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Comparison of Transmission and Internal Reflection Infrared Spectra of Cocaine

ABSTRACT: Comparing the infrared transmission spectrum of cocaine HCl to its attenuated total reflection (ATR) spectrum has raised questions about the use of ATR spectra for forensic drug analysis. Whenever infrared spectra are collected using different modes or sample preparation methods, small variations in peak intensity ratios or peak positions are possible. These variations in infrared spectra are small and do not interfere with qualitative analysis, but they can cause confusion when unrecognized as normal effects of the different spectroscopic techniques. Comparison of the absorption and ATR spectra of cocaine hydrochloride illustrates the type of differences that can be expected. These differences are explained by the fundamental differences in the collection techniques. For the best quantitative results, only spectra collected by the same technique should be compared.

KEYWORDS: forensic science, chemistry, infrared spectroscopy, attenuated total reflection, internal reflection spectroscopy, transmission spectroscopy, cocaine salt, cocaine base, drug analysis, dispersion

The two most common methods for collecting infrared spectra are transmission and internal reflection. While transmission is the traditional method for recording infrared absorption spectra, today, ATR spectra collected using the internal reflection method are rapidly displacing the older technique. Most collections of infrared absorption spectra were recorded using the transmission method, but libraries of ATR spectra are available (1,2).

Absorption spectra report the fraction of the incident radiant energy transmitted through a uniform thickness of a material. If the material is a gas or liquid, then the sample is contained between infrared transmissive windows. Solids are often mixed with KBr and pressed into a "pellet," or ground in mineral oil (Nujol) to form a paste that is spread into a thin film between salt windows. The absorption spectral energy is reported as the fraction of radiant energy, Transmittance (T) or Absorbance [$A = \log (1/T)$].

Transmission measurements have their unique factors that cause variations in peak shapes and intensities. A small bubble in a liquid film will distort intensity ratios. Water in KBr introduces its own bands and can cause ion exchange in some organic compounds. When mineral oil or other liquids are used to make mulls, they introduce interfering absorption bands. Particle size in KBr pellets or mulls can also cause band distortions (Christiansen Effect). Many libraries of infrared absorption spectra contain spectra collected using mulls. The Nujol bands distort the carbon-hydrogen absorption region. Spectral differences in these regions commonly are ignored. All of these effects are well documented and accepted as routine artifacts in recording transmission spectra. The qualitative infrared analysis is based on the presence of numerous spectral band correlations and is not limited by known artifacts.

Until Fahrenfort's pioneering work in 1959, all infrared spectra were collected using the transmission method. He introduced "Attenuated Total Reflection—a new principal for the production of useful infrared reflection spectra of organic compounds" (3). It is very important to note that ATR spectra are reflection spectra, not absorption spectra. The principles of internal reflection were rigorously described by N.J. Harrick (4,5). Since the early 1960s, the collection of ATR spectra, using the principals of internal reflection, has gained wide acceptance. Because of simplicity, the advantages of FT-IR spectrometers and the availability of efficient internal reflection accessories, ATR spectra are commonly recorded for infrared analysis.

Cocaine (base) and cocaine hydrochloride were selected to illustrate the comparisons between transmission and ATR spectra. The spectra of these drugs are reported in Figs. 1 and 2. Each figure contains both the absorption spectrum and the ATR spectrum. The similarities between these ATR spectra and the corresponding absorption spectra are evident. Because of these similarities, its is often written that ATR spectra are absorption-like spectra. Closer examination reveals differences. The carbonyl absorption bands found between 1800 and 1650 cm^{-1} are notable for their differences in intensities and peak shifts (Figs. 3, 4). These spectral differences are real; they are the direct result of differences between the reflection and transmission processes. The complex role the sample's refractive index plays in internal reflection is of primary importance in understanding the spectral differences.

Experimental

Spectra were collected using three different techniques. Absorption spectra were recorded using the KBr pellet method to prepare samples of the solid drugs. ATR spectra were collected using a small diamond internal reflection element with a 45° incident angle. Specular reflection spectra were collected on single crystals using a microspectrometer. Unless stated differently, spectra were collected at 4 cm^{-1} spectral resolution.

The cocaine salt standard was obtained from Sigma Chemical Company. The cocaine base reference was synthesized by disassociating the cocaine salt standard using a basic extraction.

Transmission spectra were obtained using a Mattson Genesis I Fourier Transform Infrared Spectrometer (FT-IR) (Madison, WI) with a Gateway 2000 Computer equipped with the WINFIRST program, and a Hewlett Packard printer. The following parameters
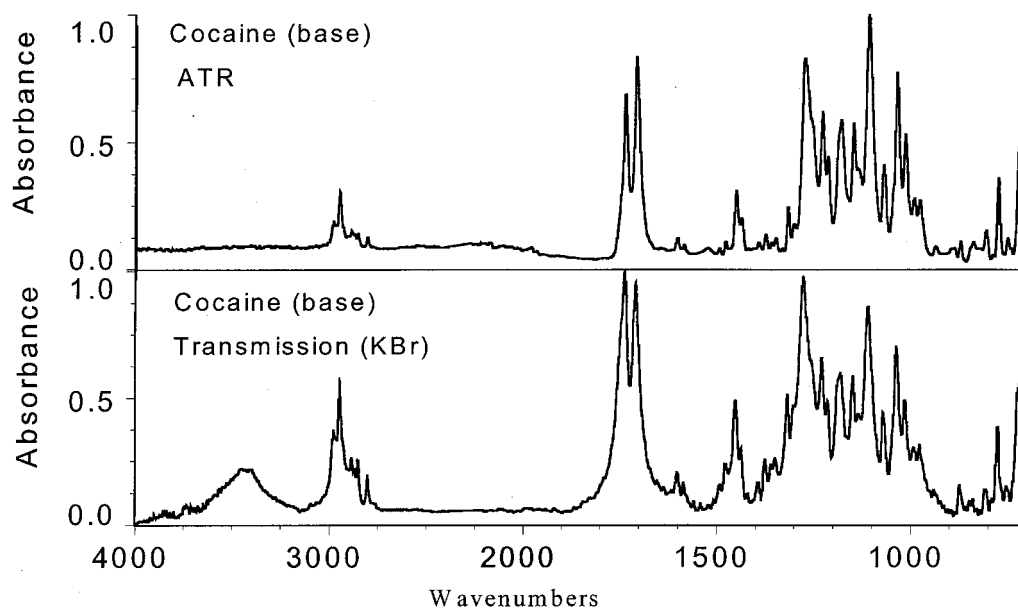
FIG. 1—*The ATR and absorption spectra of cocaine (base) are presented to illustrate the correspondence and differences between internal reflection and transmission measurements.*
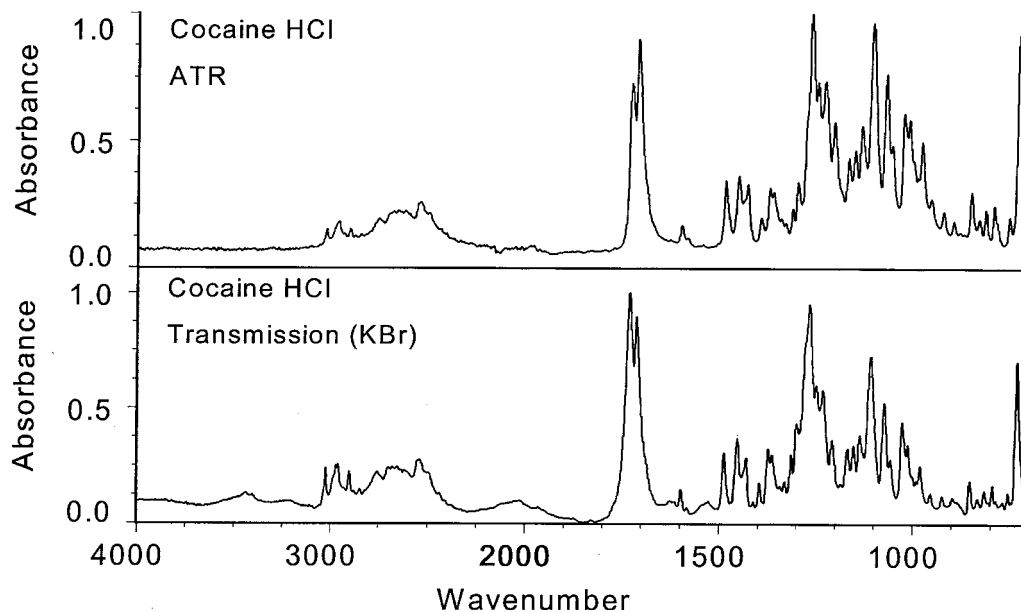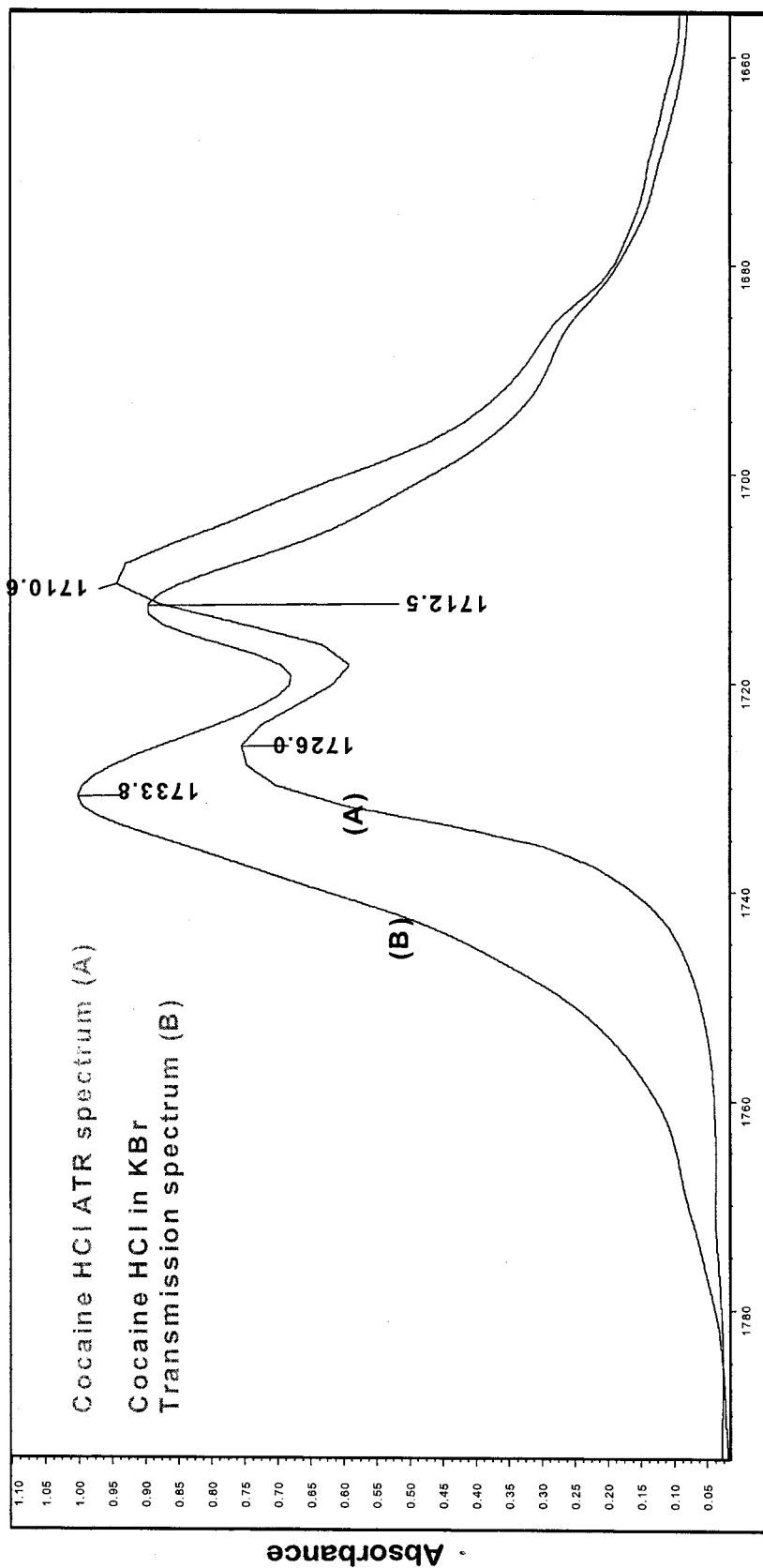

FIG. 2—*The ATR and absorption spectra of cocaine HCl are presented to illustrate the correspondence and differences between internal reflection and transmission measurements.*

were used: 4 cm-1 resolution, 10 scans, and a gain of 1. Each cocaine sample was prepared as a pellet using infrared grade KBr from Mallinkrodt.

ATR spectra were obtained with cocaine salt and cocaine base, using the same instrumentation as listed above with the addition of the Golden Gate (Graseby Specac Inc., Fmyrna, GA) or a DuraSampler (SensIR Technologies, Danbury, CT) attachment. The following parameters were used: 4 cm-1 resolution, 36 scans, and a gain of 1.

External reflection spectra from individual single crystals of cocaine salt and cocaine base were collected using an IRμs micro-

spectrometer system, (Nicolet Instrument Corp., Madison, WI). The smooth, flat surface of the single crystals produced pure specular reflection. The intensity weighted average incidence angle was 29° (15 times magnification, 0.58 NA) for the Schwarzschild objective. A gold reference mirror was used when collecting all external reflection spectra. The specular reflection spectra were processed with a Kramers-Kronig transformation program available on the IRμs spectra processing software. Using this Kramers-Kronig transformation, the refractive index as a function of wavelength was directly calculated from the specular reflection spectra.

FIG. 3—*The carbonyl band absorption of cocaine (base) showing the difference between ATR and absorbance spectra.*
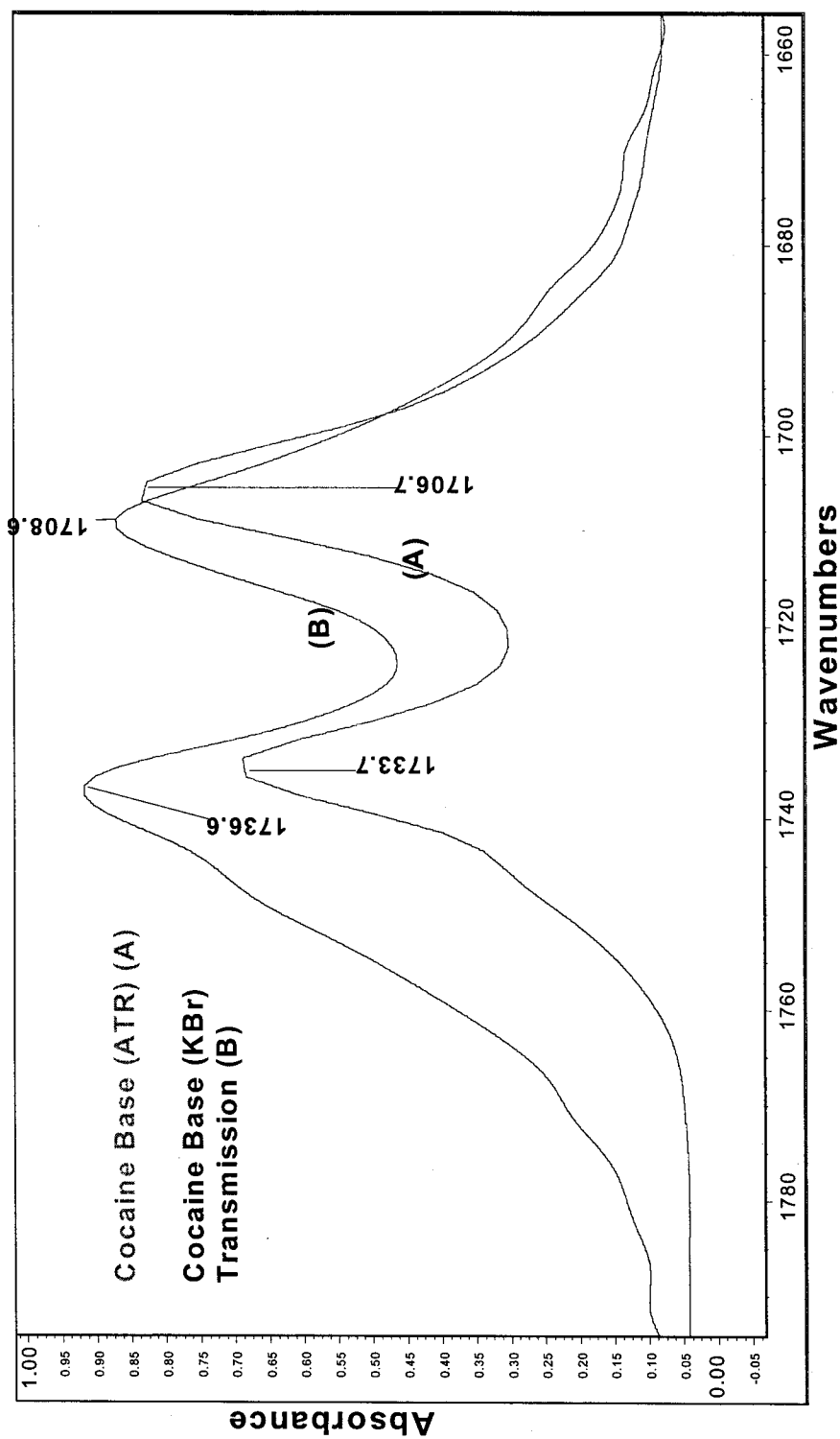
FIG. 4—*The carbonyl band absorption of cocaine HCl showing the difference between ATR and absorbance spectra.*

Theory

The theory of the reflection and refraction of radiant energy at the boundary between two materials of different refractive index is well established and will not be repeated. However, some specific details about internal reflection are important to understanding ATR spectra. The attenuation of the total internal reflection is what produces ATR spectra. Internal reflection occurs when radiant energy, traveling through a high index of refraction (n_h) material, is incident on the interface of a lower index (n_l) material. Total internal reflection occurs when the incidence angle exceeds a specific value. The critical angle (α_c) is the angle of incidence where the radiant energy is refracted into the low index media at 90°. If the lower refractive index material is nonadsorbing, then all radiant energy incident on this interface at angles greater than the critical angle is totally reflected. If the lower index media absorbs radiant energy, then the intensity of the internally reflected energy is reduced. In an internal reflection apparatus, the higher index medium is known as the internal reflection element (IRE) or the ATR crystal. The sample is the lower index medium. Generally, the IRE-air interface is used when collecting the background.

The internal reflectance can be calculated using the Fresnel equations after substituting the complex refractive index. In the following expressions, R_s and R_p are the reflectance for the s (perpendicular) and p (parallel) polarization states, α is the angle of incidence, $I_o = 1$ is the incident radiant intensity, $A(\lambda)$ is the absorption index, $n_h(\lambda)$ is the refractive index of the IRE, $n_l(\lambda)$ is the refractive index of the sample and i is the square root of -1.

$$R_p = \left(\cfrac{ \{n_l(\lambda) - i A(\lambda)\}^2 \cos\alpha - n_h(\lambda) [\{n_l(\lambda) - i A(\lambda)\}^2 - n_h(\lambda)^2 \sin^2\alpha]^{1/2} }{ \{n_l(\lambda) - i A(\lambda)\}^2 \cos\alpha + n_h(\lambda) [\{n_l(\lambda) - i A(\lambda)\}^2 - n_h(\lambda)^2 \sin^2\alpha]^{1/2} } \right)^2$$

$$R_s = \left[\frac{ n_h(\lambda)\cos\alpha - [\{n_l(\lambda) - i A(\lambda)\}^2 - n_h(\lambda)^2 \sin^2\alpha]^{1/2} }{ n_h(\lambda)\cos\alpha + [\{n_l(\lambda) - i A(\lambda)\}^2 - n_h(\lambda)^2 \sin^2\alpha]^{1/2} } \right]^2$$

The internal reflectance value, measured at any single wavelength, depends on both sample and apparatus dependent factors. The sample dependent factors are the refractive index and the absorption index. The apparatus dependent factors are the index of refraction of the IRE, polarization, and the angle of incidence α. Internal reflection is more dependent on refractive index factors than are absorption spectra recorded in transmission.

Additional factors effecting the reflectance values are the depth of penetration of the electromagnetic field into the sample and the area of the sample in optical contact with the IRE. Liquid samples normally make full contact with the IRE's surface. Solid samples need some force to bring them into full contact. The force applied to solid samples should be minimal to avoid possible spectral artifacts. The electromagnetic field penetrating into the sample is commonly referred to as an evanescent wave. Using the following formula, the depth of penetration (D_p) can be calculated.

$$D_p = \lambda / \{ 2\pi [n_h^2 \sin^2\alpha - n_l^2]^{1/2} \}$$

In internal reflection measurements of ATR spectra, the depth of penetration has been accepted as the average effective sample thickness. Since D_p is directly proportional to the wavelength of the incident radiation, the effective thickness is greater at longer wavelengths (smaller wavenumbers). Most commercial spectrometer software provides ATR correction routines that adjust absorbance values for this wavelength factor. However, these ATR correction routines do not account for the sample's refractive index, dispersion or angle of incidence.

Results

The differences between transmission spectra and ATR spectra of cocaine are a direct product of the differences in the measuring processes. The differences in the spectral data, shown in Figs. 3 and 4, can be understood qualitatively by reviewing the internal reflection factors.

Internal reflectance values depend on the refractive index of both the IRE and the sample. The refractive index of both the IRE and
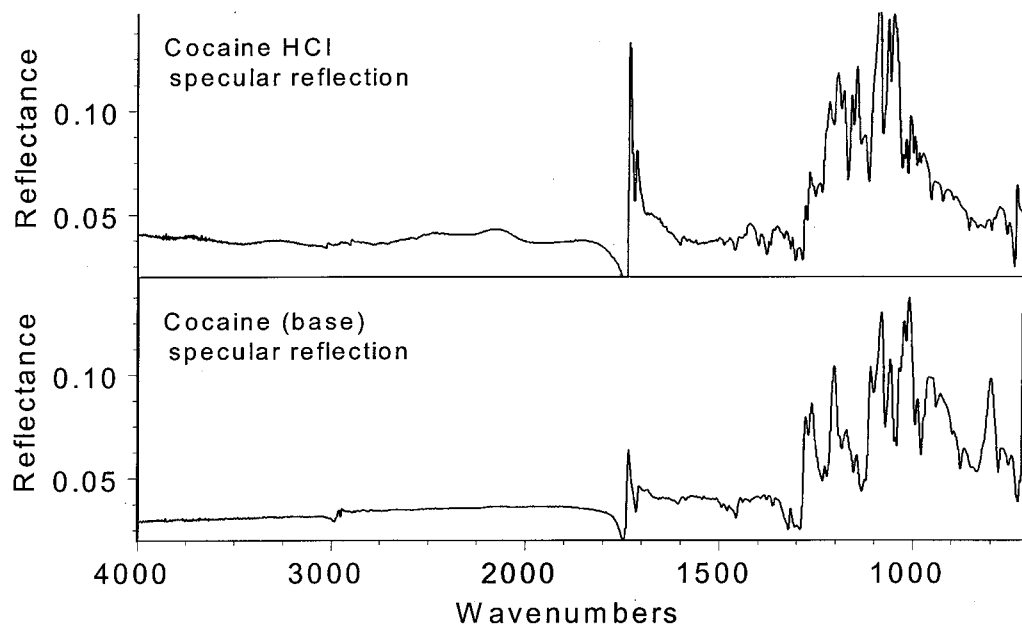

FIG. 5—*Specular reflection spectra of cocaine (base) and cocaine HCl recorded from the surface of single crystals.*

the sample vary with wavelength, an optical property of materials known as dispersion. In wavelength regions where the sample absorbs energy, dispersion is anomalous, varying in a complex, discontinuous way.

Dispersion can be calculated from specular-reflection spectra using the Kramers-Kronig transformation. Specular reflection spectra of cocaine (base) and cocaine hydrochloride are reported in Fig. 5. The specular reflections from the surface of single crystals of these drugs were recorded using an infrared microspectrometer and dispersion of the refractive index calculated. The dispersion data, in the region of the carbonyl absorption, are reported in Fig. 6. This illustrates anomalous dispersion. Since the penetration depth is determined in part by the index of refraction, the effective sample thickness follows the anomalous dispersion. The variations of the penetration depth, calculated from the refractive index dispersion for cocaine base, are found in Fig. 7.
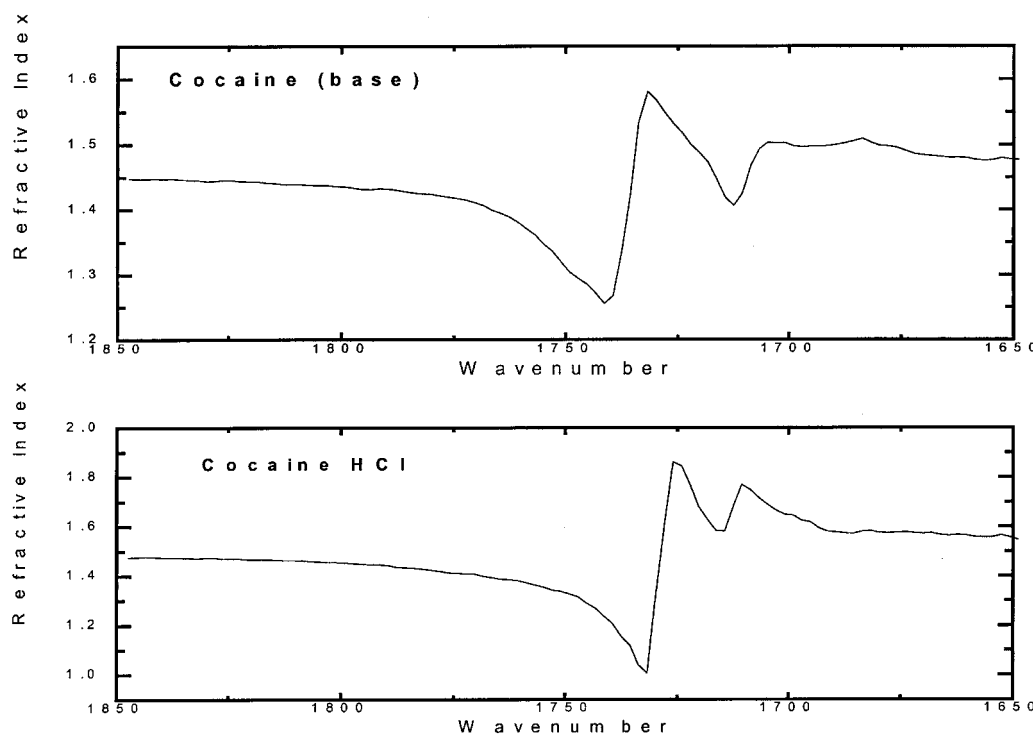

FIG. 6—*Dispersion values for cocaine (base) and cocaine HCl calculated from the specular reflection spectra using the Kramers-Kronig transformation.*
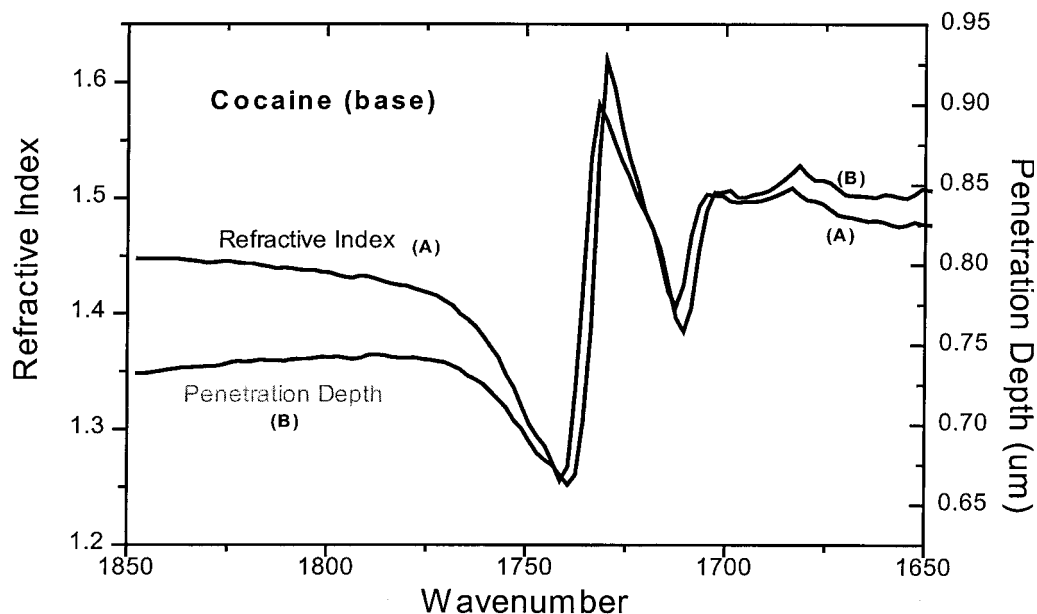

FIG. 7—*The penetration depth and the refractive index for cocaine (base) are plotted to show the change in effective sample thickness with wavelength of the incident radiation.*

This shows how the effective sample thickness varies with wavelength.

The experimentally recorded reflectance values in ATR spectra are a direct product of the absorption and effective thickness (or penetration depth). The changes in absorbance values caused by the thickness variations are illustrated by normalizing the effective thickness in the ATR spectrum. The thickness normalized ATR spectra of cocaine (base) and cocaine salt are shown in Figs. 8 and 9. The dispersion effects are greater for cocaine salt than they are for the cocaine base. Correspondingly, there are larger differences between the transmission and ATR spectra for cocaine salt. The normalized ATR spec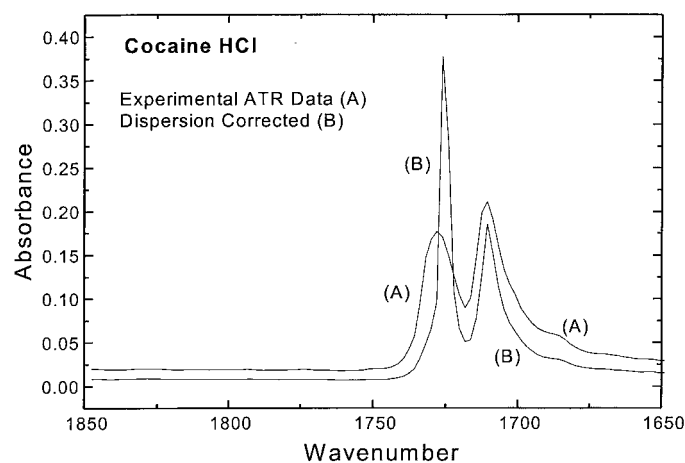tra have band intensities and locations that are different from the experimental ATR spectra. Because of experimental variables and approximations in these calculations, quantitative agreement is not expected, but the qualitative effect of the anomalous dispersion are demonstrated.

Discussion

The absorption and refractive index are the two major optical properties of dielectric materials. Infrared absorption spectra are very useful for qualitative and quantitative analysis of molecular compounds. In recording infrared spectra, both the absorption and refractive index effect the experimentally recorded absorbance values. If different methods are used to record infrared spectra, then small differences can occur. While transmission and internal reflection are reliable methods, the experimental spectra they generate are not identical. Refractive index has a minor effect on the absorbance values measured by transmission. However, refractive index plays a significant role in the absorbance values measured by internal reflection. Although the differences are small, they can cause confusion when directly comparing ATR to transmission spectra. Since direct spectral comparisons are commonly used in forensic evidence analyses, it is important to know that the cause for minor differences is optical and not compositional.

When using the internal reflection method to collect ATR spectra, refractive indexes are an important factor in the effective sample thickness. Since the value of the refractive index changes abruptly in the region of an absorption band, so does the effective sample thickness. This change in thickness causes shifts in the peak position of ATR spectral absorption. These shifts are optical effects caused by the anomalous change in refractive index. The ATR spectra of cocaine (base) and cocaine salt, in the region of the carbonyl absorption band, qualitatively demonstrates the effect of dispersion on internal reflection measurements.

FIG. 8—*The experimental ATR spectrum and the dispersion corrected ATR spectrum of cocaine (base), illustrating the changes in absorbance and peak positions caused by anomalous dispersion.*
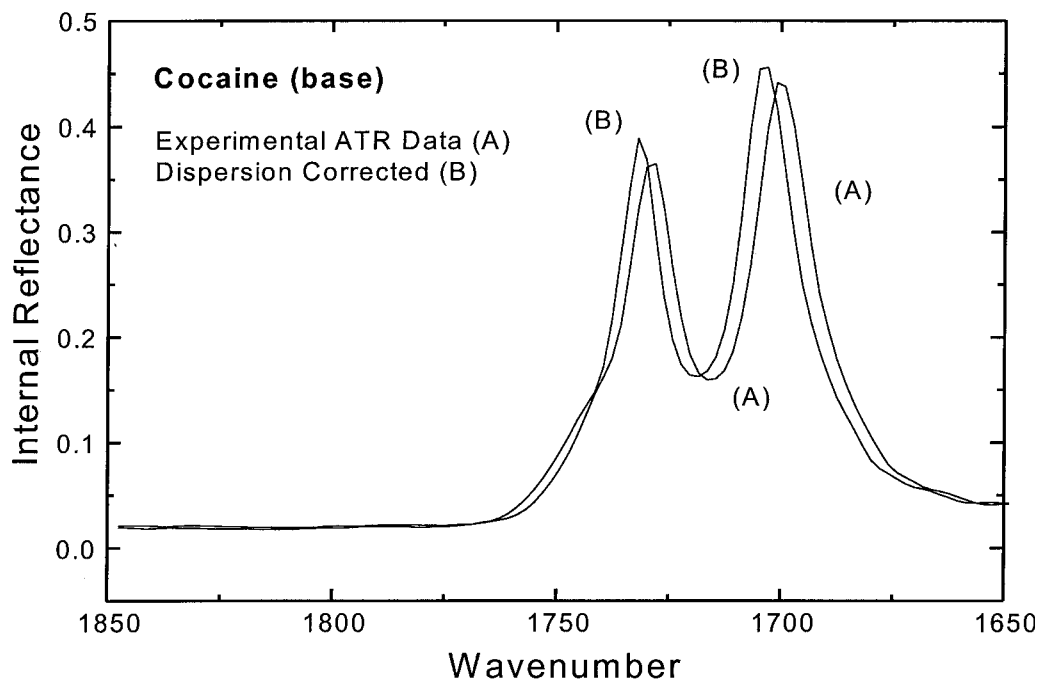

FIG. 9—*The experimental ATR spectrum and the dispersion corrected ATR spectrum of cocaine HCl, illustrating the changes in absorbance and peak positions caused by anomalous dispersion.*

Either the transmission or internal reflection method can be used to evaluate forensic evidence. When ATR spectra are searched using a transmission spectral library, reliable matches can be made. The match index of this search will not be as high as when transmission spectra are used, but qualitative analysis is possible. When making a direct comparison of spectra, it is important that the spectra are collected using the same method. Internal reflection is becoming the most popular method for infrared analysis. While ATR spectra are similar to transmission spectra, they are not identical. New libraries of ATR spectra are needed. Transmission and internal reflection are two different methods for infrared analysis. While similar, each method produces unique infrared data. Transmission produces absorption spectra; internal reflection produces ATR spectra.

Acknowledgment

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