APPLICATION NOTE

Infrared Spectroscopy

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Analysis of Pharmaceutical Raw Materials Using the Tri-Range Spectrum 3 Infrared Spectrometer

Introduction

Infrared Spectroscopy has become a ubiquitous tool in the analysis of pharmaceutical raw materials, with both Mid- and Near-infrared spectroscopy functioning as commonplace techniques for the investigation of both excipients and active pharmaceutical ingredients (APIs). The PerkinElmer Spectrum™ 3 tri-range infrared spectrometer (Figure 1) provides a high-performance solution for pharmaceutical analysis across the infrared spectrum.

Figure 1. The PerkinElmer Spectrum 3 FT-IR Spectrometer.

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Near-Infrared Spectroscopy

Near-infrared spectroscopy is arguably the most common spectroscopic technique for the interrogation of incoming raw materials. This is due to the robust nature of the technique and simple sampling methods which allow for quick and simple data collection. There are two main sampling accessories which may be used to investigate pharmaceutical raw materials in the near-infrared region; the Near-Infrared Sampling Module (NIRM) and Remote Sampling Module (RSM).

Each of these sampling accessories offers its own unique advantages. The remote sampling module allows the user to measure the spectrum of a material by pressing the tip of the fibre probe against the sample. This accessory is also able to measure spectra through plastic bags, reducing the need to expose the material to the potentially unclean conditions found in a warehouse. The NIRM accommodates glass vials and petri dishes. The large sampling area and sample spinner improves the repeatability of measurements of inhomogeneous samples.

Near-Infrared Reflectance Module (NIRM)

One particularly useful application of near-infrared reflectance spectroscopy is demonstrated by the discrimination between spectroscopically similar materials with the aid of principal component analysis (PCA). An example of this is the discrimination between different grades of Avicel®, a microcrystalline cellulose. The different grades of this material do not vary in chemical composition but in particle size and moisture content. NIR diffuse reflectance spectra vary slightly with particle size and moisture content and SIMCA (soft independent modelling of class analogies) may be utilized to differentiate between different grades of Avicel®.

Experimental and Results

Near-Infrared Spectra of seven different grades of Avicel® were collected using the Spectral parameters shown in Table 1.

Table 1. Parameters used for Near-infrared measurement of Avicel® samples.

Parameter	Value
Spectral Range	10000 – 4000 cm^{-1}
Spectral Resolution	8 cm^{-1}
Scan Time	40 seconds
Corrections	AVI, AVC, Stray Light and Reference Correction

Overlaid spectra of the seven different grades of Avicel® are shown in Figure 2.

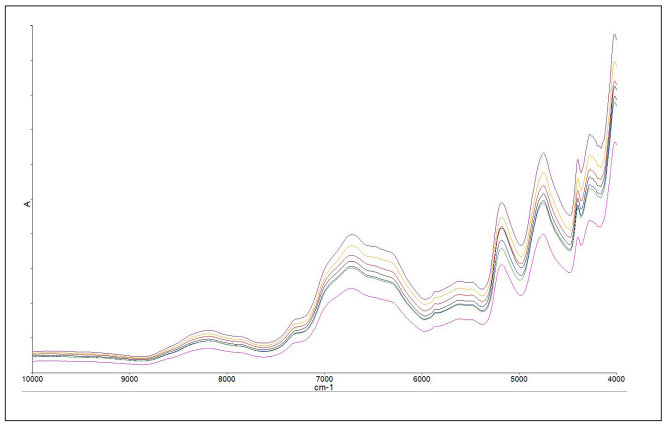

Figure 2. Overlaid spectra of different grades of Avicel®.

This clearly shows that there are very few defining spectral features which could be used to visibly differentiate between the different grades. In this scenario, SIMCA modelling is incredibly useful for differentiation between grades of Avicel, as shown in Figure 3.

The pre-processing used to create this model was a noise filter and nine point smoothing.

Each sphere in this plot represents the data from one grade of Avicel®. The further apart the spheres (larger intermaterial distance) the higher the probability of discrimination. Table 2 shows the intermaterial distances between each set of samples.

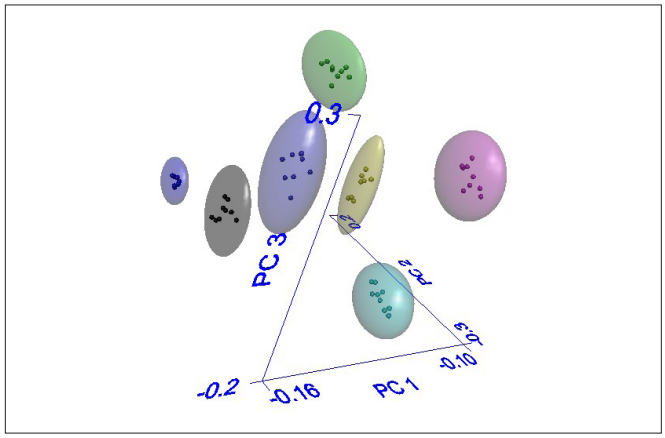

Figure 3. SIMCA model created using Avicel® spectra.

Table 2. Intermaterial distance between various grades of Avicel®.

Material	PH101	PH102	PH103	PH105	PH113	PH301	PH302
PH101	-	7.65	6.44	11.10	11.20	18.90	19.40
PH102	-	-	7.65	15.00	12.50	12.80	12.70
PH103	-	-	-	8.78	6.46	16.50	17.80
PH105	-	-	-	-	10.00	20.80	22.70
PH113	-	-	-	-	-	16.80	19.30
PH301	-	-	-	-	-	-	5.11

The model was able to correctly recognise 100% of samples within each grade. This, coupled with the large intermaterial distances, demonstrate the low probability of sample misclassification.

Validation

To validate the SIMCA model, samples measured using the same instrumental parameters as those used to build the model were identified. The results from this validation are shown in Table 4.

Table 3. Results from the Avicel® SIMCA validation.

Sample ID	Result (Pass/Fail)	Total Distance Ratio	Distance Ratio Limit
PH101	Pass	0.7490	1.0000
PH102	Pass	0.6152	1.0000
PH103	Pass	0.6195	1.0000
PH105	Pass	0.5669	1.0000
PH113	Pass	0.6826	1.0000
PH301	Pass	0.6222	1.0000
PH302	Pass	0.5243	1.0000

Remote Sampling Module (RSM)

The Remote Sampling Module, also referred to as the Fiber Probe, gives the user enhanced flexibility with regards to measuring spectra. This provides the opportunity to measure spectra of raw materials in a loading bay or warehouse setting with no sample preparation. The RSM can be easily used to verify the identity of incoming raw materials via a simple compare algorithm. This calculates the correlation between a sample spectrum and that of a known reference standard.

Experimental and Results

In order to verify raw material spectra collected using the RSM, spectra of several different raw materials were measured using the parameters shown in Table 4.

Table 4. Spectral parameters used to measure raw material samples with the Remote Sampling Module (RSM).

Parameter	Value
Spectral Range	10000 – 4000 cm^{-1}
Spectral Resolution	16 cm^{-1}
Number of Scans	32

The samples were measured, as received, by simply pressing the tip of the fiber probe against the surface of the sample and using the trigger to start the data collection.

Once measured, samples may be compared against a folder of reference spectra in order to verify their identity. The Spectrum 10 COMPARE™ algorithm features allows the user to calculate a correlation coefficient between a sample and reference spectra, as well as customize various parameters such as pass/fail boundaries.

As an example, D-mannitol (commonly used as a sweetener for medications) was measured and compared to a collection of reference spectra. The resulting sample and reference spectra are shown in Figure 4.

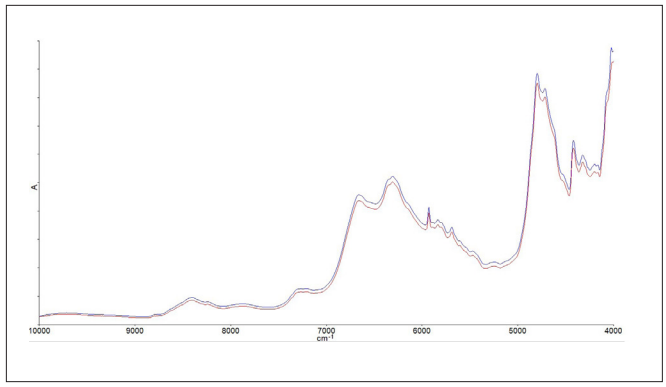

Figure 4. Measured raw material spectrum (blue) and best match reference spectrum (D-mannitol, red).

Mid-Infrared Spectroscopy

Mid-infrared spectroscopy generally contains the most information regarding the chemical structure of an analyte. Therefore, it is particularly useful for identification and analysis of active materials in pharmaceutical samples. The 'Search' function in Spectrum 10™ may be used to identify APIs by comparing the sample spectra against reference spectra in a library.

In one example, the commonly used anti-inflammatory acetylsalicylic acid (the API in aspirin) can be successfully identified using a library of APIs.

Experimental

Spectra of both the APIs and packaging materials were measured using the PerkinElmer Spectrum 3™ tri-range infrared spectrometer with attenuated total reflectance accessory. The parameters used are shown in Table 5.

Table 5. Spectral parameters used for analysis of APIs and packaging materials.

Parameter	Value
Spectral Range	4000 - 450 cm^{-1}
Resolution	4 cm^{-1}
Number of Scans	8

The overlaid spectra from this experiment can be seen in Figure 5.

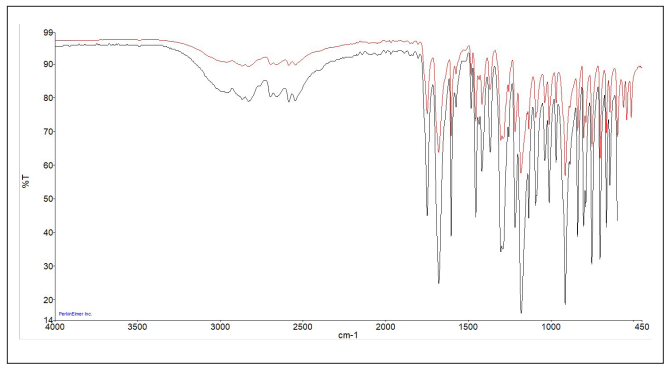

Figure 5. Overlaid spectra of a sample of acetylsalicylic acid (red) and an aspirin reference spectrum (black).

The search score of 0.998 indicates a very strong match between the sample spectrum and the reference spectrum.

Another common part of pharmaceutical quality control is identification of materials used in the packaging of pharmaceutical products. Generally, the packaging of pharmaceuticals consists of one or more polymer layers. Analysis of the packaging is crucial for ensuring the material is suitable for its intended use, according to USP <661.1>.

The PerkinElmer Spectrum 3 with universal attenuated total reflectance (UATR) accessory provides the ideal utility for measurement of these materials. In this application, ATR sampling offers a significant advantage over transmission measurement, both of which are permitted according to USP <661.1>. The main advantage offered by ATR is the ability to measure different sides of the same multilayer packaging and, in doing so, get individual spectra of the outer layer and the inner layer.

The most important features of pharmaceutical packaging are related to the inner and outer layers. The inner layer must be inert in such a way that it does not interact with the material it is surrounding. The outer layer must be resistant to the external environment, protecting the contents from contamination.

As with identification of active pharmaceutical ingredients, the main process of verifying packaging materials involves comparing spectra to a reference library.

An incredibly common material used in pharmaceutical packaging is Nylon-6. This is most often used to form an external layer as it gives the packaging a desirable level of flexibility. Figure 6 shows the overlaid spectra of the outer layer of multi-layered blister packaging and the best-hit reference spectrum.

Both the examples demonstrate the utility of mid-infrared spectroscopy as a tool for simple identification of raw materials used in the pharmaceutical industry.

Far-Infrared Spectroscopy

Although rarely used in a routine Pharma QA/QC laboratory, far-infrared spectroscopy has some uses within the pharmaceutical end-market pertaining more towards research and academia. Far-infrared spectroscopy is frequently referred to as a technique for investigating large structure such as crystals. In a pharmaceutical setting this would be most relevant to the examination of polymorphism in active ingredients.

Polymorphism occurs when a material can exhibit more than one crystal structure. This phenomenon is incredibly important in drug design as it can affect multiple physicochemical properties such as solubility, bioavailability, efficacy and even toxicity.

One active ingredient which exhibits this property is sulfathiazole, a drug formerly used as both a topical and oral antimicrobial. There are 5 different polymorphs of sulfathiazole with slightly differing crystal structures. Two of the most visibly different polymorphs of sulfathiazole are forms I and II, images of which are shown in Figure 7.

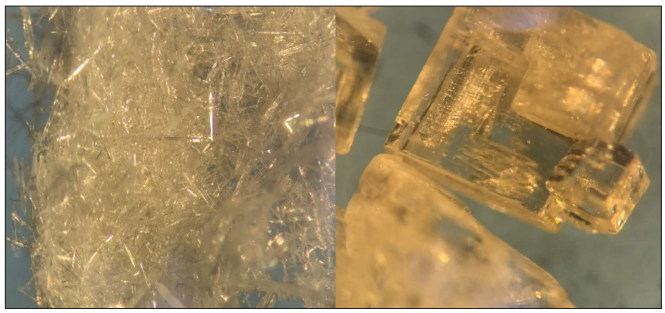

Figure 7. Magnified images of the crystal structures of sulfathiazole form I (left) and form II (right).

Experimental

Sulfathiazole form I was produced by cooling a saturated solution of sulfathiazole in isopropanol from 85 °C to ambient temperature, as described by Higuchi et al. Form II was produced by cooling a saturated solution of sulfathiazole in methanol from 65 °C to ambient temperature, as described by Parmar et al.

Far-infrared spectra of the two polymorphs were collected using the PerkinElmer Spectrum 3 tri-range infrared spectrometer with an ATR accessory containing an uncoated diamond crystal. The data collection parameters are shown in Table 6.

Table 6. Data collection parameters used to measure far-infrared spectra of different sulfathiazole polymorphs.

Parameter	Value
Spectral Range	200 – 80 cm^{-1}
Resolution	4 cm^{-1}
Number of Scans	128

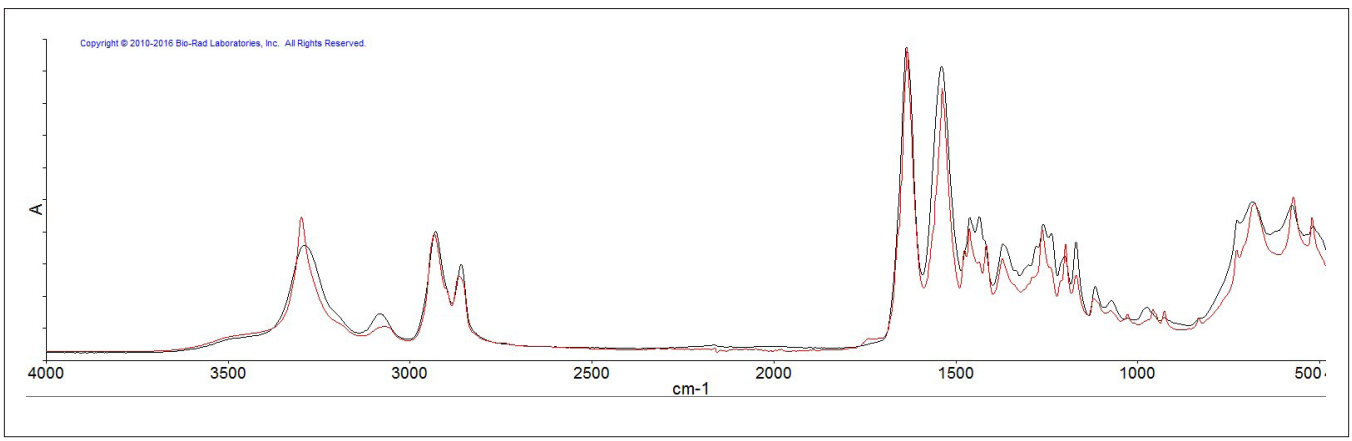

Figure 6. Overlaid spectra of the measured packaging (black) and the reference (red).

The infrared spectra of these two polymorphs are also fairly similar other than some significant bands in the Far-infrared region of the spectrum. Below 200 cm^{-1} the spectra begin to deviate from one another. At 97 cm^{-1}, polymorph I has a strong band whereas polymorph II does not. Between 90 cm^{-1} and 85 cm^{-1}, polymorph II has two strong bands (at 90 cm^{-1} and 86 cm^{-1}) whereas polymorph I only has one, relatively weak band at 89 cm^{-1}. The far-infrared spectra of the two polymorphs of sulfathiazole are shown in Figure 8.

Summary

The PerkinElmer Spectrum 3 FT-IR spectrometer provides a high-performance solution for infrared analysis of materials used in the pharmaceutical industry. Spectrum 10™ software gives the user a wide variety of powerful data processing tools allowing for in depth analysis of data. Furthermore, the enhanced security (ES) version of the software allows for analysis to be carried out in compliance with 21 CFR part 11 regulations.

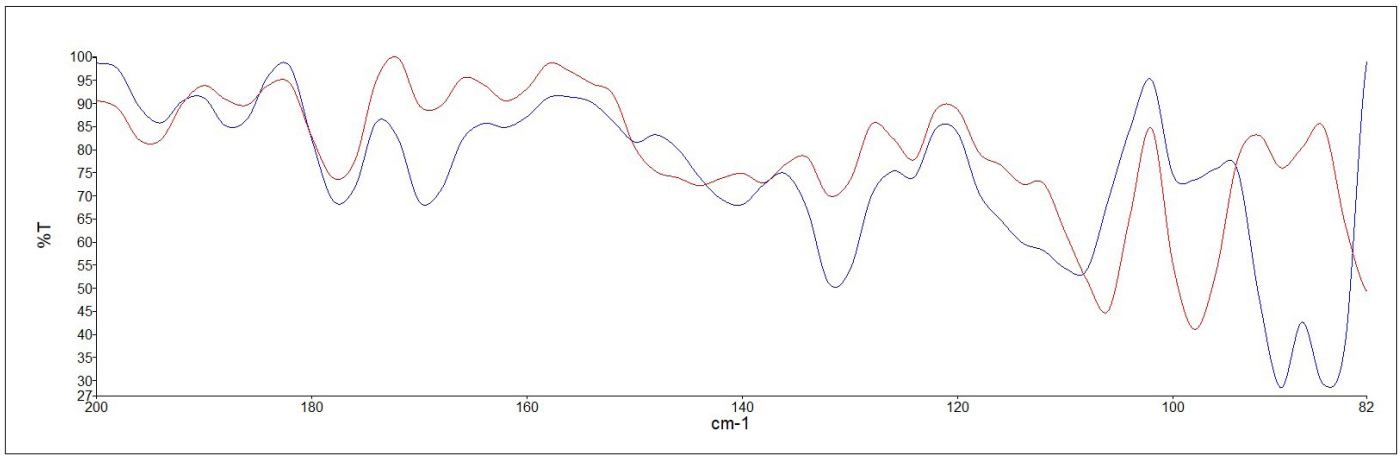

Figure 8. Sulfathiazole polymorph forms I (red) and II (blue).

References

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2. M. M. Parmar, O. Khan, L. Seton, J. L. Ford, Polymorph selection with morphology control using solvents, Cryst. Growth Des., 2007, 7, 1635-1642

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