Evaluation of 5 Kinds of 2 µm and Sub 2 µm C18 Columns Based on Separation Behavior

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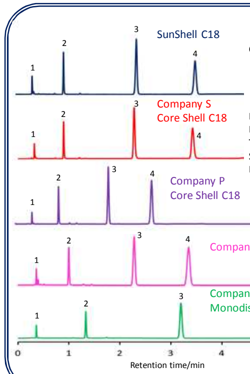
A column packed with 2.6 µm or 2.7 µm superficially porous particle has been widely used on HPLC and UHPLC, because it showed not only excellent column efficiency but also lower back pressure than sub-2 um column. Recently 2.0 µm and less than 2.0 µm superficially porous C18 columns were developed and have been available. In this study, 3 kinds of 2.0 µm and 1.7 µm superficially porous C18s and one totally porous hybrid C18, one totally porous monodisperse C18 were evaluated regarding efficiency, hydrogen bonding capacity, hydrophobicity, steric selectivity as well as peak shape of acidic, basic and metal chelating compounds. Compared C18 columns were SunShell C18 2 µm, Ascentis Express C18 2 µm, Kinetex C18 1.7 µm, Acquity BEH C18 1.7 µm and Titan C18 1.9 µm. Furthermore, efficiency loss due to frictional heating which yielded under high pressure and at high flow rate was observed. This efficiency loss was larger for a totally porous C18 than a superficially porous C18. Especially totally porous hybrid C18 showed the largest efficiency loss because of the lowest thermal conductivity.

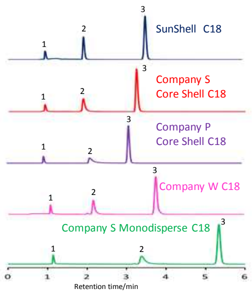
Theoretical plate and tailing factor



Column: SunShell C18, 2 µm 100 x 2.1 mm
Company S Core Shell C18, 2 µm 100 x 2.1 mm
Company P Core Shell C18, 1.7 µm 100 x 2.1 mm
Company W C18, 1.7 µm 100 x 2.1 mm
Company S Monodisperse C18, 1.9 µm 100 x 2.1 mm
Mobile phase: CH_3CN/H_2O=60/40
Flow rate: 0.5 mL/min
Temperature: 40 °C
Sample: 1 = Uracil, 2 = Ethylbenzoate, 3 = Acenaphthene, 4 = Butylbenzene

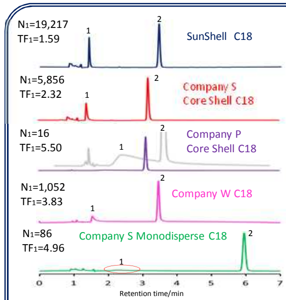
	Acenaphthene		Pressure (MPa)	Theoretical plate /pressure (MPa^{-1})
	Theoretical plate	Tailing factor		
SunShell C18	29,721	1.04	48.5	613
Company S Core Shell C18	25,533	1.19	53.5	477
Company P Core Shell C18	24,700	0.97	53.8	458
Company W C18	14,511	1.01	54.1	269
Company S Monodisperse C18	26,592	1.05	43.9	605

Comparison of Pyridine (2) as a basic compound



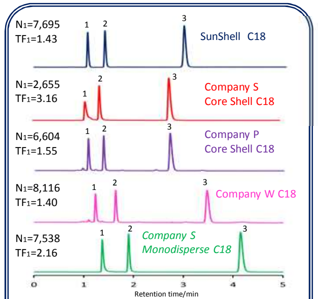
Column dimension: 100 x 2.1 mm
Mobile phase: CH_3OH/H_2O=30/70
Flow rate: 0.2 mL/min
Temperature: 40 °C
Detection: UV@250nm
Sample: 1 = Uracil
2 = Pyridine
3 = Phenol

Comparison of Oxine (1) as a metal chelating compound



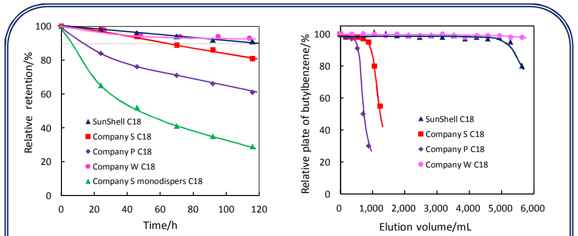
Column dimension: 100 x 2.1 mm
Mobile phase: CH_3CN/20mM H_3PO_4=10/90
Flow rate: 0.2 mL/min
Temperature: 40 °C
Detection: UV@250nm
Sample: 1 = 8-Quinolinol (Oxine)
2 = Caffeine

Comparison of Formic acid (1) as an acidic compound



Column dimension: 100 x 2.1 mm
Mobile phase: CH_3CN/0.1% H_3PO_4=2/98
Flow rate: 0.2 mL/min
Temperature: 40 °C
Detection: UV@210nm
Sample: 1 = Formic acid
2 = Acetic acid
3 = Propionic Acid

Stability under pH1 and 10 Conditions



Durable test condition
Column size: 50 x 2.1 mm
Mobile phase: CH_3CN/1.0% TFA, pH1=10/90
Flow rate: 0.4 mL/min
Temperature: 80 °C

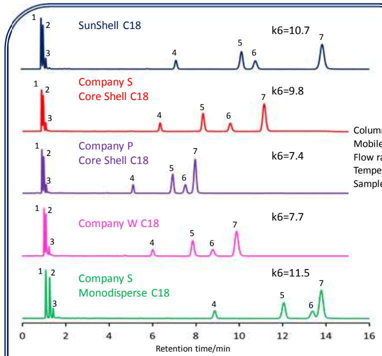
Measurement condition
Column size: 50 x 2.1 mm
Mobile phase: CH_3CN/H_2O=60/40
Flow rate: 0.4 mL/min
Temperature: 40 °C
Sample: 1 = Uracil
2 = Butylbenzene

Durable test condition
Column size: 50 x 2.1 mm
Mobile phase: CH_3OH/20mM Sodium borate/10mM NaOH=30/21/49 (pH10)
Flow rate: 0.4 mL/min
Temperature: 50 °C

Measurement condition
Column size: 50 x 2.1 mm
Mobile phase: CH_3OH/H_2O=70/30
Flow rate: 0.4 mL/min
Temperature: 40 °C
Sample: 1 = Butylbenzene

*Company S monodisperse C18 was not evaluated under pH10 condition because its pH stability is listed to be 2 to 8 in their catalog .

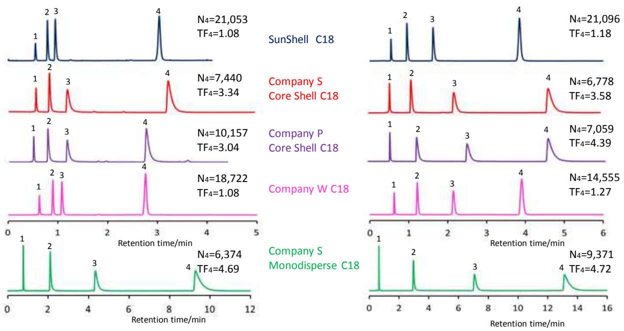
Separation of standard samples



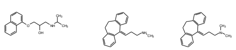
Column dimension: 100 x 2.1 mm
Mobile phase: CH_3OH/H_2O=75/25
Flow rate: 0.2 mL/min
Temperature: 40 °C
Sample: 1 = Uracil
2 = Caffeine
3 = Phenol
4 = Butylbenzene
5 = o-Terphenyl
6 = Amylbenzene
7 = Triphenylene

	Hydrogen bonding (Caffeine/Phenol)	Hydrophobicity (Amylbenzene/Butylbenzene)	Steric selectivity (Triphenylene/o-Terphenyl)
SunShell C18	0.43	1.59	1.41
Company S Core Shell C18	0.37	1.59	1.38
Company P Core Shell C18	0.45	1.57	1.17
Company W C18	0.35	1.55	1.30
Company S Monodisperse C18	0.53	1.58	1.16

Comparison of Amitriptyline (4) as a strong basic compound

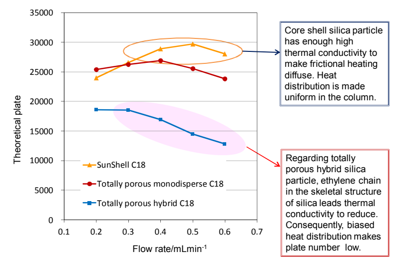


Column dimension: 100 x 2.1 mm
Mobile phase: CH_3CN/20 mM Phosphate buffer pH 7.0=60/40
Flow rate: 0.3 mL/min
Temperature: 40 °C
Detection: UV@250 nm
Sample: 1 = Uracil, 2 = Propranolol, 3 = Nortriptyline, 4 = Amitriptyline



Column dimension: 100 x 2.1 mm
Mobile phase: CH_3CN/10 mM ammonium acetate pH 6.8=40/60
Flow rate: 0.3 mL/min
Temperature: 40 °C
Detection: UV@250 nm
Sample: 1 = Uracil
2 = Propranolol
3 = Nortriptyline
4 = Amitriptyline

Decreasing of theoretical plate due to frictional heating effect



Column: 100 x 2.1 mm
Mobile phase: H_3CN/H_2O=60/40
Temperature: 40 °C
Sample: Acenaphthene

Conclusion

- SunShell C18 column provided highly inert phases that deliver excellent chromatographic performance for acidic, basic, neutral and metal chelating analytes.
- SunShell C18 column showed excellent stability under both acidic and basic pH conditions as well as company W C18.
- A core shell C18 like SunShell showed much lower frictional heating effect than a totally porous C18 or totally porous hybrid C18.