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Technical Report

Separation Characteristics of the Shim-pack Reversed Phase Column Series

Reversed Phase Columns C4/C8/C18 CoreFocus

Tomoaki Shimpo[1]

Abstract:

Reversed phase mode is the most frequently used method of separation in high-performance liquid chromatography. This method of separation uses hydrophobic interactions and can accommodate the separation of various compounds. A number of columns for reversed phase mode are commercially available from multiple manufacturers. It is crucial that basic separation characteristics of each column is assessed to determine which column is the most suitable for analysis. Using the Shim-pack reversed phase column series as examples, this article describes the basic separation characteristics of each column and functional group, and introduces the differences in usage of the columns within the Shim-pack series.

Keywords: Shim-pack™ series, Shim-pack Arata™ series, Shim-pack Scepter™ series, Shim-pack Velox™ series, reversed phase chromatography, C18, ODS, C8, C4, Tanaka Test method

1. Introduction

1-1. Reversed Phase Mode

In high-performance liquid chromatography (hereinafter HPLC), when the multiple compounds injected into the column pass through, the components are separated based on differences in the transit speed for each compound. The difference in transit speed is produced by differences in the strength of the interactions between the compound and the stationary, and the compound and mobile phases. In this case, compounds refer to the compounds that are dissolved in the solution being analyzed, mobile phase refers to the liquid circulated by a pump, and stationary phase refers to the chemically modified functional groups on the spherical particles with which the column is packed. Accordingly, the separation type in HPLC analysis is determined by the power relationships between three elements: the compounds, the mobile phase, and the stationary phase. Table 1 shows typical separation types and their interactions.

Table 1 General Separation Types and Interactions

Main Separation Types	Main Interactions	Typical Analytes
RP (RP: Reversed phase chromatography)	Hydrophobic interaction	Low molecular weight pharmaceuticals, pesticides, and vitamins
NP (NP: normal phase chromatography) HILIC (HILIC: Hydrophilic interaction liquid chromatography)	Hydrophilicity	Sugars, nucleic acids
IEX (IEX: Ion exchange chromatography)	Electrostatic properties	Inorganic ions, amino acids, proteins
SEC (SEC: Size exclusion chromatography)	Molecular size	Synthetic polymers, biopolymers, polysaccharides

Reversed phase chromatography (reversed phase mode) is the most frequently used HPLC separation type (Table 1). In this separation, the polarity of the stationary phase is low and the polarity of the mobile phase is high. Hydrophobic interactions predominate when compounds are retained in the column. The next section describes the generally used column base packing materials.

1-2. Column Base Packing Materials

Table 2 shows the generally used column base packing materials currently in use in HPLC and their features.

Table 2 Features of Generally Used Column Base Packing Materials

Item/Base Material Name	Silica	Polymer	Organosilica
Mechanical Strength Impact on Pressure Resistance and Separation	High	Low	High
Number of theoretical plates Impact on Separation	High	Low	High
Silanol activity End capping has an impact	Diverse	None	Diverse
Generally Used pH Range Impact on the chemical durability of the column	About 2-8	0-14	About 1-12
Resistance to organic solvents	High	Diverse	High

The three main base materials used in commercially available columns are silica, polymers, and organosilica. Of these, silica and organosilica are often used as the base packing material for columns in reversed phase mode. Organosilica is relatively new in comparison to silica and polymers, which are more established. This column base material is designed to make up for the low chemical durability of silica and the low mechanical strength of polymers. Recently, there has been a focus on techniques for end capping and functional group binding, leading to the appearance of silica columns that maintain high mechanical strength while extending the pH range from 1 to 12. These column base materials combine chemical durability and high mechanical strength to be used for a wide range of analysis.
Note that porous silica (hereinafter core shell silica) could be called a derivative column base material, as it makes use of these silica and organosilica base materials. A feature of this base material is that it is designed so compounds only pass through the surface layer of the packing material particles, giving it a higher number of theoretical plates per unit of pressure compared to fully porous columns with the same sized particles. (See Fig. 1)

[1] Analytical & Measuring Instruments Division

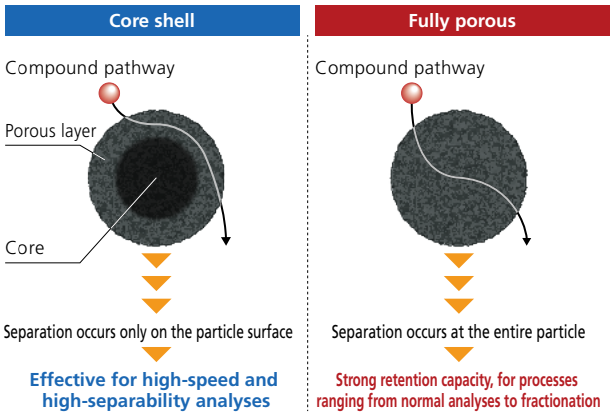


Fig. 1 Diagram of Core Shell and Fully Porous Characteristics

1-3. Reversed Phase Columns

With reversed phase columns, base materials introduced in 1-2. are chemically modified with alkyl chains to form the substrate used as the base packing materials. Note that silica and organosilica are often used as the base materials in reversed phase columns. Reversed phase columns with a polymer material are typically selected when the mobile phase pH must be set beyond the range of use of silica or organosilica base materials, or when poor separability due to an un-reacted silanol group on the silica surface cannot be resolved.

Typical functional groups to be modified chemically on reversed phase column base materials are as follows.

- ·C30
- ·C1
- ·C18
- ·Phenyl
- ·C8
- ·PFPP (Pentafluorophenylpropyl)
- ·C4
- and other functional groups

C18 columns are the most commonly used reversed phase columns. In a C18 column, the base material is chemically modified with carbon chains that have 18 carbons (C) in the chain. Note that columns in which a silica or organosilica base material is modified with an octadecylsilyl group (Octadecylsilyl: ODS, C18 group) are referred to as ODS columns, and are the most commonly used reversed phase columns.

Conversely, the mobile phase used in reversed phase mode is aqueous in nature- typically an organic solvent including methanol, acetonitrile, or tetrahydrofuran. In reversed phase mode, the first thing to consider is the use of a C18 column, followed by changes to the mobile phase, and adjustments to the ratio of aqueous and organic solvents in the mobile phase. This is done to optimise separation conditions.

In this case, if favorable separation is not obtained even after changing the mobile phase type and the ratio of water to organic solvents in the mobile phase, columns with other functional groups should be considered, corresponding to the hydrophobic retention strength and chemical properties of the target compounds.

For example, if the compound is not eluted within an appropriate period even under the strongest elution capacity mobile phase conditions (such as a 100 % organic solvent), consider using a column with a weaker retention capacity. Conversely, if the compound is not retained for an appropriate period even under the weakest elution capacity conditions, consider using a column with a stronger retention capacity. From another perspective, you might also consider a column with bonded phenyl or PFPP functional groups if you wish to achieve favorable separation using an interaction based on something other than hydrophobicity.

As seen in Fig. 2, separation behavior varies with the functional group of the reversed phase column. In comparing C4, C8, and C18 functional groups that differ in the length of their carbon chains, it is evident that the shorter the carbon chain the shorter the retention time for hydrophobic compounds. Additionally, with phenyl and PFPP columns, in comparison with C18 columns, there are multiple interactions with the compounds from the functional group, causing the compound retention times and separation patterns to vary significantly.

The reversed phase columns examined in this article are mainly C4, C8, and C18 columns in which the alkyl chains are chemically modified on the base material. As noted above, changing the functional group bonded to the base material changes the separation behavior. Additionally, differences in separation behavior occur as the result of differences in the base packing material used and the manufacturing methods even in C18 and other columns with the same chemically modified functional group.

Examples of where the separation patterns differ when comparing the same functional groups are shown in Table 3. Note that differences in separation behavior other than those shown in Table 3 occur due to the functional group binding type, variations in particle size, and differences in specific surface area.

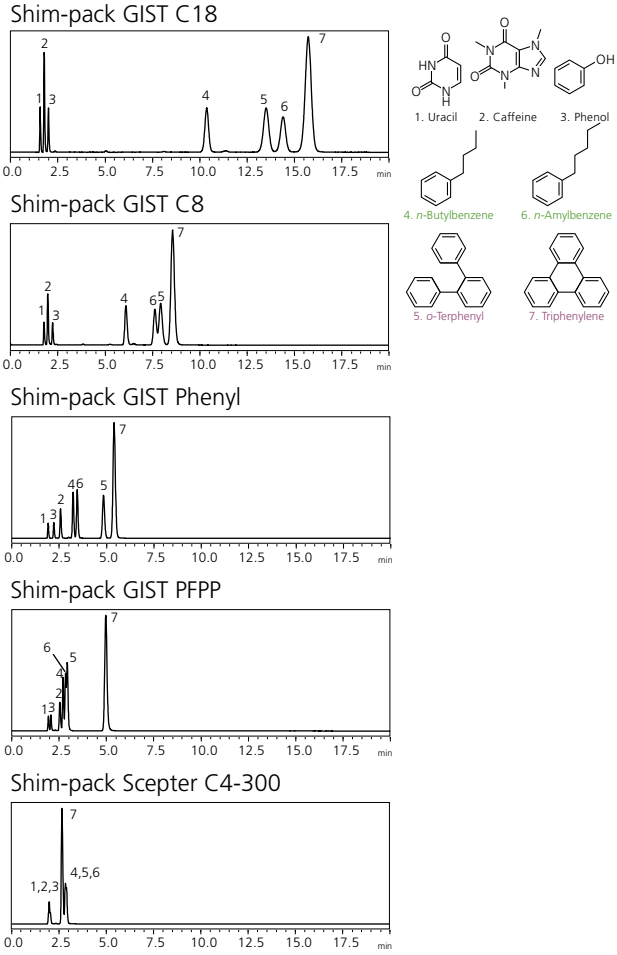


Fig. 2 Examples of the Differences in Separation Behavior Due to the Functional Groups in Reversed Phase Columns
The analysis conditions are the same as in Table 4.

Table 3 Examples Where Separation Behavior Differs Even for Reversed Phase Columns with the Same Functional Groups

Examples	Remarks
Differences in Carbon Content	Generally, the larger the carbon content, the stronger the retention capacity for compounds in reversed phase mode.
Differences in Pore Size	Depending on the relationship between pore size and compound size, the diffusion of the compound within the packing material changes, and so the retention capacity for the compound changes.
End Capping	· End capping reduces the residual silanol in the base material in columns in which silica base materials and organosilica base materials are used · The chemical interactions with the compounds change
Differences in Column Tube Materials	Depending on differences in the materials in the column's wetted parts, interactions with the compounds change, which can have a particular impact on the degree of compound adsorption.

There are numerous reversed phase columns currently available that differ in design concept, based on differences in separation behavior. It is often the case that given the number of columns that are commercially available, it is difficult to assess how the basic separation characteristics of each column differ, further complicating column selection.
In this article, Shim-pack C18 columns as well as columns with other functional groups were evaluated consistently using a C18 column evaluation method[1]. Here, we describe differences in the usage and features of each column, through highlighting differences in separation selectivity of C4, C8, and C18 columns available in the Shim-pack series of reversed phase columns.

2. Test Conditions and Evaluative Indices

The test conditions were implemented with reference to the comparative evaluation method for reversed phase C18 columns known as the Tanaka Test method[1], which is shown in Table 4.

Table 4 Analysis Conditions

System	:	Nexera™ X2
Column*	:	150 mm × 4.6 mm I.D., 5 µm
Mobile phase	:	(1) Water / Methanol = 20 : 80 (2) Water / Methanol = 70 : 30
Flow Rate	:	1.0 mL/min
Column Temp.	:	40 ˚C
Injection vol.	:	1 µL
Detection	:	254 nm
Sample	:	1. Uracil 2. Caffeine 3. Phenol 4. Butylbenzene 5. Terphenyl 6. *o*-Amylbenzene 7. Triphenylene
Vial	:	TORAST™ Vial ** (Shimadzu GLC)

* Shim-pack FC-ODS only, 150 mm x 4.6 mm I.D., 3 µm used
** P/N: GLCTV-801 (vial) + GLCTV-803 (cap)

In the Tanaka Test Method, four attributes were calculated for each column. The basic characteristics of the respective attributes are shown in Table 5. Note that the retention index k for each compound was calculated from the retention time t for each compound with the retention time for Uracil used as t0.

Table 5 Explanation of the Attributes in the Tanaka Test Method Implemented in this Article

Attribute	Index	Mobile Phase Conditions	Explanation
Hydrophobic Retention Capacity	k 6.Amylbenzene	Mobile Phase (1) (See Table 4)	Indicates the strength of the retention capacity for hydrophobic compounds
Responsiveness to Hydrophobic Differences	k 6.Amylbenzene / k 4.Butylbenzene	Mobile Phase (1) (See Table 4)	Indicates the level of the capacity to recognize differences in hydrophobicity between compounds
Responsiveness to Structure	k 7.Triphenylene / k 5.*o*-Terphenyl	Mobile Phase (1) (See Table 4)	This indicates the level of the capacity to recognize differences between plane structures and three-dimensional structures.
Responsiveness to Hydrogen Bonds	k 2.Caffeine / k 3.Phenol	Mobile Phase (2) (See Table 4)	Indicates the level of the capacity to recognize hydrogen bonds (or the degree of polarization of compounds)

A general explanation and a list of cautions regarding these attributes are shown below. In terms of cautions, the analysis conditions in Table 4, which made use of the Tanaka Test Method as a reference, are generally used in evaluations of C18 columns. Also, each attribute indicates the general features of the various columns. The strength of the interaction between compounds and columns varies according to compounds, mobile phase conditions and other analysis conditions. This means that these features are not expressed in the same way, regardless of the analysis.

■ Hydrophobic Retention Capacity:
This indicates the strength of the retention capacity with respect to hydrophobic compounds. In particular, this indicates the strength of the retention capacity for low molecular weight hydrophobic compounds under water and methanol conditions. In general, the strength of the retention capacity for hydrophobic compounds is said to be related to the number of alkyl chains in the column and the specific surface area. However, if a mobile phase with π electrons such as acetonitrile is used, the interaction between the column and the compounds will change, and the retention capacity could change significantly depending on the types of column and compound. The size of the pores also has an impact on the compound retention mechanism. If compounds of larger molecular weight are analyzed, retention might be weak even when the hydrophobic retention capacity value is high, if a column with a small pore size is used. For details, see "3-4 Wide Pore Reversed Phase Columns."

■ Responsiveness to Hydrophobic Differences:
This indicates the level of the capacity to recognize differences in hydrophobicity between compounds. Columns with a high responsiveness to hydrophobic differences demonstrate a difference in elution time between amyl benzene and butyl benzene. In particular, the level of recognition of hydrophobic differences between compounds in the low molecular weight region is shown under water and methanol conditions. Responsiveness to hydrophobic differences is related to the length and amount of alkyl chains in the column.

■ Responsiveness to Structure:
This indicates the level of the capacity to recognize differences between plane structures and three-dimensional structures. The greater the expression of the difference in elution time between o-terphenyl and triphenylene, the higher the responsiveness to structure. In general, this is related to the functional group binding density and the functional group binding treatment method. For example, in columns where a high octadecyl group density has been introduced, triphenylene, which has a plane structure, fits between the alkyl chains and is strongly retained. In contrast, *o*-terphenyl, which has a bulky, three-dimensional structure, tends not to fit between the alkyl chains, and is retained comparatively weakly[2].

■ **Responsiveness to Hydrogen Bonds:**
This indicates the level of responsiveness to hydrogen bonds. C18 columns, which have silica and organosilica base materials, tend to have a lower responsiveness to hydrogen bonds compared to columns less affected by residual silanol groups. For this reason, the value tends to be higher for C18 columns without end capping. Note that in this article, the height of the value for responsiveness to hydrogen bonds is not necessarily related to the strength of the retention capacity for hydrophilic compounds.

In terms of the characteristics of each column, spider charts based on these four attributes are presented (see page 6). Note that these spider charts were calculated from more than 30 test results for reversed phase columns in the Shim-pack lineup.
One note of caution in the explanation of these spider charts is that the size of the area occupied in the spider chart does not necessarily indicate the level of performance by that column. For example, the larger the area occupied in the spider chart, the easier it is for various interactions to have an impact on separation. As such, even if all the column attributes are high, this does not mean that favorable separation is achieved. On the contrary, predictions about separation behavior are difficult, and the desired degree of separation is not always obtainable.
In addition, the hydrophobic retention capacity value for the Shim-pack Scepter HD-C18-80 does not fit into the spider charts. Furthermore, the hydrophobic retention capacity value for the Shim-pack Scepter HD-C18-80 does not fit into the spider charts. This was due to the difficulties surrounding spider chart calculations of other columns, particularly concerning hydrophobic retention capacity.
The attributes and spider charts shown in this article are not characteristics that are necessarily demonstrated in all applications. They should instead be treated as column characteristics, and used as a reference.

3. Results of the Tanaka Test for Shim-pack Reversed Phase Columns and Their Features

This article will introduce the results of the Tanaka Test for Shim-pack reversed phase columns, and the features of each column.
Note that in this evaluation method, a conventional HPLC analysis column size of 150 mm x 4.6 mm I.D., 5 µm was used. A 150 mm x 4.6 mm I.D., 3 µm column size was only used for the Shim-pack FC-ODS. The specifications for the Shim-pack reversed phase columns are shown in Table 6.

3-1. Shim-pack Reversed Phase C18 Columns

Chromatograms for each column under mobile phase (1) conditions, and spider charts for their four attributes are shown in on page 6.
In comparison to columns with other functional groups, C18 columns tend to have a high hydrophobic retention capacity and a high responsiveness to hydrophobic differences, due to the presence of alkyl chains with octadecyl groups.
In particular, with their limited responsiveness to structure and hydrogen bonds, separation behavior in C18 columns is mainly due to hydrophobic interaction. Such columns easily demonstrate separation in accordance with the hydrophobicity of the compounds, and their comparative separation behavior is easy to predict. This allows them to be the superior choice as a general-purpose column. The general purpose Shim-pack C18 columns include the following.

- Shim-pack Scepter C18-120 (Fully Porous Organosilica)
- Shim-pack GIST C18 (Fully Porous Silica)
- Shim-pack GIS C18 (Fully Porous Silica)
- Shim-pack GWS C18 (Fully Porous Silica)
- Shim-pack VP-ODS/XR ODS, ODS II, ODS III (Fully Porous Silica)
- Shim-pack FC-ODS (Fully Porous Silica)

On the other hand, if the separation selectivity of a general purpose C18 column is the be changed, using a C18 column with high levels of responsiveness to structure and hydrogen bonds, such as polar embedded types and high-density carbon chain types is an effective approach. There are also special columns although not evident through the spider charts, demonstrate their effectiveness under special analysis conditions. Such columns make an effective choice as a No. 2 C18 column. The special Shim-pack columns include the following.

- Shim-pack Scepter C18-300 (Wide Pore Column)
- Shim-pack Scepter HD-C18-80 (High-Density Carbon Chain Column)
- Shim-pack GIST C18-AQ (Adjustable Bond Spacing Carbon Chain Column)
- Shim-pack GISS C18 (Fully Porous Column for High-Speed Analysis)
- Shim-pack Arata C18 (Special End Capping)
- Shim-pack GIS C18-P (High-Density Carbon Chain Column)
- Shim-pack GIS RP-Shield (Polar Group Embedded Column)
- Shim-pack MAqC-ODS I (Metal Containing Column)
- Shim-pack Velox C18 (Surface Porous Silica Column)
- Shim-pack Velox SP-C18 (Surface Porous Silica Column)

The other features of these columns that are not expressed by the spider charts are noted beside the spider charts for each column.

Table 6 List of Specifications for Shim-pack Reversed Phase Columns

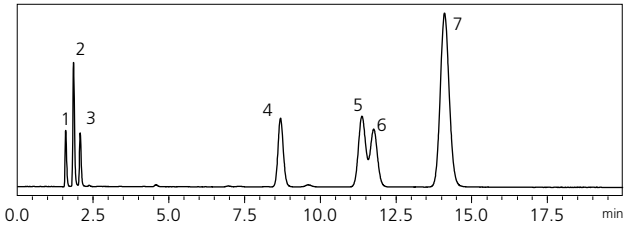
Column Base Packing Material	Column Series Name	Functional Group	Particle size (μm)	Pore Size (Å)	Surface Area (㎡/g)	Carbon Content (%)	End Capping	Operating pH Range	Use Under 100 % Aqueous Mobile Phase Conditions
Fully porous organosilica	Shim-pack Scepter	C18-120	1.9, 3, 5	120	360	20*	Yes	1-12	○
		C18-300	1.9, 3, 5	300	N.D.	N.D.	Yes	1-12	○
		HD-C18-80	1.9, 3, 5	80	430	25*	Yes	1-12	
		C8-120	1.9, 3, 5	120	360	17*	Yes	1-12	
		Phenyl-120	1.9, 3, 5	120	360	17*	Yes	1-10	○
		PFPP-120	1.9, 3, 5	120	360	15*	No	1-8	○
		C4-300	1.9, 3, 5	300	N.D.	N.D.	Yes	1-10	○
Fully porous silica	Shim-pack Arata	C18	2.2, 5	120	340	17	Yes	2-7.5	○
	Shim-pack GIST	C18	2, 3, 5	100	350	14	Yes	1-10	
		C18-AQ	1.9, 3, 5	100	350	13	Yes	1-10	○
		C8	2, 3, 5	100	350	8	Yes	1-10	○
		Phenyl	2, 3, 5	100	350	10	No	2-7.5	○
		Phenyl-Hexyl	3, 5	100	350	9	Yes	1-10	○
		PFPP	3, 5	100	350	10	Yes	2-7.5	○
	Shim-pack GISS	C18	1.9, 3, 5	200	200	9	Yes	1-10	○
		C8 (metal free body only)	1.9, 3, 5	200	200	6	Yes	1-10	○
	Shim-pack GIS	C18	2,3,4,5,10	100	450	15	Yes	2-7.5	
		C18-P	3, 5	100	450	29	No	2-7.5	
		RP-Shield	5	100	450	9	No	2-7.5	○
	Shim-pack GWS	C18	5	100	450	9.5	Yes	2-7.5	
	Shim-pack VP**	C18	5	120	410	20	Yes	2-7.5	
		C8	5	120	410	12.5	Yes	2-7.5	
		Phenyl	5	120	410	12.3	Yes	2-7.5	
	Shim-pack MAqC	ODS I	5	120	N.D.	13	Yes	2-4	
	Shim-pack FC	ODS	3	120	315	18	Yes	1.5-9	
Core shell silica	Shim-pack Velox	C18	1.8, 2.7, 5	90	125, 130, 100	9, 7, 5	Yes	2-8	
		SP-C18	1.8, 2.7, 5	90	125, 130, 100	7, 7, 5	No	1-8	
		Biphenyl	1.8, 2.7, 5	90	125, 130, 100	7, 7, 5	Yes	1.5-8	○
		PFPP	1.8, 2.7, 5	90	125, 130, 100	4, 4, 3	No	2-8	○

* Includes the percentage carbon content from the organosilica base material.

** The Shim-pack XR series is the high-speed analysis column series in the Shim-pack VP series.

Shim-pack Scepter C18-120

General-purpose fully porous organosilica C18 | Prep size available



Features:

- Uses a highly chemically durable organosilica base material
- A first-choice general-purpose C18 column
- A reversed phase C18 column that can be used in a variety of circumstances, with three column bodies and high pH resistance
- A first-choice column for reversed phase nucleic acid pharmaceuticals analysis

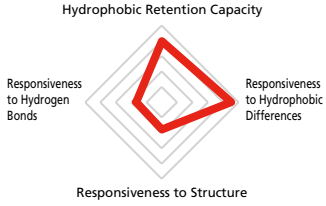
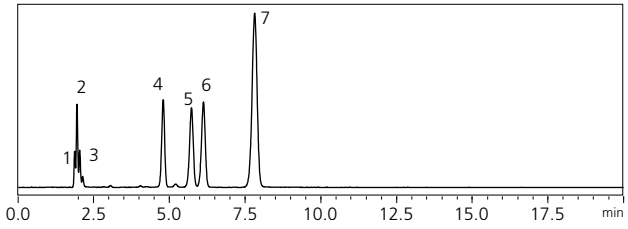


Fig. 3 Results for the Shim-pack Scepter C18-120

Shim-pack Scepter C18-300

Special fully porous organosilica C18 | Prep size available



Features:

- Uses a highly chemically durable organosilica base material
- This wide pore C18 column is suitable for the analysis of medium to high molecular weight substances including peptides, proteins, and oligonucleic acids
- Recommended for reversed phase nucleic acid pharmaceuticals analysis, due to its high pH resistance and two types of metal-free column body

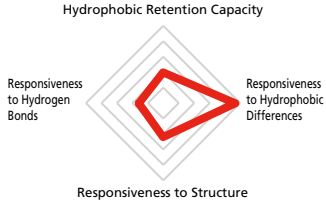
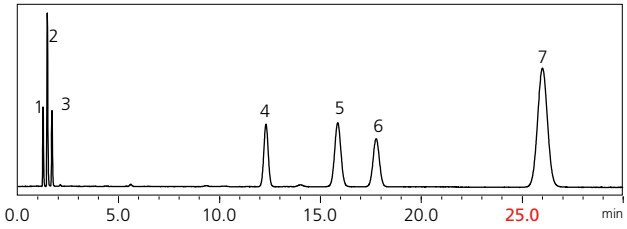


Fig. 4 Results for the Shim-pack Scepter C18-300

Shim-pack Scepter HD-C18-80

Special fully porous organosilica C18 | Prep size available



Features:

- Uses a highly chemically durable organosilica base material
- Of all the Shim-pack columns, this one has the highest hydrophobic retention capacity due to its high carbon content
- With its high-density modified C18, this column features excellent hydrophobic retention capacity, responsiveness to hydrophobic differences, and responsiveness to structure. It is intended for the separation of analogues and constitutional isomers with C18

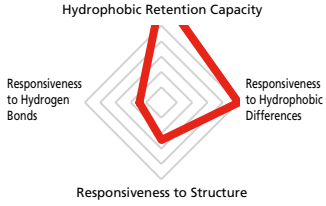
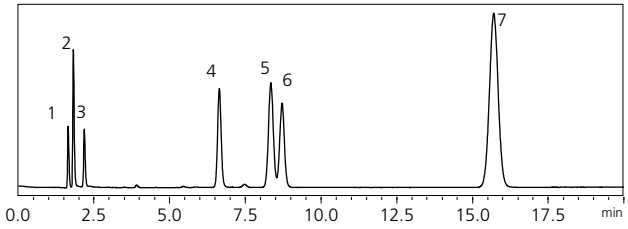


Fig. 5 Results for the Shim-pack Scepter HD-C18-80

Shim-pack Arata C18

Special fully porous silica C18



Features:

- Column with proprietary end capping
- Fast column equilibration under formic acid/acetic acid mobile phase conditions, with sharp peaks obtainable for acidic, neutral, and basic compounds
- It has unique separation characteristics, with low responsiveness to hydrogen bonds and high responsiveness to structure. It is recommended when you wish to change the separation pattern but still use a C18 column

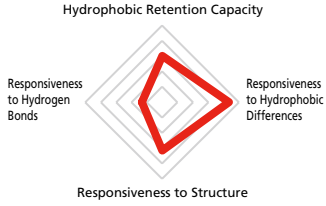
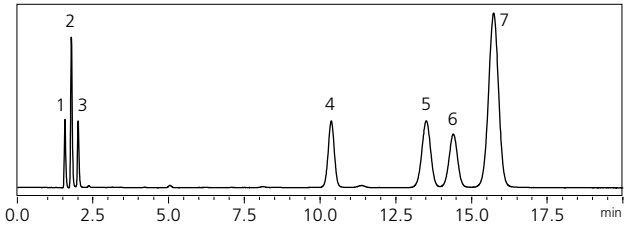


Fig. 6 Results for the Shim-pack Arata C18

*1. These test results will not necessarily be obtained in all applications.
*2. If you would like fractionation size in a column series that does not list "Prep size available," contact your local Shimadzu representative.

Shim-pack GIST C18

General-purpose fully porous silica C18 | Prep size available



Features:
- Highly pH resistant (pH 1 to 10) with a highly inert silica base material
- A first-choice general-purpose C18 column
- Of all the Shim-pack reversed phase columns, this one has the highest values for hydrophobic retention capacity and responsiveness to hydrophobic differences

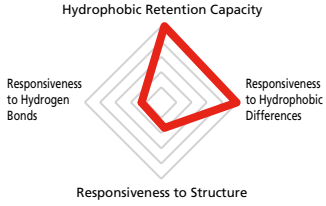
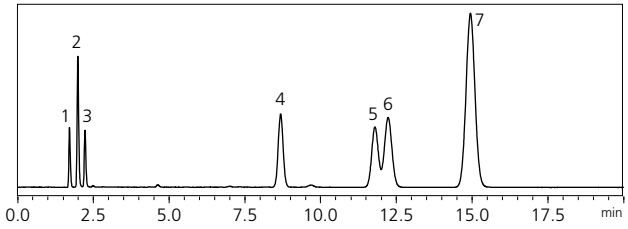


Fig. 7 Results for the Shim-pack GIST C18

Shim-pack GIST C18-AQ

Special fully porous silica C18 | Prep size available



Features:
- Highly pH resistant (pH 1 to 10) with a highly inert silica base material
- The bond spacing of the C18 is adjustable to suit the retention of hydrophilic compounds
- Recommended if you wish to retain hydrophilic compounds as well with a Shim-pack C18 column

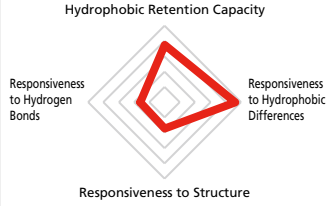
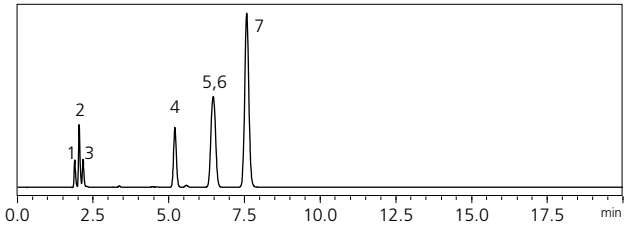


Fig. 8 Results for the Shim-pack GIST C18-AQ

Shim-pack GISS C18

Special fully porous silica C18 | Prep size available



Features:
- Highly pH resistant (pH 1 to 10) with a highly inert silica base material
- This is a wide pore column, with a low hydrophobic retention capacity, intended for high-speed analysis
- The pore size is wider than in general C18 columns, so it is recommended for the analysis of peptides and other fairly large molecular weight compounds

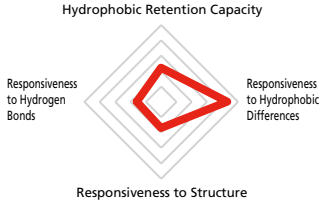
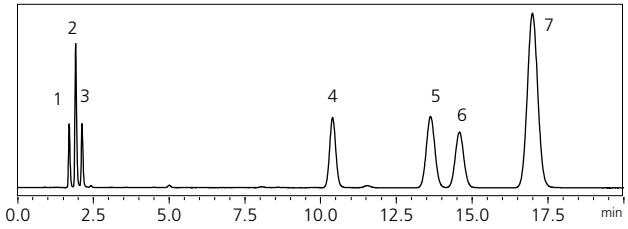


Fig. 9 Results for the Shim-pack GISS C18

Shim-pack GIS C18

General-purpose fully porous silica C18 | Prep size available



Features:
- Uses a standard silica base material
- This is the most standardized of the Shim-pack C18 columns, with strong retention, limited adsorption of ionic compounds, and a good balance of attribute values
- A large I.D. size is available, so it is recommended for those considering large capacity fractionation

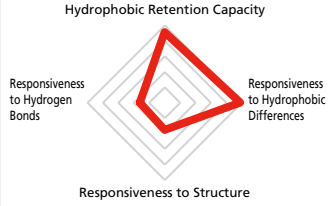
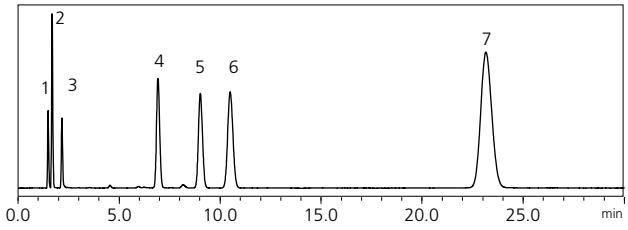


Fig. 10 Results for the Shim-pack GIS C18

*1. These test results will not necessarily be obtained in all applications.
*2. If you would like fractionation size in a column series that does not list "Prep size available," contact your local Shimadzu representative.

Shim-pack GIS RP-Shiled

Special fully porous silica C18 | Prep size available



Features:

- In this column, the standard silica base material is modified with polar group embedded C18
- The interaction between compounds and residual silanol groups is reduced, and the responsiveness to hydrogen bonds is low
- The polar group has unshared electron pairs, so mutual interactions are seen depending on the electron profile of the compound, which is recommended if you wish to change the separation pattern while using a C18 column

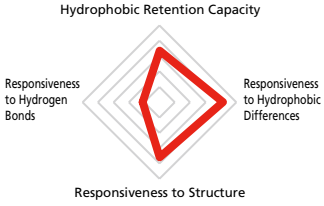
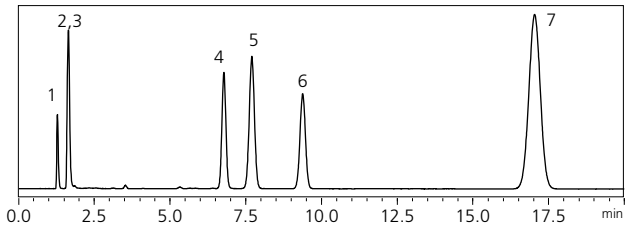


Fig. 11 Results for the Shim-pack GIS RP-Shield

Shim-pack GIS C18-P

Special fully porous silica C18 | Prep size available



Features:

- In this column, the standard silica base material is modified with high-density C18
- This features excellent responsiveness to hydrophobic differences and responsiveness to structure, and is intended for the separation of analogues and constitutional isomers with C18
- Of all the Shim-pack reversed phase columns, this one has the highest carbon content

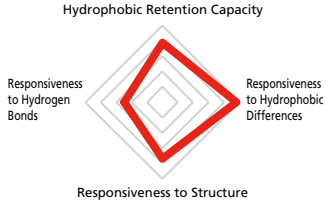
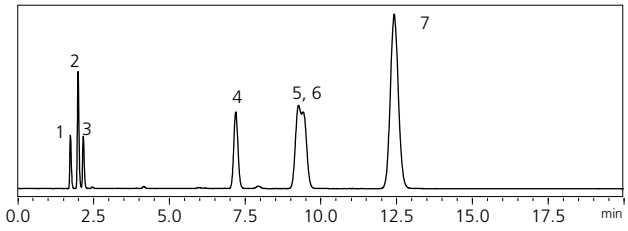


Fig. 12 Results for the Shim-pack GIS C18-P

Shim-pack GWS C18

General-purpose fully porous silica C18



Features:

- Uses a standard silica base material
- This standard C18 column has a good balance of attribute values
- Recommended for those trying to control costs

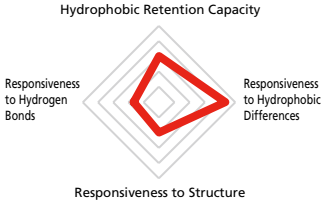
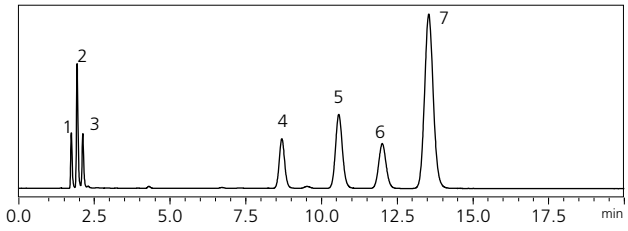


Fig. 13 Results for the Shim-pack GWS C18

Shim-pack VP-ODS

General-purpose fully porous silica C18



Features:

- Uses a standard silica base material meeting the strictest quality controls
- This standard C18 column has a good balance of attribute values, and is optimal for standard analyses
- The Shim-pack XR ODS, ODS II, and ODS III columns are suitable for the high-speed analysis applications of a Shim-pack VP-ODS

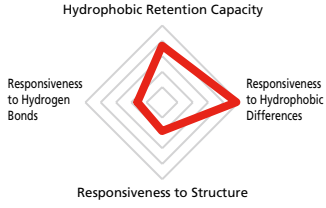
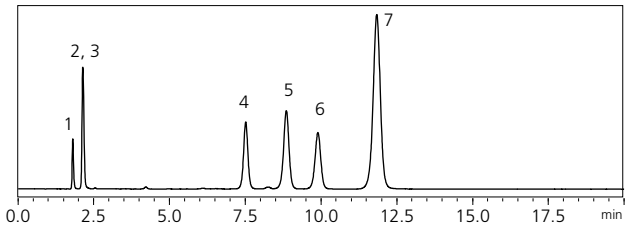


Fig. 14 Results for the Shim-pack VP-ODS

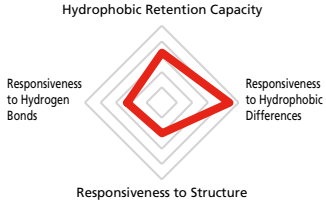
*1. These test results will not necessarily be obtained in all applications.
*2. If you would like fractionation size in a column series that does not list "Prep size available," contact your local Shimadzu representative.

Shim-pack MAqC-ODS I*

Special fully porous silica C18



Features:
- Uses a silica base material containing metal as the packing material
- This column has cation exchangeability via aquo-ions centering around the metal ions, and is excellent for retaining highly polar basic compounds without using ion pair reagents
- Lineup with 5 µm only

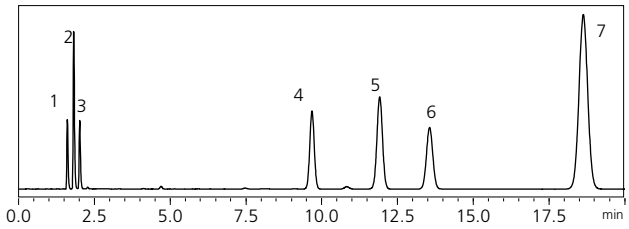


* The analysis conditions implemented with this evaluation method were outside the recommended operating range (outside the recommended operating pH range) noted in the instruction manual for the Shim-pack MAqC-ODS I. Please be careful to read the instruction manual thoroughly before actually using this product.

Fig. 15 Results for the Shim-pack MAqC-ODS

Shim-pack FC-ODS

General-purpose fully porous silica C18



Features:
- This column uses a highly inert silica base material to limit hydrophilic interactions to the utmost
- This has a high responsiveness to hydrophobic differences and a low responsiveness to hydrogen bonds, so it acts as a simple reversed phase C18 column
- Lineup with 3 µm only

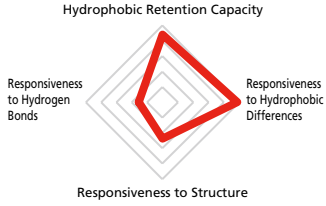
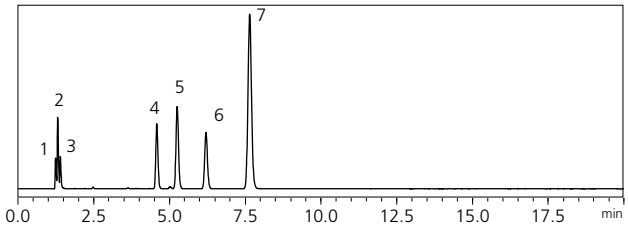


Fig. 16 Results for the Shim-pack FC-ODS

Shim-pack Velox C18

Special core shell silica C18



Features:
- Uses a core shell silica base material
- This is a general-purpose C18 core shell silica column, and is recommended for high-speed analysis
- Of all the Shim-pack columns, this one has balanced separation selectivity including responsiveness to hydrogen bonds, and acts as a simple reversed phase C18 column

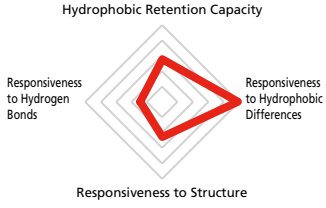
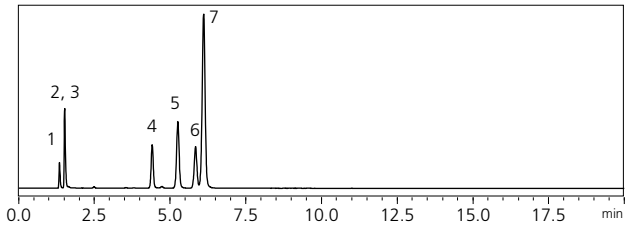


Fig. 17 Results for the Shim-pack Velox C18

Shim-pack Velox SP-C18

Special core shell silica C18



Features:
- Uses a core shell silica base material
- This is a general-purpose C18 core shell silica column, and is recommended for high-speed analysis using a strongly acidic mobile phase
- This column has a balance separation selectivity different from the Shim-pack Velox C18

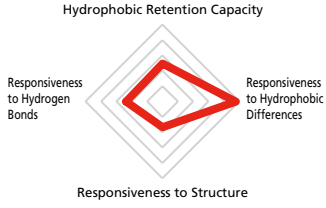


Fig. 18 Results for the Shim-pack Velox SP-C18

*1. These test results will not necessarily be obtained in all applications.
*2. If you would like fractionation size in a column series that does not list "Prep size available," contact your local Shimadzu representative.

3-2. Shim-pack Reversed Phase C4 and C8 Columns

Chromatograms for each column under mobile phase (1) conditions, and spider charts for their four attributes are shown below.

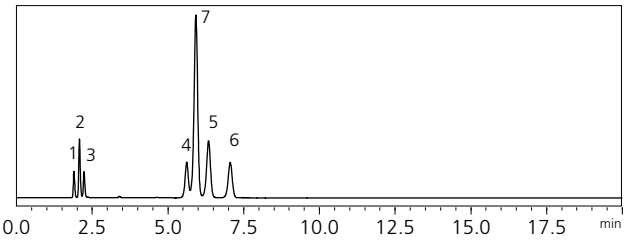
In terms of the C4 and C8 functional groups, the length of a single carbon chain is shorter than with C18, and the C4 and C8 columns contain less carbon in comparison to C18 columns. This means that the hydrophobicity for C4 and C8 is reduced when compared to a C18 column. Accordingly, they feature reduced values for hydrophobic retention capacity and responsiveness to hydrophobic differences in comparison to a C18 column. As a result, C4 and C8 columns are used for the purpose of improving retention and increasing analysis throughput when retention is too strong with a C18 column

The following C4 and C8 Shim-pack columns are available.

- **Shim-pack VP-C8 (Fully Porous Silica)**
- **Shim-pack Scepter C8-120 (Fully Porous Silica)**
- **Shim-pack GIST C8 (Fully Porous Silica)**
- **Shim-pack GISS C8 (Fully Porous Silica)**
- **Shim-pack Scepter C4-300 (Fully Porous Silica)**

Shim-pack VP-C8

General-purpose fully porous silica C8



Features:
- Separation selectivity is different from the Shim-pack VP-ODS
- In these results, the elution order was different from the Shim-pack VP-ODS
- The separation selectivity was comparable to the 2.2 µm particle size Shim-pack XR-C8

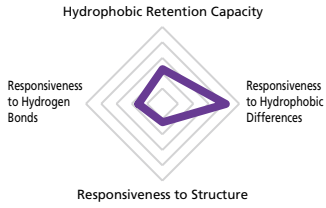
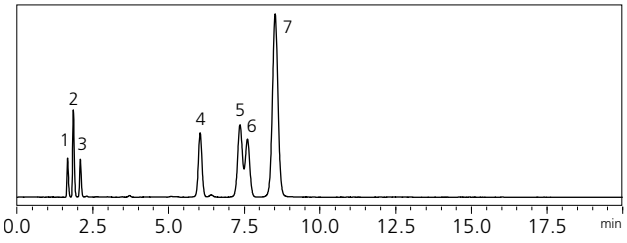


Fig. 19 Results for the Shim-pack VP-C8

Shim-pack Scepter C8-120

General-purpose fully porous organosilica C8 | Prep size available



Features:
- Separation selectivity is different from the Shim-pack Scepter C18-120
- From the spider charts, it appears similar to the Shim-pack GIST C8, but the elution pattern is different
- In these results, the elution order was similar to the Shim-pack Scepter C18-120
- High pH resistance (Operating pH range: 1 to 12)

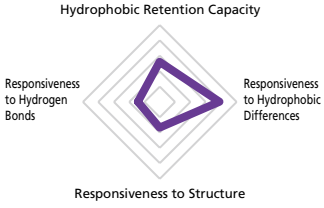
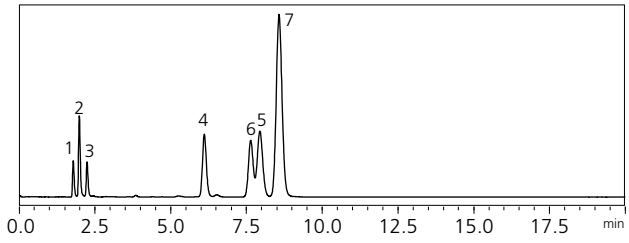


Fig. 20 Results for the Shim-pack Scepter C8-120

Shim-pack GIST C8

General-purpose fully porous silica C8 | Prep size available



Features:
- Separation selectivity was different from the Shim-pack GIST C18
- From the spider charts, it appears similar to the Shim-pack Scepter C8-120, but the elution pattern is different
- In these results, the elution order was different from the Shim-pack GIST C18
- High pH resistance (Operating pH range: 1 to 10)

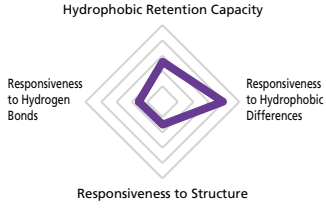
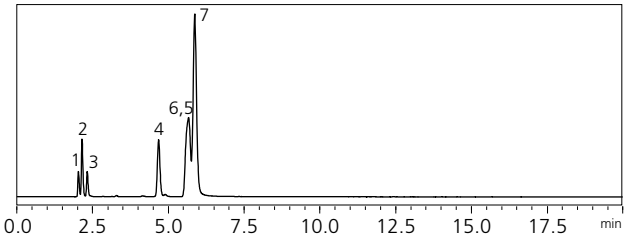


Fig. 21 Results for the Shim-pack GIST C8

Shim-pack GISS C8

Special fully porous silica C8



Features:
- Separation selectivity was different from the Shim-pack GISS C18
- Wide pore C8 columns that are also suitable for the analysis of peptides
- High pH resistance (Operating pH range: 1 to 10)
- In terms of the analysis scale, the lineup only includes metal-free inert columns in which PEEK is used

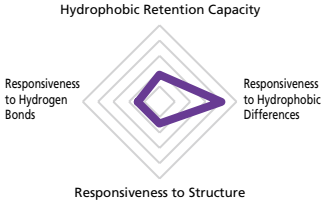
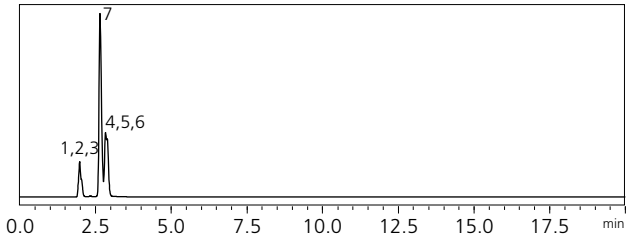


Fig. 22 Results for the Shim-pack GISS C8

Shim-pack Scepter C4-300

Special fully porous organosilica C4 | Prep size available



Features:
- Of all the Shim-pack reversed phase columns, this one has the lowest hydrophobic retention capacity
- This wide pore C4 column is suitable for the analysis of large molecular weight compounds (such as peptides and proteins)

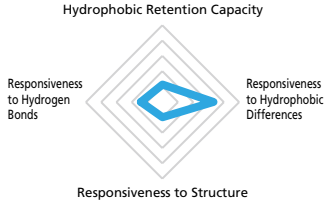


Fig. 23 Results for the Shim-pack Scepter C4-300

*1. These test results will not necessarily be obtained in all applications.
*2. If you would like fractionation size in a column series that does not list "Prep size available," contact your local Shimadzu representative.

3-3. Summary of the Shim-pack Reversed Phase Column Series

To highlight the hydrophobicity of the columns used in this evaluation, the hydrophobic retention capacity and responsiveness to hydrophobic differences for each column are shown in Fig. 24.
From Fig. 24, it is evident from comparing C4, C8, and C18 columns, that the shorter the carbon chain, the lower the responsiveness to hydrophobic differences and hydrophobic retention capacity. In comparing the C18 columns themselves, it is evident that the hydrophobic retention capacity varies from column to column, and that they have significantly different values.
This is a feature of each column, for example the Shim-pack GISS C18 for high-speed analysis applications and the Shim-pack Velox C18 and Velox SP-C18 core shell columns all have a low hydrophobic retention capacity.
As an example, when selecting a column, if you are having difficulty with separation using the Shim-pack GIST C18 (Fig. 24), even after changing the analysis conditions, consider the Shim-pack GISS C18, the core shell silica Shim-pack Velox C18, and Velox SP-C18, or a C4 or C8 column when reducing the retention. Conversely, for higher retention than the Shim-pack GIST C18, consider using the Shim-pack Scepter HD-C18-80.

As indicated above, even for C18 columns in the Shim-pack lineup, differences in the packing material result in significant differences in separation behavior. Accordingly, in method development for HPLC analysis, an effective approach is to consider the use of columns from series with different packing materials even though the functional groups are the same.
Note however, that the evaluation method implemented in this article is just one aspect of the characterization of the various reversed phase columns. In actual analysis, separation might be hard to predict in some cases even after assessing the basic column characteristics, due to the analysis conditions and compatibility with the compounds. At the same time, in HPLC analysis, assessing the basic column characteristics before implementing an analysis leads to an understanding of the separation results obtainable.
Actual reversed phase column selection can be complicated in some cases, but make your selection in accordance with the analysis conditions, while bearing in mind the basic characteristics as introduced in this article. If you run into difficulties in selecting from the Shim-pack reversed phase columns, refer to the selection chart for shim-pack reversed phase columns shown in Fig. 39 on page 12 of this article.

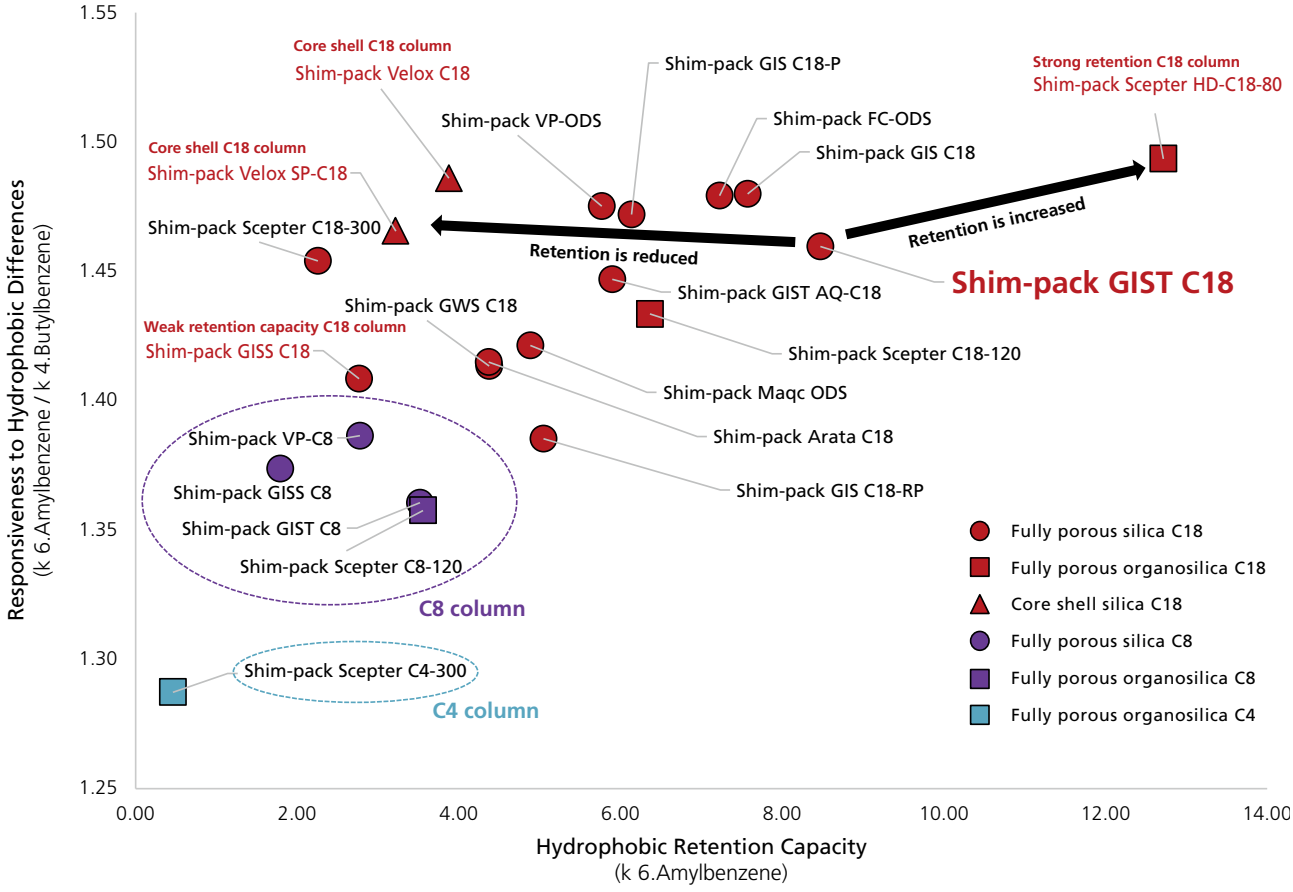


Fig. 24 Relationship Between Hydrophobic Retention Capacity and Responsiveness to Hydrophobic Differences in the Shim-pack C4, C8, and C18 Columns

3-4. Wide Pore Reversed Phase Columns

So-called wide pore columns are one type of commercially available reversed phase column. As mentioned previously, pore size in a general reversed phase column is approximately 90 to 120 Å. In contrast, there are columns (hereinafter referred to as wide pore columns) with a larger pore size than usual, on the order of 200 to 1,000 Å.

There are three series of wide pore Shim-pack reversed phase columns.

- **Shim-pack Scepter C18-300 (Pore size: 300 Å)**
- **Shim-pack GISS C8 (Pore size: 200 Å)**
- **Shim-pack Scepter C4-300 (Pore size: 300 Å)**

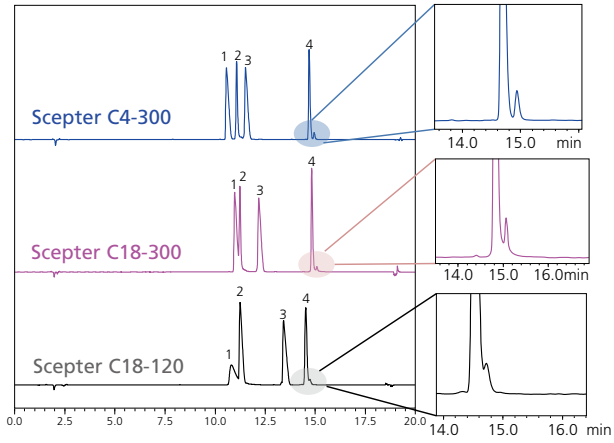
In comparison to columns with the same functional group in other series, wide pore columns have weaker retention for low molecular weight hydrophobic compounds. Wide pore columns feature a stronger retention of peptides, proteins, and other compounds with a larger molecular weight in comparison to smaller molecular weight compounds.

Fig. 25 shows the results of peptide analysis with different molecular weights using the Shim-pack Scepter C18-120 (pore size: 120 Å), the Shim-pack Scepter C18-300 (pore size: 300 Å), and the Shim-pack Scepter C4-300 (pore size: 300 Å).

In terms of the Tanaka Test results, the hydrophobic retention capacities of the Shim-pack Scepter C18-300 and Shim-pack Scepter C4-300 are less than with the Shim-pack Scepter C18-120. In Fig. 25 however, the retention times for 1. Ribonuclease A, 2. Angiotensin II, and 4. Insulin were equivalent or longer in comparison to the Shim-pack Scepter C18-120. At the same time, the retention time for 3. Leusin-Enkepharin, which has a molecular weight under 1,000, is longer with the Shim-pack Scepter C18-120, meaning that the retention has become stronger according to the Tanaka Test hydrophobic retention capacity value.

This separation behavior is believed to be due to a size exclusion effect which depends on the size of the compound. If the compound can penetrate the column pores, it is strongly retained, but if it cannot, then it is weakly retained. Compound retention is also performed within the pores, so the pore size must enable the compounds to penetrate into the pores. (See Fig. 26)

In actual reversed phase analysis, whether the strength of the interaction between the column and compounds dominates or whether the size exclusion effect dominates will depend on the compound characteristics and the analysis conditions, which can be varied. Consider using a wide pore column in accordance with the size of the compound to be retained.



System	:	Nexera XR
Column	:	150 mm × 4.6 mm I.D., 5 µm (each)
Mobile phase	:	A) 0.1% formic acid in water B) Acetonitrile
Flow Rate	:	1.0 mL/min
Time program	:	B conc. 5% (0-2 min) → 40% (17 min) → 95% (17.01-22 min) → 95% (22.01-27 min)
Column Temp.	:	40 ˚C
Injection vol.	:	10 µL
Detection	:	220 nm
Vial	:	TORAST -H Bio Vial* (Shimadzu GLC)
Sample	:	1. Ribonuclease A (100 mg/L), Molecular weight = 13,680 2. Angiotensin II (100 mg/L), Molecular weight = 1,046 3. Leusin-Enkepharin (100 mg/L), Molecular weight = 555 4. Insulin (100 mg/L), Molecular weight = 5,808

* P/N :370-04350-00

Fig. 25 Comparative Peptide Analyses Using Columns with Different Pore Sizes and Hydrophobic Retention Capacities

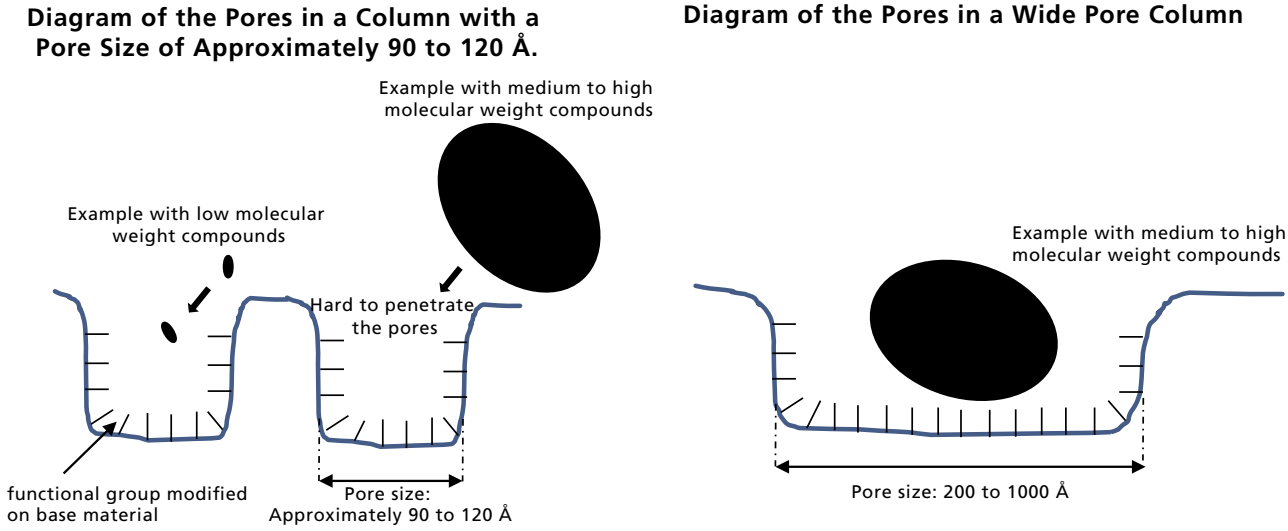


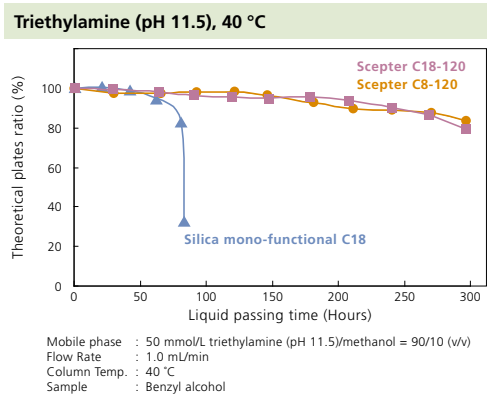
Fig. 26 Diagrams of the Pores in a General Column and a Wide Pore Column

4. Introduction to the Shim-pack Column Series

4-1. Shim-pack Scepter Series

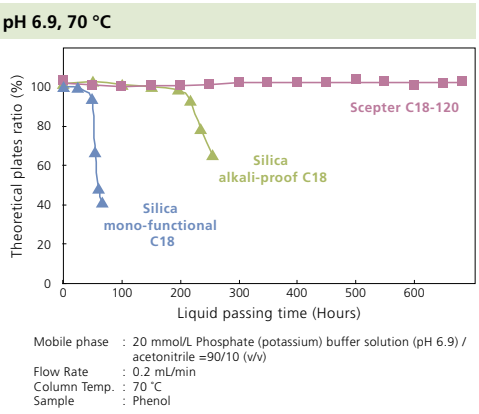
The Shim-pack Scepter series, which uses an organosilica base material, features excellent durability and performance across a wide range of conditions. The Shim-pack Scepter series is recommended as a first-choice of column series.

[Durability at High pH]



Mobile phase : 50 mmol/L triethylamine (pH 11.5)/methanol = 90/10 (v/v)
Flow Rate : 1.0 mL/min
Column Temp. : 40 °C
Sample : Benzyl alcohol

[Durability at high temperatures]



Mobile phase : 20 mmol/L Phosphate (potassium) buffer solution (pH 6.9) / acetonitrile =90/10 (v/v)
Flow Rate : 0.2 mL/min
Column Temp. : 70 °C
Sample : Phenol

Fig. 27 Excellent Durability of the Shim-pack Scepter

With extensive column stationary phases (seven reversed phases and one HILIC) differing in separation selectivity, as well as a wide range of particle and column sizes in the lineup, these are compatible with a wide range of applications from analysis to fractionation. (See Fig. 28)

Shim-pack Scepter	Reversed phase				
	C18-120	C18-300	HD-C18-80	C8-120	C4-300
Functional group type	Trifunctional bond C18	Trifunctional bond C18	Trifunctional bond C18	Trifunctional bond C8	Trifunctional bond C4
	General-purpose type	General-purpose type	High functional group density type		
Base Material	Organosilica hybrid				
Particle size	1.9 µm、3 µm、5 µm				
Pore size	12 nm	30 nm	8 nm	12 nm	30 nm
End capping	Proprietary				
Operating pH range	1 - 12				1 - 10
Used with a 100 % aqueous mobile phase	○	○	×	×	○
USP category	L1	L1	L1	L7	L26

Shim-pack Scepter	Reversed phase		HILIC
	Phenyl	PFPP	Diol-HILIC
Functional group type	Trifunctional bond Phenylbutyl	Trifunctional bond Pentafluorophenylpropyl	Trifunctional bond Dihydroxypropyl
Base Material	Organosilica hybrid		
Particle size	1.9 µm、3 µm、5 µm		
Pore size	12 nm		
End capping	Proprietary	None	
Operating pH range	1 - 10	1 - 8	2 - 10
Used with a 100 % aqueous mobile phase	○	○	–
USP category	L11	L43	L20

Fig. 28 List of the Shim-pack Scepter Series Lineup

Furthermore, two types of column body are available to accommodate adsorption suppression when analyzing biopharmaceuticals, so different column bodies can be utilized for the analysis conditions.

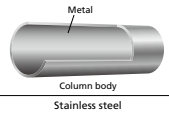
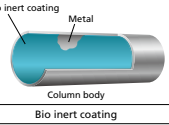
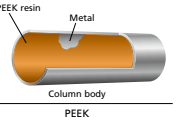
	Scepter	Scepter Claris	Scepter [metal-free]
			
Body wetted materials	Stainless steel	Bio inert coating	PEEK
Frit wetted materials	Stainless steel	Bio inert coating	PEEK

Fig. 29 Differences in Column Bodies in the Shim-pack Scepter Series

Shim-pack Scepter series

This is recommended as a first-choice of column series.

Use this QR code for details.

Fig. 30 Shim-pack Scepter Series

4-2. Shim-pack GIST Series

While demonstrating different separation behavior from Shim-pack Scepter series columns, the Shim-pack GIST series lineup, can be used with a wide range of analysis conditions, due to its extensive column chemistries (six reversed phase types and two HILIC) and particle and column sizes in the lineup. (See Fig. 31) This is as the GIST series uses a highly durable, highly inert silica base material.

In particular, the Shim-pack G series (including the Shim-pack GIST/GISS/GIS/GWS series), are well balanced columns with excellent cost performance, and are the most popular series of Shim-pack column. Of these, the Shim-pack GIST series is recommended as a first-choice of column series.

A distinctive reversed phase column in the Shim-pack GIST series is the Shim-pack GIST C18-AQ. The Shim-pack GIST C18-AQ is an effective choice if you are considering a reversed phase mode C18 column for highly polar compounds. Fig. 32 shows a comparison of the retention of hydrophilic compounds by the major Shim-pack C18 columns. The Shim-pack GIST C18-AQ is a C18 column designed for stronger retention of hydrophilic compounds in comparison to general-purpose C18 columns. The packing material surface is appropriately hydrated even under 100 % aqueous mobile phase conditions, and the column features excellent retention and repeatability. As shown in Fig. 8, it combines ample hydrophobic interaction while maintaining highly polar compounds, making it suitable for the batch analysis of a variety of polar components.

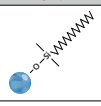
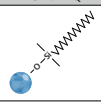
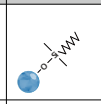
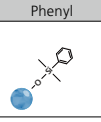
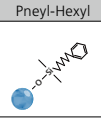
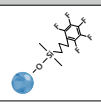
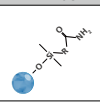
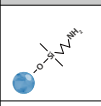
	Shim-pack GIST							
	Reversed Phase						HILIC	
	C18	C18-AQ	C8	Phenyl	Pneyl-Hexyl	PFPP	Amide	NH2
Chemistry								
Bonded Phase	Octadecyl groups	Octadecyl groups	Octyl groups	Phenyl groups	Phenyl-Hexyl groups	Pentafluorophenylpropyl groups	Carbamoyl groups	Aminopropyl groups
Features	Ultra-high inertness and high stability	Excellent retentivity of highly polar compounds	Ultra-high inertness and high stability	Extremly strong π-π interactions	Alternative selectivity to C18 columns	Excellent retentivity of highly polar base	First choice HILIC column	Sugar analysis
Particle Size (µm)	2, 3, 5	1.9, 3, 5	2, 3, 5	2, 3, 5	3, 5	3, 5	1.9, 3, 5	3, 5
Pore Size (nm)	10	10	10	10	10	10	10	10
Surface Area (m^2/g)	350	350	350	350	350	350	350	350
Carbon Loading (%)	14	13	8	10	9	10	15	7
End Cap	Yes	Yes	Yes	No	Yes	Yes	No	No
pH Range	1-10	1-10	1-10	2-7.5	1-10	2-7.5	2-8.5	2-7.5
USP Code	L1	L1	L7	L11	L11	L43	L68	L8

Fig. 31 List of the Shim-pack GIST Series Lineup

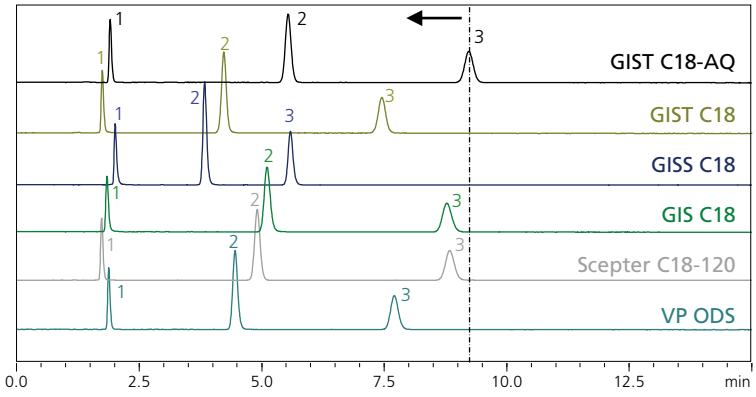


Fig. 32 Comparison of the Retainability of Hydrophilic Compounds by C18 Columns
(The analysis conditions are as shown in Table 4, with Mobile phase (2) used and samples: 1. Uracil, 2. Caffeine, and 3. Phenol.)

Shim-pack GIST series

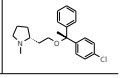
Use this QR code for details.

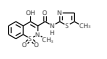
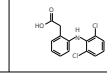
Fig. 33 Shim-pack GIST series

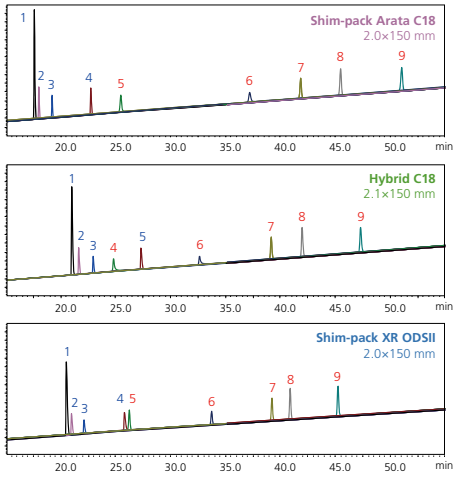
4-3. Shim-pack Arata series

Suitable separation is sometimes unattainable even with columns demonstrating improved peak shapes for basic compounds, due to leading of highly polar basic compounds and peak shape deterioration for acidic compounds. The Shim-pack Arata C18 is an effective solution to such issues. (See Fig. 34)

Type of analyte

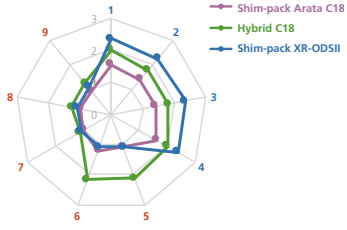
	Basic pharmaceuticals			
No.	1	2	3	4
Compound name	Cyproheptadine	Hydroxyzine	Mequitazine	Clemastine
Structural formula				

	Acidic pharmaceuticals				
No.	5	6	7	8	9
Compound name	Piroxicam	Meloxicam	Flurbiprofen	Diclofenac	Mefenamic acid
Structural formula					



Analysis conditions

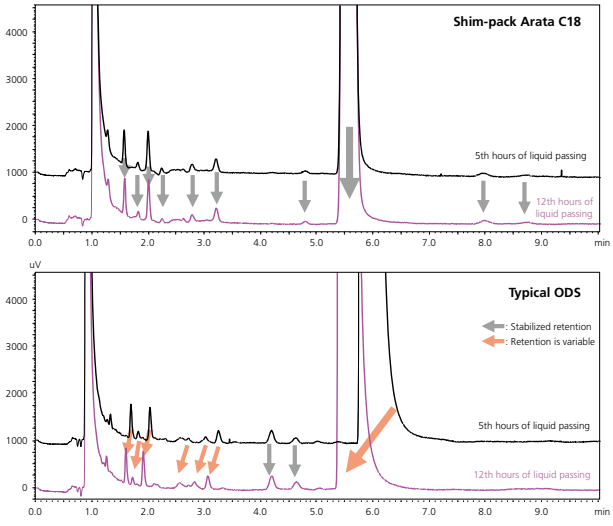
LC system : Nexera™ X2_SPD20A (Semi-micro Cell)
LC Column :
Shim-pack Arata C18 (2.0 × 150 mm, 2.2 µm)
Hybrid C18 (2.1 × 150 mm, 2.5 µm)
Shim-pack™ XR ODSII (2.0 × 150mm, 2.2 µm)
Mobile phase : 0.1%HCOOH in H_2O
Mobile phase B : 0.1%HCOOH in CH_3CN
Gradient : 10%B (0 min) →70%B (50-60 min)
→10%B (60.01-70 min.)
Flow rate : 0.2 mL/min.
Detection : 226 nm
Column temperature : 40 °C
Injection volume : 1 µL



Symmetry coefficient for pharmaceuticals

Fig. 34 Comparison of Columns when Analyzing Basic and Acidic Pharmaceutical Mixtures

With general C18 columns, column equilibration can take longer under weak acidic mobile phase conditions. Fig. 35 shows a comparison of the analysis of basic compounds using a general C18 column and the Shim-pack Arata C18 column. When comparing the respective peak retention times at the fifth and twelfth hours of liquid passing, it is evident that with general C18 column there are fluctuations in the retention times for the major components and many impurities. In contrast, fluctuations in the retention times for the major components and their impurities are not evident with the Shim-pack Arata C18. If column equilibration takes a long time, this not only increases the analysis time and the consumption of mobile phase, but also reduces the reliability of the analysis results. With the Shim-pack Arata C18 however, quick column equilibration increases the analysis efficiency and at the same time contributes to increasing the reliability of the analysis results.



Analysis conditions

LC system : NexeraX2MP_M30A (STD Cell)
Column : Shim-pack Arata C18 (3.0 × 75 mm, 2.2 µm)
Typical ODS column (3.0 × 75 mm, sub 2 µm)
Mobile phase : 0.1% H_3PO_4 in H_2O / CH_3CN
= 76 / 24 (Shim-pack Arata C18)
= 70 / 30 (Typical ODS)
Flow rate : 0.4 mL/min
Detection : 210 nm
Temperature : 40 °C
Injection volume : 1 µL
Sample : Amitriptyline

Fig. 35 Retention of Basic Pharmaceuticals and Impurities in 0.1 % Phosphoric Acid Mobile Phase Conditions

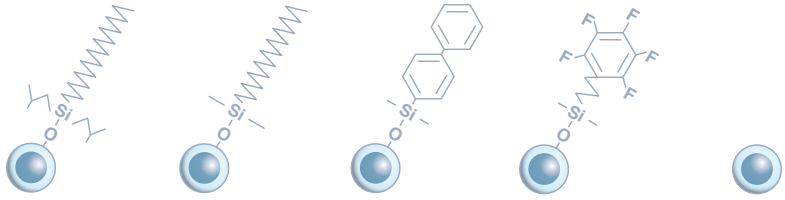
Shim-pack Arata series

Use this QR code for details.

Fig. 36 Shim-pack Arata Series

4-4. Shim-pack Velox Series

The Shim-pack Velox column is designed to optimize LC separation performance by using core shell technology to improve separation, and shorten the analysis time at pressures suited to the LC instruments used. Columns in the Shim-pack Velox series have a high theoretical plate number per unit pressure, and are designed to optimize LC separation performance. They can also be used for analysis transfer with the aim of improving throughput while maintaining the separation with existing analysis methods. With extensive column chemistries (four reversed phase and one HILIC) differing in separation selectivity, as well as a wide range of particle and column sizes in the lineup, they can be used for a wide range of applications. (See Fig. 37)



		SP-C18	C18	Biphenyl	PFPP	HILIC
USP category		L1	L1	L11	L43	L3
Functional group type		Sterically protected C18	C18	Biphenyl	Pentafluorophenyl propyl	None
Particle size (µm)		1.8, 2.7, 5	1.8, 2.7, 5	1.8, 2.7, 5	1.8, 2.7, 5	2.7
Pore size		90 Å	90 Å	90 Å	90 Å	90 Å
Surface area	1.8 µm	125 m^2/g	125 m^2/g	125 m^2/g	125 m^2/g	
	2.7 µm	130 m^2/g	130 m^2/g	130 m^2/g	130 m^2/g	130 m^2/g
	5 µm	100 m^2/g	100 m^2/g	100 m^2/g	100 m^2/g	
Carbon content ratio	1.8 µm	7 %	9 %	7 %	4 %	
	2.7 µm	7 %	7 %	7 %	4 %	N/A
	5 µm	5 %	5 %	5 %	3 %	
End capping		No	YES	YES	No	N/A
pH range		1.0-8.0	2.0-8.0	1.5-8.0	2.0-8.0	2.0-8.0
Pressure resistance	1.8 µm	100 MPa*	100 MPa*	100 MPa*	100 MPa*	
	2.7 µm	60 MPa	60 MPa	60 MPa	60 MPa	60 MPa
	5 µm	40 MPa	40 MPa	40 MPa	40 MPa	

* To maximize the column operating life, 1.8 µm columns are normally used at 80 MPa or less.

Fig. 37 List of the Shim-pack Velox Series Lineup

The Shim-pack Velox series includes the Velox SP-C18 (SP: Sterically-protected), which is designed for use under low pH conditions, and the Velox C18 for standard use, so varied separation selectivity can be obtained.
The Shim-pack Velox series is a lineup having biphenyl groups. The biphenyl group demonstrates stronger π interactions than other phenyl groups, and more varied interactions in comparison to C18, so it is advantageous for the retention of dipoles, unsaturated compounds, conjugates, and other compounds that are normally weakly retained in reversed phase mode.

There are two cautions when using core shell columns.

(1) There is a significant difference in back pressure between fully porous columns and core shell columns

A feature of core shell columns in comparison to fully porous columns with the same particle size is that they have a higher theoretical plate number per unit pressure, so it is easier to attain a given theoretical plate number while keeping the pressure low. The column back pressure itself does not change if you change from a fully porous column to a core shell column without changing the analysis conditions, particle size, or column size.

(2) They tend to have a larger impact than fully porous columns on the volume and sampling rate of HPLC instruments

With general core shell columns, there is less diffusion within the column than for fully porous columns with the same particle size, and the compound travel speed is faster. This means that a higher theoretical plate number can be obtained. At the same time, since the diffusion within the column is less for core shell columns in comparison to fully porous columns, this tends to have an impact on diffusion outside the column. As a result, the HPLC instrument's system volume (including piping volume and cell volume) and sampling rate must be adjusted. If the expected theoretical plate number is not obtained even when a core shell column is used, check the LC system configuration.

Use this QR code for details.

Fig. 38 Shim-pack Velox Series

5. Conclusion

This article has focused on columns in thphase lineup with C4, C8, or C18 carbon chain packing material modifications. If you run into difficulties in making a selection from the Shim-pack reversed phase columns, refer to the selection chart for Shim-pack reversed phase columns shown in Fig. 39.

In terms of actual analysis, column selection is not always a problem, particularly when the column for use is specified. When unspecified, column selection in analysis method development is both important and difficult.

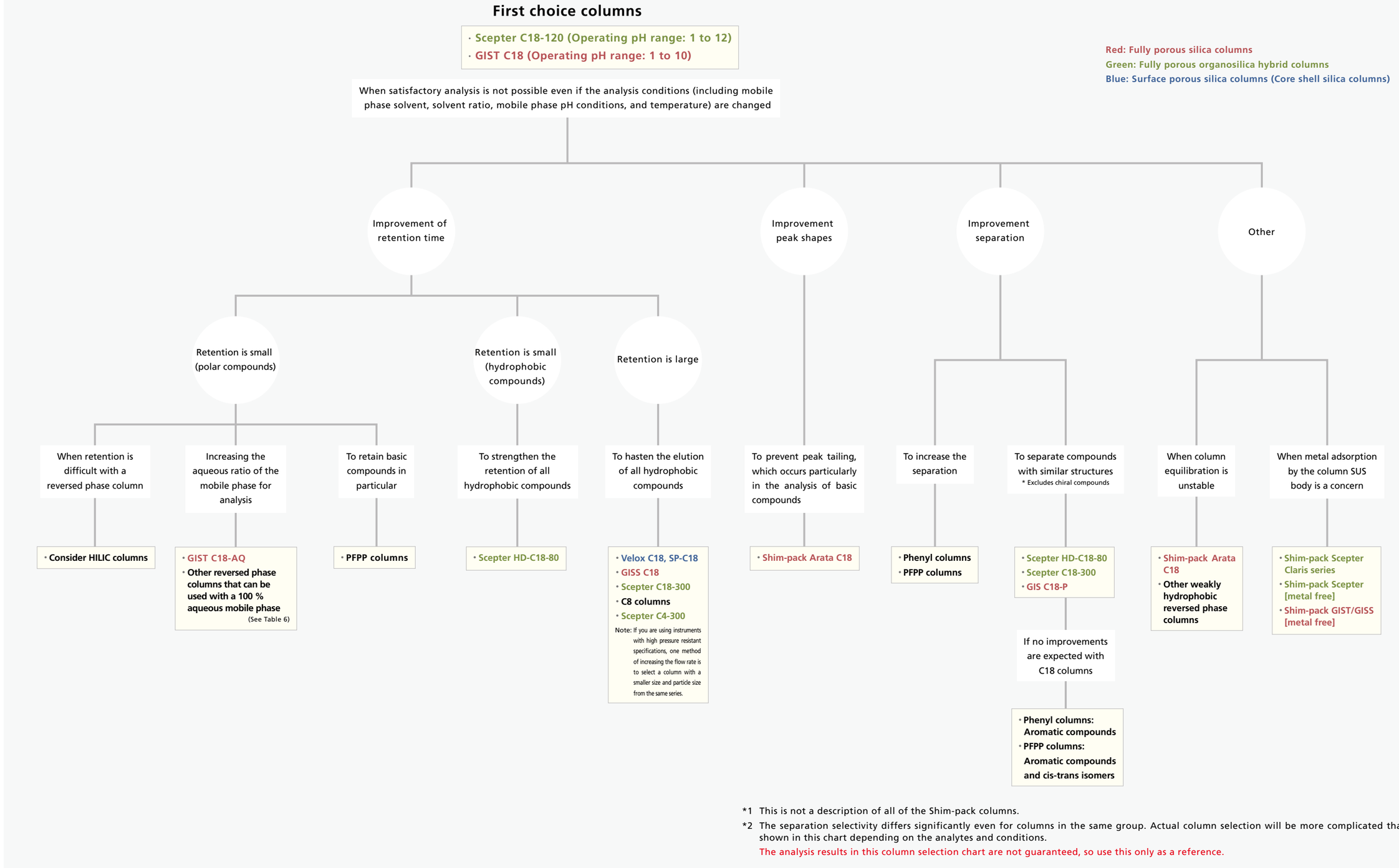
Even the same C18 column can be designed with different concepts in mind, so the separation selectivity obtainable will vary significantly if the column is changed. For this reason, in analysis method development, the screening of multiple columns is recommended, with consideration of suitable columns and analysis conditions. In such cases, the Shim-pack reversed phase column lineup provides a variety of separation selectivities, so it is important to consider its use during method development.

For screening and optimization of separation conditions including columns, consider LabSolutions™ MD and Method Scouting Solution, software provided by Shimadzu to support method development.

References

[1] K. Kimata, K. Iwaguchi, S. Onishi, K. Jinno, R. Eksteen, K. Hosoya, M Araki And N.Tanaka, ; J. Chromatogr. Sci., Vol. 27 (1989)

[2] Yoshihisa Sudo, Hiroshi Sakamaki; CHROMATOGRAPHY, Vol. 32 (2011)



*1 This is not a description of all of the Shim-pack columns.

*2 The separation selectivity differs significantly even for columns in the same group. Actual column selection will be more complicated than shown in this chart depending on the analytes and conditions.
The analysis results in this column selection chart are not guaranteed, so use this only as a reference.

Fig. 39 Chart for the Selection of Recommended General-Purpose Reversed Phase Columns from the Shim-pack Column Lineup

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