







# Let your application define your system solution.

phase and separation is based on differences in only the method but also the HPLC system.

In HPLC, components of a mixture are carried migration rates among the sample components. through the stationary phase by the flow of a mobile Therefore, the nature of your analytes defines not

Main characteristics of the analytes:

**MOLECULAR WEIGHT** defines the pore size of the stationary phase.

**SOLUBILITY** defines the HPLC mode, chemistry of stationary phase and eluent.

**CONCENTRATION AND MATRIX** defines the detection parameters and column dimensions.



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The solubility of your analytes defines the HPLC mode. The elutropic series defines the solvent strength for the most often used chromatography modes normal phase and reversed phase.



### **Most common HPLC modes**

HPLC mode	Mobile phase	Stationary phase	Analytes
Separation of small molec	<b>cules</b> (up to 2000 [	Da)	
Reversed Phase RP (<200 Å)	Polar (e.g. mixtures from water and acetonitrile)	Nonpolar (e.g. C18)	Mid-polar - mid-nonpolar soluble in polar and aqueous solvents
Normal Phase NP	Nonpolar (e.g. heptan)	Polar (e.g. SiOH)	Nonpolar soluble in nonpolar solvents, insoluble in water
Hydrophilic Interaction Liquid Chromatography HILIC	Polar organic sol- vents + water	Polar (e.g. SiOH)	hydrophilic and highly polar not retained by RP
	Water layer betwee	,	
Separation of biomolecul	<b>es</b> (larger than 200	00 Da)	
Wide Pore Reversed phase RP (>200 Å)	Polar	Nonpolar	Mid-polar - mid-nonpolar soluble in polar and aqueous solvents
Polymer gel based station	nary phases		
Ion Exclusion & Ligand exchange	Water, acidic water	Organic resin with charged groups	Sugars, organic acids, alcohols

# **AZURA®** Analytical HPLC/UHPLC

### Efficient and adaptable to your needs

The analytical HPLC and UHPLC systems of the tool to overcome your analytical challenges. KNAUER AZURA liquid chromatography instruments are designed to support and facilitate your work. Whether doing routine analysis or demanding separation tasks, AZURA systems are the right

Choose between different gradient forming technologies and maximum flow rates to find the best configuration for your task.

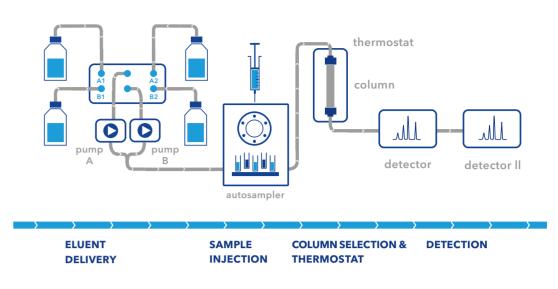


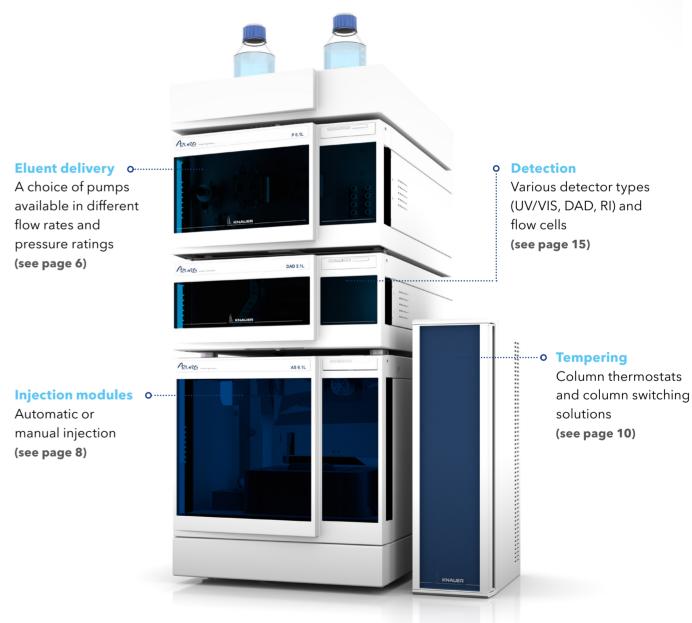




### **Features**

- Isocratic binary high pressure gradient (HPG) or quaternary low pressure gradient (LPG) pump
- Pump heads allowing flow rates up to 10 ml/min at 700 bar or 1000 bar up to 2 ml/min
- Autosampler with 0.1 µl sample aspiration at max. 1240 bar with zero sample loss
- Choice of highly sensitive UVD, MWD, DAD, or RID detectors with intelligent temperature control
- Wide range of flow cells available, including remote cells
- Finger-tight high pressure stainless steel capillary connections
- Extensive safety features such as leak management and sensors
- Frontal access of detector lamp and pump head for easy maintenance

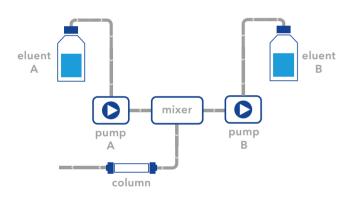


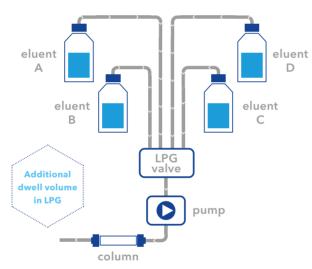


# **Gradient HPLC**

### Advantages and disadvantages of HPG and LPG

HPG (Binary HPG pump)	<b>LPG</b> (Quaternary LPG pump)
Small gradient delay volume	Higher gradient delay volume
Fast methods	For classical HPLC methods: no difference For UHPLC: slower gradients
Only binary gradients possible	Ternary and quaternary gradients possible
2 pumps that have to be maintained	Only 1 pump, but shorter maintainance intervalls because of higher usage





#### The dwell volume

The dwell volume is responsible for the time delay for a gradient. By definition it is the volume of a gradient HPLC system between the mixing chamber and the column inlet. It is typically higher in LPG systems caused by the setup of the pump. Therewith, gradients are faster with HPG systems.



Whether an LPG or an HPG pump is recommended highly depends on the user's preferences.

Only if very fast UHPLC gradients are needed, an HPG will be recommended.

If a quaternary gradient is inevitable, an LPG pump has to be applied.

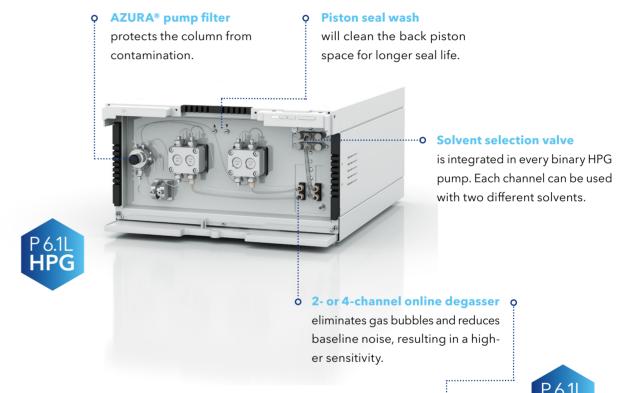
In any other case: The user decides!

# **AZURA®** Pump P 6.1L

Choose your HPLC pump according to your application's needs. Gradient formation, mixer size and pulsation compensation will have an extensive influence.

Choose between pump heads with a maximum flow rate of 10 ml/min and 700 bar back pressure,

or pump heads with a flow rate of 5 ml/min and 1000 bar backpressure. Special pump heads for normal phase applications will help to deliver robustly even demanding eluents like heptane or hexane. Pumps without a degasser offer a cost effective alternative.



#### AZURA® mixer

Highly efficient microfluidic solvent mixer combines high mixing performance with a low dead volume. The user-changeable mixer is available in different volumes (50/100/200  $\mu$ l) allowing best possible adaption to any application.

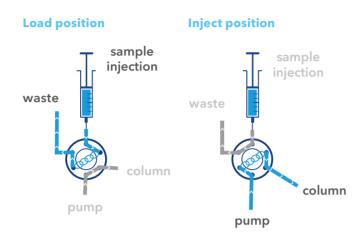


# Sample injection

### Principle of manual injection

Manual injection valves are the most cost-effective option to introduce samples. Normally, valves with 6 ports and 2 positions - for loading and injection - are used. In the load position a sample loop is filled with sample while the system is equilibrating.

When turning to the inject position, the sample loop is switched to the high pressure part of the HPLC system. The flow delivered by the pump flows through the loop and feeds the sample onto the column.





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KNAUER valves feature a wide back pressure range of up to 1200 bar with a 0.3 mm bore size. All valves can be equipped with an automatic valve drive. In addition, an integration into the assistant module housing AZURA ASM 2.1L is possible. For automated injection of numerous samples, we recommend to use the autosampler AS 6.1L. The injection principle - based on a valve and a sample loop - is similar, but the AS 6.1L additionally features several injection modes as described below.

### Injection modes of the autosampler AS 6.1L

Device	Key features
Full loop	<ul> <li>Sample loop is completely filled with the sample.</li> <li>Maximum possible reproducibility but not the maximum accuracy is achieved as the specified sample loop volume may deviate by 10 %.</li> <li>Maximum injection volume equals the loop volume.</li> <li>Sample loss caused by the need of overfilling of the loop.</li> </ul>
Partial loop	<ul> <li>Sample loop is filled with both sample and mobile solvent.</li> <li>High precision of the sample volume with minimal loss of sample</li> <li>Maximum injection volume equals 50 % of the loop volume</li> <li>Sample loss per injection equals the adjusted flush volume of the needle tubing.</li> </ul>
Microliter pick-up	<ul> <li>Sample loop is filled with a very small amount of sample and transport liquid or wash solution (mobile phase).</li> <li>Very high precision</li> <li>No loss of sample</li> </ul>

# **Autosampler AS 6.1L** for automated injection

The AZURA Autosampler AS 6.1L is a high precision device available for a maximum back pressure of 1 240 bar. This autosampler can inject from up to 768 positions when equipped with microtiter plates or from up to 108 standard 2 ml sample vials. The AS 6.1L is equipped with an ILD™ valve, consisting of a rotor-stator combination with a central port for depressurizing the sample loop before receiving the sample. This way, the sample is not diluted with solvent. The AS 6.1L is available in two analytical version for 700 and 1240 bar, respectively. In addition, also a biocompatible version and one for preparative purposes is available.

- Up to 1240 bar (700 bar)
- Optional cool/heat function (4 40 °C)
- 0.1 μl 10 000 μl (depending on configuration) sample injection volume
- Up to 768 samples (microtiter plates) or 108 standard vials
- Intermediate Loop Decompression ILD™

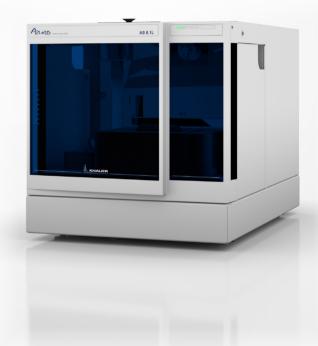


Rack for 108 standard 2 ml vials



the user.

Most important factors in injection are precision, accuracy and carryover. They are effected by the injection technique and equipment and in case of manual injection also by





The Integrated Intermediate Loop decompression valve reduces sample dilution and increases measurement reproducibility.

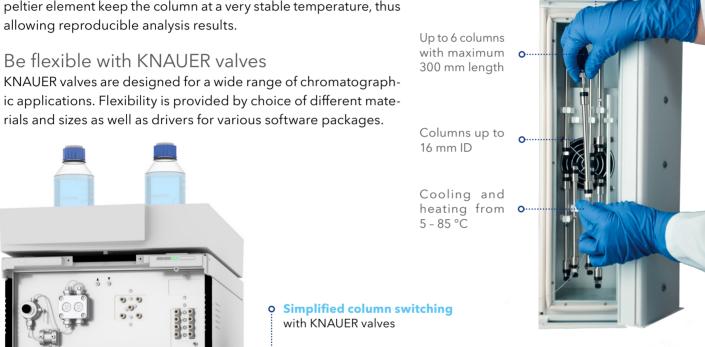
# **Column tempering** and switching

### Column Thermostat CT 2.1

The AZURA CT 2.1 is a forced air column thermostat capable of heating or cooling from 5 to 85 °C. The powerful fan and robust peltier element keep the column at a very stable temperature, thus allowing reproducible analysis results.

#### Be flexible with KNAUER valves

ic applications. Flexibility is provided by choice of different materials and sizes as well as drivers for various software packages.



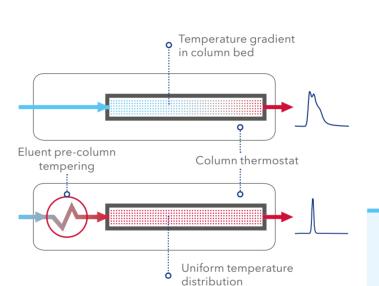
 Valves are driven either manually or automatically through a valve drive.

> Column thermostats are an essential part of each HPLC system since most HPLC separation parameters are temperature-sensitive. Hence, the control of the column temperature is one of the most important prerequisites for reproducible results in HPLC.

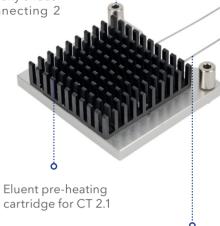
## **Eluent pre-column tempering**

When running HPLC at temperatures above 40 °C, a significant temperature gradient within the column is always present when mobile phase preheating is not applied in air-conditioned labora-

This device will converge solvent temperature with column temperature before entering the column and will therefore reduce temperature gradients within the column.



The Eluent pre-heating cartridge can easily be retrofitted to the oven chamber o. of the CT 2.1 by any enduser by just connecting 2 screws.



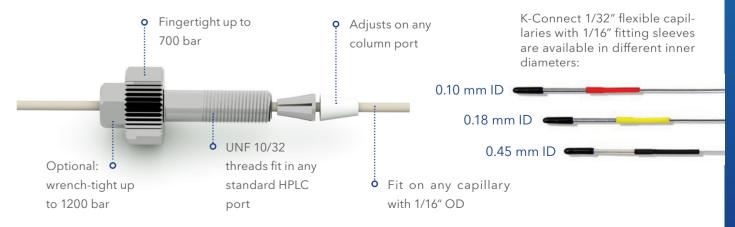
Available with 0.10 or 0.18 mm inner diameter (5 µl/18 µl inner volume)

#### **Rule of thumb**

For flow rates higher than 500 µl/min or temperatures above 50°C, the use of a pre-heating cartridge is recommended.

# **K-Connect fittings and capillaries**

Flexible capillaries and finger-tight connections provide easy handling while ensuring high pressure stability. Precision manufactured surfaces and edges significantly enhance fluid transfer.



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• Wide space.

easy handling

# **Columns** for HPLC and UHPLC



#### STAY FLEXIBLE

Any commercially available standard HPLC column can be used in an AZURA analytical system. KNAUER HPLC columns are compatible with any LC system.



#### BE SAFE

Find the matching guard column solution for your analytical column and extend its lifetime even if more agressive eluents or samples are used.

#### FIND YOUR PERFECT MATCH

We offer a wide range of stationary phases for nearly every application field.



#### MAKE IT SIMPLE

Easiest up- and downscaling with the same selectivity in many different particle- and column dimensions as well as pressure ranges.



#### Which column dimensions are recommend?

While the stationary phase always depends on the application, the recommended column dimensions are mainly dependent on the pump integrated in your AZURA analytical system:

Analysis	AZURA pump	Typical column dimensions
Classical HPLC	P 6.1L 700 bar	250 x 4 mm ID / 250 x 4.6 mm ID, 5 μm
HPLC Plus	P 6.1L 700 bar	150 x 3 mm ID, 3 μm
UHPLC	P 6.1L 1000 bar	100 x 2 mm ID, 2 μm



Smaller column dimensions

Shorter analysis time

Higher back pressure and performance

# Safety solutions for every column

Inner diameter of the analytical column	Recommended safety solution	Advantages
2 mm	UHPLC precolumn filter	Minimized dead volume via sintermetal filter instead of a packed precolumn cartridge, easy exchange.
3 - 4.6 mm, KNAUER Vertex III column hardware	Integrated precol- umn	Low dead volume because no capillary is used between analytical column and precol- umn. Can easily be installed later on any KNAUER Vertex III column without a precolumn.
3 - 4.6 mm, any column hardware	External precol- umn cartridge and holder	Precolumn can easily be flushed in reversed flow for cleaning.
8 mm	External precol- umn	High capacity for analytical and semi- pre- parative purposes, easy cleaning by flushing in reversed flow.

A guard column system is always recommended to protect the valuable analytical column, but especially when:

- Agressive eluents are used.
- Injected samples are dirty.
- Not only standards but different samples are analyzed.





An integrated precolumn can be exchanged very easily by the user.

### HPLC/UHPLC of small molecules:

## Silica based phases: Eurospher II

Modification	USP code
C18 P	L1
C18 H	L1
C18	L1
C18 A	L1
Phenyl	L11
C8	L7
C8 A	L7
C4	L26
NH₂	L8
CN	L10
HILIC	-
Diol	L20
Si	L3
	•

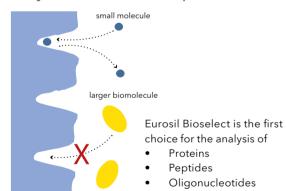
### Highest flexibility

13 different modifications are available for a wide range of small molecule applications (< 2000 Da)

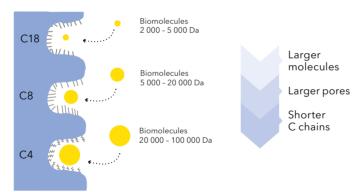
- Reversed phase mode for samples, soluble in water and water/organic solvent mixes
- Normal phase mode for water insoluble samples
- HILIC mode for highly polar and water soluble samples that are not retained by RP chromatography
- Ion pair chromatography for acids and bases that are poorly retained in RP mode

### KNAUFR columns for the determination of biomolecules: **Eurosil Bioselect**

Why do I need wide pores?



Which modification should I choose?



## HPLC of sugars, organic acids and alcohols: Eurokat

A complex separation consisting of ligand ex- fonated cross-linked styrenedivinylbenzene change, ion exclusion, ion exchange, size exclusion and partition mechanisms. Based on a sul-

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copolymer, 4 ionic forms are available for special applications:

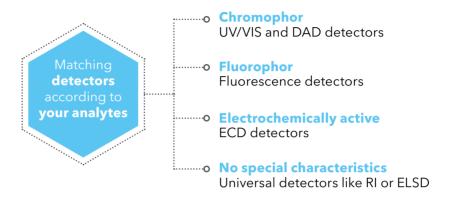
Н	Sugars, organic acids and alcohols	
РВ, Са	Small carbohydrates (mono- and disasccharides)	
Na	Sugar oligomers (up to DP 8)	

Eurokat is ideal for the organic solvent free analysis of:

- Fruit beverages
- Soft drinks
- Wine
- Fermentation broths

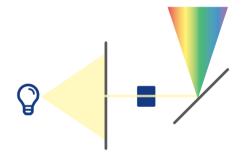
# **Detection**

When do I use which detector? The detection technique depends on the characteristics of the analyte.



### Variable or multiple wavelength detectors (MWD or VWD)

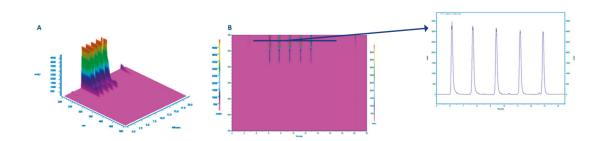
Classical UV detectors record 2D data as a chromatogram. These devices are called VWD or MWD. Nevertheless, 3D-like measurements can be done by scanning the wavelength range. VWD or MWD detectors cannot record 3D data over the whole analysis time but just in programmed time frames.



### Diode array detectors (DAD or PDA)

Devices called DAD or PDA detectors are able to record 3D data in addition to the 2D data over the whole analysis time.

3D data means that full UV spectra are measured and these spectra are plotted over the whole time. This is useful for unknown analytes or target compounds with different UV characteristics. Additionally 2D chromatograms can be extracted from 3D-data at any wavelength.



# **Special detection**

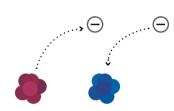
# Highly sensitive and specialized detectors

#### Fluorescence detection

These detectors allow to develop highly selective methods with sensitivities three to six orders of magnitude greater than UV detection.

- + Very sensitive method
- + Easy handling
- Only for fluorescent molecules





#### Electrochemical detection

Very sensitive, selective detection of oxidizable/reducable compounds. In amperometric electrochemical detection the electrical current is measured resulting from oxidation or reduction reactions.

- + Very sensitive method
- + Lowest LODs
- Only for special applications
- Handling is not for beginners

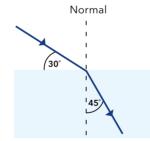
# **Universal detection**

### Refractive index detection (RI)

RI detection is a cost-effective solution for the analysis of sugars, polymers, surfactants and other compounds that do not contain a chromophore. These detectors measure the ability of analyte molecules to bend or refract light.

- + UV absorbing solvents usable
- + Very price attractive
- + Easy to use

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## Ev

Scattered light + Gr

Evaporative light scattering detection (ELSD)

As an universal detector, an ELSD offers numerous possibilities for detecting substances that have few or no chromophores. Since the eluents are evaporated, the use of non-UV-compatible solvents poses no problems and the ELSD is gradient compatible.

+ Gradient elution possible

- No gradient elution

- Comparably low sensitivity

- + Comparably high sensitivity
- Comparably cost-intensive
- Very clean solvents needed
- Nitrogen source needed
- Not suitable for volatile compounds

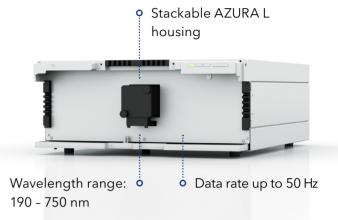
# **Sensitive UV/VIS detectors**

Choose between single variable wavelength, multiple variable wavelength and 3D diode array detectors.

### Single variable wavelength detectors

#### **AZURA® UVD 2.1L**

Target analytes: Molecules carrying a chromophore, all analytes in a sample that absorb at the same wavelength



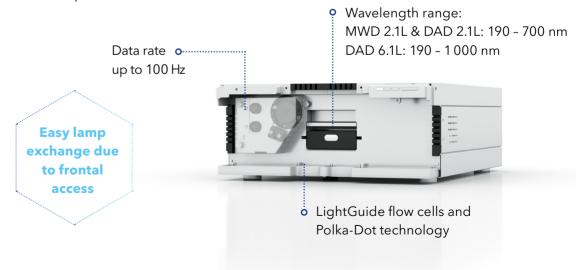


Modern detectors can record data rates up to 100 Hz or even more. These high data rates are needed in very fast UHPLC. For classical HPLC, 20 or 50 Hz are typically sufficient.

Multiple variable wavelength detectors and diode array detectors

#### AZURA® MWD 2.1L, DAD 2.1L and DAD 6.1L

Target analytes: Molecules carrying a chromophore, adsorption at different wavelengths or for method development



# Flow cell cartridges for MWD and DAD detectors

#### PressureProof Flow cells

- Price attractive and robust
- Possibility to couple a second detector or LC dimension caused by the pressure stability up to 300 bar
- Also suited for higher flow rates up to 20 ml/min



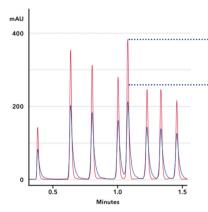
### LightGuide Flow cells

- Total reflection technology for high sensitivity
- Minimized cell volume for high resolution



Comparison of flow cells with 10 mm path length:

PressureProof vs. LightGuide



Higher peaks with LightGuide caused by total reflection technology

 Sharper peaks with LightGuide caused by lower cell volume

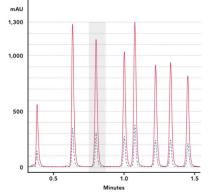


**Blue:** Analytical PressureProof

Red: Standard LightGuide

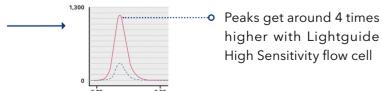
The great advantage of LightGuide flow cells and their low dispersion volume seen in UHPLC does not play a major role in classical HPLC as the complete system dispersion volume is much larger.

### Comparison of LightGuide flow cells: Standard vs. High Sensitivity



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**Blue:** Standard LightGuide with 10 mm path length **Red:** High Sensitivity LightGuide with 50 mm path length



## Which flow cell fits your application best?

Analysis	Typical HPLC column dimension	Main objective	Recommended flow cell
Classical HPLC 25	250 x 4 mm ID / 250 x 4.6 mm ID, 5 μm	Robust method	Analytical PressureProof
Classical HPLC		High sensitivity	High sensitivity LightGuide
HPLC Plus	150 x 3 mm ID, 3 μm	Robust method	Analytical PressureProof
		High resolution and fast runs	Standard LightGuide
		High sensitivity	High sensitivity LightGuide
UHPLC	100 x 2 mm ID, 2 μm	High resolution and fast runs	Standard Lightguide
		High sensitivity	High sensitivity LightGuide
Micro and Nano LC	100 x 0.3 mm ID, 5 μm 50 x 0.075 mm ID, 3 μm	Lowest volume	Nano flow cell with fiber optics

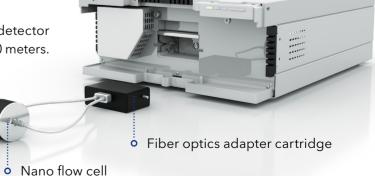




KNAUER flow cells in cartridge design are very easy exchangeable by just clicking them out without any tools.

## **Remote flow cells**

Separate the flow cell spatially from the detector via fiber optic cables in a distance up to 10 meters.

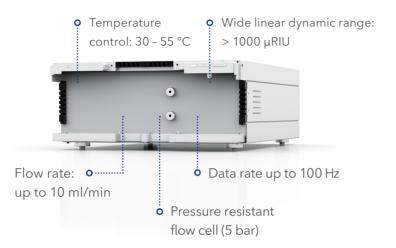


# **Universal detectors**

#### **AZURA® RID 2.1L**

The AZURA RID 2.1L is a highly competitive and sensitive refractive index detector, ideal for fast and reliable routine analysis of non-UV absorbing substances. The intelligent temperature control guarantees fast baseline stabilization and stable operation.

Target analytes: Alcohols, sugars, saccharides, fatty acids and polymers



Light Scattering Detector Sedex 85LT, Sedex 90LT and Sedex 100LT Sensitive universal detection with the possibility to run gradients



As an universal detector, an ELSD offers numerous possibilities for detecting substances that have few or no chromophores. Since the eluents are evaporated, the use of non-UV-compatible solvents poses no problems and the ELSD is gradient compatible.

Target analytes: Carbohydrates and similar compounds, detergents, ionic and non-ionics, artificial sweeteners, antioxidants, amino acids, lipids, peptides, polymers, pestizides, proteins, steroids



ELSD is a good replacement for a Charged Aerosol Detector (CAD). Just choose the right fitting nebulizer and get the best out of the ELSD for your analyte detection.

# **Special detectors**

One of the smallest single variable wavelength detectors on the market

#### **AZURA® UVD 2.1S**

The AZURA UVD 2.1S offers excellent technical specifications for routine laboratory work. With its small footprint, it is one of the smallest detectors for HPLC on the market.





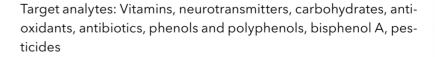
#### Fluorescence Detector RF-20A

The fluorescence detector RF-20A provides world-class sensitivity, excellent maintainability and diverse validation / support functions. It supports a wide range of applications in the wavelength range of 200 to 650 nm from conventional analysis to high-performance analysis.

Target analytes: Fluorecent molecules like polycyclic aromatic hydrocarbon, fluorescence tagged analytes like amino acids or proteins

#### **Electrochemical Detector AZURA® ECD 2.1**

With its measurement frequency up to 100 Hz, this electrochemical detector is specially designed for super-fast highly sensitive and selective measurement of oxidizable and reducible substances in (U)HPLC. The AZURA ECD 2.1 comprises of a thermostat-controlled Faraday's cage, accommodating column and flow cell.







#### **Interface Box IFU 2.1 LAN**

The KNAUER interface box IFU 2.1 LAN allows highly precise analog data acquisition of third party modules over analog and relay outputs. Example: MALS-detectors for molecular weight determination.

# **Software**

### **Mobile Control**

KNAUER's monitoring and control software for AZURA® devices and systems

With the hand-held Mobile Control (Chrom) option you have your AZURA devices at your fingertips. Remotely control and monitor your system and enjoy the touchscreen-optimized user interface. Choose Mobile Control as an easy-to-use and cost-effective software solution!



Mobile Control provides full access to AZURA devices. Change device settings, set operating parameters, automate device control or check the system status... Mobile Control features all functionalities of a display. Do you want more than a display without using an advanced chromatographic data system?

#### **Mobile Control Chrom**

Mobile Control Chrom features data acquisition from AZURA detectors in addition to full device control.

#### **OpenLAB® CDS EZChrom Edition**

OpenLAB CDS EZChrom Edition is the next generation of chromatography data systems. It provides chromatography data acquisition, processing and control of GC and LC chromatographs and is used in chromatography operations ranging from single user/single instrument to multi-user/multi-instrument laboratories.

#### ClarityChrom® CDS

ClarityChrom is an easy-to-use chromatography data system for workstations. The optional GPC-and Knauer FRC control modules and extensions for PDA, SST, SEC/GPC and MS allow using the software for a wide range of applications.



#### **Chromeleon™ 7.2 Drivers**

Thermo Scientific™ Dionex™ Chromeleon™ is one of the most wide-spread chromatography data systems. Its intuitive handling benefits laboratory workflow and the highly developed algorithms simplify data processing. It offers a broad range of third-party drivers and can be easily used with existing HPLC systems. KNAUER offers drivers for a lot of its devices.



# **AZURA® Analytical HPLC systems**

Product	Features	Page
AZURA Analytical UHPLC system	This system features a AZURA P 6.1L HPG pump, an autosampler 6.1L, a column thermostat CT 2.1, a DAD 6.1L UV/VIS detector with 8-variable UV measuring channels from 190 - 1000 nm, as well as a Tablet PC with AZURA Mobile Control. It is the UHPLC system with a backpressure range of 1000 bar. It perfectly fits the demanding needs of a method development system, as well as the robust fitness of a routine analysis machine.	24
AZURA Educational system	Complete isocratic analytical HPLC system, UV/VIS detector with one variable wavelength, pump unit with pressure sensor for a low pulsation eluent supply, optional AZURA Mobile Control app allows direct control of all integrated modules.	25
AZURA Analytical system for analysis of carbonyl emissions in air	HPLC up to 700 bar in LPG version for quaternary gradients, 10 mL pump head, 8-channel DAD detector with 3D UV/Vis detection at 190-700 nm and up to 100 Hz, column thermostat, automated injection via autosampler, Mobile Control for monitoring and control of the integrated modules, Clarity-Chrom CDS for instrument control, data acquisition and data processing.	26
AZURA GPC Cleanup system	The system features 15 sample loops controlled by two 16 port multiposition valves. With an additional multiposition valve 15 fractions and waste can be collected. All multiposition valves and the manual injection valve are integrated into a compact AZURA® Assistant ASM 2.1L. Elution of separated substances/ standard components is monitored by a variable single wavelength UV detector. A second assistant harbors the small detector, the system pump with a pressure sensor and a valve to bypass the GPC column or select between two columns, Mobile Control for system control.	27
AZURA sugar system	HPLC up to 700 bar in isocratic version, 10 mL pump head, refractive index detector, column thermostat, manual injection via an injection valve, Mobile Control for monitoring and control of the integrated modules, ClarityChrom CDS for instrument control, data acquisition and data processing.	28

# **Polyphenol analysis**

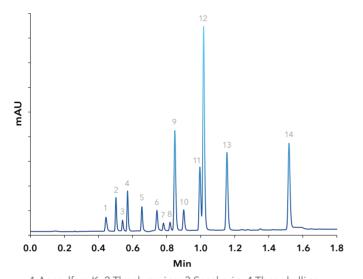
# in chocolate

Determination of 14 compounds in 1.5 minutes using the AZURA® UHPLC system

#### **UHPLC** for quality control of dark chocolate

 Polyphenols and alkaloids like caffeine or theobromine





#### **UHPLC** system

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Analytical HPG configuration with DAD detection

1 Acesulfam K; 2 Theobromine; 3 Saccharin; 4 Theophylline; 5 Caffeine; 6 Chlorogenic acid; 7 Catechin; 8 Epicatechin; 9 4-Hydroxbenzoic acid; 10 Vanillin; 11 Guaiacol; 12 Sorbic acid; 13 Methylparaben; 14 Propylparaben



Key features
HPG pump, 0.001-5 ml/min, sst, max. 1000 bar
Diode array detector, D <sub>2</sub> lamp, 190-700 nm, max. 100 Hz
Forced air column thermostat, 5 - 85 °C
Analytical autosampler, 0.1 μl - 10 ml injection volume, cool/heat option, 108 vial positions, max. 1240 bar

# **How do you explain HPLC?**

# AZURA® Educational system

### For tomorrow's HPLC professionals

Your AZURA Educational System includes printed training material that explains several applications of HPLC. For example, the quantitative determination of caffeine and paracetamol in samples of your choice (e.g. coffee, soft drinks, medicine) is explained in detail.



In simple steps, students are introduced to basic concepts and terms used in HPLC such as calibration curve and internal standard. The easy to understand instructions guide them through the complete experiment, including setup, run and analysis.



### What is included?

Devices	Key features	
HPLC system AZURA Educational System	AZURA Compact HPLC with pump P 4.1S and detector UVD 2.1S with flow cell (10 mm path length, 1/16" connectors)	
Sample application	Manual injection valve including 10 μl sample loop HPLC injection syringe (straight needle), 100 μl volume injection valve mounting bracket	
Start-up kit AZURA	Capillaries and fittings (PEEK, 1/16")	
Accessories	Eluent tray E 2.1L set of mobile phase bottles (2 x 1000 ml) tool kit for HPLC system setup	
PC	Laptop with pre-installed Windows	
PC communication	Router for LAN connection (8 x)	
Software for controlling and recording	g ClarityChrom software and licence for the AZURA Compact HPLC system	
HPLC Column	Eurospher II 100-5 C18 with integrated precolumn, 125 mm x 4 mm ID, magnetic clip to attach column	

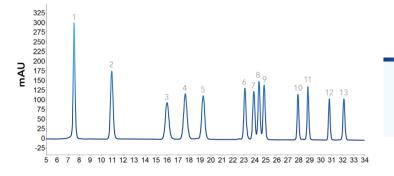
# **Analysis of carbonyl emissions** in air

In the monitoring of industrial air, the determination of carbonyl (aldehyde and ketone) emissions is crucial to prevent respiratory, pulmonological, autoimmune diseases or cancer.

#### **HPLC solution by KNAUER**

According to the analytical method described in DIN ISO 16000-3, the carbonyls must be converted to their corresponding hydrazones with 2,4-dinitrophenylhydrazine (DNPH) in order to be detected via UV detector and analyzed by reversed phase HPLC.

For method optimization DryLab® has been used and the detailed application note (VEV0078) is available on the KNAUER website: "Systematic HPLC method development and robustness evaluation of 13 carbonyl DNPH derivatives using DryLab®".



1 Formaldehyde-DNPH; 2 Acetaldehyde-DNPH; 3 Acetone-DNPH; 4 Acroleine-DNPH; 5 Propionaldehyde-DNPH; 6 Crotonaldehyde-DNPH; 7 2-Butanone-DNPH; 8 Methacroleine-DNPH; 9 n-Bytaldehyde-DNPH; 10 Benzaldehyde-DNPH; 11 Valeraldehyde-DNPH; 12 m-Tolualdehyde-DNPH; 13 m-Tolualdehyde-DNPH

#### **System** (A46001)

HPLC Plus System with 8-channel DAD detector, LPG pump (700 bar), autosampler and column thermostat, DNPH-column

#### **Optional hardware and software**

Chromatography workstation for OpenLab/Chromeleon, Mini PC, WIN 7, 64 bit, 24" Monitor

Chromatography workstation for ClarityChrom®, Mini PC, WIN 10, 64 bit, 24" Monitor

ClarityChrom® single instrument license one time base

OpenLAB® CDS EZChrom edition workstation for one system with SMA and 4x system suitability

Chromeleon™ 7.2 driver

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Optional service: Installation



This gradient HPLC system perfectly meets even sophisticated analysis demands.



# **Sample preparation**

# for the analysis of pesticides

Preparative system for general cleanup tasks relying on gel permeation

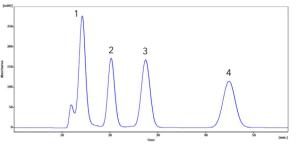
#### **GPC Cleanup system by KNAUER**

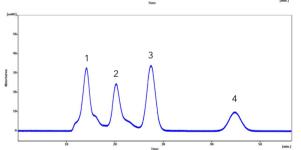
- Saves time with automated sample preparation
- Improves the reproducibility and quality of your analytes
- Fully automatically processes up to 15 samples while requiring minimal bench space
- Can be customized to fulfill individual requirements



The AZURA GPC cleanup system was selected by the Analytical Scientist for "The Innovators 2017" product.







Four pesticide standards with US EPA method 3640A and a chromatogram of spiked olive oil.



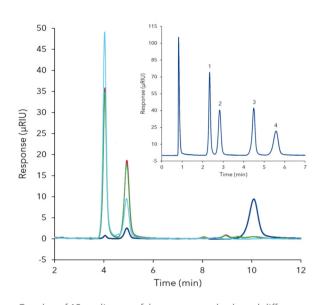
Devices	Key features
AZURA Assistant ASM 2.1L	UVD 2.1S 6 port 2 pos valve, 1/16", sst, 300 bar P 4.1S, 10 ml, sst, 50 bar
AZURA Assistant ASM 2.1L	3 valves: 16 port 16 pos, 1/16", sst, 50 bar
AZURA GPC tubing guide	16 sample loops (1 ml each)
Column	BioBeads SX-3

# **Sugar system**

# for the determination of sugar monomers

### **Honey and its substitutes**

- Differentiate between natural honey and possible substitutes of food industry
- Determination of sugar monomers like fructose (1), glucose (2), sucrose (3) and maltose (4)
- Highest reproducibility in shortest time



Overlay of 12 replicates of the sugar standards and different honey and honey substitute samples.



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Devices	Key features
AZURA Pump P 6.1L	LPG pump, 0.001-10 ml/min, sst, max. 700 bar
AZURA Detector RID 2.1L	Refractive index detector, max. 5 bar, max. 100 Hz
AZURA Column Thermostat CT 2.1	Forced air column thermostat, 5 - 85 °C, 2 °C/min
AZURA Valve V 2.1S	6 port 2 pos injection valve, 1/16", sst, 400 bar
Vertex Plus column Eurospher II NH2	150x4 mm, 100 Å, 3 μm

HILIC amino phase for fast and high resoluti-

on separation of sugars.

# **Accessories**

### **AZURA Eluent Tray E 2.1L**

The eluent tray E 2.1L for AZURA devices with a capacity of 6 x 1000 ml bottles is stackable onto all AZURA devices. The inlay is removeable for cleaning. The eluent tray possesses a drainage system and a removable front that facilitates access to a capillary guide.



The AZURA Tubing kit with solvent filter (stainless steel, 10  $\mu$ m) is suitable for all analytical HPLC systems (isocratic, HPG, LPG). The filter can be used with all flow rates of the pump heads provided.

#### Safety caps

Why should I use safety caps?

- Stop the liquid flow line via stopcocks whenever no flow is delivered. Be safe from eluent loss!
- Filters and air-inlet valves prevent the eluent from evaporating and result in maximized stability of retention times due to stable eluent composition.
- Filters at the waste bottle keep the air around your AZURA system clean.







# **Technical data**

AZURA Pump P 6.1L	Low pressure gradient HPLC Plus	Low pressure gradient UHPLC	High pressure gradient HPLC Plus	High Pressure gradient UHPLC
Max. flow rate	10 ml/min	5 ml/min	10 ml/min	5 ml/min
Flow rate range	0.001 - 10 ml/min	0.001 - 5 ml/min	0.001 - 10 ml/min	0.001 - 5 ml/min
Flow rate increment	0.001 ml/min	0.001 ml/min	0.001 ml/min	0.001 ml/min
Maximum delivery pressure [psi]	10 150 psi	14 500 psi	10 150 psi	14 500 psi
Maximum delivery pressure [bar]	700 bar	1 000 bar	700 bar	1 000 bar
Maximum delivery pressure [MPa]	70 MPa	100 MPa	70 MPa	100 MPa
Pump head materials	Stainless steel	Stainless steel	Stainless steel	Stainless steel
Gradient	Low pressure gradient (quaternary)	Low pressure gradient (quaternary)	High pressure gradient (binary)	High pressure gradient (binary)
Leak management	Yes	Yes	Yes	Yes
Best working conditions	0.1 - 8.0 ml/min	0.02 - 5 ml/min	0.1 - 8.0 ml/min	0.02 - 5 ml/min
Continous working conditions	0.1 - 4.0 ml/min	0.1 - 4.0 ml/min	0.1 - 4.0 ml/min	0.1 - 4.0 ml/min

Heating and cooling system	microprocessor controlled Peltier element for heating and cooling, fan supported 2-way air circulation
Temperature range	5 - 85 °C
Heating/cooling rate	2 °C/min
Temperature accuracy	± 0.2 °C
	± 0.1 °C
	90 x 390 x 47 mm (W x H x D)
Safety	self-check and auto-calibration at power-on, selectable turn-off temperature

Method	HPLC Plus	UHPLC
Autosampler Flow Path	Analytical	Analytical
Maximum back pressure	700 bar	1240 bar
Vial/plate dimensions	max. plate/vial height: 47 mm (incl. septa or capmat)	max. plate/vial height: 47 mm (incl septa or capmat)
Injection volume range	0.1 μl - 10 ml depending on sample loop	0.1 μl - 10 ml depending on sample loop
Sample loop	100 μΙ	10 μΙ
Dispenser syringe	250 μΙ	250 μΙ
Headspace pressure	built-in compressore, only for sample vials with septum	built-in compressor, only for sample vials with septum
Switching time inj. valve	< 100 ms	< 100 ms
Piercing needle precision	± 0.6 mm	± 0.6 mm
Sample tray cooling	none	none
Vial detection	missing vial/well plate detection by sensor	missing vial/well plate detection by sensor
Needle wash	programmable: wash between injections and wash between vials	programmable: wash between injections and wash between vials
Wetted materials	Tefzel® (ETFE), Glass, Teflon® (PTFE), Kel-F® (PCTFE), stainless steel, PEEK	Tefzel® (ETFE), Glass, Teflon® (PTFE) Kel-F® (PCTFE), stainless steel, PEEK
Injection modes	full loop filling, partial loop filling and microliter pickup; PASA™ (pressure-assisted sample aspiration)	full loop filling, partial loop filling and microliter pickup; PASA™ (pressure-as sisted sample aspiration)
Injection precision	RSD (Relative Standard Deviation): full loop injection: <0.3%partial loop injection at injection volume>5 $\mu$ l: <0.5%microliter pickup injection at an injection volume>5 $\mu$ l: <1.0%	RSD (Relative Standard Deviation):ful loop filling: <0.3%partial loop injection at injection volumes>5 μl: <0.5%micro liter pickup at injections>5 μl: <1.0%
Sample carryover	< 0.05 % with needle washing	< 0.05 % with needle washing
Injections per vial	max. 9 injections	max. 9 injections
Injection cycle time	minimum 7 s from the same vial, 14 s from different vials;< 60 s for>100 µl sample injection in all injection modes, incl. 300 µl needle wash	minimum 7 s from the same vial, 14 from different vials;< 60 s for>100 µ sample injection in all injection modes incl. 300 µl needle wash
Analysis time	max. 9 h, 59 min, 59 s	max. 9 h, 59 min, 59 s
Tray Tempering	Optional (4-40°C)	Optional (4-40°C)

# **Technical data**

AZURA Detector	DAD 2.1L	DAD 6.1L
Detector type	Diode array detector	Diode array detector
Number of diodes	256	1024
Pixel pitch	2 nm/diode	0.8 nm/diode
Detection channels	8 (Digital)/4 (Analog)	8 (Digital)/4 (Analog)
Light source	Deuterium (D²) lamp with integrated GLP chip	High brightness deuterium (D²) lamp and halogen lamp with integrated GLP chip
Wavelength range	190 - 700 nm	190 - 1000 nm
Spectral bandwidth	<10 nm at Ha line (FWHM) /Note: digital bandwidth 1 - 32 nm	< 3.5 nm at Ha line (FWHM) /Note: digital bandwidth 1 - 32 nm
Wavelength accuracy	± 1 nm	± 1 nm
Noise	± 5 μAU at 254 nm	± 3.5 μAU at 254 nm
Drift	400 μAU/h at 254 nm	300 μAU/h at 254 nm
Linearity	> 2.0 AU at 274 nm	> 2.5 AU at 274 nm
Maximum data rate	100 Hz (LAN)/12.5 Hz (analog)	100 Hz (LAN)/12.5 Hz (analog)
Flow cell	Not included (see Accessories / Spare parts)	Not included (see Accessories / Spare parts)
Time constants	0.00 / 0.01 / 0.02 / 0.05 / 0.1 / 0.2 / 0.5 / 1.0 / 2.0 / 5.0 / 10.0 s	0.00 / 0.01 / 0.02 / 0.05 / 0.1 / 0.2 / 0.5 / 1.0 / 2.0 / 5.0 / 10.0 s
Integration time	Automatic	Automatic

AZURA Detector	MWD 2.1L	UVD 2.1L	UVD 2.1S	
Detector type	Variable multiwave- length detector	Variable single wave- length UV detector	Variable single wave- length UV detector	
Detection channels	8 (Digital)/4 (Analog)	1	1	
Light source	Deuterium (D²) lamp with integrated GLP chip	Deuterium (D²) lamp with integrated GLP chip	Deuterium (D²) lamp with integrated GLP chip	
Wavelength range	190 - 700 nm	190 - 750 nm	190 - 500 nm	
Spectral bandwidth	< 10 nm at Ha line (FWHM) /Note: digital bandwidth 1 - 32 nm	11 nm at Ha line (FWHM)	13 nm at Ha line (FWHM)	
Wavelength accuracy	± 1 nm	± 2.5 nm	± 3 nm	
Noise	± 5 μAU at 254 nm	± 1.5 x 10-5 AU at 254 nm (ASTM E1657-98)	± 2.0 x 10-5AU at 254 nm (ASTM E1657-98)	
Drift	400 μAU/h at 254 nm	3.0 x 10-4 AU/h at 254 nm (ASTM E1657-98)	3.0 x 10-4AU/h at 254 nm (ASTM E1657-98)	
Linearity	> 2.0 AU at 274 nm	> 2.0 AU at 270 nm (ASTM E1657-98)	> 2.0 AU at 270 nm (ASTM E1657-98)	
Maximum data rate	100 Hz (LAN)/12.5 Hz (analog)	50 Hz (LAN)/20 Hz (Analog)	50 Hz (LAN)/20 Hz (Analog)/10 Hz (RS-232)	
Flow cell	Not included	Not included	Not included	
Time constants	0.00 / 0.01 / 0.02 / 0.05 / 0.1 / 0.2 / 0.5 / 1.0 / 2.0 / 5.0 / 10.0 s	0.0 / 0.1 / 0.2 / 0.5 / 1.0 / 2.0 / 5.0 / 10.0 s	0.00 / 0.02 / 0.05 / 0.1 / 0.2 / 0.5 / 1.0 / 2.0 s	
AZURA Detector	RID 2.1L			
Detector type	Refractive index detector			
Light source	Long-life LED			
Detection channels	1			
Refractive index range	1.00 - 1.75 RIU			
Noise	± 2.5 nRIU			
Drift	200 nRIU/h			
Linearity	> 1000 μRIU			
Flow cell	5 bar back pressure resistance Flow cell included			
Maximum flow rate	10 ml/min (pure water)			
Flow cell volume	15 μΙ			
Wetted materials	Stainless steel / fused sili	ca / PTFE / PEEK		
Temperature control	OFF, 30 - 55 °C (1 °C increment)			
Time constants	0.00 / 0.01 / 0.02 / 0.05 / 0.1 / 0.2 / 0.5 / 1.0 / 2.0 / 5.0 / 10.0 s			
Maximum data rate	100 Hz (LAN)/20 Hz (Anal	og)		

# **Science Together**







Worldwide partner in science since 1962

Based in Berlin, KNAUER is a medium-sized, owner-managed company that has been serving the sciences since 1962. We develop and manufacture scientific instruments of superior quality for liquid chromatography. The range includes sys-

tems and components for analytical HPLC / UHPLC, preparative HPLC, fast protein liquid chromatography (FPLC), multi-column chromatography / simulated moving bed (SMB), and osmometry.





Roswitha are still active as advisers in the company to this day. The couple's daughter, Alexandra Knauer, has been managing director and KNAUER a "leading employer".

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The founder Dr. Herbert Knauer and his wife owner of the company since 2000. Several awards for outstanding products and innovations as well as entrepreneurial excellence make

> We separate molecules and unite people.

www.knauer.net f in 6







# **System configurator** HPLC/UHPLC by KNAUER

#### MAKE YOUR PRESELECTION

☐ UHPLC

(551, max. 1000 bar)	(551, max. 700 bar)	(metal-free, max. 400 bar)		
pump pump		column 2x/6x	detector I	detector II

$\rightarrow$ $\rightarrow$ $\rightarrow$	$\rightarrow$	<b>&gt;</b>	<b>&gt; &gt;</b>	$\rightarrow$
ELUENT SELECTION & DELIVERY	SAMPLE INJECTION	COLUMN SELECTION & THERMOSTAT	DETECTION	
☐ 5 ml/min binary gradient pump P 6.1L (UHPLC)	Manual injection valve	☐ 2 columns	UV/VIS single wavelength	☐ DAD 2.1L☐ DAD 6.1L
☐ 5 ml/min quaternary gradient pump P 6.1L (UHPLC)	☐ Autosampler AS 6.1L ☐ Autosampler AS 6.1L cool/heat	☐ Column thermostat	☐ UV/VIS multiple wavelength	☐ Fluorescence Detector RF-20 A
□ 10 ml/min binary gradient pump P 6.1L	cooi/neat	☐ Column kit HPLC	☐ Conductivity	☐ Fluorescence Detector RF-20 Axs
☐ 10 ml/min quaternary pump P 6.1L		☐ Eluent preheater 0.1 mm ID UHPLC	☐ Refractive index ☐ Light Scattering	☐ ECD 2.1
<b>x</b> solvent selection valve (6 further inlets)		☐ Eluent preheater 0.18 mm ID HPLC	☐ A/D-converter (integration of further detectors)	
ACCESSORIES				
□ 0.1 mm tubing	□ 0.18 mm tubing	☐ PEEK tubing	<b>x</b> Back pressure regulator	☐ Workstation (Windows)
FLOW CELLS FOR UV-DE	TECTOR			
□ 10 mm/10 µl Pressure proof	□ 10 mm/2 μl LightGuide®	□ 50 mm/6 µl LightGuide®	3 mm/2 μl (up to 100 Pressure proof	ml/min)
SOFTWARE				
☐ ClarityChrom®	☐ OpenLAB®	☐ Chromeleon <sup>™</sup>	☐ Mobile Control	
COMMON APPLICATIONS				
☐ Reversed phase	☐ Normal phase	other	☐ System Qualification	



# Analytical HPLC

Multi-Column Chromatography, SMB

Preparative HPLC

**FPLC** 

Osmometry

Dosing, Metering, Pumping

Detection

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