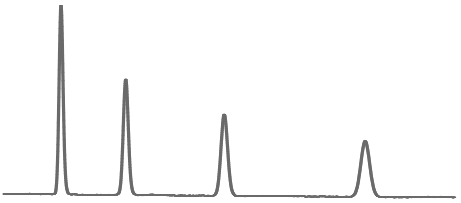
GL Sciences



Inertsil Technical Library

Inertsil® ODS-4, C8-4
Comparison of Performance

- List of Compared Columns
- Experimental Explanation, Analytical Conditions
- Comparison between commercially available ODS Columns
- Comparison between commercially available C8 Columns
- Comparison of Separation Pattern between ODS and C8 Columns
- Comparison of Retentivity
- Comprehensive Evaluation

GL Sciences

Introduction of Inertsil 4 Series

About Inertsil

Inertsil, the name of our brand comes from the key word " INERT", which is the most important factor in the separation analysis. GL Sciences' Inertsil columns delivers the maximum performance of an HPLC column and we would like to support you and your critical analysis.

Inertsil 4 series columns are the most recent addition to the Inertsil line and the most inert columns available.

Inertsil ODS-4 - A Must-Have column in your laboratory

Reversed phase ODS and C8 columns comprise the vast majority of columns used in the HPLC separation industry. In this publication, we selected a wide variety of popular brands and compared them with GL Sciences' Inertsil ODS-4. As you review this data we believe that you will conclude that Inertsil ODS-4 is a must-have column in your laboratory.

Comparison Table

ODS（C18）Columns

Column	Serial No.	Specific Surface Area (m^2/g)	Pore Diameter (Å)	Pore Volume (mL/g)	Carbon Content (%)
Inertsil ODS-4	**8JF37043**	**450**	**100**	**1.05**	**11**
Atlantis T3	011138164102 08	330	100	1.0	14
SunFire C18	011235104117 14	340	100	0.88	16
XBridge	012739246136 08	185	135	0.74	18
CAPCELLPAK MG Ⅱ	A4AD 01490	260	100	0.95	15
CAPCELLPAK MG Ⅲ	A15AD 01145	260	100	0.91	15
Zorbax Eclipse Plus C18	USUXA01612	160	95	-	8
YMC-Pack Pro C18	0425016137(W)	340	120	1.05	16
Luna 5u C18(2)	442336-10	400	100	-	17.5
Cadenza 5CD-C18	HJ17M3I	-	120	-	-
TSKgel ODS-100V	N0098	450	100	1.1	15
Mightysil RP-18GP	8027128	325～380	110～140	1.0～1.2	19.5～21.5
Hypersil GOLD	1084135E	220	175	-	10
Shim-pack VP-ODS	7052777	410	120	1.2	20

※ Column Size: 5um, 250×4.6 mm I.D.

C8 Columns

Column	Serial No.	Specific Surface Area (m^2/g)	Pore Diameter (Å)	Pore Volume (mL/g)	Carbon Content (%)
Inertsil C8-4	**9JF39013**	**450**	**100**	**1.05**	**5**
YMC-Pack Pro C8	011039142121 03	340	100	0.86	11.5
Zorbax Eclipse Plus C8	010739093130 03	185	135	0.77	13
Luna C8(2)	USUTA01326	160	95	-	8
SunFire C8	0425020742(W)	325	120	0.97	10
XBridge C8	462435-6	400	100	-	13.5

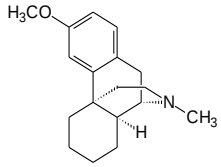
※ Column Size: 5um, 250×4.6 mm I.D.

Explanation of Analytical Tests and Conditions

By conducting the 4 tests below, we can evaluate the degree of inertness of the column and whether it can be used in a 100% aqueous mobile phase condition. To strictly evaluate all columns, the below mentioned samples were used, which are all known as strong adsorptive compounds.

Basic Compound Test

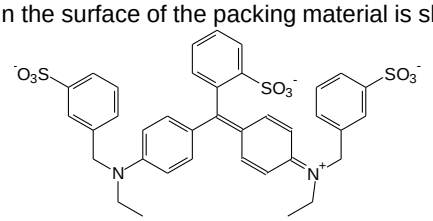
Dextromethorphan is a strong basic compound. Severe tailing can be confirmed when the packing material contains residual silanol groups.



Mobile Phase: A) CH_3CN
B) 25 mM Phosphate buffer (pH 7.0)
A/B = 40/60
Flow Rate: 1.0 mL/min
Col. Temp.: 40 ℃
Detection: UV 220 nm
Injection Vol.: 1 ul
1. Phenol (0.015 mg/mL)
2. Dextromethorphan HBr (0.1 mg/mL)

Acidic Compound Test

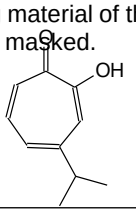
Sharp peaks can be obtained when analyzing Phenol or Salicylic Acid. However, as Brilliant Blue FCF has three sulfonic groups in its chemical structure, tailing will occur when the surface of the packing material is slightly basic.



Mobile Phase: A) CH_3CN
B) 0.1 % H_3PO_4
A/B = 25/75
Flow Rate: 1.0 mL/min
Col. Temp.: 40 ℃
Detection: UV 254 nm
Injection Vol.: 3 ul
1. Brilliant Blue FCF (0.05 mg/mL)
2. Phenol (0.3 mg/mL)
3. Salicylic acid (0.2 mg/mL)

Chelating Compound Test

Hinokitiol is a strong chelating compound, which coordinately binds with the surface of residual trace metal impurities, resulting in severe tailing.
However, the peak shape improves as the injection increases since the surface of the packing material of the adsorption active sites eventually become masked.



Mobile Phase: A) CH_3CN
B) 0.1 % H_3PO_4
A/B = 40/60
Flow Rate: 1.0 mL/min
Col. Temp.: 40 ℃
Detection: UV 254 nm
Injection Vol.: 1 ul
1. Hinokitiol (0.1 mg/mL)

Dewetting Test

When analyzing hydrophilic compounds under water rich mobile phase condition, once the pump is stopped, the hydrophobic bonded group pushes the aqueous mobile phase out off the pore in an irreversible fashion, in what has become known as the dewetting phenomenon. Inertsil ODS-4 demonstrates excellent resistance to dewetting.

Testing Procedure:

1) 100% water is introduced into column over 20 minutes.
2) Conduct Analysis (Upper chromatogram in the following pages)
3) Stop flow for 15 minutes.
4) 100% water is introduced again into column over 30 minutes.
5) Stop flow for 15 minutes again.
6) Conduct Analysis (Lower chromatogram in the following pages)

Mobile Phase: 100 % H_2O
Flow Rate: 1.0 mL/min
Col. Temp. : 40 ℃
Detection: UV 254 nm
Injection Vol.: 2 ul
1. Cytosine (0.01 mg/mL)
2. Uracil (0.01 mg/mL)
3. Guanine (0.01 mg/mL)
4. Thymine (0.01 mg/mL)
5. Adenine (0.002 mg/mL)

References

Test compounds for detecting the silanol effect on the elution of ionized amines in reversed-phase LC

Nobuo Tanaka, Kensuke Okusa, Yuki Suita, Yukio Otsuka, Mineo Tahara, Tohru Ikegami, Masayoshi Ohira, Masakazu Takahashi
Journal of Separation Science, Volume 33 Issue 3, Pages 348 - 358

The effectiveness of several basic compounds for testing silica-based stationary phases was reviewed by applying them to recent columns for reversed-phase HPLC. Most octadecylsilylated (C18) stationary phases, prepared as a base-deactivated material from high-purity silica gel with endcapping, provided excellent peak shape and column efficiency for the bases including benzylamine and amitriptyline that once caused problems and were subsequently employed for testing silanol activities. However, a cyclic tertiary amine, dextrometorphan, was eluted as an acceptable peak from only a few columns at neutral pH. Such a more sensitive probe is expected to contribute to further improvement of the stationary phase for reversed-phase HPLC.

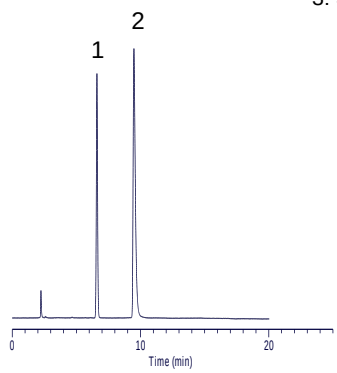
ODS Columns Comparison (1)

Basic Compound Test	Acidic Compound Test	Chelating Compound Test	Dewetting Test

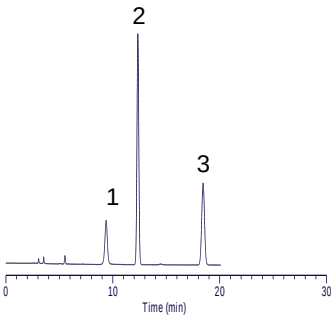
Inertsil ODS-4

Basic Compound : ◎ Acidic Compound : ◎ Chelating Compound : ○ Dewetting : ◎

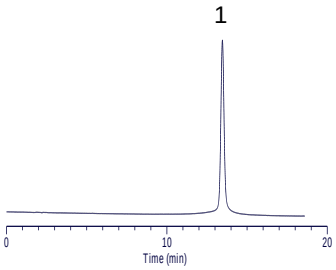
1: Phenol
2: Dextromethorphan

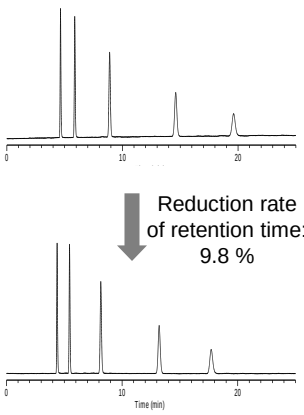


1: Brilliant Blue FCF
2: Phenol
3: Salicylic acid



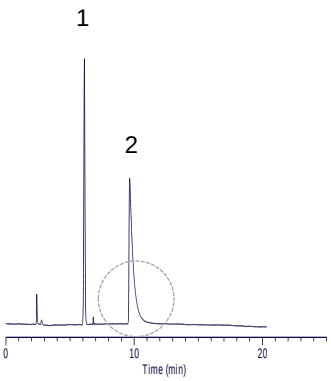
1. Hinokitiol

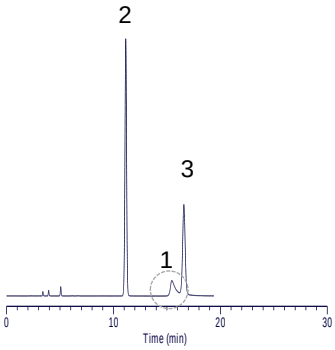


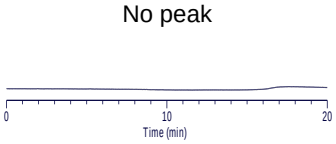


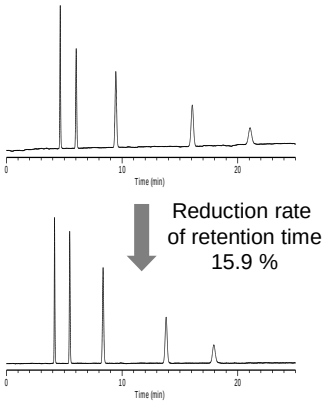
Atlantis T3

Basic compound : ○ Acidic compound : ○ Chelating compound : × Dewetting : ○



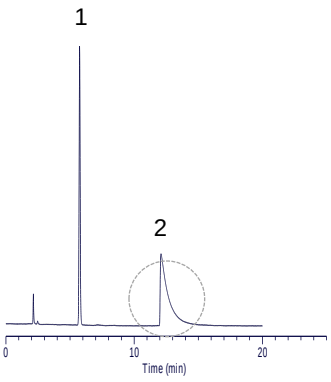


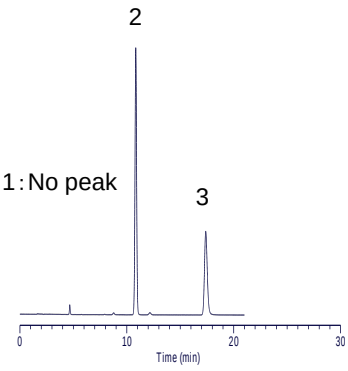


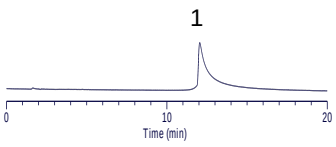


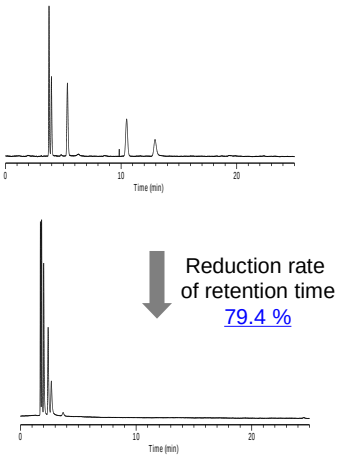
SunFire C18

Basic compound : △ Acidic compound : × Chelating compound : △ Dewetting : ×







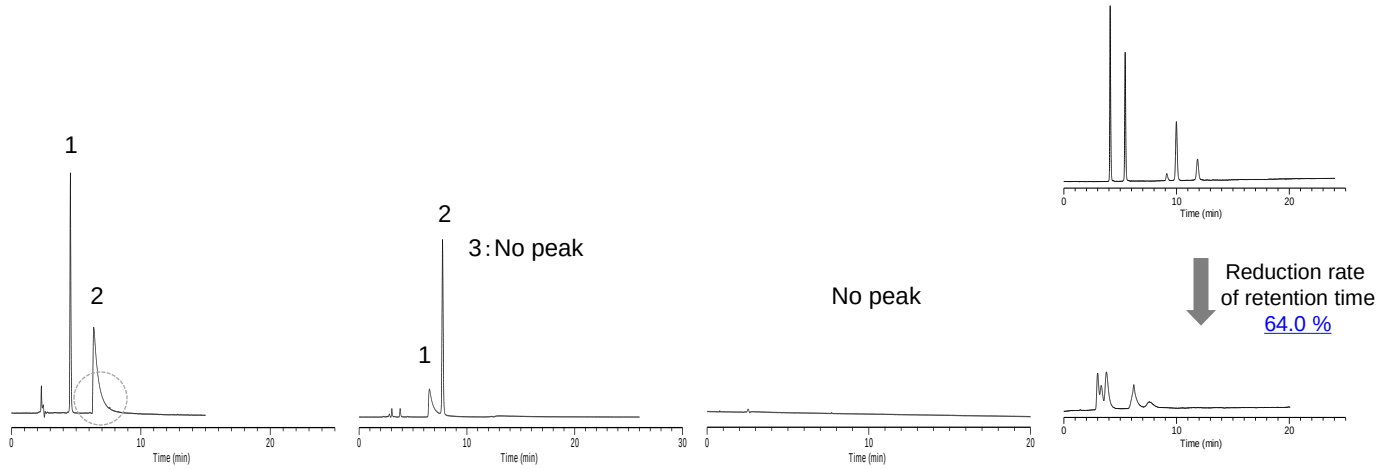


Inertsil ODS-4	Proved superior peak shapes for all various compounds along with extreme stability under 100% aqueous mobile phases. Inertsil ODS-4 is just compatible with any type of compounds in any mobile phase conditions delivering highly stable chromatograms for qualitative and quantitative analysis.
Atlantis T3	Compatible for the analysis of hydrophilic compounds, but not for the analysis of chelating compounds.
SunFire C18	Overall, it is not an easy-to-use column as adsorption of peaks can be concerned especially when conducting a trace analysis.

Basic Compound Test	Acidic Compound Test	Chelating Compound Test	Dewetting Test

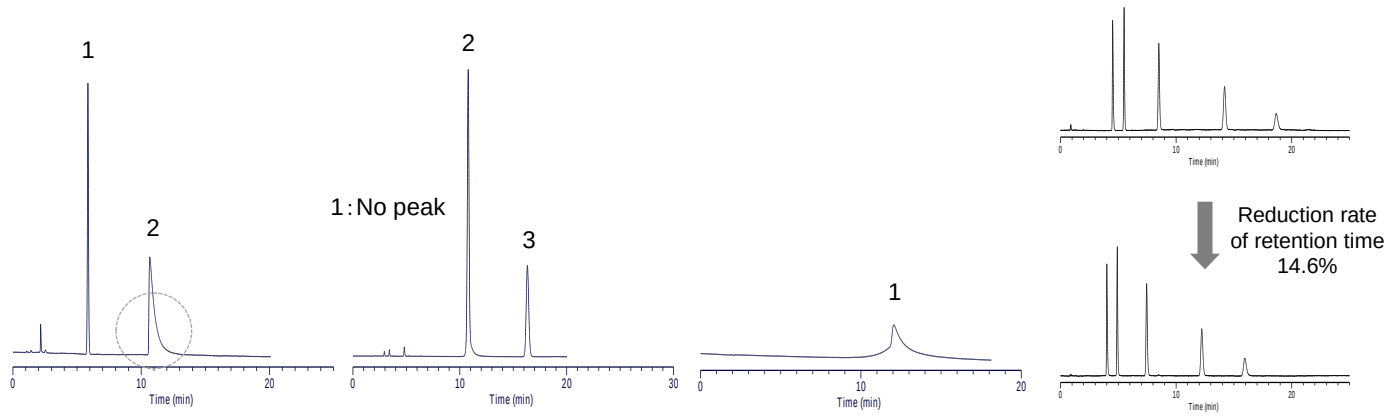
XBridge

Basic compound: △ Acidic compound: △ Chelating compound: × Dewetting: ×



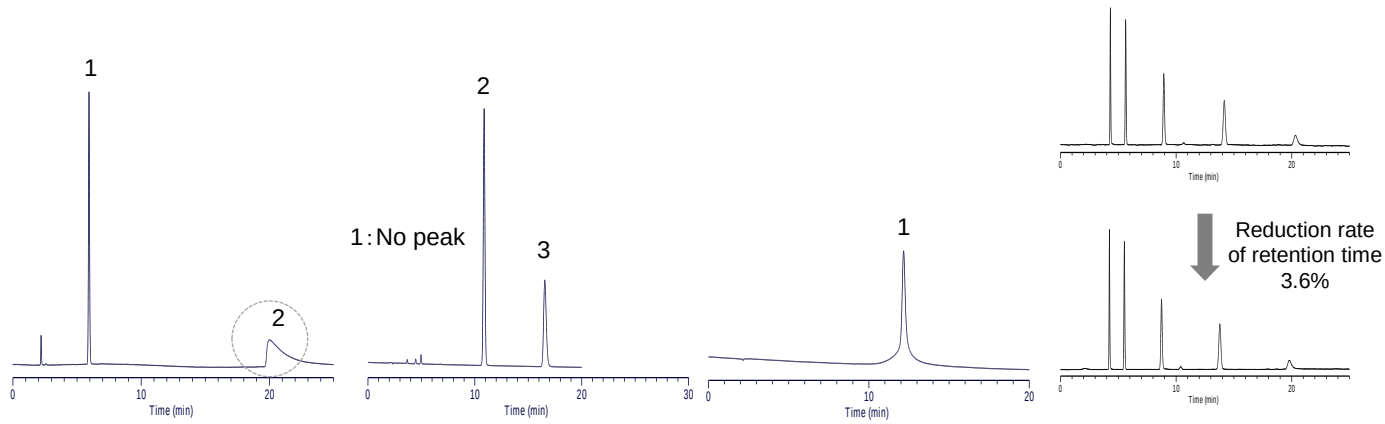
CAPCELLPAK MG II

Basic compound: △ Acidic compound: × Chelating compound: △ Dewetting: ○



CAPCELLPAK MG III

Basic compound: × Acidic compound: × Chelating compound: ○ Dewetting: ◎



XBridge	Adsorption of acidic and chelating compounds were confirmed. In addition, it is apparently not compatible with 100% aqueous mobile phase as it showed a sudden loss of retention due to a dewetting phenomenon.
CAPCELLPAK MG II	Compatible for analysis of hydrophilic compounds, but caution needs to be taken when analyzing acidic and chelating compounds.
CAPCELLPAK MG III	Can obtain high reproducibility and strong retentivity under 100% aqueous mobile phases. Precision of analysis can be concerned as adsorbed peaks were confirmed in the analysis of basic and acidic compounds.

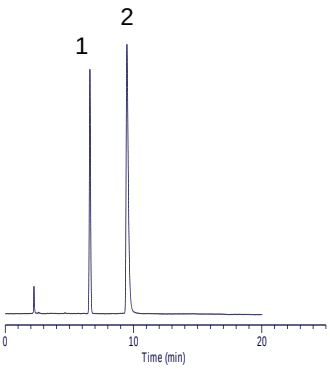
ODS Columns Comparison (2)

Basic Compound Test | **Acidic Compound Test** | **Chelating Compound Test** | **Dewetting Test**

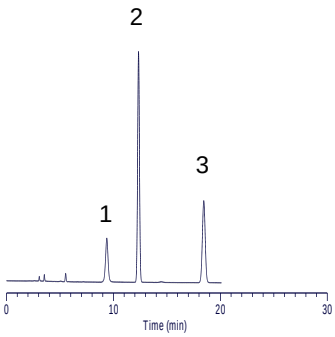
Inertsil ODS-4

Basic compound: ◎ Acidic compound: ◎ Chelating compound: ○ Dewetting: ◎

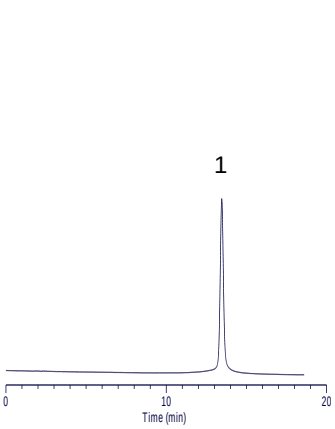
1: Phenol
2: Dextromethorphan

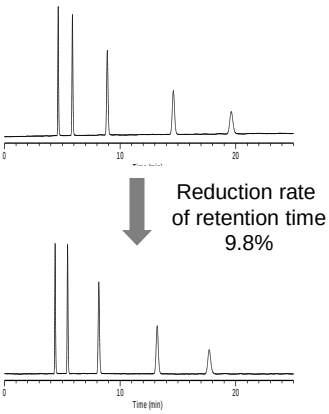


1: Brilliant Blue FCF
2: Phenol
3: Salicylic acid



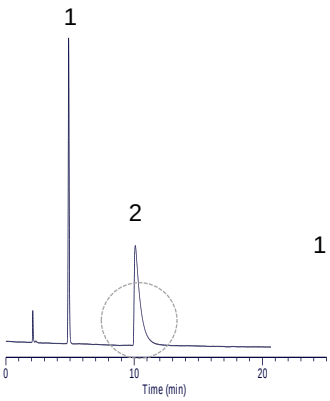
1: Hinokitiol



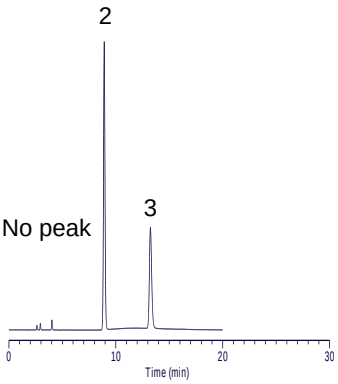


Zorbax Eclipse Plus C18

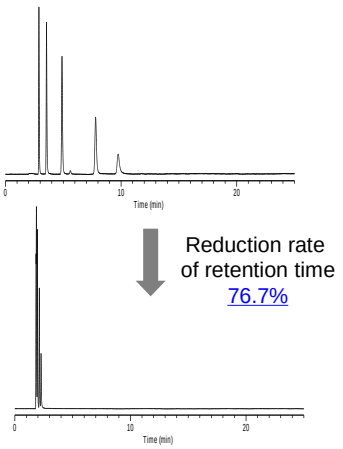
Basic compound: △ Acidic compounds: × Chelating compound: × Dewetting: ×



1: No peak

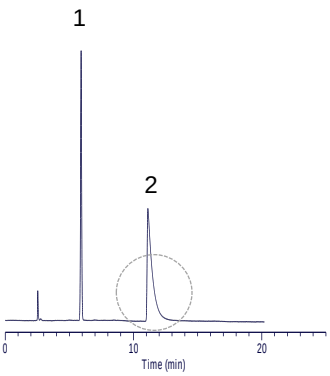


No peak

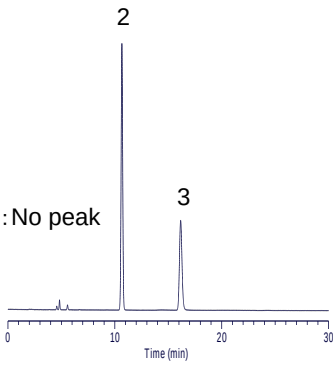


YMC-Pack Pro C18

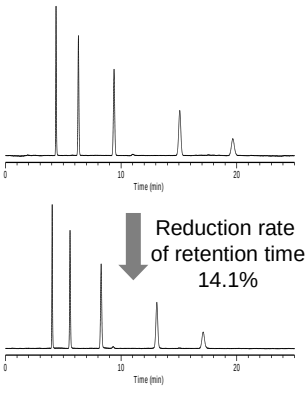
Basic compound: △ Acidic compound: × Chelating compound: × Dewetting: ○



1: No peak



No peak

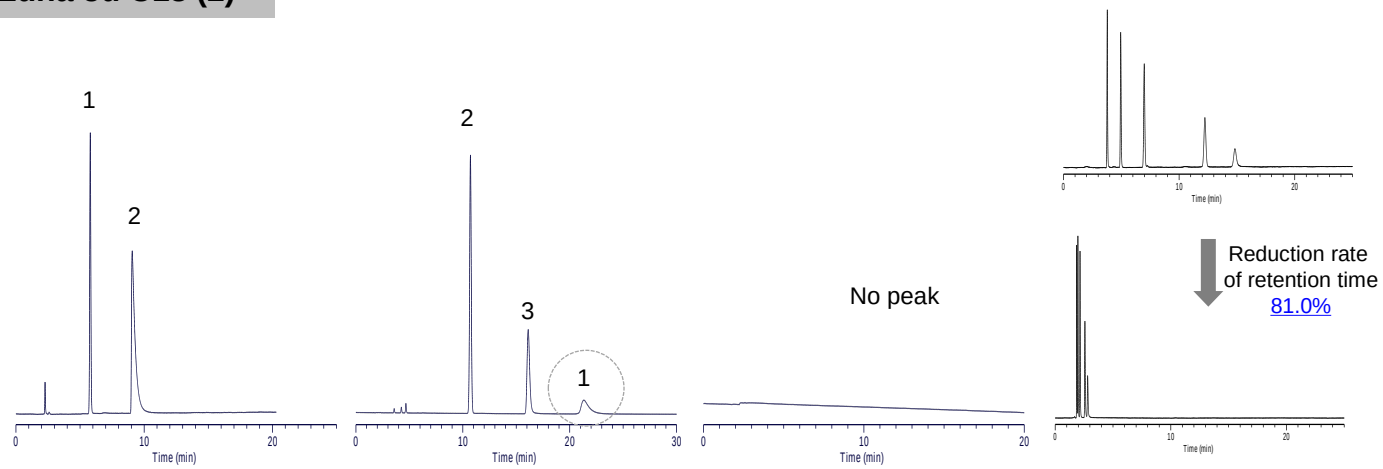


Inertsil ODS-4	Proved superior peak shapes for all various compounds along with extreme stability under 100% aqueous mobile phases. Inertsil ODS-4 is just compatible with any type of compounds in any mobile phase conditions delivering highly stable chromatograms for qualitative and quantitative analysis.
Zorbax Eclipse Plus C18	Overall, indications of severe chromatographic issues seem apparent.
YMC-Pack Pro C18	Retentivity is relatively high and excellent peak shapes were obtained for hydrophilic compounds. However, quantitative analysis can be a source of concern since the adsorption of acidic and chelating compounds were confirmed.

Basic Compound Test	Acidic Compound Test	Chelating Compound Test	Dewetting Test

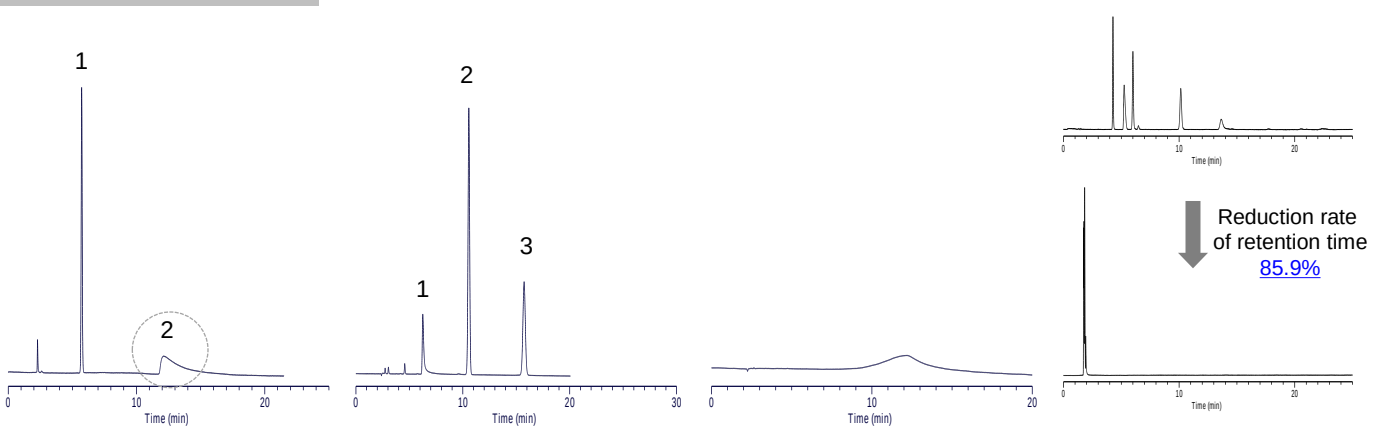
Luna 5u C18 (2)

Basic compound: ○ Acidic compound: △ Chelating compound: × Dewetting: ×



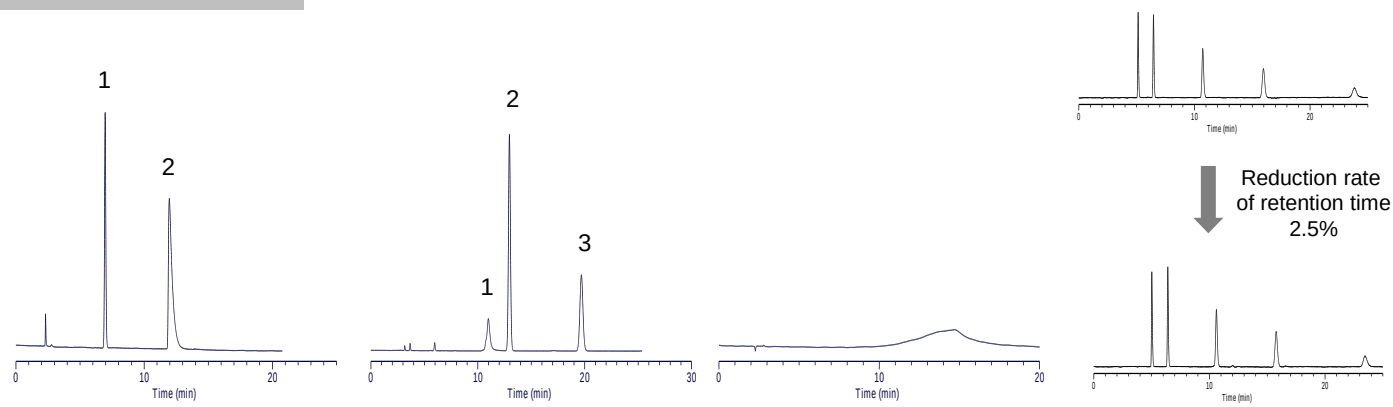
Cadenza 5CD-C18

Basic compound: × Acidic compound: ○ Chelating compound: × Dewetting: ×



TSKgel ODS-100V

Basic compound: ○ Acidic compound: ◎ Chelating compound: × Dewetting: ◎



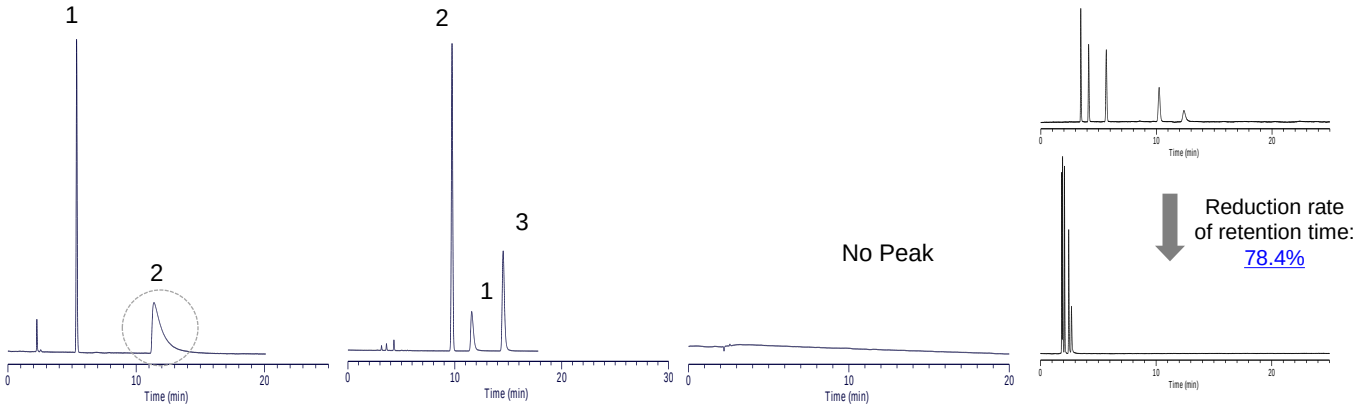
Luna 5u C18 (2)	Compatible for the analysis of basic compounds. On the other hand, adsorption of chelating compounds and unstable results seem apparent for the analysis of hydrophilic compounds under 100% aqueous mobile phases.
Cadenza 5CD-C18	Compatible for the analysis of acidic compounds, but not for basic, acidic and hydrophilic compounds under 100% aqueous mobile phases.
TSKgel ODS-100V	Compatible for the analysis of acidic and hydrophilic compounds under 100% aqueous mobile phases. Caution needs to be taken when analyzing chelating compounds.

ODS Columns Comparison (3)

Basic Compound Test	Acidic Compound Test	Chelating Compound Test	Dewetting Test

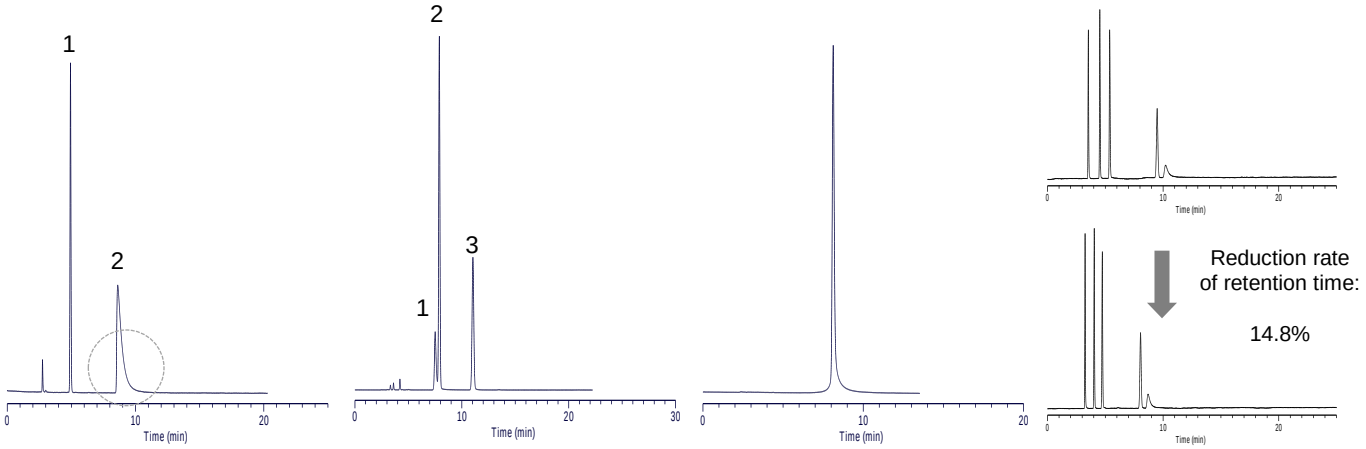
Mightysil RP-18GP

Basic compound: △ Acidic compound: ◎ Chelating compound: × Dewetting: ×



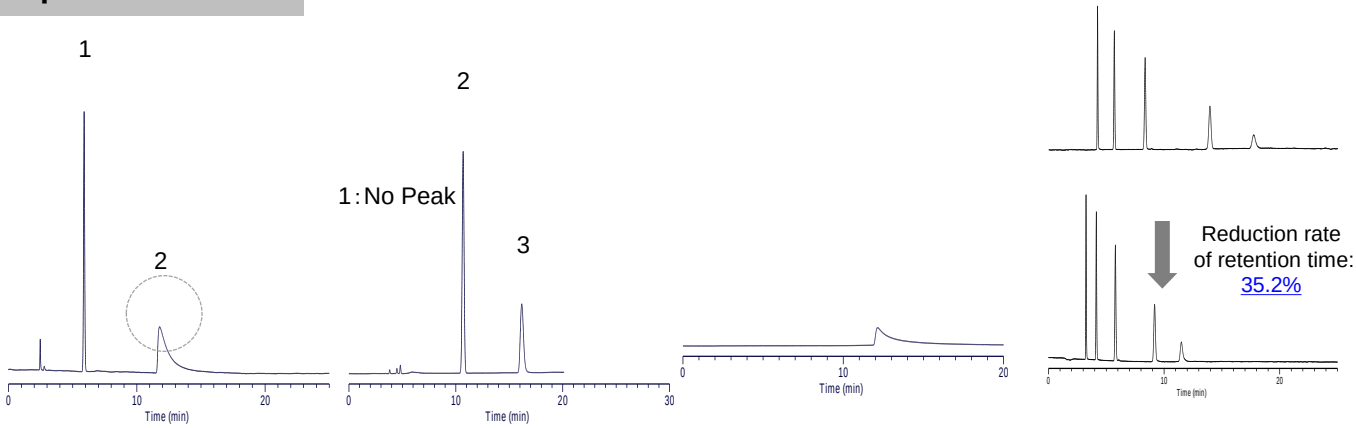
Hypersil GOLD

Basic compound: △ Acidic compound: ◎ Chelating compound: ○ Dewetting: ○



Shim-pack VP-ODS

Basic compound: △ Acidic compound: × Chelating compound: × Dewetting: △



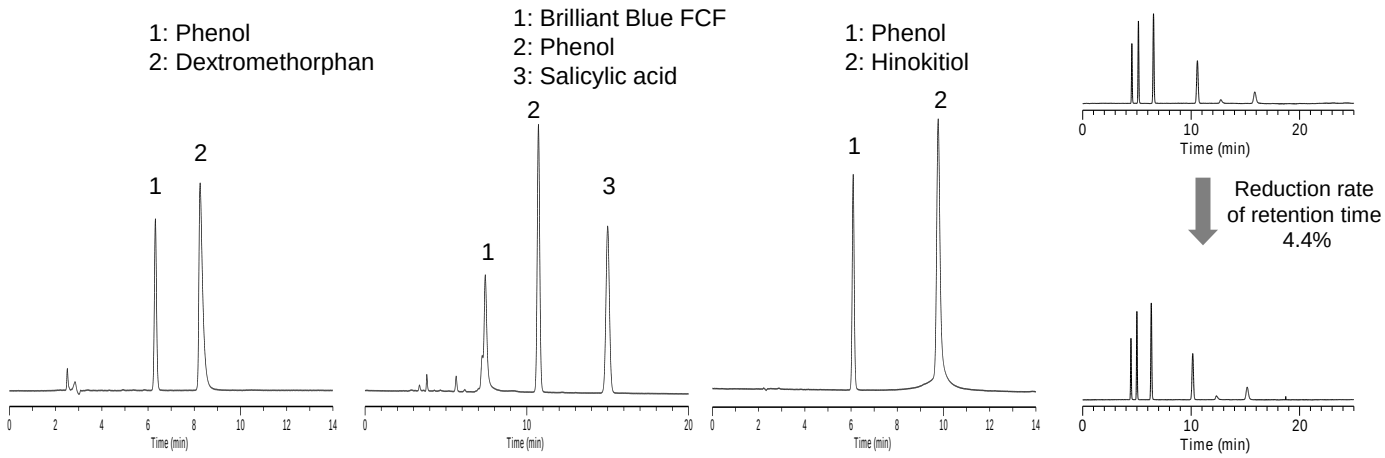
Mightysil RP-18GP	Compatible for the analysis of acidic compounds, but not for basic, chelating and hydrophilic compounds under 100% aqueous mobile phases.
Hypersil GOLD	Compatible for the analysis of chelating compounds. As the retentivity of hydrophilic compounds were weak, separation of multi-compounds may be difficult to achieve.
Shim-pack VP-ODS	Overall, severe chromatographic issues seem apparent.

Comparison of C8 Columns

Basic Compound Test | **Acidic Compound Test** | **Chelating Compound Test** | **Dewetting Test**

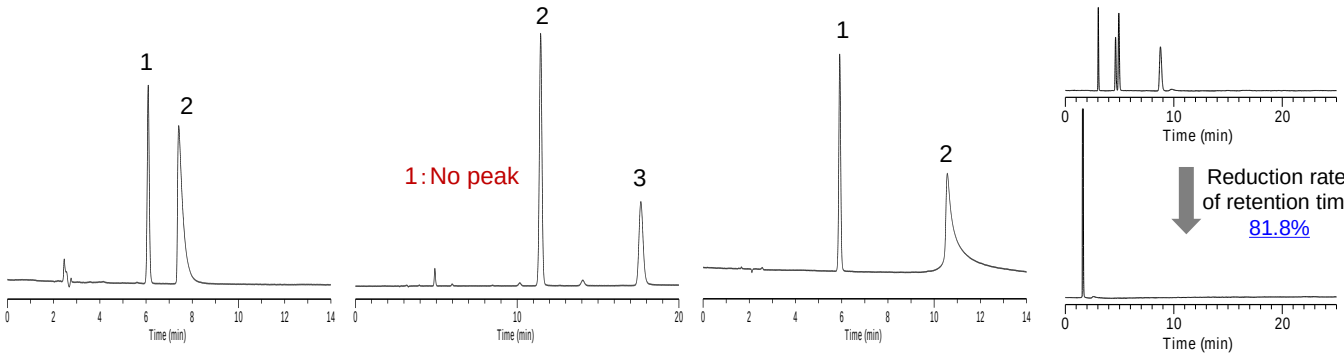
Inertsil C8-4

Basic compound: ◎ Acidic compound: ◎ Chelating compound: ○ Dewetting: ◎



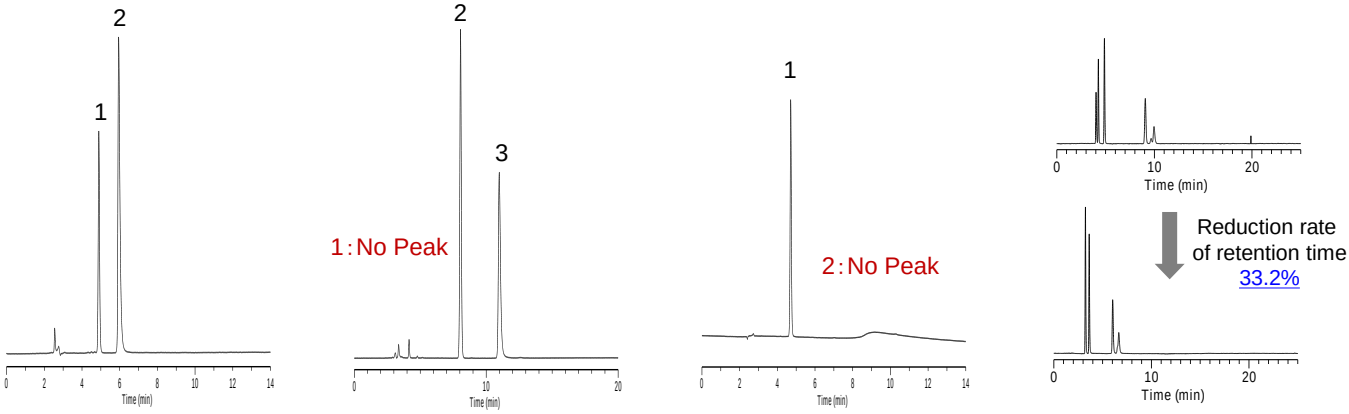
SunFire C8

Basic compound: ○ Acidic compound: × Chelating compound: △ Dewetting: ×



Xbridge C8

Basic compound: ◎ Acidic compound: × Chelating compound: × Dewetting: △

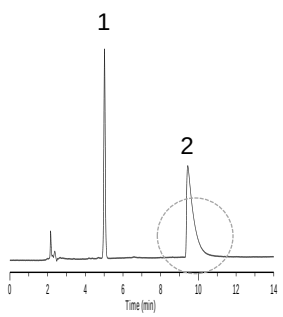


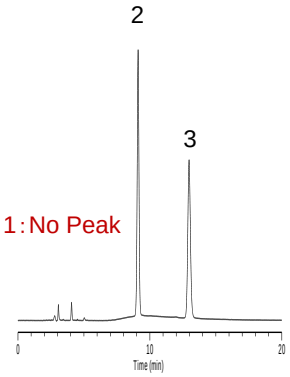
Inertsil C8-4	Delivers the same separation pattern and extreme inertness to any type of compounds just like Inertsil ODS-4 along with extreme stability under 100% aqueous mobile phases for qualitative and quantitative analysis.
SunFire C8	Compatible for the analysis of basic compounds, but adsorption of acidic compounds and unstable results were obtained under 100% aqueous mobile phases.
XBridge C8	Compatible for the analysis of basic compounds. As the retentivity is relatively weak when comparing to other commercially available C8 columns, separation of multi-compounds may be difficult to achieve.

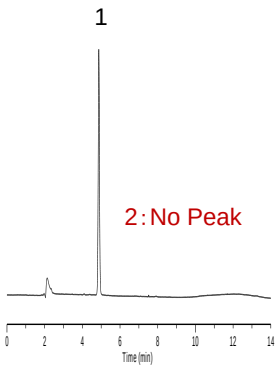
Basic Compound Test	Acidic Compound Test	Chelating Compound Test	Dewetting Test

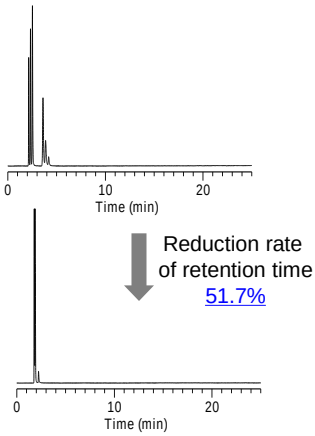
Zorbax Eclipse Plus C8

Basic compound: △ Acidic compound: × Chelating compound × Dewetting: ×



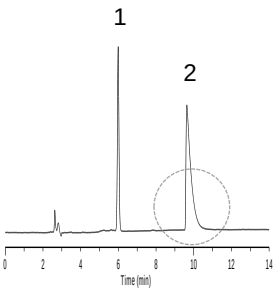


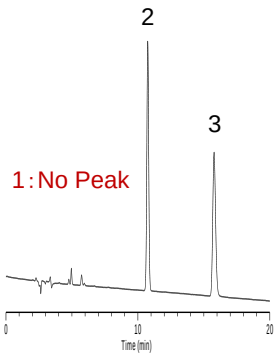


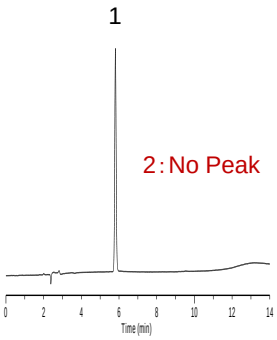


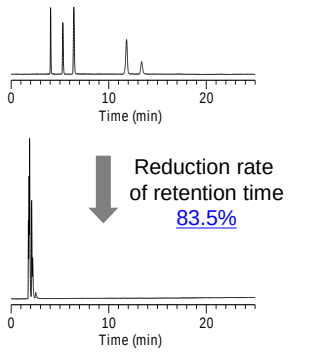
YMC-Pack Pro C8

Basic compound: ○ Acidic compound: × Chelating compound: × Dewetting: ×



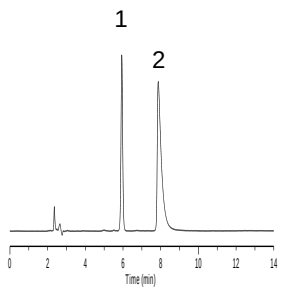


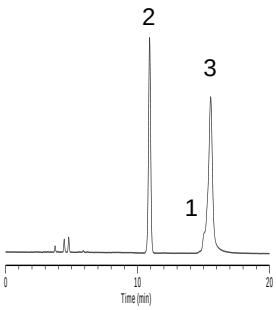


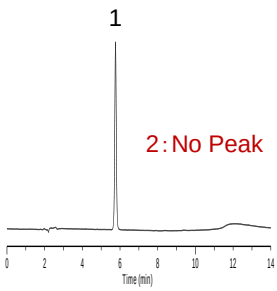


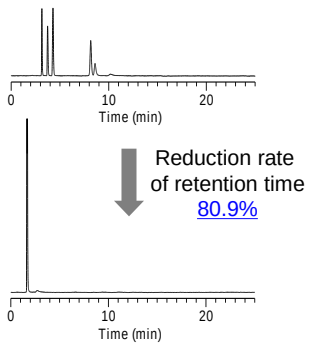
Luna C8 (2)

Basic compound: ○ Acidic compound: △ Chelating compound: × Dewetting: ×









Zorbax Eclipse Plus C8	Adsorption of peaks were confirmed and was not compatible under 100% aqueous mobile phases as the retention was lost.
YMC-Pack Pro C8	Overall, it is not an easy-to-use column.
Luna C8(2)	Apparently, shows limited compatibility with acidic and chelating compounds and a tendency towards severe dewettting under highly aqueous conditions.

Comparison of Separation Pattern between ODS and C8 columns

Shortening the analysis time without changing the separation pattern

Many chromatographers prefer a C8 column when and ODS phase shows excessive retention values.
Inertsil C8-4 provides the same separation pattern (selectivity) and extreme inertness to any type of compounds just like Inertsil ODS-4, which enables easy method transfer from ODS-4 to C8-4.

Other commercially available ODS and C8 columns can show dramatically different selectivity even though they are part of the same brand/series. For example, the elution position of Sample No. 7, Triphenylene wasdifferent between commercially available ODS and C8 columns from the same brand/series. This indicates that the stereoselectivty between those ODS and C8 columns are different as the bonding density of the functional group is high on their C8 column when comparing from their ODS column. Therefore, caution needs to be taken when changing the column from their ODS to C8 column.

Experimental Explanation of Selectivity Test

- Sample No.1, Uracil cannot be retained in a reversed phase mode when using the mobile phase condition described on the right. Therefore, Uracil was used to determine the t0 position.

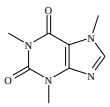
t0: Void Volume

- Sample No.2, 3, Caffeine and Phenol are used to confirm the amount of residual silanol on the surface of the silica gel. Caffeine elutes later against Phenol when there is a lot of residual silanol on the surface of the silica gel.
- Sample No.4, n-Butylbenzene and Sample No.6, n-Amylbenzene were used to determine the hydrophobic property of the column. n-Amylbenzene elutes later against n-Butylbenzene when the hydrophobicity of the column is high.
- Stereoselectivity is indicated by Sample No.5, o-Terphenyl and Sample No.7, Triphenylene. O-Terphenyl has a twisted tertiary structure and Triphenylene has a planar structure. Triphenylene elutes later against o-Terphenyl when the stereoselectivity of the column is high.

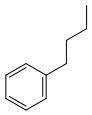
Mobile Phase: A) CH_3OH
B) H_2O
A/B = 80/20
Flow Rate: 1.0 mL/min
Col. Temp.: 40 ℃
Detection: UV 254 nm
Injection Vol.: 5 ul

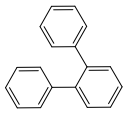
1) Uracil	(0.005 mg/mL)
2) Caffeine	(0.04 mg/mL)
3) Phenol	(0.08 mg/mL)
4) *n*-Butylbenzene	(1.12 mg/mL)
5) *o*-Terphenyl	(0.04 mg/mL)
6) *n*-Amylbenzene	(1.37 mg/mL)
7) Triphenylene	(0.014 mg/mL)

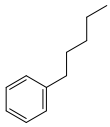

1. Uracil

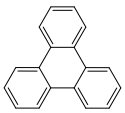

2. Caffeine


3. Phenol


4. *n*-Butylbenzene


5. o-Terphenyl


6. *n*-Amylbenzene


7. Triphenylene

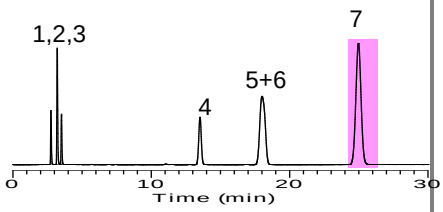
It is necessary to set the bonding density of the function group of an ODS and C8 column to obtain the same separation pattern. Theoretically, the carbon loading ratio shall be 0.44 =8/18. The carbon loading ratio of Inertsil 4 series is 0.45 and we have successfully obtained the same separation pattern between ODS-4 and C8-4. Now, not only the analysis time can be shortened, but the same separation pattern can be obtained using Inertsil 4 series.

	Carbon Loading of ODS (%)	Carbon Loading of C8 (%)	Ratio
The Length of Alkyl Chain	C18	C8	8/18 = 0.44
Inertsil 4 Series	11.00%	5.00%	5.0/11.0 = 0.45
YMC-Pack Pro	16	10	10/16 = 0.63
Zorbax Eclipse Plus	8	8	8/8 = 1.0
Luna	17.5	13.5	13.5/17.5 = 0.77
SunFire	16	11.5	11.5/16 = 0.72
XBridge	18	13	13/18 = 0.72

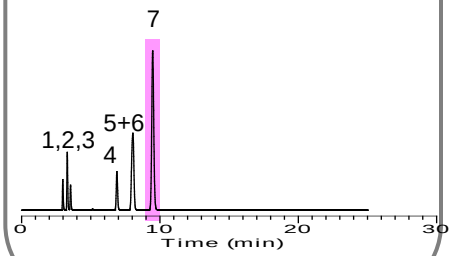
Inertsil 4 Series

ODS-4

Carbon Loading Ratio
5/11=0.45



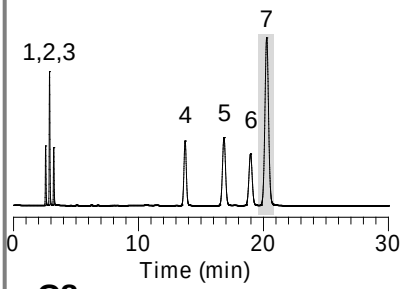
C8-4



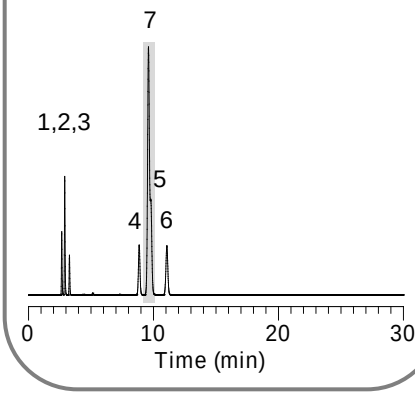
SunFire

ODS

Carbon Loading Ratio
11.5/16=0.72



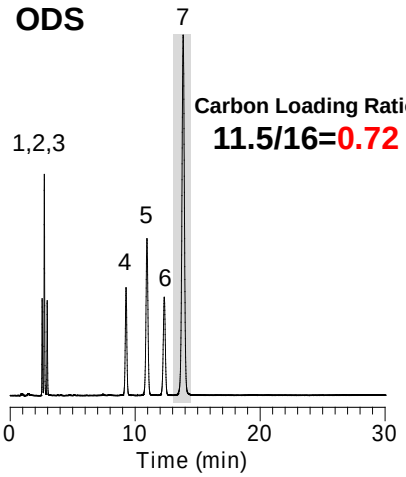
C8



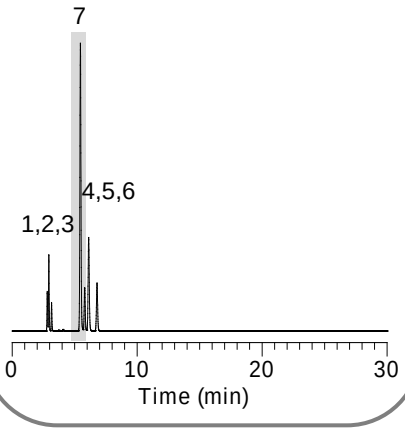
XBridge

ODS

Carbon Loading Ratio
11.5/16=0.72



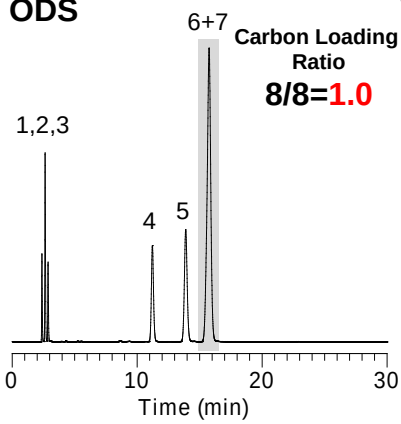
C8



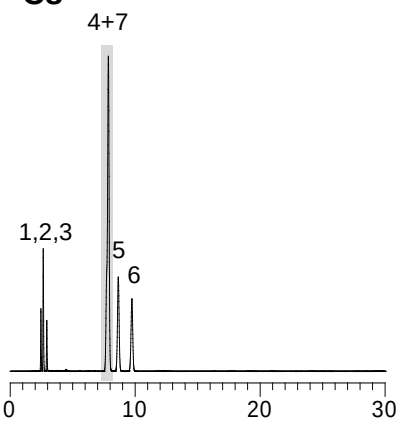
Zorbax Eclipse Plus

ODS

Carbon Loading Ratio
8/8=1.0



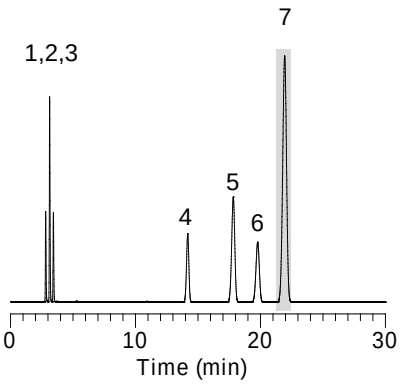
C8



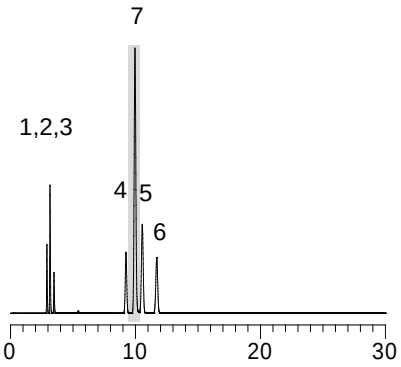
YMC-Pack Pro

Carbon Loading Ratio
10/16=0.63

ODS



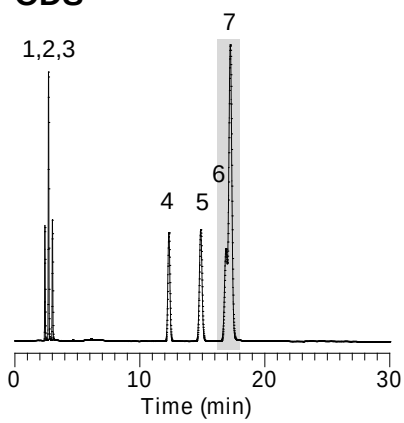
C8



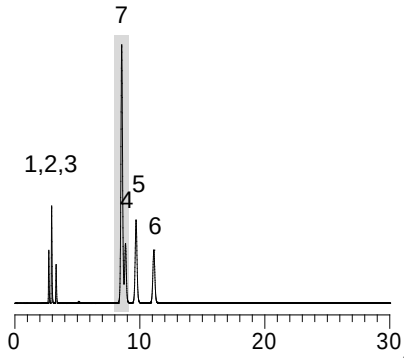
Luna (2)

Carbon Loading Ratio
13.5/17.5=0.77

ODS

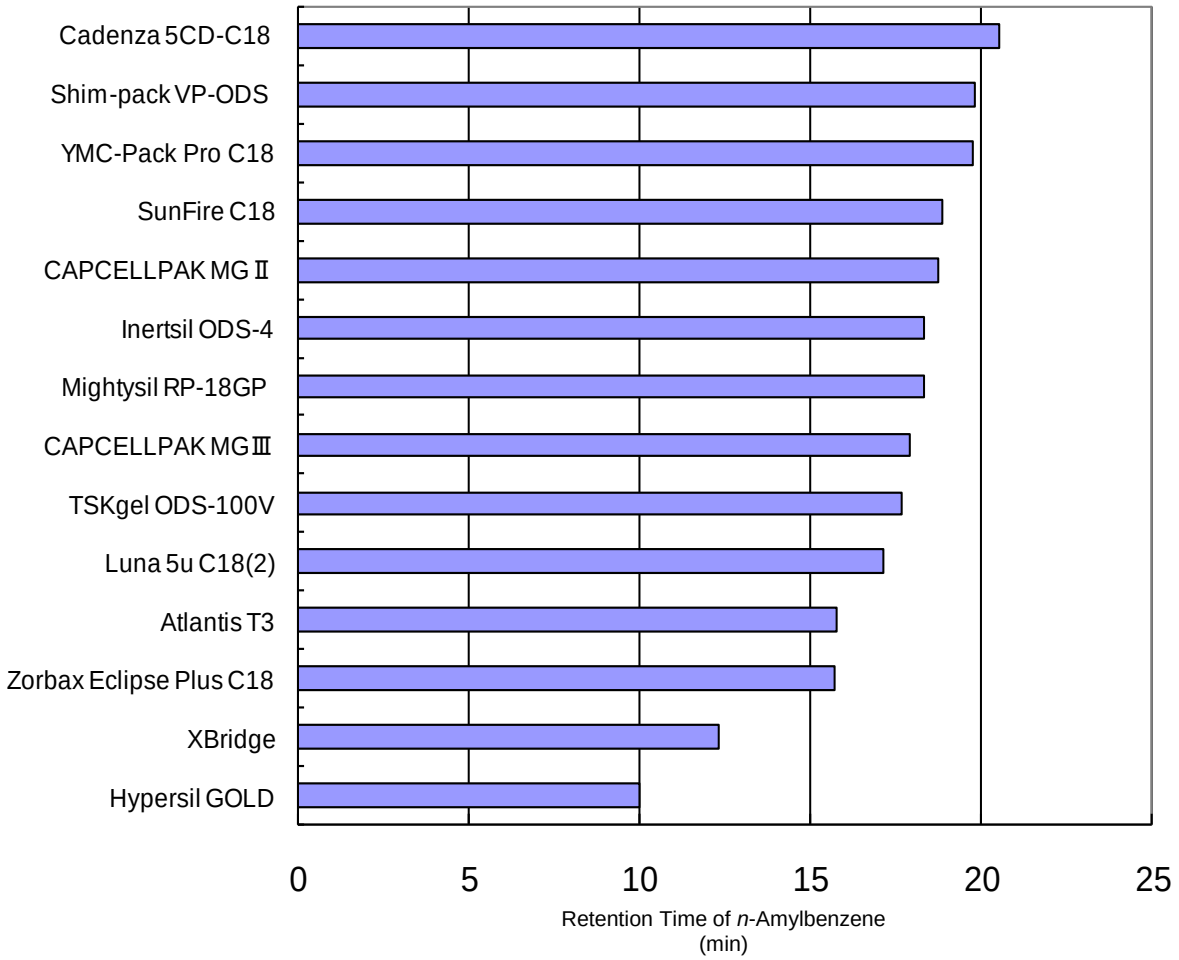


C8

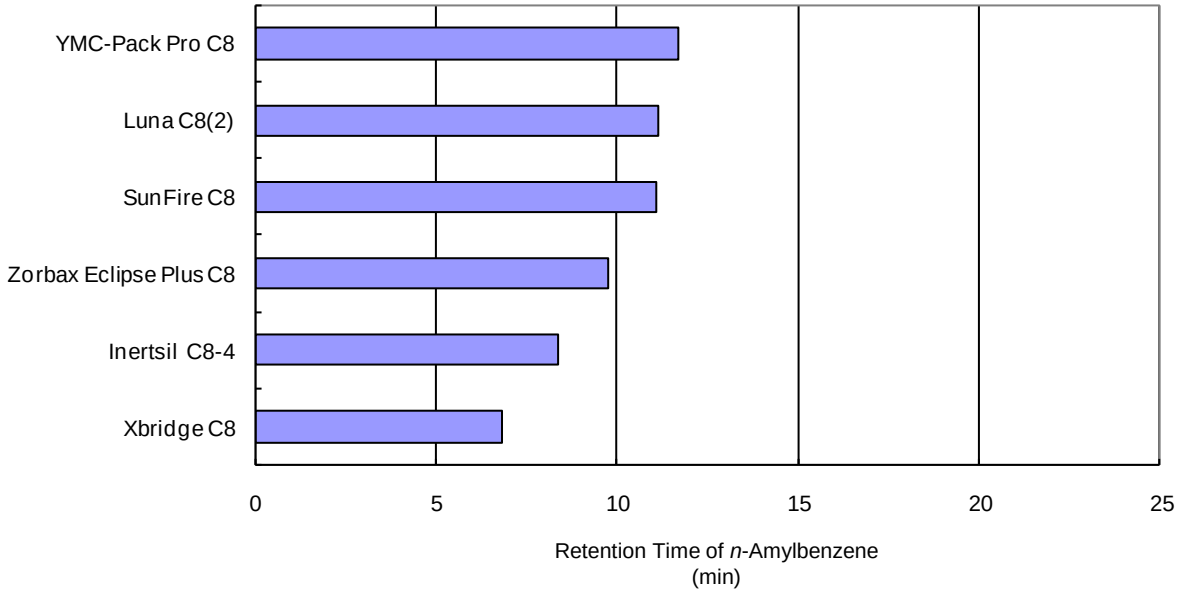


Comparison of Retentivity

List of ODS（C18）Columns



List of C8 Columns



Comprehensive Evaluation of ODS(C18) Columns

Column	Basic compound	Acidic compound	Chelating compound	Dewetting
Inertsil ODS-4	◎	◎	○	◎
Atlantis T3	○	○	×	○
SunFire C18	△	×	△	×
Xbridge	△	△	×	×
CAPCELLPAK MG Ⅱ	△	×	△	○
CAPCELLPAK MG Ⅲ	×	×	○	◎
Zorbax Eclipse Plus C18	△	×	×	×
YMC-Pack Pro C18	△	×	×	○
Luna 5u C18(2)	○	△	×	×
Cadenza 5CD-C18	×	○	×	×
TSKgel ODS-100V	○	◎	×	◎
Mightysil RP-18GP	△	◎	×	×
Hypersil GOLD	△	◎	○	○
Shim-pack VP-ODS	△	×	×	△

Comprehensive Evaluation of C8 Columns

Column	Basic compound	Acidic compound	Chelating compound	Dewetting
Inertsil C8-4	◎	◎	○	◎
YMC-Pack Pro C8	○	×	△	×
Zorbax Eclipse Plus C8	◎	×	×	△
Luna C8(2)	△	×	×	×
SunFire C8	○	×	×	×
Xbridge C8	○	△	×	×