2019 EASTERN ANALYTICAL SYMPOSIUM & EXPOSITION
ENHANCING ANALYTICAL CHEMISTRY WITH SUSTAINABLE SOLUTIONS
Crowne Plaza Princeton
Conference Center
Plainsboro, NJ
November 18-20, 2019

Successful Modernization of a USP Monograph Method using Fusion QBD® Software resulted in an Optimized LC/MS Method and Eliminated the Need for a Separate GC Method

Richard Verseput
S-Matrix Corporation
1594 Myrtle Avenue
Eureka, CA 95501
USA
Phone: 707-441-0404
URL: www.smatrix.com

Presentation Outline

1. Rapid Chemistry System Screening
    a. Automation Makes it Simple
    b. Automation Maintains Data Integrity
2. Robust Method Optimization
    a. UV & MS Spectra Based Peak Tracking
    b. Robustness Modeling Throughout MODR
3. New Degradants Discovered
    a. Automated Forced Degradation Study
    b. Method Re-optimization

S-Matrix®

Presentation Outline

1. Rapid Chemistry System Screening
    a. Automation Makes it Simple
    b. Automation Maintains Data Integrity
2. Robust Method Optimization
    a. UV & MS Spectra Based Peak Tracking
    b. Robustness Modeling Throughout MODR
3. New Degradants Discovered
    a. Automated Forced Degradation Study
    b. Method Re-optimization

S-Matrix®

Initial Chemistry Screening – Gradient Time / pH / Column Type

Gradient Time – 5 Levels:	10.0, 13.8, 17.5, 21.3, 25.0
pH – 5 levels:	2.7, 3.2, 3.7, 4.3, 5.0
Column Type – 4 Levels:	BEH C18, BEH Shield RP18 HSS T3, CSH Phenyl Hexyl

---

All Possible Combinations = 5 x 5 x 4 = 100 methods

Fusion QbD Screening Design = 30 methods (plus 6 repeats)

> 3x efficiency.

S-Matrix®

Rapid Chemistry System Screening

Fusion QbD

Organizes the experiment for fully automated efficient execution:

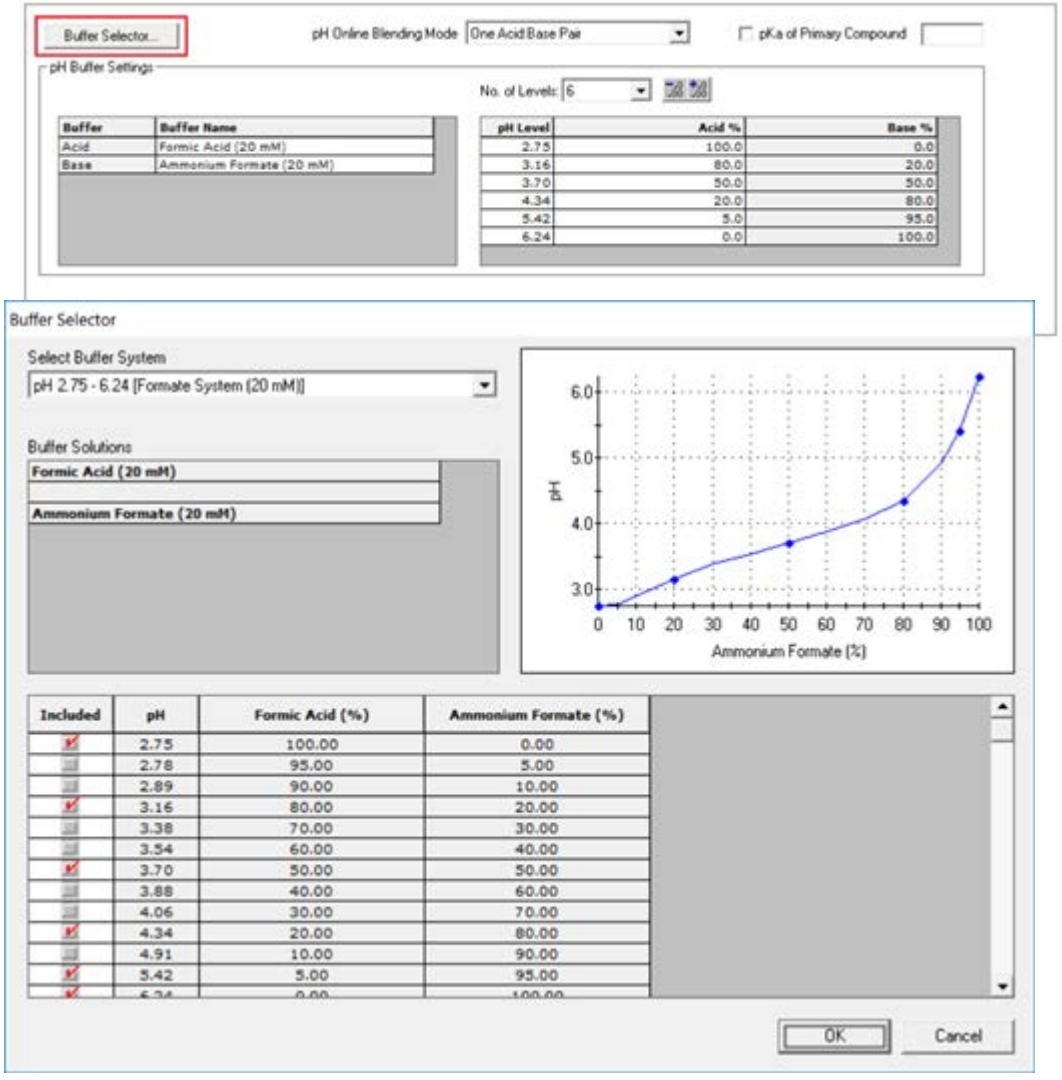
- **Ramps on Temperature**
- **Blocks on Mobile Phase Chemistries**
- **Conditions Columns between Chemistry Changes**

	Run No.	Gradient Time	pH	Column Type
1	Condition Column - 1	2	2.73	BEH C18
2	Condition Column - 2	2	2.73	BEH Shield RP18
3	Condition Column - 3	2	2.73	HSS T3
4	Condition Column - 4	2	2.73	CSH Phenyl Hexyl
5	1	25	2.73	BEH C18
6	2	10	2.73	BEH C18
7	3	25	2.73	BEH Shield RP18
8	4	10	2.73	BEH Shield RP18
9	5	25	2.73	HSS T3
10	6	10	2.73	HSS T3
11	7	17.5	2.73	CSH Phenyl Hexyl
12	8	17.5	2.73	CSH Phenyl Hexyl
13	Condition Column - 5	2	3.2	BEH C18
14	Condition Column - 6	2	3.2	BEH Shield RP18
15	Condition Column - 7	2	3.2	HSS T3
16	Condition Column - 8	2	3.2	CSH Phenyl Hexyl
17	9	21.3	3.2	BEH C18
18	10	13.8	3.2	HSS T3
19	11	17.5	3.2	BEH Shield RP18
20	12	17.5	3.2	CSH Phenyl Hexyl
21	Condition Column - 9	2	3.69	BEH C18
22	Condition Column - 10	2	3.69	BEH Shield RP18
23	Condition Column - 11	2	3.69	HSS T3
24	Condition Column - 12	2	3.69	CSH Phenyl Hexyl
25	13	17.5	3.69	BEH C18
26	14	17.5	3.69	BEH Shield RP18
27	15	17.5	3.69	HSS T3
28	16	17.5	3.69	CSH Phenyl Hexyl
29	17	25	3.69	CSH Phenyl Hexyl
30	18	10	3.69	CSH Phenyl Hexyl
31	19	17.5	3.69	BEH C18
32	20	17.5	3.69	BEH Shield RP18

Auto-built for walk-away execution in the CDS.

S-Matrix®

Initial Chemistry Screening – Built In pH Automation

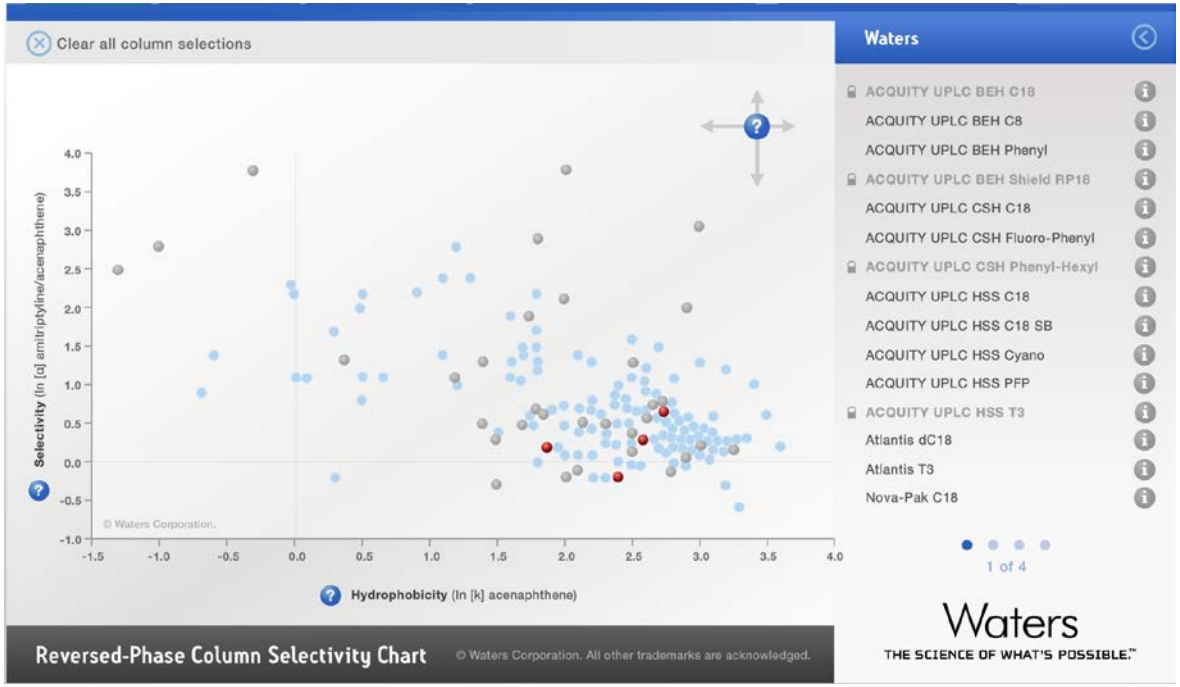


Built in Buffer System Titration Curves

Extremely Precise!

S-Matrix®

Initial Chemistry Screening – Built In Column Switching Automation



Fusion QbD Automation Supports Column Compartments with Multi-position Valves!

Column Settings

	Name	Valve Position	Condition Column Time	pH Upper Bound
1	BEH C18	Position 1	2.0	14.00
2	BEH Shield RP18	Position 2	2.0	14.00
3	HSS T3	Position 3	2.0	14.00
4	CSH Phenyl Hexyl	Position 4	2.0	14.00

S-Matrix®

Fusion QbD Automation – Supports All Waters LC Systems

Empower™

- ✓ Solvent Selection Valves
- ✓ Column Switching Valves

Alliance HPLC

Acquity Binary

Acquity H-Class

Acquity Arc

Acquity UPC²

S-Matrix®

Fusion QbD Automation – Supports All Agilent LC Systems

Empower™

CHROMELEON 7.2

ChemStation
OpenLab

- ✓ Solvent Selection Valves
- ✓ Column Switching Valves

Agilent 1100s And 1200s

Agilent 1260 Infinity Series

Agilent 1260 Infinity II Series

Agilent 1290 Infinity Series

Agilent 1290 Infinity II Series

S-Matrix®

Fusion QbD Automation – Supports Thermo LC Systems

- ✓ Solvent Selection Valves
- ✓ Column Switching Valves

UltiMate LCs

Vanquish Horizon And Flex LCs

S-Matrix®

FDA Statement –

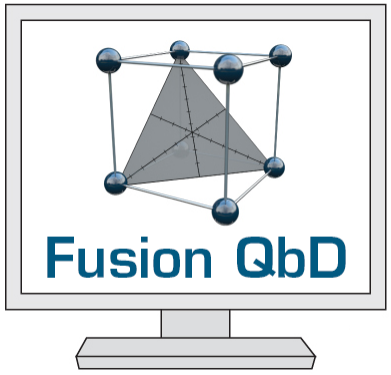
As long as the **data integrity** associated with the method development work matches what would be done in a formal Validation Robustness effort, then the results are acceptable.

USP Workshop – Enhanced Approaches for Analytical Procedure Lifecycle: An Alternative to Traditional Validation

(Sept. 24-25, 2018)

S-Matrix®

Fusion QbD Automation – Maintains Data Integrity



Generates QbD-aligned DOE Experiment

Automatically Builds Sequence and All Instrument Methods

Chromatography Data Software (CDS)

Text Mix pH in S-Matrix - MD Demo\LC Tutorial - Sample Workup as System/Administrator - Sample Set Method Editor

File Edit View Help

Apply Table Preferences: Sample Set Method

	Vial	Inj Vol (uL)	# of Injs	Label	SampleName	Level	Function	Method Set / Report Method	Label Reference	Processing	Run Time (Minutes)	Data Start (Minutes)	Next Inj. Delay (Minutes)	Column Position	Auto Additions	SampleWeight	Dilution
1							Condition Column				6.70	0.00	0.00	No Change			
2							Condition Column	Text Mix pH 001_017			0.10	0.00	0.00	No Change			
3							Equilibrate	Text Mix pH 001_017			3.00	0.00	7.95	No Change			
4	1	2.0	1	Unk-000-000	Blank - 1		Inject Samples	Text Mix pH 001_017		Normal	10.50	0.00	1.50			1.00000	1.00000
5							Condition Column	Text Mix pH 001_001			0.10	0.00	0.00	No Change			
6							Equilibrate	Text Mix pH 001_001			3.00	0.00	0.00	No Change			
7	2	2.0	1	Unk-001-001	1.a.1.a		Inject Samples	Text Mix pH 001_001		Normal	10.50	0.00	1.50			1.00000	1.00000
8							Condition Column	Text Mix pH 001_002			0.10	0.00	0.00	No Change			
9							Equilibrate	Text Mix pH 001_002			3.00	0.00	0.00	No Change			
10	2	2.0	1	Unk-001-002	2.a.1.a		Inject Samples	Text Mix pH 001_002		Normal	10.50	0.00	1.50			1.00000	1.00000
11							Condition Column				6.70	0.00	0.00	No Change			
12							Condition Column	Text Mix pH 001_003			0.10	0.00	0.00	No Change			
13							Equilibrate	Text Mix pH 001_003			3.00	0.00	0.00	No Change			
14	2	2.0	1	Unk-001-003	3.a.1.a		Inject Samples	Text Mix pH 001_003		Normal	10.50	0.00	1.50			1.00000	1.00000
15							Condition Column				6.70	0.00	0.00	No Change			
16							Condition Column	Text Mix pH 001_004			0.10	0.00	0.00	No Change			
17							Equilibrate	Text Mix pH 001_004			3.00	0.00	0.00	No Change			
18	2	2.0	1	Unk-001-004	4.a.1.a		Inject Samples	Text Mix pH 001_004		Normal	10.50	0.00	1.50			1.00000	1.00000
19							Condition Column	Text Mix pH 001_005			0.10	0.00	0.00	No Change			
20							Equilibrate	Text Mix pH 001_005			3.00	0.00	0.00	No Change			
21	2	2.0	1	Unk-001-005	5.a.1.a		Inject Samples	Text Mix pH 001_005		Normal	10.50	0.00	1.50			1.00000	1.00000
22							Condition Column				6.70	0.00	0.00	No Change			
23							Condition Column	Text Mix pH 001_006			0.10	0.00	0.00	No Change			
24							Equilibrate	Text Mix pH 001_006			3.00	0.00	0.00	No Change			

Eliminate Transcription Errors.
Maintain Data in Audited Environment.

S-Matrix®

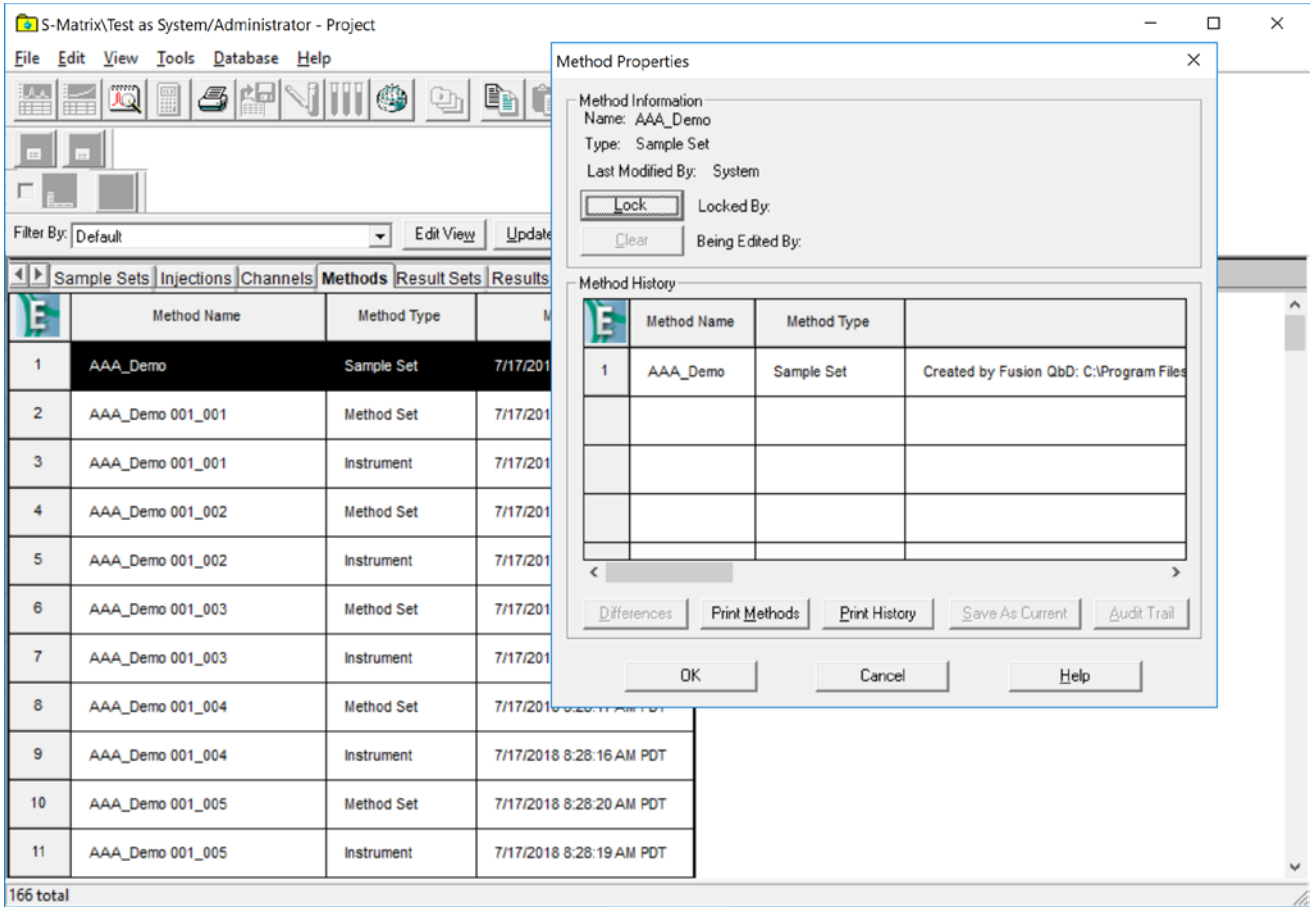
Fusion QbD Automation Supports Full Data Integrity

Full integration of **all e-record** and **all e-signature** features and functions required to support full 21 CFR 11 compliance.

Secure **Workflow** and **Project Management** systems.

Full audit trail, including **bi-directional auditing** of all data exchanges with the CDS.

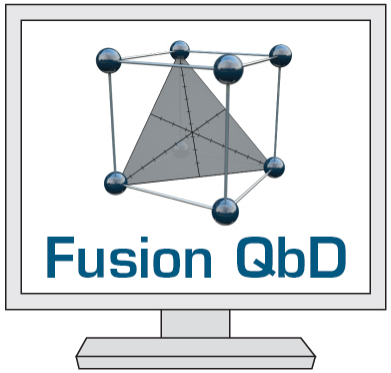
Fusion QbD Automation – Maintains Data Integrity



Audited Data Exchange – Assures Data Integrity

S-Matrix®

Fusion QbD Automation – Maintains Data Integrity

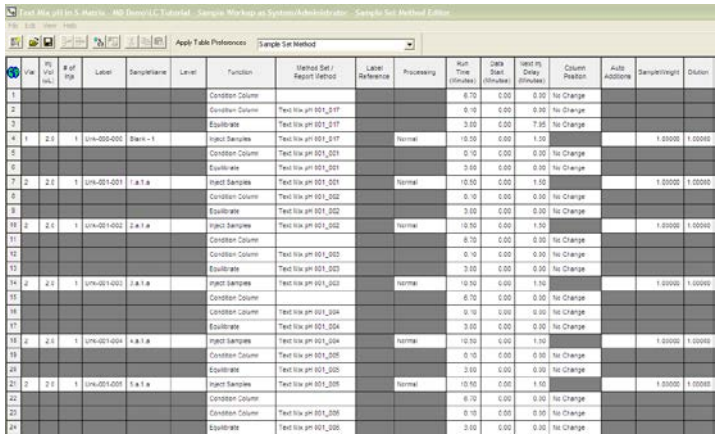


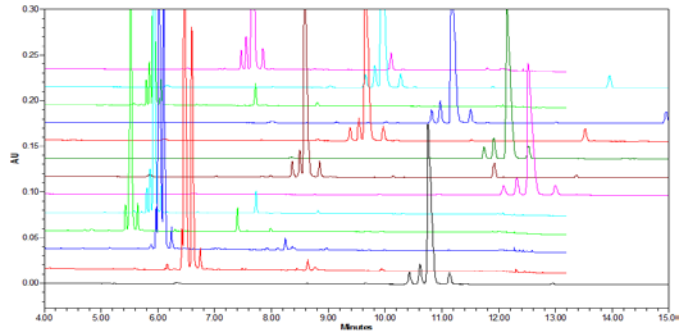
Automatically Retrieves
All Chromatogram
Results Data

Automated analysis, graphing, and reporting.

Report formats:
RTF, DOC, HTML, PDF, XLSX, XML

Chromatography Data Software (CDS)

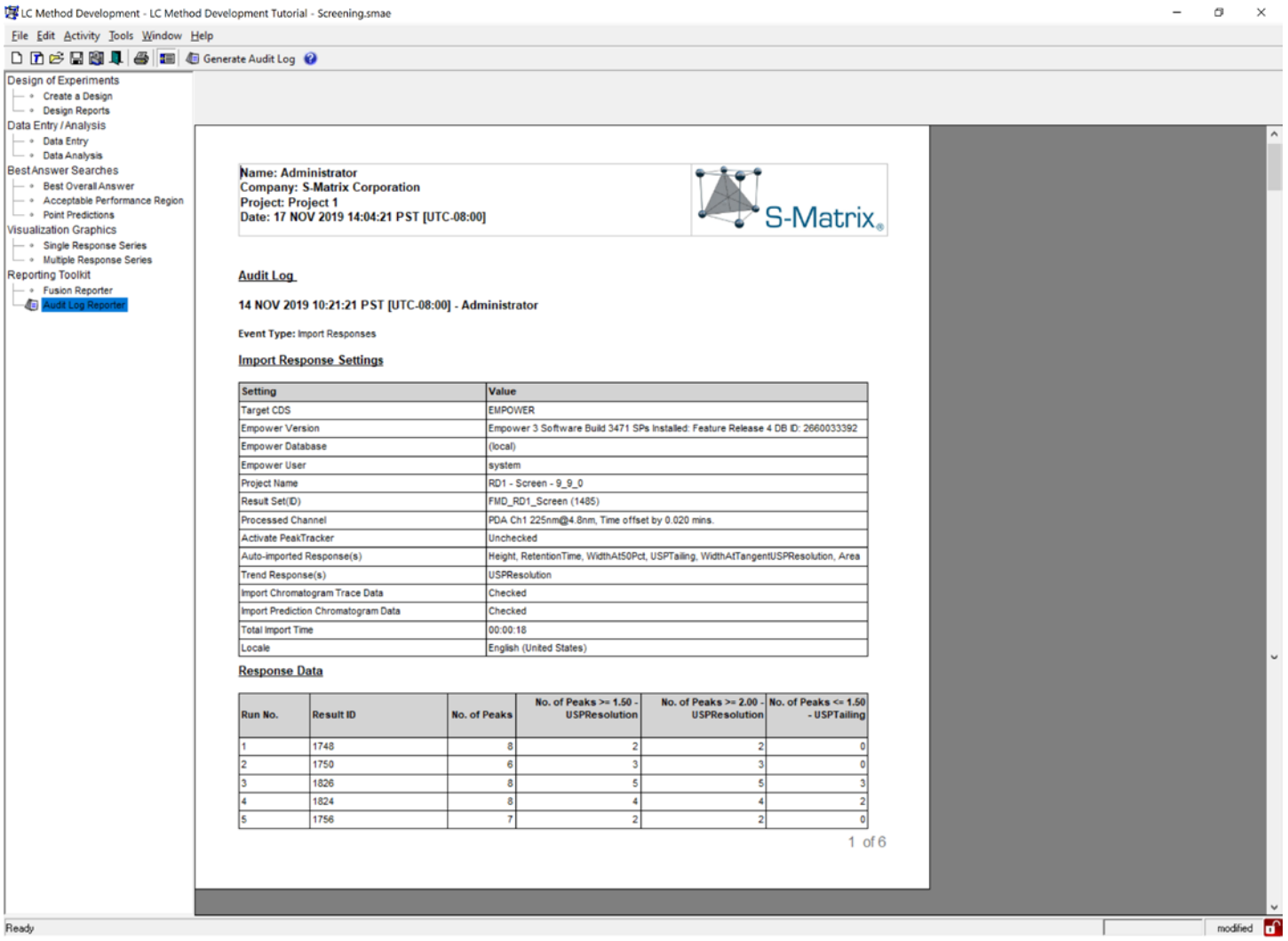




Eliminate Transcription Errors.
Maintain Data in Audited Environment.

S-Matrix®

Fusion QbD Automation – Maintains Data Integrity



Audited Data Exchange – Assures Data Integrity

S-Matrix

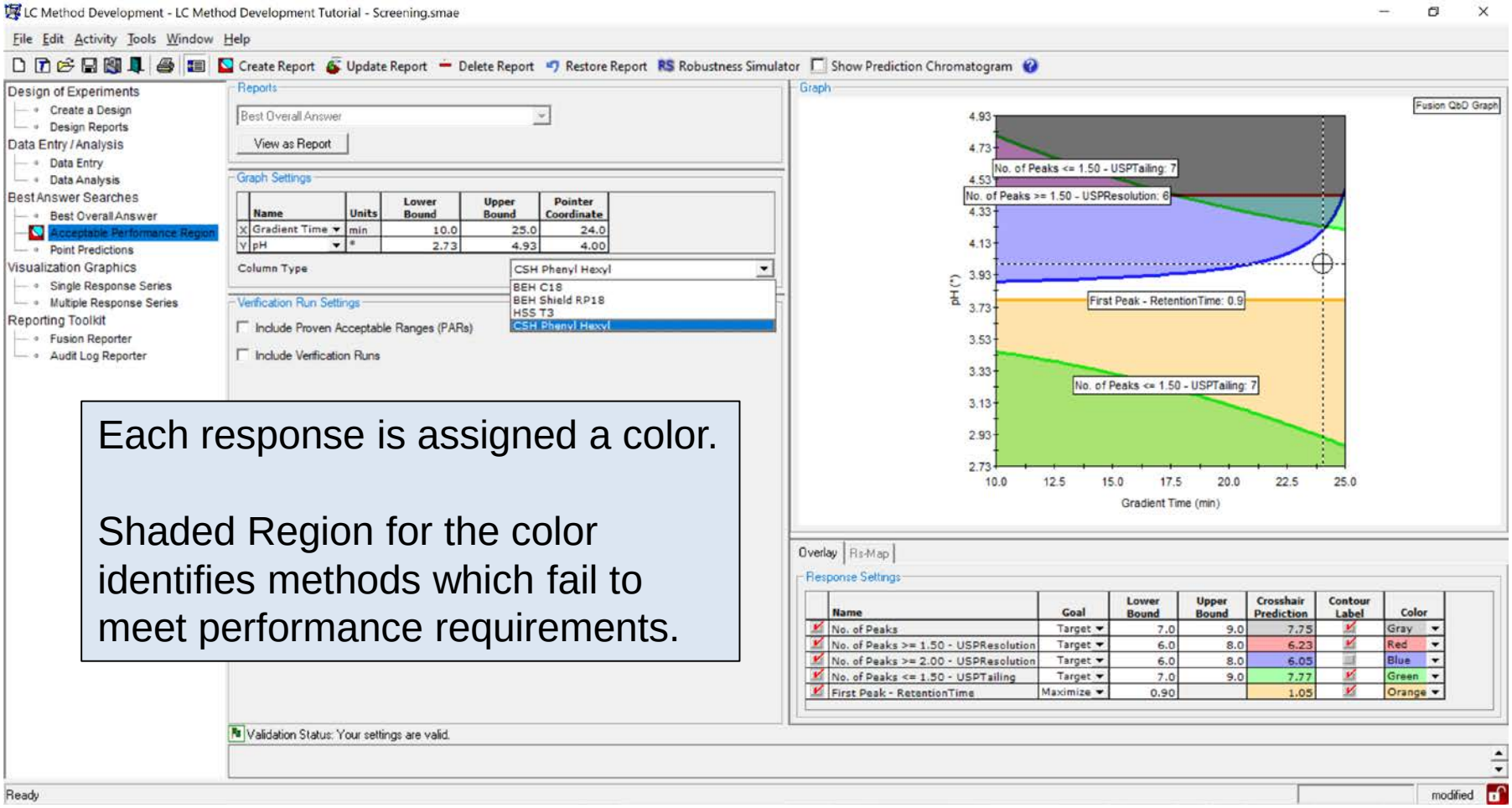
Fusion QbD Trend Responses –

Automatically Derived from Integrated Chromatograms:

- No. of Separated Peaks
- No. of Baseline Resolved Peaks
- No. of Peaks with Good Tailing
- Resolution of the Main Peak
  - from the immediately ***earlier*** eluting peak
  - from the immediately ***later*** eluting peak

S-Matrix®

Chemistry Screening Study – Acceptable Performance Region



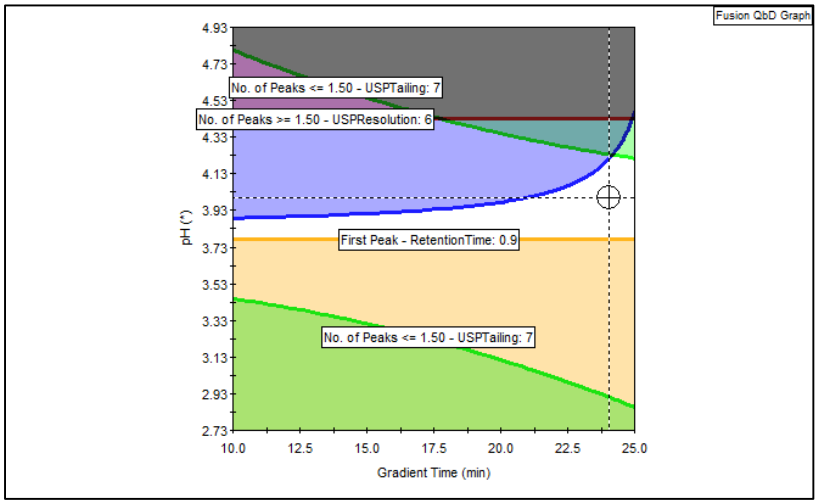
Each response is assigned a color.

Shaded Region for the color identifies methods which fail to meet performance requirements.

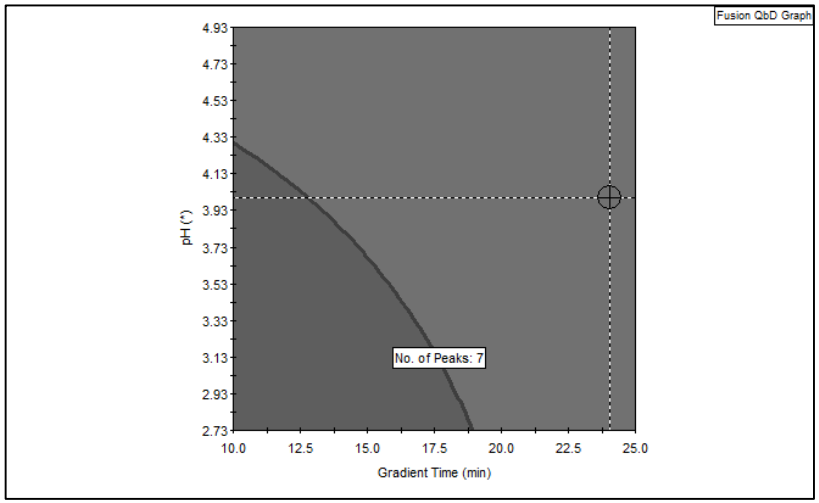
S-Matrix®

Chemistry Screening Study – Acceptable Performance Region

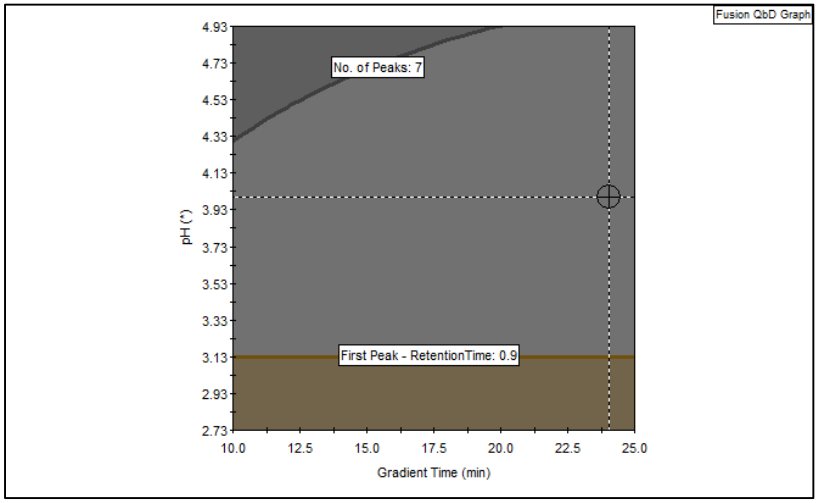
CSH Phenyl Hexyl



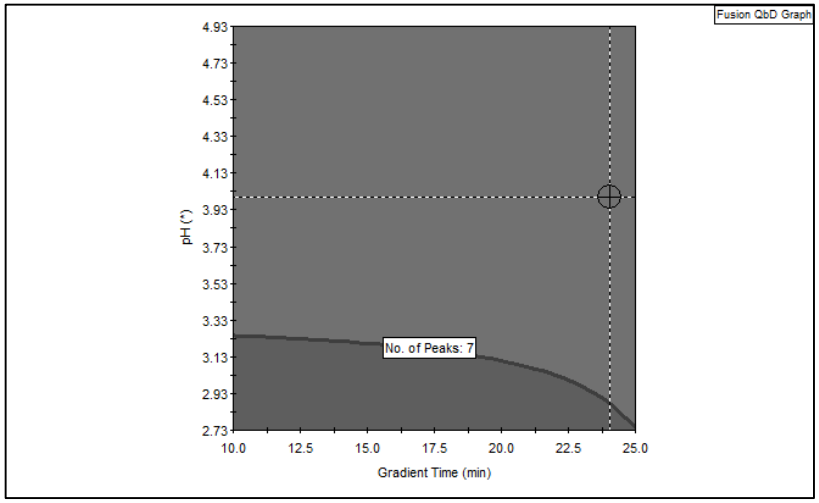
BEH C18



BEH Shield RP18



HSS T3



S-Matrix®

Presentation Outline

1. Rapid Chemistry System Screening
    a. Automation Makes it Simple
    b. Automation Maintains Data Integrity
2. Robust Method Optimization
    a. UV & MS Spectra Based Peak Tracking
    b. Robustness Modeling Throughout MODR
3. New Degradants Discovered
    a. Automated Forced Degradation Study
    b. Method Re-optimization

S-Matrix®

Robust Method Optimization – Gradient Time / pH / Column Type

Pump Flow Rate – 3 Levels:	0.35, 0.45, 0.55
Gradient Time* – 3 Levels:	8.0, 12.0, 16.0
Oven Temp – 3 levels:	40.0, 45.0, 50.0
pH – 3 levels:	3.6, 3.9, 4.2
Column Type – Constant:	CSH Phenyl Hexyl

All Possible Combinations = 3 x 3 x 3 x 3 = 81 methods

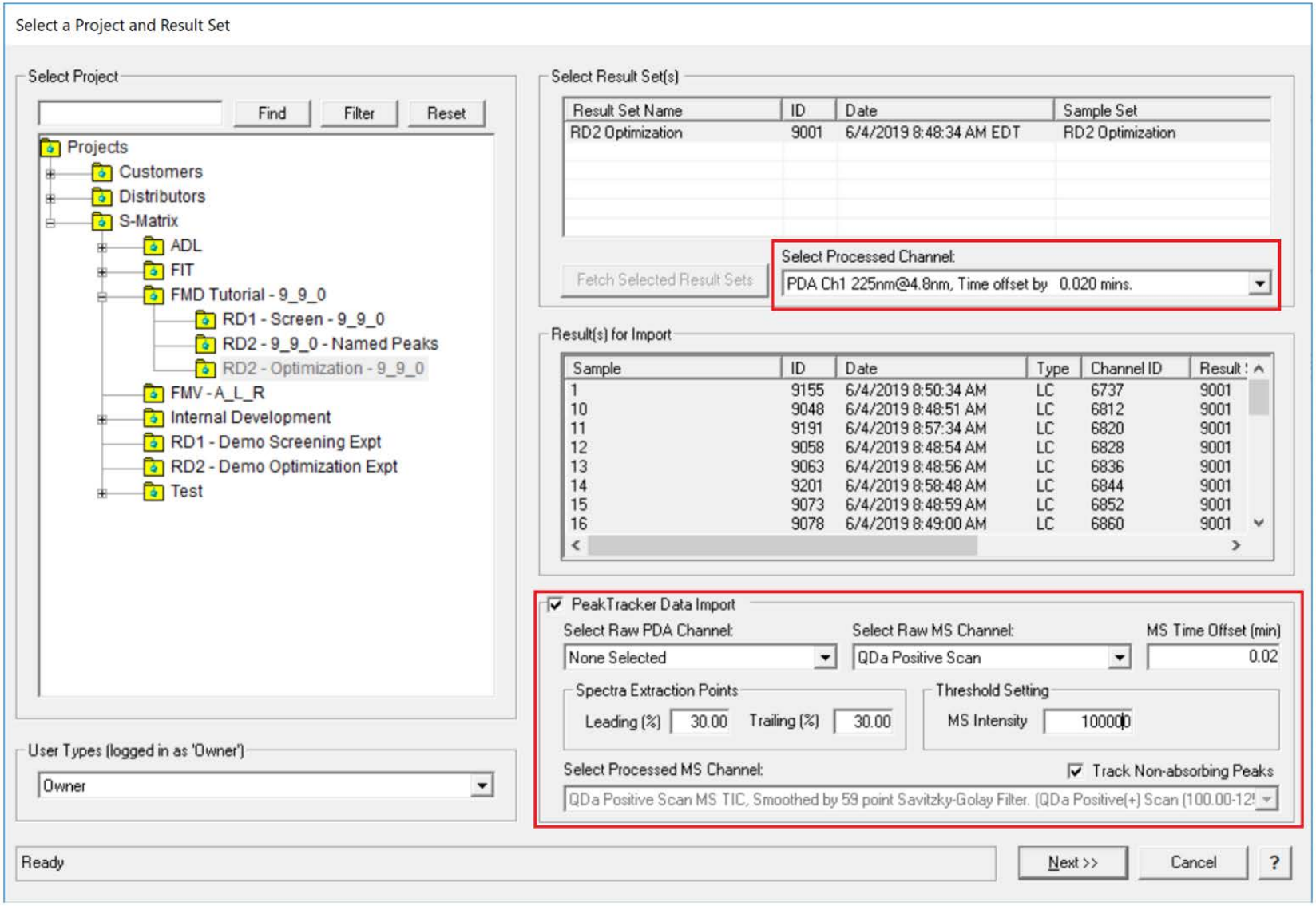
Fusion QbD Optimization Design = 25 methods (plus 5 repeats)

> 3x efficiency.

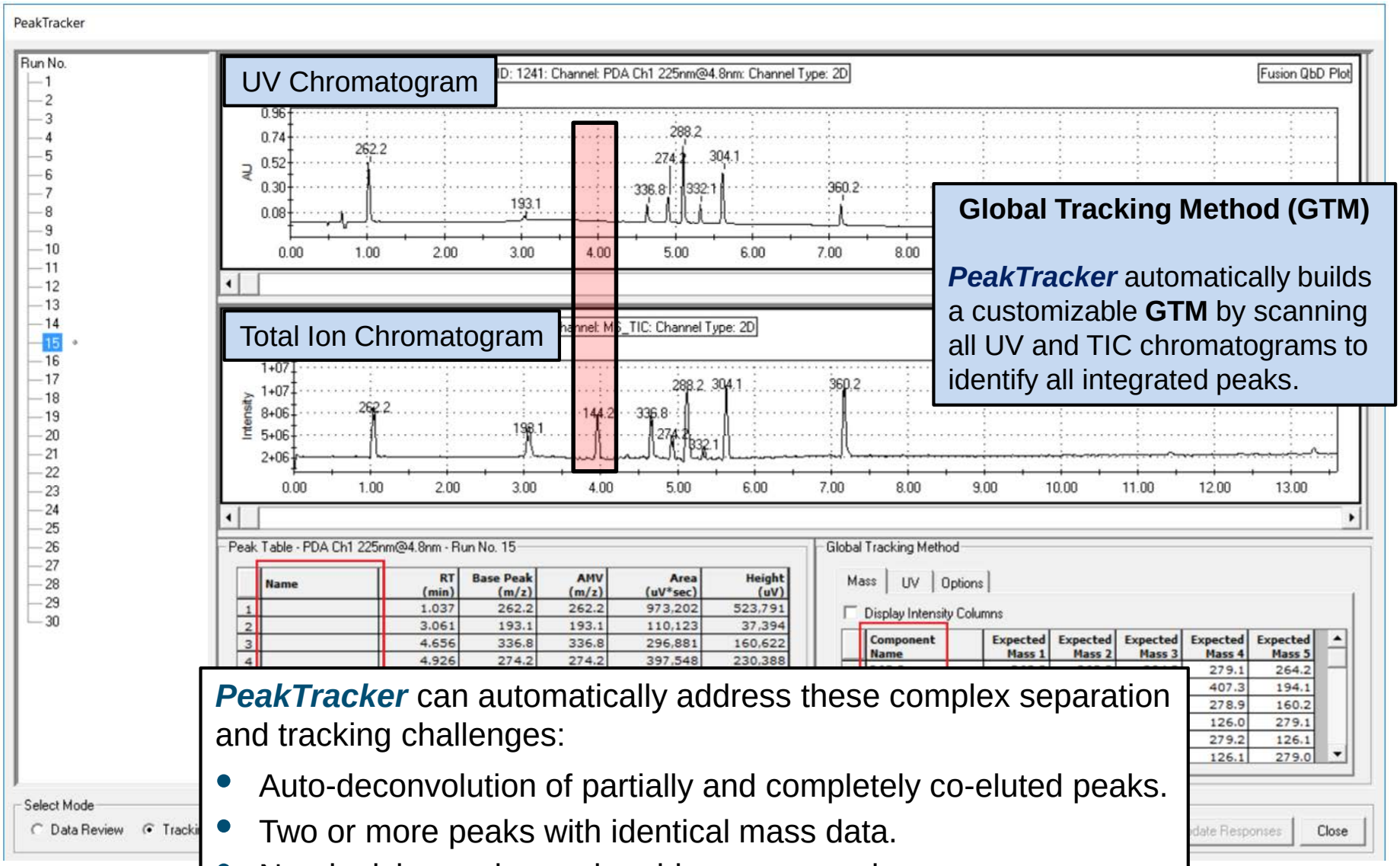
* – levels adjusted for new endpoint of 50% organic

S-Matrix®

Fusion QbD – *PeakTracker*™



Robust Method Optimization – *PeakTracker*



Global Tracking Method (GTM)

PeakTracker automatically builds a customizable **GTM** by scanning all UV and TIC chromatograms to identify all integrated peaks.

PeakTracker can automatically address these complex separation and tracking challenges:

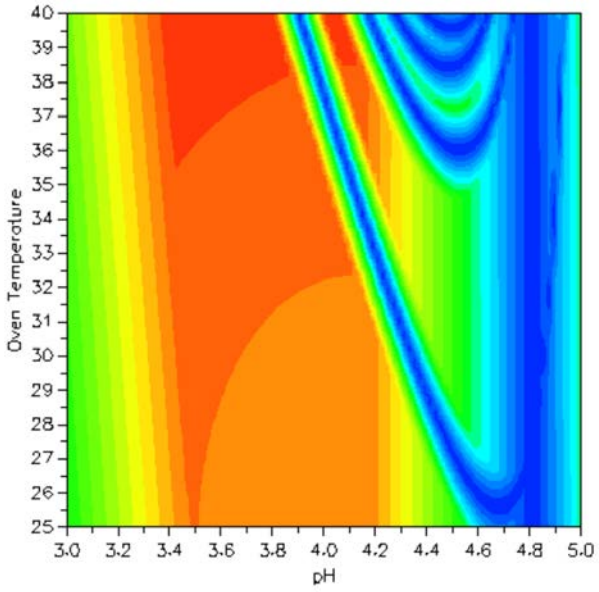
- Auto-deconvolution of partially and completely co-eluted peaks.
- Two or more peaks with identical mass data.
- Non-ionizing and non-absorbing compounds.

S-Matrix®

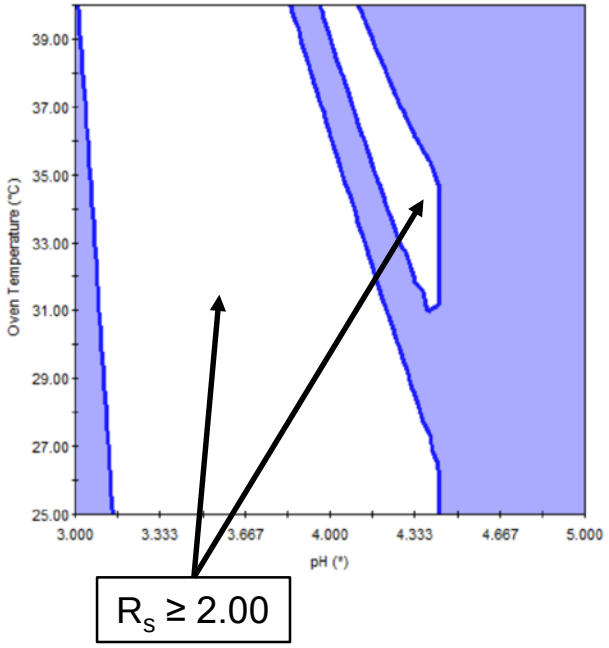
Fusion QbD – *Rs-Map Response*

Fusion QbD now uses its hyper-accurate modeling technologies to predict USP or EP Resolutions for all peaks for any method conditions using standard Resolution equations. Graphical and numerical displays update in real time as you change method conditions.

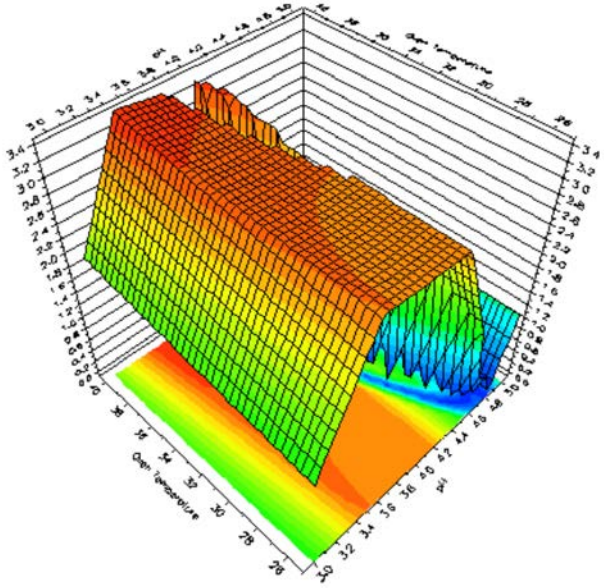
Contour Graph



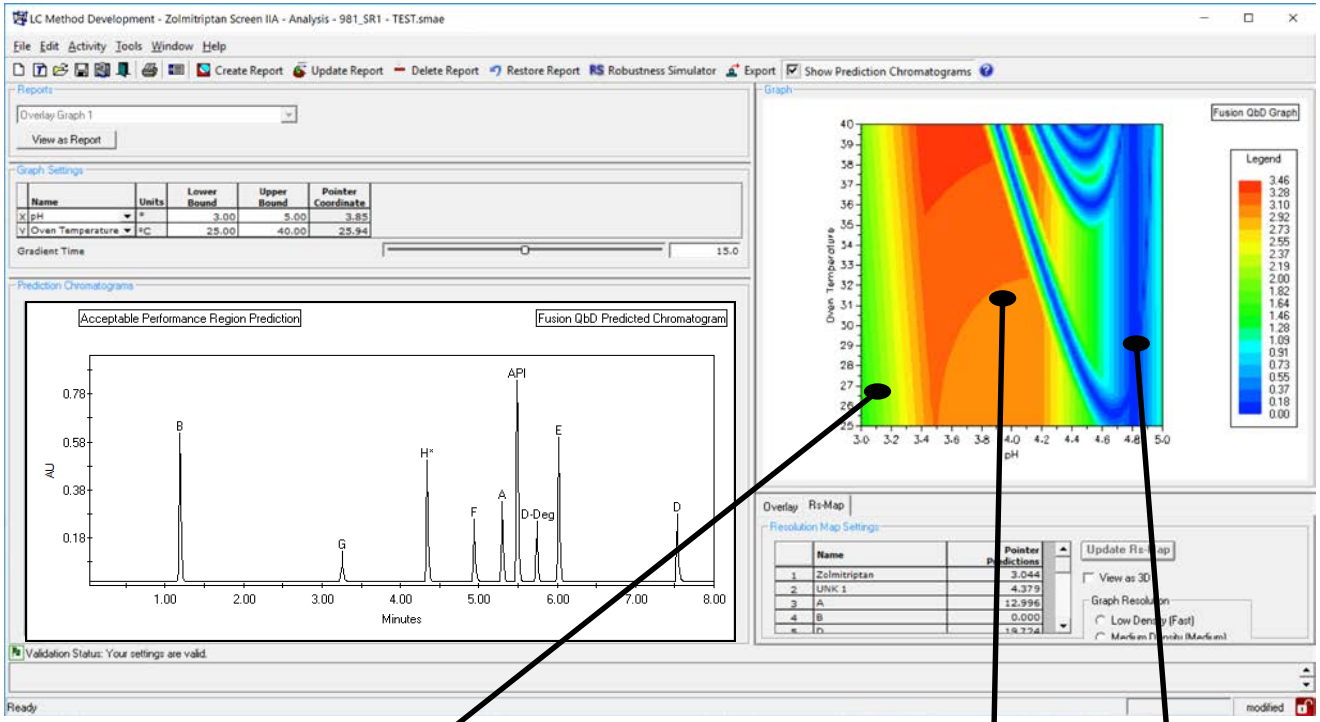
Overlay Graph



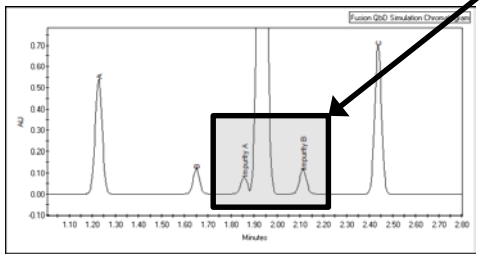
Response Surface Graph



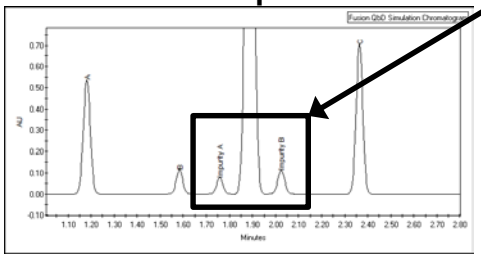
Robust Method Optimization – *Rs-Map Response*



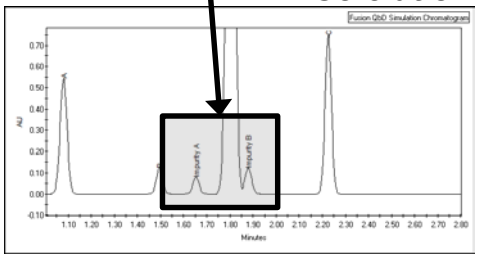
Co-elution



Robust Separation



Co-elution

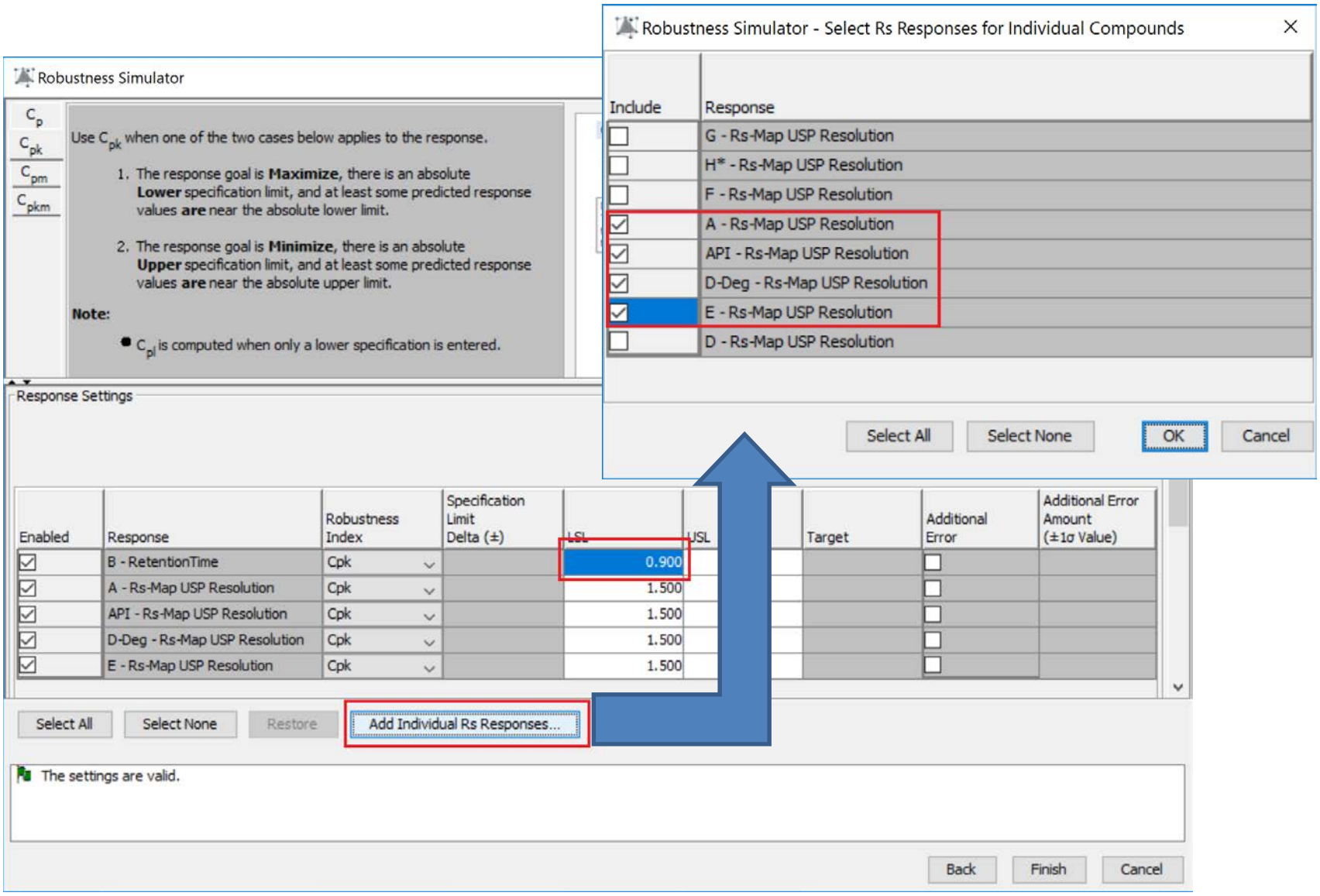


Presentation Outline

1. Rapid Chemistry System Screening
    a. Automation Makes it Simple
    b. Automation Maintains Data Integrity
2. Robust Method Optimization
    a. UV & MS Spectra Based Peak Tracking
    b. Robustness Modeling Throughout MODR
3. New Degradants Identified
    a. Automated Forced Degradation Study
    b. Method Re-optimization

S-Matrix®

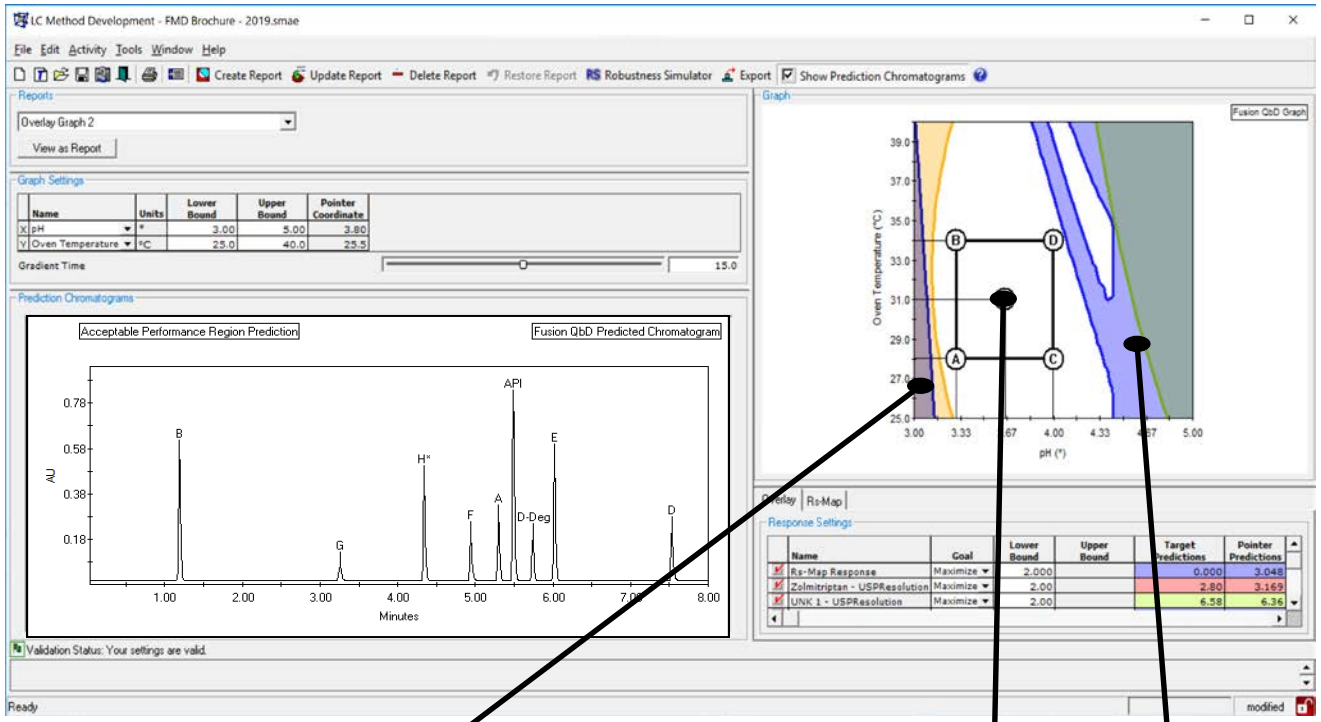
Robust Method Optimization – Robustness Modeling Throughout MODR



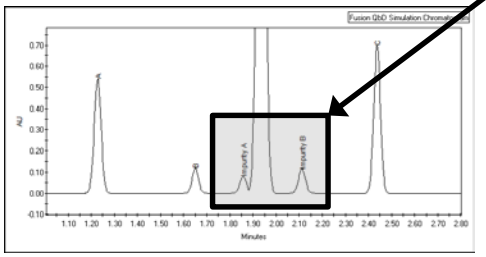
Robustness is calculated for each key performance metric.

Robust Method Optimization – Robustness Modeling Throughout MODR

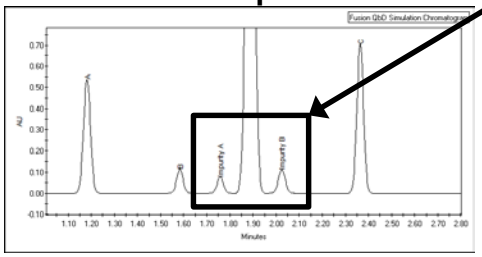
- Mean Performance and Robustness Throughout MODR.
- Include Performance Requirements for Critical Peaks and Peak Pairs.



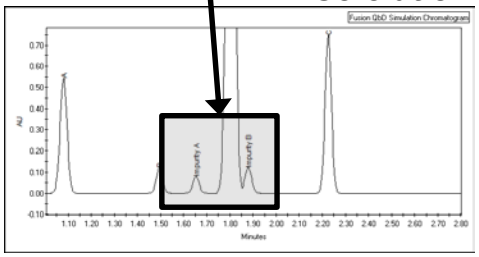
Co-elution



Robust Separation



Co-elution



Monte Carlo Robustness Simulation

"*Statistical treatments (e.g., **Monte Carlo simulations**) can help evaluate the effects of uncertainty.*"

Points to Consider for Design Space – A Regulatory Perspective, Elaine Morefield, Ph.D., 2012 Annual Meeting, AAPS.

Statistical Robustness Metrics

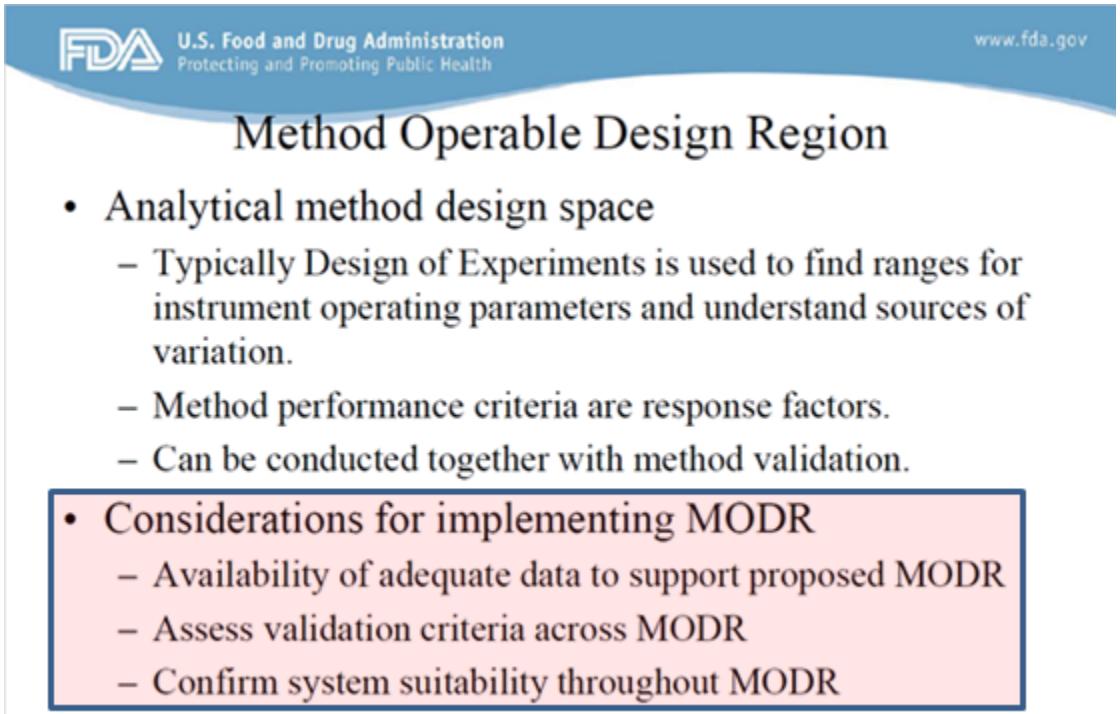
*The FDA has stated that accepted process capability indexes such as **C_p, C_{pk}, C_{pm}, and C_{pkm}** are also part of the QbD toolset.*

US FDA, Quality by Design: Objectives, Benefits, and Challenges, Lawrence X. Yu, Ph.D., 2012 Annual Meeting, AAPS.

Characterizing Robustness Throughout the Design Space

"*Statistical Confidence* ***Shown Throughout the MODR***."

QbD Considerations for Analytical Methods – FDA Perspective, Sharmista Chatterjee, Ph.D., 2013 Annual Meeting, IFPAC.



John F. Kauffman, Ph.D. and Daniel J. Mans, Ph.D., "*Experimental Design and Modeling to Improve HPLC Method Performance for Small Molecules*", FDA Division of Pharmaceutical Analysis, CASSSCMC Strategy Forum Europe 2015

S-Matrix®

Analytical Procedure Lifecycle Management

European Compliance Agency, Analytical Quality Control Group, July 2018, Final_r1

3. Process Capability

Process capability refers to the performance of the process when it is operating under statistical control. Two capability indices are usually computed: C_p and C_{pk} in a similar way as was described with P_p and P_{pk}. However, C_p measures the **potential** capability in the process, if the process was centred, while C_{pk} measures the actual capability in a process which is off-centre or biased. If a process is centred, then $C_p = C_{pk}$.

$$C_{pk} = \min\left[\frac{U-\bar{\bar{X}}}{3S_w}, \frac{\bar{\bar{X}}-L}{3S_w}\right] \qquad (1.5)$$

The critical thing to note is that whilst the formulae for P_{pk} and C_{pk} look very similar, the standard deviation used to calculate the reference interval for C_{pk} is not S_t but S_w.

S_w is the within batch standard deviation (called the within sub group standard deviation in ISO) not the overall process standard deviation. It is usually estimated from a Shewhart mean and range control chart using the formula

Robust Method Optimization – Optimized Method Predictions

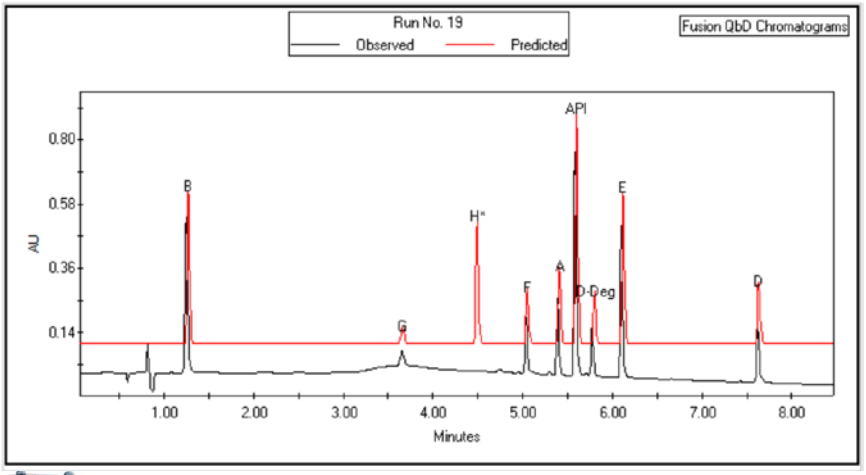
Predicted Point - 19

Variable Settings

Variable	Level Setting
Pump Flow Rate	0.450
Gradient Time	12.0
Oven Temperature	45.0
pH	3.90

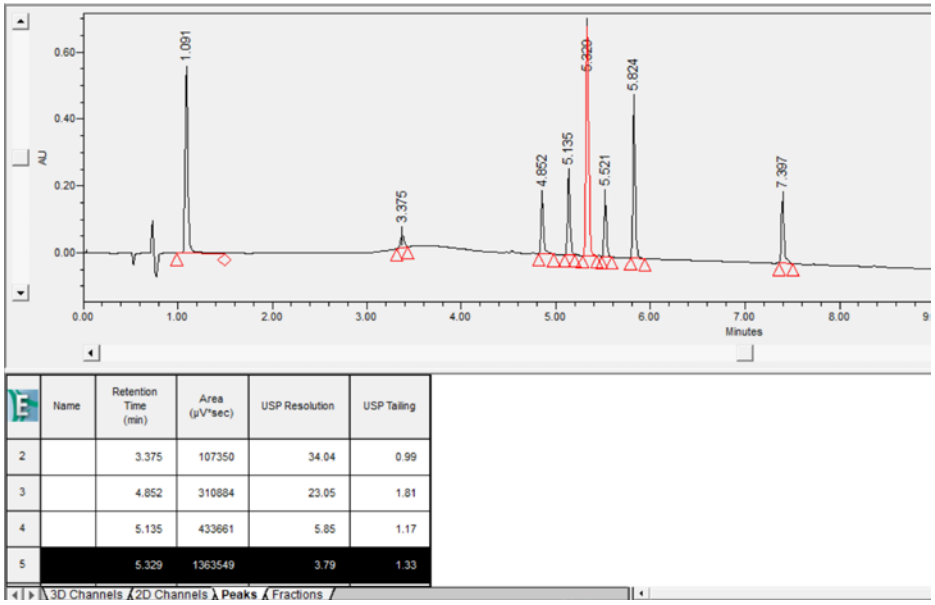
Predicted Results

Response Name	Predicted Result	Observed Result	-2 Sigma Conf. Limit	+2 Sigma Conf. Limit
Rs-Map Response	3.756	3.920		
B - Rs-Map USP Resolution	0.000	0.000		
G - Rs-Map USP Resolution	37.981	37.521		
H* - Rs-Map USP Resolution	13.532	13.541		
F - Rs-Map USP Resolution	11.242	11.781		
A - Rs-Map USP Resolution	7.539	7.649		
API - Rs-Map USP Resolution	3.760	3.920		
D-Deg - Rs-Map USP Resolution	3.756	3.945		
E - Rs-Map USP Resolution	6.465	6.899		
D - Rs-Map USP Resolution	29.898	30.575		
B - RetentionTime	1.27	1.27	1.25	1.29



Internal Verification – Predicted vs. Observed

External Verification – Export to CDS



	Name	Retention Time (min)	Area (µV*sec)	USP Resolution	USP Tailing
2		3.375	107350	34.04	0.99
3		4.852	310884	23.05	1.81
4		5.135	433661	5.85	1.17
5		5.329	1363549	3.79	1.33

S-Matrix®

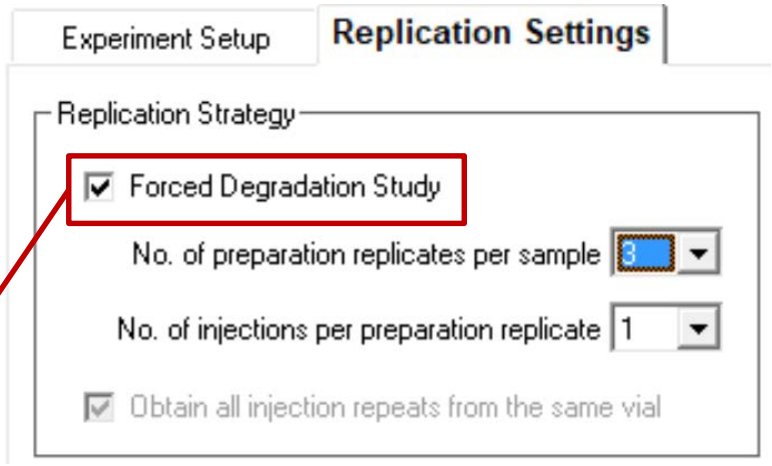
Presentation Outline

1. Rapid Chemistry System Screening
    a. Automation Makes it Simple
    b. Automation Maintains Data Integrity
2. Robust Method Optimization
    a. UV & MS Spectra Based Peak Tracking
    b. Robustness Modeling Throughout MODR
3. New Degradants Discovered
    a. Automated Forced Degradation Study
    b. Method Re-optimization

S-Matrix®

Fusion QbD – *Forced Degradation Studies*

Fusion QbD enables users to set up an experiment execution protocol in which each experiment run will be repeated according to user-specified Replication Strategy which can be any combination of Sample Preparation and/or Injection replicates – with the replicates taken from the same vial or assigned different vials.



S-Matrix has now created a new "Forced Degradation Study" mode to support experiments in which the different Sample Preparation replicates represent different degradation paths such as photo degradation, acid degradation, and peroxide degradation. This activates an entirely new technology which aggregates peak data from the replicates for each run into a "Composite Chromatogram" data set for the run to be used in robust method optimization and prediction chromatogram visualization.

S-Matrix®

New Degradants Identified – Forced Degradation Study

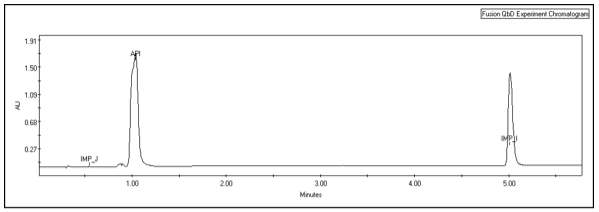
Forced Degradation Studies – Full Automation Support

Experiment Design Matrix

Run No.	Pump Flow Rate (mL/min)	Gradient Time (min)	Oven Temperature (°C)	pH
Condition Column - 1	0.400	2.0	35.0	3.20
1.a	0.300	5.0	35.0	3.20
1.b	0.300	5.0	35.0	3.20
1.c	0.300	5.0	35.0	3.20
2.a	0.500	5.0	35.0	3.20
2.b	0.500	5.0	35.0	3.20
2.c	0.500	5.0	35.0	3.20
3.a	0.300	15.0	35.0	3.20
3.b	0.300	15.0	35.0	3.20
3.c	0.300	15.0	35.0	3.20

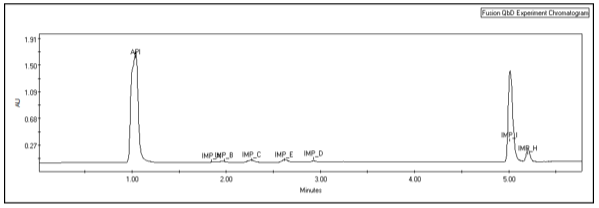
Vial a

Photo Degraded Sample (Vial a)



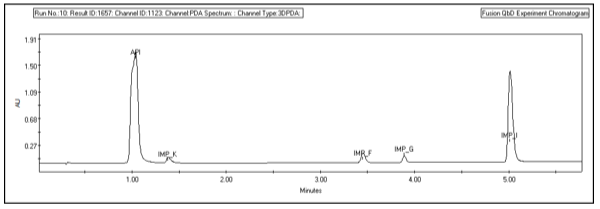
Vial b

Acid Degraded Sample (Vial b)



Vial c

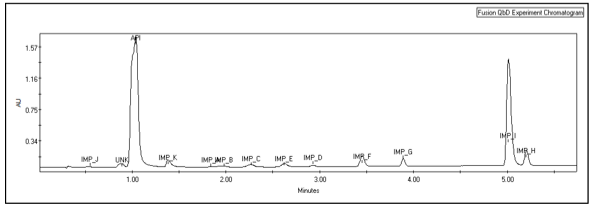
Peroxide Degraded Sample (Vial c)



Full Automation Support

- ✓ Each experiment run is replicated for each degradation path sample.
- ✓ Each peak is tracked in each degradation path sample chromatogram.
- ✓ All peaks from all degradation path sample chromatograms are aggregated into one chromatogram for the run.

Composite Chromatogram – Run 1



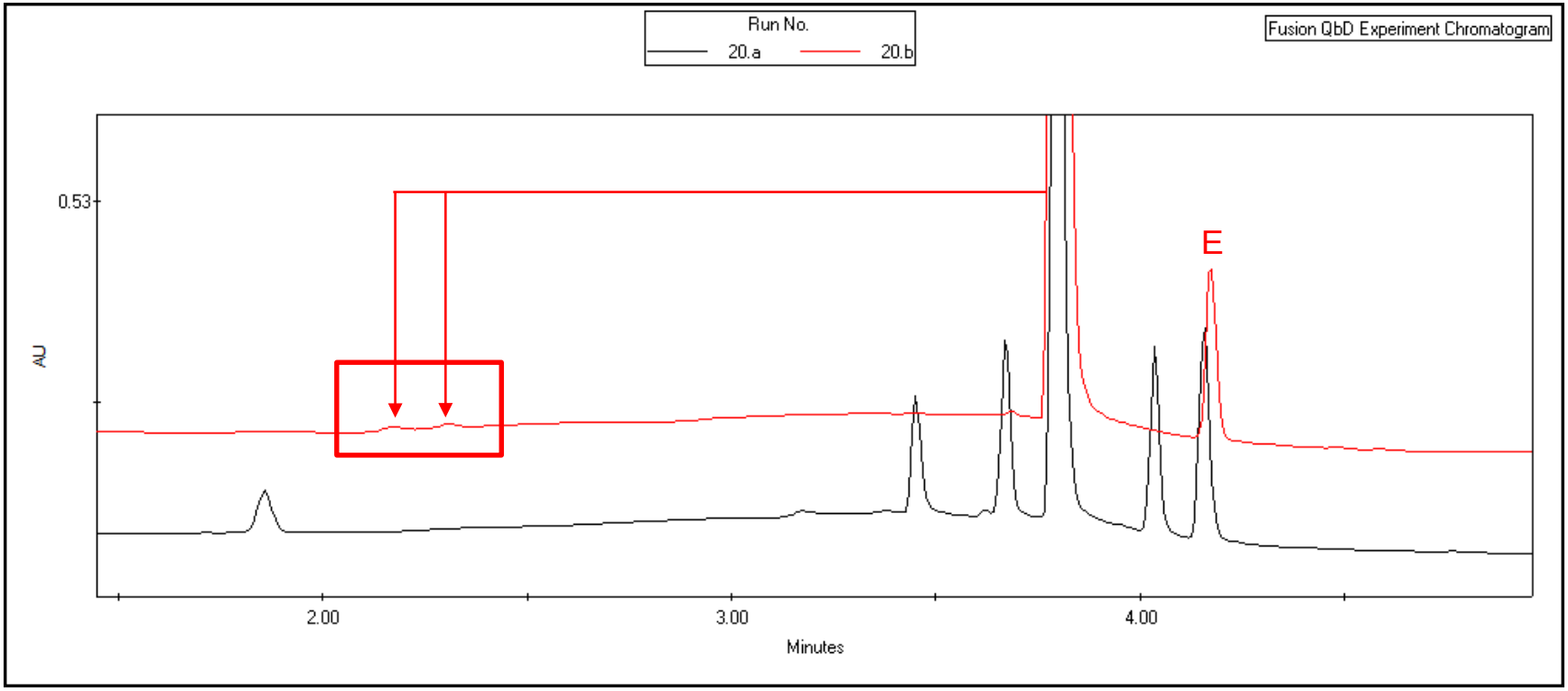
S-Matrix®

Forced Degradation Study (FDS) – 2 New Degradants Identified

Black Trace – Undegraded Sample (Note – API material contained Related Compound “E”)

Red Trace – Peroxide Degraded API (generated 2 new degradants)

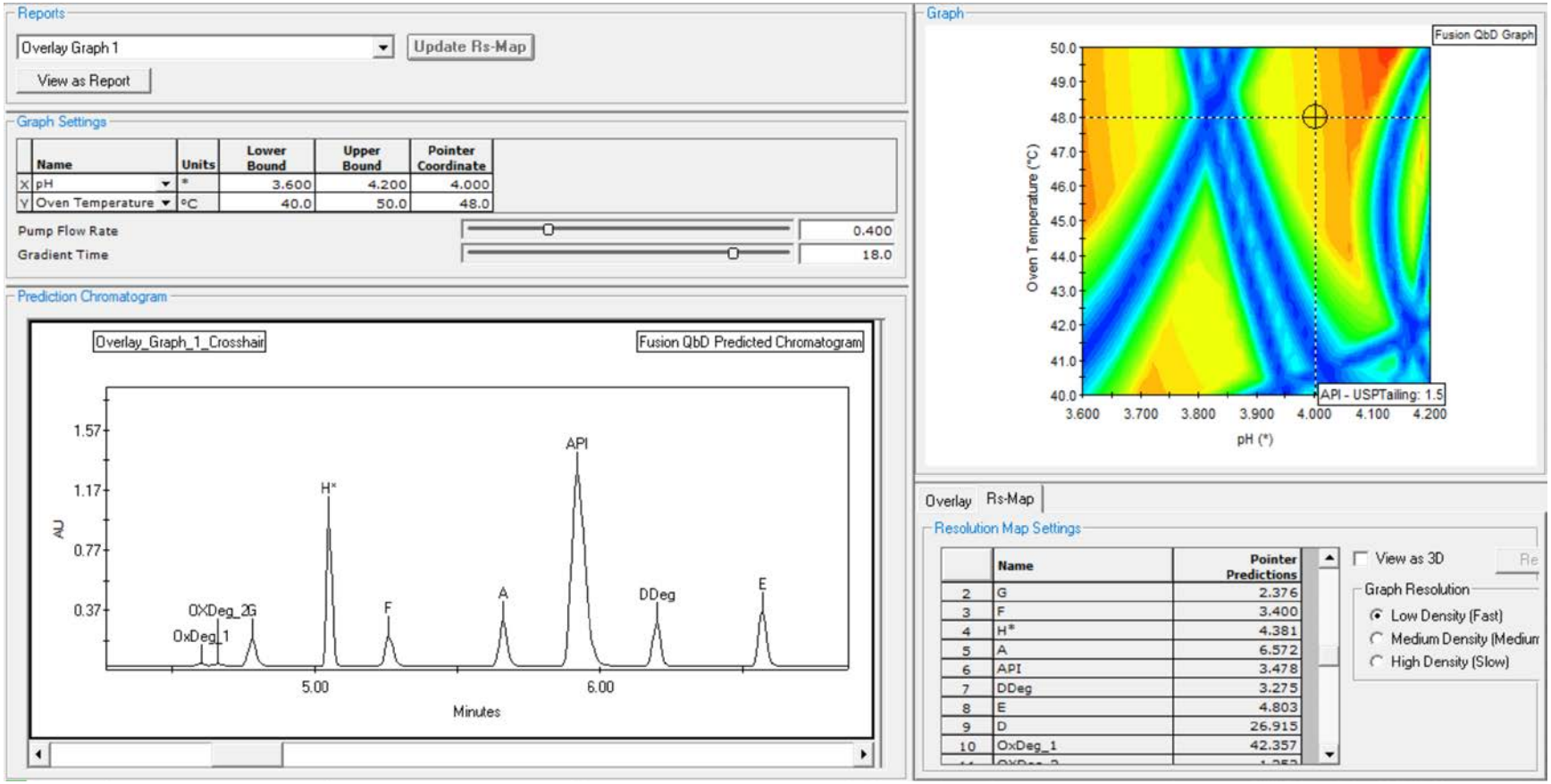
Chromatogram formatting – zoom, stack, offset, and annotate to get best presentation view.



S-Matrix®

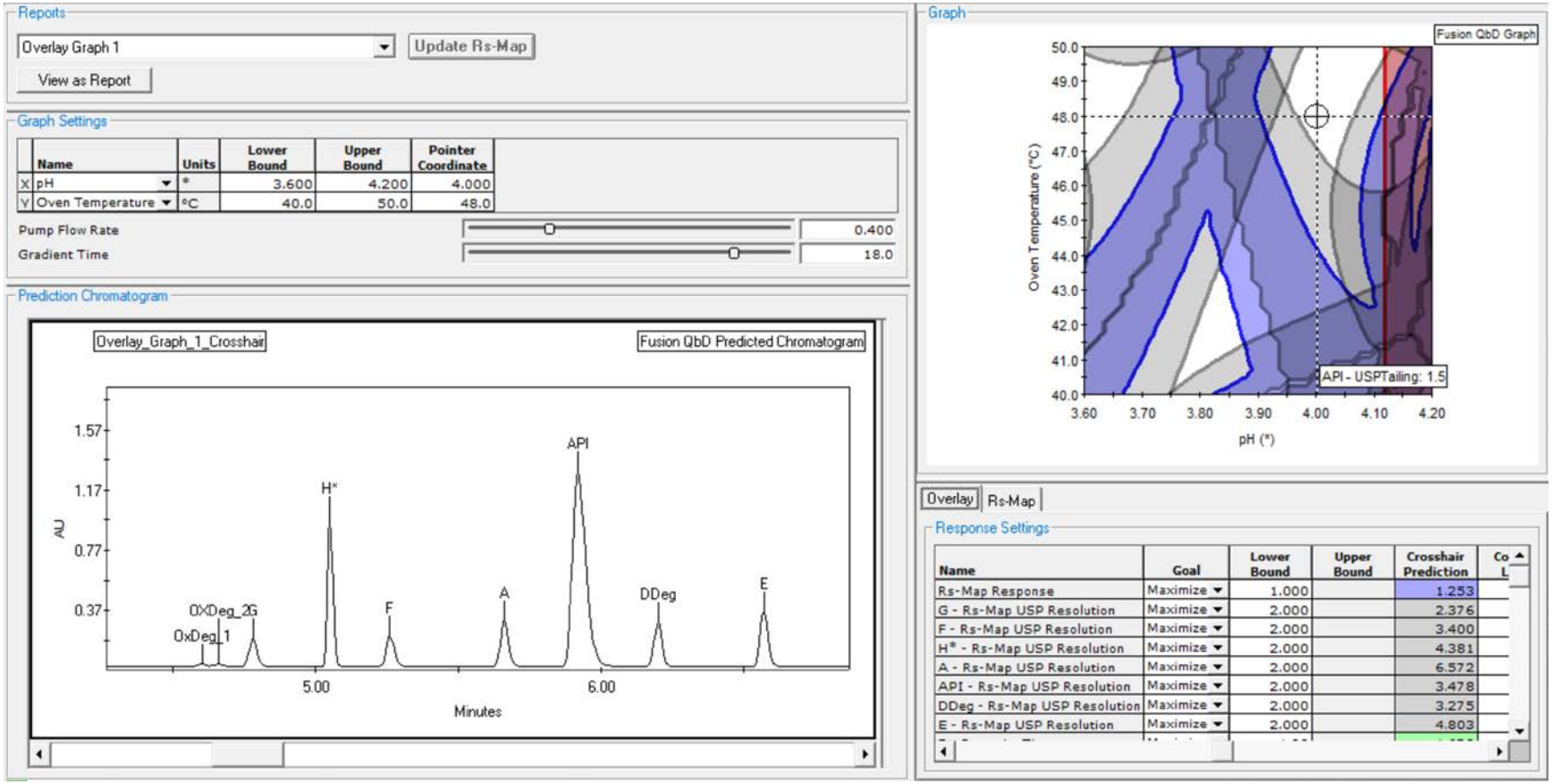
New Degradants Identified – Forced Degradation Study (FDS)

Fusion QbD automatically creates a data set of composite chromatograms for peak tracking, modeling, and simulation.



New Degradants Identified – Forced Degradation Study (FDS)

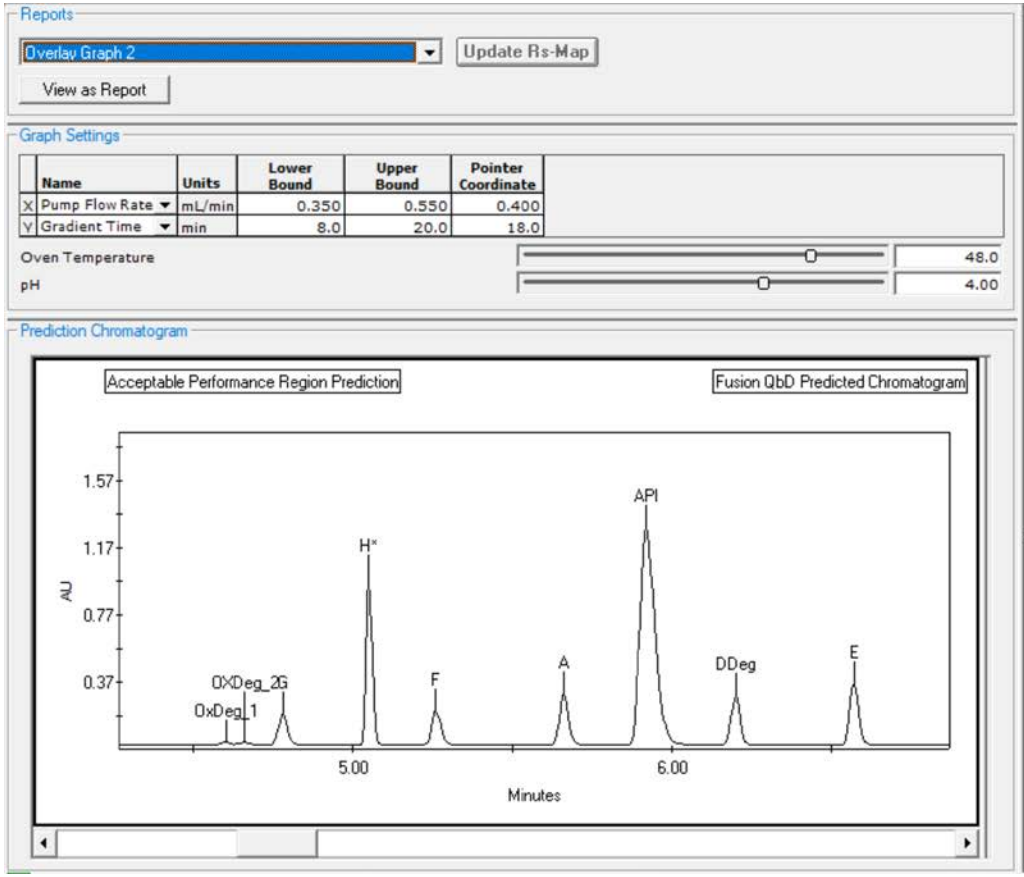
Overlay lets you include mean performance and robustness for any critical performance characteristic across the MODR.

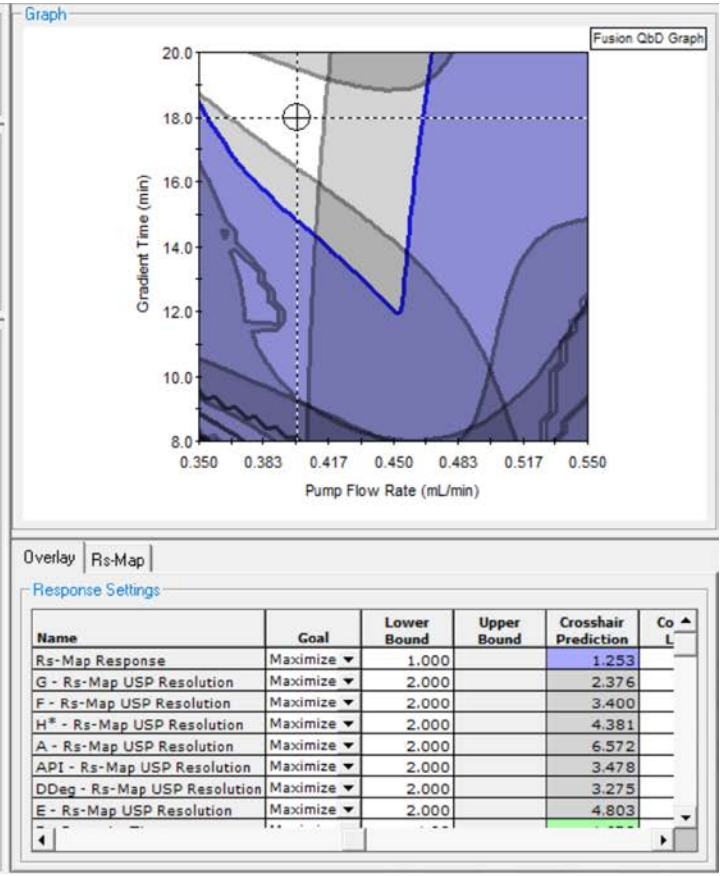


S-Matrix®

New Degradants Identified – Forced Degradation Study (FDS)

Switch graphed and non-graphed variables – find most robust method overall.





S-Matrix®

Fusion QbD – Completely Aligned with ICH Q8(R2)

ICH Q8(R2) – Page 22

C. Presentations of Design Space

Example 1: Response graphs for dissolution are depicted as a surface plot (Figure 1a) and a contour plot (Figure 1b). Parameters 1 and 2 are factors of a granulation operation that affect the dissolution rate of a tablet (e.g., excipient attribute, water amount, granule size.)

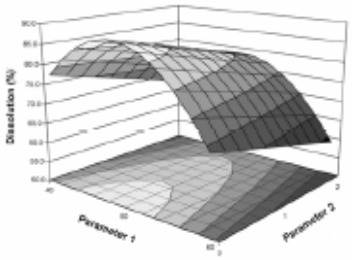


Figure 1a: Response surface plot of dissolution as a function of two parameters of a granulation operation. Dissolution above 80% is desired.

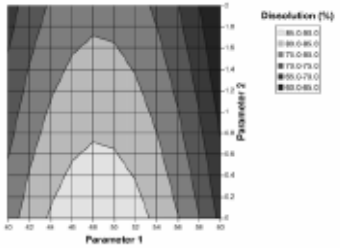


Figure 1b: Contour plot of dissolution from example 1a.

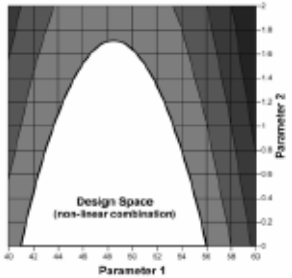


Figure 1c: Design space for granulation parameters, defined by a nonlinear combination of their ranges, that delivers satisfactory dissolution (i.e., >80%).

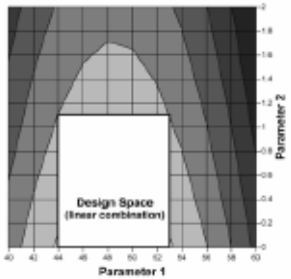


Figure 1d: Design space for granulation parameters, defined by a linear combination of their ranges, that delivers satisfactory dissolution (i.e., >80%).

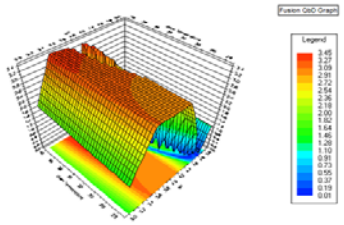
Fusion QbD

Name: Administrator
Company: S-Matrix
Project: Project 1
Date: 08 APR 2018 18:38:09 PDT [UTC-07:00]

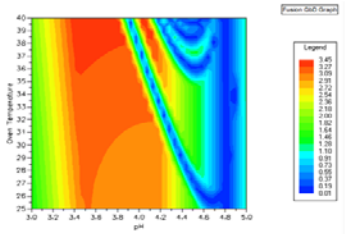
S-Matrix

Final Design Space + PARs

Rs-Map Response Surface Graph

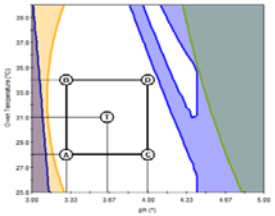


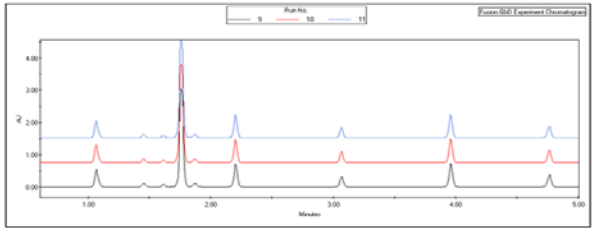
Rs-Map Response Contour Graph



Response	Performance Goal	Color
API – USP Resolution	> 2.00	Red
API – USP Tailing	> 2.00	Blue
Impurity A – USP Resolution	> 2.00	Blue
Impurity B – USP Resolution	> 2.00	Green
API – USP Resolution - Cpk	> 1.33	Orange
Impurity A – USP Resolution - Cpk	> 1.33	Teal
Impurity B – USP Resolution - Cpk	> 1.33	Purple
API – USP Tailing - Cpm	> 1.33	Sky

Multi-Response Overlay with Design Space





1 of 3

S-Matrix

END

S-Matrix®

www.smatrix.com