



# UV-visible spectroscopy solutions for chemical analysis



**Agilent Technologies**

# Agilent's UV-visible systems

UV-visible spectroscopy solutions that satisfy the diverse needs of analysts in routine QA/QC, research, and method development laboratories.

The Agilent 8453 UV-visible spectro-photometer offers the latest in diode-array technology:

- compliance with all requirements of the European Pharmacopoeia (EP) and United States Pharmacopoeia (USP)
- small footprint to save bench space
- prealigned deuterium and tungsten lamp light sources for trouble-free maintenance
- built-in buttons to measure sample, standard, and blank for convenient measurements even when wearing gloves
- thermally stable ceramic spectrograph for a wide operating temperature range
- communication through GPIB or LAN for dedicated or networked PC control
- firmware upgrade from PC for easy participation in future developments
- built-in GPIO interface for control of accessories

## Good Laboratory Practice

The Agilent 8453 supports compliance with GLP regulations:

- serial and firmware revision number held in firmware
- own clock for time and date stamps of the spectra
- extensive self-test procedures that check the electronics and key optical characteristics to ensure consistent performance between validation
- built-in electronic logbooks which contain the results of self tests, notes on instrument
- maintenance, events and errors

## Optical performance

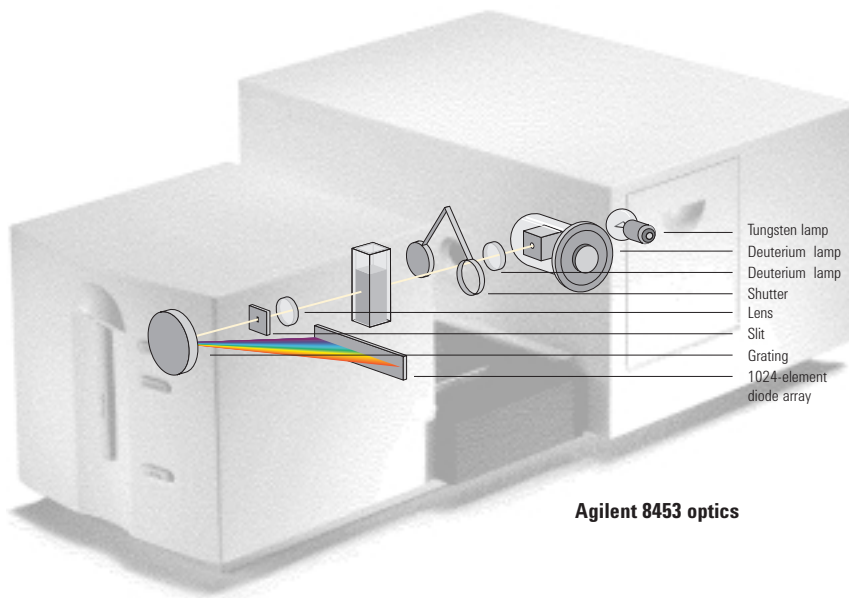
The Agilent 8453 offers the advantages of a diode array plus significant improvements in optical performance:

- 190–1100-nm wavelength range
- 1-nm slit width
- <0.03 % stray light

## The diode-array advantages

Backed by over two decades of experience in the development and manufacture of diode-array spectrophotometers, the Agilent 8453 gives you clear advantages.

- Fast spectral scanning for complete spectral information useful for:
  - proof of identity and pureness of the sample
  - additional information for “out of specification test result” investigation in a regulated environment
  - re-evaluation with different method
  - multi-wavelength applications such as user-defined equations
- Open sample area for convenient sample handling.
- High throughput optics guarantees excellent signal-to-noise for high sensitivity.
- Virtually absolute wavelength resettability allows selection of optimum wavelength and use of electronic standards.
- Exceptional ruggedness and reliability.



## Specifications – Agilent 8453 UV-Visible spectrophotometer

### Optical performance

Wavelength range	190–1100 nm	
Slit width	1 nm	
EP resolution test	>1.6	toluene in hexane, ratio abs. at 269 nm/266 nm
Stray light	<0.03 %	at 340 nm (NaNO <sub>2</sub> , ASTM)
	<0.05 %	at 220 nm (NaI, ASTM)
	<1 %	at 200 nm (KCl, EP)
Wavelength accuracy	<±0.5 nm	0.5-second scan (NIST 2034)
	<±0.2 nm	at 486.0 and 656.1 nm
Wavelength reproducibility	<±0.02 nm	ten consecutive scans (NIST 2034)
Photometric accuracy	<±0.005 A	at 440.0, 465.0, 546.1, 590.0, and 635.0 nm, 1 A (NIST 930e)
	<±0.01 A	at 235, 257, 313, 350 nm, (potassium dichromate, EP method)
Photometric noise	<0.0002 A	sixty 0.5-second scans at 0 A, 500 nm, rms
Photometric stability	<0.001 A/h	at 0 A, 340 nm, after 1-hour warm up, measured over 1 hour, every 5 seconds, constant ambient temp.
Baseline flatness	<0.001 A	0.5-second blank, 0.5-second scan, rms
Typical scan time	1.5 second	full range
Shortest scan time	0.1 second	full range
Time until next scan	0.1 second	full range, 0.1-second scan, up to 150 consecutive scans

### Physical dimensions

Height x width x depth	185 x 344 x 560 mm (7.3 x 13.5 x 22.0 inches)
Weight	16.5 kg (36.3 lb)

### Power requirements

Line voltage	90–264 V AC
Line frequency	47–63 Hz
Power consumption	70 VA typical

### Environmental conditions

Operating temperature	0–50 °C (32–122 °F)
Non-operating temperature	-40–70 °C (-4–158 °F)
Humidity	<95 %, at 25–40 °C (77–104 °F)



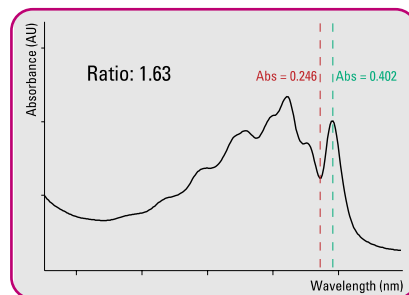
Sipper operation is easy with built-in measurement buttons



Side door for easy lamp change



Open sample area means large accessories are easy to use—here the Peltier cell holder



Spectrum of a 0.02 % v/v solution of toluene in hexane

# Solutions for affordable performance

## Easy, ergonomic, exact, excellent

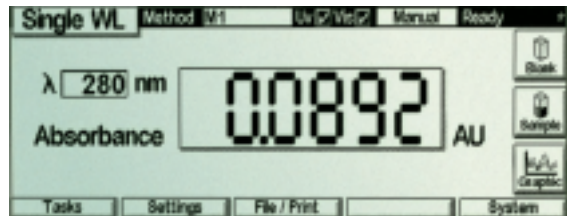
Whether you are an experienced spectroscopist or just a beginner, making measurements with the 8453E system could not be easier.

- Intuitive user interface suits beginners and experienced users.
- Structured task concept prompts user for necessary parameters.
- Extensive online information provides extra help when needed.
- Plug-and-go installation for easy addition of accessories.
- Early maintenance feedback (EMF) guarantees reliable results.

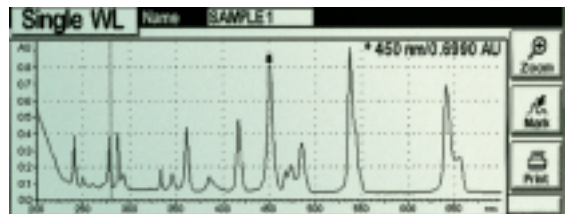
## Simplicity in the palm of your hand

Simple dialogs guide you through blank, standard and sample measurements, and the results are shown numerically and graphically.

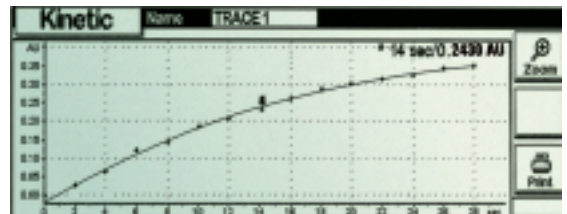
**Get fast results – start the measurement and within seconds you get a complete spectrum and the result is shown in clear, easy-to-read digits.**



**Get even more information – the liquid crystal display gives you more than just digital readout. Display the complete spectrum for review and then print a complete report.**



**Watch measurement in progress – monitor enzyme kinetic measurements during the run and view rate calculations graphically.**



*"The power and flexibility to develop the optimum method and solve the toughest problems"*

### The challenge

Provide performance and simplicity at an affordable price.

### The solution

The Agilent 8453E UV-visible spectroscopy system combines optical performance of the Agilent 8453 with operational simplicity of a handheld controller at a price that fits today's tight laboratory budgets.

## Specifications – handheld controller for 8453E

Handheld controller with numeric and softkeys, high resolution LCD screen (640 x 200 pixels). PCMCIA memory card for storage of data and methods. Built-in tasks for measurements at single or multiple wavelengths for data evaluation based on user-defined equation, for spectral measurements, for quantification, or for time-based measurements as used in kinetics experiments.

Note that with exception of kinetics all measurements contain full spectra.

- Wavelength settings can be changed without remeasuring
- Data analysis mode can be changed without remeasuring, for example, quantification and equation.
- Spectral information can be used for identification, peak/valley find, or to diagnose measurement problems.

<b>Data types</b>	Abs., %T, first through fourth derivative.
<b>Multiple wavelengths</b>	Up to four wavelengths, additional background correction.
<b>Equation</b>	User defined with up to four wavelengths, +, -, /, x, ln, log, sqr, sqrt, exp.
<b>Spectrum</b>	Interactive data type conversions and spectral arithmetic
<b>Quantification</b>	Single component quantification; additional background correction; four calibration curve types, calibration equation and statistics.
<b>Kinetics</b>	Absorbance time trace at a single wavelength with online graphic display; delta absorbance, zero and first-order rate calculation; conversion factor for multiplication.
<b>Common tasks features</b>	Meaningful names for samples and methods; graphic display of spectrum including zoom, cursor, peak/valley find and print; load and save of data to PCMCIA card; load and save parameters as method; print result report.
<b>Data transfer</b>	Export of spectra or time traces as ASCII files by PCMCIA card to other applications running on a PC.
<b>Diagnostic and tests</b>	Selftest; lamp intensity, stability, dark current; procedures for performance testing; early maintenance feedback (EMF) for lamps; electronic logbooks.
<b>Supported accessories</b>	Sipper and multicell transport; printers that support control by printer command language PCL 3 or higher.
<b>Language</b>	English and French (user-selectable) or Chinese.

*"The power and flexibility to develop the optimum method and solve the toughest problems"*



Choose what you want to do in the first screen – a variety of tasks are available which match the types of task performed in almost any laboratory.

# Solutions for general purpose tasks

## Easy to use

The Agilent 8453 UV-visible spectroscopy system may well be the easiest UV-visible system you will ever use. Three steps are all you need to get results.

## Graphical user interface

Usability tests performed with actual users in their own labs initiated a completely new approach to user interface design. The Agilent 8453 UV-visible spectroscopy system uses symbolic graphics to:

- make the system intuitive, easy to learn, and easy to use,
- provide confirmation of current status at a glance,
- provide visual confirmation of actions, and
- provide fast access to frequently performed activities.

## Automation

A single dialog box makes setup for automated analyses easy and quick. You can use automation to guide an operator through a series of manual measurements, or combine it with sampling accessories to provide semi- or fully automated analysis. The automation function controls

- the Agilent sipper system
- Agilent XY autosampler or Gilson autosampler
- the Agilent multicell transport

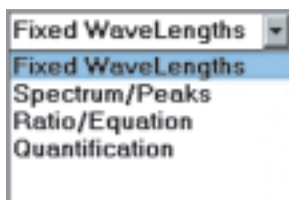
The optional control samples allow you to run an automatic system suitability test before quantitative analyses.

## Compatibility

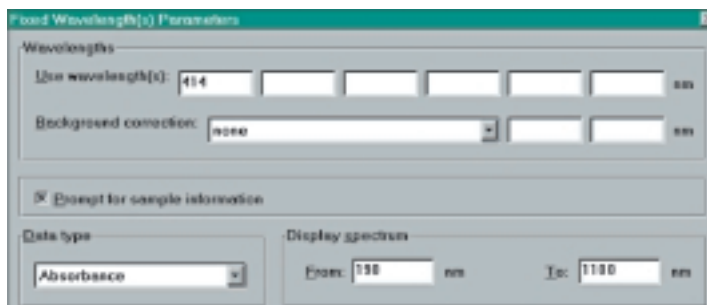
The Agilent ChemStation uses the Microsoft Windows environment so you can run other programs, such as word processors and spreadsheets, at the same time, and transfer data using copy-and-paste.

*"Easy to learn and easy to use for increased productivity"*

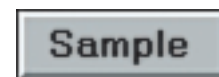
1. **Select the task you want**



2. **Enter the parameters**



3. **Do the measurement**



### The challenge

*Provide a powerful and productive system for routine and occasional users that is easy-to-use and integrates seamlessly in the modern laboratory.*

### The solution

*The Agilent 8453 UV-visible spectroscopy system, combining the PC-controlled Agilent 8453 spectrophotometer with the Microsoft Windows based Agilent ChemStation software.*

### Clear and simple reports

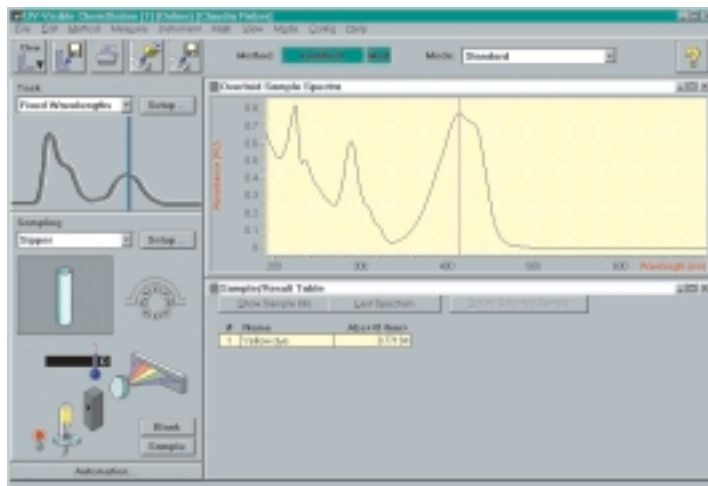
Generating reports could not be easier. Simply click the printer icon in the toolbar and a standard report is printed. When possible this standard report is printed on a single sheet of paper. The report format depends upon the task in use but all reports include essential GLP information such as method and data file name, date, time, operator, overlay of sample spectra and result table.

### Productivity

The Agilent 8453 system is a highly productive UV-visible spectroscopy system. Its combination of ease of learning, ease of use, fast spectral acquisition, fast data evaluation, report generation, and automation lets you get results fast – even if you only use the system occasionally.

Add diagnostic tools such as performance verification, system documentation, and the extreme reliability of the system and you will spend the minimum time with administration or maintenance and repair.

### Your results are displayed automatically



### Print a report



# Solutions for research and method development

## Powerful data analysis

The advanced software provides unprecedented interactive and programmable data analysis capability:

- 21 mathematical functions for processing spectra that you can combine in any sequence
- flexible use of any number of single or multiple wavelengths, or average a range of values
- evaluation using user-entered equations, single-component or multi-component analysis
- up to four different types of data analysis performed on one set of data in parallel
- use confirmation analysis with quantitative methods to check the identity and purity of samples, and to detect if measurements are being made outside the linear range of the analysis

## Method development tools

Four special utilities are provided to assist the user in developing the best parameters for quantitative analysis:

- Evaluate Standards for linearity – performs a single component calibration at each wavelength over a user-specified wavelength. It determines the correlation coefficient and uncertainty at each wavelength.
- Compare Calibrations – puts the results from two independent calibrations side by side on the screen for comparison.
- Optimize Wavelength for selectivity and accuracy – quantifies a user-selected sample at all wavelengths and plots the quantification results against wavelength.
- Test Method for precision – calculates the average and standard deviation of multiple analyses of an identical sample.

## Multicomponent analysis

Multicomponent UV-visible analysis can provide a real alternative to time-consuming separations. Fast electronic scanning, spectral curve fitting of standards to unknowns, excellent wavelength reproducibility, and maximum-likelihood statistics make sure that results are superior to those obtained with conventional mechanical-scanning spectrophotometers. Calibration is simple and fast using pure standards or mixtures of standards. Statistics for the fit of the standards to the sample spectra give you confidence in your results.

## Customization

Virtually every aspect of the system can be customized through macro programming.

A series of predefined reports are available in the software. However, to fulfill specific needs, the user can define their own customized reports based on the predefined reports or user defined template.

## Electronic records and signatures

The advanced mode can be combined with the security pack to support 21 CFR part 11.

*"The power and flexibility to develop the optimum method and solve the toughest problems"*

### The challenge

Provide a powerful and flexible UV-visible spectroscopy system for the method developer with easy transfer of the developed methodologies to routine laboratory systems.

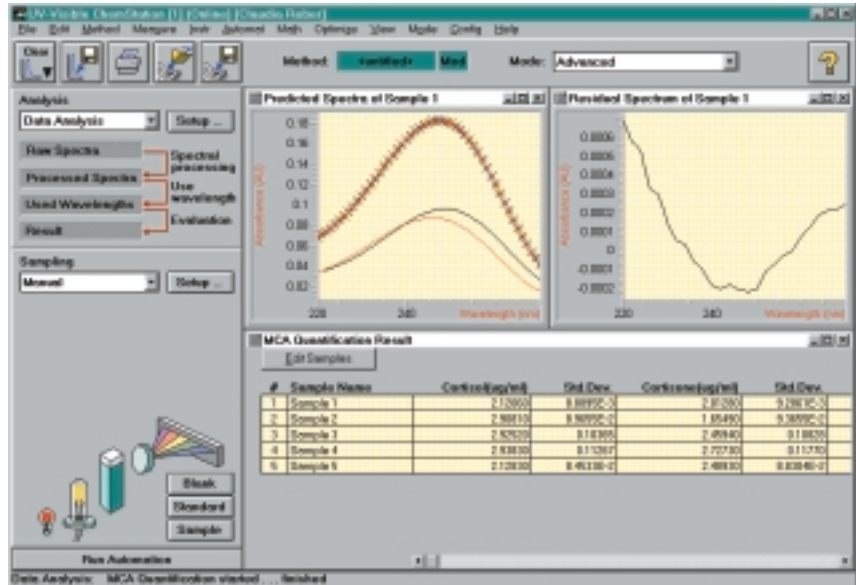
### The solution

Add the advanced software to the Agilent ChemStation system to provide special development tools and powerful analysis and automation capabilities.

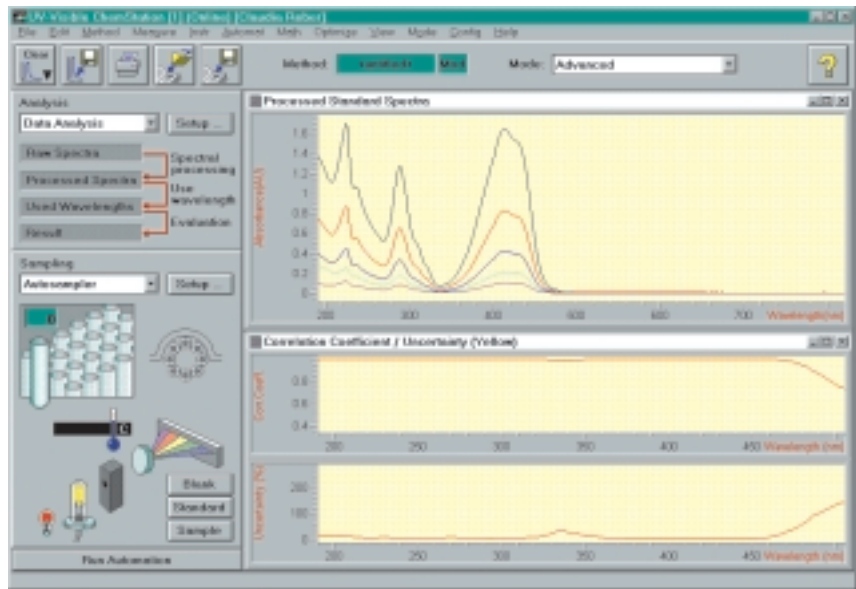
### Advanced automation

With the advanced automation capability you can perform fully automatically complex measurement, data evaluation, and report generation sequences using single or multiple methods for different types of samples. It can also be used to guide an operator through a complex sequence of manual measurement operations. Complex methods or automation processes that you develop on the advanced system can be transferred to and executed on the general purpose system.

Multi-component analysis with diagnostic tools that show the quality of the results



The Optimize Standards function shows the wavelengths that give the best calibration



# Solutions for biochemical analysis

## Ease of use

The biochemical analysis software builds on the Agilent 8453 UV-visible spectroscopy system. It provides the additional tasks that the biochemist needs and uses the same easy-to-use graphical user interface. Preprogrammed methods for protein and nucleic acid analysis help you get up and running fast.

## Proteins and nucleic acids

Standard methods are provided for the qualitative and quantitative analysis of nucleic acids and proteins:

- 260/280 nm ratios (with optional reference at 320 nm) for nucleic acid purity
- Warburg-Christian calculation of concentration of nucleic acid or protein in mixtures
- Biuret, Lowry, Modified Lowry, Bradford, Bicinchoic acid, and Trinitrobenzene sulfonate protein quantification methods

## Microsampling

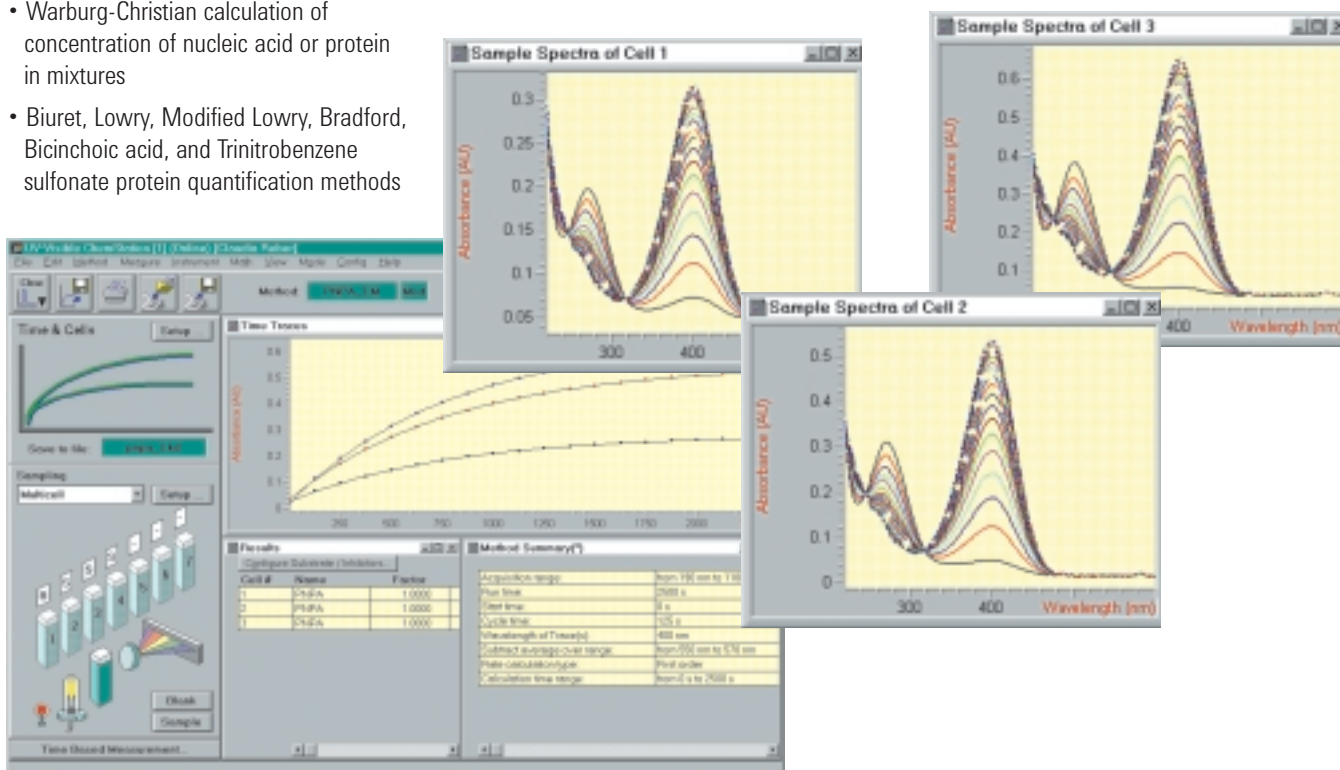
A frequent problem for the biochemist is the small sample volumes available for analysis. With the Agilent 8453 spectrophotometer you can use microcells requiring as little as 15  $\mu\text{L}$  of sample for qualitative and precise quantitative analysis.

## Multicell kinetics

For productivity with enzyme kinetic measurements the optional eight-position multicell transport can be used. You can freely configure which positions contain samples and blanks and you can select to subtract the rate of one cell from the others.

You can evaluate kinetic rates at a single wavelength or subtract a reference wavelength. Rate calculation methods include initial rate, zero order, first order, and delta absorbance. For a single cell you can also display and calculate rates at up to six individual wavelengths.

**Full spectra are always acquired, even in multicell kinetics, so you can re-evaluate results at leisure**



### The challenge

Provide an easy-to-use UV-visible system that meets the specific needs of the biochemist.

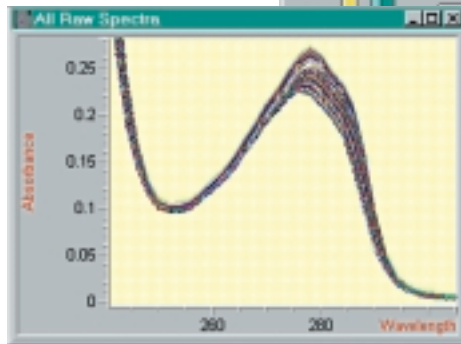
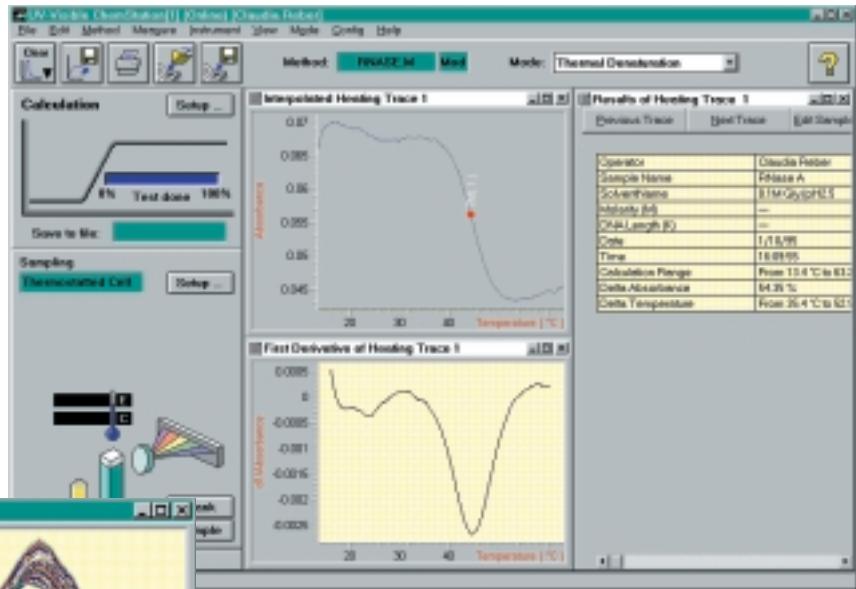
### The solution

Add biochemical analysis software to the Agilent 8453 UV-visible spectroscopy system for multicell kinetics, thermal denaturation, and protein/nucleic acid analysis capabilities.

## Thermal denaturation

With the Peltier temperature controller you can perform thermal denaturation experiments such as DNA melt testing and protein unfolding.

You can select your own multi-step heating and cooling program and, while the run is in progress, you are kept up to date with display of actual and target temperature, and the thermal denaturation trace at your specified wavelength. Results can be evaluated using average or derivative methods to calculate transition temperatures and standard or user-entered equations to calculate %GC.



Thermal denaturation results are calculated using your own equation

*"Key biochemical applications in a single, easy-to-use system"*

# Agilent's UV-visible spectroscopy solutions for chemical analysis – at a glance

## Selecting a controller

The 8453 can be operated through the handheld controller or the PC-based ChemStation. Both configurations provide the same quality of data. Therefore from a data quality point of view both configurations are equivalent.

Select the 8453E system with handheld controller if:

- bench space is critical
- standard data evaluation features are required
- sample throughput is low to medium
- budgets are tight

Select the 8453 system with the PC-based ChemStation if:

- flexibility and scalability is required
- powerful data evaluation features are required
- sample throughput can be high
- networking is required

## Upgrading a handheld-controlled system to a ChemStation system

Upgrading an 8453E system to a PC-controlled ChemStation system is easy:

1. Disconnect the handheld controller.
2. Connect the PC to the 8453 through GPIB or LAN (to connect through LAN your 8453 needs the optional interface board).
3. Start the ChemStation software and make your measurements.

## Selection of software modules

Adapt the 8453 spectroscopy system to your needs by adding different software modules to the general-purpose software. One or more modules can be added to at the same time to support different applications.

Depending on the task, select:

- general-purpose software, including verification and diagnostics, for standard applications
- advanced software for more capabilities in data evaluation, method development and automation
- biochemical analysis software, including kinetics and thermal denaturation
- dissolution testing software for single and multibath dissolution testing
- security pack software to support compliance with 21 CFR Part 11



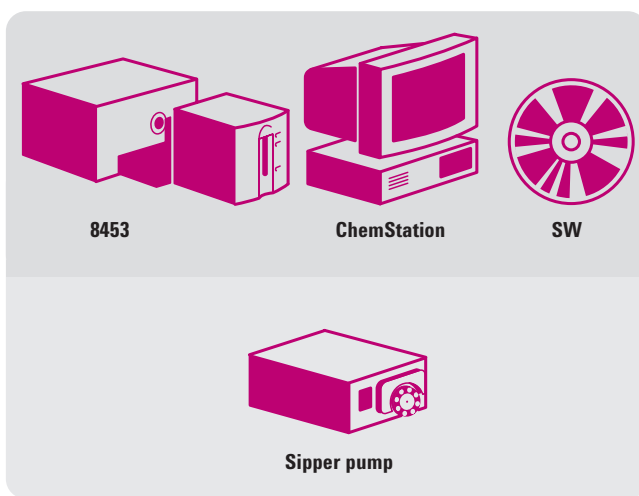
Software modules	<b>Advanced</b>	<b>Dissolution</b> Dissolution testing Multibath dissolution testing Combined report	<b>Biochemical Analysis</b> Kinetics Thermal denaturation
8453 spectroscopy system with standard software	<b>General Purpose</b> Verification and diagnostics		

### Increasing productivity

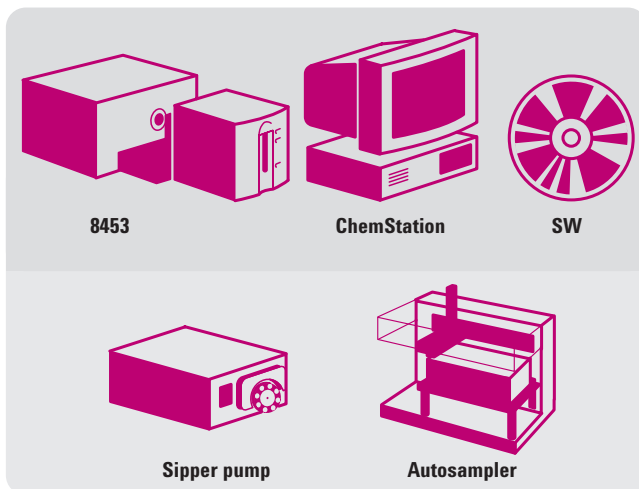
The ChemStation software allows control of different sampling systems to increase throughput. A common feature of the general purpose, advanced and dissolution software is that the predefined methods work with different sampling systems – without changing parameters.

Therefore, a method developed for manual sampling can easily be used with a sipper or autosampler, depending on the actual demand.

For convenient operation and medium sample throughput the sipper system is recommended. A sipper system consists of:



For unattended operation and high sample throughput, the autosampler system is recommended. It consists of:



# Accessories

## Cell holders

Agilent Technologies offers a range of single cell holders, providing you with the ability to analyze a wide variety of samples. Unless otherwise stated the holders accept cells with path lengths up to 10 mm.

**Standard cell holder:** Ensures quick and precise cell positioning for each measurement. Supplied standard with all Agilent Technologies spectrophotometers.



**Thermostatable cell holder:** When connected to a circulating-water bath, ensures constant sample temperature for temperature-sensitive analyses. An optional accessory provides water-driven stirring with magnetic stirring bar for 10-mm cells.

**Long-path cell holder:** Holds rectangular and cylindrical cells with path length up to 100 mm.

## Multicell transport



The multicell transport provides a significant increase in productivity when you need to do simple, repetitive measurements on a small number of samples, or when you want to follow changes in several samples (for example, enzyme kinetic studies).

The multicell transport features:

- full control through software,
- eight cell positions (for example, one blank and seven samples),
- random access to all positions,
- water thermostatable using external water bath, 5–90 °C temperature range and  $\pm 0.3$  degrees temperature difference between any two cells at 37 °C,
- <1 second to move between adjacent cells, and
- $\pm 0.1$  mm position repeatability.

## Peltier thermostatted cell holder

Use the Peltier controller and cell holder when you require precise or variable temperature control:

- built-in magnetic stirrer,
- heat exchanger for sample pre-heating when using a sipper system,



- fully controlled through software and GPIB interface,
- external sensor for precise sample temperature (optional),
- typical range of 10–100 °C (-10 to +80 degrees relative to ambient) without need for water cooling,
- accuracy of  $\pm 0.2$  degrees at 20–40 °C,  $\pm 0.3$  degrees at 0–20 °C and 40–60 °C,  $\pm 0.5$  degrees at 60–100 °C,
- reproducibility of  $\pm 0.1$  degrees at 0–60 °C,  $\pm 0.2$  degrees at >60 °C, and
- stirrer speed of 40–1000 rpm.

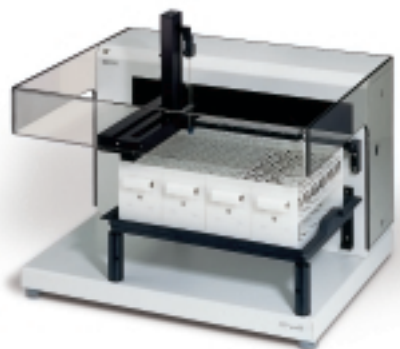
## Sipper system



For repetitive measurement of liquid samples a sipper system improves productivity and eliminates errors caused by manual cell handling. The Agilent sipper system comprises a peristaltic pump and a quartz flow cell:

- full control through software,
- variable pump, delay, and return time,
- flow cell with 10-mm path length, 3-mm diameter, and 80- $\mu$ L volume,
- constant-speed peristaltic pump with Tygon pump tubing,
- minimum sample volume of approximately 1 mL, and
- typical sampling time of 20 seconds (with <1% cross-contamination).

## Autosampler



Combine an XY autosampler with a sipper system and you can measure large numbers of samples fully automatically in unattended mode. The XY autosampler features:

- full control through software,
- capacity of up to 240 samples,
- wash station for optional rinsing between samples,
- 10–13-mm diameter sample tubes with maximum height of 100 mm,
- typical transit time (adjacent samples) of approximately 6 seconds (including raising and lowering probe),
- typical time per sample of 25 seconds (including sipper operation time), and
- minimum sample volume of 2 mL.

## Cells

Agilent Technologies offers a range of quartz cells for standard and flow-through operation including:

- Regular rectangular cells with path lengths of 1, 2, and 10 mm.
- Cylindrical cell with 100-mm path length.
- Stopped cells to protect your sample from air and ideal for use with the external sensor of the Peltier temperature controller.
- Flow cells with rectangular, oval or circular apertures with screw fittings and path lengths of 1, 2, 5 and 10 mm.
- Semi-micro and micro cells with path lengths of 2 and 10 mm for minimum sample volumes of 15 and 60  $\mu\text{L}$  respectively.



For more details see the Agilent Columns and Supplies Catalog.

[www.agilent.com/chem/supplies](http://www.agilent.com/chem/supplies)

## Third-party accessories

A wide range of accessories, designed for or compatible with Agilent spectrophotometers are available:

- **Custom Sensors & Technology** fiber optics coupler, fibers and sampling devices,
- **DBS** Peltier devices for single cell and multicell transport,
- **Gilson** autosamplers that are controlled directly from the Agilent ChemStation software,
- **Labsphere** diffuse reflectance accessories,
- **Applied Photophysics** stopped-flow accessory for fast kinetics,
- **Hi-Tech** stopped-flow accessory for fast kinetics,
- **Distek** dissolution baths, and
- **Caliper Lifesciences** MultiDose dissolution workstation.

Ask your Agilent representative for more details.

## Agilent support

Agilent has support centers in 85 countries supplying a wide range of support products that can be tailored to your needs.

For example, the Agilent network of Analytical Response Centers provides direct access to support professionals who help you resolve operational difficulties, and offer assistance and advice on running Agilent chemical analysis software. They also help you to solve analytical problems, to keep your equipment up to date, and to maintain GLP standards.

All of us at Agilent are here to help you meet your chemical analysis goals, not just now but also long into the future.

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