

Analytical, Preparative Supercritical Fluid Chromatograph and Supercritical Fluid Extraction System




# Nexera UC Nexera UC Prep



# Unified Chromatography...

## Just another chromatographic technique... or the only technique you'll need?

Conventional LC/MS and GC/MS face these challenges...

-  Unseparated peaks or low-intensity peaks
-  Labor-intensive sample preparation
-  Time-consuming post run processing



Nexera UC received a 2015 "R&D 100" award from the American technology information magazine, "R&D Magazine" and a 2015 Pittcon Editors' Gold Award.  
Nexera UC Prep received a 2019 Pittcon Editors' Gold Award.

SFE (Supercritical Fluid Extraction) : An extraction method using supercritical fluid. It is available as a pretreatment method for solid sample analyses.  
SFC (Supercritical Fluid Chromatography) : A chromatographic technique using supercritical fluids as mobile phases. With its unique properties, it enables high-speed, high-resolution analyses.

**Analysis**

▶ P.4

**Purification**

▶ P.10

**Unified Chromatography**

**Extraction**

▶ P.16

# Nexera™ UC and Nexera UC Prep

provide uncompromising solutions to these problems.

## **Unified speed of analysis, sensitivity, and resolution**

Supercritical CO<sub>2</sub> enables highly efficient sample extraction and high-resolution chromatographic analysis. The result; improved sensitivity and throughput for multi-analyte analyses.

## **High recovery rate of preparative purification with excellent operability**

Reliable small volume fractionation is ensured through Shimadzu's unique collection technology.

## **Fully automated on-line sample preparation, analysis and fractionation**

Target compounds are automatically extracted, analyzed, and fractionated.

Supercritical fluid is a fluid over its critical point. It has unique properties like liquid and gas. Low viscosity, high diffusion coefficient, liquid-like dissolving power. CO<sub>2</sub> is most popular for use.

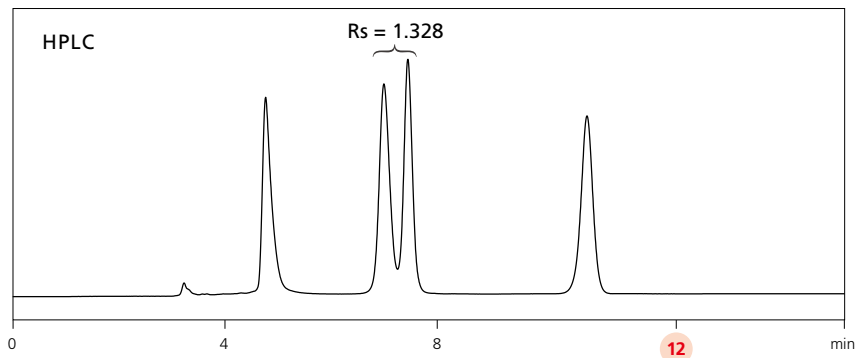
## Unified speed of analysis, sensitivity, and resolution

Supercritical fluid chromatography (SFC) is a separation method that uses supercritical CO<sub>2</sub> as the mobile phase. The benefits of supercritical CO<sub>2</sub> are the low polarity similar to n-hexane, the lower viscosity, and the high diffusion coefficient, which offers potential advanced separation methods that are different from conventional HPLC separation. Thanks to this feature, SFC enables unique analyses, such as high resolution chromatography, simultaneous analysis of compounds over a wide polarity range, and improvement of MS detection sensitivity.

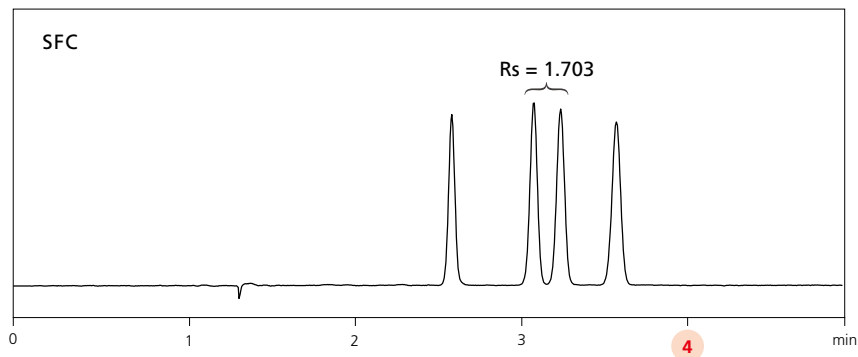
- Very fast separation speed due to the relatively low viscosity of supercritical CO<sub>2</sub>
- Improved peak capacity and chromatographic resolution
- Efficient separation of analogues and/or chiral compounds by high penetration mobile phase
- Different separation mode leads to high sensitivity

### Higher resolution

Improved separation and detection capabilities result from the low viscosity and high diffusion coefficient of supercritical CO<sub>2</sub>. As shown below, Nexera UC demonstrates high-separation selectivity for isomeric compounds that are difficult to separate by conventional HPLC.



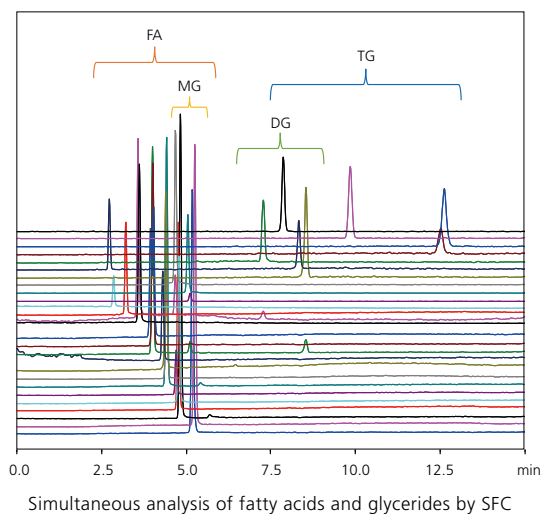
Approx. 1/3



Comparison of separation acquired by Conventional HPLC and SFC  
(sample:  $\alpha$ -tocopherol, column: Shim-pack™ UC-X Sil)

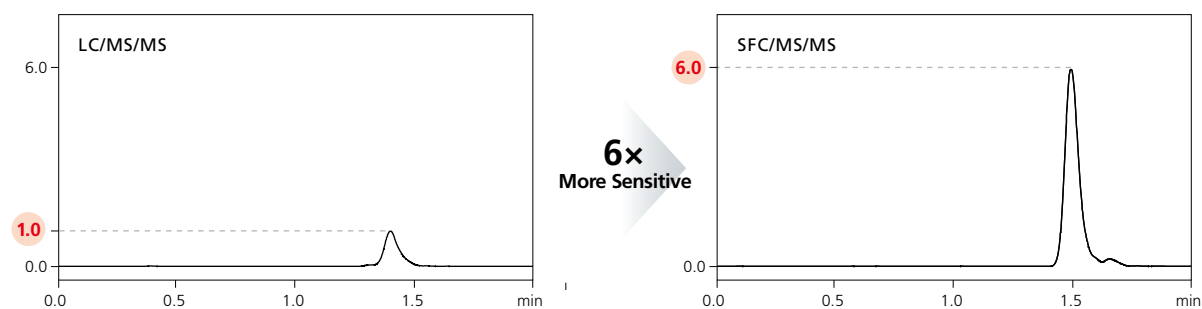
## Ideal for simultaneous analysis of compounds with a wide range of polarities

SFC can achieve a wide variety of separation patterns. This allows comprehensive analysis of compounds over a wide polarity range not possible with HPLC. Different separation methods are generally used for fatty acids, which are typically analyzed by GC, and glycerides, which are typically analyzed by HPLC. However, because supercritical CO<sub>2</sub> has properties similar to hexane, SFC is well-suited for analyzing compounds with different polarities, providing an ideal solution for the simultaneous analysis of fatty acids and glycerides.



## Sensitivity results from different separation modes in HPLC vs SFC

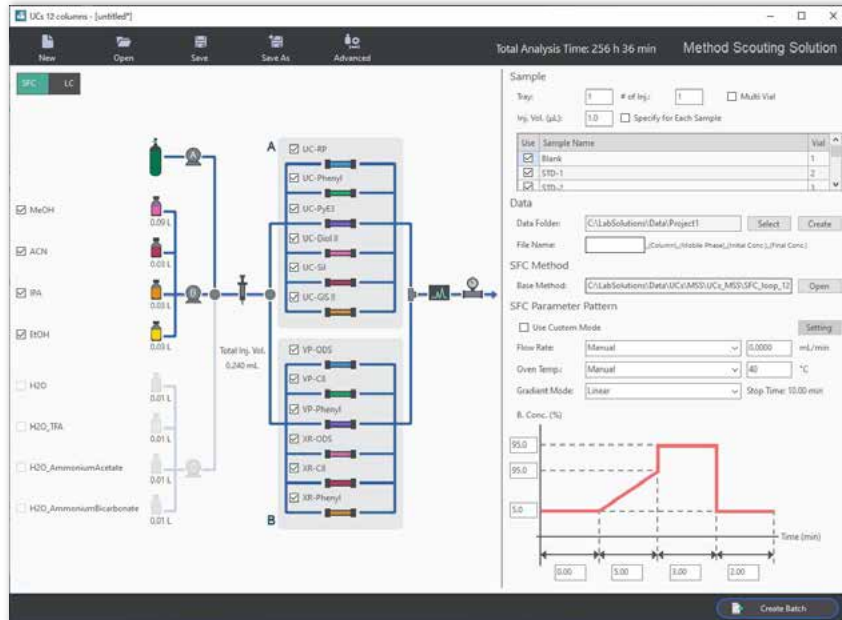
Supercritical CO<sub>2</sub> has unique properties that differ from liquid. Using SFC in front of a mass spectrometer offers greater sensitivity than achieved with LC/MS/MS.



## Ensures high-throughput method development and scaling up

### Automatically performs a variety of method scouting processes

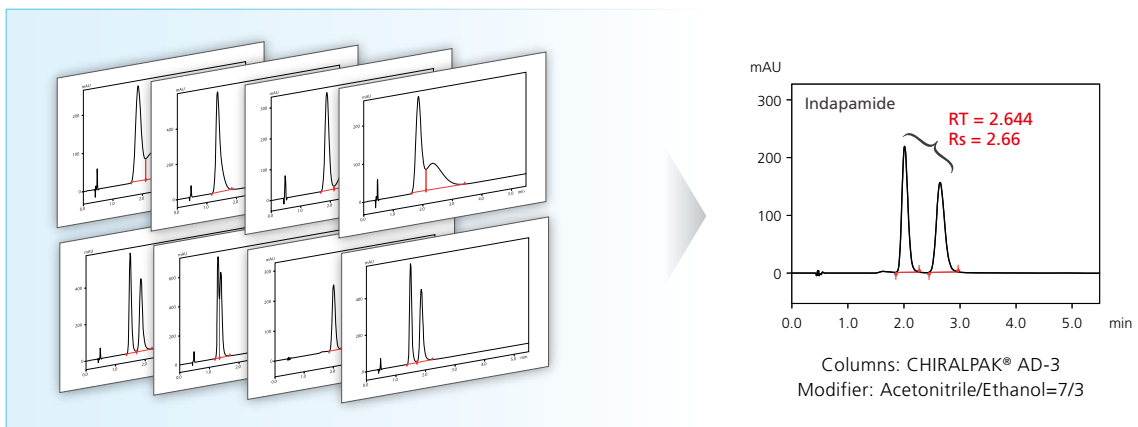
The high-speed performance of SFC can shorten the time required for method scouting. The system automatically generates a large number of methods by utilizing combinations of up to 12 columns, four modifiers, and different ratios of modifiers to mobile phase. With the UHPLC / SFC Switching system, screening by UHPLC and SFC can be carried out in the same batch. Moreover, Method Scouting Solution, optional software for method scouting, makes it easy to set up multiple conditions.



A screen shot of Method Scouting Solution for Nexera UC user interface.

### Chiral analysis with “Nexera UC Chiral Screening System”

CHIRALPAK® Series and CHIRALCEL® Series columns (Daicel Corporation) for chiral analysis are capable of resolving a wide variety of compounds by showing complementary separation targets. The combination of the Nexera UC Chiral Screening System and these columns simplifies method scouting for chiral analysis.



## Method scouting for optimization of separation conditions and scaling up to preparative size

For high-purity isolation, target peaks need to be adequately separated, for which the user must determine optimum column and separation parameter settings (method scouting). The Nexera UC chiral screening system and Method Scouting Solution can be used to screen columns more quickly and accurately (Step 1). Once the optimum column has been identified, smoothly scale up to preparative scale flow rates, preserving the peak separation while increasing the mass load (Step 2).

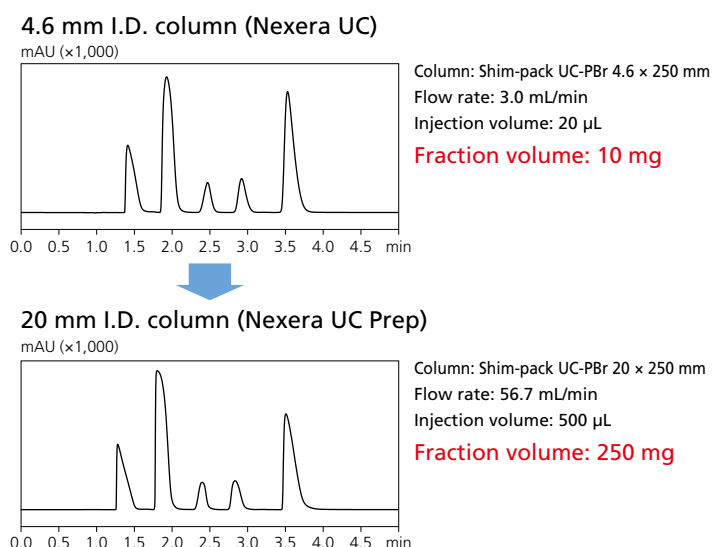
\*Preparative SFC: page 10 and following.

### Step 1 Method scouting at analytical scale

Method scouting is easy even for first-time users—simply execute the batch table generated automatically by Method Scouting Solution. The system can automatically switch between different settings to run the scouting process continuously day or night, even for multiple modifiers and columns. Various types of data can be displayed in the data browser and multi-data reports generated with resolution values for all data to assist in evaluating separation conditions.

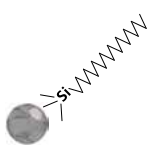

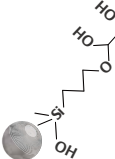
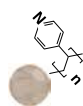
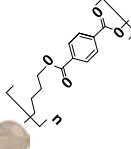
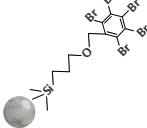
### Step 2 Scaling up

Using a Shim-pack UC series column enables you to increase mass load while maintaining separation performance. The optimum preparative column determined from Step 1 can be used to scale up the column size, flow rate and injection volume based on the desired fraction volumes.



## Selecting an SFC Column

The HPLC mobile phase for reverse phase analysis is very different from the one used for normal phase analysis. In contrast, SFC uses a mixture of supercritical CO<sub>2</sub> and a modifier (an organic solvent such as methanol) regardless of the stationary phase used. Therefore, the same mobile phase composition can be used for serial analysis through all columns. Column scouting is effective by using the following set of six columns, each providing a different separation selectivity.

		6 columns set					
		UC-ODS	UC-Sil II	UC-Diol II	UC-PolyVP	UC-PolyBT	UC-PBr
Chemistry							
Features		The separation mode is reverse phase. Retention is provided through hydrophobic interaction.	This is excellent for retention of basic compounds and recognition of their tertiary structures.	The separation mode is normal phase. This inhibits non-specific interactions.	A favorable peak shape is obtained even without acid-base additives.	This is excellent for resolving aromatic compounds through π-π interactions.	With ODS, separation of poorly retained compounds is improved.

## Add fractionation capabilities to the analytical SFC system

### Upgrade to an analytical fraction system

By adding a fraction collector to the analytical scale SFC, small-volume fractionation is possible. A complete workflow, from method development to fractionation, can be carried out seamlessly on one system.



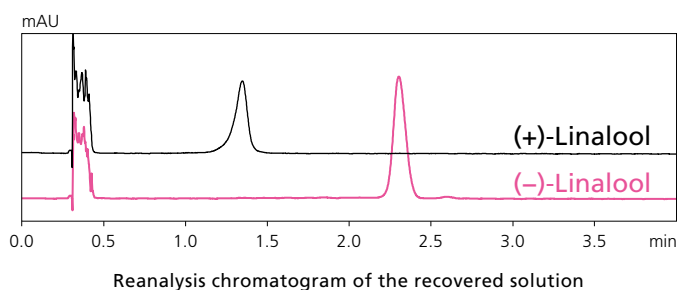
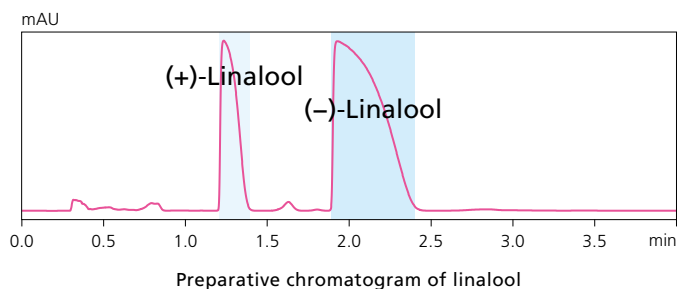
Nexera UC



Fraction collector  
FRC-40 SF

### Small-volume fractionation is possible

The patented gas-liquid separator, the LotusStream™ successfully reduces sample dispersion due to the volumetric expansion of CO<sub>2</sub>. It allows fractionation of small volumes, even into vessels such as 1.5 mL vials. The optical isomers of linalool were fractionated with high purity.





## System configuration examples

### SFC-UV / SFC-MS System

This is the minimum setup of Nexera UC and can replace both normal phase and reverse phase HPLCs. A wide range of analyte polarity can be covered by the combination of supercritical CO<sub>2</sub> and modifiers (for example, MeOH). Hazardous organic solvents such as hexane or chloroform are eliminated. In addition, the system reduces environmental impact by utilizing low-toxicity mobile phases and completing analyses in a shorter time. Both UV-Vis and MS detectors such as the triple quad LCMS-8050 can be successfully coupled with this SFC system.



### Chiral Screening System

This system is best for developing methods to separate chiral compounds. It automatically generates a large number of methods by utilizing combinations of up to 12 columns, four modifiers, and a different ratio of modifiers to mobile phase.



### Nexera UC/s UHPLC / SFC Switching System

This system can switch automatically between SFC analysis and UHPLC analysis and make measurements on a single sample in each separation mode. It enhances user-friendliness and operability by allowing the investigation of separation conditions and performing reverse-phase high-speed analysis in a single system. Shimadzu also provides a kit to upgrade from your current UHPLC system to the UHPLC/SFC switching system.

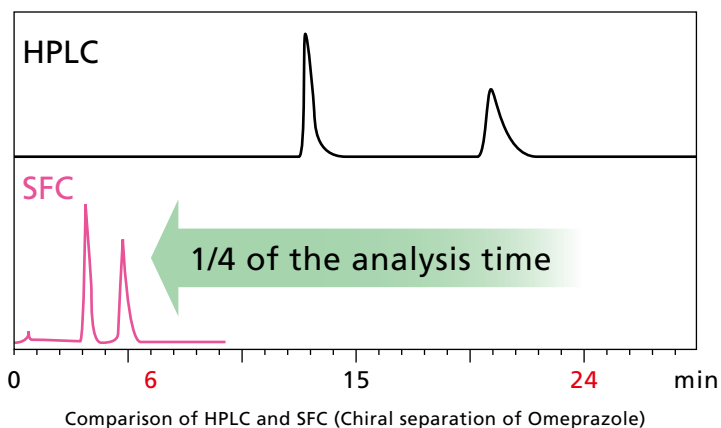


## High recovery rate of preparative purification with excellent operability

In purification using SFC, the target compounds are recovered in high concentrations in an organic solvent, which saves time not only during analysis, but also during post-run processing after preparative tasks are complete. The Nexera UC Prep maximizes fractionation output with its high recovery rates and ability to carry out continuous preparative work that further shortens the user's waiting time.

### SFC shortens analysis times

Due to the low viscosity and high diffusivity of supercritical CO<sub>2</sub>, column backpressure for SFC is low even at high flow rates which enables faster analysis without sacrificing column efficiency. This allows significantly shorter analysis times than HPLC.



### High recovery rates

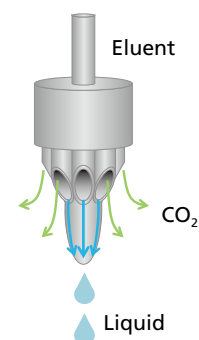
In preparative SFC, one factor that results in lower recovery rates is increased scattering of the eluent when the CO<sub>2</sub> returns from a supercritical to a gaseous state. The Nexera UC Prep's patented gas-liquid separator, the LotusStream separator, successfully reduces sample dispersion and carryover, while also achieving high recovery rates. These high recovery rates can be obtained regardless of flow rate or modifier concentration, even for volatile compounds such as the fragrance linalool.

Comparison for 1% linalool

Equipment	Recovery rate
Conventional separator	78.0%
LotusStream Separator	96.7%

#### LotusStream separator (patented technology)

Decreases flow rate without increasing the pipe diameter by splitting flow through multiple channels. The CO<sub>2</sub> is discharged externally while the liquid travels along the column and drips directly into the collection vessel without dispersing or scattering the eluate.



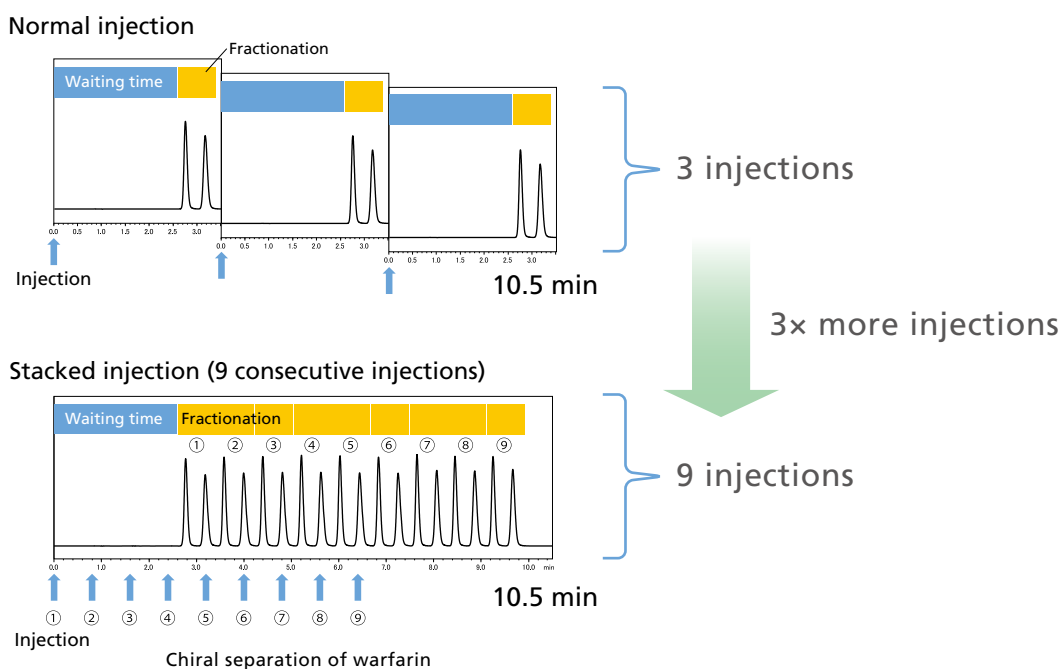
Watch videos >>





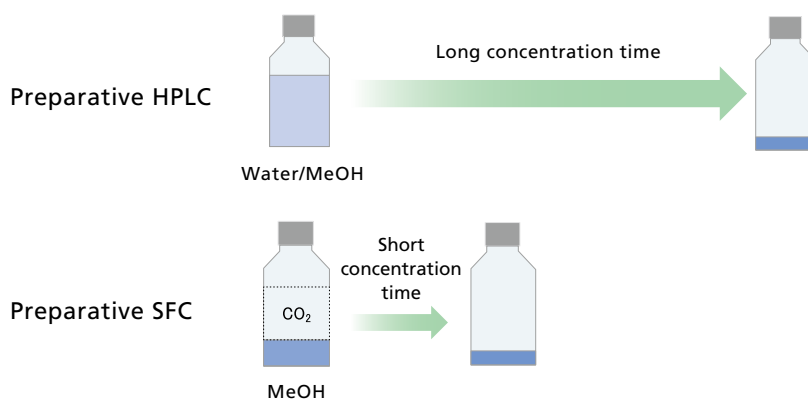
## Stacked injection function eliminates waiting time

Normal injection wastes time between peak elutions. Using the Nexera UC Prep's stacked injection function, samples can be injected continuously without any waiting time, enabling more samples to be processed. Settings for this function can be specified easily in the dedicated software Prep Solution.



## Simple post-run processing

Because most of the mobile phase is vaporized supercritical CO<sub>2</sub>, only the organic solvent (modifier) added to change the polarity of the mobile phase remains after preparative work. Since there is no water content in the recovery fraction, the concentration time is significantly shorter.



# Prep Solution enables a seamless preparative workflow

Prep Solution, ensures that it is simple to scale up from an analytical to a preparative workflow, and makes it easy to configure parameter settings. It greatly increases the efficiency of preparative workflows.

## Easy to understand even for first-time users

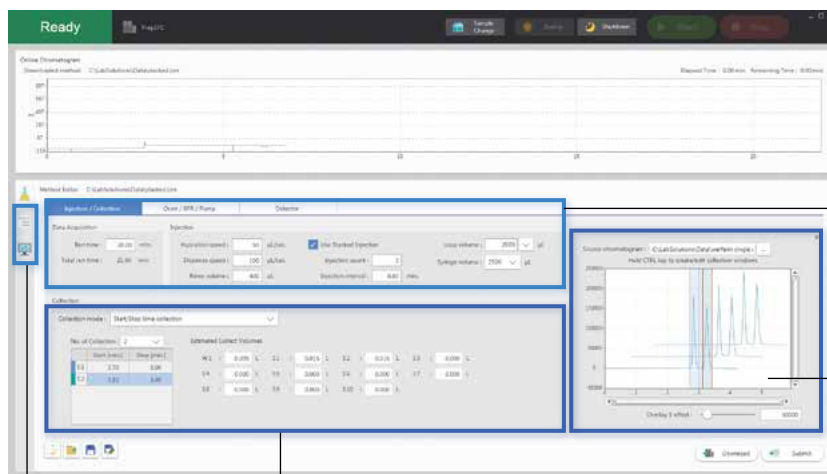
The parameter settings in Prep Solution are concise and intuitive, so that all users can operate the system with minimal training. This also avoids the risk of wasting samples due to human error.

### 1 Single analysis (peak check)

A trial analysis, or scouting run, prior to fractionation confirms component peak shapes and retention times. Analysis can be started by simply inputting basic parameters in the three onscreen tabs.

### 2 Simulation

The chromatogram obtained from the single analysis can be displayed in the simulation window, so that the collection start and stop times for each fraction can be selected with just a few mouse clicks. These settings can be applied to methods automatically.



Tabs to switch windows  
Toggle between windows with a single click

### Stacked Fraction System

The fractionation method can be selected from four options (manual fractionation, time fractionation, peak integration fractionation with/without time program) depending on the purpose of the analysis. Using the "peak integration mode", it is possible to assign individual slope and level values for fractionation start and end points, even for tailing peaks or other asymmetrical peaks.

Parameter settings  
Input parameters for injection, fractionation, etc.

Simulation window  
Simulations reflecting various parameter settings can be displayed, or peaks from the window can be selected to apply their parameter settings to a new analysis.



## 🕒 Easily isolate the target peaks

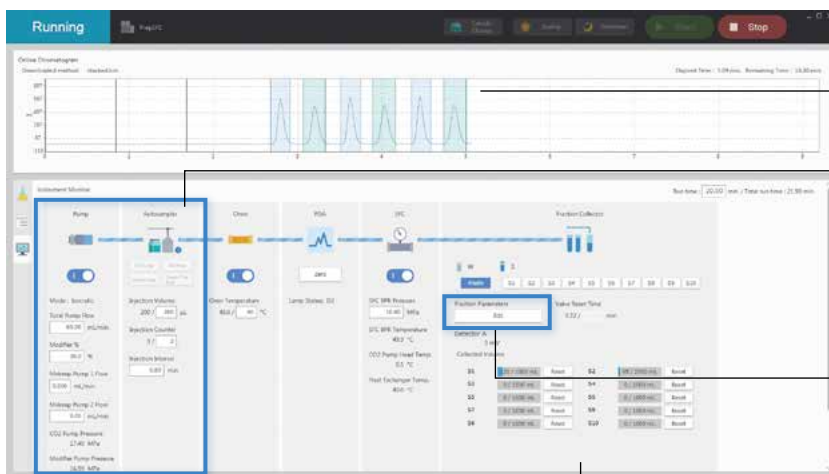
In case of peak shapes that are different from predictions or other unexpected situations, it is possible to change the preparative parameter settings while viewing the chromatogram. This eliminates the time and hassle involved in changing parameters and reanalyzing samples later on, as well as preventing the waste of valuable samples.

### 3 Fractionation

Samples are fractionated based on the user-selected parameters. The fraction range is displayed on the chromatogram, which can be checked in real time.

### 4 Adjust parameters during fractionation

Parameter settings for fractionation and injection can be adjusted during stacked injection (“on-the-fly” function).



The fractionation range is displayed on the chromatogram.

Parameters currently being applied to fractionation are displayed here. Settings for modifier concentration and stacked injection parameters (injection volume, number and interval) can be changed while viewing the chromatogram.

Fractionation time range and threshold values can be changed mid-analysis. In addition to a fractionation mode for target peaks, there is a mode for fractionation of “waste” intervals between peaks.

Stacked Fraction System

For the Multi-Fraction System, fraction collector racks are displayed here, with different display colors depending on the current status (fractionation complete / in progress / not started).

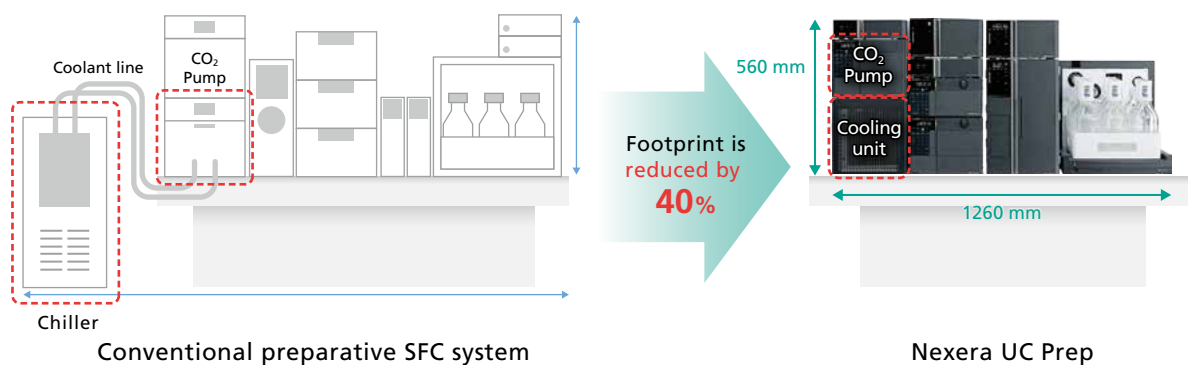


## Compact, benchtop design

This space-saving benchtop model includes a carbon dioxide pump that does not require an external chiller (cooling system for heat generated when pumping CO<sub>2</sub> at high flow rates). In addition, one unit can handle a wide range of flow rates, lowering installation costs.

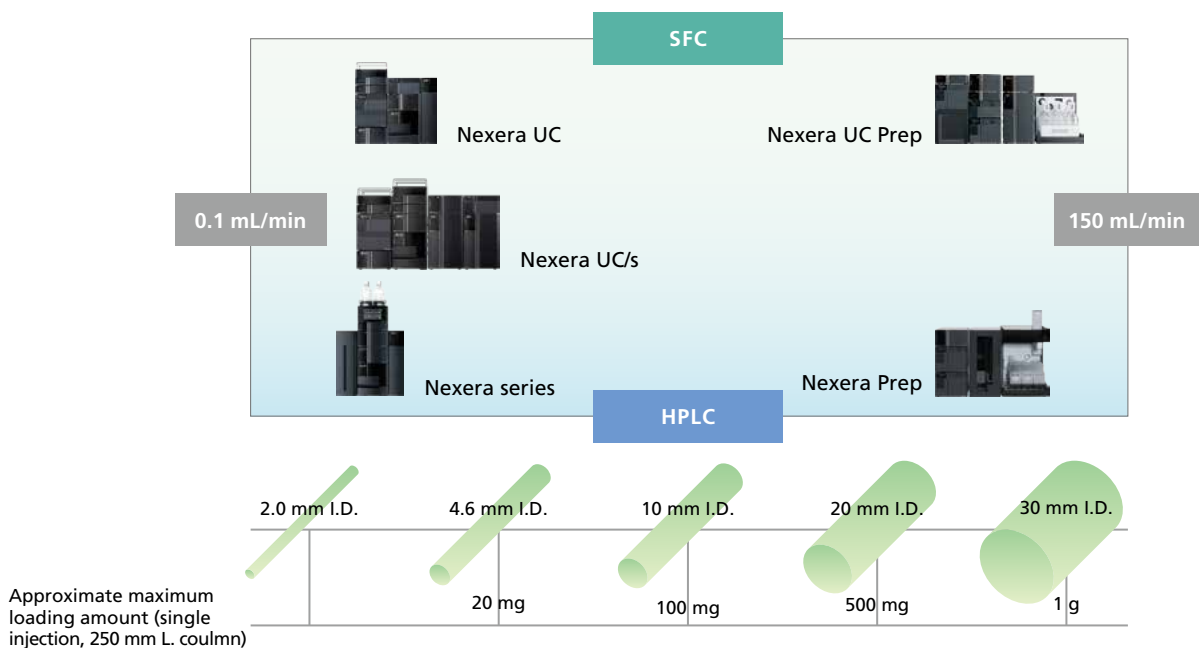
### ● Benchtop system that can be installed anywhere

Usually a chiller is required to cool the solvent delivery pump when pumping CO<sub>2</sub> at high flow rates. However, the Nexera UC Prep features a compressor-type cooling unit, reducing the size of the system and allowing it to be installed anywhere. Its footprint is equivalent to an analytical scale SFC system.



### ● Wide range of flow rates available

In a single system, the Nexera UC Prep can handle flow rates from 10 to 150 mL/min for fractionation of a range of sample sizes, from a few hundred mg up to a few grams. Shimadzu provides various preparative systems, such as Nexera UC/s, which can perform SFC analysis and UHPLC analysis using a single system. The system corresponding to the application can be selected.



## System configuration examples

### Stacked Fraction System: supports injection volumes up to 20 mL for large volume fractionation

This system is optimized for large volume fractionation involving repeated injection of samples which may contain several compounds. The FRS-40 unit includes both injector and fraction collector functionality, so that the same unit can be used for repeated sample injections and gram-level preparative work. It supports injection volumes up to 20 mL\* and collection of ten fractions. Compatible with flow rates from 10 to 150 mL/min and 10 to 30 mm I.D. columns.

\*optional



### Multi-Fraction System: for multiple peak fractionation of impurities and natural products

This system is suitable for preparative tasks involving samples with many peaks detected, such as impurities in pharmaceutical compounds. Volumes of up to 2 mL\* can be injected using an autosampler that holds up to 162 samples (using 1.5 mL vials). Three rack types can be selected for the FRC-40 SF fraction collector, which can recover up to 540 fractions (using 10 mL vials). With support for flow rates from 10 to 150 mL/min, columns with an internal diameter from 10 to 30 mm can be used.

\*optional



### Analytical Fraction System: analytical flow and fraction collection in one system

This system is intended for analytical scale fractionation only requiring fraction volumes of several mL to recover up to 20 mg of material (used to check synthesis for example). By connecting an FRC-40 SF fraction collector to the Nexera UC system, analytical columns from 2.1 to 4.6 mm I.D. can be run at flow rates up to 5 mL/min for small volume fraction collection.



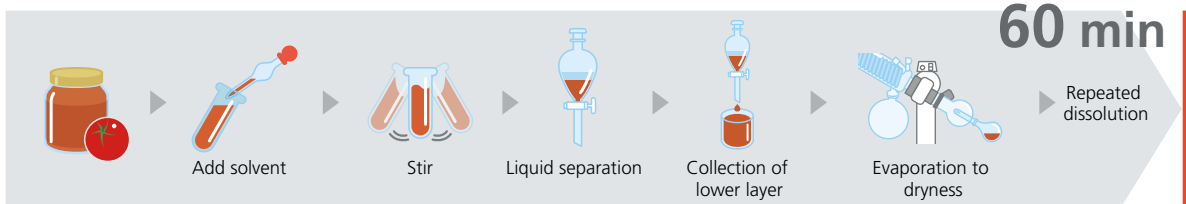
## Fully automated on-line extraction and purification

Nexera UC on-line SFE-SFC is a revolutionary system that combines on-line SFE, SFC and fraction collector in a single flow path. Target compounds are extracted from solid samples and then automatically transferred to SFC/MS so that no human intervention is required. The Nexera UC on-line SFE-SFC system reduces sample preparation time and acquires highly accurate data. Preparation can be carried out by connecting a fraction collector.

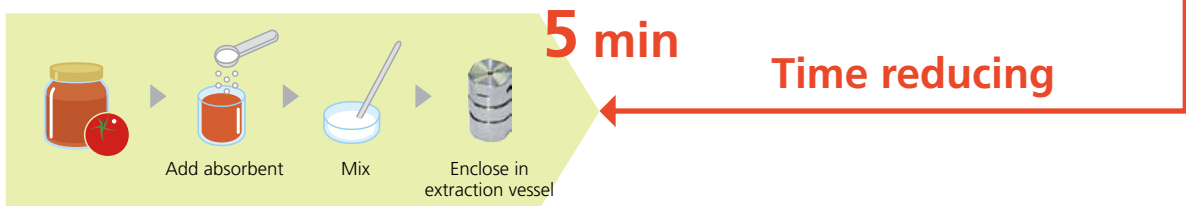
### On-line automation of the extraction and analysis of functional components in natural products

Target compounds are extracted from solid samples and then automatically transferred to SFC so that no human intervention is required. In the case of tomato paste, after preparing for analysis in as little as 5 minutes, extraction, separation, and detection of Lycopene are automatically carried out. In addition to a shorter sample preparation time, Nexera UC can automate the extraction, analysis, and fractionation process with the addition of a fraction collector.

**Typical sample preparation** ... Requires **60 minutes** of sample preparation

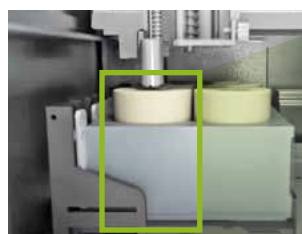


**Nexera UC** ... As little as **5 minutes** to be ready for analysis



### On-line SFE-SFC technology achieves a high recovery rate

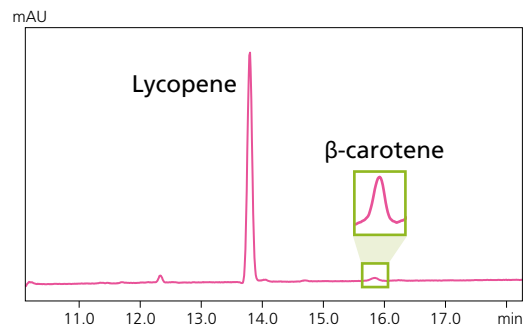
After placing the extraction vessel in position on the SFE unit and starting analysis in LabSolutions™ (analysis software), extraction and purification are performed seamlessly.



Inside of the extraction unit



Inside of the extraction vessel encapsulating the sample



Chromatogram of the tomato paste extract

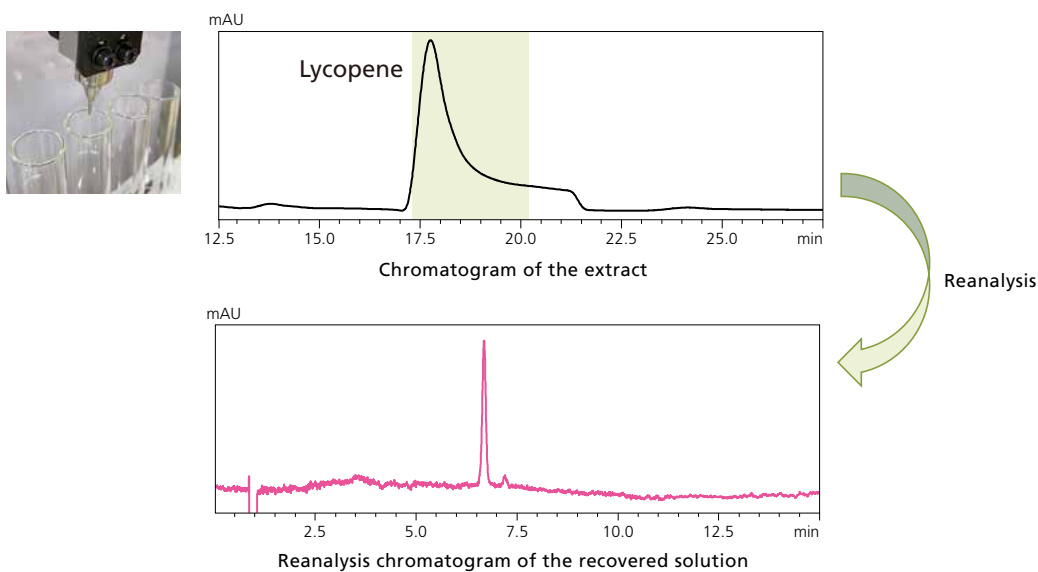
On-line SFE-SFC enables the functional component "lycopene" to be automatically extracted from tomato paste. The amount of extracted lycopene is equivalent to the value indicated on the product packaging.





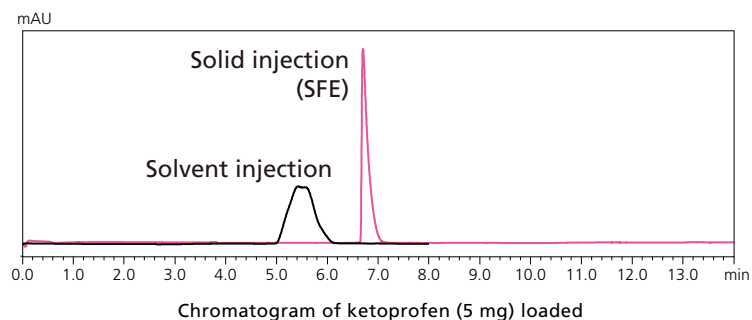
## ☉ Maximize fractionation output and labor-savings

On-line extraction, separation, and fractionation are possible by connecting a fraction collector at the end of the detector. Furthermore, scale-up from analytical to preparative scale increases the volume of fractions. The Nexera UC Prep SFE unit (maximum flow rate: 150 mL/min) with a preparative column enables high-volume extraction. On-line SFE-SFC can automatically run the extraction, purification and fractionation continuously day or night. The target compounds are recovered in high concentrations in an organic solvent, saving time during post-run processing after preparative tasks are complete.



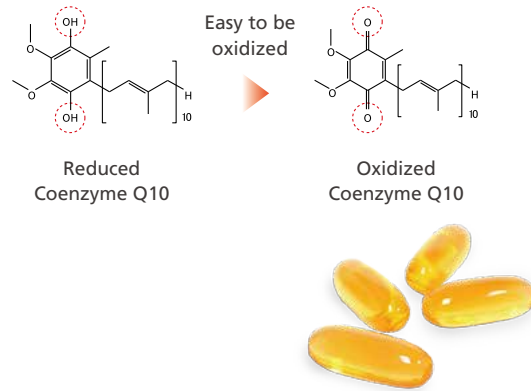
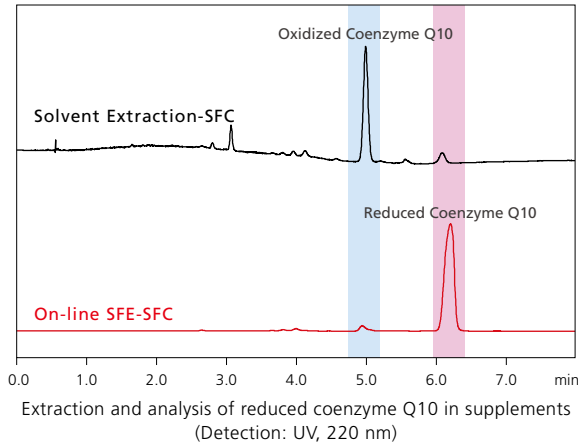
## ☉ Solid autosamplers for increasing loading capacity

More of the target compound can be fractionated at once by loading a large volume. However, this can sometimes lead to poor separation and broad peak shapes because of sample solvent effects. By performing fractionation with the on-line SFE-SFC, solid samples can be enclosed in an extraction vessel and extracted directly without dissolving in a solvent. A good peak shape prevents simultaneous elution of contaminants and improves the purity of the target component.



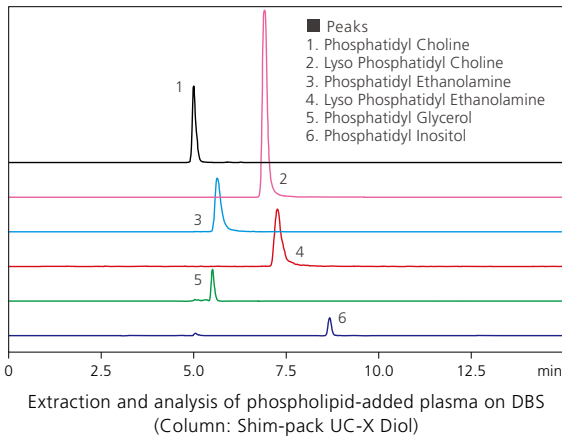
### Prevent degradation of labile compounds

With conventional solvent extraction, labile compounds may react with extraction solvents or could be oxidized and/or degraded.



### Analysis of biomarkers from dried blood spots (DBS)

The preparation of dried blood spots (DBS) from extraction to analysis requires simply enclosing a blood spot in an extraction vessel.

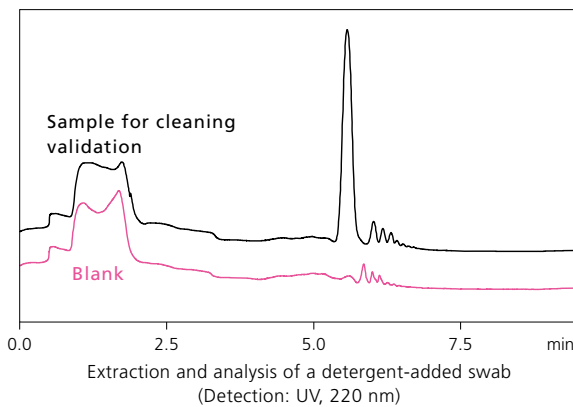


■ Detection

PC : 778.40>255.20 Negative  
LPC : 496.30>184.10 Positive  
PE : 716.40>255.20 Negative  
LPE : 454.30>313.20 Positive  
PG : 747.40>255.20 Negative  
PI : 835.50>255.20 Negative

### Only a few simple preparation steps for cleaning validation

Nexera UC can be applied to cleaning validation which is performed in the pharmaceutical industry to confirm that manufacturing equipment has been properly cleaned. Nexera UC automatically runs a series of steps from extraction to analysis by only putting the sample swab in the extraction vessel. In conventional cleaning validation, the sample swab needs to be extracted with water, and then the extraction is analyzed by TOC. However, when a target compound is hydrophobic, swab extraction is performed with ethanol and TOC is not applicable. Nexera UC can perform both types of cleaning validation.



An extraction vessel enclosing sample swab

The sample is provided by Daiichi Sankyo Co., Ltd.

## System configuration examples

### ☉ On-line SFE-SFC-MS System

In this system, solid samples are extracted by supercritical CO<sub>2</sub> and introduced to SFC on-line. The time for pretreatment of samples is drastically shortened. In addition, samples are extracted under light-shielding and anaerobic conditions to protect labile analytes from degradation. By adding a fraction collector to this system, small-volume fractionation of the extract is also available.



### ☉ SFE Pretreatment System

This system allows the pretreatment of samples using supercritical CO<sub>2</sub>. An extraction operation that changes the types of modifiers (up to four types) and concentrations to mix with supercritical CO<sub>2</sub> can be performed on each sample. The extracted material is collected in a test tube using a FRC-40 SF fraction collector. The LotusStream separator allows fractionation of small volumes, even into vessels such as 1.5 mL vials, without dispersion of the solvent. In addition to analysis by SFC, the system is ideal for measurements using other analytical systems such as GC-MS and NMR.



### ☉ On-line SFE-SFC Preparative System

In this system, solid samples are extracted by supercritical CO<sub>2</sub> and introduced to SFC on-line. The efficiency of fractionation can be improved, because this system can automatically run the extraction even at night.



# Extensive column lineup opens up analysis options

Shim-pack UC series columns are designed specifically for Nexera UC series SFC systems. When using supercritical fluids for analysis, retention behavior can vary significantly depending on the type of stationary phase. To optimize separation, a variety of columns should be used to determine which phase gives the best resolution in the shortest time. The extensive choice of column sizes available means that operations can be scaled up seamlessly from analytical SFC to preparative SFC.

	Functional group	Particle size	I.D. × L (mm)
Shim-pack UC-ODS	Octadecyl	3 μm	2.1×150, 3.0×50, 3.0×100, 3.0×150
		5 μm	4.6×250, 10×50, 10×250, 20×50, 20×250, 28×250
Shim-pack UC Sil II	-	3 μm	2.1×150, 3.0×50, 3.0×100, 3.0×150
		5 μm	4.6×250, 10×50, 10×250, 20×50, 20×250, 28×250
Shim-pack UC-Diol II	Diol	3 μm	2.1×150, 3.0×50, 3.0×100, 3.0×150
		5 μm	4.6×250, 10×50, 10×250, 20×50, 20×250, 28×250
Shim-pack UC-Py	Pyridinyl	3 μm	2.1×150, 3.0×50, 3.0×100, 3.0×150
		5 μm	4.6×250, 10×50, 10×250, 20×50, 20×250, 28×250
Shim-pack UC-PolyVP	Poly (4-vinylpyridine)	3 μm	2.1×50, 2.1×100, 2.1×150, 3.0×50, 3.0×100, 3.0×150, 4.6×50, 4.6×100, 4.6×150, 4.6×250
		5 μm	4.6×150, 4.6×250, 10×250, 20×250
Shim-pack UC-Triazole	Triazole	3 μm	2.1×150, 3.0×50, 3.0×100, 3.0×150
		5 μm	4.6×250, 10×50, 10×250, 20×50, 20×250, 28×250
Shim-pack UC-Choles	Cholesteryl	3 μm	2.1×150, 3.0×50, 3.0×100, 3.0×150
		5 μm	4.6×250, 10×50, 10×250, 20×50, 20×250, 28×250
Shim-pack UC-PolyBT	Polybutylene terephthalate (coated on silica gel)	3 μm	2.1×50, 2.1×100, 2.1×150, 3.0×50, 3.0×100, 3.0×150, 4.6×50, 4.6×100, 4.6×150, 4.6×250
		5 μm	4.6×150, 4.6×250, 10×250, 20×250
Shim-pack UC-NaE	Naphtylethyl	3 μm	2.1×150, 3.0×50, 3.0×100, 3.0×150
		5 μm	4.6×250, 10×50, 10×250, 20×50, 20×250, 28×250
Shim-pack UC-PyE	Pyrenylethyl	3 μm	2.1×150, 3.0×50, 3.0×100, 3.0×150
		5 μm	4.6×250, 10×50, 10×250, 20×50, 20×250, 28×250
Shim-pack UC-HyP	3-Hydroxyphenyl	3 μm	2.1×150, 3.0×50, 3.0×100, 3.0×150
		5 μm	4.6×250, 10×50, 10×250, 20×50, 20×250, 28×250
Shim-pack UC-PBr	Pentabromobenzyl	3 μm	2.1×150, 3.0×50, 3.0×100, 3.0×150
		5 μm	4.6×250, 10×50, 10×250, 20×50, 20×250, 28×250

Other Columns and Details  
Shim-pack UC series brochure >>>  
(C190-E251)



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