

# Fusion QbD<sup>®</sup> – Advanced LC & LC/MS Method Development Software

## Protein and Peptide Examples



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# Fusion LC Method Development (FMD)

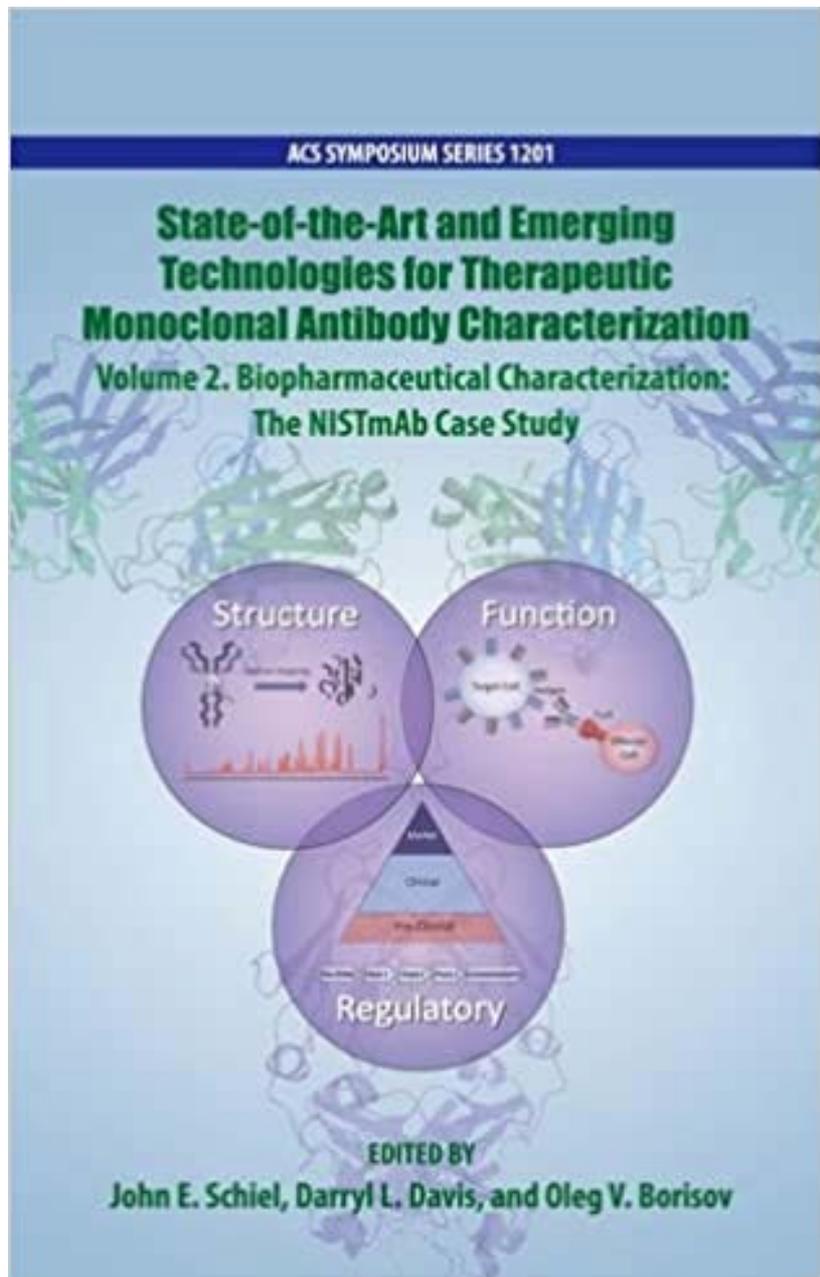
Full Support  
for Part 11  
Compliance



Citrix-Ready  
Certified

## LC METHOD DEVELOPMENT

- Quality by Design Complete
- Automated DOE & Peak Tracking
- Full Robustness Integration
- 100% Aligned with QbD Guidances



# Only Fusion QbD – Supports Small and Large Molecule Method Development

**NIST mAb Case Study**  
**Work was done at Amgen  
using Fusion QbD**

John Schiel, Darryl Davis, Oleg Borisov. ed. (2015), *State-of-the-Art and Emerging Technologies for Therapeutic Monoclonal Antibody Characterization Volume 2. Biopharmaceutical Characterization: The NIST mAb Case Study*, American Chemical Society

# Use of Fusion QbD for Automated Method Screening for Biotherapeutics

Joshua Woods<sup>1</sup>, Marguerite Arechederra<sup>2</sup>, Barbara Kelly<sup>1</sup>, and Justin Sperry<sup>1</sup>

<sup>1</sup>Analytical R&D, Pfizer Inc. Chesterfield MO 63017

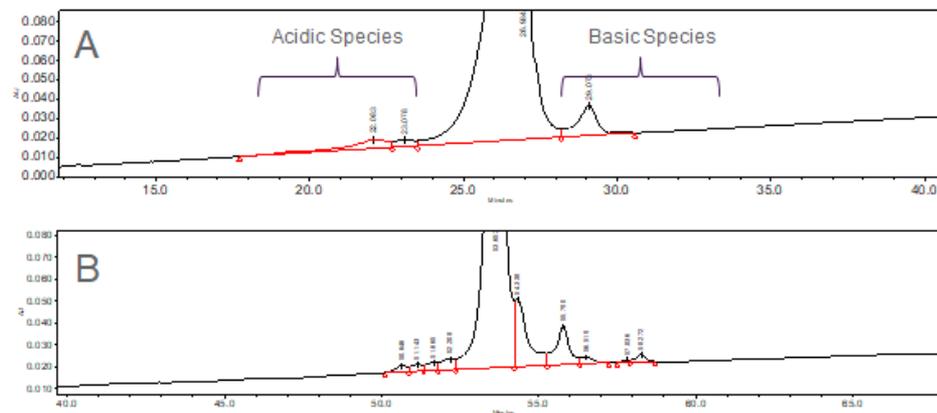
<sup>2</sup>Waters, Milford MA 01757



## Case Study 1 - WCX Development

### *Fusion QbD Screening and Optimization*

- Variables: pH, gradient time, mobile phase composition, organic additive, salt concentration, and column temperature.
- Resulting method showed no fronting, better resolution of acidic species, and better resolution of basic species.



## Productivity Gain

Resulting method comparable to method developed in 5 months prior to use of Fusion QbD.

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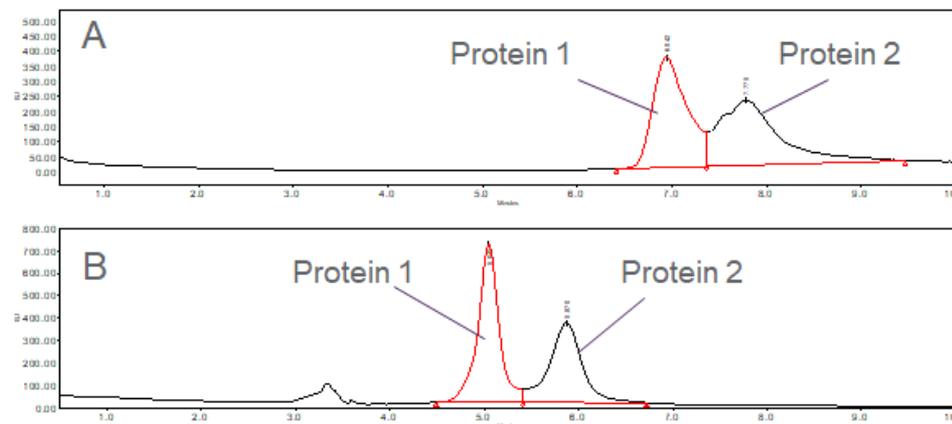
<sup>2</sup>Waters, Milford MA 01757



## Case Study 2 - HILIC Development

### Fusion QbD Screening

- Variables in DOE: pH, column, temperature, gradient time.
- Resulting method shows increased resolution between Protein 1 and Protein 2 in addition to less tailing of both protein peaks.
- 5 Full time employee (FTE) hours, 120 instrument hours.

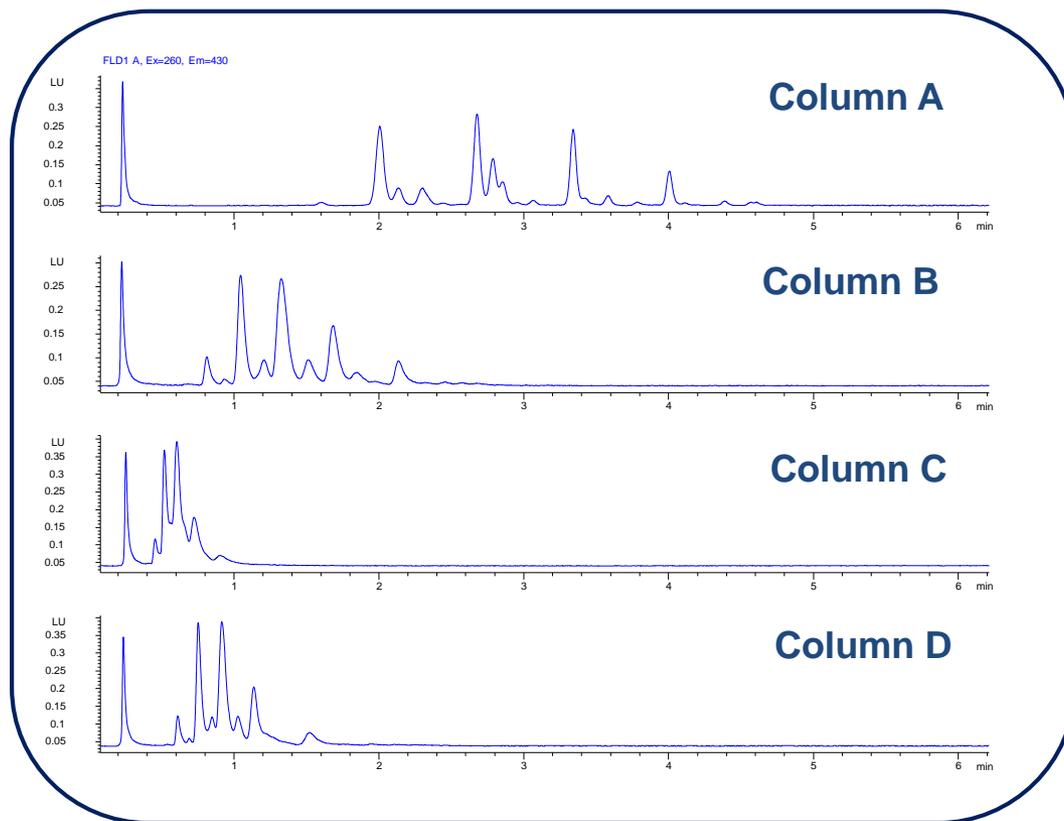


## Overall Productivity Gain – Both Case Studies:

The amount of time saved using Fusion QbD is estimated at 2.5 full time employees (FTE's) over the course of a month.

# Separation of a Critical Glycan Pair using Fusion QbD<sup>®</sup> software

## Chemistry Screening Study – Example Results



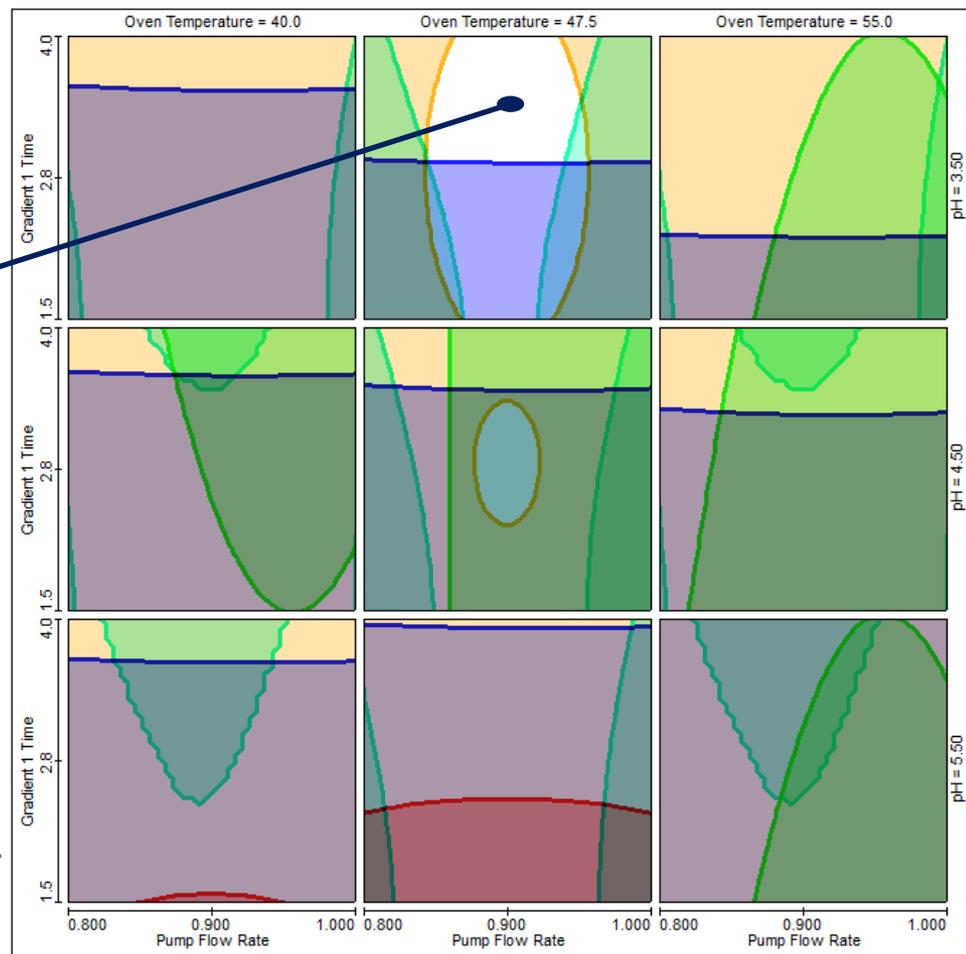
# Separation of a Critical Glycan Pair using Fusion QbD<sup>®</sup> software

## Optimization Study Results

### Best Performing Method

Variable	Level Setting
Pump Flow Rate	0.85
Gradient 1 Time	3.3
Methanol	2.8
Oven Temperature	47.5
pH	3.50

Trellis Graphs  
Cover Full  
Experiment  
Ranges

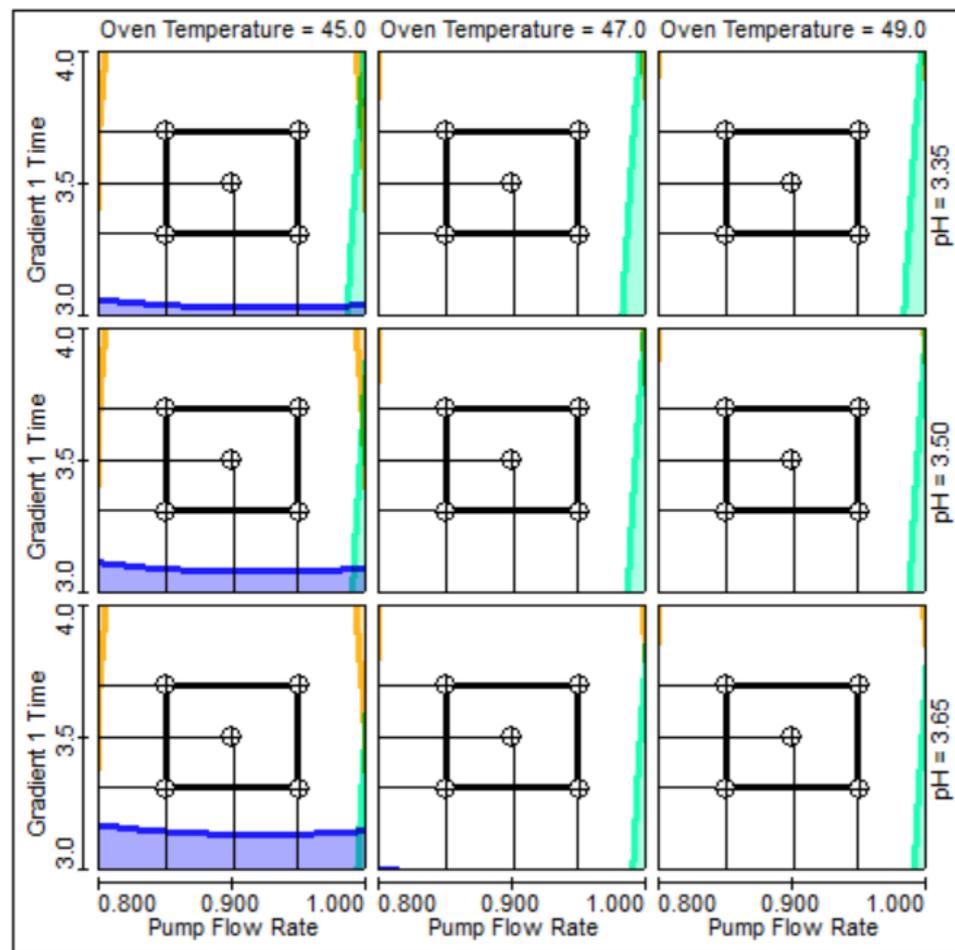


# Separation of a Critical Glycan Pair using Fusion QbD<sup>®</sup> software

## Optimization Study Results

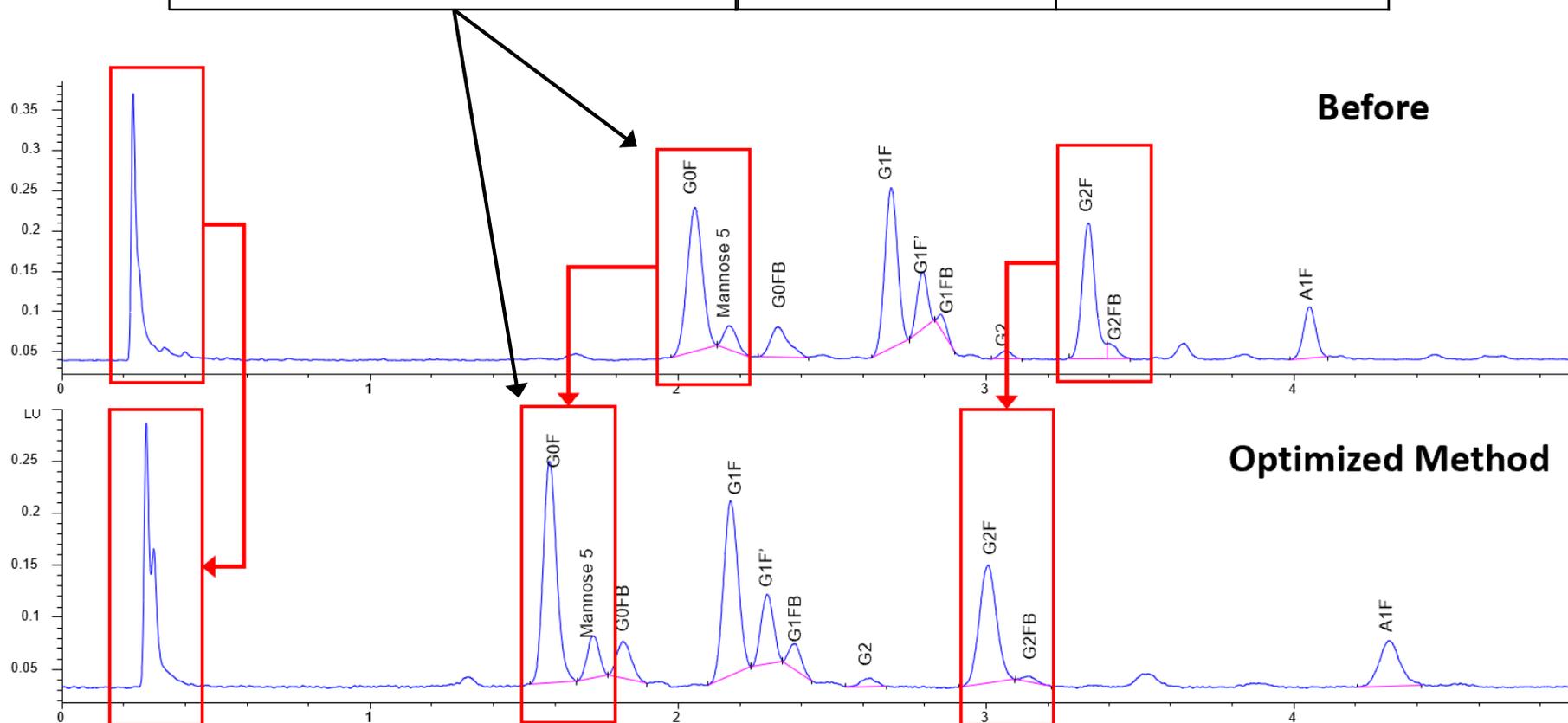
Variable	Level Setting
Pump Flow Rate	0.85
Gradient 1 Time	3.3
Methanol	2.8
Oven Temperature	47.5
pH	3.50

Trellis Graphs  
Zoomed in on  
Robustness  
Ranges



# Separation of a Critical Glycan Pair using Fusion QbD<sup>®</sup> software

Critical Pair Resolution	Initial Method	Optimized Method
		1.30



# Modernization of the USP Monograph method for Human serum albumin (HSA) using Fusion QbD<sup>®</sup> software

## How To Use QbD Software To Improve An Existing Identification Method

Source: Pfizer CentreOne

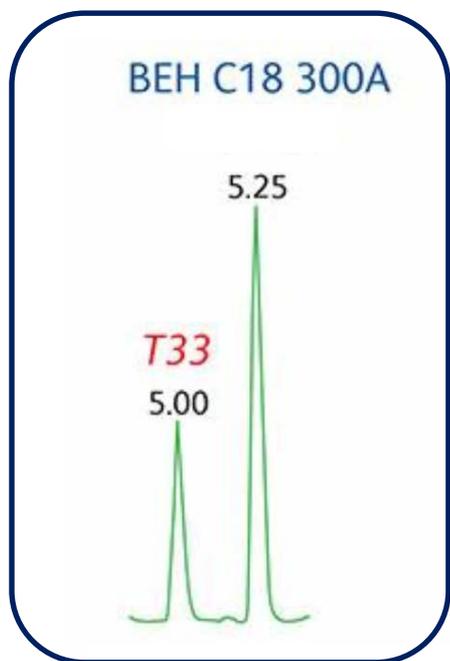
*By Ashraf Madian, Ph.D., Sr Group Leader, Pfizer Global Technology Services Biomanufacturing Sciences and Shen Chen, Ph.D., Director, R&D, Lisa Cherry, Ph.D., Pharmaceutical Sciences Manager, Irish Gibson, Ph.D., Associate Research Scientist, all three from Pfizer CentreOne*

Pharmaceutical Online, November 15, 2017

# Modernization of the USP Monograph method for HSA using Fusion QbD<sup>®</sup>

## Chemistry Screening Study – Example Results

BEH C18 Column separates a critical marker peak from a co-eluting peak.



BEH C18 130A



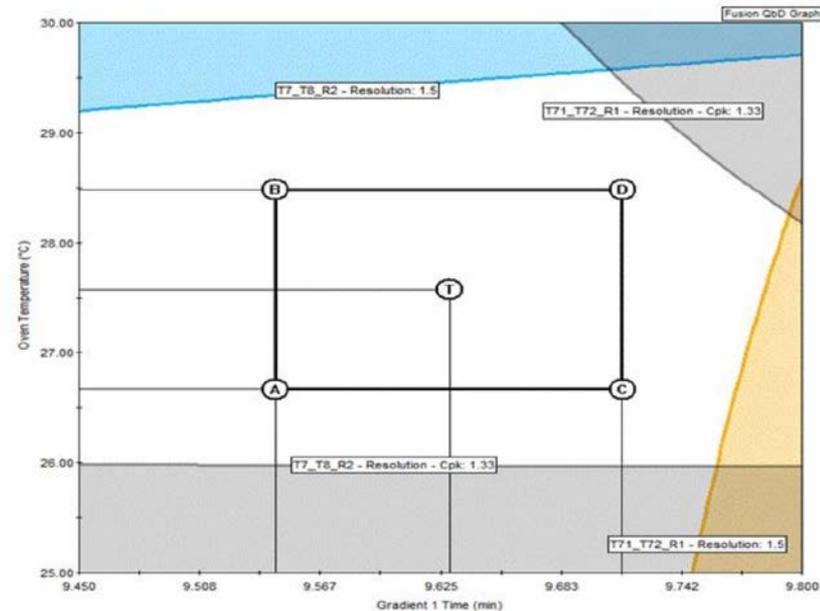
CSH C18 130A



# Modernization of the USP Monograph method for HSA using Fusion QbD<sup>®</sup>

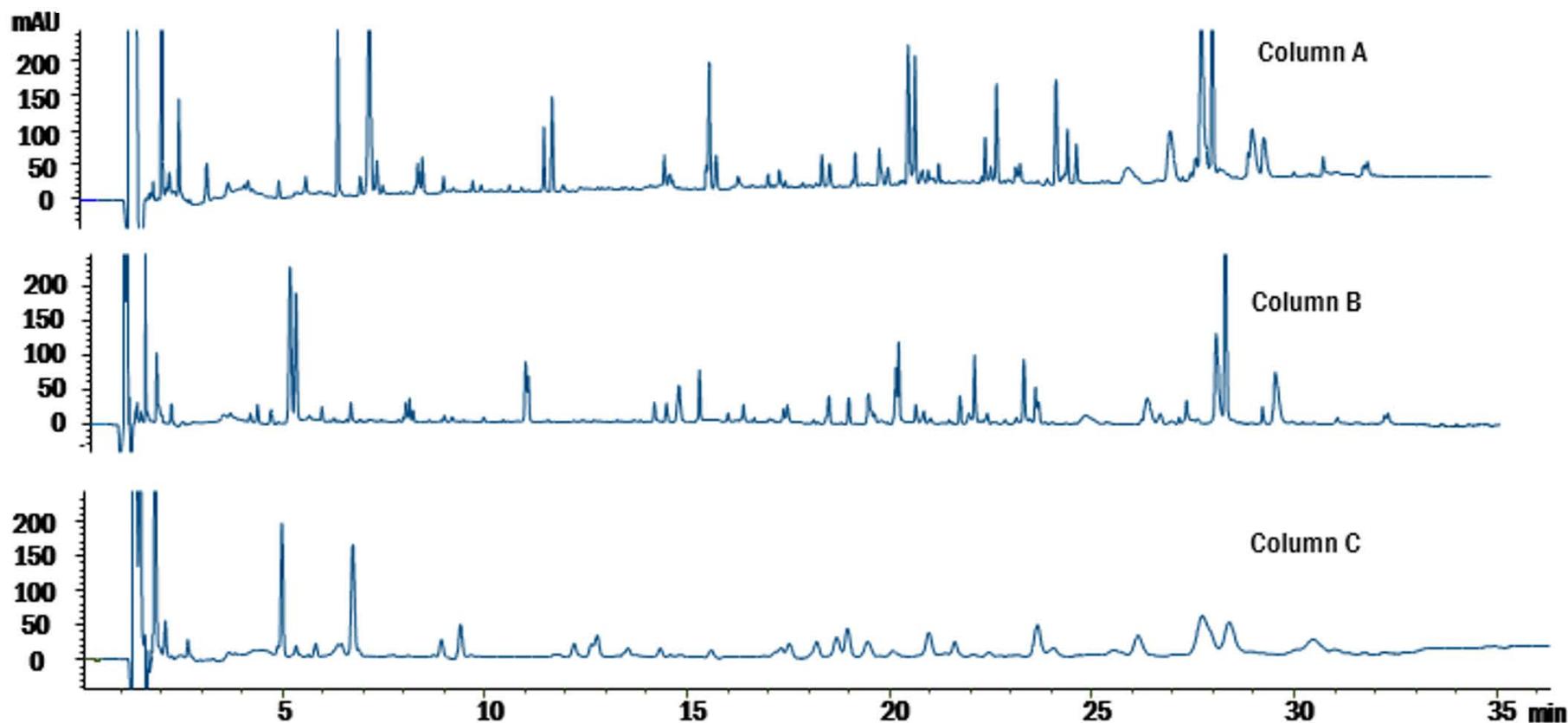
## Optimization Study Results

- Optimized column selection, column temperature, gradient time, and Mobile Phase Composition.
- Reduced Gradient Time from 120 minutes to < 15 minutes
- Achieved a Final Method with Robust Resolution and Excellent Peak Shape for All Seven Marker Peaks.



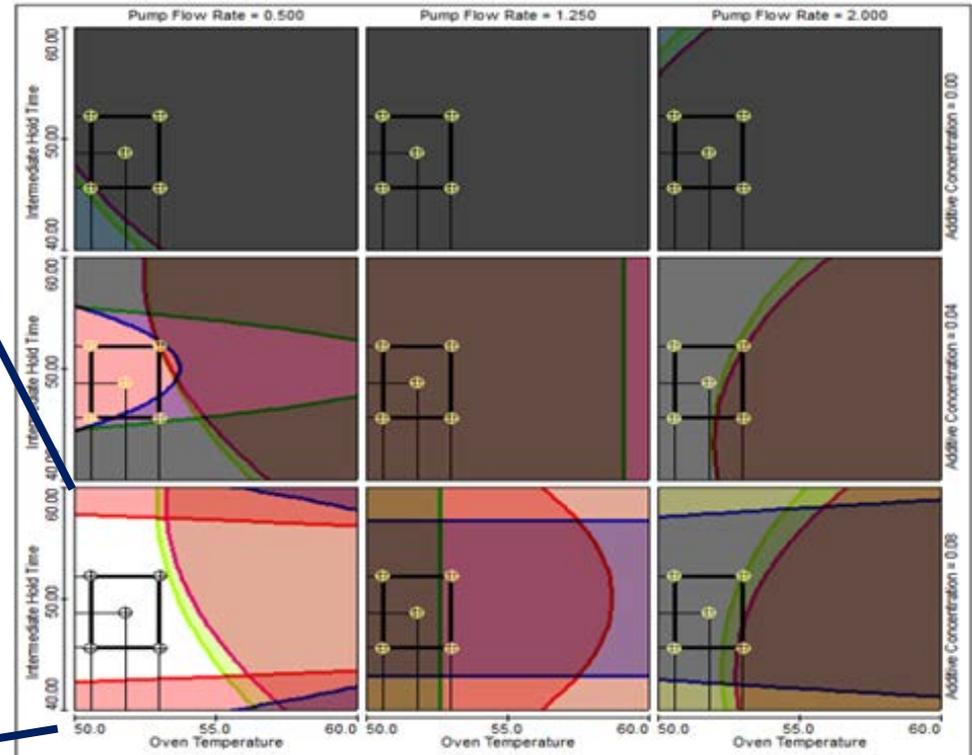
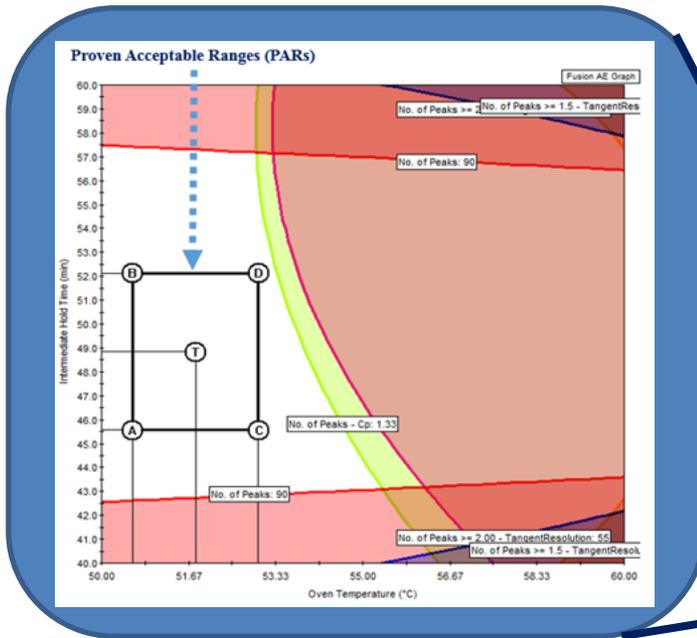
# Peptide Mapping using Fusion QbD<sup>®</sup>

## Chemistry Screening Study – Example Results



# Peptide Mapping using Fusion QbD<sup>®</sup>

## Optimization Study Results

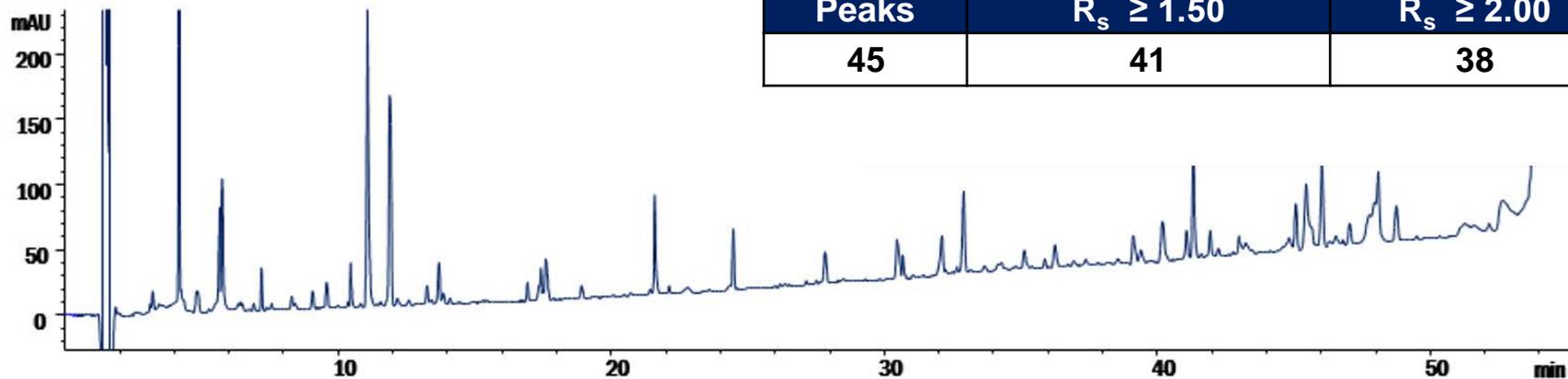


# Peptide Mapping using Fusion QbD<sup>®</sup>

## Optimization Study Results

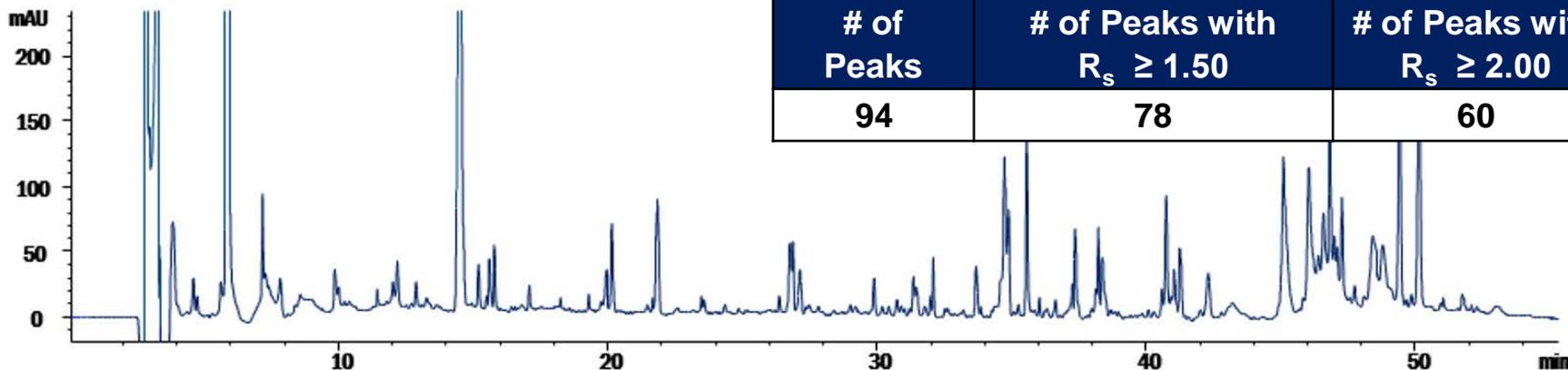
Before

# of Peaks	# of Peaks with $R_s \geq 1.50$	# of Peaks with $R_s \geq 2.00$
45	41	38



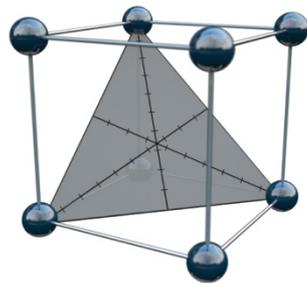
Optimized Method

# of Peaks	# of Peaks with $R_s \geq 1.50$	# of Peaks with $R_s \geq 2.00$
94	78	60



# END

THANK YOU!



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