Solutions for Biomedical and Biological Applications

AGILENT FTIR SPECTROSCOPY, MICROSCOPY AND IMAGING SOLUTIONS

The Measure of Confidence

Agilent Technologies

identify confirm solve…explore

SEE MORE. SEE CLEARLY. SEE FASTER.

When you're considering how to approach your next analytical challenge, consider FTIR. From new drug product development to automating QA/QC SOPs, from the study of protein structure/folding to bioavailability studies, and from the investigation of cellular and sub-cellular components to the study of diseased tissues, you can do it all with FTIR.

World-Class Solutions for Biomedical and Biological Applications

Agilent FTIR systems can be adapted and customized to your specific experiments, enabling you to collect more sample information than you ever thought possible. Agilent has a complete suite of FTIR solutions to meet all your biospectroscopy needs. Our spectrometers, microscopes, and chemical imaging systems deliver:

- Thermally-controlled accessories for use in aqueous environments, simplifying the analysis of tissues and cells under native conditions.
- Straightforward kinetics software to study changes in biosystems such as protein folding, enzyme dynamics, and drug uptake.
- Highest signal-to-noise (S/N) performance — up to four times better than any available FTIR — providing the highest sensitivity and productivity and the ability to analyze samples that may not be possible with other FTIRs.
- Intelligent electronics for accessory and component recognition, providing seamless changeovers and automatic method optimization.
- Micrometer to meter measurements using Agilent FTIR large sample microscope objective, to analyze a wide range of samples.
- Multiple measurement modes including transmission, reflection, ATR and grazing angle.
- ATR micro- and macro-imaging, which extend traditional imaging measurements to new boundaries, reducing sample preparation and improving spatial resolution.
- Full upgrade path for spectrometers and microscopes for the ultimate in flexibility, to meet your changing application needs.
- Versatile, easy-to-use software, making FTIR spectroscopy, microscopy and imaging accessible to users of all levels.

Agilent FTIR spectrometers deliver speed, ease of use and the highest quality data for biomedical and biological researchers.

Molecular Spectroscopy Innovations

1947 First commercial recording UV-Vis, the Cary 11 UV-Vis	**1954** Release of the Cary 14 UV-Vis-NIR	**1969** First rapid-scanning fourier transform infrared spectrometer, the FTS-14	**1971** First use of a mercury cadmium telluride (MCT) detector in an FTIR	**1982** First FTIR microscope, the UMA 100	**1989** Release of the acclaimed Cary 1 and 3 UV-Vis	**1991** First infinity corrected infrared microscope
1997 Cary 50 Series released to coincide with 50th anniversary of Cary 11	**1995** Launch of the 8453A, the first small-footprint, full-featured diode-array	**1999** Launch of the Cary Eclipse Fluorescence Series	**2000** First ATR chemical imaging system	**2002** Cary 4000/5000/ 6000i research grade UV-Vis-NIR series released	**2008** Launch of the 600 Series FTIR spectrometers, microscopes and imaging systems	**2011** Agilent offers out-of-lab FTIR solutions

FOR YOUR APPLICATION

Agilent is committed to providing solutions for your application. We have the technology, platforms, and expert guidance you need to be successful.

Identify Biological Samples and Optimize Bioprocesses

- Obtain detailed chemical and structural information about biological samples in seconds without using tedious wet chemistry methods.
- Perform QA/QC of raw materials, intermediates and finished goods.
- Simplify method development for infrared analyses.
- Characterize materials and synthesis products.
- Study the structure, architecture and function of biofilms.
- Monitor and optimize microbial fermentation processes to increase production efficiency.

Study reaction dynamics in real-time

- Monitor drug-synthesis reactions or study the stability of pharmaceutical formulations across a range of drug concentrations.
- Probe fluid dynamics in real-time using microfluidic devices.
- Characterize cell lipids and membranes or study drug uptake through membranes using Attenuated Total Reflectance (ATR) chemical imaging.
- Study the structural conformations of proteins and enzymes under variable conditions.
- Characterize induced structural changes on a microsecond or nanosecond time scale using innovative step-scan instrumentation.

Investigate and characterize biological samples with a high spatial resolution over large areas of analysis

- Quickly visualize chemical heterogeneity without selective-compound characterization and cell fractioning – simply select FTIR spectral peaks of interest.
- Develop rapid detection techniques for the examination of human diseases such as cancer and other pathologies.
- Study the fine chemical structure of cellular and sub-cellular components such as phospholipids, proteins, nucleic acids and carbohydrates in plant and animal tissues.
- Rapidly differentiate and identify microorganisms such as bacteria and yeasts.
- Investigate the chemical makeup of live cells in-situ on a microscopic scale and probe their response in aqueous environments.

identify

IDENTIFY BIOLOGICAL SAMPLES AND OPTIMIZE BIOPROCESSES

Quickly characterize raw materials and synthesized products

Obtain detailed chemical and structural information about your biological samples in seconds. Unlike time-consuming wet chemistry methods, with an Agilent FTIR you can simultaneously obtain quantitative and qualitative data from virtually any type of biological material. Use our dedicated ATR accessories to study the structure and function of biofilms in a non-destructive manner or investigate how monolayers of biomolecules bind to the surface of a substrate using Polarization Modulation-Infrared Reflection Absorption Spectroscopy (PM-IRRAS).

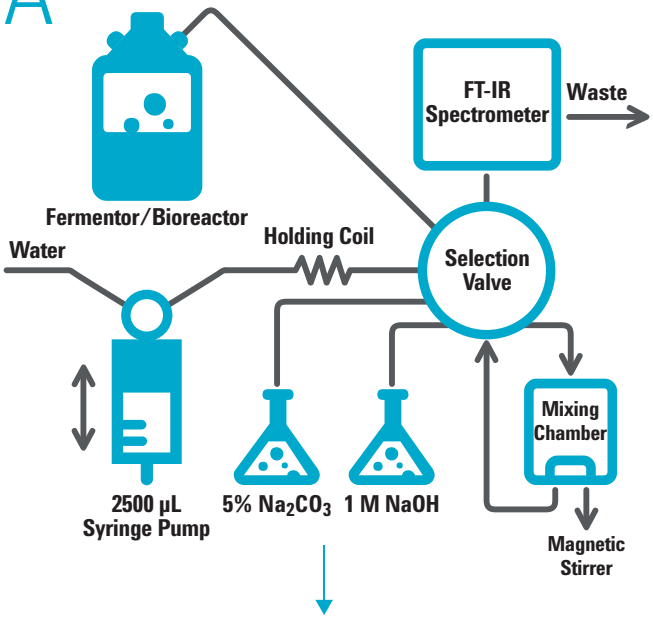
Automate your lab-based instrument for bioprocesses that provide rich chemical information by searching in-house spectral databases or the largest FTIR libraries in the world.

The example here illustrates the simplicity of using Agilent FTIR spectrometers for optimizing solvent production during a microbial fermentation process.

The Agilent 600 Series FTIR delivers:

- Thermally-controlled accessories for the study of biological processes under user-defined environmental conditions.
- Superior data quality and sensitivity to detect and identify even the smallest sample concentrations faster.
- Easily searchable in-house spectral libraries of proprietary data and commercially-available libraries to identify and verify your sample with a single mouse click.

A



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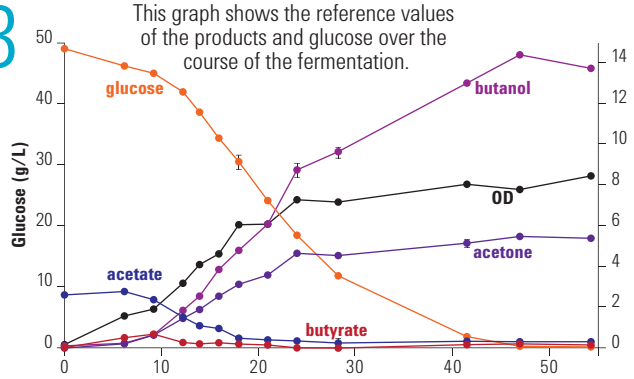

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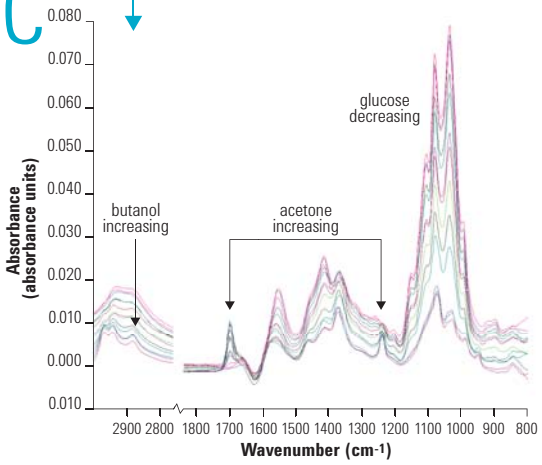

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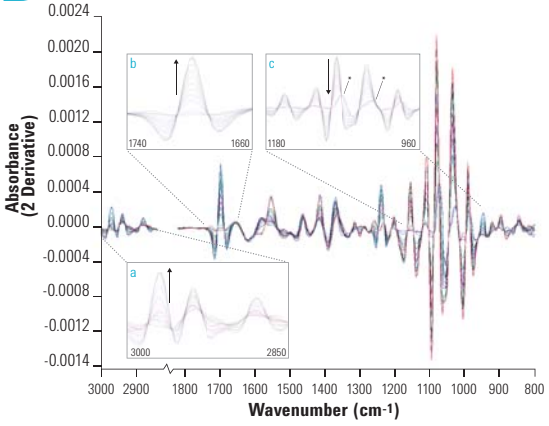


Background

Typical commercial fermentation processes use microorganisms to convert chemical substrates to products of higher value. Bioprocess products are of vital importance to modern society, ranging from traditional products such as beer, wine and vinegar, to fine chemicals such as amino acids, and antibiotics and other pharmaceuticals.

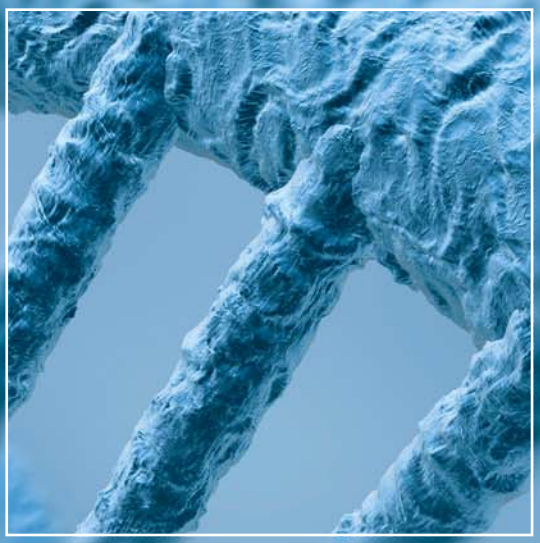
The Challenge

Develop a method to actively monitor solvent production during a 55 h Clostridium beijerinckii acetone-butanol fermentation (shown in A) to increase the efficiency of the bioprocess. Monitor the concentration of key chemical components such as glucose, acetone, acetic acid, butyric acid and butanol (shown in B), and physiochemical parameters such as temperature, turbidity and pH value, to ensure that minimal waste is generated during solvent production.

The Solution

With Agilent's software, simplify complex bioprocesses by automating the on-line monitoring using an in-bench ATR with a 660 FTIR spectrometer (results shown in C and D). This rapid method allowed the concentrations of acetone, acetate, n-butanol, butyrate and glucose to be determined simultaneously at the level of accuracy and precision required for process monitoring. Using Agilent's systems, it is possible to optimize fermentation and industrial processes that require the rapid measurement of one or more analytes in-situ.

confirm



STUDY REACTION DYNAMICS IN REAL-TIME

Perform dynamic studies and kinetics experiments that other systems cannot

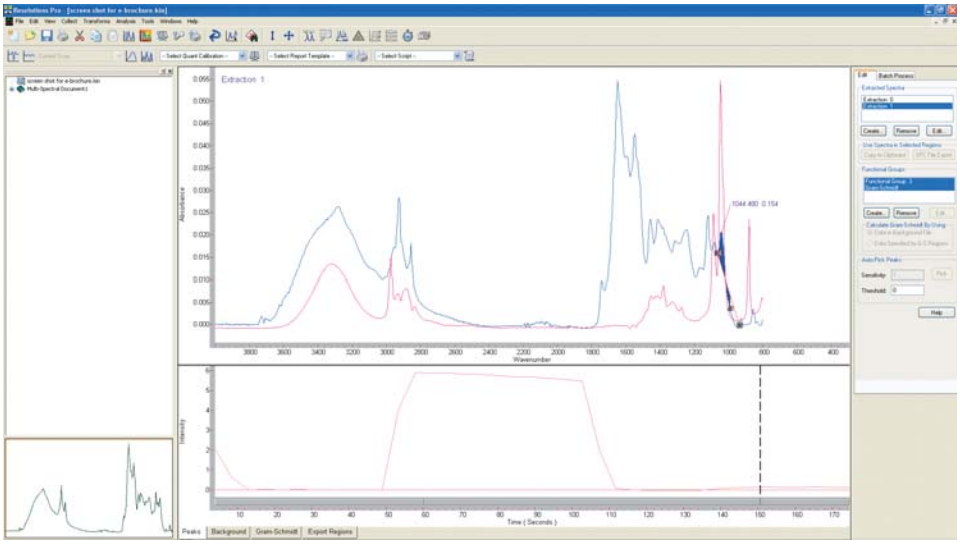
Characterize dynamic biological processes such as protein folding or cell membrane transitions and quickly monitor conformational changes that are involved in functional modifications using Agilent's unique combination of hardware and software.

If you need to mimic real-world sample conditions by controlling the temperature, pH, and atmospheric environment of your sample during analysis, you can with our unique accessories. With flexible FTIR solutions that implement both rapid scan and step scan approaches, Agilent's systems allow you to investigate the widest range of dynamic processes — whether they occur within hours to seconds, or on the micro- or nano-second time scale – making them ideal for the analysis of stimulus-induced samples.

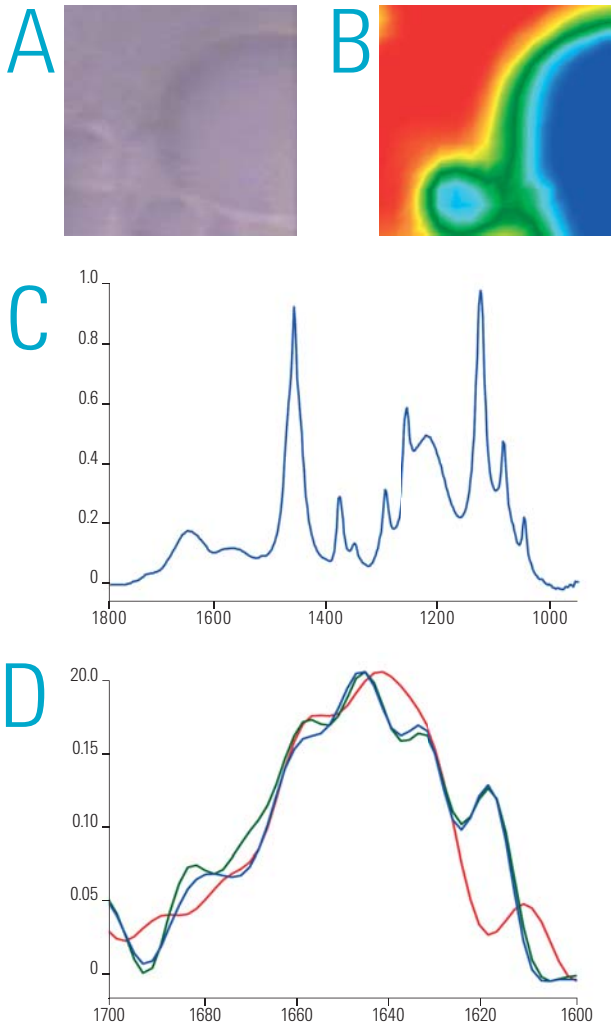
In these examples, our easy-to-use Resolutions Pro software and unique hardware give you the edge by allowing you to see what others are missing.

Agilent's complete solution delivers:

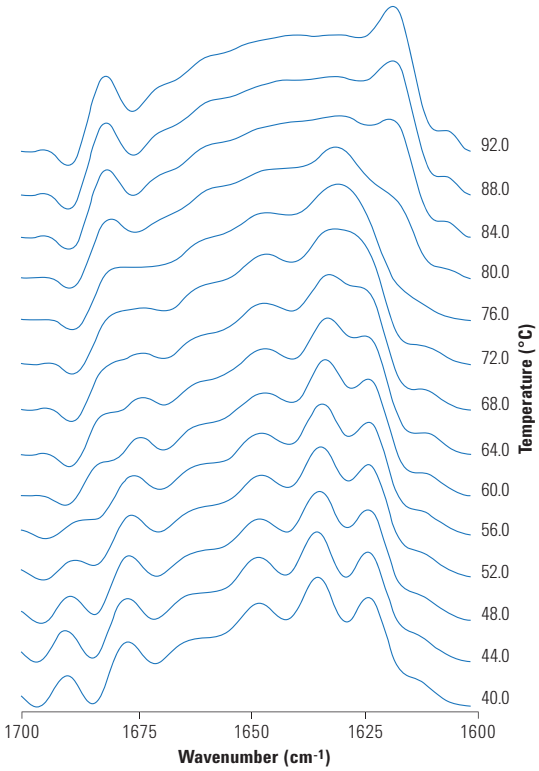
- An unsurpassed ability to probe fluid dynamics in biological systems using microfluidic devices
- The fastest kinetics speeds to investigate changes that occur within a fraction of a second as well as slow kinetics to study reaction dynamics over hours or days
- The unique capability to investigate processes such as molecular diffusion and drug uptake in real time via time-based imaging kinetics using a focal plane array detector.



Left: Study the physicochemical properties of proteins and enzymes in real-time using intuitive kinetics software. In this example, the thermal properties of β-lactoglobulin A (5% w/v) in deuterated phosphate buffer at pH 8.6 were investigated, by monitoring changes in the secondary structure. Shown above is the stacked plot of deconvolved infrared spectra during heating from 40 to 92 °C. This information provides a useful model for studying the relationship between protein structure and function, in order to improve manufacturing processes.



Left: Study enzyme catalysis and mixing reactions between fluids in small-volume environments in real-time, enabling you to better understand the reaction intermediates and products. Lipoxygenase is an important class of enzymes used in industrial applications. Chemical imaging was used to study lipoxygenase conformations during reactions in organic solvent media (visual and chemical images are shown in A and B respectively). FPA-FTIR imaging allowed for the simultaneous investigation of the substrate, enzyme, and product partitioning during the reaction. A representative spectrum was selected from the reaction interface (shown in C) and the secondary structure of lipoxygenase conformations were compared in the presence of different solvents (shown in D).



Above: Agilent provides dedicated software to monitor reaction dynamics and kinetics experiments such as protein folding or cell membrane transition studies in real time. Create functional group plots with a single click to extract the data that you want.

solve

INVESTIGATE AND CHARACTERIZE BIOLOGICAL SAMPLES

Distinguish biological samples with a high spatial resolution over large analysis areas

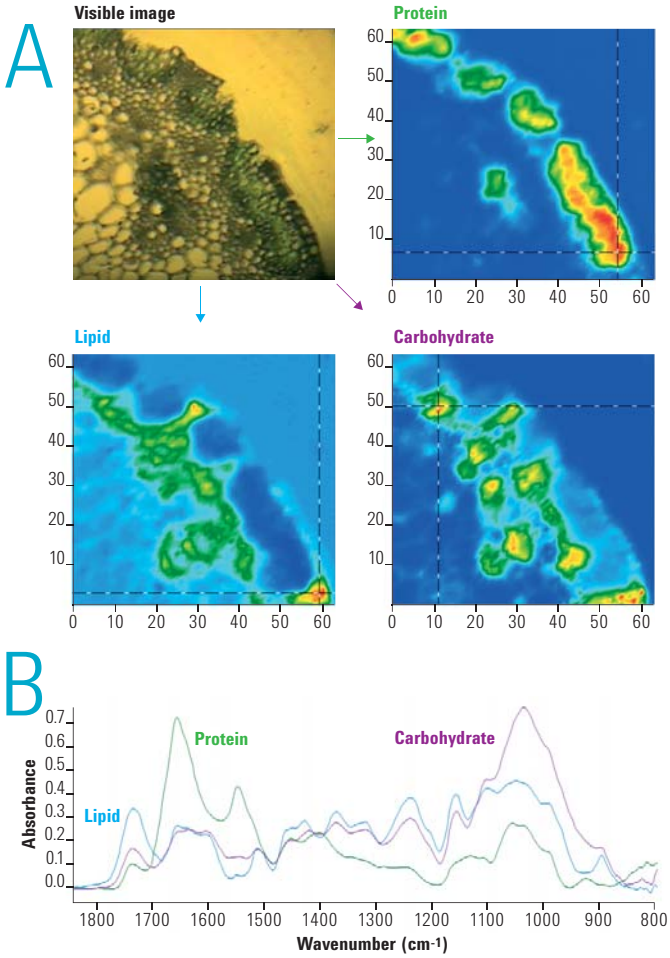
With Agilent FTIR systems, see data that other systems cannot. Using an Agilent microscope, featuring a wide range of spatial resolution options, combined with quality data produced by an Agilent spectrometer, characterize the spatial distribution of components within heterogeneous materials and identify the specific nature of a sample.

Perform both microscopic and macroscopic measurements using the multiple measurement modes of Agilent's infrared microscopes, including transmission, reflection, attenuated total reflectance (ATR), grazing angle reflection analysis and 'large sample' mode. With infrared mapping and chemical imaging using a Focal Plane Array (FPA) detector, Agilent's systems give you superior quality information — even from the most challenging of samples — in the shortest possible time.

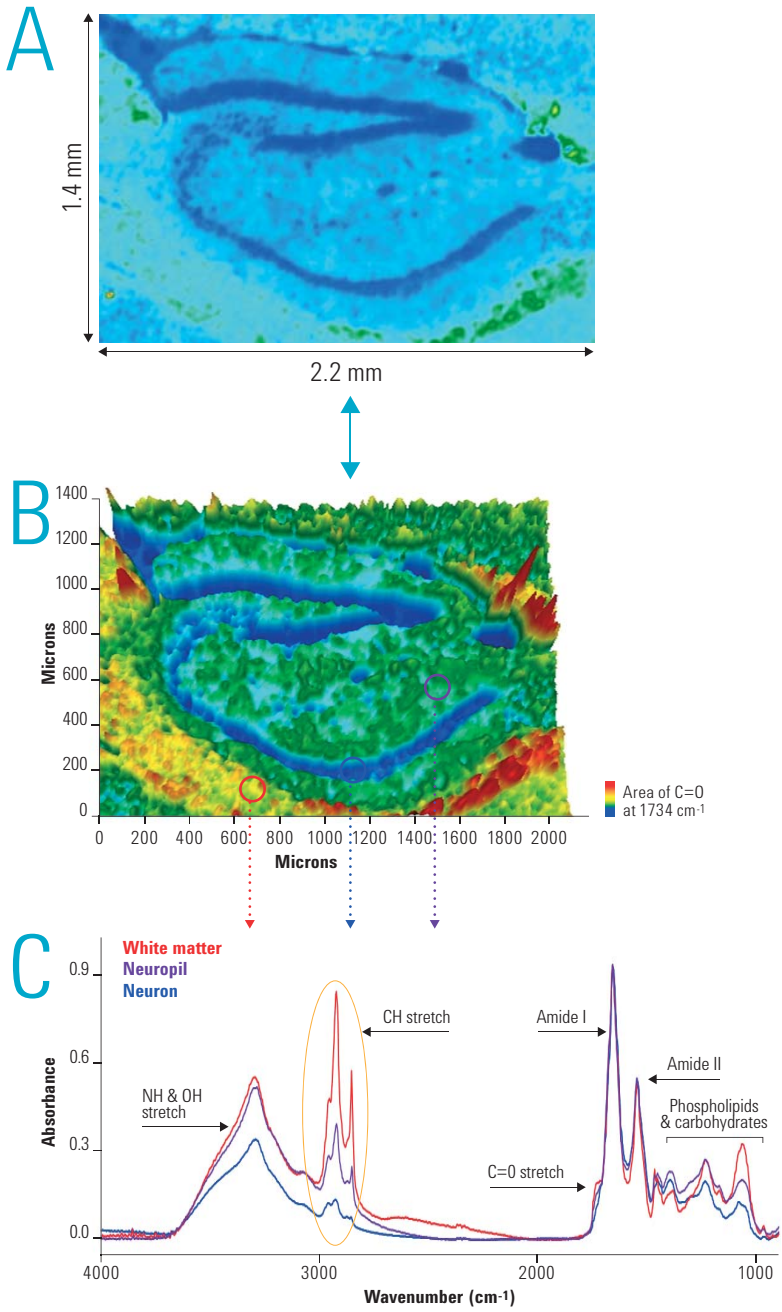
In the following examples, tissue heterogeneity across large areas was investigated within minutes, providing a comprehensive understanding of sample chemistry on the micron scale.

The Agilent 600 Series FTIR delivers:

- Dedicated hardware that is ideal for both simple and advanced biological sample characterization.
- The best sensitivity to detect the smallest sample features quickly.
- Simple to use, versatile microscopes that can be customized to suit any required area of analysis.



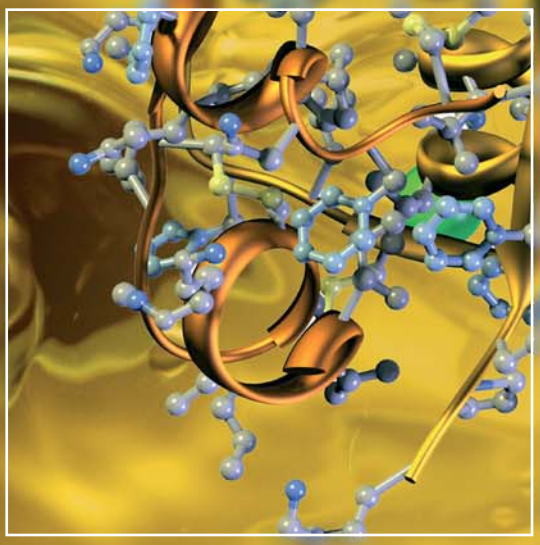
Plant tissue analysis using chemical imaging easily reveals the distribution of cellular and sub-cellular components. Unlike traditional analysis methods that rely on selective-compound characterization and cell fractioning, chemical heterogeneity can be quickly visualized by selecting FTIR spectral peaks of interest – in this instance the distribution of proteins (peak at ~1650 cm^{-1}), lipids (peak at ~1730 cm^{-1}) and carbohydrates (peak at ~1050 cm^{-1}) can be easily displayed, as shown in A. Representative spectra from the infrared image in B reveal differences in the chemical composition at selected location across the cross-section.



Infrared imaging analysis of a mouse hippocampus used to study Alzheimer's disease. Approximately 100,000 spectra were acquired at high spatial resolution (5.5 um) from the 1.4 mm x 2.2 mm tissue section. This technology enables scientists to rapidly study the chemical composition and development of plaques – the main pathological feature of Alzheimer's disease.

2-D and 3-D chemical images are shown in A and B respectively, while representative spectra from the white matter, neuropil and neuron are shown in C. The highlighted changes in CH2 & CH3 peaks illustrate differences in lipid content between white matter and grey matter.

solve



INVESTIGATE AND CHARACTERIZE BIOLOGICAL SAMPLES

Investigate cellular chemistry without the use of reagents

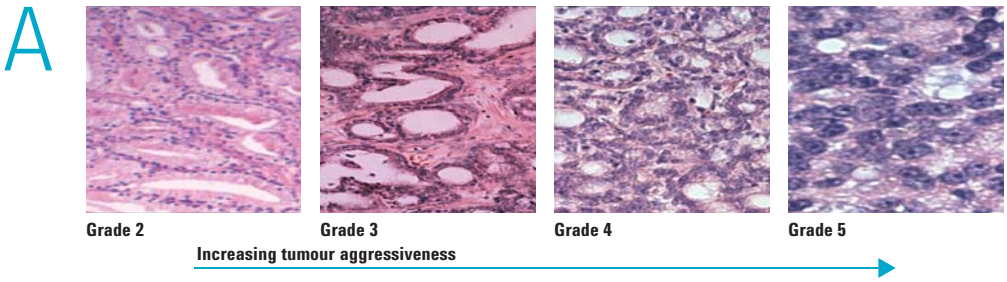
With Agilent's unrivalled ability to quickly analyze large tissue sections at high spatial resolution, you can study the fine chemical structure of cellular and sub-cellular components such as phospholipids, proteins, nucleic acids and carbohydrates in plant and animal tissues. You can also resolve morphological features in cells and tissues to differentiate between them according to their macromolecular chemistry.

Interested in investigating diseased states in animal and human tissue? Analyze biopsied tissues in order to understand the mechanism of pathologies and characterize subtle chemical differences. Use Agilent's FTIR imaging microscopy systems to distinguish between cancerous and non-cancerous tissue, develop novel approaches to assess disease grade and stage, or study the mechanisms of how cells move through tissues.

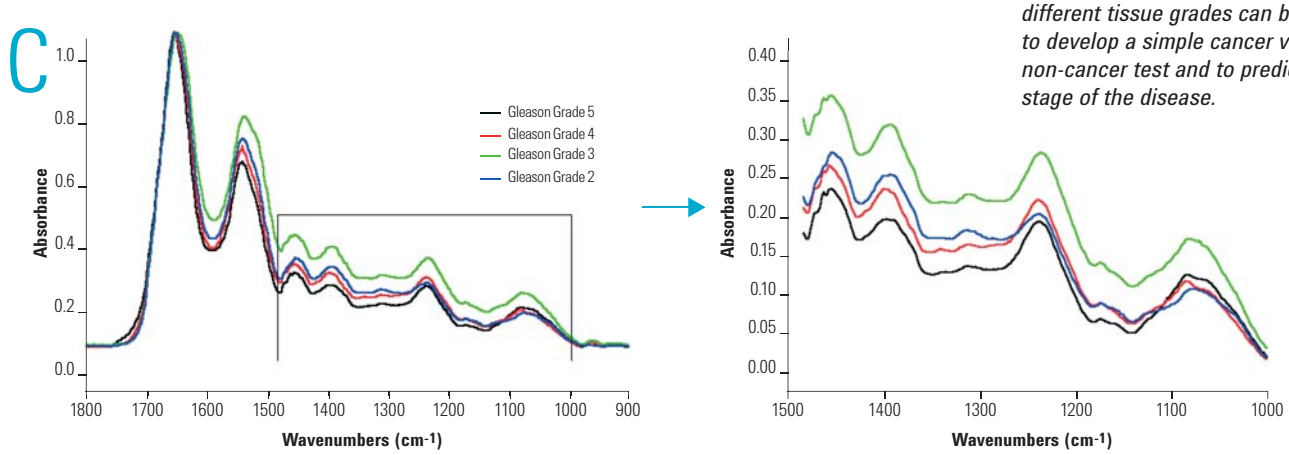
In these examples, Agilent's systems were used to study tissue heterogeneity at a high spatial resolution across several millimetres rapidly. This unique technology reveals structural features that traditional techniques and other systems overlook, and allows for unmatched experimental control and performance.

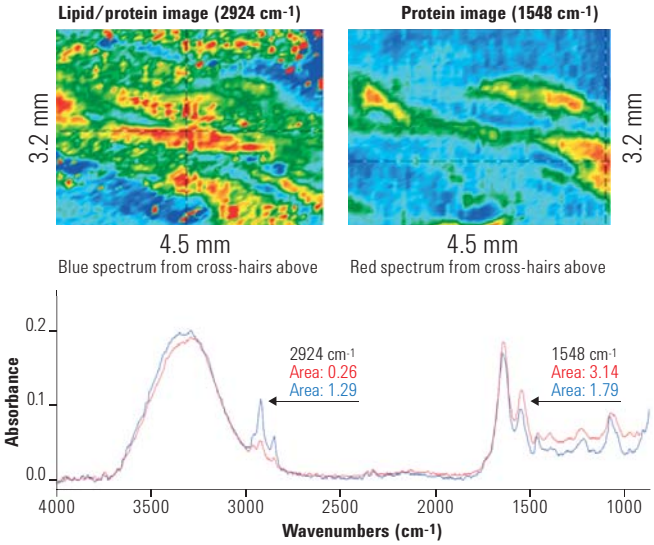
For these applications, the Agilent 600 Series FTIR delivers:

- Unsurpassed spectral collection of hundreds to thousands of spectra within seconds to characterize any sample, providing you with more information from a single collection
- Intelligent imaging with the most comprehensive spatial resolution modes of 1.1, 5.5, 11, and >22 µm for information rich detection of even the smallest sample features
- The ability to develop simple and rapid screening methods for the analysis of biopsied tissues and the study of human pathologies.

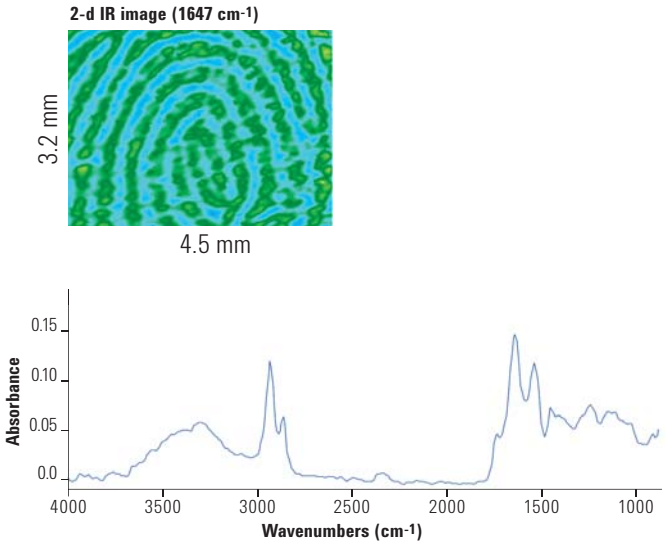


Research new methods for the rapid detection of cancer by developing spectral databases of cancer tissue samples using FTIR imaging. (A) Using optical microscopes representative tissue sections of prostate tissue can be graded according to the Gleason score based on tissue architecture. (B) Infrared imaging microscopes allow for the rapid screening of large tissue samples. An area of 7 mm x 7 mm (representing 100 tiled images) was recorded with four pixel aggregation at 8 cm^{-1} spectral resolution by co-adding 16 scans with 11 x 11 micron spatial resolution. The entire image took just 40 minutes to collect. (C) Representative spectra from different tissue grades can be used to develop a simple cancer vs non-cancer test and to predict the stage of the disease.



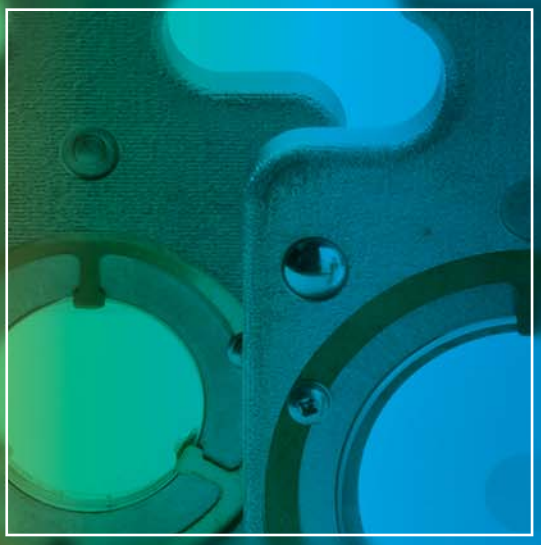


Use Agilent's ATR Focal Plane Array imaging system to investigate large tissue samples quickly, with little sample preparation. This lamb brain cross section was measured without the need for tissue microtoming. Representative chemical images show the distribution of different components, while the spectra below highlight some of the chemical differences seen in the tissue.



Unique accessories allow thin biological films such as this fingerprint residue to be investigated with ease. The chemical image was created based on the amide I peak centered at 1640 cm^{-1}. Several thousand spectra were collected in seconds, and a representative spectrum from the residue is shown below.

explore

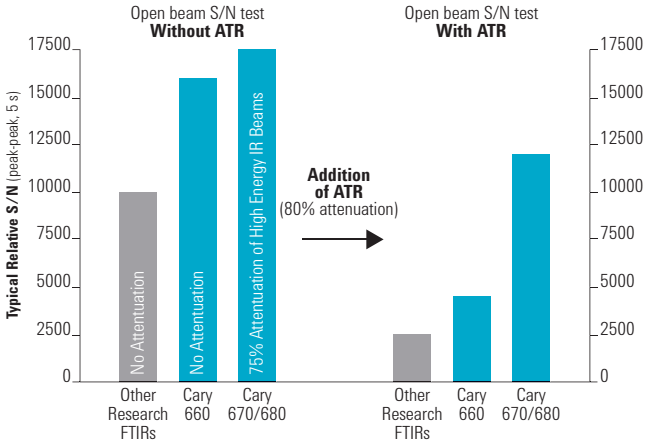


THE WORLD'S BEST FTIR

Every component of the Cary 600 Series is engineered for performance and usability, ensuring you get the right answer every time.

Real results. Real conditions.

The Agilent Cary 600 Series FTIR provides enhanced source throughput, beamsplitter and detector efficiencies and reduced instrument noise effects. The result is superior performance and sensitivity, up to four times better than any other research FTIR.



NOTE: Measured at 4cm^{-1} resolution using a DLaTGS detector.
ATR measurements performed using a diamond single bounce ATR.

The S/N difference

Traditional S/N tests
Performed without a sample or sampling accessory in the instrument, so they are effectively measuring air.

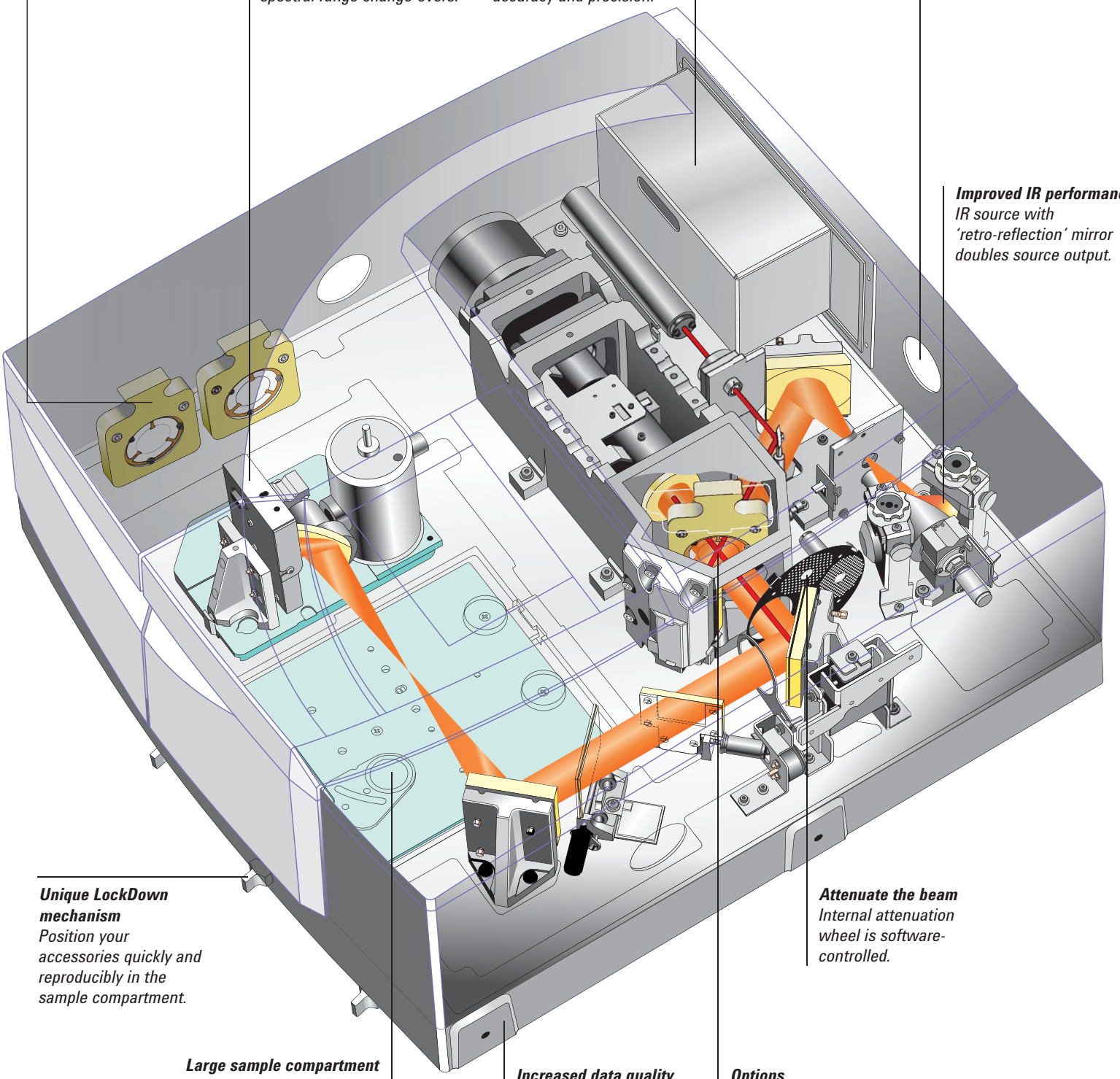
Agilent S/N tests
Measured under real-world conditions, giving you a true indication of performance.

Internal beamsplitter storage
Provides a dry, protected storage location.

Increased productivity
Easy-to-change detectors and beamsplitters enable quick and reproducible spectral range change-overs.

True digital 24-bit dynamic range Delta-Sigma A/D converter delivers maximum S/N, sensitivity, spectral accuracy and precision.

Experimental flexibility
With multiple external/ emission ports.



Improved IR performance
IR source with 'retro-reflection' mirror doubles source output.

Unique LockDown mechanism
Position your accessories quickly and reproducibly in the sample compartment.

Attenuate the beam
Internal attenuation wheel is software-controlled.

Large sample compartment
Includes removable floor plate for maximum flexibility when mounting samples.

Increased data quality and reproducibility
The sealed and desiccated enclosure, or full purge with purge shutter options, minimize environmental disturbances.

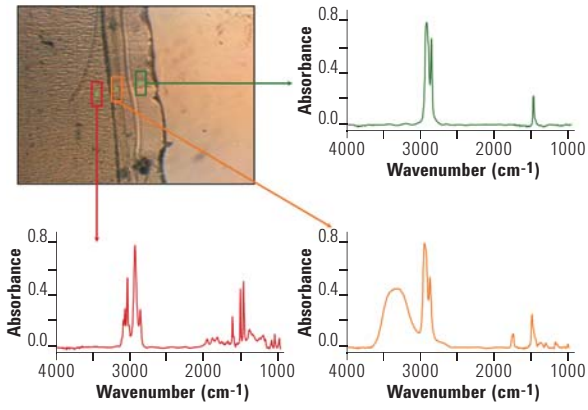
Options
57 mm dynamically aligned 60° air-bearing interferometer maximizes throughput and S/N performance (Cary 670/680 only).

Step-scan extends the capabilities for probing samples (advanced PAS, polymer stretching, and TRS), resulting in comprehensive sample characterization.

SPEED AND SENSITIVITY BY DESIGN

See more than ever – fast

The Cary 610/620 FTIR microscopes provides superior quality information in the shortest time, even with challenging samples. The microscopes offer the highest available optical throughput for the best S/N performance. The control panel enables all common software actions to be performed at the microscope, and aperture changes are quick and simple.



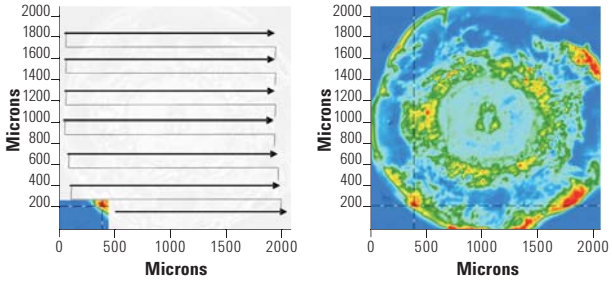
Set the Cary 610 microscope aperture to your sample size to obtain specific, exceptional quality spectra within seconds. Shown is a three-layer polymer laminate visible image (top left) and the resulting spectra, allowing for comprehensive characterization.

True chemical imaging

The Cary 620 provides the most sensitive and fastest available chemical imaging. The FPA[1] detectors enable simultaneous collection of up to 16,384 spectra within seconds. With a range of detector options (16x16, 32x32, 64x64, 128x128) and spatial resolution modes of 1.1, 5.5, 11 and >22 µm, you can characterize any sample.

Large sample analysis

Extend chemical imaging beyond the microscope with macro imaging, using Agilent's Large Sample (LS) accessory. With a field of view of up to 5x5 mm, you get more information from a single collection. Combine this with our range of macro ATR solutions for even simpler sampling.

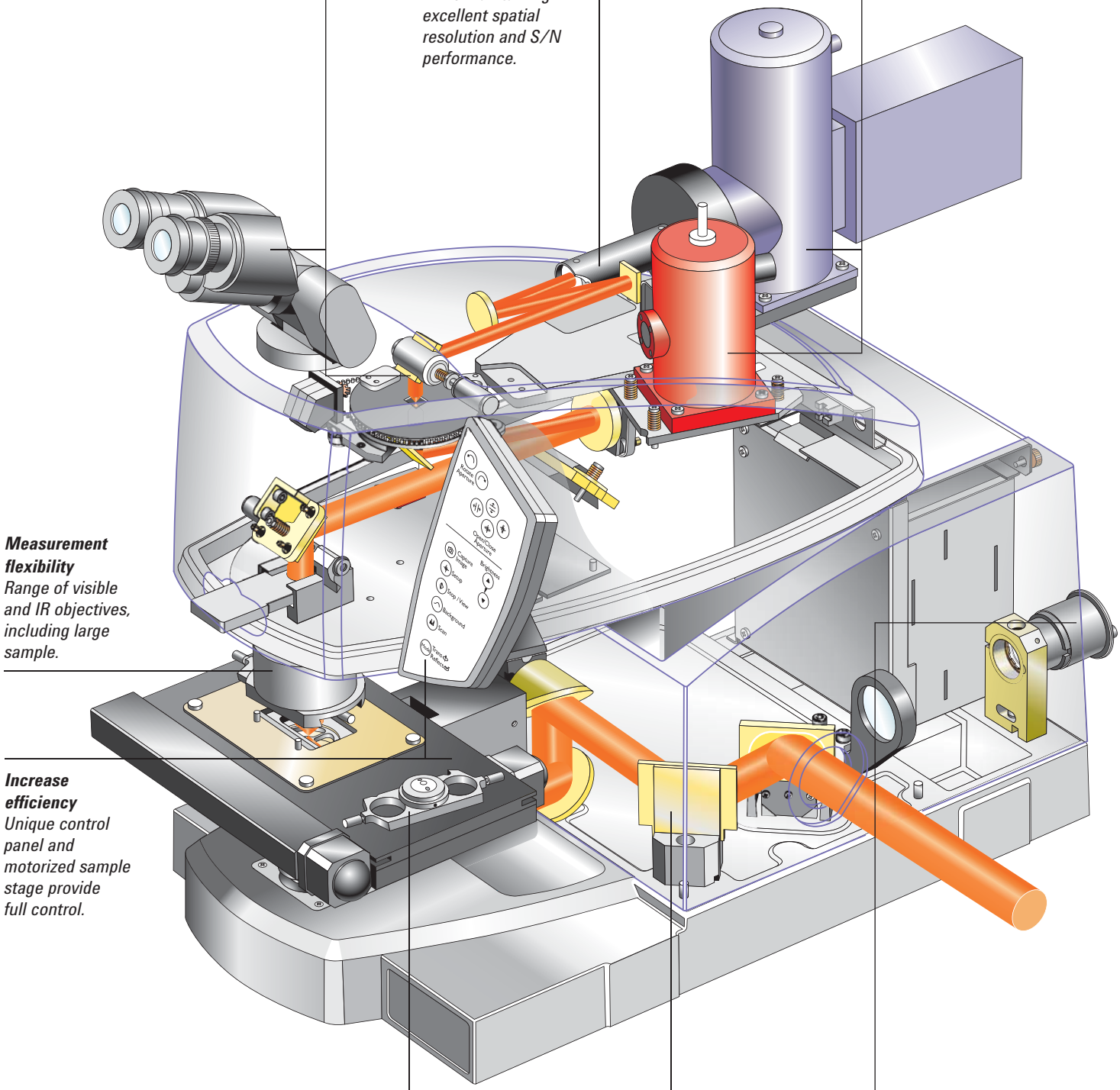


Get the full picture — fast. Left: Linear array mapping. In 20 minutes, only 5% of this large, high spatial resolution image is collected. Right: Agilent Cary 610 chemical imaging. In 20 minutes, 100% of the image is collected.

Simplify sample analysis
Binoculars and internal video CCD camera make sample analysis more flexible.

Quadruple the area of analysis
With field-expanding optics you can analyze larger samples in significantly less time, while maintaining excellent spatial resolution and S/N performance.

Sample flexibility
A range of detectors, from single element to FPA[1] imaging (Cary 620).



Measurement flexibility
Range of visible and IR objectives, including large sample.

Increase efficiency
Unique control panel and motorized sample stage provide full control.

Multiple modes
Transmission, reflection, ATR or grazing angle analysis mode.

Information-rich results
Advanced optical design provides maximum IR throughput and better S/N performance.

Fast setup
Ultra-bright white LED visual illumination ensures the highest quality sample visualization, even with difficult to see samples.

POWERFUL, INTUITIVE SOFTWARE

Whether you are performing routine measurements or cutting edge research, with Resolutions Pro software you will be able to acquire, process, analyze and manage your FTIR data quickly and easily.

Intuitive

- Use 'Method Editor' to easily set up a method and start a measurement from one window.
- Spend less time on setup — the accessory and component recognition detects instrument configurations and automatically optimizes the method.
- Customize — use the built-in scripting tool to simplify analytical tasks for the multi-user laboratory, or to develop advanced routines for challenging applications.

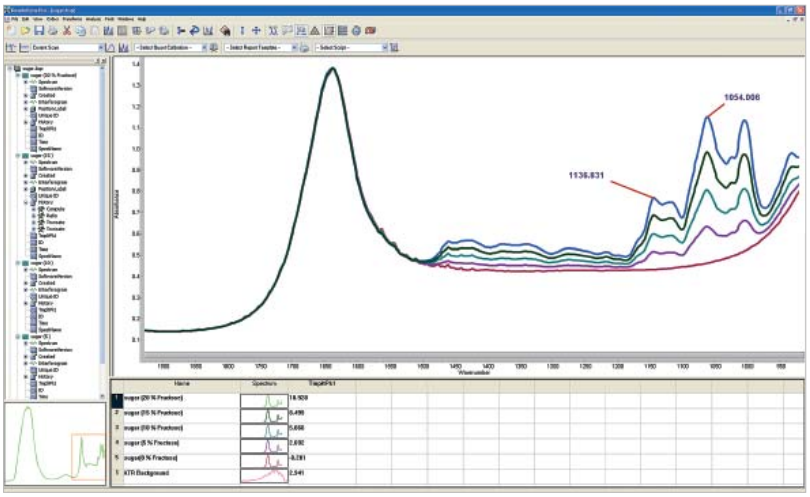
Data security and integrity

- 'User Manager' enables administrators to set user privileges, providing protection of data and methods from change or deletion.
- Access to ALL original data — including sample and background interferograms and post-collection — ensures data integrity, and allows for data reprocessing.
- Built-in instrument performance tests provide proof of performance and confidence in your results.

Intelligent imaging

For chemical imaging experiments, Resolutions Pro provides:

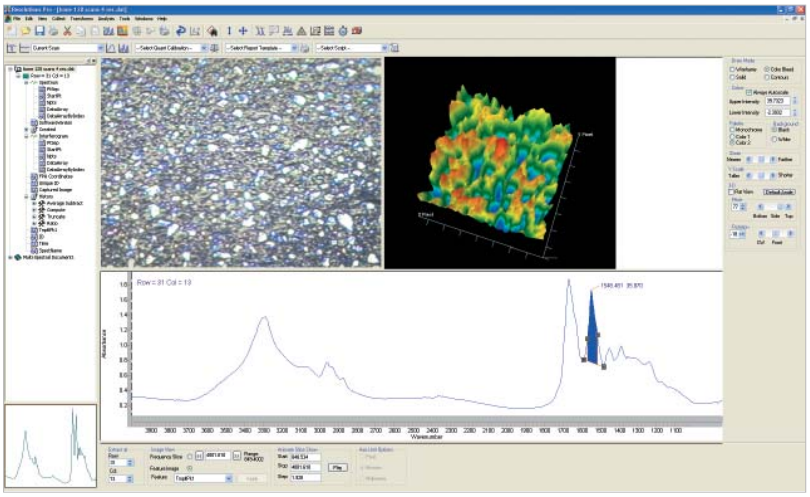
- Unsurpassed spectral collection of hundreds to thousands of spectra.
- Mosaic option to extend the field of view for unlimited image size.
- Individual spectra corresponding to a selected part of the image and conversely, image region corresponding to a selected wavenumber. This is useful as a quick check of a sample's heterogeneity.
- Control of chemical imaging detector integration time so you can maximize dynamic range and S/N performance to increase the quality of data for difficult to analyze samples.
- 2-D and 3-D views, which simplify the interpretation of spatially-resolved components.

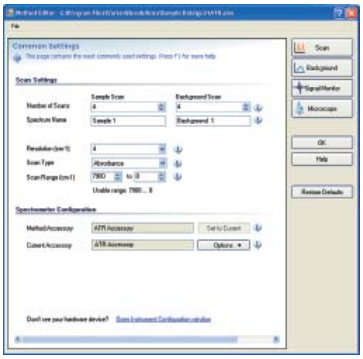


Left: Unique 'Spectral Spreadsheet' view allows multiple spectra to be overlaid, compared, and their parameters simultaneously tabulated with ease and speed.

Below left: Powerful Resolutions Pro software has multiple views including image, 3-D chemical image and spectrum,for comprehensive confirmation.

Below: 'Method Editor' enables users of all levels to easily set up a method.





Confirm what you see

For single point and mapping experiments, Resolutions Pro provides:

- Fully automated mapping for consecutive, unattended analysis of large sample areas or multiple samples.
- Grid mapping templates customized to your sample to create chemical contour maps for speedy analysis.
- Ability to create application-specific methods to simplify routine experiments.

Advanced data analysis

Resolutions Pro features sophisticated post-run analysis capabilities.

- Easily relate spectral information to a corresponding image.
- 'Play', 'Extract' and 'Image Peak' functions to easily explore imaging results.
- Full access to all collection and processing parameters for simple reprocessing of spectra and chemical images.

flexible

MEET ALL YOUR ANALYSIS CHALLENGES

Agilent has a range of complementary UV-Vis-NIR and Fluorescence Solutions for Polymers and Materials applications.

The Agilent Cary routine QA/QC to research-grade Cary UV-Vis-NIR and fluorescence spectrophotometers combine leading edge technology with flexibility and ease of use. Offering performance and versatility, the Agilent Cary 60, 100, 300, 4000, 5000, 6000i and Cary Eclipse spectrophotometers are equipped to handle the most demanding biomedical and biological and biomedical applications:

- DNA & protein melts
- GFP expression
- Intracellular ion measurements assays
- DNA/Protein quantitation investigations and purification using 260/280 ratios
- Protein folding studies using FRET/TR-FRET
- Enzyme kinetics
- Cytochrome P450
- Bioluminescence
- Drug binding using Fluorescence Polarization (FP)

Flexible and functional

Spend more time on analysis and less on set-up with Agilent's comprehensive suite of LockDown accessories. The unique Lockdown mechanism guarantees fast, reproducible accessory changeover. In addition, Agilent's application-focused software means you can obtain results at the push of a button. Whether you're a new or expert user, you'll be collecting data within minutes.

Move your analysis beyond the traditional cuvette. Agilent offers Fiber Optics solutions for both fluorescence and absorbance assays, so you can bring the instrument to your sample.

Application

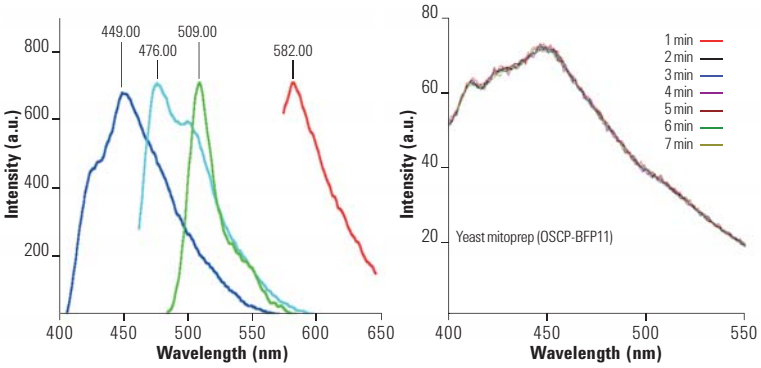
Measuring photosensitive samples without photobleaching

Solution

Agilent Cary Eclipse and Cary 50

Providing sensitivity without photobleaching

Multiple emission spectra of the photosensitive Blue Fluorescence Protein (BFP) collected at very slow scan speeds every 1 minute, exciting in the UV at 370 nm. The intense, narrow pulse of the Xe flash lamp delivers enough energy to measure minute concentrations of sample without causing photobleaching.



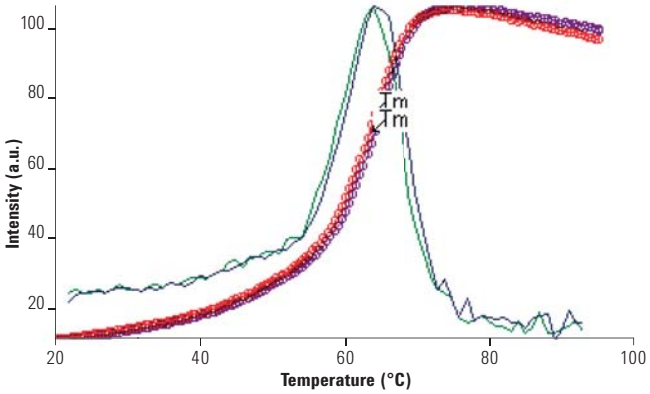
Application

DNA or Protein thermal melts

Solution

BioMelt UV or Fluorescence package

Ensure the Tm value of your sample is accurate by measuring the actual temperature of the solution within the cuvette, using Agilent's temperature probes.
Use the Thermal application to calculate Tm values and ΔG, ΔH and ΔS thermodynamic data within seconds of completing each experiment.



Application

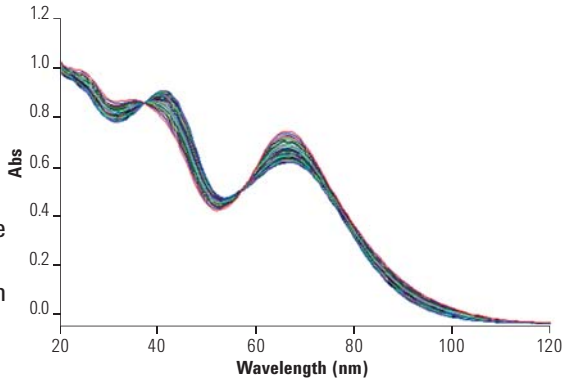
Investigating rapid photochemical and thermal kinetics

Solution

Agilent Cary 50

Analyze reactions that are over in less than a second

Monitoring a platinum organo-amide anti-cancer drug during photo-irradiation using an external source. While the reaction is complete within a second, the unparalleled fast data acquisition of the Agilent Cary 50 enables this reaction to be followed to completion.



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