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Attenuated Total Reflectance

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Attenuated Total Reflectance (ATR)

Theory of ATR

Data Collection for ATR

Troubleshooting Spectral Results

ATR Correction

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ATR Microscopy

- **Use Micro ATR crystal to direct beam**
- **Surface Sampling (0.2 - 4.0 μm)**
- **Available with ZnSe, Ge, diamond and Silicon crystals**
- **Ideal for "non-destructive" analysis of samples**
- **Sampling area = Aperture size/Refractive Index of the crystal**

There are differences between macro-ATR and micro-ATR. In macro analysis there is much more variation and flexibility.

Microscopically the crystals are much smaller and only provide a one point analysis.

Due to the smaller sampling size, the intensity of the interaction tends to be smaller making it more susceptible to noise.

Because of the small surface area being analyzed, the amount of pressure being applied to the sample needs to be monitored closely so as not to damage the crystal.

Slide-On ATR

With the slide-on ATR there are concessions that need to be made.

Due to the bracket that must be installed, there will be a slight loss in working distance when using the objective for non-ATR sampling. The implication is that larger samples may not fit into the available space needed for focusing.

Only Ge and Si crystals are available for this accessory. With both of these materials having a relatively high refractive index, this is a good approach for strongly absorbing samples.

No view-through is possible with the design of this device. Samples are first positioned under the objective with the crystal removed, using the normal 15x view. The crystal is then slid into position and contact is made by raising the stage. The X, Y position of the crystal must match that of the 15x view mode. This requires alignment of the crystal once after installation.

Also, due to the need for focusing the light on the ATR crystal there needs to be an adjustment to the focal point of the crystal. This may be accomplished through the use of the compensation ring or by adding shims to the mounting process so the crystal is positioned at the focal point of the 15x objective.

ATR - How It Works

η_s

η_c

For ATR to work, $\eta_c > \eta_s$

Total Internal Reflectance

ϕ Critical Angle
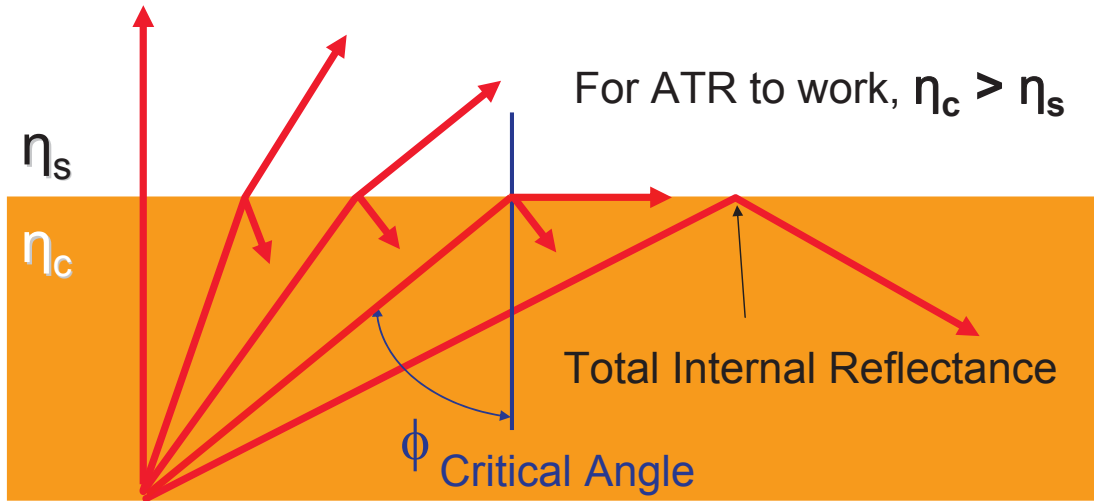
Where:

η_c = Refractive Index of Crystal

η_s = Refractive Index of Sample or Air

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When light transitions between materials of different refractive index, refraction occurs. If the transition is from a high refractive index material to a low refractive index material, the incidence angle, or internal angle, is smaller than the transmitted, or external angle in the low refractive index material.

When the internal angle reaches a critical level, the external angle is 90 degrees and the propagation of light is parallel to the surface of the interface between the sample. This is the critical angle.
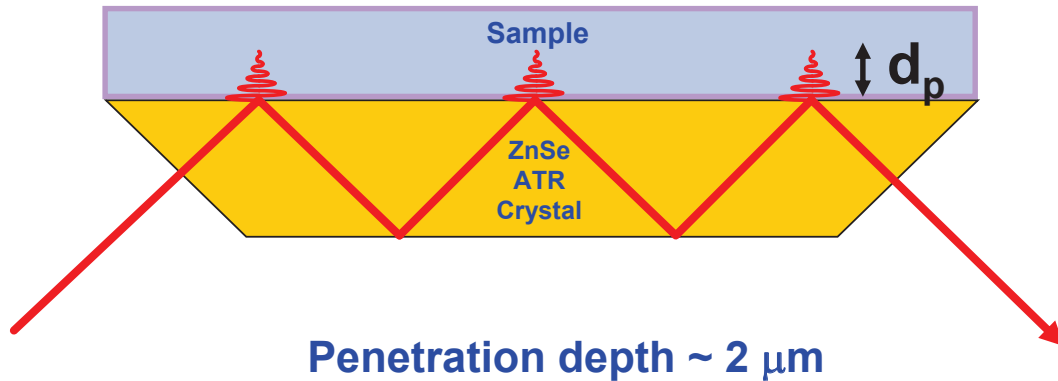
With ATR, the infrared beam enters a crystal at an angle that is greater then the critical angle of the crystal. This causes the IR energy to internally reflect off the parallel surfaces of the crystal. Since the crystal has a refractive index much higher then the refractive index of the sample, the beam never really leaves the crystal.

The critical angle can be calculated:

$$\phi_c = \sin^{-1}\frac{\eta_s}{\eta_c}$$

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For ATR effect to work $\eta_{crystal} > \eta_{sample}$

Sample

$\updownarrow$ **d_p**

ZnSe ATR Crystal

Penetration depth ~ 2 μm

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In the ATR experiment, the light propagates inside of an infrared transparent internal reflecting element. The sample is typically applied to the top of the ATR crystal. As the light bounces off the inside surface of the crystal, a small amount of radiation, an evanescent wave, will interact with the sample.

This schematic of the ATR crystal from a macro ATR accessory, shows the evanescent wave, the path and number of reflections for IR radiation.

Based on the refractive index of the ZnSe, the average penetration depth for a single reflection was 2.0 μm. In a multi-reflection ATR crystal the penetration depth would be multiplied by the number of interactions with the sample to calculate the pathlength.

Depth of Penetration*

$$\text{Pathlength} = \Sigma \, (d_p)_n$$

Depth of penetration at each reflection point is controlled by many factors and is wavelength dependant.

Longer wavelength waves penetrate deeper than shorter wavelength waves.

*In a spectrum, an increase in the depth of penetration translates to increased absorbance intensity

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Depth of penetration can also be considered to be the depth of interaction with the sample. This is equivalent to the pathlength (sample thickness) in a transmission experiment.

The wavelength dependence of this interaction produces, in effect, a variation in sample thickness across the spectrum.

ATR Spectral Influences

1. **Distortion of relative peak intensities in ATR compared to transmission data**

   - **Depth of penetration is influenced by wavelength**
   - **Relative intensity of bands in ATR spectrum increases with wavelength**
     resulting in poor match values in library Search

2. **Shift of absorption bands in ATR compared to transmission data**

   - **Absorption bands shift to lower wavenumbers**
   - **Will affect Search results**
   - **Deviation from Beer's Law caused by non-polarization effects**

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While the ATR technique yields different spectral data than the transmission technique, the data is no more or less valid than transmission, IT'S JUST DIFFERENT!!

Depth of Penetration Relationships

$$d_p = \frac{\lambda}{2\pi\eta_c \sqrt{\sin^2\theta - \left(\dfrac{\eta_s}{\eta_c}\right)^2}}$$

λ = wavelength of the energy
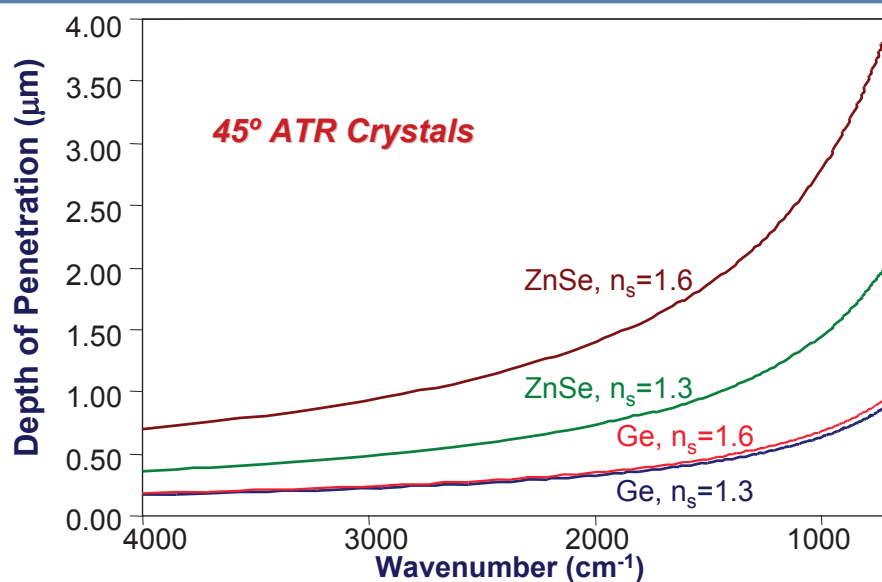θ = angle of incidence
η_s = refractive index of the sample
η_c = refractive index of the crystal

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Remember the relationships rather than the equation.

d$_p$ vs. Wavenumber

| η_c	η_s	d_p (µm)		
		4000 cm^{-1}	2000 cm^{-1}	700 cm^{-1}
2.4	1.6	0.70	1.41	4.04
2.4	1.3	0.36	0.73	2.09
4.0	1.6	0.17	0.34	0.98
4.0	1.3	0.16	0.32	0.91

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This shows both the relationships between wavelength vs. pathlength (depth of penetration) and crystal refractive index vs. pathlength.

The maximum depth of penetration can be seen for the ZnSe crystal in the lower wavenumber region of the spectrum. This barely reaches the 4 micron depth with a high refractive index sample.

The two samples shown in this example represent the extremes seen for common organic compounds. They range is from 1.3 in hydrocarbons up to 1.7 for halogen substituted materials.

Depth of Penetration - Changing Crystal
(Changing Refractive Index)
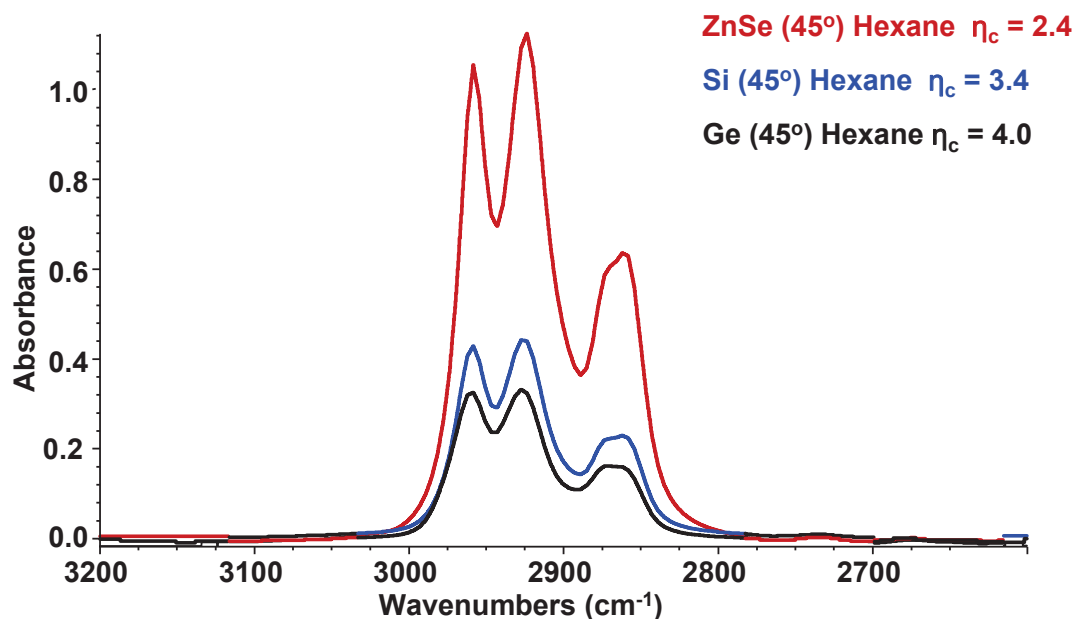
Infrared spectra of hexane were obtained using three different ATR crystal materials at the same angle of incidence (45 degrees). The spectra above illustrate that the intensity of the absorption of the hexane increases as the depth of penetration increases. If a quantitative result was desired, ZnSe would be a poor choice since the intensity of the bands shown are greater than 0.8 absorbance units and therefore outside the typical linear range of the FT-IR spectrometer. Silicon or germanium would provide similar results.

	η	d_p
Germanium	4.0	0.66
Silicon	3.4	0.85
Zinc Selenide	2.4	2.0

The above table illustrates that as the refractive index (η) of the crystal increases, the depth of penetration (d_p) decreases. The d_p value shown assumes the angle of incidence was 45 degrees for all the crystals, the sample refractive index was 1.5, and the wavelength of the radiation was 10 μm (1000 cm^{-1}). By varying the crystal material you may customize the depth of penetration to your experimental needs. An alternate method to vary the depth of penetration is to change the angle of incidence.

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Properties of Internal Reflection Elements (IREs)

Material	Useful Spectral Range (cm^{-1})	Refractive Index (η) (@ 1000 cm^{-1})	Depth of Penetration (d_p) (@ 45° & 1000 cm^{-1})	Hardness (Knoop)	Notes
Diamond	25,000 – 100 reduced signal 2500 - 1667	2.4	2.01 µ	7000	Good for most samples, extremely caustic or hard samples, aqueous samples, very hard, chemically inert, water insoluble, attacked by $K_2Cr_2O_7$ & conc. H_2SO_4
ZnSe	15,000 - 650	2.4	2.01 µ	137	General use, good for aqueous samples, very resistant to most solvents, water insoluble, scratches easily, attacked by strong alkaline solutions
AMTIR GeAsSe	11,000 - 725	2.5	1.77 µ	170	Good for aqueous samples, very resistant to acidic solutions, water insoluble, scratches easily, attacked by strong alkaline solutions
Silicon	8,900 – 120 reduced signal 1,500 - 360	3.4	0.85 µ	1150	Good for hard samples, aqueous samples, resistant to basic solutions water insoluble, withstands thermal & mechanical shock, attacked by HF & HNO_3
Germanium	5,500 - 675	4.0	0.66 µ	550	Good for most samples, especially strong absorbing samples, such as dark polymers, aqueous samples, water insoluble, hard & brittle, attacked by H_2SO_4 & aqua regia, temperature sensitive

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This slide illustrates the differences between ATR crystal material. This information can be used to help evaluate which crystal material will best suit the needs of the experiment.

When the samples are heavily absorbing (as in carbon filled rubber materials) a germanium crystal will better limit the absorption. For weakly absorbing materials (like hydrocarbons) the ZnSe or diamond crystals are better choices.

The hardness of each crystal should also be noted when selecting the appropriate crystal for the experiment. If the sample is as hard or harder than the crystal, the risk of damaging the crystal when brought in contact should be considered.

AMTIR stands for Amorphous Material Transmitting Infrared Radiation (really!). This material is not available on microscopes.

Recommended Cleaning

Do **NOT** use Kimwipes® to clean the crystal surfaces. This will cause severe scratching of the surfaces of all materials except diamond.
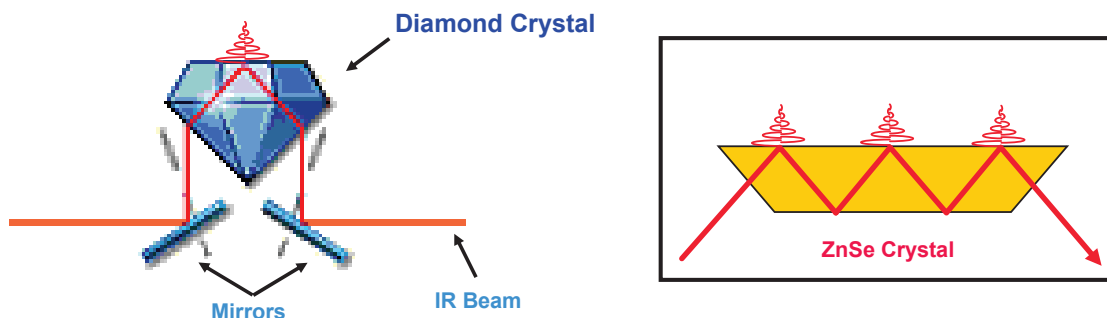
Use lens cleaning tissue or a q-tip to wipe surface.

Use isopropyl alcohol (IPA) or methanol as cleaning agents. Do **NOT** use large quantities of solvent

Do NOT use acetone as this may attack the epoxy holding the crystal.

Avoid strong acids and bases.

ATR - Single Bounce vs Multi-bounce

Diamond Crystal

Mirrors

IR Beam

ZnSe Crystal

Single Bounce ATR

- Small sampling area

- Use for strong absorbers

- Solid samples

Multi-Bounce ATR (HATR)

- Broad sampling area provides greater contact with the sample

- Use for weak absorbers or dilute solutions
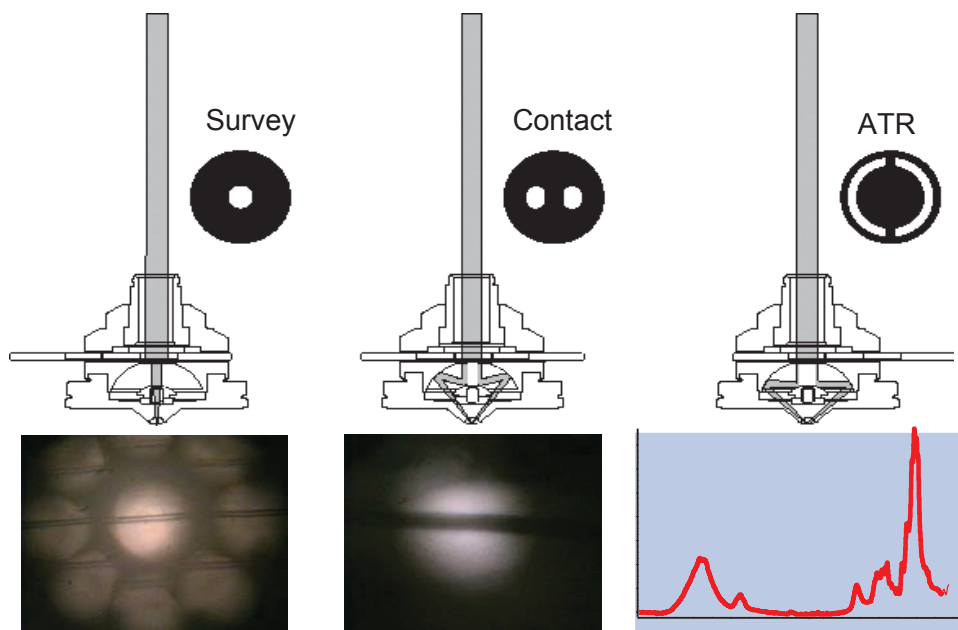
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In the ATR experiment, the light propagates through the inside of an infrared transparent internal reflection element. The sample is typically applied to one side of the ATR crystal. As the light bounces off the inside surface of the crystal, a small amount of radiation will penetrate into the sample via an evanescent wave.

The schematic of the ATR crystal showing the evanescent wave also showed the path and number of reflections for IR radiation.

Based on the refractive index of the ZnSe, the average penetration depth using a single reflection ATR was 2 μm. In a multi-reflection ATR crystal the penetration depth would be multiplied by the number of reflections to calculate the pathlength. The ATR experiment theoretically maximizes out at about 200 μm total pathlength.

All the microscope ATR objectives are of the single bounce design.

Diamond ATR Objective - Modes

Survey   Contact   ATR

Can only view through Diamond and Zinc Selenide

ATR objectives are preferred over slide-on ATR for small samples or irregularly shaped samples that could be moved out of the beam by the pressure of the ATR.

On the left of the slide, we see the diamond ATR in survey mode. The image is the view through the center of the ATR crystal! In the center of the image is a fiber. The ghost images are back reflection from the diamond facets. The field stop on the Continuum eliminates most of the back reflected light and gives a clean image.

After selecting the fiber we are interested in, the slider is moved into contact mode. The middle position shows Contact mode. As the ATR is brought into contact with the fiber, the critical angle is exceeded, the light is not reflected back and the sample appears dark. This is called "wetting".

The slider is then moved to the ATR mode and the spectrum of the fiber is collected. Note that the phonon bands from the diamond are ratioed out leaving only a slight increase in noise near 2300 cm^{-1} as an artifact. ZnSe crystals may also be used but diamond is more durable, provides a clearer visual image, and has spectral coverage below 750 cm^{-1}.

Collection Parameters

	ATR
Scans	32 – 64
Resolution	4, 8
Final Format	% Reflection or Log1/R
Bkg. Material	Clean crystal with ***nothing*** making contact
Spectral Range	4000 – Cutoff of Crystal

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The ATR experiment is considered to be a reflection analysis. As with all reflection techniques on the microscope, less energy is available at the sample due to the design of the system.

Note the Y-axis units are reflection units. This is the correct format for ATR data.

Peak Shape and Position

$$A = (\log_{10} e) \frac{\eta_s \varepsilon_0^2 d_p}{2 \eta_c \cos \phi} \, \alpha$$

α = absorption coefficient
ε_0 = electric field at interface
ϕ = angle of incidence
η_s = refractive index of the sample
η_c = refractive index of the crystal

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The relative intensity of bands in an ATR spectrum increases with wavelength. If η_S/η_C is constant or a smooth function over the frequency range of observations, the penetration depth increases linearly with wavelength. Most spectroscopic software packages include an ATR correction function that simply applies a linear ramp to the ATR spectrum in order to approximate the relative band intensities that would be found in a transmission experiment.

The refractive index of the sample, n_S, changes sharply around absorption bands, the wavelength dependence of d_p is further modified according to $(\sin^2\phi - (\eta_S/\eta_C)^2)$ -1/2.

The second characteristic feature is the shift of absorption bands to lower frequency. This equation indicates, the ATR spectrum is influenced by the refractive index of the sample.

According to the first equation, penetration depth, d_p, depends on n^2. Thus, instead of α, (n^2 d_p α) determines the absorption band positions. Because of the dispersion of the refractive index, (n^2 d_p α) always introduces a band shift to lower frequency and this shift becomes particularly noticeable for strong bands.

Consequently, the presence of absorptions in the infrared spectrum represents a change in the refractive index of the sample across the band and results in the shift to lower frequency.
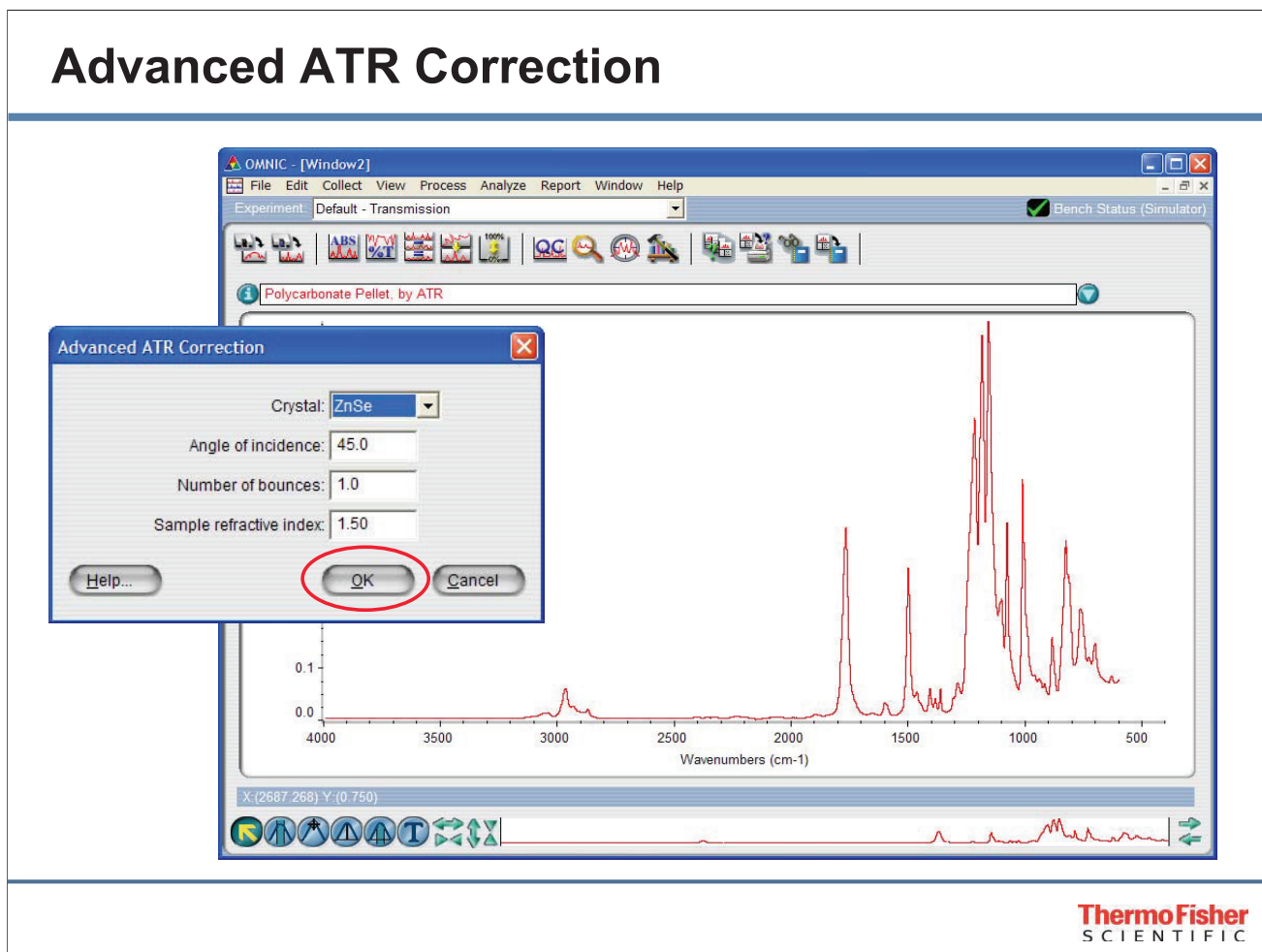
Advanced ATR Correction
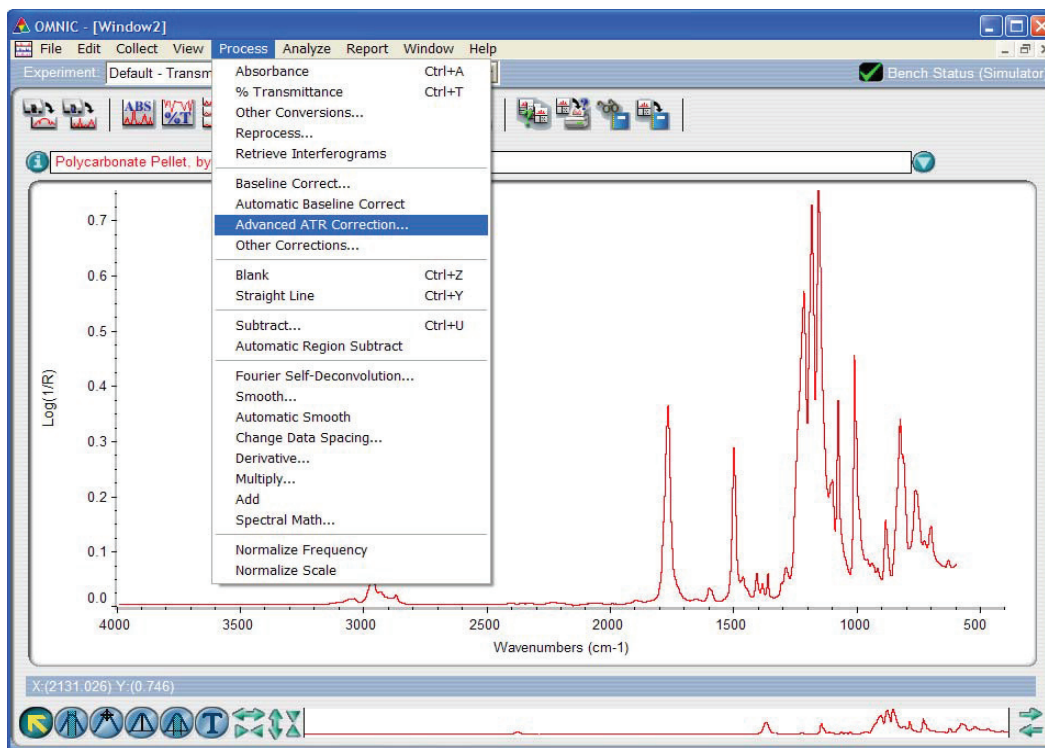
The advanced ATR correction utilizes both equations to bring the spectral data into a format that more closely resembles data that would have been collected in a transmission experiment.

This is useful when comparing data collected to transmission data. either by visual inspection or through search techniques.

Advanced ATR Correction

The Advanced ATR correction requires all the information contained in the two equations shown previously.

All of the accessory information is readily available, crystal type (hence refractive index), angle of incidence and number of interactions with the sample.

In many cases, the experiment is being performed to identify an unknown material. The request for the sample refractive index cannot be fulfilled because it is not known. If this is the case, the entry of a general refractive index will still produce a corrected spectrum, but the result of the correction will not be as accurate.

Most organic materials have refractive index values that range from about 1.3 to 1.7. Using an average 1.5 refractive index for organic unknowns will yield a correction that will be very close to the ideal.
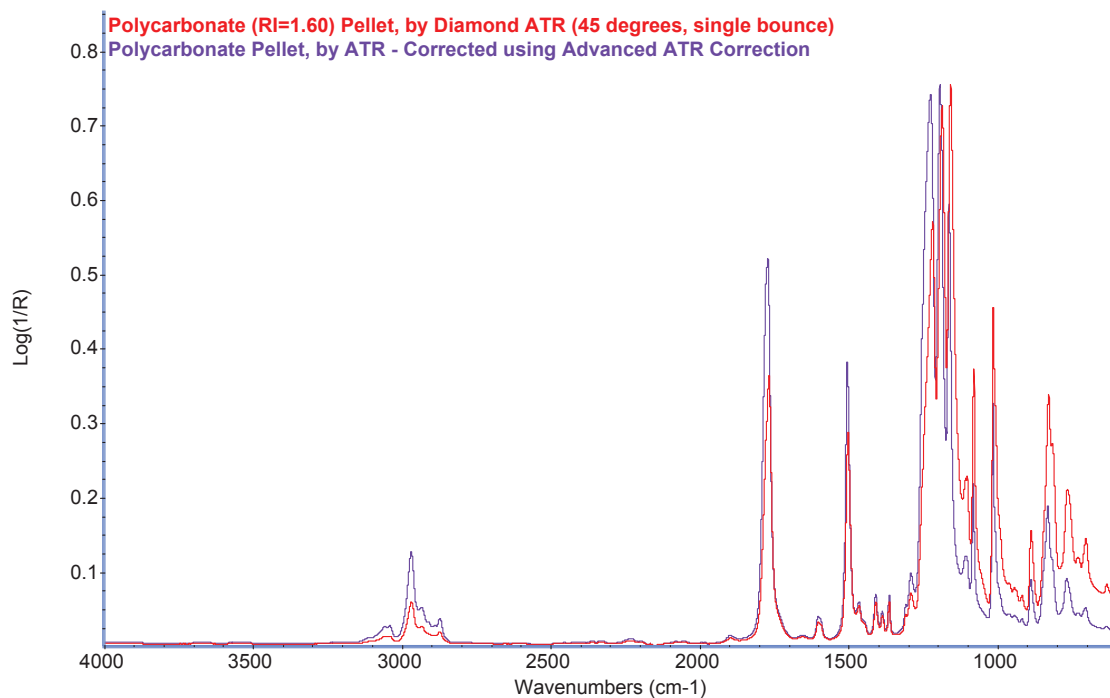
Remember – A corrected ATR spectrum is still only an approximation to the data that would have been collected in transmission mode on the material analyzed!!!

Advanced ATR Correction - HELP
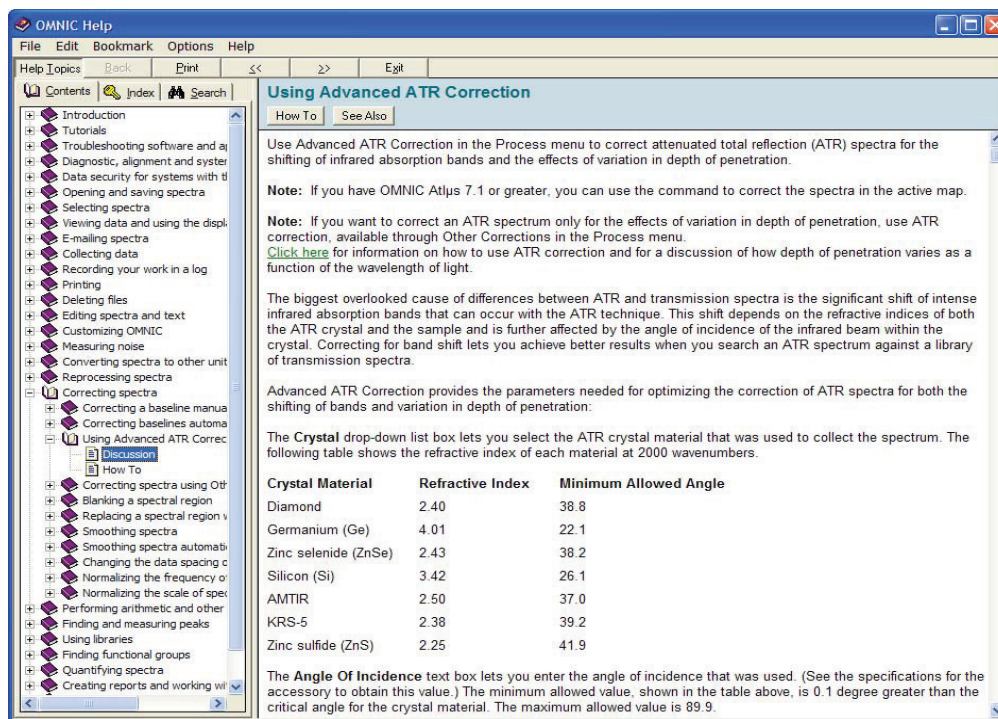
A quick reference is available through the Help button in the software. Refractive index and other useful information are also included.

Advanced ATR Correction Result

Polycarbonate (RI=1.60) Pellet, by Diamond ATR (45 degrees, single bounce)
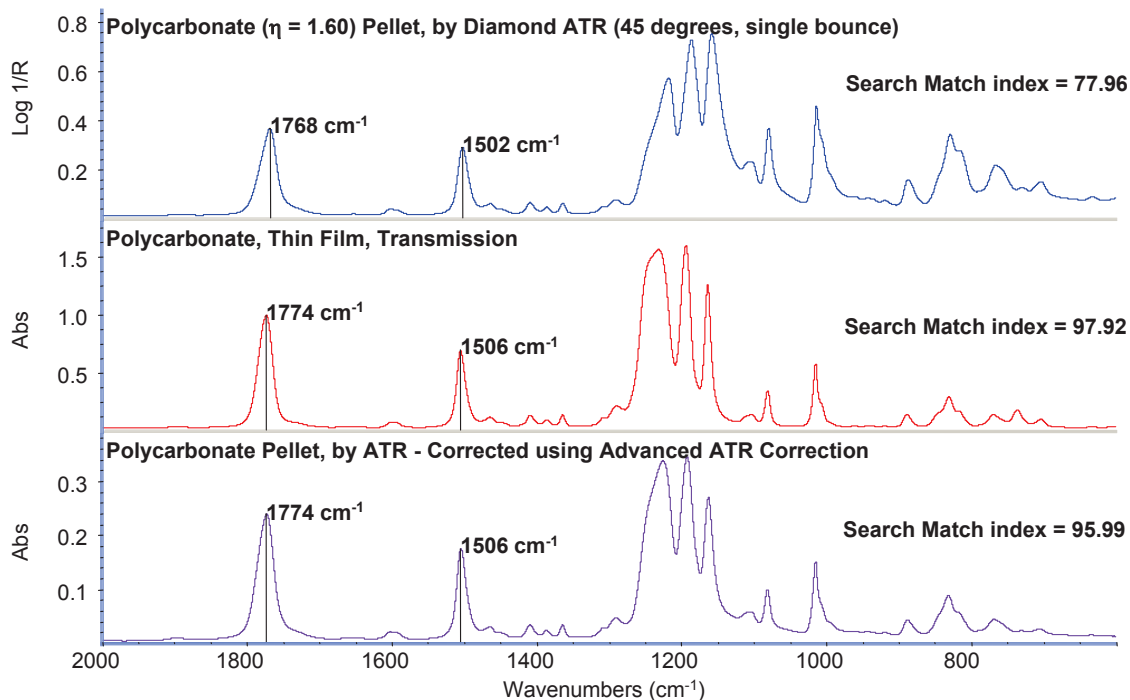**Polycarbonate Pellet, by ATR - Corrected using Advanced ATR Correction**

The intensity of the peaks before and after the correction can be seen in this data. The wavelength dependence of the Depth-of-Penetration can be seen here.

After the correction –

> The features that occur at the shorter wavelengths (higher frequencies) have been intensified.

> The features that occur at the longer wavelengths (lower frequencies) have been minimized.

Advanced ATR Correction Result

Polycarbonate (η = 1.60) Pellet, by Diamond ATR (45 degrees, single bounce)

Search Match index = 77.96

1768 cm^{-1} 1502 cm^{-1}

Log 1/R

Polycarbonate, Thin Film, Transmission

Search Match index = 97.92

1774 cm^{-1} 1506 cm^{-1}

Abs

Polycarbonate Pellet, by ATR - Corrected using Advanced ATR Correction

Search Match index = 95.99

1774 cm^{-1} 1506 cm^{-1}

Abs

Wavenumbers (cm^{-1})

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Expanding the fingerprint region (2000-600 cm^{-1}), the effects on peak positions can be seen relative to the transmission data collected on this sample.

Note that after applying the correction, the peak positions and absorbance values better match those of the transmission data.

When searching both the corrected and uncorrected ATR data against a spectral library that was assembled from transmission data, the match quality increased almost to that of the data collected in transmission mode.

Attenuated Total Reflectance (ATR) - Review

Theory of ATR

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