



SRC Spectra
Research
Corporation

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Please join Tornado Spectral Systems at the

IFPAC 2018 Annual Meeting

ADVANCING THE UNDERSTANDING & CONTROL OF
MANUFACTURING PROCESSES

February 12-14, 2018

Bethesda North Marriott | Bethesda, MD

Meet our experts and learn more about Tornado's HyperFlux™ PRO Plus

Visit us at **booth 209** and learn more about our **HyperFlux™ PRO Plus** Raman Spectroscopy system! Our team will demonstrate how you can achieve the best possible combinations of **signal strength** and **spectral resolution** in a dispersive spectrometer.



High Throughput Virtual Slit (HTVS™) Performance Advantage

Sensitivity, Speed and Safety

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Tornado's proprietary HTVS design eliminates spectrometer slit losses while maintaining high spectral resolution.



What can be done with the HTVS performance advantage is game-changing:

(A) 10X Improvement in Spectrometer Throughput and Signal Strength

(B) 3X to 6X Improvement in SNR - more accurate identification and quantitation for challenging mixtures and low concentration levels

(C) 10X to 30X Faster Measurement - for precise tracking of rapid changes and transient features

(D) Lower Laser Power Operation - for improved safety and reduced sample damage

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Featured Application Note

Featured Video

9 Component Multivariate Calibration For
The Analysis Of Metabolites

Demonstrating the Speed & Sensitivity of
Tornado's HyperFlux PRO Plus: Real-time

Raman Spectroscopy of Propylene Glycol in Water

An assessment by Sanofi UK of the performance of the HyperFlux™ PRO Plus Raman Spectroscopy System for the quantitative analysis of biochemical components in a simplified chemically defined pseudo growth medium for mammalian cell culture.

To demonstrate the sensitivity of the PRO Plus for making real-time process measurements or for reaction monitoring, we've performed a simple experiment where we add small quantities of propylene glycol to water and then monitor the mixture with a PRO Plus analyzer and immersion probe.



Rapid Development of a 9 component Multivariable Calibration for the analysis of Metabolites in Chemically Defined Cell Culture Media

Marjorie Maquasias, Sibhan Hennessy and Dylan Jones
Chemical and Biotechnology Development, Sanofi, Research, Suffolk, CB9 6PL, UK

This study assessed the performance of the HyperFlux™ PRO Plus Raman Spectroscopy System (Tornado Spectral Systems) for the quantitative analysis of biochemical components in a simplified chemically defined pseudo growth medium for mammalian cell culture. An experiment was designed to evaluate the ability of Raman Spectroscopy to directly measure individual components in a complex mixture at concentrations at or below the level of quantification of conventional Raman spectrometers. A set of samples with varying amounts of glucose, lactate, glycine, glutamate, aspartate, arginine, histidine, leucine, and phenylalanine were prepared so that covariance between components was close to zero. The spectral collection and model development were completed in one day. The sample spectra were collected in a moving using fast acquisition times and generating calibrations were developed in the afternoon using basic pre-treatments such as derivatives and normalisation.

Background

Real time analysis of biochemical metabolites in bioreactors using Raman Spectroscopy has been widely discussed in the literature [1-4]. Finding the ability to accurately monitor the composition of bioreactor media, the formation of waste products, or need to optimize the culture conditions, provides valuable information to better control the process and thereby improve process yield and consistency.

Raman spectroscopy has not yet been widely implemented in industrial bioreactors and this is due mainly to the complexity of the sample under analysis. There are many interfering signals arising from a cocktail of metabolites which makes calibration model development challenging. A second factor is that in bioreactors, there are high levels of turbidity between samples to be processed, making it unclear whether either the components of interest can be measured directly or via some differential means which in turn has implications for scalability. Finally, the accuracy of Raman instruments can be in doubt as sample concentrations tend to vary often and Raman spectrometers are operating at the lower end of their capabilities. Measurement errors of 10-20 minutes to obtain a good calibration is the reality.

In this analysis, a design space was generated to produce a multivariable calibration set for a chemically defined growth medium which contained dependencies between variables. The samples were then synthesized in the laboratory and analysed using the HyperFlux™ PRO Plus 700 via Raman Spectroscopy System.

Method

Sample Preparation:

Using an experimental design which ensured orthogonality between the variables, a design space was mapped out with nine variables and five levels. A sample covariance plot between two variables is shown in Figure 1. Scatter graphs could be created for all combinations of variables.

Covariance plot between glucose and lactate

Level	Glucose	Lactate
1	0.00	0.00
2	0.05	0.00
3	0.10	0.00
4	0.15	0.00
5	0.20	0.00
6	0.00	0.05
7	0.00	0.10
8	0.00	0.15
9	0.00	0.20



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