SHIMADZU
Excellence in Science

Solution for Method Development and Analytical Quality by Design

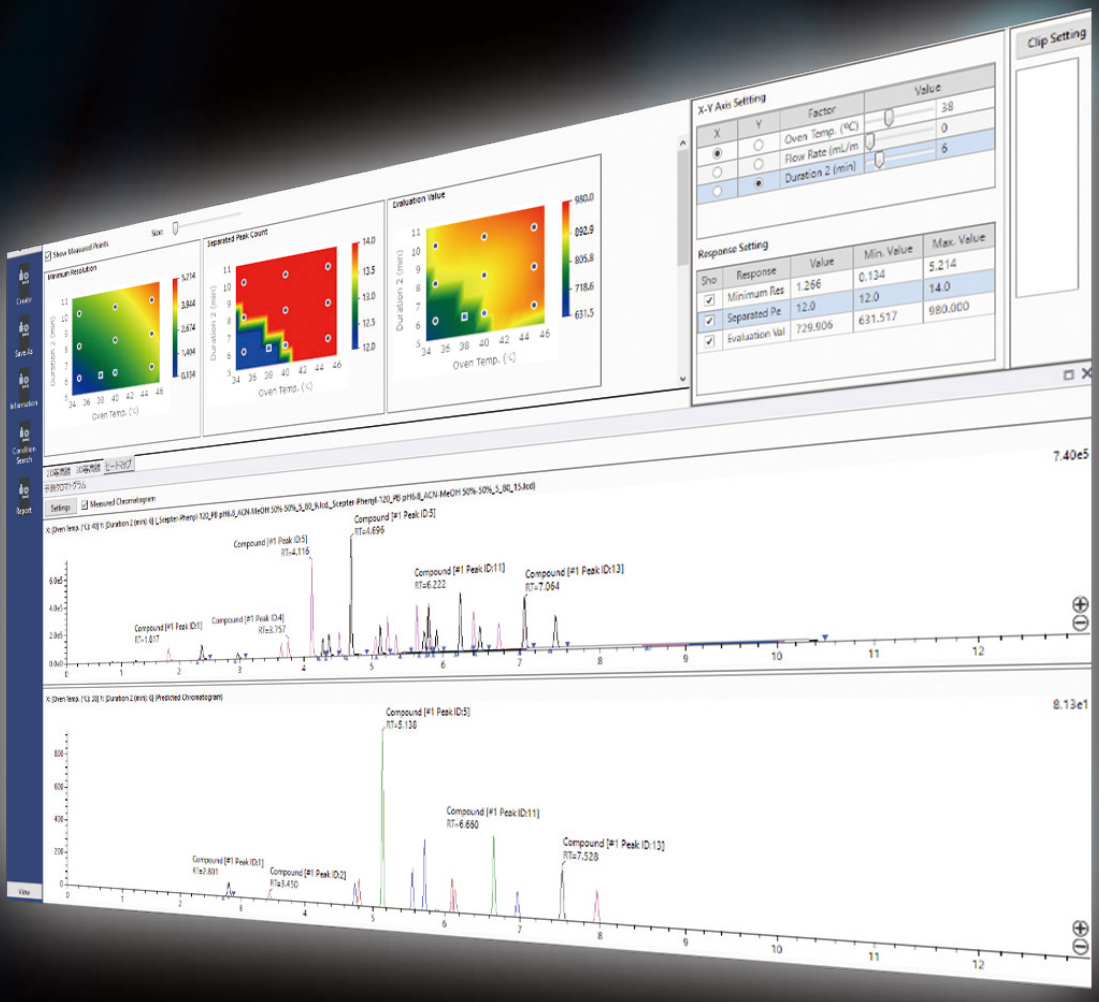
LabSolutions MD


Improve the Efficiency of Analytical Condition Screening with Experimental Design
Analytical condition settings can be efficiently screened in fewer attempts using an experimental process design to collect data.
Screening Phase
Optimization Phase
Use Design Space to Visualize the Robustness of Analysis Methods
The software can graph factor-response relationships and suggest the most robust analytical conditions. It even supports chromatogram simulation.
Validation Phase
Centrally Manage All Experiment Results in a Database
The software outputs a report that summarizes the experimental design, design space, chromatograms, and other relevant information. It also manages the information in a database to ensure data integrity.

AQbD

Develop Analysis Methods with Higher Reliability More Efficiently

LabSolutions MD uses an "Analytical Quality by Design" (AQbD) approach for achieving efficient method development by designing analysis methods based on science and risk. All workflow steps can be completed using LabSolutions MD, including analyzing samples using the experimental design, building a design space by using the analytical results, and evaluating robustness after deciding the optimal analytical conditions.



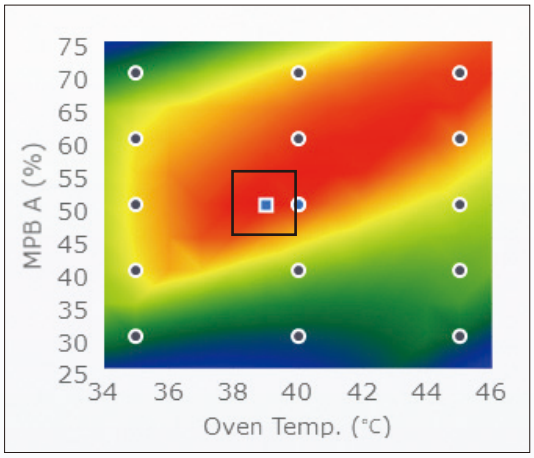
LabSolutions MD Features

for Each Phase of Analysis Method Development using the AQbD Approach

All steps involved in the screening, optimization, and validation phases of the analysis method development workflow can be completed using LabSolutions MD. These include analyzing samples using the experimental design, building a design space using the analytical results, and evaluating robustness after deciding the optimal analytical conditions.

			Response	
Column Nick Name	MPA pH	MPB A (%)	Evaluation Value	Minimum Resolution
Scepter-Phenyl-120	6.8	50	546.000	3.224
Scepter-C8-120	6.8	0	469.894	0.093
GIST-C18-AQ	2.7	0	465.124	1.075
GIST-C18-AQ	6.8	50	443.580	1.826
Scepter-C8-120	6.8	50	436.241	0.026
Scepter-Phenyl-120	2.7	50	419.659	1.743
Scepter-C18	2.7	0	419.338	1.518

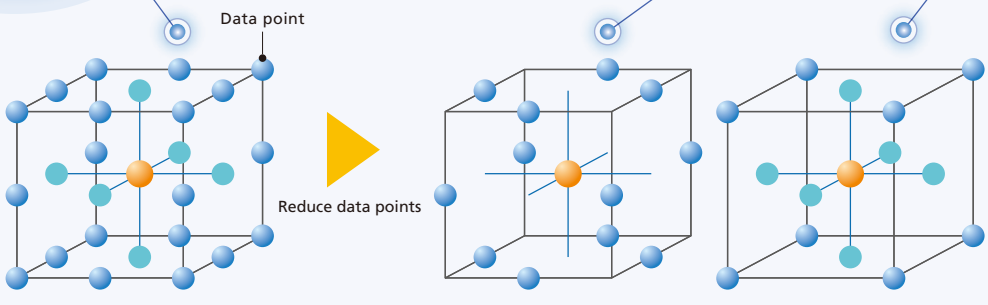
Quickly Screen for Optimal Analytical Conditions by Ranking Chromatograms ▶ p.9



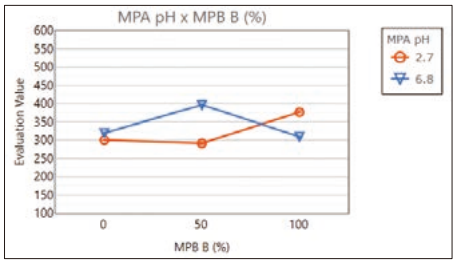
Visualize the Most Robust Analytical Conditions Using the Design Space ▶ p.11

ANALYTICAL INTELLIGENCE

Screening Phase



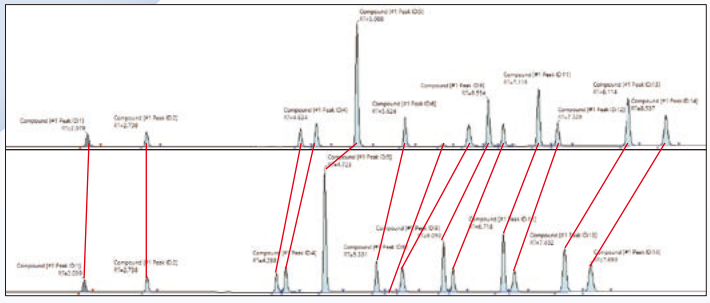
Reduce Data Points Using Experimental Design ▶ p.10



Verify Robustness by Variance Analysis ▶ p.13

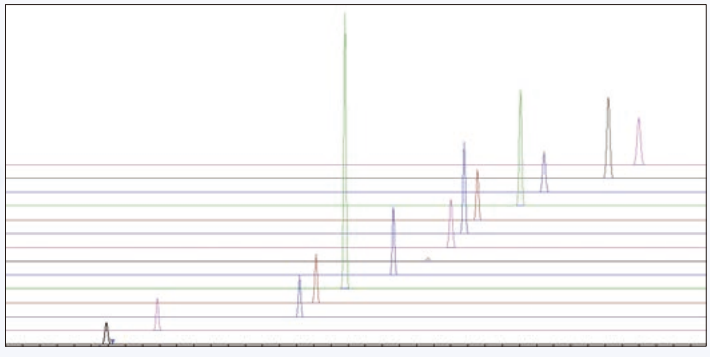
Validation Phase

Optimization Phase



Automatically Identify Compounds by Peak Tracking ▶ p.10

ANALYTICAL INTELLIGENCE



Chromatogram Simulation ▶ p.11

ANALYTICAL INTELLIGENCE

ANALYTICAL INTELLIGENCE

Automated support functions utilizing digital technology, such as M2M, IoT, and Artificial Intelligence (AI), that enable higher productivity and maximum reliability. Allows a system to monitor and diagnose itself, handle any issues during data acquisition without user input, and automatically behave as if it were operated by an expert. Supports the acquisition of high quality, reproducible data regardless of an operator's skill level for both routine and demanding applications.

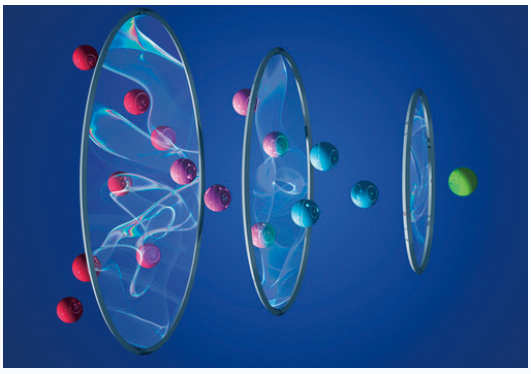
Workflow of AQbD Approach for Analysis Method Development

The International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use (ICH) advocates using the AQbD approach for analysis method development. For AQbD-based analysis method development, it is recommended that data gets acquired by efficient experiments, such as experiments planned by experimental design; that factors which affect analytical results are identified by statistical methods; and that the effective range of parameter settings is visualized as a design space. The science and risk-based approach ensures robust, low-risk analysis methods can be developed by comprehensively evaluating candidate analysis methods without relying on experience or intuition.

Screening Phase

Initial Screening of Analysis Methods

▶p.8

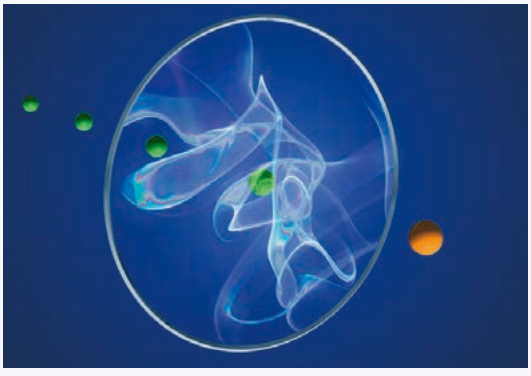


Initial screening is based on parameters that have a major effect on peak retention time and separation, such as the pH of aqueous mobile phases, the mixture ratio of organic mobile phases, and the type of column.

Optimization Phase

Optimization of Analysis Methods

▶p.10



With the analytical conditions determined by initial screening results as a starting point, optimal setting levels are verified for other parameters, such as pump gradient and column oven temperature conditions.

Validation Phase

Robustness Validation

▶p.12

Validation verifies that small variations in the optimized analytical condition settings affect measurement values only within an allowable range.

The following pages describe the various functions of LabSolutions MD software based on an example of using the workflow indicated on the left page to screen analytical conditions for simultaneous analysis of small-molecule drugs (12 types).

Analytical Conditions for Simultaneous Analysis

Analytes
(12 types of small-molecule drugs)

1: Probenecid
2: (S)-(+)-Naproxen
3: Acetylsalicylic acid
4: Diclofenac sodium
5: Papaverine hydrochloride
6: Dibucaine hydrochloride
7: Amitriptyline hydrochloride
8: Indometacin
9: Antipyrine
10: Lidocain
11: Quinidine
12: Metoclopramide

Mobile phase:
Pump A: A: 20 mmol/L (Sodium) phosphate buffer (pH 2.7)
B: 20 mmol/L (Sodium) phosphate buffer (pH 6.8)
→ 2 aqueous types

Pump B: A: Acetonitrile
B: Acetonitrile / Methanol = 1 : 1
C: Methanol
→ 3 organic types

Column: 1: Shim-pack Scepter C18-120 (100 mm × 3.0 mm I.D., 1.9 µm)
2: Shim-pack Scepter C8-120 (100 mm × 3.0 mm I.D., 1.9 µm)
3: Shim-pack Scepter C4-300 (100 mm × 3.0 mm I.D., 1.9 µm)
4: Shim-pack Scepter Phenyl-120 (100 mm × 3.0 mm I.D., 1.9 µm)
5: Shim-pack Scepter PFPP-120 (100 mm × 3.0 mm I.D., 1.9 µm)
6: Shim-pack GIST C18 AQ HQ (100 mm × 3.0 mm I.D., 2.0 µm)
→ 6 column types

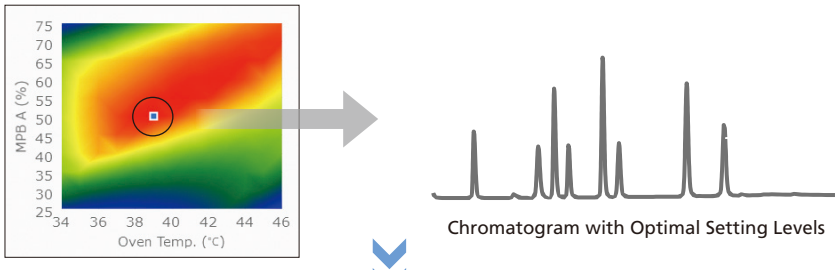
Analytical condition: Time program : B.Conc. 5%(0 min)→80%(8.01-11 min)→5%(11.01-15 min)
Flow rate : 0.7 mL/min
Inj.vol. : 1.0 µL
Temperature : 40 °C
Detection : Max plot 220- 400 nm (SPD-M40)
→ Basic conditions

STEP 1 Initial Screening of Analysis Methods
▶p.8

Using full factorial design, select the optimal combination of mobile phase (from 2 aqueous types and 3 organic types) and column (6 types).

STEP 2 Optimization of Analysis Methods
▶p.10

Create a design space in terms of three parameters: organic mobile phase mixture ratio, pump gradient conditions, and column oven temperature. Then specify analytical conditions by determining the optimal level of each.



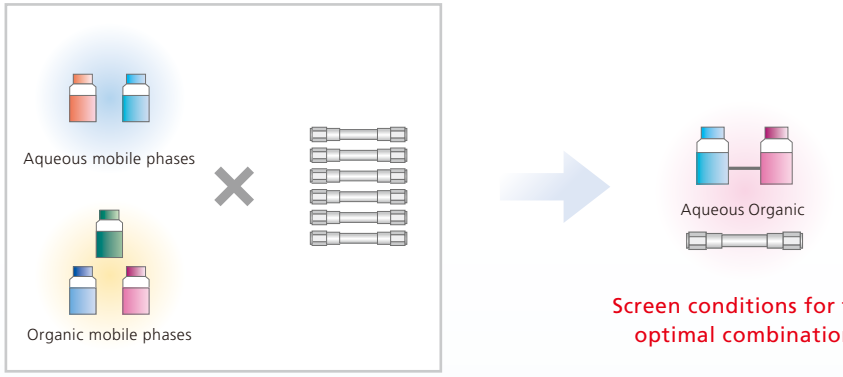
Chromatogram with Optimal Setting Levels

STEP 3 Robustness Validation
▶p.12

Using iterative experimental design, evaluate robustness with respect to variations in the organic mobile phase mixture ratio and column oven temperature levels.

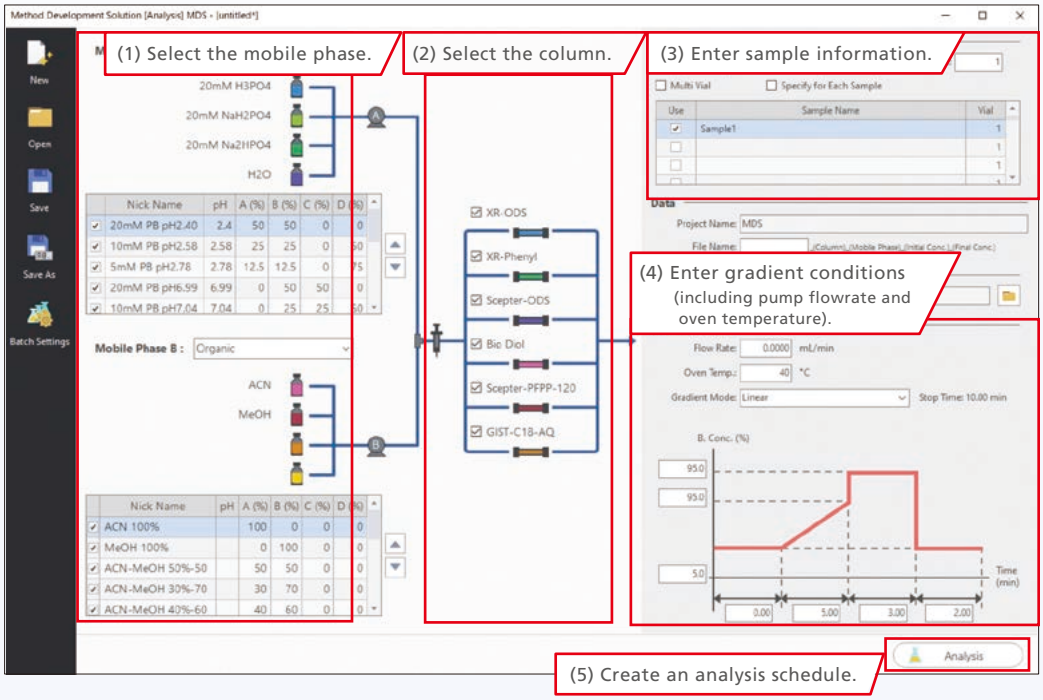
Initial Screening of Analysis Methods

Use the two types of aqueous mobile phases, three types of organic mobile phases, and six types of columns to acquire a total of 36 data points (full factorial design) for screening mobile phase and column conditions.



Easily Create an Analysis Schedule

The process of creating the vast numbers of method files and analysis schedules required for screening can be completed quickly by simply following steps (1) to (6) below. The mobile phase and column to be used can be selected with a single click and a comprehensive schedule reflecting column equilibration and blank analysis is generated automatically. That not only improves operational efficiency, but also can reduce human errors. The experimental design to be used can also be selected with a single click.



Experimental Design Selection Window

Quickly Find Optimal Analytical Conditions among Vast Amounts of Data

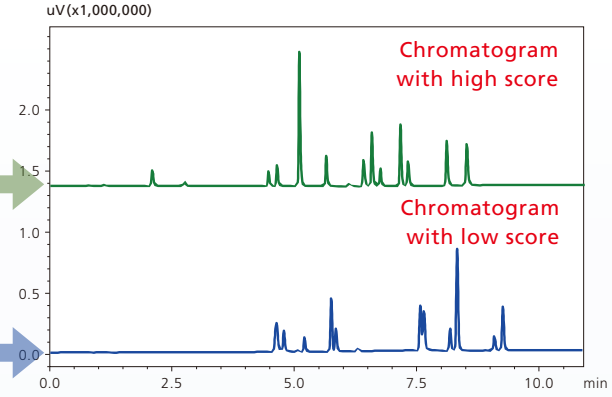
Because screening generates as many chromatograms as the number of conditions considered, they must be evaluated to determine which is optimal. If all the chromatograms had to be scrutinized by a human, it would be very tedious. LabSolutions MD can quickly and easily find optimal analytical conditions using equation (1) below to quantitatively evaluate the separation status resulting from each analytical condition.

$E = P \times (Rs_1 + Rs_2 + \ldots Rs_P)$ ... (Eq. 1)

The evaluation value (E) is calculated as the number of peaks detected (P) multiplied by the sum of the separation level (Rs) for all peaks.

			Response	
Column Nick Name	MPA pH	MPB A (%)	Evaluation Value	Minimum Resolution
Scepter-Phenyl-120	6.8	50	546.000	3.224
Scepter-C8-120	6.8	0	469.894	0.093
GIST-C18-AQ	2.7	0	465.124	1.075
GIST-C18-AQ	6.8	50	443.580	1.826
Scepter-C8-120	6.8	50	436.241	0.026
Scepter-Phenyl-120	2.7	50	419.659	1.743
Scepter-C18	2.7	0	419.338	1.518
Scepter-C18	6.8	50	396.000	4.326
Scepter-C4-300	2.7	0	394.239	0.402
Scepter-C18	6.8	100	384.553	2.046

Ranking Based on Each Analytical Condition and Evaluation Score Value



Comparison of Chromatograms with High and Low Evaluation Score Values

Identify Parameters with a Large Effect on Separation Using Variance Analysis

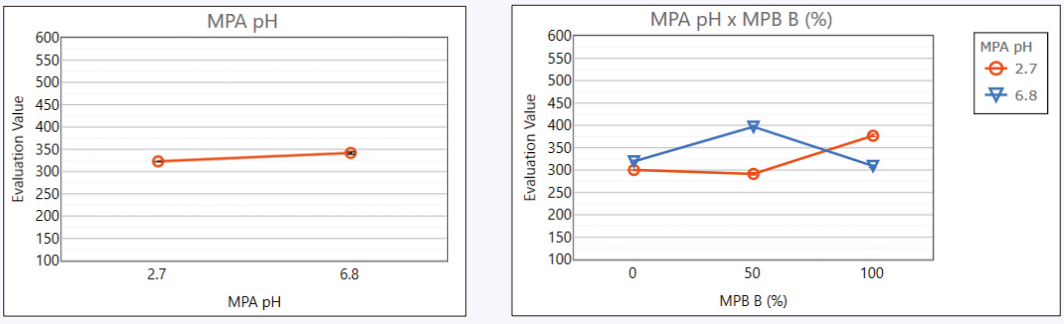
How much each parameter used for screening affects separation can be confirmed by variance analysis. Identifying the parameters with a large effect on separation can reduce the number of target parameters to be optimized during the analysis method optimization phase, which enables even more efficient experiments.

Because factors with a p-value of 0.05 or less can be assumed to have a variance value that is 95 % of the error variance, it is safe to assume that, if results differ for each setting level, then that factor has a large effect on separation. The analysis can also confirm the effect of interactions.

ANOVA Table

Display Plots	Effect	SSR	df	MS	F Value	p Value
☑	MPA pH x MPB B (%)	44817.9	2	22408.9	6.72	0.0141
☑	Column Nick Name	66312.0	5	13262.4	3.98	0.0302
☑	Column Nick Name x MPA pH	35853.2	5	7170.6	2.15	0.142
☑	Column Nick Name x MPB B (%)	50149.0	10	5014.9	1.50	0.265
☑	MPB B (%)	9123.7	2	4561.9	1.37	0.298
☑	MPA pH	3243.6	1	3243.6	0.973	0.347
	Error	33336.5	10	3333.7		
	Total	242835.8	35			

Using Variance Analysis to Determine How Much Each Parameter Affects Separation

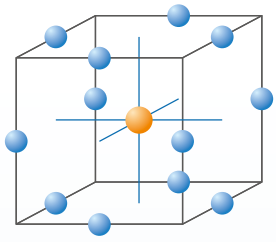


Factorial Response Graph

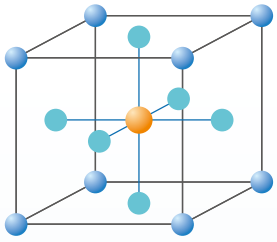
Reduce the Number of Data Points Using Experimental Design

Box-Behnken design and central composite design can shorten analysis times because they require fewer data points than full factorial design. For example, if determining the three optimal levels for the organic mobile phase mixture ratio, pump gradient conditions, and column oven temperature, full factorial design requires 27 data points (3 × 3 × 3) for optimization whereas Box-Behnken design requires 13 points and central composite design requires 15 points.

Box-Behnken Design



Central Composite Design

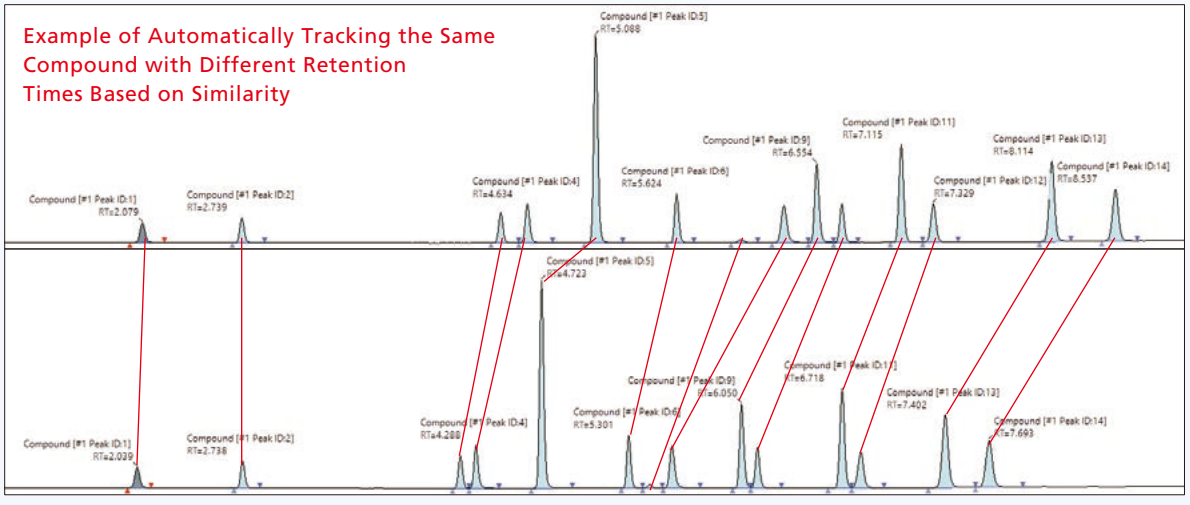


	Required Experiment Data Points
Full Factorial Design	27 points
Box-Behnken Design	13 points (52 % reduction of analysis time)
Central Composite Design	15 points (44 % reduction of analysis time)

Comparison of Data Points Required for Each Experimental Design Method (Given 3 Levels of 3 Parameters)

Automatic Identification of Target Compounds by Peak Tracking

ANALYTICAL INTELLIGENCE



Parameter	Torelance
Peak# ±	3
Area ±	10000
Height ±	10000
Area% ±	10
Height% ±	10
Similarity	0.9
Ret. Time ±	1

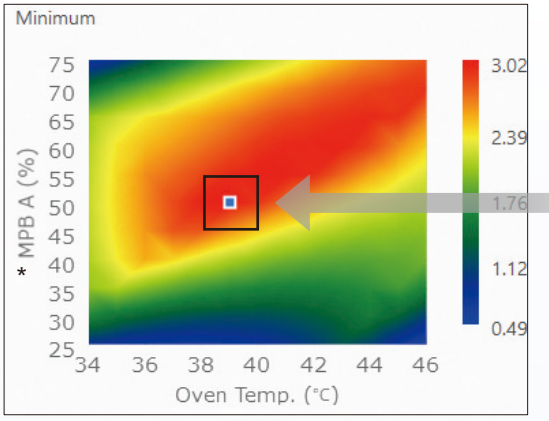
Similarity scores, area values, or other user-specified parameters can be used to automatically track variations in retention time for each compound in response to changes in analytical conditions. Even if analytical conditions change peak elution positions, peak tracking can quickly and automatically recognize peaks from all data and without requiring additional work for identification.

Parameters Usable for Automatic Peak Tracking

Identify the Most Robust Analytical Conditions by Graphing Factor-Response Relationships

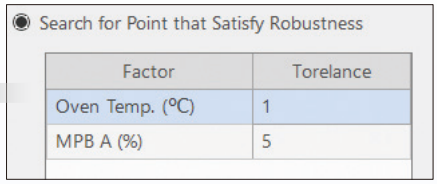
ANALYTICAL INTELLIGENCE

After the aqueous mobile phase pH level and column were selected by initial screening, analytical conditions were further optimized between five organic mobile phase mixture ratio settings (30, 40, 50, 60, or 70 %), three column oven temperature settings (35, 40, or 45 °C), and three gradient final-concentration settings (75, 80, or 85 %). The effect on separation by variations in parameter settings can be visually shown by graphing separation in terms of the organic mobile phase mixture ratio on the vertical axis and column oven temperature on the horizontal axis. That makes it easy to see at a glance that the most robust analytical conditions have an organic solvent mixture ratio of 50 %, a column oven temperature of 39 °C, and a gradient final concentration of 80 %. Consequently, robust analytical conditions can be specified without relying on experience or intuition. It is also possible to visually display variations in the distribution of minimum separation levels in response to arbitrary changes in the gradient final concentration as an additional parameter to the organic mobile phase mixture ratio on the vertical axis and column oven temperature on the horizontal axis.



Design Space for Minimum Separation Level
(given a gradient with 80 % final concentration)

* Mobile Phase B A: Mobile phase A connected to pump B (Refer to p. 7.)

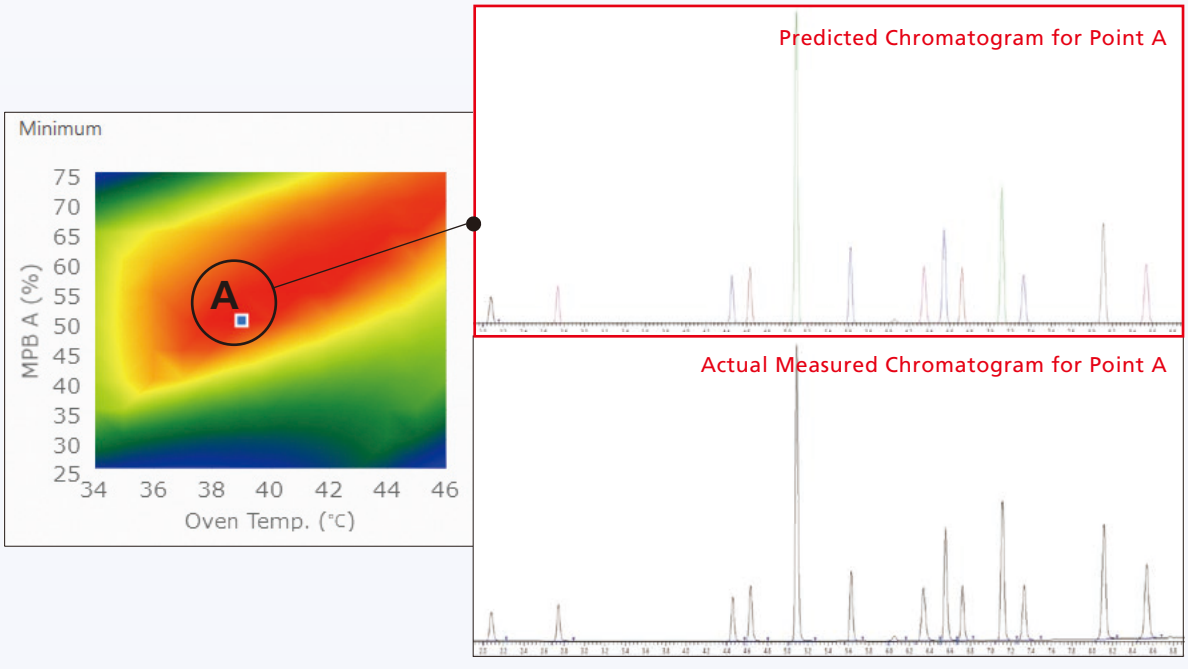


By entering the allowable fluctuation range in response to parameter (factor) changes, the software can also suggest robust analytical conditions that satisfy that allowable range (black box in figure to the left).

Chromatograms Resulting from Any Given Analytical Conditions Can Be Predicted

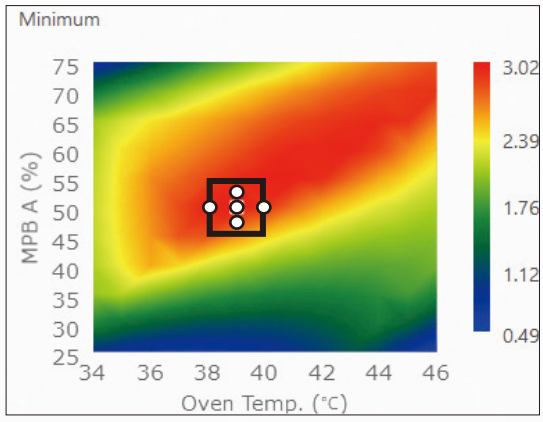
ANALYTICAL INTELLIGENCE

By clicking any analytical condition point (point A in the figure below), chromatogram changes in response to analytical condition changes can be visually predicted. That allows confirming how separation will behave in response to arbitrary changes to analytical conditions prior to starting an analysis.

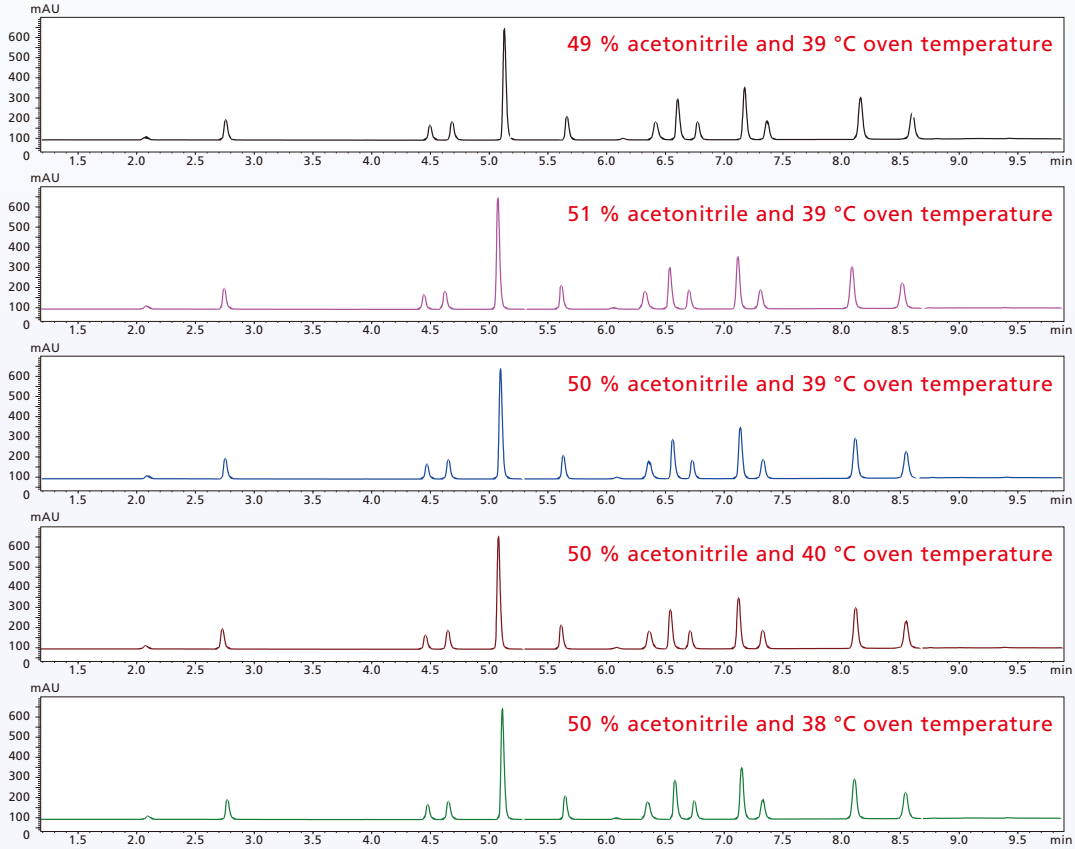


Robustness Validation by Iterative Experimental Design

Whenever analytical conditions are varied, it is important to validate the robustness of the conditions to better understand how those variations will affect measurement values and to ensure the reliability of the analysis method. Iterative experimental design automatically generates a schedule for analyzing a small range of variations in each parameter in the analytical conditions that were determined as optimal. That schedule can be used to evaluate how those variations affect separation. Specifically, it varies the organic mobile phase mixture ratio in 1 % increments (49, 50, and 51 %) and the column oven temperature in 1 °C increments (38, 39, and 40 °C) (area circled in white below) to verify the effect the analytical condition variations have on retention times and separation levels.



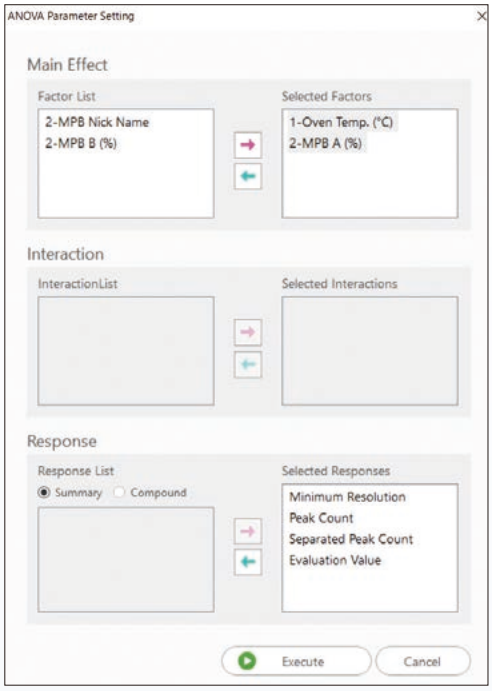
The resulting chromatograms obtained for robustness validation are shown below. That makes it easy to check all evaluation points with the chromatograms lined up next to each other. In this case, the setting variations result in very small separation level and retention time fluctuations and quickly show the robustness of the optimized analytical conditions.



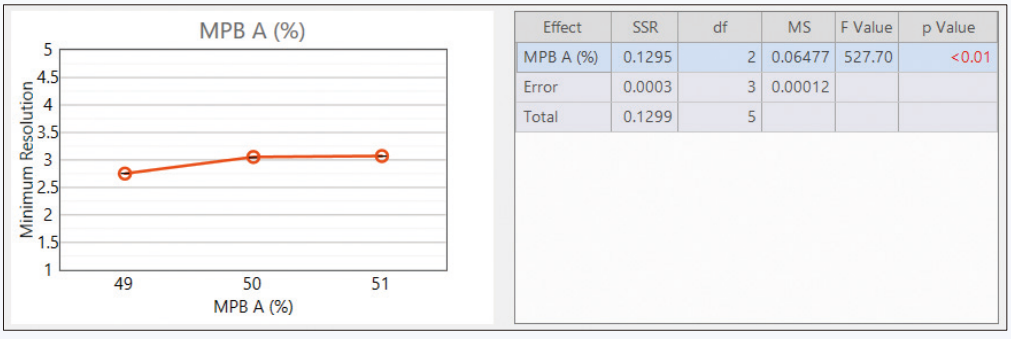
Chromatograms Obtained at Each Robustness Validation Data Point

Confirm Separation Effects Are Small Using Variance Analysis

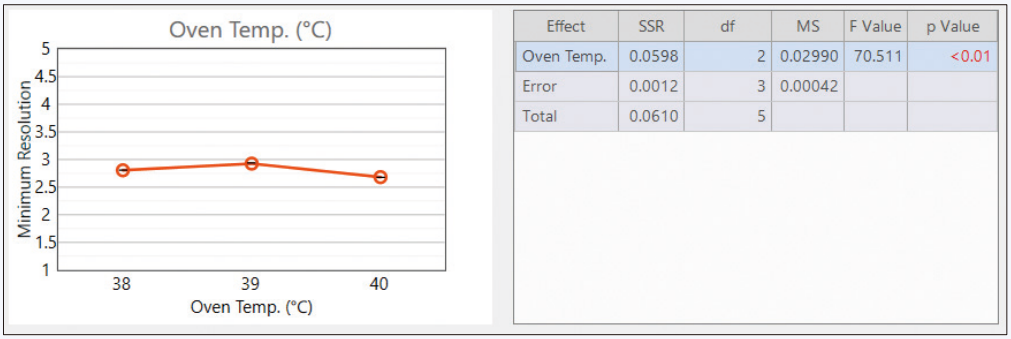
Robustness levels can be quantitatively calculated by performing a one-way analysis of variance with respect to robustness validation results. Specifically, the analysis can confirm that fluctuations in the organic mobile phase mixture ratio and column oven temperature will not significantly affect the separation level and it can quantitatively indicate the robustness level of the final analytical conditions. Any parameter or response can be selected for analysis of variance.



Parameter Settings for Analysis of Variance



Effect	SSR	df	MS	F Value	p Value
MPB A (%)	0.1295	2	0.06477	527.70	<0.01
Error	0.0003	3	0.00012		
Total	0.1299	5			



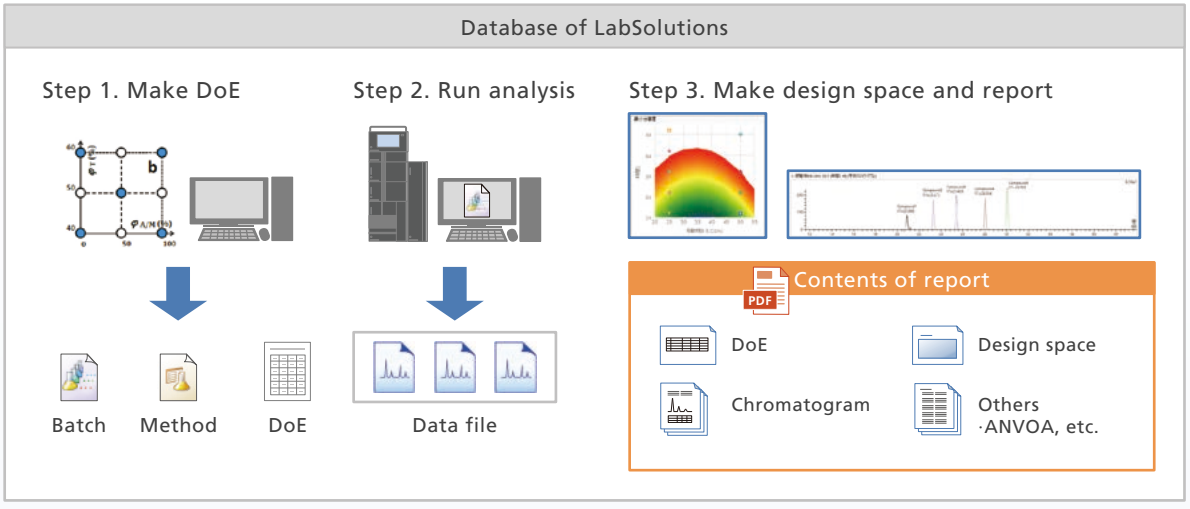
Effect	SSR	df	MS	F Value	p Value
Oven Temp.	0.0598	2	0.02990	70.511	<0.01
Error	0.0012	3	0.00042		
Total	0.0610	5			

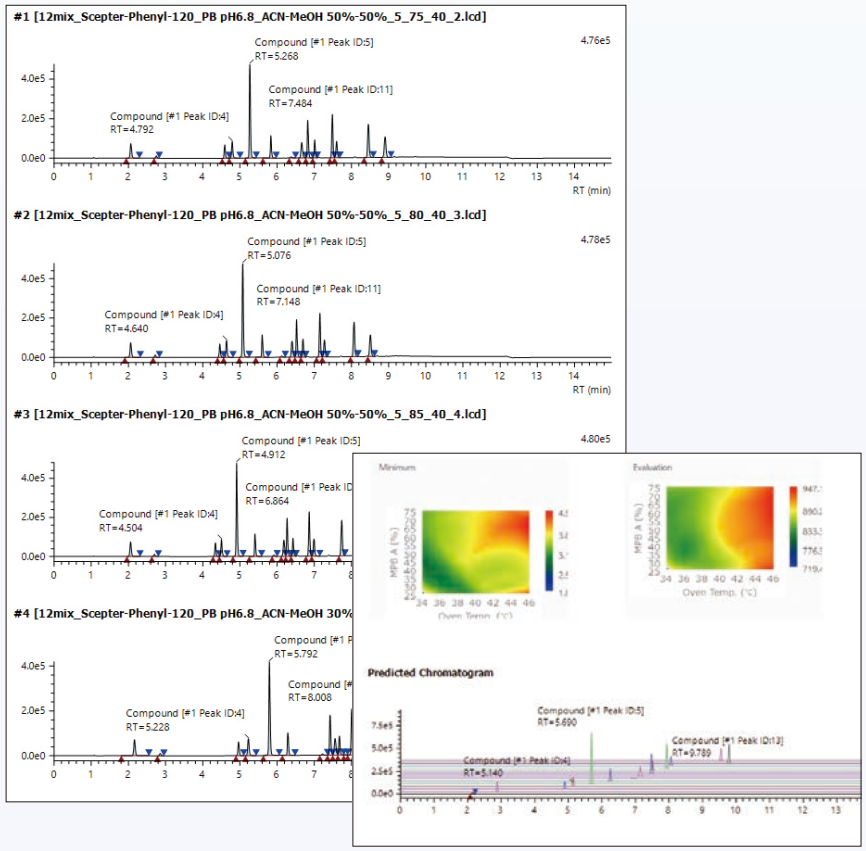
Using Variance Analysis to Determine How Much Each Parameter Affects Separation

Ensure Data Integrity by Database Management

It is easy to check analysis results using LabSolutions MD because all associated information, such as the experimental design, design space, and chromatograms, are output in a report. It also ensures data integrity by managing outputted reports together with the corresponding experimental design file, method file, batch file, and data file within a LabSolutions database.

Due to the seamless integration of all process steps, including creating an experimental design, acquiring data, and all method development steps in the design space, LabSolutions MD eliminates the need for any time-consuming file importing or exporting steps.





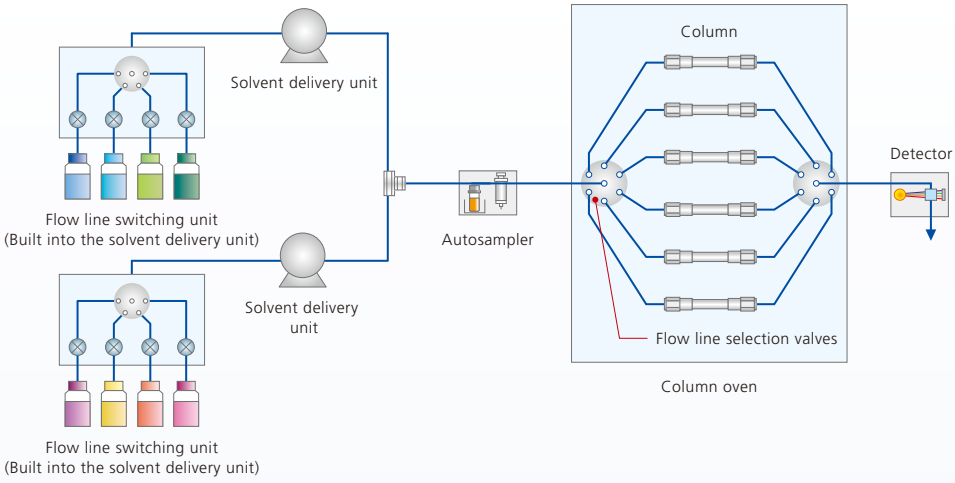
Example of Analysis Method Development Report

Save Labor by Switching between Mobile Phases and Columns Automatically

In addition to automatically switching between multiple mobile phases and multiple columns, mobile phase blending functionality can also save labor by automating mobile phase preparation. LabSolutions MD is compatible with Nexera series and i-Series systems.

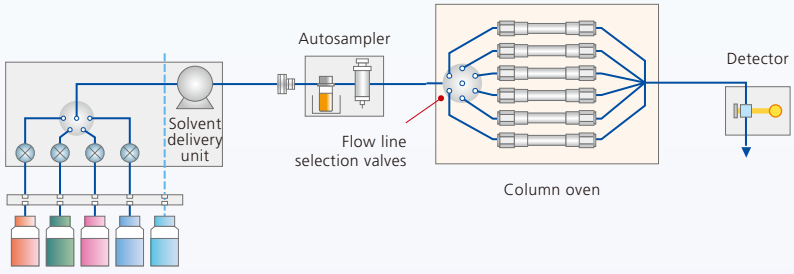
Nexera™ Series

These ultra high performance liquid chromatographs have a maximum pressure capacity of 130 MPa and support up to 192 combinations of 8 types of mobile phases and 12 types of columns (4 × 4 × 12).



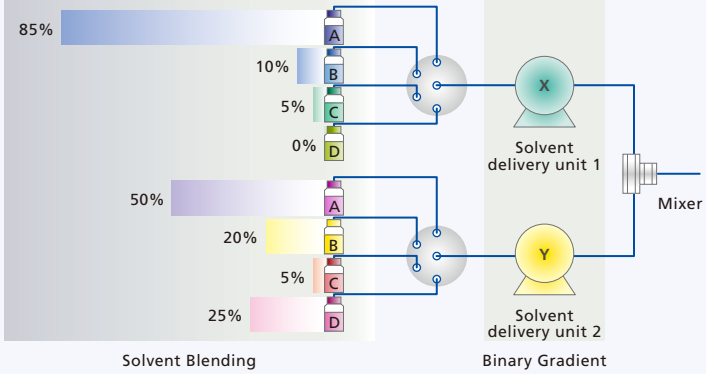
i-Series

These space-efficient and cost-efficient integrated LC systems support pressures up to 70 MPa.



Mobile Phase Blending Functionality Reduces Labor

The mobile phase blending functionality can dramatically reduce the amount of time previously required for mobile phase preparation by automatically preparing mobile phases with the user-specified pH level or the organic mobile phase mixture ratio using only a few types of mobile phases prepared in advance. i-Series systems can use a low-pressure gradient to blend up to two types of aqueous and two types of organic mobile phases.



Automatic Mobile Phase Preparation Using the Mobile Phase Blending Functionality (in Nexera systems configured with high-pressure gradient capability)

LabSolutions MD Package Contents

License of Method Development Solution
CD for installation (Instruction manual, Technical explanation)

LabSolutions, Analytical Intelligence logo and Nexera are trademarks of Shimadzu Corporation or its affiliated companies in Japan and/or other countries.

Shimadzu Corporation
www.shimadzu.com/an/

For Research Use Only. Not for use in diagnostic procedures.
This publication may contain references to products that are not available in your country. Please contact us to check the availability of these products in your country.
Company names, products/service names and logos used in this publication are trademarks and trade names of Shimadzu Corporation, its subsidiaries or its affiliates, whether or not they are used with trademark symbol "TM" or "®".
Third-party trademarks and trade names may be used in this publication to refer to either the entities or their products/services, whether or not they are used with trademark symbol "TM" or "®".
Shimadzu disclaims any proprietary interest in trademarks and trade names other than its own.

The contents of this publication are provided to you "as is" without warranty of any kind, and are subject to change without notice. Shimadzu does not assume any responsibility or liability for any damage, whether direct or indirect, relating to the use of this publication.

© Shimadzu Corporation, 2021 / First Edition: November 2021, 3655-10103-PDFIK, C190-E278