

Raman spectroscopy is an ideal Process Analytical Technology (PAT) to ensure Quality by Design (QbD) for drug development, process development, and manufacturing. It is also ideal for optimizing the synthesis of a new drug, gaining process understanding through real-time monitoring of reactors and crystallizers, and verifying raw materials and assuring product quality. Here we describe these applications with a focus on drug synthesis and process optimization using our *RamanPro* (Figure 1), a fluorescence free and x-axis stable Fourier transform Raman spectrometer.

Drug Development – Once a potentially new drug is identified, the method to synthesize the drug is developed. Raman spectroscopy is ideal for monitoring reactant, intermediate and product concentrations, determining pathways, kinetics, mechanisms, end-points, and yields for a variety of reaction types, such as Diels-Alder, Fischer esterification, Grignard, and hydrogenation.



Figure 1. RTA's *RamanPro*.

Interferometer-Based Raman
Advantages
No Fluorescence Interference
Wavenumber Stability (x-axis never shifts)
No Analyzer Re-Calibration
Lab-to-Production Calibration Transfer
Reproducible and Selectable Resolution (4-32 cm ⁻¹)
No Wavelength Dependent Resolution
Full Spectrum Every Scan
Temperature & Vibration Immune (RTA Only)
Dispersive-Based Raman
Advantages
Relatively Easy to Design & Build
Sensitivity (785 nm ~ 10X 1064 nm Sensitivity)

Fischer esterification is used in the synthesis of many drugs, such as benzocaine, but this reaction is more often used as an intermediate step to protect carboxylic acid groups, while functionalizing phenyl rings. As an example of this important pharmaceutical reaction, the esterification of benzoic acid was performed to produce methyl benzoate (Figure 2A). The reactant and product have unique spectra, with peaks at 780 and 817 cm⁻¹, respectively, which are ideal for real-time monitoring (Figure 2B). An emersion probe in a batch reactor was used to collect spectra every 45 seconds using 0.5W of 1064 nm laser excitation. *Rate constants, activation energy, end-point and yield were easily determined*, while optimizing the effects of reactor temperature, catalyst concentration and type (Figure 2C).

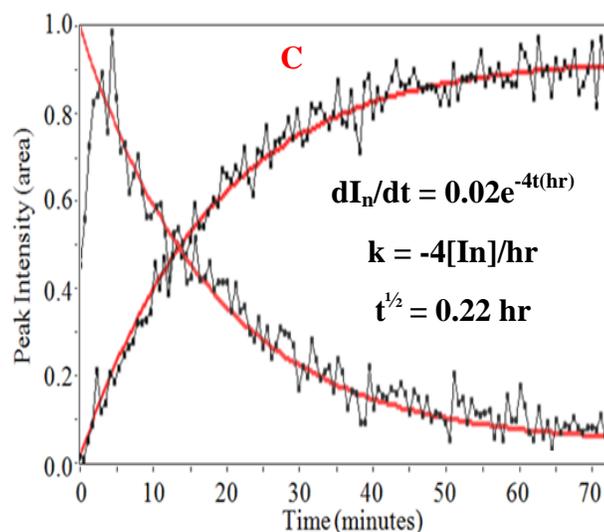
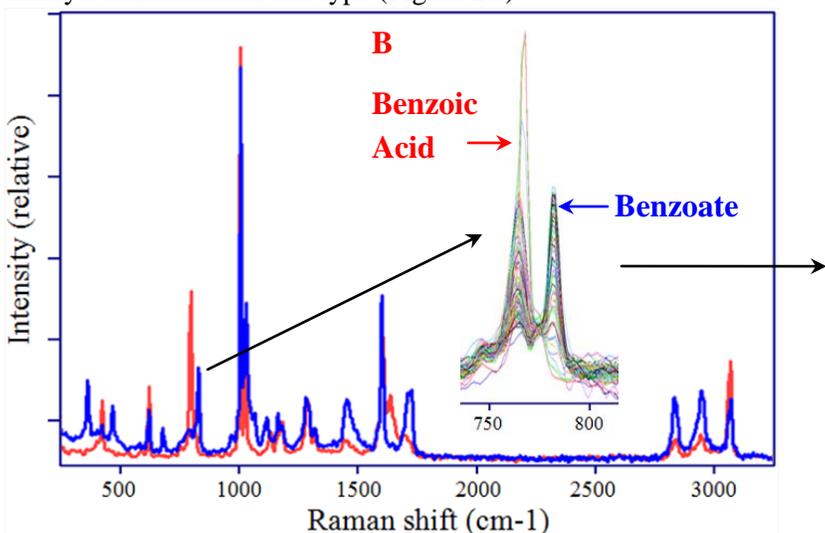
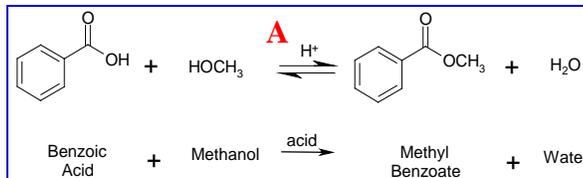
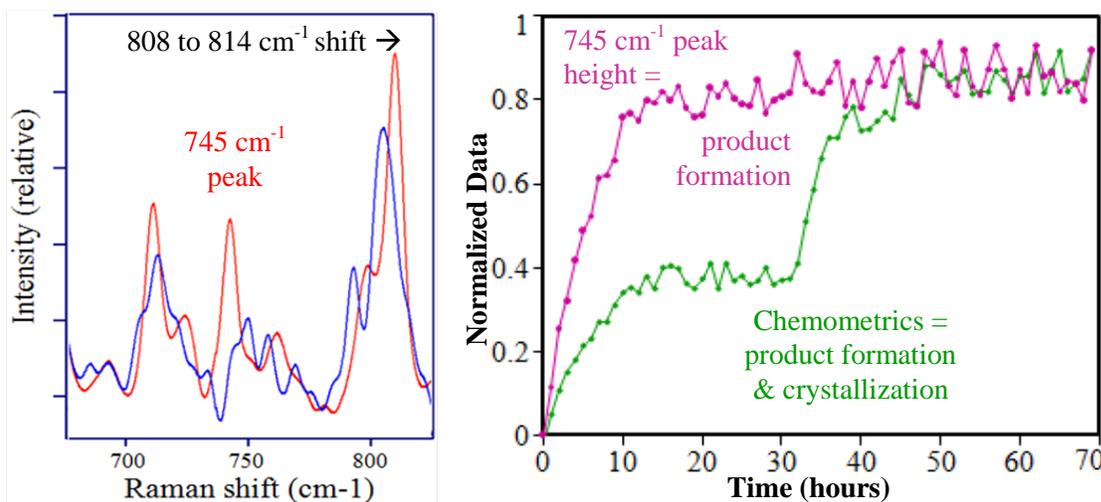


Figure 2. A) Esterification of Benzoic Acid, B) Raman spectra of **Benzoic Acid** and **Benzoate**, and C) Plot of their respective peak intensities as a function of time. 90% yield achieved at 60 °C in 70 minutes.

Process Understanding and Optimization – Once a synthesis route for a drug has been established, the reaction is scaled-up to the production level. RTA's *RamanPro* is ideal for process understanding, optimization, monitoring, and ultimately, control. Since the x-axis of all interferometer-based Raman spectrometers never shift (as opposed to dispersive spectrometers that do), the same spectral parameters used to optimize the lab-scale reactors, can be used to optimize the pilot-scale, and production-scale reactors. This means that concentration calibration curves used to monitor the reactants, intermediates, and products, as well as complex chemometric models, can be transferred from the lab to production. RTA provides both immersion and remote fiber optic probes (in reactor and through reactor sight glass, respectively) that are tolerant to wide pressure and temperature ranges for challenging production conditions.

Crystallization, often the final step in drug synthesis, is used to separate a drug from a solvent matrix so that it is suitable for final form manufacturing. This process must also be optimized, not only to maximize separation, but in many cases to ensure that the correct polymorph is formed. For some drugs various polymorphs, or crystalline structures, exist with dramatically different solubilities that effect bioavailability, which determines appropriate dosage. Process conditions, such as temperature, mixing rate, and concentrations can affect crystalline formation kinetics, and which polymorph dominates. Again, Raman spectroscopy is well suited for understanding and optimizing process conditions. In fact, drug synthesis and crystallization are often carried out in a single batch reactor. Figure 3A shows the Raman spectra for the initial reactants and the final crystalline product. In this case the product has a unique Raman peak at 745 cm^{-1} suitable for monitoring its formation, while the shift in the peak at 808 to 814 cm^{-1} can be used to monitor crystallization. Although these spectral changes can be used, a chemometrics model that correlates these properties to the entire Raman spectrum allows monitoring both the synthesis and crystallization process with high accuracy and precision (green trace, Figure 3B). *The importance of x-axis stability cannot be overemphasized when using peak shifts or chemometrics.*

Figure 3. A) Raman spectra of the reactant mixture and the crystallized product. The peak height is used to monitor product formation, while the shifts are used to follow crystallization. B) Plot of the 745 cm^{-1} peak height and a chemometrics correlation to both product formation and crystallization (onset initiated by a (temperature drop).



Quality Manufacturing – Quality by design in drug manufacturing begins with verification of the purity of raw materials and ends with the quality of the product. The latter requires assurance that the product, as a gel cap, pill, tablet, etc., contains the correct amount of active, its polymorph (if appropriate), excipient and other additives (e.g. dyes). Dyes are often used to provide product uniqueness for the pharmacist and consumer alike. Verification and quality assurance of drug products containing dyes require 1064 nm laser excitation to avoid fluorescence interference. Figure 4 compares the Raman spectra obtained for a pink Amoxicillin pill obtained using 785 and 1064 nm lasers. In addition to this obvious example, 785 nm excitation causes fluorescence in the majority of full scale processes (due to by-products and contaminants) and many raw materials (due to aging). *The RamanPro is the only process Raman analyzer that employs 1064 nm laser excitation.*

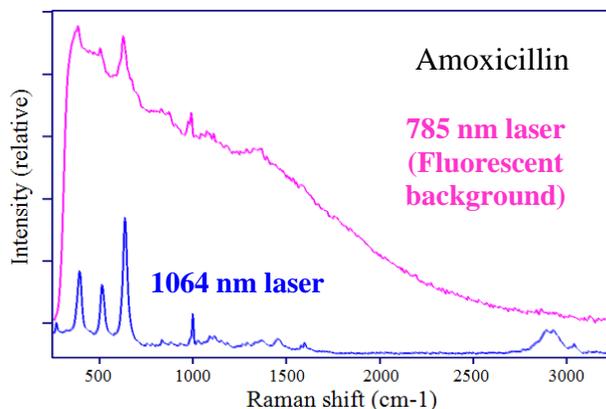


Figure 4. Raman spectra of pink Amoxicillin pill.