



2019 EASTERN ANALYTICAL SYMPOSIUM & EXPOSITION

Crowne Plaza Princeton  
Conference Center  
Plainsboro, NJ  
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ENHANCING ANALYTICAL CHEMISTRY  
WITH SUSTAINABLE SOLUTIONS

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



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# 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

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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 =  $5 \times 5 \times 4 = 100$  methods

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

**> 3x efficiency.**

## 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.**

# Initial Chemistry Screening – Built In pH Automation

Buffer Selector...      pH Online Blending Mode: One Acid Base Pair       pKa of Primary Compound

pH Buffer Settings:      No. of Levels: 5

Buffer	Buffer Name	pH Level	Acid %	Base %
Acid	Formic Acid (20 mM)	2.75	100.0	0.0
Base	Ammonium Formate (20 mM)	3.16	80.0	20.0
		3.70	50.0	50.0
		4.34	20.0	80.0
		5.42	5.0	95.0
		6.24	0.0	100.0

Buffer Selector

Select Buffer System  
pH 2.75 - 6.24 [Formate System (20 mM)]

Buffer Solutions

Formic Acid (20 mM)

Ammonium Formate (20 mM)

Included	pH	Formic Acid (%)	Ammonium Formate (%)
<input checked="" type="checkbox"/>	2.75	100.00	0.00
<input checked="" type="checkbox"/>	2.78	95.00	5.00
<input checked="" type="checkbox"/>	2.89	90.00	10.00
<input checked="" type="checkbox"/>	3.16	80.00	20.00
<input checked="" type="checkbox"/>	3.38	70.00	30.00
<input checked="" type="checkbox"/>	3.54	60.00	40.00
<input checked="" type="checkbox"/>	3.70	50.00	50.00
<input checked="" type="checkbox"/>	3.88	40.00	60.00
<input checked="" type="checkbox"/>	4.06	30.00	70.00
<input checked="" type="checkbox"/>	4.34	20.00	80.00
<input checked="" type="checkbox"/>	4.91	10.00	90.00
<input checked="" type="checkbox"/>	5.42	5.00	95.00
<input checked="" type="checkbox"/>	6.24	0.00	100.00

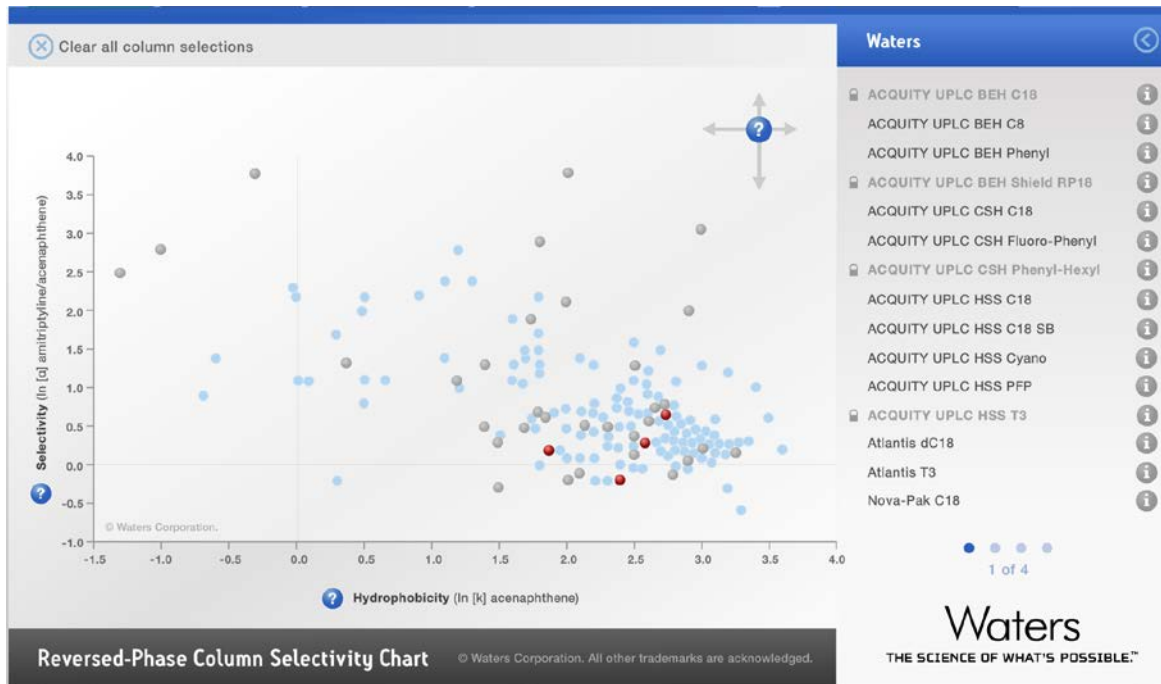
OK      Cancel

Built in Buffer System  
Titration Curves

Extremely Precise!



# 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

# Fusion QbD Automation – Supports All Waters LC Systems



- ✓ Solvent Selection Valves
- ✓ Column Switching Valves

Alliance HPLC



Acquity Binary



Acquity H-Class



Acquity Arc



Acquity UPC<sup>2</sup>





# Fusion QbD Automation – Supports All Agilent LC Systems



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



# Fusion QbD Automation – Supports Thermo LC Systems



- ✓ Solvent Selection Valves
- ✓ Column Switching Valves

## UltiMate LCs



## Vanquish Horizon And Flex LCs

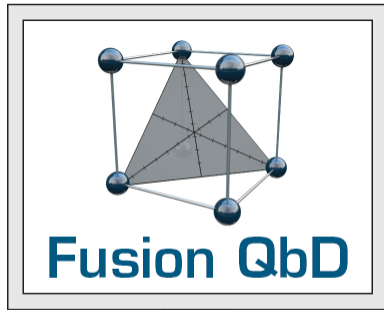


## **FDA Statement –**

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)**



Fusion QbD

Generates QbD-aligned  
DOE Experiment

Automatically Builds  
Sequence and All  
Instrument Methods

## Chromatography Data Software (CDS)

Text Mix pH in S-Matrix - MD DemoLC 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.**

## 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

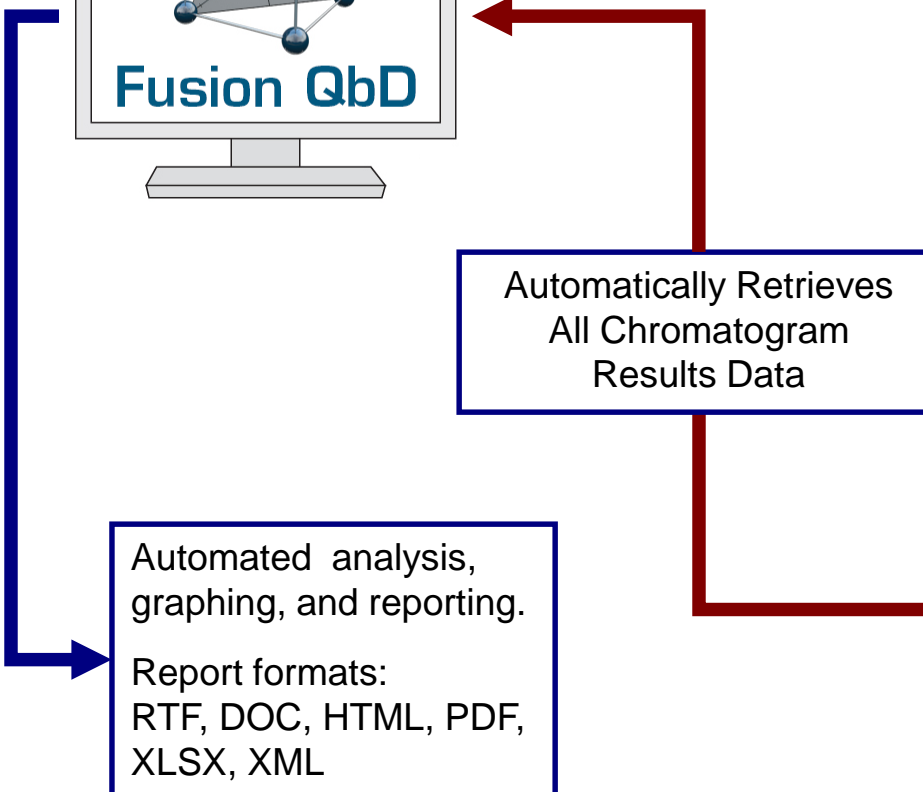
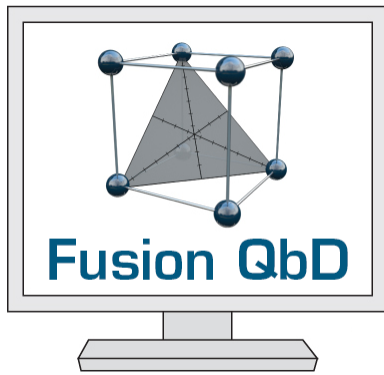
The screenshot displays the S-Matrix software interface. A 'Method Properties' dialog box is open, showing details for a method named 'AAA\_Demo'. The dialog includes a 'Lock' button and fields for 'Locked By' and 'Being Edited By'. Below this, a 'Method History' table shows a single entry for 'AAA\_Demo' created by 'Fusion QbD: C:\Program Files'. The background shows a 'Methods' table with 11 rows of method entries, including names like 'AAA\_Demo 001\_001' and 'AAA\_Demo 001\_005', their types (Sample Set, Method Set, Instrument), and timestamps. The status bar at the bottom indicates '166 total'.

Method Name	Method Type	
1 AAA_Demo	Sample Set	7/17/2018
2 AAA_Demo 001_001	Method Set	7/17/2018
3 AAA_Demo 001_001	Instrument	7/17/2018
4 AAA_Demo 001_002	Method Set	7/17/2018
5 AAA_Demo 001_002	Instrument	7/17/2018
6 AAA_Demo 001_003	Method Set	7/17/2018
7 AAA_Demo 001_003	Instrument	7/17/2018
8 AAA_Demo 001_004	Method Set	7/17/2018 8:28:16 AM PDT
9 AAA_Demo 001_004	Instrument	7/17/2018 8:28:16 AM PDT
10 AAA_Demo 001_005	Method Set	7/17/2018 8:28:20 AM PDT
11 AAA_Demo 001_005	Instrument	7/17/2018 8:28:19 AM PDT

Method Name	Method Type	
1 AAA_Demo	Sample Set	Created by Fusion QbD: C:\Program Files

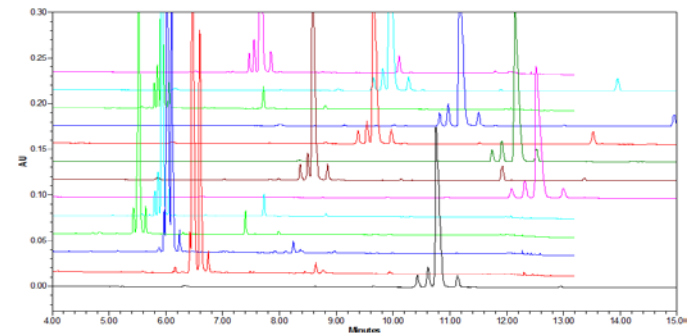
## Audited Data Exchange – Assures Data Integrity

# Fusion QbD Automation – Maintains Data Integrity



## Chromatography Data Software (CDS)

Run	File No.	# of PIP	Label	Sample Name	Lot#	Function	Method Set / Report Method	Label Reference	Processing	Run Time (minutes)	QMS Start (minutes)	QMS End (minutes)	Column Position	Auto Addition	Sample/Injct	Column
11						Condition Column	Test file pH 01_217			8.70	0.00	0.00	No Change			
21						Condition Column	Test file pH 01_217			0.10	0.00	0.00	No Change			
31						Equilibrate	Test file pH 01_217			3.00	0.00	0.00	No Change			
41	1	2.0	1	UPL-001-001	2.a.1.a	Injct Samples	Test file pH 01_217		Normal	10.50	0.00	1.50	No Change		1.00000	1.00000
51						Condition Column	Test file pH 01_201			0.10	0.00	0.00	No Change			
61						Equilibrate	Test file pH 01_201			3.00	0.00	0.00	No Change			
71	2	2.0	1	UPL-001-001	1.a.1.a	Injct Samples	Test file pH 01_201		Normal	10.50	0.00	1.50	No Change		1.00000	1.00000
81						Condition Column	Test file pH 01_202			0.10	0.00	0.00	No Change			
91						Equilibrate	Test file pH 01_202			3.00	0.00	0.00	No Change			
101	2	2.0	1	UPL-001-002	2.a.1.a	Injct Samples	Test file pH 01_202		Normal	10.50	0.00	1.50	No Change		1.00000	1.00000
111						Condition Column	Test file pH 01_202			8.70	0.00	0.00	No Change			
121						Condition Column	Test file pH 01_202			0.10	0.00	0.00	No Change			
131						Equilibrate	Test file pH 01_202			3.00	0.00	0.00	No Change			
141	2	2.0	1	UPL-001-001	2.a.1.a	Injct Samples	Test file pH 01_202		Normal	10.50	0.00	1.50	No Change		1.00000	1.00000
151						Condition Column	Test file pH 01_204			8.70	0.00	0.00	No Change			
161						Condition Column	Test file pH 01_204			0.10	0.00	0.00	No Change			
171						Equilibrate	Test file pH 01_204			3.00	0.00	0.00	No Change			
181	2	2.0	1	UPL-001-004	1.a.1.a	Injct Samples	Test file pH 01_204		Normal	10.50	0.00	1.50	No Change		1.00000	1.00000
191						Condition Column	Test file pH 01_205			0.10	0.00	0.00	No Change			
201						Equilibrate	Test file pH 01_205			3.00	0.00	0.00	No Change			
211	2	2.0	1	UPL-001-005	1.a.1.a	Injct Samples	Test file pH 01_205		Normal	10.50	0.00	1.50	No Change		1.00000	1.00000
221						Condition Column	Test file pH 01_205			8.70	0.00	0.00	No Change			
231						Condition Column	Test file pH 01_205			0.10	0.00	0.00	No Change			
241						Equilibrate	Test file pH 01_205			3.00	0.00	0.00	No Change			



**Eliminate Transcription Errors.  
Maintain Data in Audited Environment.**

# Fusion QbD Automation – Maintains Data Integrity

LC Method Development - LC Method Development Tutorial - Screening.smae

File Edit Activity Tools Window Help

Generate Audit Log

Design of Experiments

- Create a Design
- Design Reports

Data Entry / Analysis

- Data Entry
- Data Analysis

Best Answer Searches

- Best Overall Answer
- Acceptable Performance Region
- Point Predictions

Visualization Graphics

- Single Response Series
- Multiple Response Series

Reporting Toolkit

- Fusion Reporter
- Audit Log Reporter**

**Name: Administrator**  
**Company: S-Matrix Corporation**  
**Project: Project 1**  
**Date: 17 NOV 2019 14:04:21 PST [UTC-08:00]**

**Audit Log**

14 NOV 2019 10:21:21 PST [UTC-08:00] - Administrator

Event Type: Import Responses

**Import Response Settings**

Setting	Value
Target CDS	EMPOWER
Empower Version	Empower 3 Software Build 3471 SPs Installed: Feature Release 4 DB ID: 266003392
Empower Database	(local)
Empower User	system
Project Name	RD1 - Screen - 9_9_0
Result Set(ID)	FMD_RD1_Screen (1485)
Processed Channel	PDA Ch1 225nm@4.8nm, Time offset by 0.020 mins.
Activate PeakTracker	Unchecked
Auto-imported Response(s)	Height, RetentionTime, WidthAt150Pct, USPTailing, WidthAtTangentUSPResolution, Area
Trend Response(s)	USPResolution
Import Chromatogram Trace Data	Checked
Import Prediction Chromatogram Data	Checked
Total Import Time	00:00:18
Locale	English (United States)

**Response Data**

Run No.	Result ID	No. of Peaks	No. of Peaks >= 1.50 - USPResolution	No. of Peaks >= 2.00 - USPResolution	No. of Peaks <= 1.50 - USPTailing
1	1748	8	2	2	0
2	1750	6	3	3	0
3	1826	8	5	5	3
4	1824	8	4	4	2
5	1756	7	2	2	0

1 of 6

Ready modified

## Audited Data Exchange – Assures Data Integrity



## 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

# Chemistry Screening Study – Acceptable Performance Region

LC Method Development - LC Method Development Tutorial - Screening.smae

File Edit Activity Tools Window Help

Create Report Update Report Delete Report Restore Report Robustness Simulator Show Prediction Chromatogram

Design of Experiments

- Create a Design
- Design Reports

Data Entry / Analysis

- Data Entry
- Data Analysis

Best Answer Searches

- Best Overall Answer
- Acceptable Performance Region**
- Point Predictions

Visualization Graphics

- Single Response Series
- Multiple Response Series

Reporting Toolkit

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Reports

Best Overall Answer

View as Report

Graph Settings

Name	Units	Lower Bound	Upper Bound	Pointer Coordinate
X Gradient Time	min	10.0	25.0	24.0
Y pH	#	2.73	4.93	4.00

Column Type

CSH Phenyl Hexyl

BEH C18

BEH Shield RP18

HSS T3

CSH Phenyl Hexyl

Verification Run Settings

Include Proven Acceptable Ranges (PARs)

Include Verification Runs

Graph

Overlay | Ris-Map

Response Settings

Name	Goal	Lower Bound	Upper Bound	Crosshair Prediction	Contour Label	Color
No. of Peaks	Target	7.0	9.0	7.75		Gray
No. of Peaks >= 1.50 - USResolution	Target	6.0	8.0	6.23		Red
No. of Peaks >= 2.00 - USResolution	Target	6.0	8.0	6.05		Blue
No. of Peaks <= 1.50 - USPTailing	Target	7.0	9.0	7.77		Green
First Peak - RetentionTime	Maximize	0.90		1.05		Orange

Validation Status: Your settings are valid.

Ready

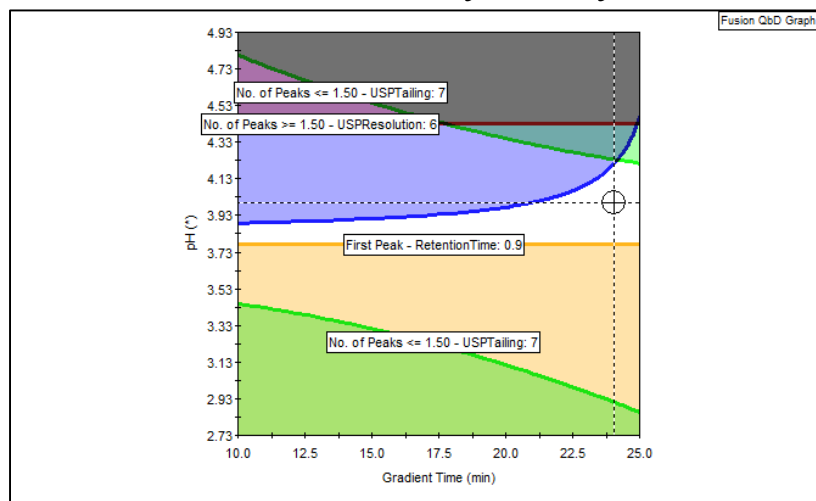
modified

Each response is assigned a color.

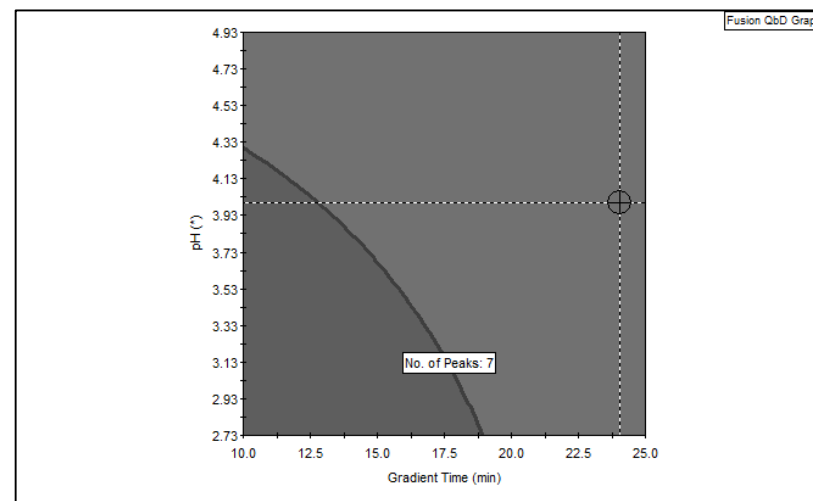
Shaded Region for the color identifies methods which fail to meet performance requirements.

# Chemistry Screening Study – Acceptable Performance Region

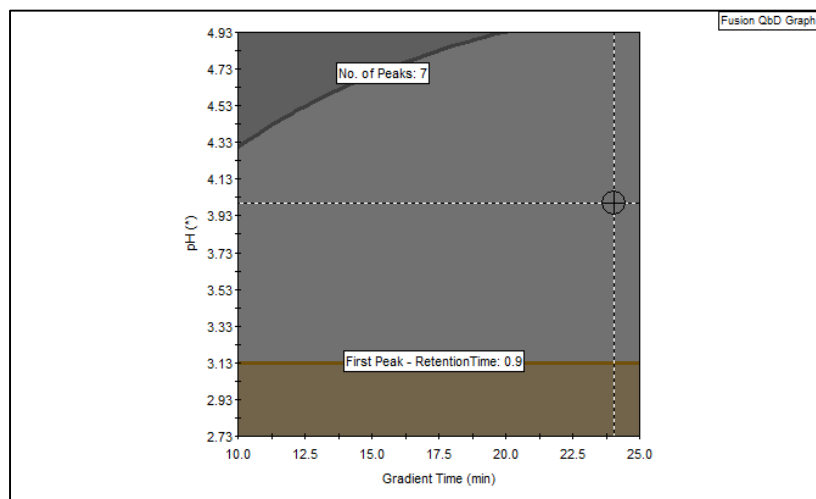
## CSH Phenyl Hexyl



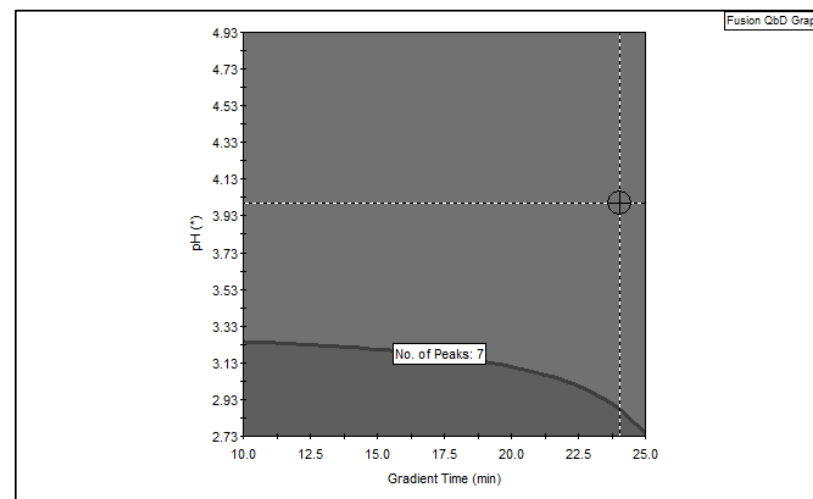
## BEH C18



## BEH Shield RP18



## HSS T3



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3. New Degradants Discovered
  - a. Automated Forced Degradation Study
  - b. Method Re-optimization

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 =  $3 \times 3 \times 3 \times 3 = 81$  methods

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

**> 3x efficiency.**

\* – levels adjusted for new endpoint of 50% organic

# Fusion QbD – PeakTracker™

Select a Project and Result Set

Select Project

Find Filter Reset

- Projects
  - Customers
  - Distributors
  - S-Matrix
    - ADL
    - FIT
    - FMD Tutorial - 9\_9\_0
      - RD1 - Screen - 9\_9\_0
      - RD2 - 9\_9\_0 - Named Peaks
      - RD2 - Optimization - 9\_9\_0
    - FMV - A\_L\_R
    - Internal Development
    - RD1 - Demo Screening Expt
    - RD2 - Demo Optimization Expt
    - Test

Select Result Set(s)

Result Set Name	ID	Date	Sample Set
RD2 Optimization	9001	6/4/2019 8:48:34 AM EDT	RD2 Optimization

Fetch Selected Result Sets

Select Processed Channel:  
PDA Ch1 225nm@4.8nm, Time offset by 0.020 mins.

Result(s) for Import

Sample	ID	Date	Type	Channel ID	Result
1	9155	6/4/2019 8:50:34 AM	LC	6737	9001
10	9048	6/4/2019 8:48:51 AM	LC	6812	9001
11	9191	6/4/2019 8:57:34 AM	LC	6820	9001
12	9058	6/4/2019 8:48:54 AM	LC	6828	9001
13	9063	6/4/2019 8:48:56 AM	LC	6836	9001
14	9201	6/4/2019 8:58:48 AM	LC	6844	9001
15	9073	6/4/2019 8:48:59 AM	LC	6852	9001
16	9078	6/4/2019 8:49:00 AM	LC	6860	9001

PeakTracker Data Import

Select Raw PDA Channel: None Selected      Select Raw MS Channel: QDa Positive Scan      MS Time Offset (min): 0.02

Spectra Extraction Points:      Threshold Setting

Leading (%) 30.00      Trailing (%) 30.00      MS Intensity 10000

Select Processed MS Channel: QDa Positive Scan MS TIC, Smoothed by 59 point Savitzky-Golay Filter. (QDa Positive(+) Scan (100.00-12'       Track Non-absorbing Peaks

User Types (logged in as 'Owner')

Owner

Ready

Next >> Cancel ?

# Robust Method Optimization – *PeakTracker*

PeakTracker

Run No. 15

UV Chromatogram D: 1241: Channel: PDA Ch1 225nm@4.8nm: Channel Type: 2D Fusion QbD Plot

Total Ion Chromatogram Channel: MS TIC: Channel Type: 2D

Peak Table - PDA Ch1 225nm@4.8nm - Run No. 15

Name	RT (min)	Base Peak (m/z)	AMV (m/z)	Area (uV*sec)	Height (uV)
1	1.037	262.2	262.2	973,202	523,791
2	3.061	193.1	193.1	110,123	37,394
3	4.656	336.8	336.8	296,881	160,622
4	4.926	274.2	274.2	397,548	230,388

Global Tracking Method

Mass UV Options

Display Intensity Columns

Component Name	Expected Mass 1	Expected Mass 2	Expected Mass 3	Expected Mass 4	Expected Mass 5
				279.1	264.2
				407.3	194.1
				278.9	160.2
				126.0	279.1
				279.2	126.1
				126.1	279.0

Select Mode  
 Data Review  Tracking

Global Tracking Method

Update Responses Close

**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:

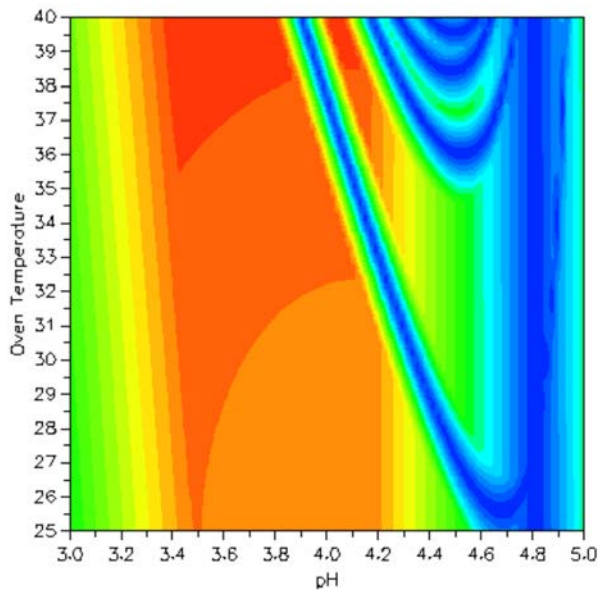
- Auto-deconvolution of partially and completely co-eluted peaks.
- Two or more peaks with identical mass data.
- Non-ionizing and non-absorbing compounds.



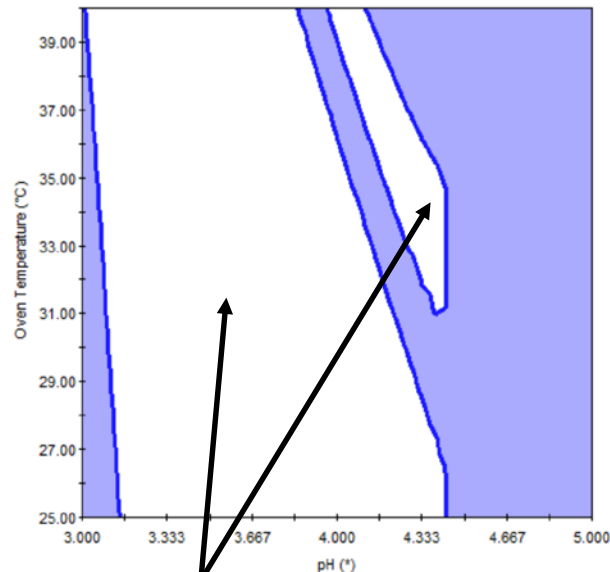
# Fusion QbD – *Rs-Map Response*

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.

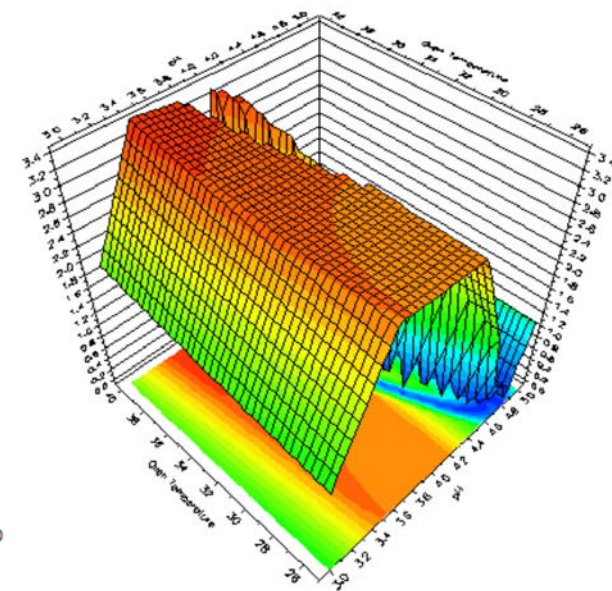
### Contour Graph



### Overlay Graph

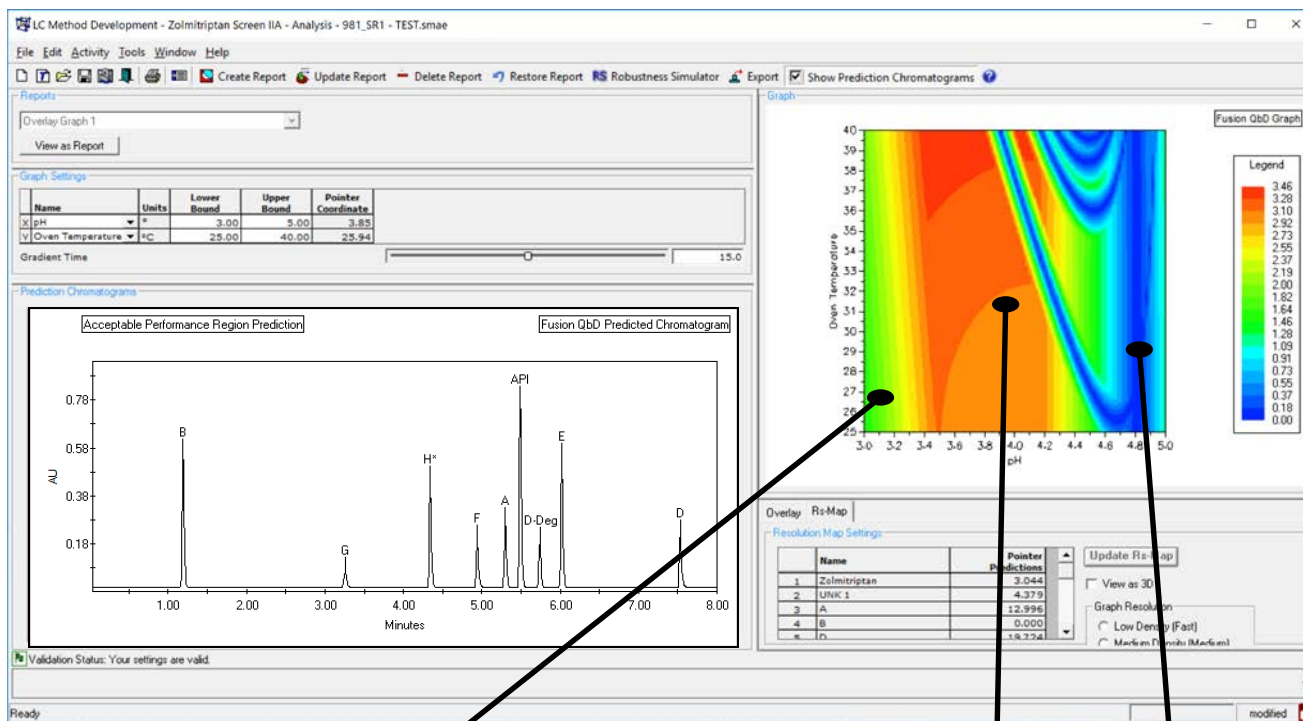


### Response Surface Graph

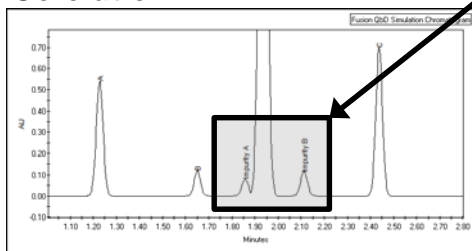




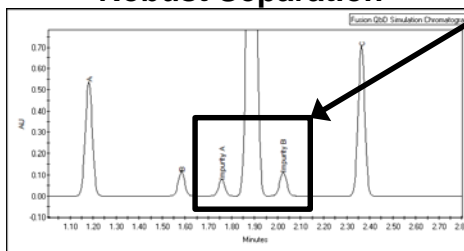
# Robust Method Optimization – Rs-Map Response



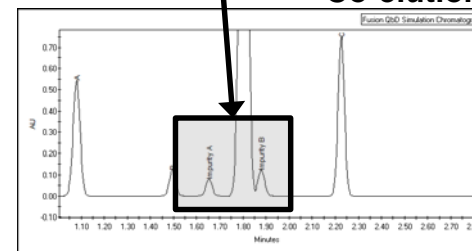
Co-elution



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

# Robust Method Optimization – Robustness Modeling Throughout MODR

The screenshot displays the Robustness Simulator interface. The main window shows response settings for various metrics, and a dialog box titled "Robustness Simulator - Select Rs Responses for Individual Compounds" is open, allowing the user to select specific responses for inclusion.

**Robustness Simulator - Response Settings**

Enabled	Response	Robustness Index	Specification Limit Delta ( $\pm$ )	LSL	USL	Target	Additional Error	Additional Error Amount ( $\pm 1\sigma$ Value)
<input checked="" type="checkbox"/>	B - RetentionTime	Cpk		0.900			<input type="checkbox"/>	
<input checked="" type="checkbox"/>	A - Rs-Map USP Resolution	Cpk			1.500		<input type="checkbox"/>	
<input checked="" type="checkbox"/>	API - Rs-Map USP Resolution	Cpk			1.500		<input type="checkbox"/>	
<input checked="" type="checkbox"/>	D-Deg - Rs-Map USP Resolution	Cpk			1.500		<input type="checkbox"/>	
<input checked="" type="checkbox"/>	E - Rs-Map USP Resolution	Cpk			1.500		<input type="checkbox"/>	

**Robustness Simulator - Select Rs Responses for Individual Compounds**

Include	Response
<input type="checkbox"/>	G - Rs-Map USP Resolution
<input type="checkbox"/>	H* - Rs-Map USP Resolution
<input type="checkbox"/>	F - Rs-Map USP Resolution
<input checked="" type="checkbox"/>	A - Rs-Map USP Resolution
<input checked="" type="checkbox"/>	API - Rs-Map USP Resolution
<input checked="" type="checkbox"/>	D-Deg - Rs-Map USP Resolution
<input checked="" type="checkbox"/>	E - Rs-Map USP Resolution
<input type="checkbox"/>	D - Rs-Map USP Resolution

The dialog box includes buttons for "Select All", "Select None", "OK", and "Cancel". The "OK" button is highlighted with a blue dashed border. A blue arrow points from the "Add Individual Rs Responses..." button in the main window to the dialog box.

**Robustness Simulator - Instructions**

Use  $C_{pk}$  when one of the two cases below applies to the response.

- The response goal is **Maximize**, there is an absolute **Lower** specification limit, and at least some predicted response values **are** near the absolute lower limit.
- The response goal is **Minimize**, there is an absolute **Upper** specification limit, and at least some predicted response values **are** near the absolute upper limit.

**Note:**

- $C_{pi}$  is computed when only a lower specification is entered.

**Robustness Simulator - Status**

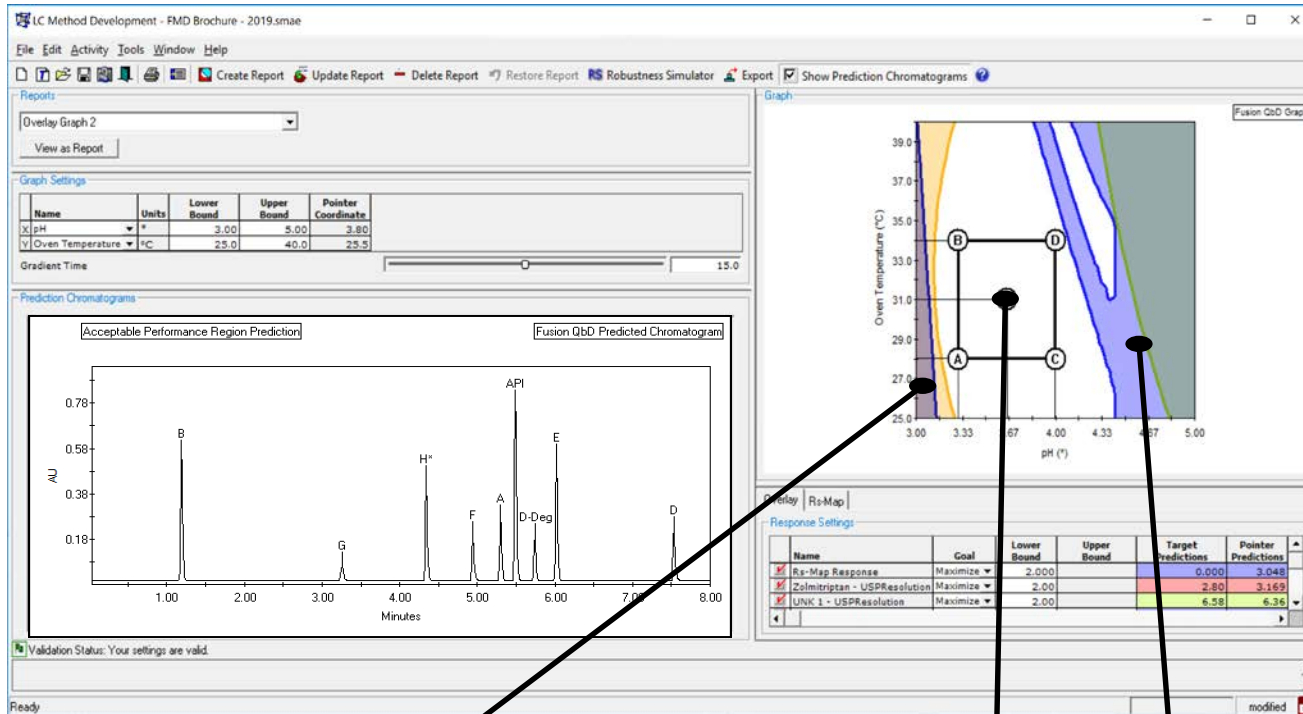
The settings are valid.

Buttons: Back, Finish, Cancel

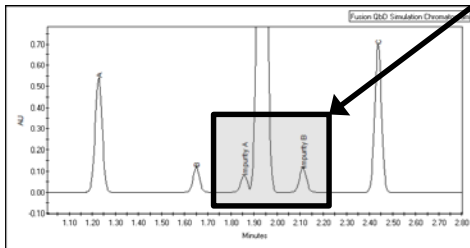
Robustness is calculated for each key performance metric.

# Robust Method Optimization – Robustness Modeling Throughout MODR

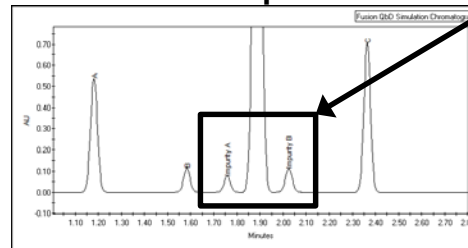
- Mean Performance and Robustness Throughout MODR.
- Include Performance Requirements for Critical Peaks and Peak Pairs.



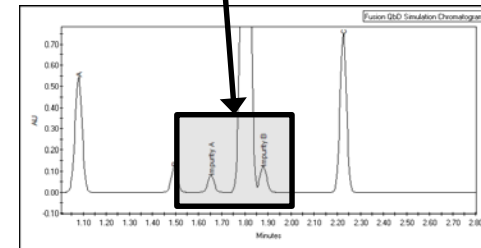
Co-elution



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.

 U.S. Food and Drug Administration  
Protecting and Promoting Public Health www.fda.gov

## Method Operable Design Region

- Analytical method design space
  - Typically Design of Experiments is used to find ranges for instrument operating parameters and understand sources of variation.
  - Method performance criteria are response factors.
  - Can be conducted together with method validation.
- Considerations for implementing MODR
  - Availability of adequate data to support proposed MODR
  - Assess validation criteria across MODR
  - Confirm system suitability throughout MODR

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

## 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  $C_p = C_{pk}$ .

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

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

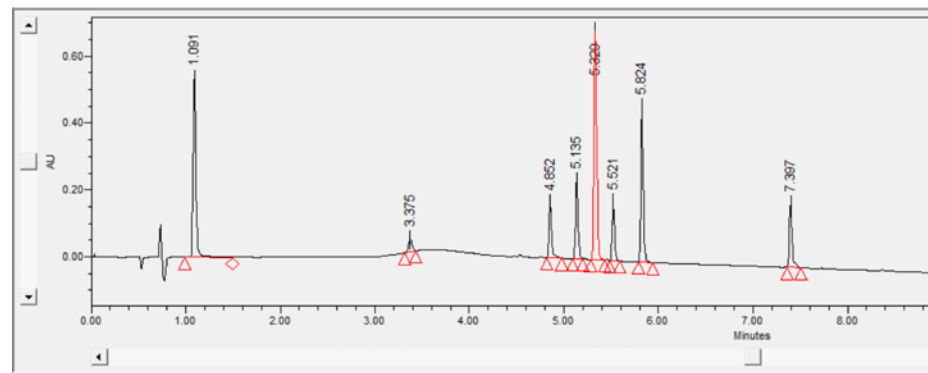
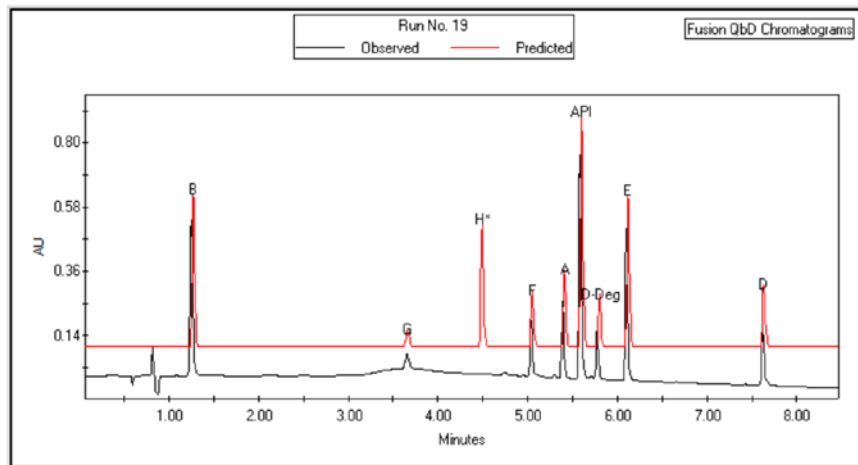
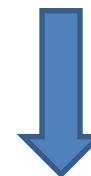


Internal Verification – Predicted vs. Observed

### 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

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

3D Channels 20 Channels Peaks Fractions



# 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

# 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.

Experiment Setup | **Replication Settings**

Replication Strategy

- Forced Degradation Study
- No. of preparation replicates per sample: 3
- No. of injections per preparation replicate: 1
- Obtain all injection repeats from the same vial

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.

# 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

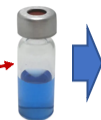
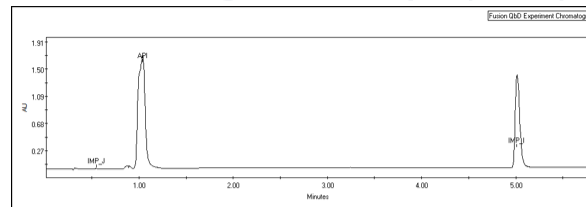
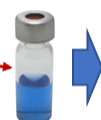


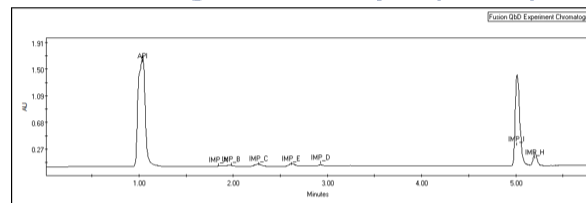
Photo Degraded Sample (Vial a)



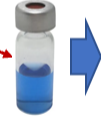
Vial b



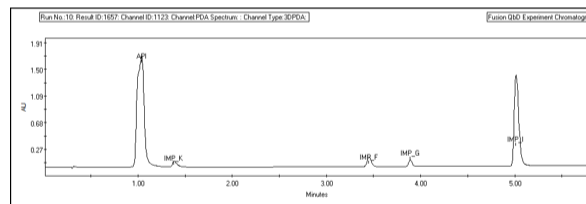
Acid Degraded Sample (Vial b)



Vial c



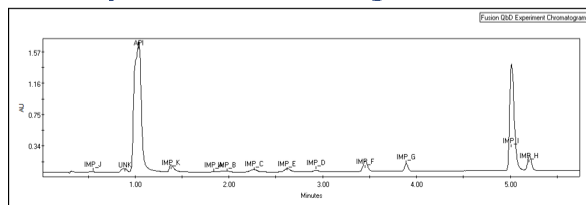
Peroxide Degraded Sample (Vial c)



### 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

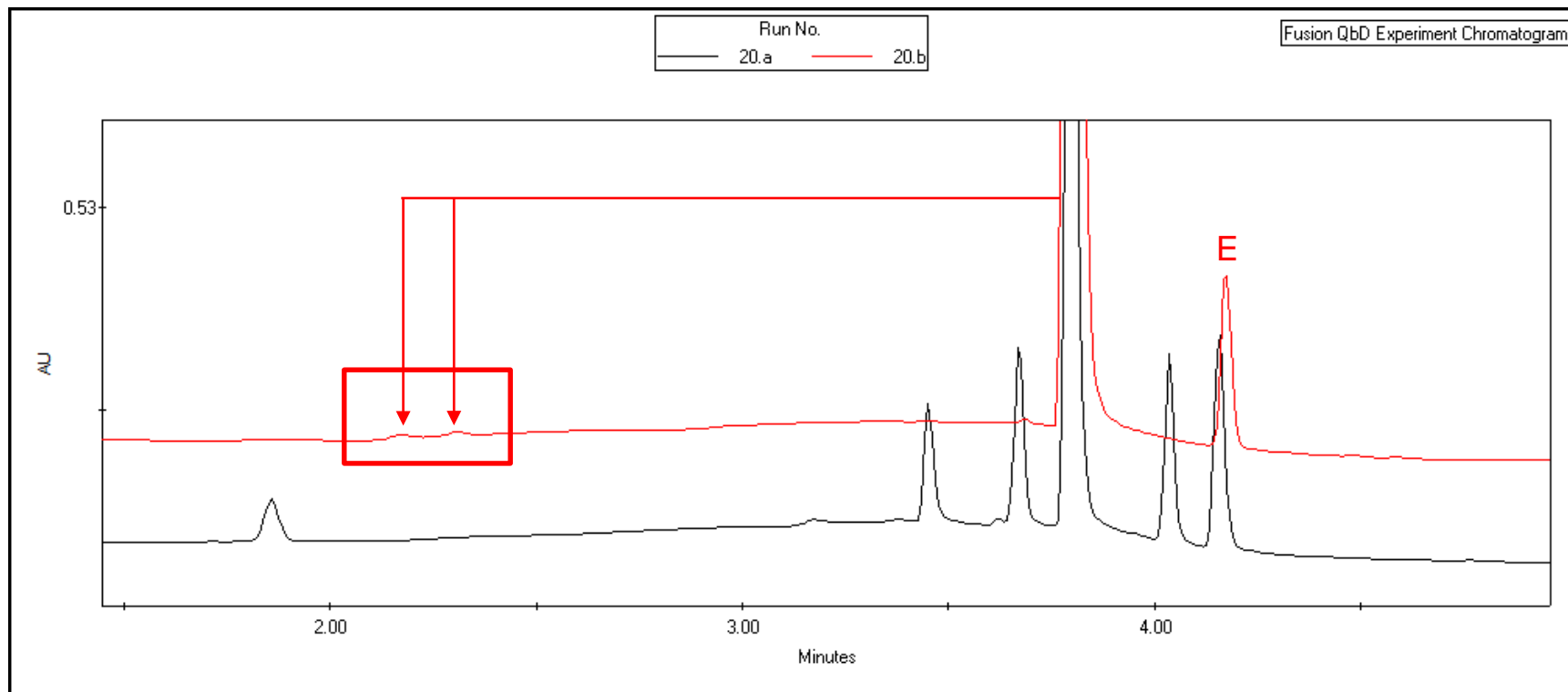


# Forced Degradation Study (FDS) – 2 New Degradants Identified

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

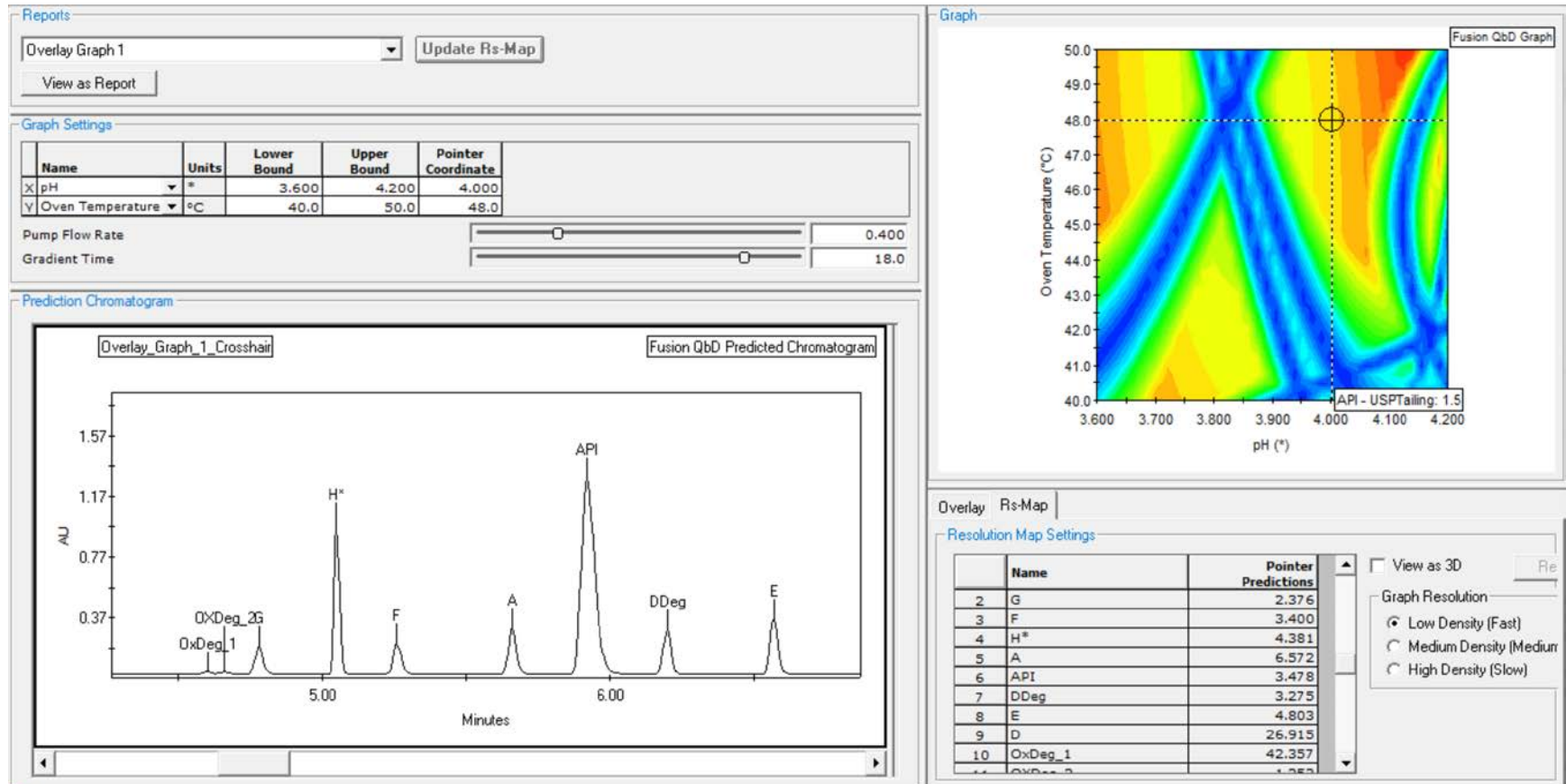
Red Trace – Peroxide Degraded API (generated 2 new degradants)

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



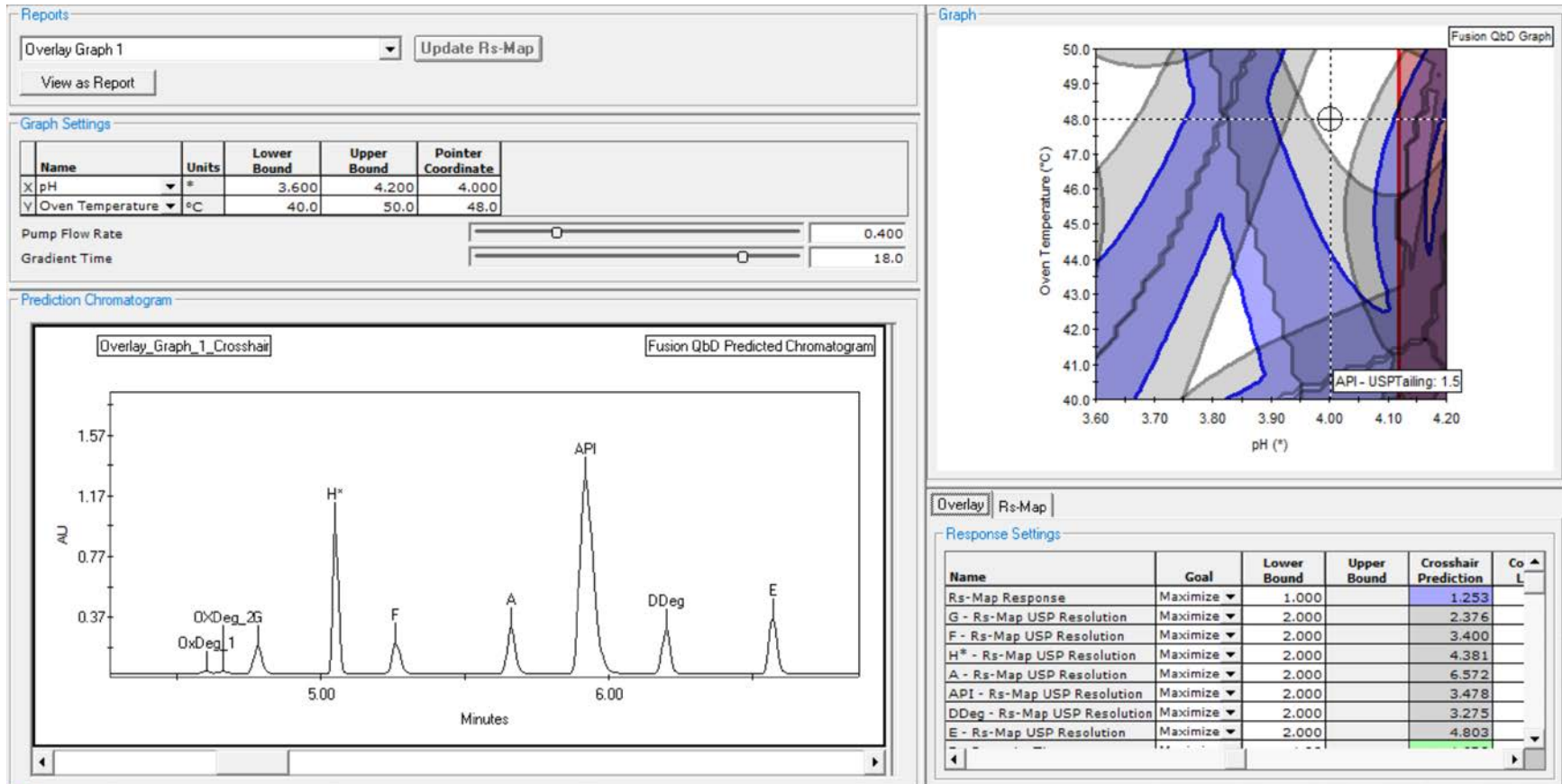
# New Degradants Identified – Forced Degradation Study (FDS)

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



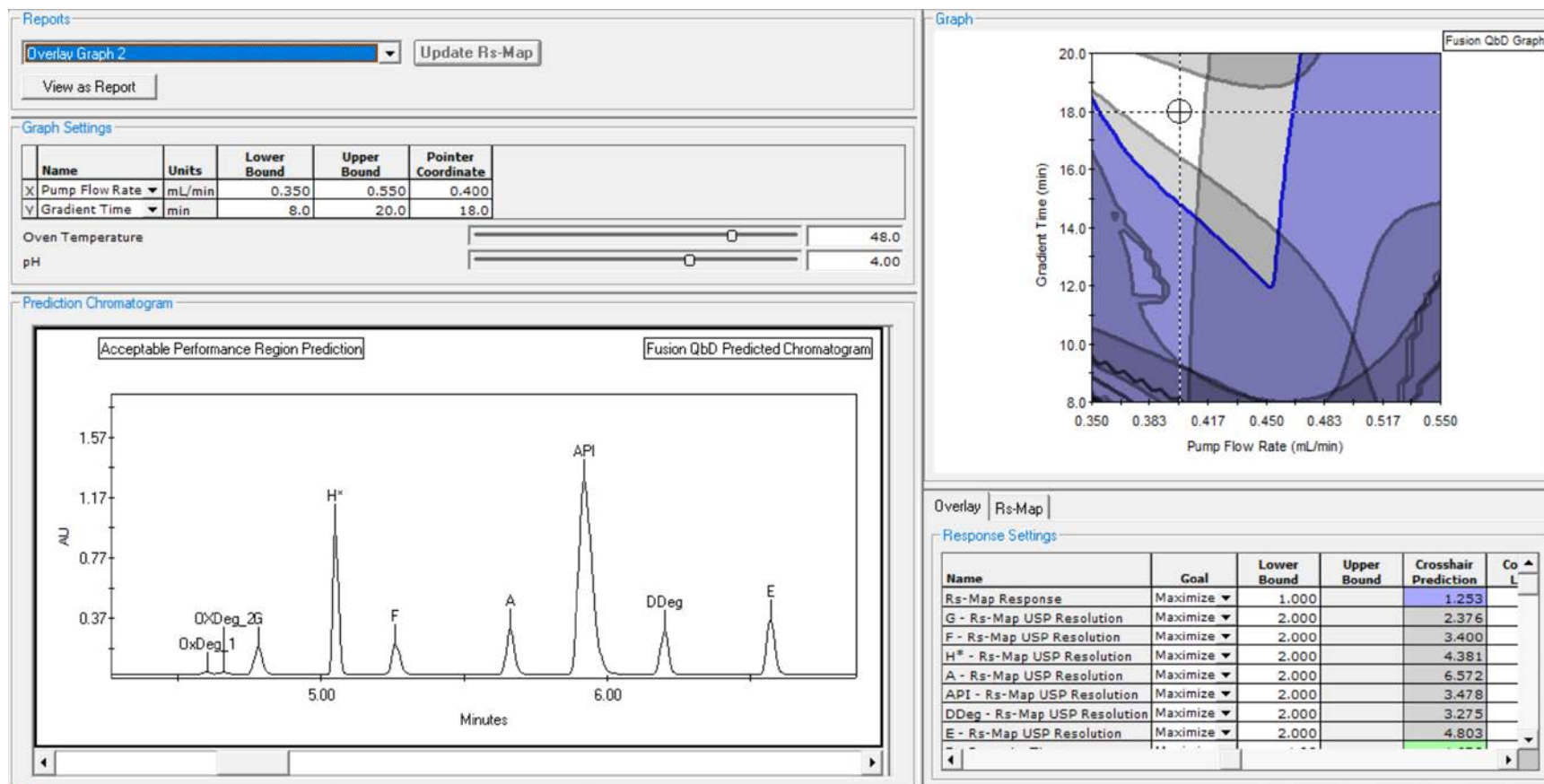
# New Degradants Identified – Forced Degradation Study (FDS)

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



# New Degradants Identified – Forced Degradation Study (FDS)

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





### 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.)

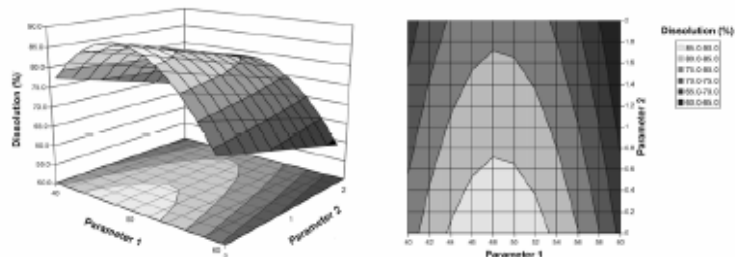


Figure 1a: Response surface plot of dissolution as a function of two parameters of a granulation operation. Dissolution above 80% is desired.

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

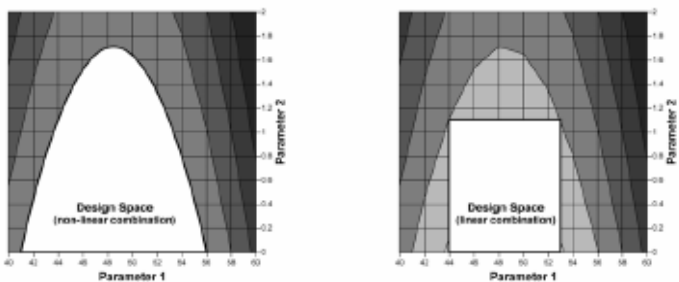


Figure 1c: Design space for granulation parameters, defined by a nonlinear combination of their ranges, that delivers satisfactory dissolution (i.e., >80%).

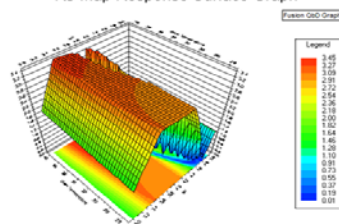
Figure 1d: Design space for granulation parameters, defined by a linear combination of their ranges, that delivers satisfactory dissolution (i.e., >80%).

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

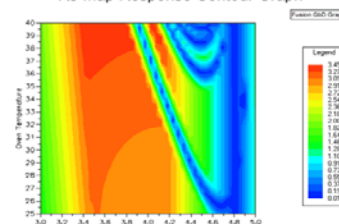


### Final Design Space + PARs

Rs-Map Response Surface Graph

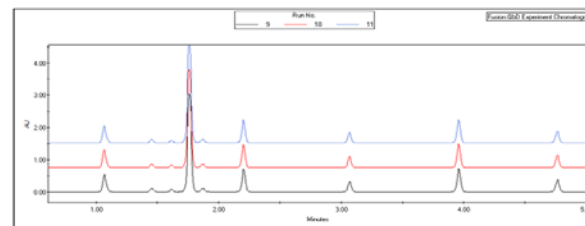
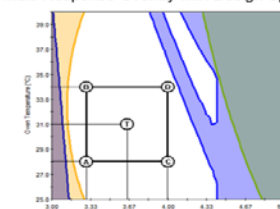


Rs-Map Response Contour Graph



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

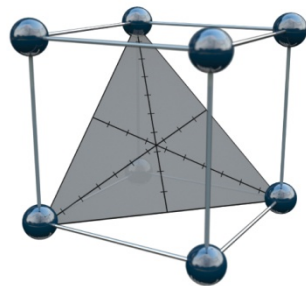
Multi-Response Overlay with Design Space





END

THANK YOU!



S-Matrix®

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