Absolute Virtual Instrument (AVI) Technology to achieve more robust and dependable analytical results

Introduction

When an infrared spectrum of a sample is measured, the shape of the spectral features are a convolution of the bands due to the sample itself and the influence of the spectrometer, including any sampling accessory which may be present. In practice, the relative contributions of the sample line shapes and the instrument line shape (ILS) can vary enormously; to the extent where sometimes the instrument line shape dominates the appearance of the bands (usually when working at relatively low spectral resolutions), and sometimes the sample line shape dominates.

Considering the line shapes in mid-IR and NIR spectra of condensed phase materials, it is often the case that the sample absorption bands (full width at half height or FWHH) are broader than the instrument resolution used, and slight changes in the ILS do not greatly influence the shapes of the measured spectral bands. In many situations however, this is not the case and slight changes in the ILS can lead to discernable changes in the measured spectra. Whether this variation is important depends entirely upon the application. For example, in an application which simply discriminates between a few chemically very distinct materials, the ability to discriminate may not be adversely affected by small ILS variation. On the other hand, a chemometric method which determines active ingredient concentrations with high reproducibility, using highly overlapped spectral regions may be adversely affected.

There can be various manifestations of poorly controlled instrument line shape. First, in a single instrument, changes due to replacement or even aging of certain optical components (e.g. the source) can change the ILS, and calibration models can subsequently produce biased results, predict less reproducibly or even break down completely. Second, attempting to transfer a calibration model or library developed using one spectrometer to a different spectrometer can fail. Often, quantitative and qualitative models rely on the accurate measurement of very small spectral differences; and upsetting the ILS can have disastrous consequences.

PerkinElmer®
For the Better

The Absolute Virtual Instrument™ (AVI) from PerkinElmer has been designed to address the problem of inconsistency of instrument line shape and wavenumber calibration[1]. This problem exists for all commercial IR and NIR spectrometers. The PerkinElmer FT-IR and FT-NIR spectrometers are inherently capable of the highest levels of wavelength reproducibility (typically better than 0.01 cm^{-1} at 2800 cm^{-1}). However what is often not mentioned in the wider literature is that regardless of instrument manufacture, many user actions can inadvertently affect this performance for the worse – and quite dramatically. AVI is intended to allow users to effectively lock the instrument back into calibration after these events as required. The result is improved instrument measurement consistency which in turn leads to more robust and dependable analytical results. This paper outlines the concept and performance of AVI as implemented on PerkinElmer FT-IR an FT-NIR spectrometers.

Approaches to improving consistency of measurement

The aim is to achieve consistent measurement both over time and between instruments as much as possible. In addition, if possible, the actions necessary to achieve this should not require unduly specialized instrument skills (for example for the physical re-adjustment of components) or hardware. Measurements should also be traceable to known standards.

One approach to achieving this is the "Master/Slave" instrument concept currently used in a number of commercial instruments. Here a "master" instrument is nominated and 'slave' instruments are adjusted so that the spectra of certain transfer standards match those of the master instrument as close as possible. There are some limitations of this approach; a major concern is that the master instrument itself can vary over time. In addition the calibration procedure is often dependent on specialized transfer standards and/or the skills of an instrument service engineer. In practice this can mean that the procedure is only performed at the instrument factory or during service calls once the instrument is in use. In every day use, situations arise when it is desirable to be able to check and re-calibrate a system on demand – for example after user replacement of an optical component or even changing the sampling configuration.

The AVI eliminates these concerns by replacing the measured spectrum of a transfer standard (the "transfer spectrum") from the master instrument with a synthetic master spectrum calculated for the instrument to be calibrated. In the case of a FT-IR or FT-NIR instrument, it is possible and relatively easy to calculate such a spectrum from a known material such as methane. This calculation is possible given two requirements; (a) the "raw" or un-convoluted "stick" spectrum of the material which is known independently, and (b) a thorough understanding of how the spectrometer affects the ILS. In the case of FT-IR and FT-NIR spectrometers, contributions to the ILS include the nominal resolution determined by the optical path difference of the interferometer, the apodisation function and the beam divergence in the interferometer.

The former two components are easily calculated, the latter is determined empirically as it is affected by several components in the beam, including the sampling system.

Methane is a suitable standard for calculating the synthetic master spectrum as the HITRAN database[2] provides the frequencies and intensities of selected methane bands to very high accuracy. As the spectra have very narrow lines, they are very sensitive to the ILS. Therefore, for a given instrument, it is possible to (a) simply measure a spectrum of methane, (b) calculate a synthetic master spectrum from the HITRAN database for that set of instrument conditions, temperature and pressure and (c) derive a correction function which converts the measured spectrum to the synthetic master spectrum.
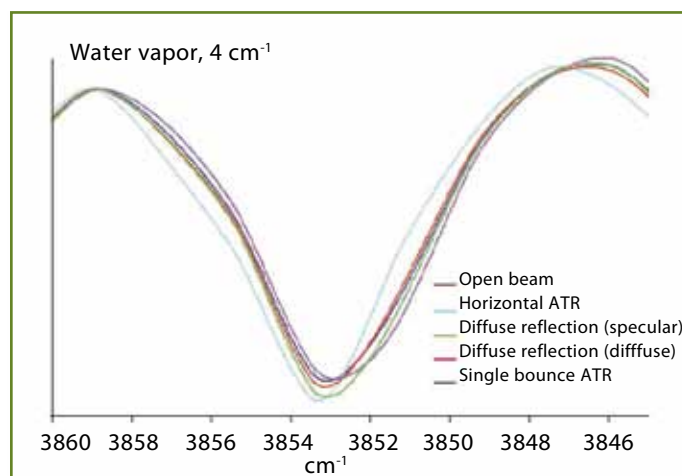

Figure 1. Effect of sampling accessories on lineshape.

This correction function can then be applied to every measured spectrum to optimize both line shape and wavenumber calibration to that of a "theoretical" master. The implementation can be such that the determination of the correction function can be performed at the touch of a button i.e. as often as desired – and the correction function applied automatically to all spectra measured under those conditions. Furthermore, the response correction can be easily generated and stored for different sampling configurations provided it allows the measurement of the methane spectrum. This is important as it is known that changing instrument sampling geometry can significantly alter the instrument wavenumber calibration and lineshape and is true for all commercial interferometers.

AVI in practice

In PerkinElmer FT-IR and FT-NIR spectrometers equipped with AVI, such as the Spectrum Two and Frontier systems, the implementation is designed to allow the user to initiate the generation of the AVI correction function at any time (known as "AVI Calibration"). Although FT-based systems have been shown to be remarkably precise with respect to wavelength repeatability, it is known that calibration is subject to disturbance of optical elements including sampling and other effects (which in themselves may not initiate a service call). The use of AVI can ensure that wavenumber scale and ILS are optimized at any time. This offers an invaluable degree of assurance of consistent instrument operation with respect to these important parameters without the need for a service engineer. This is achieved by introducing a small gas cell containing methane into the beampath - for a given set of scan conditions (those to be used for sample measurement), the methane is measured, the correction function stored and automatically applied to all subsequent spectra generated under these conditions. If these scan conditions are changed (e.g. resolution) the user is invited to recalibrate. Automatic sample accessory recognition, where fitted, assists this procedure, so that if the user exchanges the accessory for a new accessory, the system automatically prompts for a new AVI calibration. There may be instances where AVI correction using methane does not add significant benefits; for example for measurement of very broad features in the very short wavelength NIR. In these instances, the correction can be disabled through software, and it is of course still possible to use the internal laser for wavelength calibration to deliver wavelength consistency similar to that of FT-IR and FT-NIR systems without AVI implemented.
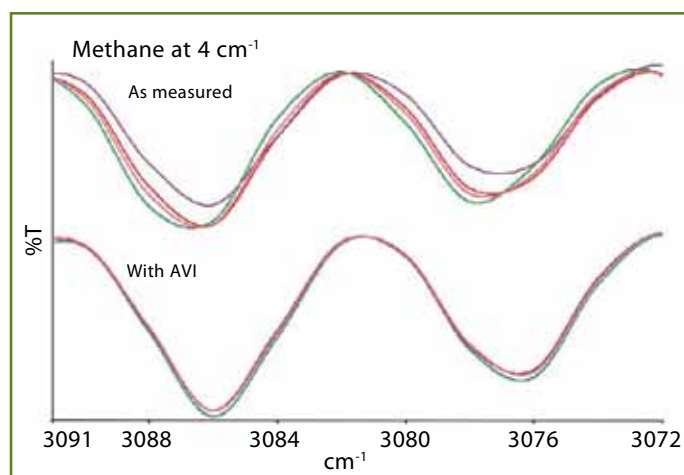

Figure 2. Correction for sampling systems.

AVI Performance

To demonstrate the need for separate corrections for different sampling arrangements, a spectrum of water vapor was collected with a number of common sampling accessories in the beam. Scan conditions, sample temperature and pressure were constant throughout, so it can be assumed the differences in the spectra are essentially due to the differences in sampling geometries. The spectra are shown in Figure 1, where it can be seen that not only have the interpolated band minima changed position, but line shapes have changed also.

Figure 2. shows the results of a similar experiment using methane bands, but with the measurements repeated with and without the AVI correction as described above. It is clear there is a significant improvement in consistency between the measurements with different accessories. Note that the corrected spectra are not perfectly overlaid. AVI does not correct for all the contributions to the measurement inconsistency. For example, the accessories used in this study are known to contribute different but measurable amounts of stray light to the overall signal and AVI does not correct for this.

To test the effectiveness of the AVI correction function, the spectra of methane were collected at 0.5, 1.0, 2 and 4 cm^{-1} resolution, spanning fully and partially resolved bands, and compared with the synthetic master spectra calculated from the HITRAN database using the same scanning parameters. The results are shown in Figure 3A and Figure 3B. The agreement between observed and calculated spectra is typically within 1% of the spectral resolution.

Using AVI provides you with the critical assurance that your instrument is optimized with respect to these important calibration parameters at all times, not just immediately following a service engineer's call.

AVI and calibration transfer

Variations in wavelength calibration and line shape are two of the most significant factors influencing the ability to transfer a multivariate calibration (a) after instrument maintenance and (b) from one instrument to another instrument. The problem of calibration transfer is actually a collective term for a range of potential issues which may be encountered in practice, from simple wavelength shifts causing bias in results, to more subtle and intractable intensity and line shape inconsistencies which may arise by a number of mechanisms. There are a number of approaches[3] which have been taken to address these issues and they can be broadly categorized as follows:

(1) Improving the robustness of the calibration/model (here we refer to either a quantitative or qualitative method, univariate or multivariate). This could include data pre-processing methods such as scatter correction and spectral derivatives.

(2) Adjusting the calibration model to improve its predictive ability on the target instrument. This could for example involve augmenting the calibration model using sample spectra recorded on the target instrument to improve the model.

(3) Adjusting the spectra of the target instrument to better match those of the source instrument or master instrument. In some instances suitable samples used in the calibration can be used as transfer standards, provided they exhibit adequate stability. A number of algorithms and patented methods exist for this process(3).

AVI does not neatly fall into these categories; as it uses a method independent transfer standard to ensure raw data is consistent as possible. However, there is no reason why it cannot be used in conjunction with the techniques described above. In our experience with FT-IR and FT-NIR methods, it has been found that for many quantitative and qualitative applications, the use of AVI standardized data with some judicious use of spectral preprocessing is all that is required to ensure calibrations transfer well – along with careful attention to the sampling presentation protocols. The following example serves as an indication of the improvements to calibration transfer which can be realized when AVI is enabled. A calibration training set consisting of 26 mixtures of aromatic hydrocarbons (ortho-xylene, para-xylene, ethyl benzene and toluene) was prepared and measured by NIR transmission using a PerkinElmer FT-NIR System equipped with a fixed 0.5 mm pathlength transmission cell. Component concentrations ranged from 5-35%. A PLS1 regression model was performed using the range 6400-4100 cm^{-1}, and full crossvalidation produced standard errors of prediction (SEP) between 0.04-0.1% for the four components. The same samples were measured using a second spectrometer that was un-calibrated.
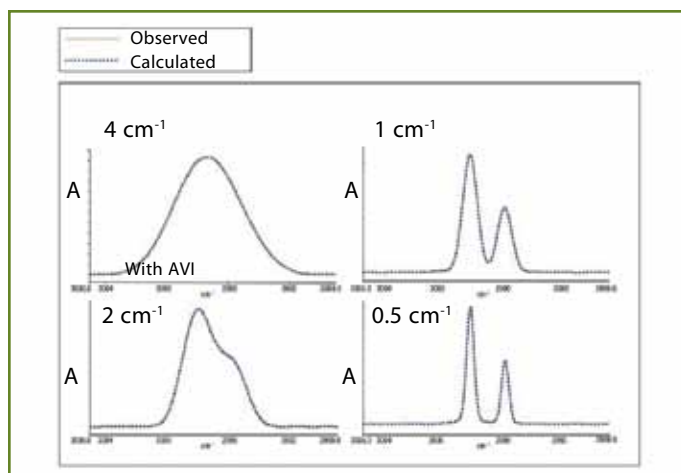

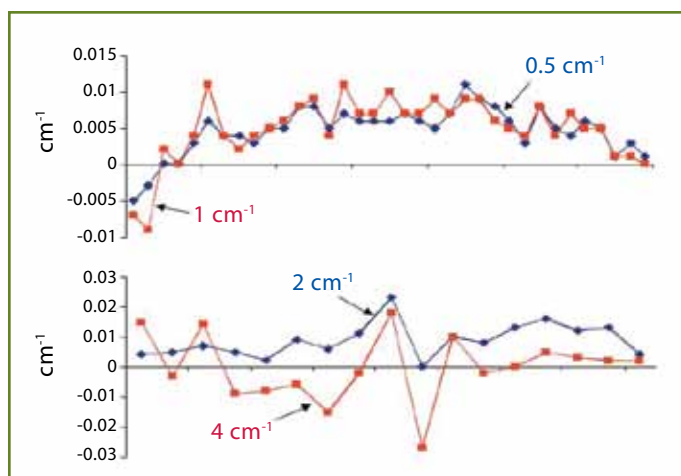*Figure 3A.* AVI corrected and HITRAN data for HCl.

Figure 3B. Deviations from HITRAN band positions.

It should be pointed out that spectrometers do not leave the factory uncalibrated. This instrument was deliberately uncalibrated to demonstrated the potential of AVI. As outlined above, in practice there are a number of scenarios where the user could render an instrument effectively uncalibrated. For these spectra the SEP values for the four components increased to 0.1-0.8% (Figure 4A). After AVI calibration of the second spectrometer the SEP values fell below 0.10% (Figure 4B) and are not significantly different from those of the original spectrometer.
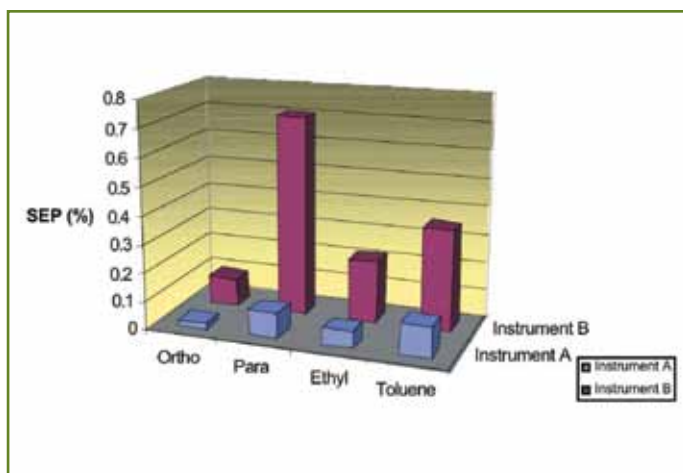
Figure 4A. SEP values for aromatic hydrocarbon mixtures – without AVI. Calibration generated using Instrument A data.
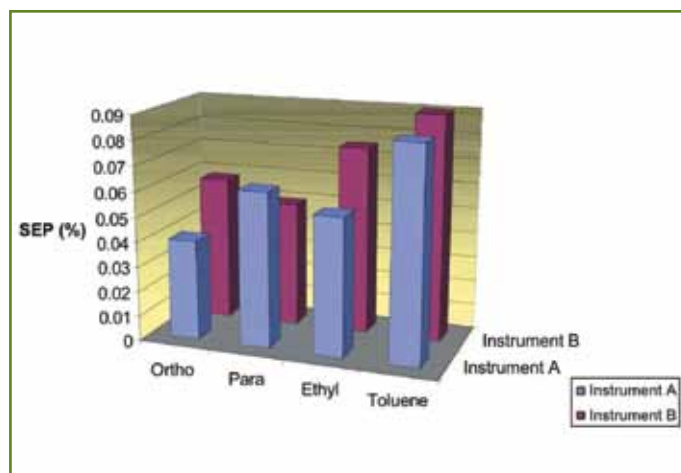

Figure 4B. SEP values for aromatic hydrocarbon mixtures – with AVI.

Conclusion

AVI has been shown to be a highly effective technique for improving the consistency of measurement between FT-IR and FT-NIR spectrometers. It should be emphasized that the purpose of the invention is not to provide the opportunity to manufacture or service the spectrometers to lower standards, although it could be argued the technique offers increased improvements to instruments of poorer wavelength and line shape stability. Well designed FT-IR and FT-NIR instruments are already inherently relatively stable with respect to these parameters compared with scanning dispersive and diode array counterparts. The design philosophy at PerkinElmer is to apply AVI to instruments which have been manufactured to the highest standards with respect to optical performance – to address issues present to some degree in all spectrometers in a way which does not rely on master instruments, troublesome transfer standards or advanced servicing skills. The implementation is simple. This in turn improves the opportunity for non-specialized operators (users) to record consistently optimized spectra with respect to two of the most important parameters (one is often overlooked) affecting the reliability of analytical results.

In the context of validation, AVI can be regarded as a more comprehensive form of instrument calibration than checking band position as outlined in many test protocols. This is because it addresses the additional problem of line shape which itself is critical to the precise determination of band maxima.

References

1. Ganz; Alan M., Wang; Yongdong, Tracy; David H., Hoult; Robert A, Cahill; Jerry E., Huppler; David A. US Patent 6,049,762.

2. L.S Rothman et al., J. Quant. Spectrosc. Radiat. Transfer 60(5), 665, (1998).

3. T Fearn; J. Near Infrared Spectrosc. 9,229-244 (2001)

PerkinElmer

For a complete listing of our global offices, visit www.perkinelmer.com/ContactUs