

Rapid Screening in Microbiology by FT-IR

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FT-IR spectroscopy has been shown to be an effective tool to classify bacteria, yeast, and fungi at the strain, species, and genus level. Compared to traditional microbiological techniques, IR sampling is less time-consuming. Microbiological applications for FT-IR analysis include a rapid method of identification and screening, quality control for food and pharmaceutical fields, and assessment of growth environment on cell and cell byproduct physiology.

An IR plate-reader for biological samples is desirable for laboratories confronted with high-throughput demands, and the need to save resources through automation. This application note illustrates the use of an in-compartment XY plate reader to analyze yeast and discusses the resource savings involved through accessory automation.



Figure 1. X, Y Autosampler by PIKE Technologies

Experimental Conditions

Yeast suspensions (30 μ l) were pipetted onto a sample well of a 96-well reusable Si plate, and dried at 45 $^{\circ}$ C for 1 h for

subsequent infrared transmission measurements. The 96-well Si plate was loaded into the PIKE Technologies XY Autosampler Accessory, an FT-IR sample compartment automated transmission/diffuse reflection accessory. The spectral range spanned from 4000 to 600 cm^{-1} using a resolution of 4 cm^{-1} . Data collection time was 1 min. For transmission measurements the accessory's integrated DTGS detector is used to maximize the signal-to-noise ratio. Automated plate movement to each sample position is controlled by PIKE AutoPRO software, which also triggers data collection via the FT-IR spectrometer.

Results

Figure 2 shows a typical transmission spectrum of the dried yeast suspension. Some main spectral features include the C-H stretching vibrations from fatty acids between 3100 – 2800 cm^{-1} , the amide I and amide II bands between 1800 – 1500 cm^{-1} , and C-O-C and C-O-P stretching vibrations of polysaccharides between 1200 – 900 cm^{-1} . A comprehensive list of absorption band assignments may be found in Naumann.¹ Application of multivariate analysis allows differentiation between unique biological materials.^{2,3}

Through automation the number of IR samples analyzed per day may be greatly enhanced and potential sample and data

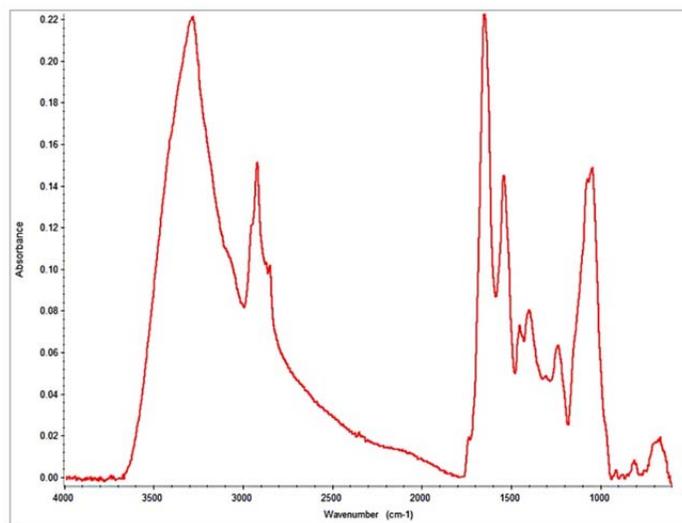


Figure 2. Infrared transmission spectrum of dried yeast suspension

entry errors decreased. Due to variability in biological materials, replicate sample measurements are often required. If 4 replicate measurements are made for 1 sample, 84 samples may be analyzed in a typical eight-hour work day equating to over 330 individual spectra collected.

AutoPro software (Figure 3), a modern Windows program, automates accessory position movement and data collection for most FTIR spectrometers. Patterns for standard configurations such as a 96-well plate are available. Also, customizable patterns are possible. Spectral file names may be unique or auto-named.

In addition to biological applications, an automated XY plate reader may be used for polymer solutions and solvent residuals analysis.

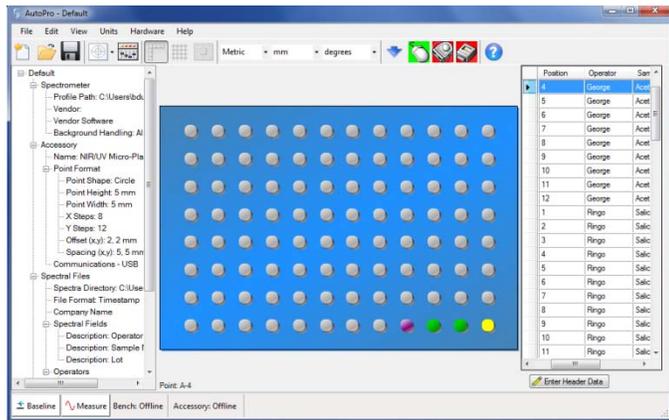


Figure 3. PIKE AutoPRO Software

Conclusions

Infrared transmission spectra of microbiological suspensions may be collected by drying the suspension on an IR transparent plate. Implementing the use of an in-compartment XY plate reader accessory allows for automated measurements via transmission or reflectance to increase laboratory capacity.

References

- ¹ D. Naumann. Encyclopedia of Analytical Chemistry. R.A. Myers (Ed.) 102-131 (2000).
- ² D. Naumann. Mikrochim Acta. 373-377 (1988).
- ³ G. Fischer et al. J Microbiol Methods. 63-77 (2006).