



## Discriminant analysis of cellulose esters using FT-NIR

### Field-to-laboratory method transfer

#### Keywords

Nicolet iS50 NIR Module, FT-NIR spectroscopy, drug delivery, cellulose acetate

#### Introduction

Near-infrared (NIR) spectroscopy is a common analysis tool for highly controlled process environments such as those found in the food or pharmaceutical industries. The near-infrared portion of the electromagnetic spectrum is easily transmitted through glass containers, windows, and fibers, which makes it ideally suited to analyze materials directly in the process stream without needing to remove (thieve) samples and analyze them in the laboratory. Fiber optic probes are commonly utilized to query the in-process materials remotely in real time. Additionally, samples that are contained in glass vials can be analyzed without exposing the analyst to the material or the analyte to the environment.

NIR analysis methods are often developed and tested in controlled laboratory settings using research-type instruments. The flexibility of the instrument and the controlled environment are best suited for collecting spectra from the ideal standard set and developing the best analysis and method parameters. Once the method has been established, it is then deployed to process NIR instruments that are typically more rugged and designed to analyze the materials in an industrial process environment routinely.

Occasionally, a deployed and accepted method is failing in the field and needs to be revisited in the laboratory. In these cases, test samples are sent back to a central laboratory as part of

the method analysis and troubleshooting. This assessment and correction process also requires that the field method in use be transferred to the analytical laboratory instrument, and that the lab/field instrument performance be closely matched. We demonstrate here the equivalency of method transfer from a process instrument, the Thermo Scientific™ Antaris™ II FT-NIR Analyzer to a laboratory instrument, the Thermo Scientific Nicolet™ iS50 Spectrometer (both shown below), for the purpose of analysis, assessment, and correction. To prove the robustness of this “reverse method transfer” (field to lab), a series of cellulose acetate esters were used as a model system.



Thermo Scientific Nicolet iS50 FTIR Spectrometer, equipped with the iS50 NIR module



Thermo Scientific Antaris II FT-NIR Analyzer

The Nicolet iS50 FTIR Spectrometer is fully configurable to perform NIR analysis and is ideally suited for lab-based development of methods intended for deployment to process instruments. The Nicolet iS50



Figure 1: iS50 NIR module

Spectrometer collects NIR data using either transmission in the sample compartment or an attached module, as shown in Figure 1, an integrating sphere or fiber optic probe. The Thermo Scientific TQ Analyst™ Chemometrics Software allows the development and optimization of methods which can then be transferred to field-based systems. In this case, the reverse was done—a developed method in the field was “returned” to the lab setting.

Cellulose acetate esters were selected as a model material set because they are heavily exploited in pharmaceutical settings where they are used for controlled drug delivery. Additionally, cellulose acetate esters are available in a wide variety of different forms. Their controlled porosity and solubility make them good excipients and coatings for solid dosage forms. Each sugar residue in cellulose has three hydroxyl groups that are amenable to esterification. Quantitative measurements of the amount of ester present are either reported by percent or by degree of substitution (DS). The DS of a particular material indicates the average number of ester groups found on each residue and will fall between 0 and 3. The cellulose acetate materials analyzed here were derivatized with propionate or butyrate groups to various degrees. Unfortunately, these derivatized materials look physically similar, making them difficult to identify. Furthermore, since they have similar chemical structures, time-consuming analytical techniques can be required to distinguish between them. Fortunately, Fourier transform near-infrared (FT-NIR) spectroscopy can be used to distinguish cellulose esters with great speed and accuracy.

For this analysis, the field method was obtained from a process environment where a series of nine different cellulose acetate esters were regularly classified. Table 1 identifies the nine cellulose materials used as well as their percent and type of ester substitution. For this process method, the samples were placed in glass vials and analyzed three or four times using the Antaris II integrating sphere module. Between scans, the contents in the vials were shaken and then compacted by gently tapping the vial on a solid surface. This ensured consistency in the density while simultaneously ensuring variety in sampled material. The samples were scanned between 10,000 and 4000  $\text{cm}^{-1}$  at a resolution of 8  $\text{cm}^{-1}$  with 16 scans per analysis. Baseline offsets were minimized by analyzing the data as first-derivative spectra. Figure 2 shows the variation between the different cellulose materials in the first derivative spectra and the regions used in the chemometric analysis.

Discriminant analysis was performed using the TQ Analyst Software. The 3-dimensional principal component scores plot, Figure 3, shows the exceptionally clear discrimination obtained in the field method between the various cellulose ester types—note the different colored sets of dots are well separated. To evaluate transfer from the process instrument to the laboratory instrument, several samples were reanalyzed using the iS50 NIR Module attached to a Nicolet iS50 Spectrometer, using the same collection parameters as the original work.

ID	%	DS	Ester	MW (kDa)
B 17	17	0.7	Butyryl	65
B 38	38	1.8	Butyryl	40
B 46	46	2.0	Butyryl	20
B 52	52	2.5	Butyryl	30
P 45	45	2.3	Propionyl	25
P 47	47	2.5	Propionyl	75
P 48	48	2.6	Propionyl	25
A 40	40	2.4	Acetyl	30
Microcrystalline cellulose	0	0	—	

Table 1: Identity and characteristics of cellulose materials used.

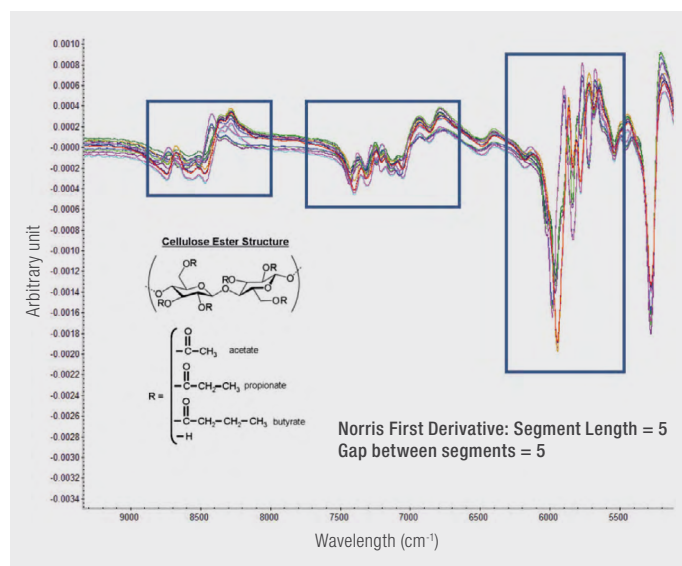


Figure 2: Variation between the different cellulose materials in the first derivative spectra and the regions used in the chemometric analysis is shown.

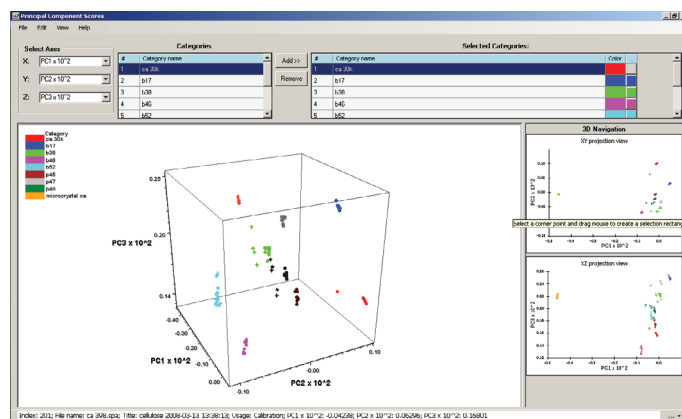


Figure 3: The 3-dimensional principal component scores plot shows the clear demonstration obtained in the field method between the various cellulose ester types. Note the different colored sets of dots are well separated.

First, samples of 38% cellulose acetate butyrate (B 38) were analyzed on the integrating sphere of the iS50 NIR module and with the fiber optic probe. The probe was tested both pressed directly in the sample and through the side of the 0.5 kg glass bottle. A comparison of the spectra obtained is shown in Figure 4. The Nicolet iS50 Spectrometer spectra, offset to make the comparison easier, are virtually identical to the spectrum collected using the Antaris process instrument regardless of the specific collection method.

The method transfer was then tested using samples from three of the nine different cellulose acetate esters—38% butyrate, 52% butyrate, or 48% propionate esterified. The samples were measured in duplicate using the integrating sphere and the fiber probe. The resulting spectra were analyzed with the field method to determine if they would be correctly classified without further method modification.

Table 2 shows the results: all spectra from samples acquired on the Nicolet iS50 were classified correctly. The Mahalanobis distance measures how well a sample matches a given class; essentially, it is the distance from the sample point to a cluster in Figure 3—the smaller the value, the more likely the sample “belongs” to that class. The Mahalanobis Ratio of the distance from the second closest class to the distance to the nearest class provides a measure of the discrimination: higher ratio values indicate the spectra are very close to the correct class and are considerably different from the nearest incorrect class – meaning they are well discriminated. The 48% propionate samples have the lowest ratio because of their extreme similarity to the 45% and 47% propionate samples. However, even these challenging samples were correctly classified. The

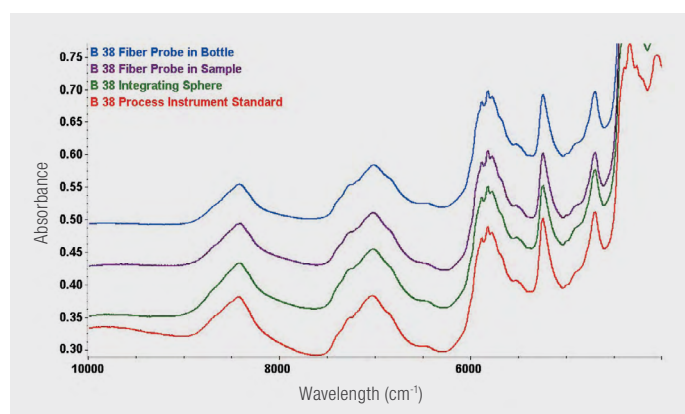


Figure 4: Samples were reanalyzed to evaluate the transfer from the process to the laboratory instrument. The Nicolet iS50 Spectrometer spectra are identical to the spectrum collected using the Antaris analyzer, regardless of the specific collection method.

fact that each sample was assigned to the correct class and these ratios are good indicates the reverse method transfer would be successful. By the same argument, the disagreement would indicate a true failure of the calibration or a true difference in the sample, providing the analyst with evidence for a need to correct the field method.

### Conclusion

This study demonstrates two Nicolet iS50 FTIR Spectrometer features for NIR method development. First, the instrument can discriminate between the various modified cellulose acetate esters, allowing its use in developing methods in the laboratory. Second, the reverse transfer shows the instrument can also provide a platform for deploying methods to the field or debugging field applications in the lab. This represents a key function of the analytical laboratory supporting process control, raw materials identification, and other field methods.

Simultaneously, the Nicolet iS50 FTIR Spectrometer includes all the tools expected from a flexible, analytical, and research-grade spectrometer system. This includes built-in ATR for materials identification, sample compartment FT-Raman for morphology studies, and complementary materials identification or even GC-IR or TGA-IR for more advanced separation and deformation analyses.

	Actual class	Calculated class	Mahalanobis ratio
Integrating sphere	B 38	B 38	5.5
	B 38	B 38	4.4
	B 52	B 52	3.4
	B 52	B 52	2.4
	P 48	P 48	1.4
	P 48	P 48	1.3
Fiber probe (through glass)	B 38	B 38	2.6
	B 38	B 38	2.2
	B 52	B 52	3.8
	B 52	B 52	3.4
	P 48	P 48	1.3
	P 48	P 48	1.4
Fiber probe (in sample)	B 38	B 38	2.0
	B 52	B 52	3.4
	P 48	P 48	1.2

Table 2: Results of the method transfer test using samples from three of the nine different cellulose acetate esters—38% butyrate, 52% butyrate, or 48% propionate esterified. All spectra from samples acquired on the Nicolet iS50 were classified correctly.

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