-isotopic labelled assays.

- UV/VIS Absorbance
- Fluorescence
- FRET
- Fluorescence Polarisation
- Time-Resolved Fluorescence
- Time-Resolved FRET
- Luminescence
- BRET
- BRET²
- AlphaScreen[®]
- AlphaLISA[®]

Monochromator Technology

Flexibility in wavelength selection for any current and future assay requirements is best met by using monochromators for absorbance/excitation and emission. The ability to scan complete absorbance and emission spectra has many benefits when measuring wavelength shifts due to, for example, changes in pH or polarity on the fluorophore's properties.

Berthold Technologies employs two double monochromators providing blocking properties needed in sensitive fluorescence assays. Both monochromators are equipped with software driven continuous bandwidth variation to optimise the instrument for the specific demands of different assay conditions.

Filter Technology

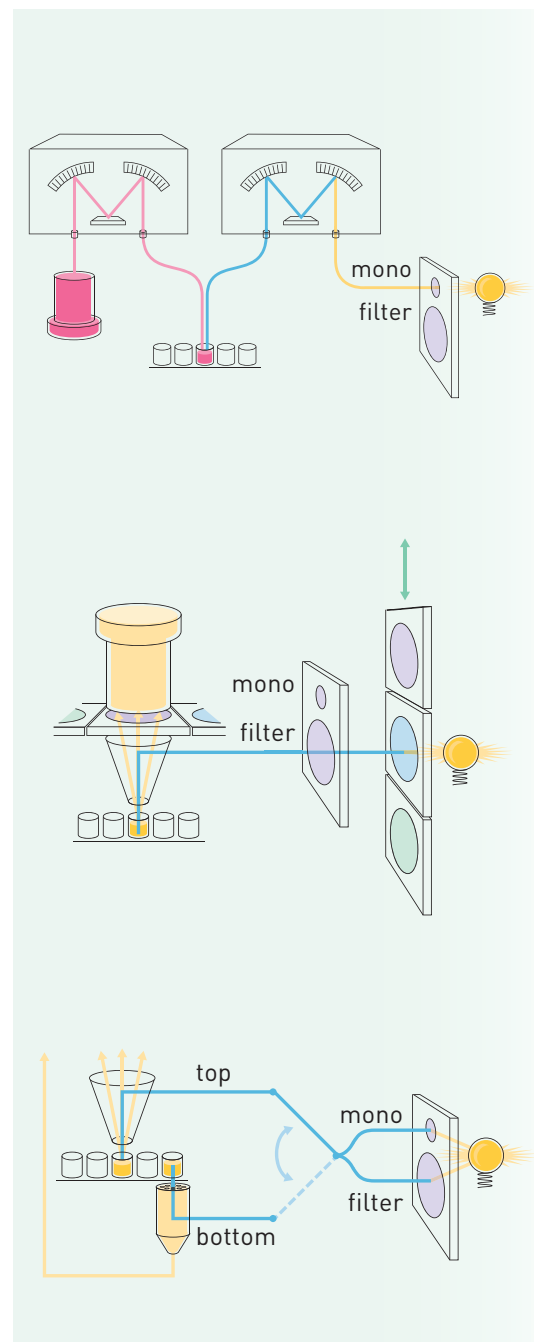
Due to their high transmission characteristics – which can be up to 25-fold that of monochromators – technologies like Fluorescence Polarisation and TR-FRET, e.g. HTRF®, can be measured more efficiently with filters. In addition filters are available with wide bandwidths making them the ideal choice for fluorophores with wide spectra and for all luminescence based assays requiring the use of filters, e.g. ChromaGlo™, BRET and BRET².

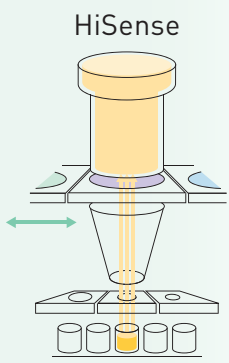
Another big advantage of filter based systems is the quick change between filters, independent of the wavelength difference, which is important for fast ratiometric measurements common in Fura 2 Calcium monitoring.

Top and Bottom Reading

Cell-based assays very often require bottom reading capability, especially when a combination of adherent cells, non-secreted label and coloured media are being used. In the Mithras² all measurements of both fluorescence and luminescence can be read from below as well as from top with high performance.

Switching from top to bottom is, of course, software driven and can be set multiple times within one measurement. Especially in migration and chemotaxis experiments this feature becomes a valuable tool.





Detectors

The Mithras² is equipped with 3 photomultipliers (PMTs) and a photodiode for absorbance measurements.

Two low noise PMTs are used with the filter and monochromator optics respectively. Both detectors have an extended wavelength range up to 850 nm covering the demands of near-infrared dyes. A third ultra-low noise PMT is employed in the HiSense position for sensitive detection of luminescence, BRET and Alpha[®] technologies.

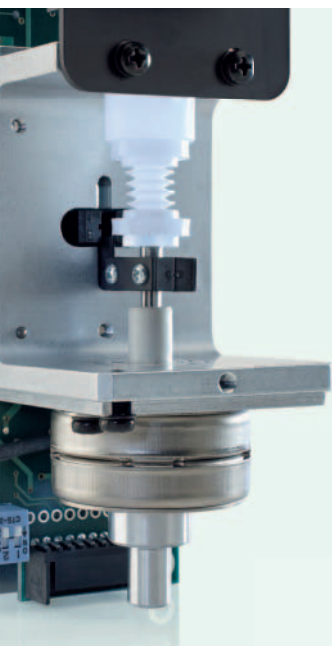
All PMTs are subject to a stringent pre-selection process to guarantee low noise, high efficiencies and outstanding stabilities throughout the temperature range of operation.

Sensitivity

Low level detection. True photon counting technology coupled with selected low noise photomultipliers is the only accepted way to achieve a low and stable background. Together with an optimised optical design this is the major parameter for high sensitivity in a measurement device enabling detection of extremely low amounts of analyte.

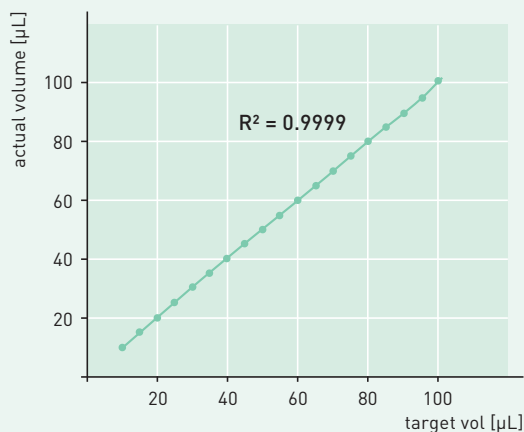
- Less than 2 amol Europium (TRF)
- Less than 0.2 fmol Fluorescein (Fluorescence)
- Less than 2 amol ATP (Luminescence)

Save money and time. The high sensitivity provides additional benefits when detecting the lowest signal levels is not the key to an assay. In these cases the consumption of expensive reagents or valuable cells can be greatly reduced. Similarly, you can significantly reduce the reading time per sample and save valuable total operation time.



JET injectors

Berthold JET injectors are based on a proprietary technology using Teflon bellows for accurate and fast injections guaranteeing most efficient mixing and ensure extreme longevity.



- **Accuracy and precision** of better than 98 % over the entire volume range
- **Frictionless** operation for extended lifetime
- **Cell-friendly materials** and negligible shear forces enable injection of cell suspensions, e.g. in Aequorin-based Calcium assays
- **Sophisticated Prime mode** reduces reagent consumption while ensuring homogeneous filling

Accuracy

The real value counts. Like all Berthold Technologies instruments the Mithras² comes to your laboratory checked against a certified light source. With this reassurance you are able to work with accurate, traceable and comparable results.

Reliability & Precision

Get the same from the same. Precise mechanics and the intrinsic stability of photon counting technology guarantee unrivalled repeatability of measurements. The instrument's performance is stable over years. Time-consuming daily calibration is thus unnecessary and you can concentrate on other more important duties.

Robustness

A companion for years. Berthold Technologies instruments are known for their quality and longevity providing a working life of decades. This is a feature gaining in importance with today's limited budgets and environmental awareness.

RFID coding of filters

Radio-Frequency Identification is the most modern and secure way for filter coding. Each filter, contained in its holder, includes a programmed tag containing filter data such as, centre wavelength, bandwidth and transmission.

Whenever the unit is switched on or a filter carrier is moved the Mithras records the filters currently inserted and compares the inventory to the requirements of the selected measurement. It is therefore no longer possible to get erroneous readings due to a wrong set of filters.



Light Protection

No influence of ambient illumination. All Berthold Technologies instruments ensure light-tightness of the housing and the doors, checked against a light source of 30,000 Lux without any influence on the measured values.

The reagents are protected from strong ambient illumination by a special attenuating reagent compartment cover.

Ergonomic design & front access

Face-to-face. All operations like plate loading, filter change and reagent connection can be performed at the front of the instrument and at an accessible level. Push buttons – also located at the front – can be engaged to load and unload microplates and start a measurement.



Reagent trough and holder. Space for reagent vials is available at the front of the instrument providing easy access and visibility. A removable trough can be filled with ice to keep the reagents at lower temperatures. A special reagent holder keeps small tubes, filled with valuable reagents, safely in place and ensures tubings recover the last droplet.



Built-in waste pump. For washing and filling the reagent lines you don't need to worry about loading microplates, waste containers or removal of injector tips and the built-in waste pump takes care of the safe removal of excess liquids.

Versatility and User-Friendliness

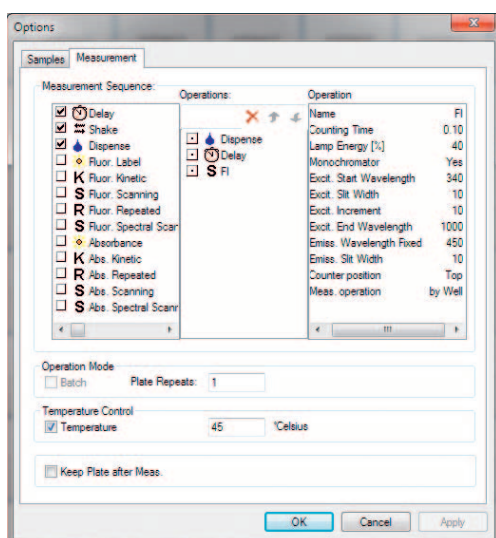
Choice of reagent injectors. The Mithras² can be equipped according to the requirements with up to 4 variable volume reagent injectors. All injectors can be used to inject in measurement position – even with bottom reading operation – for both 96 and 384 well plates.



Temperature Control & Gas Connection. For cell based assays everything has been taken care of in order to control the internal environment. A specific incubation area ensures an extremely quick attainment and homogeneous distribution of the target temperature in the samples.

Automatic Plate Height Adjustment. This feature together with z-independent optics makes any time-consuming z-scans obsolete and ensures crosstalk is minimised between adjacent wells.

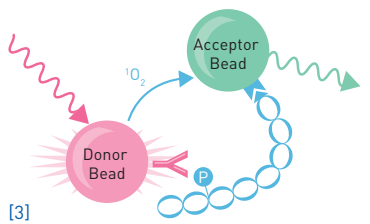
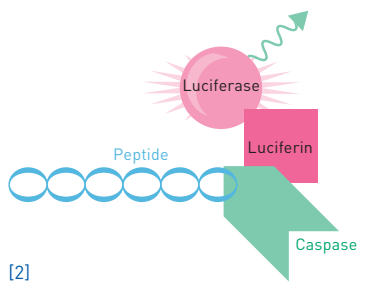
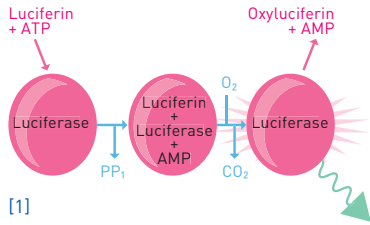
Software. The user can choose between the wizard-guided ICE software and the powerful Mikrowin software. Both support the instrument's extensive operation capabilities.



- multiple endpoint
- kinetics
- spectral scanning
- reagent injection
- shaking / delay

For further data analysis a variety of calculations may be selected, including standard curve calculation, blank subtraction, replicate statistics and kinetic parameters.

Data can be exported to multiple locations and with user-specific settings.



Applications

Reporter Gene Assay [1]

In basic research of gene regulation as well as in drug discovery the use of luciferases, β -glucuronidases, β -galactosidases and secreted alkaline phosphatases as well as GFP has become a standard tool.

Due to the high sensitivity and the possibility to use filters in the HiSens luminescence optics even colour luciferase reporter genes assay can be measured sensitively.

Caspase Assays [2]

Monitoring the activity of caspases – a group of cysteine-aspartic acid peptidases – is a key method in apoptosis research. The assays are designed around specific peptide substrates for Caspase 3, 7, 8, and 9 respectively which will be cleaved when caspases are present.

Kinase Assays [3]

The luciferase reaction can be used as well for monitoring kinases through a coupled reaction correlated by the amount of ATP. Other alternatives are being offered for HTRF[®], Fluorescence Polarisation and Alpha[®] technologies.

Detection Mode	Assay	ATP Measurement	Binding Assays	Ca ⁺⁺ Monitoring	Caspase	Cell Proliferation	Cell Viability	Cyclic AMP	Cytokine Quantification	Cytotoxicity	DNA/RNA Quantification	Dual Reporter Gene	Enzyme Activities	GPCRs with β -arrestin	Immunoassay / ELISA	IP1	Kinase	Protease	Protein – Protein Interactions	Protein Quantification	Reactive Oxygen Species (ROS)	Receptor – Ligand Binding	Receptor Dimerisation	Reporter Gene	SNPs	Spectral Scanning
Absorbance / Colorimetric					•	•	•	•	•	•	•		•		•		•	•		•	•		•		•	
BRET / BRET ²					•									•			•	•				•	•			
Luminescence		•			•	•	•	•	•	•	•	•	•		•		•	•		•	•		•		•	
Luminescence Flash with Injection		•		•		•	•			•		•	•		•						•			•		
Fluorescence				•	•		•	•	•		•		•		•		•	•		•	•		•	•	•	
Fluorescence Flash with Injection				•									•								•					
FRET			•		•			•						•				•	•			•	•			
TRF			•					•	•	•					•		•					•				
HTRF [®]			•					•							•	•	•	•	•			•	•			
Fluorescence Polarisation			•					•							•		•					•			•	
Fluorescence Polarisation Flash with Injection			•										•									•				
AlphaScreen [®] / AlphaLISA [®]			•					•							•		•		•							

GPCR Monitoring [4]

Especially in the field of G-protein coupled receptor research the BRET technology offers the opportunity to establish a homogeneous and universal functional assay.

Calcium Assays

Intracellular Ca^{++} levels are important indicators for the functioning of ion channels and G-protein coupled receptors as well as for the phases of apoptosis and cell injury. Aequorin and Fura 2 have become established detection agents.

DNA Quantification

The use of specific fluorescent labels provides lowest detection limits and widest dynamic range whereas the UV method offers label-free detection.

Protein Quantification

Traditional absorbance-based Lowry and Bradford methods are as well suited as fluorescent labels and the label-free UV measurement.

Second Messengers: IP1 and cAMP [5]

Typically, after agonist binding GPCRs trigger downstream responses via the cAMP or the Phosphoinositol pathways. Both can be monitored with HTRF® or Alpha® technologies or, using Epac, a FRET-based approach may be taken.

Cytotoxicity and Cell Viability

Resazurin-based assays can be read in absorbance or in fluorescence mode whereas assays can also be established for the sensitive luminescence read-out using Firefly reaction to determine cell viability via the ATP content.

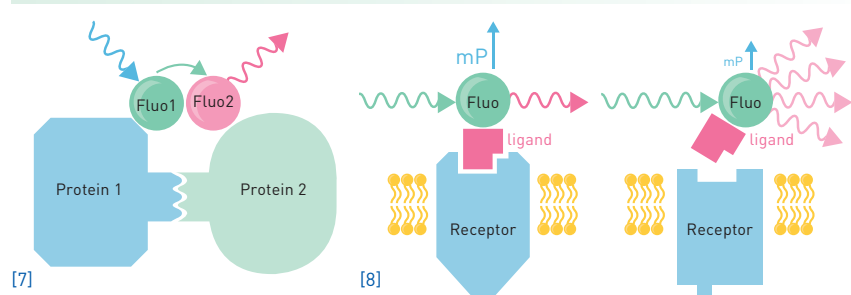
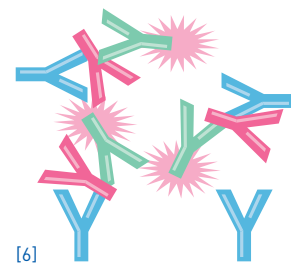
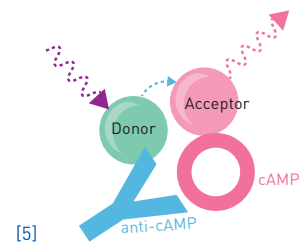
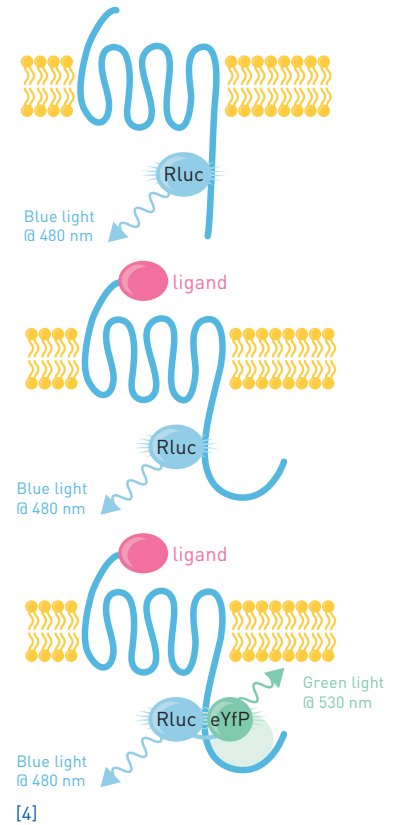
ELISAs and Immunoassays [6]

Horse radish peroxidases and phosphatases can be used with colorigenic, luminogenic or fluorogenic substrates. Using a luminescent substrate the sensitivity can be increased up to a 100-fold.

Binding assays [7, 8]

Whether protein-protein binding detected via FRET between fluorescent protein labels or small molecule binding with a fluorescent polarisation assay Mithras² offers any of these possibilities.

... and many other biochemical and cellular applications.



Quality control

The proof of the functionality and the performance of the Mithras² can be monitored by easy-to-use test devices and standardised operational procedures.

Luminescence Test Plate

The solid state test plate for luminescence is the easiest, quickest and most reliable way to periodically check the performance of an instrument. The test plate can be applied to monitor the instrument's

- efficiency
- accuracy
- reproducibility
- mechanical positioning
- crosstalk

An annual check-up and calibration of the test plate ensures its consistency.

Absorbance Test Plate

The absorbance test plate can be used over a wide range of wavelengths. It can be applied to check the instrument's

- accuracy
- reproducibility
- mechanical positioning

The test plate can be submitted to periodical checks.

Luminescence Performance Kit

With the QC luminescence performance kit (consisting of two controls and two different detection reagents) the performance of your luminometer can be checked.

It is an alternative to the test plate and an ideal quality control method as both injection and detection system can be monitored.

The reaction is a luminescence flash type one providing results within seconds.

IQ and PQ Services

The test plates as well as the kit are part of the IQ and PQ services available through Berthold Technologies engineers. All reading technologies will be checked during these services with the reagents or means appropriate.



Order Information

	Order Number
Mithras ² M2F Filter only, Luminescence, Fluorescence, Absorbance, Fluorescence Polarisation, Time-Resolved Fluorescence, top & bottom reading, temperature control	56600-001
Mithras ² M2MF Filter & Mono, Luminescence, Fluorescence, Absorbance, Fluorescence Polarisation, Time-Resolved Fluorescence, top & bottom reading, temperature control	56600-002
Mithras ² M2M Mono only, Luminescence, Fluorescence, Absorbance, Time-Resolved Fluorescence, top & bottom reading, temperature control	56600-003
Mithras ² M2Ffv Filter only, Luminescence, Fluorescence, Absorbance (vis), Fluorescence Polarisation, top & bottom reading, temperature control	56600-004
Mithras ² M2Ftu Filter only, Luminescence, Absorbance, Time-Resolved Fluorescence, top & bottom reading, temperature control	56600-005
Mithras ² M2Mfv Mono only, Luminescence, Fluorescence, Absorbance (vis), top & bottom reading, temperature control	56600-006
Mithras ² M2Mtu Mono only, Luminescence, Absorbance, Time-Resolved Fluorescence, top & bottom reading, temperature control	56600-007
Monochromator Double monochromators for excitation and emission	60532
HiSens Luminescence Additional special ultra-low-noise PMT	58903
HiSens Luminescence & Alpha technology Additional special ultra-low-noise PMT and Direct Access laser diode	60654
Injector #1, bottom reading-position	54116-21
Injector #2, monochromator position	54116-22
Injector #3, filter position	54116-23
Injector #4, HiSense position	54116-24
BRET/BRET ² package	60384
BRET "High Efficiency" package	60383
BRET ² "High Efficiency" package	60385
Chroma-Glo package	60535
HTRF [®] filter set	59542
FP Fluorescein filter set	59543
FP TAMRA & Cy3 filter set	60394
FP Cy5 filter set	60398
FP Alexa633 filter set	60403
Luminescence QC test plate	40105-10
Luminescence QC performance kit	55101
Absorbance QC test plate	50895-10
Cleanit Daily injector cleaning solution	45218
Microplates 96 well, white	23300
Microplates 96 well, black	23302
Microplates 96 well, black with white wells	55008
Microplates 96 well, white, cell culture	51838
Microplates 96 well, black, cell culture	51839
Microplates 96 well, white, clear bottom, cell culture	24910
Microplates 96 well, black, clear bottom, cell culture	38840
Microplates 96 well, white, clear bottom	60705
Microplates 96 well, black, clear bottom	60706
Microplates 384 well, white	32505

Mithras² LB 943

Technical Specification

Detection Devices	2 Low-noise photomultiplier tubes in single photon counting mode, spectral range up to 850 nm 1 Ultra-low-noise photomultiplier tube in single photon counting mode, spectral range up to 650 nm Photo diode, spectral range 200 – 1000 nm	Performance	
Excitation Sources	Halogen lamp, spectral range 340 – 1000 nm Xenon flash lamp, spectral range 230 – 1000 nm Laser diode, 680 nm	Luminescence	< 2 amol ATP
Measurement Technologies	Luminescence BRET, BRET ² Fluorescence FRET UV/VIS Absorbance Fluorescence Polarisation Time-Resolved Fluorescence TR-FRET (HTRF [®]) AlphaScreen [®] AlphaLISA [®]	Fluorescence	< 0.2 fmol Fluorescein
Measurement Optics	Top and bottom for all modes Switching by software	Absorbance	Acc. better 2 %, prec. better 0.6 %
Filters	Deep blocking RFID coding	TRF	< 2 amol Europium
Monochromators	Double monochromators for ex and em F number 2.7 (high transmission) Variable bandwidth 6 – 22 nm Increment 1 nm Blocking 10 ⁻⁶ Stray light 10 ⁻⁶	HTRF [®]	> 600 %dF
Operations	Single and multiple readings Endpoint Kinetics Repeated (long term kinetics) Scanning Spectral scanning Top and bottom reading Delay Shaking Injection Unload	Fluor. Polarisation	< 4 mP
		AlphaScreen [®]	< 12 ng
		Dynamic Range	> 6 orders of magnitude 0 – 3.5 OD
		Crosstalk	Low crosstalk due to crosstalk reduction design: 5 x 10 ⁻⁶
		Light-Tightness	30,000 Lux
		Injection Unit	Up to 4 Volume: 10 – 100 µL JET injection technology Accuracy better 2 % Precision better 2 %
		Temperature Control	+ 5 °C above RT to 45 °C Plate incubation area
		Microplate Formats	6 to 1536 well Petri dishes, Terasaki plates
		Interface	USB
		PC Operating System	Win 7
		PC Requirements	Pentium Processor, 500 MHz (or better), CD ROM drive, display 1024 x 768 (or better), USB
		Regulations	CE, UL, CSA
		Power Supply	110 – 240 V, 50/60 Hz, 650 VA
		Temperature Range	Storage: 0 – 40 °C Operation: 15 – 35 °C
		Humidity	10 – 85 % non-condensing
		Dimensions	480 x 500 x 550 mm (W x D x H)
		Weight	48 kg

Berthold Technologies reserves the right to implement technical improvements and/or design changes without prior notice.
Patents: EP 8021417.4 (pending), DE 20 2008 009 859.9 (pending)
Registered Design 20 2008 009 859.9
Certain Configurations of this product are not available for sale in the U.S.A.

BERTHOLD TECHNOLOGIES GmbH & Co. KG

P.O. Box 100 163 · 75312 Bad Wildbad · Germany
Phone: +49 7081 177-0 · Fax: +49 7081 177-100
E-mail: bio@berthold.com · www.berthold.com/bio