



Application Note AN-RS-055

# Low-frequency Raman spectroscopy

Raman spectroscopy is an analytical tool that enables the measurement of molecular structure and the identification of materials based on the rotational and vibrational modes of a molecule. Most laboratory Raman systems cover the fingerprint spectral region ranging from 200–3400  $\text{cm}^{-1}$ . However, access to the lower-frequency areas provides additional information for applications in protein characterization [1], polymorph detection and identification [2], along with material phase and

structure determination.

The lab-grade fiber optic sampling probe for laboratory spectrometers from Metrohm can access lower-frequency modes down to 65  $\text{cm}^{-1}$ , providing a cost-effective solution for wider range measurements. This Application Note describes the use of low-frequency Raman spectroscopy for studying amino acids, polymorph detection, and phase change monitoring.

The low-frequency region augments the information content from the fingerprint region of the Raman spectrum and broadens possible applications. Detection of hydrogen bonds and other structural

## EXPERIMENT

Metrohm's i-Raman Plus 785S laboratory Raman spectrometer with a 785 nm, 300 mW (maximum power output) laser and a sensitive, thermoelectrically cooled back-thinned CCD (charge-coupled detector) is used to collect Raman spectra.

The i-Raman Plus is equipped with a unique fiber

optic probe that supports data collection over a full spectral range of 65–3500  $\text{cm}^{-1}$  with a spectral resolution of 4.5  $\text{cm}^{-1}$ . Raman spectra are collected at room temperature using SpecSuite software with integration ranging from 100 ms to 10 s.

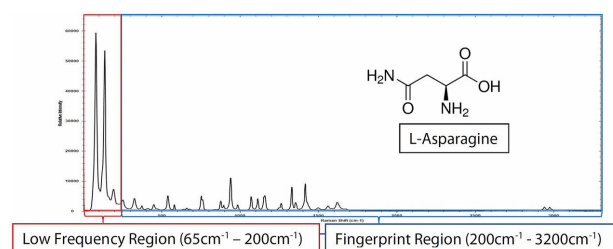
## AMINO ACIDS

Raman spectroscopy is used to study the structure and conformation of amino acids – the building blocks of proteins. The low-frequency portion of the Raman spectrum is a necessary source of information for a comprehensive study of amino acids. This is obvious in the 65–3200  $\text{cm}^{-1}$  region of the Raman spectrum of L-asparagine in **Figure 1**. Specifically, the vibrational information in a Raman spectrum can help interpret molecular interactions and biological processes [3].

**Figure 1** shows both the fingerprint region (blue) as well as the low-frequency Raman region (red) for L-asparagine; note the three dominant bands below 200  $\text{cm}^{-1}$ .

optical probe that supports data collection over a full spectral range of 65–3500  $\text{cm}^{-1}$  with a spectral resolution of 4.5  $\text{cm}^{-1}$ .

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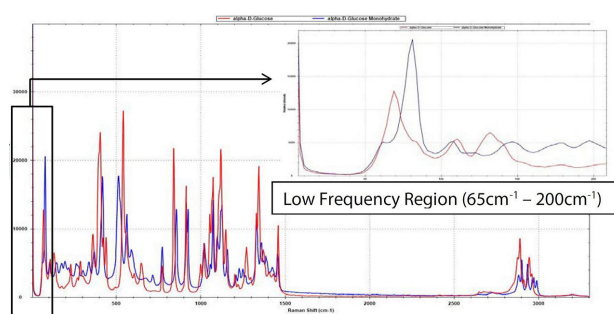
**Figure 1.** Metrohm's i-Raman Plus with standard lab-grade probe is used to collect the low-frequency spectra of L-asparagine with a total integration time of 1.2 s.

## POLYMORPH DETECTION

Determining the structural form of active pharmaceutical ingredients (APIs) is of great concern for the pharmaceutical industry. This is especially true during drug development, manufacturing, and final product quality control.

APIs exhibit polymorphism – identical chemical composition but different solid-state structures. Polymorphs may affect bioavailability and therapeutic index, so the efficacy of a drug can be compromised if the wrong form is used [2]. Pseudo-polymorphs include solvents suspended in a lattice structure.

**Figure 2** provides an example of the pseudo-polymorph D-glucose, demonstrating the ability of the i-Raman Plus probe to detect differences between monohydrate and anhydrous forms at frequencies below  $200\text{ cm}^{-1}$ .



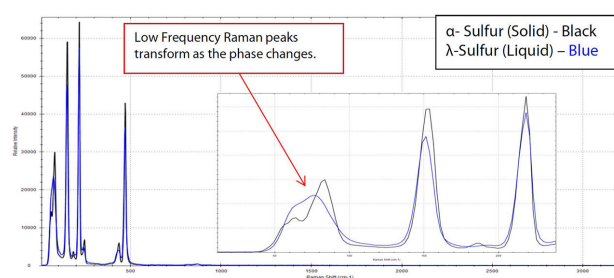
**Figure 2.** Raman spectra of  $\alpha$ -D-glucose (red) and  $\alpha$ -D-glucose monohydrate (blue), taken with 10 s integration time. Note the significant difference between the two pseudo-polymorphs within the low-frequency range (see inlay).

## MONITORING PHASE CHANGE

A change from one polymorph to another as a result of melting followed by recrystallization can be observed in the low-frequency region. Exceptional specificity is required to monitor such phase changes, and the probe of the i-Raman Plus is well-suited, as demonstrated for sulfur (**Figure 3**).

Solid  $\alpha$ -sulfur is deposited onto an aluminum tray and heated with a hot plate while Raman spectra are collected with an i-Raman Plus and fiber probe, using a laser power of 300 mW and 0.1 s integration time to measure both the solid and liquid phase.

After the sample is heated above its melting point at  $115.2\text{ }^{\circ}\text{C}$ , the low-frequency peak at  $83.6\text{ cm}^{-1}$  broadened and shifted, indicating the change from the  $\alpha$ - to the  $\lambda$ -form. Note that there are no observable changes within the fingerprint region (**Figure 3**).



**Figure 3.** Raman spectra of sulfur transitioning from the  $\alpha$ -crystalline form to the  $\lambda$ -liquid form, taken with 0.1 s integration time. Note significant broadening in the peaks located in the low-frequency region (see inlay).

## CONCLUSION

The i-Raman Plus 785S Raman spectrometer is a valuable tool for applications requiring low-frequency Raman detection down to  $65\text{ cm}^{-1}$ . The ability to characterize polymorphs and solvated forms supports manufacturing and formulation processes in pharmaceutical and biological industries.

Along with protein, polymorph, and phase characterization, low-frequency Raman spectroscopy can also be used to study semiconductor lattices [4], carbon nanotubes [5], solar cells, and an assortment of minerals, pigments and gemstones.

## REFERENCES

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## CONFIGURATION



### i-Raman Plus 785S

Das i-Raman Plus 785S ist ein hochauflösendes, forschungstaugliches Raman-Spektrometer, das für Präzision und Vielseitigkeit in einer kompakten, tragbaren Form entwickelt wurde. Ausgestattet mit einem 785-nm-Laser und einem breiten Spektralbereich von 65 bis 3350  $\text{cm}^{-1}$  bietet es eine hervorragende Leistung sowohl für qualitative als auch für quantitative Analysen. Sein TE-gekühlter CCD-Detektor sorgt für rauscharme Messungen – auch bei längeren Integrationszeiten – und damit ist es ideal für die Erfassung schwacher Raman-Signale in Anwendungen wie der Kunstkonservierung, Mineralogie und SERS.

Das System unterstützt die flexible Probennahme mit einer faseroptischen Sonde und Zubehör wie einem Küvettenhalter, einem Videomikroskop und einem XYZ-Verschiebetisch. Unterstützt von der SpecSuite Software bietet das 785S eine fortschrittliche Datenanalyse und Spektrenidentifizierung.

Das i-Raman Plus 785S ist die ideale Wahl für Teams, die zuverlässige Raman-Analysen über einen breiten Spektralbereich in anspruchsvollen Arbeitsumgebungen benötigen, von der Laborforschung bis zur Feldforschung.

Erfahren Sie, wie das i-Raman Plus forschungstaugliche Raman-Spektroskopie in einem handlichen Paket bietet:

- Breiter Spektralbereich und Spektrometer-Konfigurationen für hohe Auflösungen.
- Die kleine Grundfläche des Systems, die leichte Bauweise und der geringe Energieverbrauch sorgen überall für die Möglichkeit, Raman-Analysen auf Forschungsniveau durchzuführen.
- Das i-Raman Plus ist mit einer faseroptischen Sonde zur leichten Probennahme ausgestattet und kann mit einem Küvettenhalter, einem Videomikroskop oder einem XYZ-Verschiebetisch mit Sondenhalter verwendet werden.
- Das i-Raman Plus wird von der leistungsstarken SpecSuite Software für die einfache Erfassung von Raman-Daten sowie die quantitative Modellentwicklung, die Identifizierung mit Spektrenbibliotheken und die Routineanalytik unterstützt.



### Raman-Sonde in Laborqualität (785 nm)

Raman-Lichtleitersonde in Laborqualität mit Auslösetaste. 1.5 Meter Faserlänge. Kompatibel mit den Modellen i-Raman Plus 785S und 785HR.