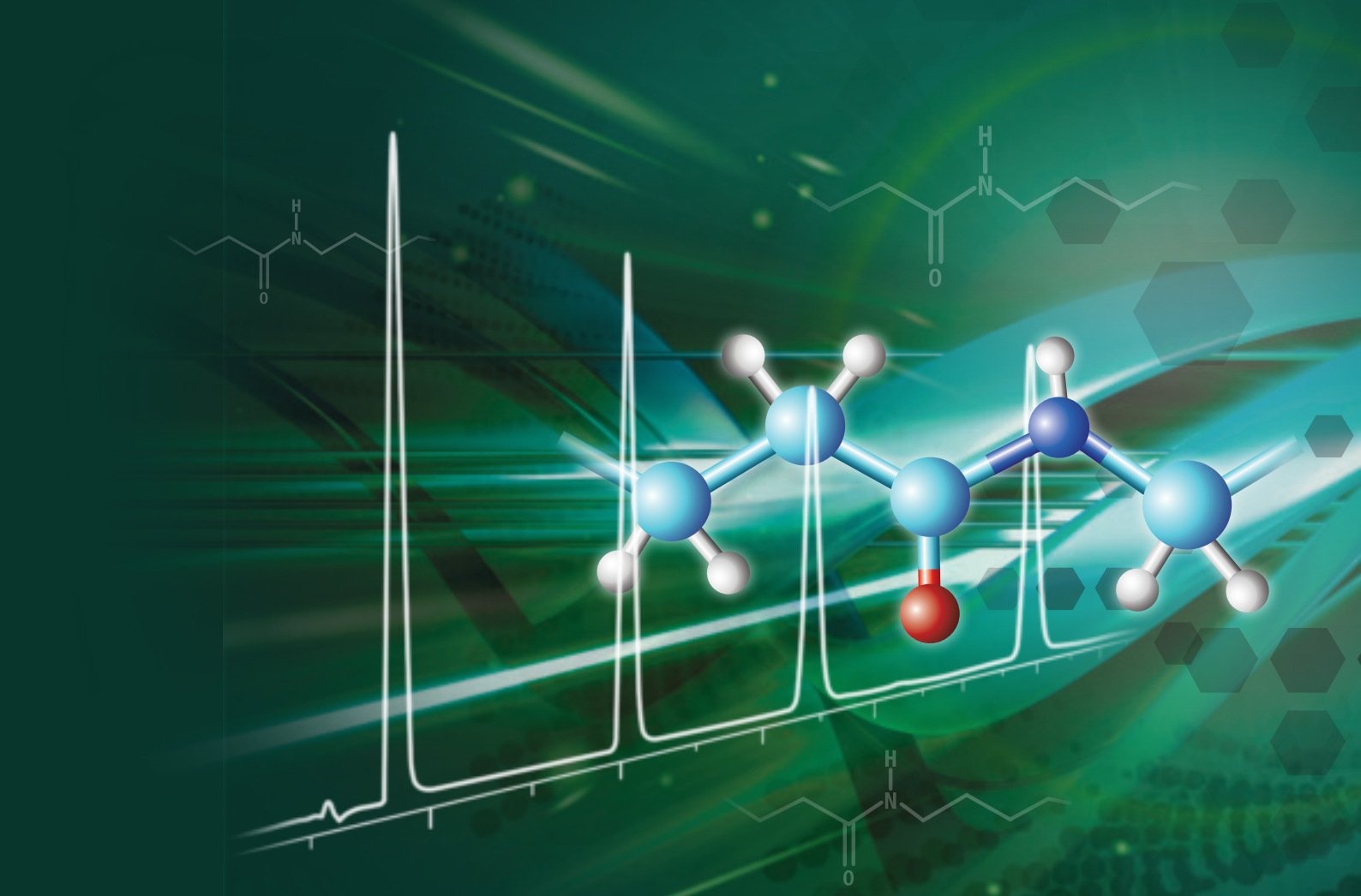
ACE® C18-Amide

For increased polar retention and alternative selectivity



- Alternative selectivity for method development
- Improved separations with polar, acidic, basic and phenolic compounds
- High efficiency 2µm, 3µm, 5µm and 10µm particles for UHPLC and HPLC
- Ultra-inert for maximum performance and reproducibility

ACE®
UHPLC and HPLC Columns

ACE® C18-Amide

For increased polar retention and alternative selectivity

Ideal Column Choice for Method Development

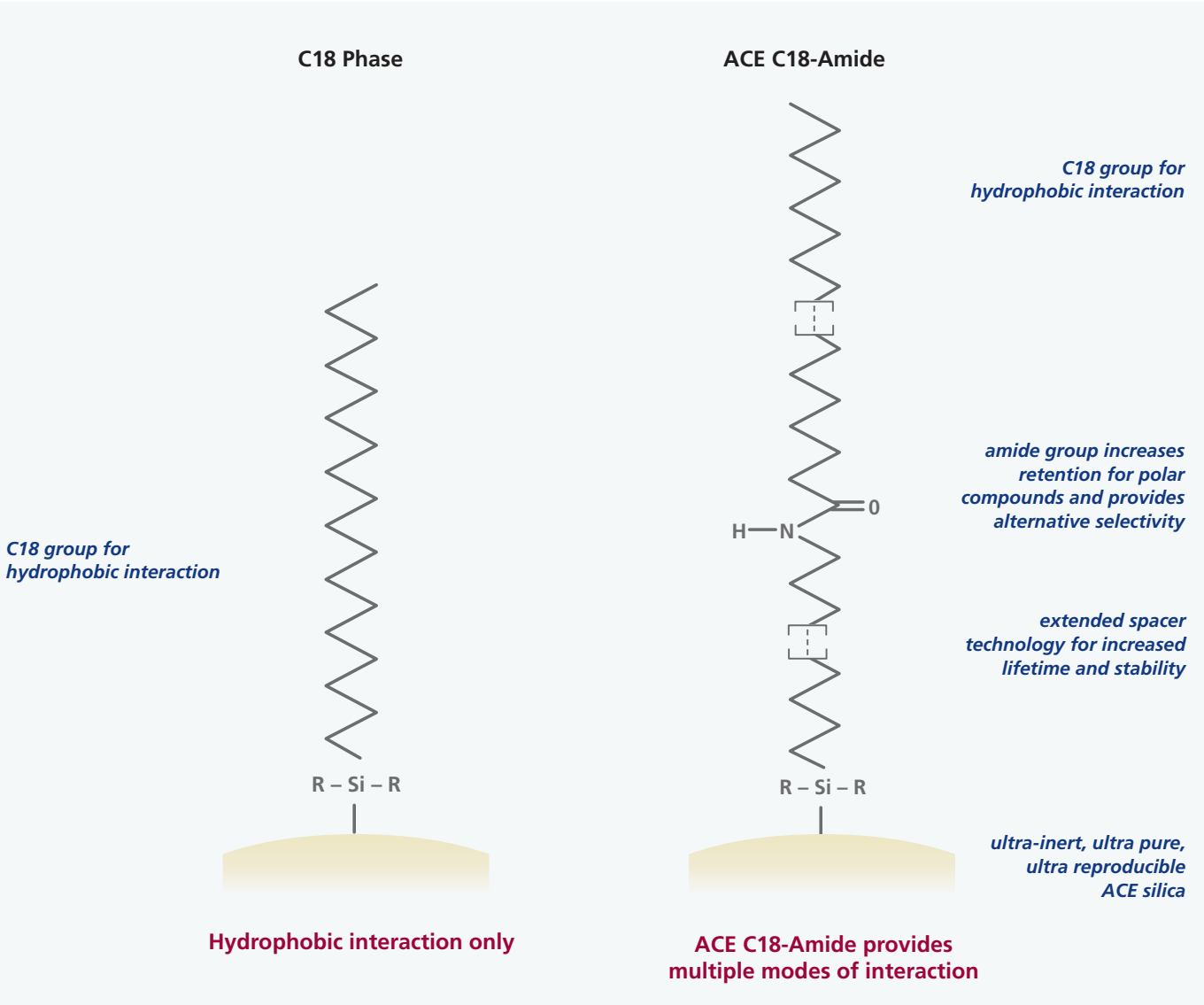
- Alternative selectivity to C18 and C8 columns with polar molecules - especially for acids
- Compatible with 100% aqueous mobile phases
- High efficiency 2µm, 3µm, 5µm and 10µm particles for UHPLC, HPLC and preparative separations
- Low bleed for UV and LC/MS compatibility

Recommended Applications

- Small water soluble analytes and polar compounds
- H-bond donors, acids, bases and phenolic compounds
- Small peptides

Why does ACE C18-Amide Provide Alternative Selectivity?

- ACE C18-Amide combines a C18 with a polar amide group on a single ligand
- Extended spacer technology additionally provides extended column lifetime



Leverage the Power of Selectivity with ACE C18-Amide to pull peaks apart

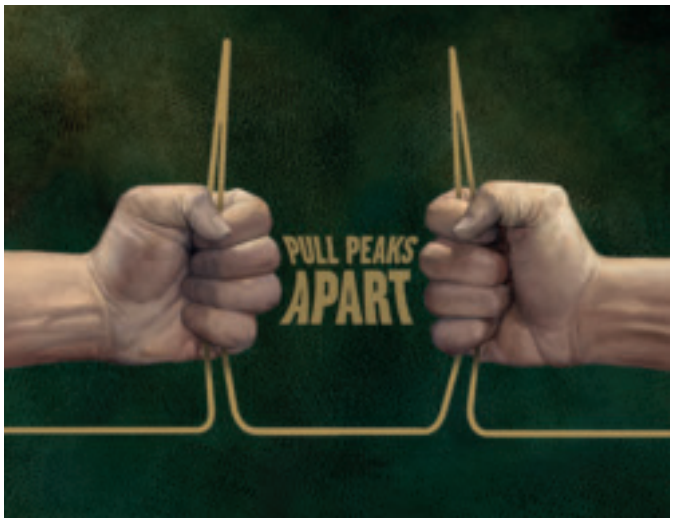
The resolution equation determines the parameters that contribute to resolution; N, α and k. In recent years there has been a significant focus on the use of ultra efficient "UHPLC" columns (such as ACE Excel 2µm columns) as a means of achieving separation goals.

$$R_s = \left(\frac{1}{4}\right) N^{0.5} \left(\frac{\alpha - 1}{\alpha}\right) \left(\frac{k}{1+k}\right)$$

Resolution | Efficiency | Selectivity | Retention Factor

However, selectivity is often overlooked. This is unfortunate as of the three parameters that affect resolution, selectivity is the most powerful (see Figure 1). By leveraging both efficiency and selectivity, better and faster separations can often be achieved.

ACE C18-Amide is the latest addition to the ACE range of bonded phases which offer both complementary selectivity and the option of 2µm, 3µm, 5µm and 10µm particle sizes - to enable development of rugged, robust UHPLC and HPLC methods.

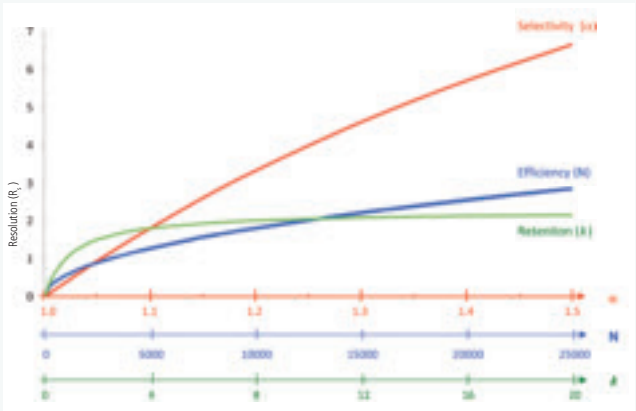


Leverage the power of bonded phase selectivity to pull peaks apart

Figure 2 illustrates the difference between two ACE bonded phases, C18-Amide and C18. Although both phases offer the possibility of strong hydrophobic interaction from their respective C18 chains, the amide group embedded within the C18-Amide phase introduces additional modes of interaction which ultimately increases retention for polar compounds and provides alternative selectivity.

Fig. 1 - The Effect of N, α and k on Resolution (R_s)

For a typical separation where N = 10,000, k = 4 and α = 1.1

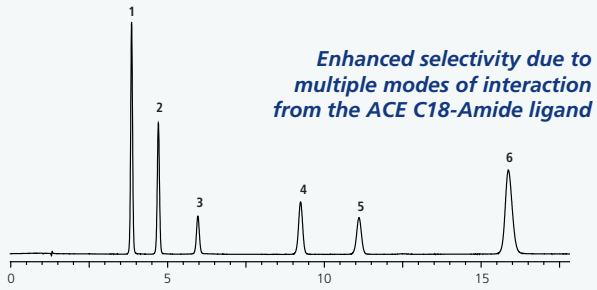


Increasing N, α or k increases resolution (R_s), but as seen from the above plots, increasing either N or k suffers from quickly diminishing returns. Increasing selectivity (α) does not have this problem and is therefore the most powerful of these three variables to optimise when developing a separation.

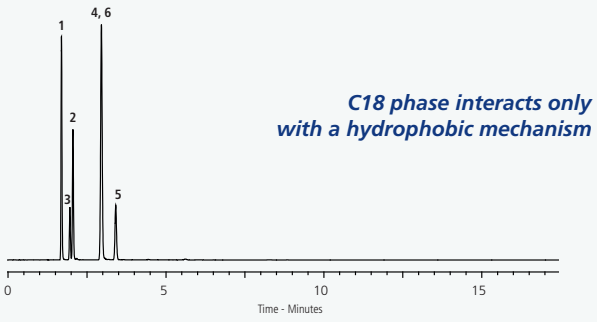
Fig. 2 - Use ACE C18-Amide to Enhance Selectivity

Application # 1601

ACE Excel 3 C18-Amide



ACE Excel 3 C18



Sample: 1) resorcinol 2) catechol 3) 2-methyl resorcinol 4) 4-methyl catechol 5) 3-methyl catechol 6) 4-nitro catechol Mobile Phase: 25:75 MeCN/27mM H_3PO_4 in H_2O Column Dimensions: 150 x 4.6mm Flow Rate: 1.50ml/min Temperature: 30°C Wavelength: 270nm

ACE C18-Amide Provides Enhanced Polar Selectivity

Fig. 3 - Comparison of ACE C18-Amide to Leading C18 UHPLC Columns Application # 1602

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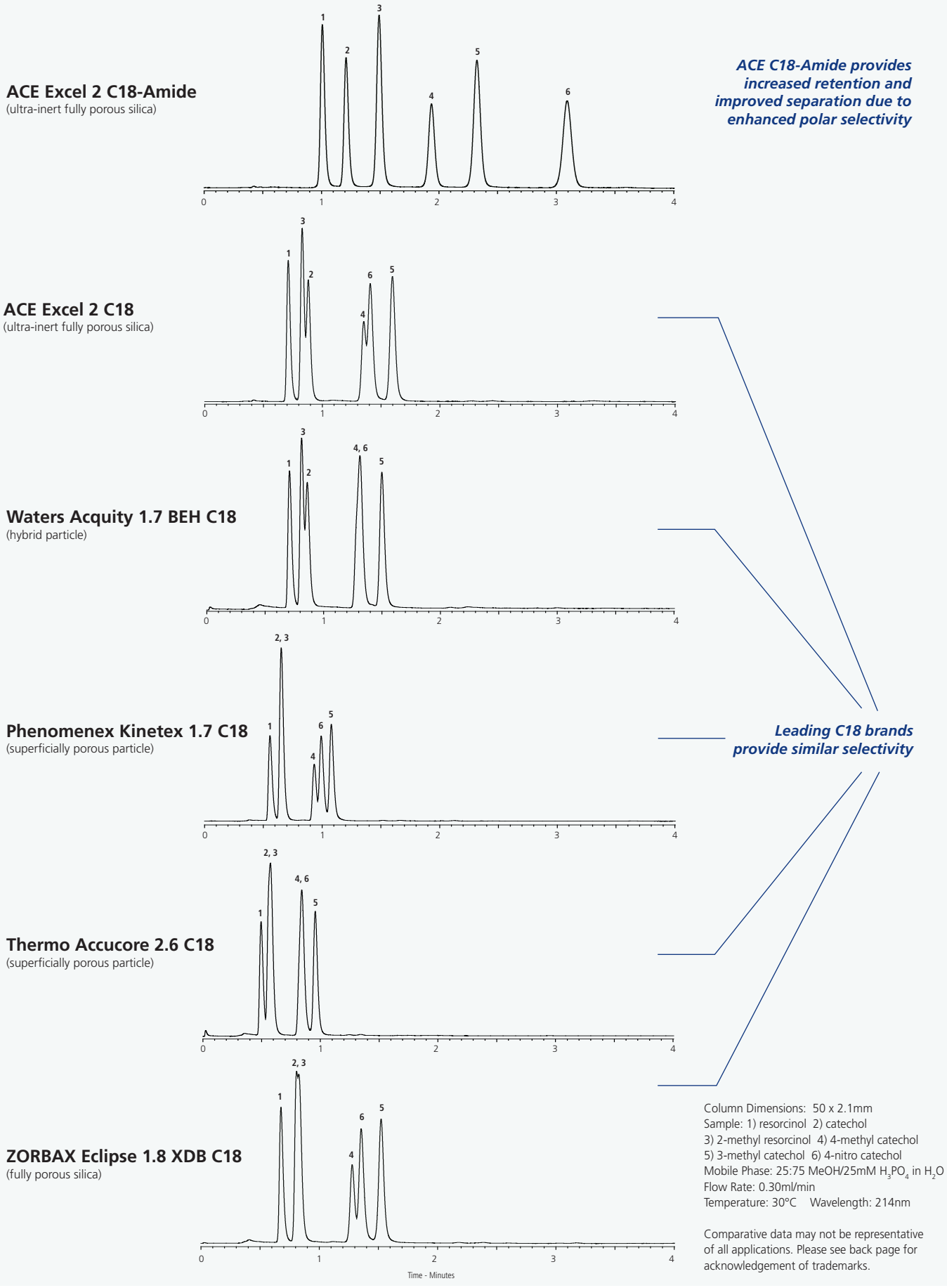


ACE C18-Amide provides increased retention and improved separation due to enhanced polar selectivity

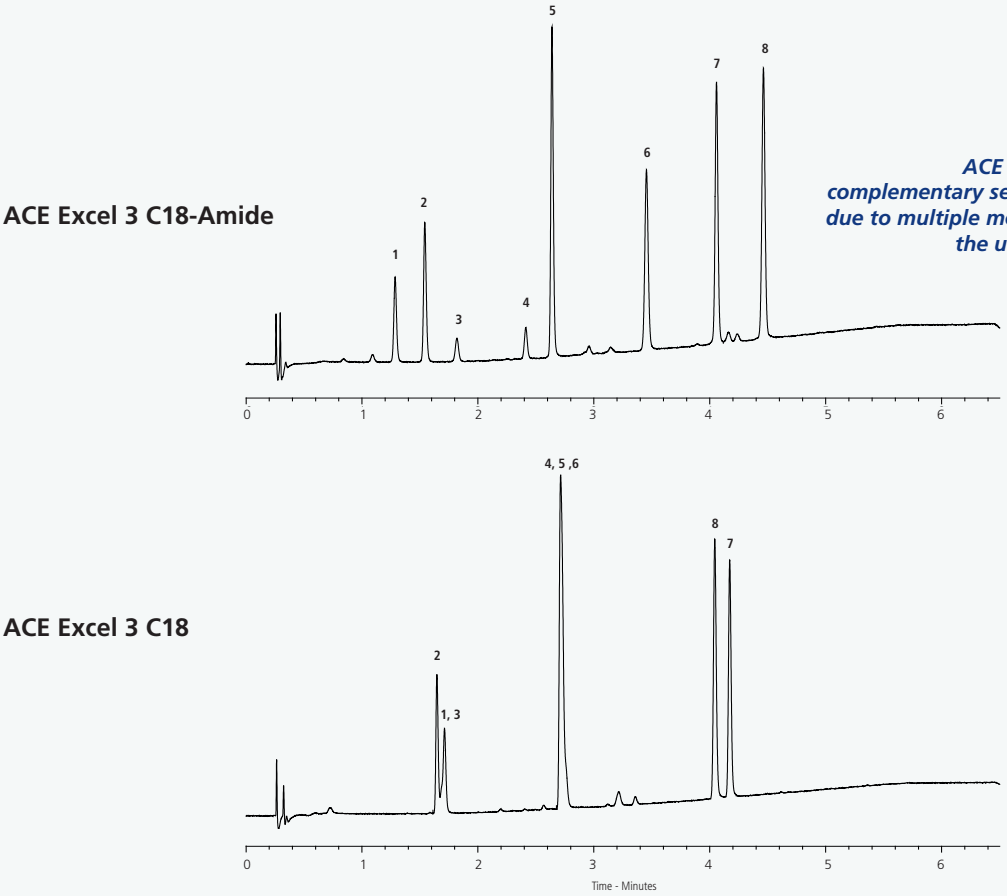
Leading C18 brands provide similar selectivity

Column Dimensions: 50 x 2.1mm
Sample: 1) resorcinol 2) catechol
3) 2-methyl resorcinol 4) 4-methyl catechol
5) 3-methyl catechol 6) 4-nitro catechol
Mobile Phase: 25:75 MeOH/25mM H_3PO_4 in H_2O
Flow Rate: 0.30ml/min
Temperature: 30°C Wavelength: 214nm

Comparative data may not be representative of all applications. Please see back page for acknowledgement of trademarks.

Exploit Alternative Selectivity for Method Development

Fig. 4 - Separation of Acidic, Basic and Neutral Compounds Application # 1603

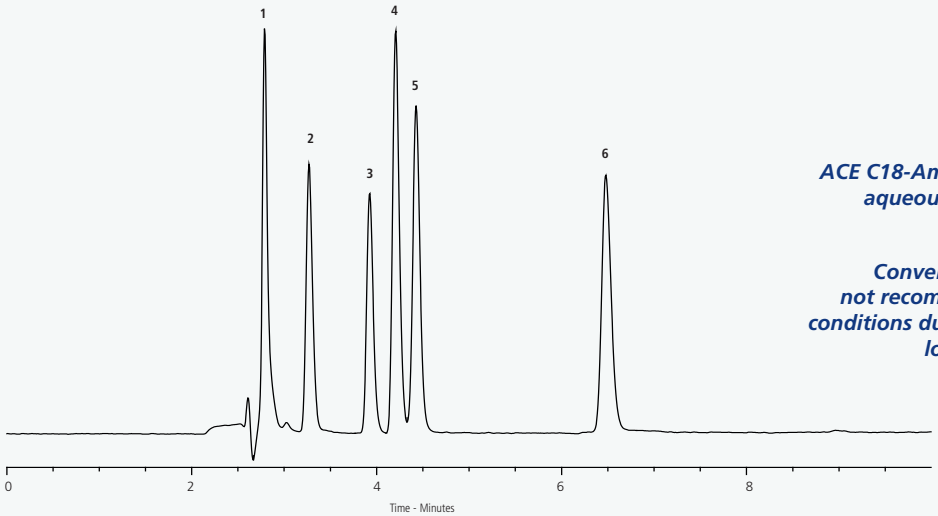


ACE C18-Amide phase offers complementary selectivity to C18 columns due to multiple modes of interaction from the unique C18-Amide ligand

Sample: 1) methylphenylsulphoxide 2) pindolol 3) 3-hydroxybenzoic acid 4) 1,2-dimethoxybenzene 5) berberine 6) myricetin 7) piperine 8) chrysin
Mobile Phase: A = 20mM ammonium formate in H_2O (pH 3.0) B = 20mM ammonium formate (pH 3.0) in 90:10 (v/v) MeOH/H_2O Gradient: 3 – 100% B in 5 minutes
Column Dimensions: 50 x 2.1mm Flow Rate: 0.60ml/min Temperature: 40°C Wavelength: 254nm

Compatible with 100% Aqueous Mobile Phases

Fig. 5 - Separation of Organic Wine Acids Application # 1604



ACE C18-Amide is compatible with 100% aqueous mobile phase for maximum retention and resolution

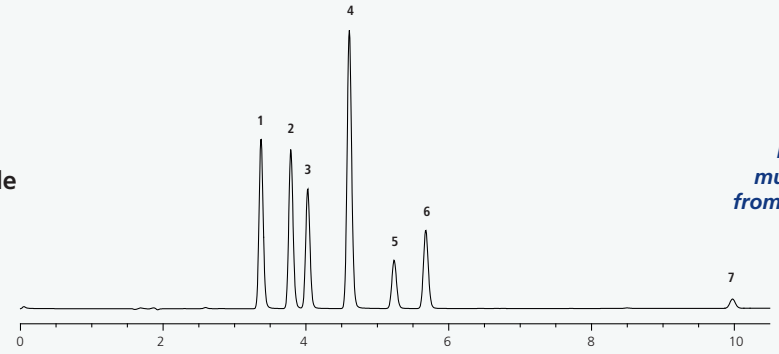
Conventional alkyl C18 columns are not recommended with 100% aqueous conditions due to irreproducible retention loss caused by pore dewetting

Sample: 1) oxalic acid 2) tartaric acid 3) malic acid 4) lactic acid 5) ascorbic acid 6) citric acid
Mobile Phase: 40mM $NH_4H_2PO_4$ in H_2O (pH 2.5)
Column: ACE Excel 3 C18-Amide, 250 x 2.1mm Flow Rate: 0.21ml/min Temperature: 25°C Wavelength: 214nm

ACE C18-Amide Provides Alternative Selectivity

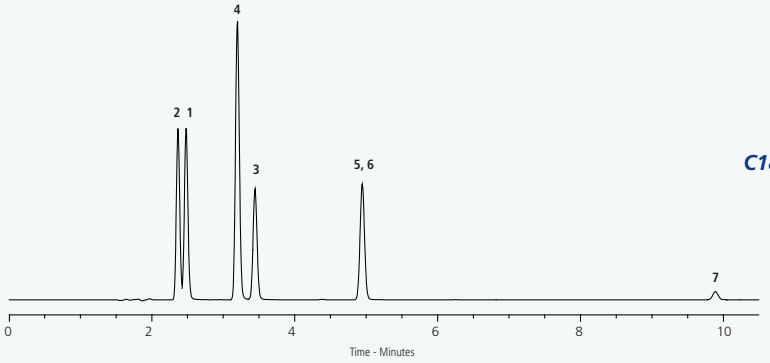
Fig. 6 - Food and Beverage Additives - Vanillins Application # 1605

ACE Excel 3 C18-Amide



Improved resolution due to multiple modes of interaction from the ACE C18-Amide ligand

ACE Excel 3 C18

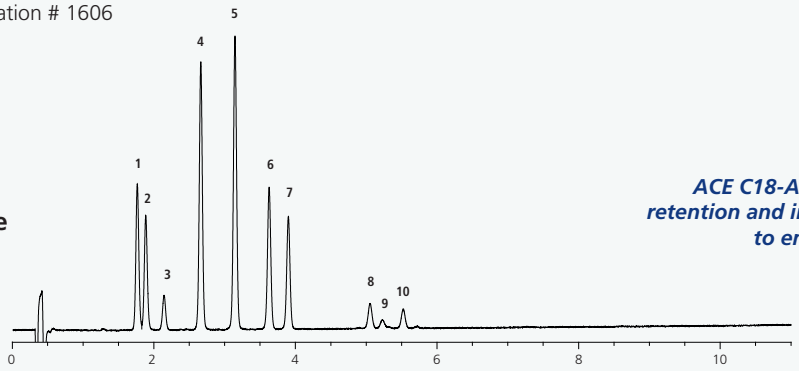


C18 phase interacts only with a hydrophobic mechanism

Sample: 1) vanillic acid 2) 4-hydroxybenzoic acid 3) vanillin 4) 4-hydroxybenzaldehyde 5) guaiacol 6) ethyl vanillin 7) eugenol
Mobile Phase: A = 0.1% formic acid in H_2O B = 0.1% formic acid in MeCN Gradient: 30 – 55% B in 10 minutes
Column Dimensions: 150 x 4.6mm Flow Rate: 1.00ml/min Temperature: 40°C Wavelength: 260nm

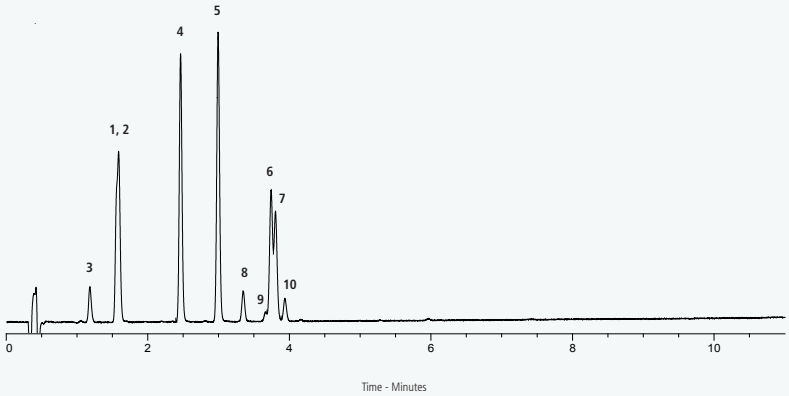
Fig. 7 - Steroids Application # 1606

ACE Excel 3 C18-Amide



ACE C18-Amide provides increased retention and improved separation due to enhanced polar selectivity

ACE Excel 3 C18

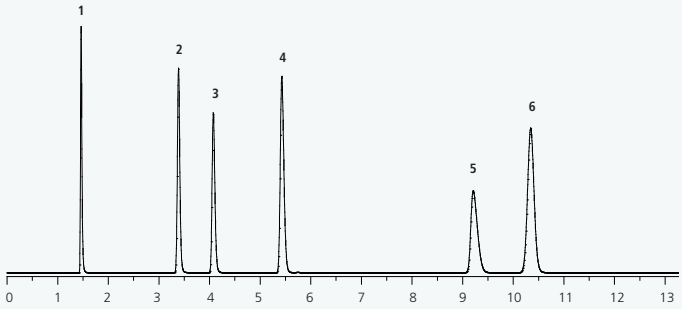


Sample: 1) prednisone 2) prednisolone 3) estriol 4) corticosterone 5) 11α-hydroxyprogesterone 6) 11-ketoprogesterone 7) 21-hydroxyprogesterone 8) β-estradiol 9) 17α-estradiol 10) 17α-ethynylestradiol
Mobile Phase: A = 0.1% formic acid in H_2O B = 0.1% formic acid in MeCN Gradient: 25 – 80% B in 10 minutes
Column Dimensions: 50 x 2.1mm Flow Rate: 0.40ml/min Temperature: 25°C Wavelength: 260nm

Guaranteed Reproducibility and Fully Scalable

Fig. 8 - Reproducible Scale Up with ACE C18-Amide Application # 1607

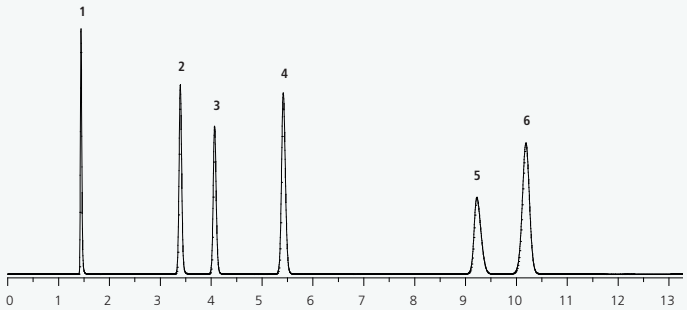
ACE Excel 2 C18-Amide



LC/MS
150 x 2.1mm
0.21ml/min

(Silane Batch #1)

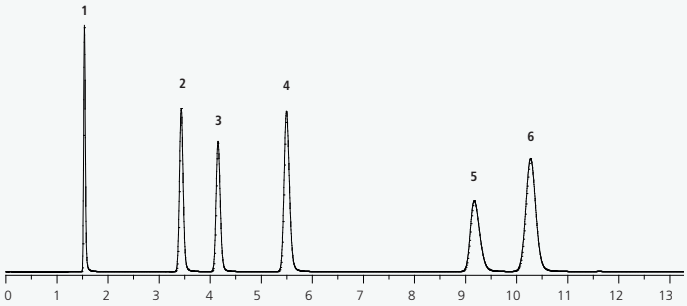
ACE Excel 3 C18-Amide



Analytical
150 x 3.0mm
0.42ml/min

(Silane Batch #2)

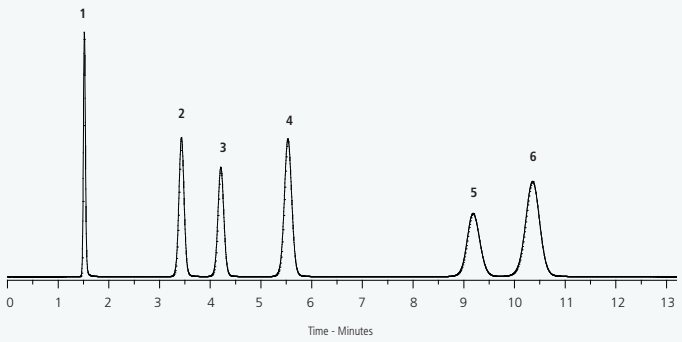
ACE Excel 5 C18-Amide



Analytical
150 x 4.6mm
1.00ml/min

(Silane Batch #3)

ACE 10 C18-Amide



Preparative
150 x 21.2mm
21.2ml/min

(Silane Batch #4)

Sample: 1) uracil 2) 4-hydroxybenzoic acid 3) acetylsalicylic acid 4) benzoic acid 5) 2-hydroxybenzoic acid 6) ethyl paraben
Mobile Phase: 35:65 MeCN/0.1% TFA in H_2O Temperature: 22°C Wavelength: 254nm

The availability of 2µm, 3µm, 5µm and 10µm particle sizes combined with a range of dimensions from UHPLC through to preparative HPLC scale ensures that methods can be reproducibly scaled up or down.

The chromatograms in figure 8 demonstrate the excellent reproducibility achieved when changing both silica batch and silane batch, and the reproducible scalability obtained when changing particle size and column diameter.

Product Availability and Specifications

Phase	Functional Group	Endcapped	Particle Size (µm)	Pore Size (Å)	Surface Area (m^2/g)	Carbon Load (%)	Maximum pH Range	USP Listing
ACE C18-Amide	Octadecyl with integral amide polar group	Yes	2, 3, 5, 10	100	300	16.4	2.0-8.0[a]	L1 / L60

[a] For optimum column lifetime, a pH range of 2-8 is recommended. To increase column lifetime at higher pH, organic buffers, low buffer concentrations, high % organic solvent and low temperatures must be considered. Further information is contained within "A Guide to HPLC and LC/MS Buffer Selection" by John Dolan – please contact your distributor to request your FREE copy or visit www.ace-hplc.com.

To further extend column lifetime under HPLC conditions (up to 5000psi/350bar), ACE guard cartridges or ACE HPLC pre-column filters are recommended.
To further extend column lifetime under UHPLC conditions (up to 15000psi/1000bar), ACE UHPLC pre-column filters are recommended.
For HPLC column connections up to 5000psi (350bar), PEEK fingertight fittings (p/n ACE-CC10) are recommended.
For UHPLC column connections up to 15000psi (1000bar), reuseable fittings (p/n EXL-CC10) are recommended.
For further details please contact your distributor or visit www.ace-hplc.com.

ACE Excel 2µm C18-Amide UHPLC/HPLC Columns (supplied in dual compatible UHPLC/HPLC "Excel" hardware format with 1000bar/15000psi pressure limit)

Column Diameter	Column Length							
	20mm	30mm	35mm	50mm	75mm	100mm	125mm	150mm
2.1mm	EXL-1012-0202U	EXL-1012-0302U	EXL-1012-3502U	EXL-1012-0502U	EXL-1012-7502U	EXL-1012-1002U	EXL-1012-1202U	EXL-1012-1502U
3.0mm	EXL-1012-0203U	EXL-1012-0303U	EXL-1012-3503U	EXL-1012-0503U	EXL-1012-7503U	EXL-1012-1003U	EXL-1012-1203U	EXL-1012-1503U
4.6mm	EXL- 1012-0246U	EXL-1012-0346U	EXL-1012-3546U	EXL-1012-0546U	EXL-1012-7546U	EXL-1012-1046U	EXL-1012-1246U	EXL-1012-1546U

ACE Excel 3µm C18-Amide UHPLC/HPLC Columns (supplied in dual compatible UHPLC/HPLC "Excel" hardware format with 1000bar/15000psi pressure limit)

Column Diameter	Column Length								
	20mm	30mm	35mm	50mm	75mm	100mm	125mm	150mm	250mm
2.1mm	EXL-1112-0202U	EXL-1112-0302U	EXL-1112-3502U	EXL-1112-0502U	EXL-1112-7502U	EXL-1112-1002U	EXL-1112-1202U	EXL-1112-1502U	EXL-1112-2502U
3.0mm	EXL-1112-0203U	EXL-1112-0303U	EXL-1112-3503U	EXL-1112-0503U	EXL-1112-7503U	EXL-1112-1003U	EXL-1112-1203U	EXL-1112-1503U	EXL-1112-2503U
4.6mm	EXL-1112-0246U	EXL-1112-0346U	EXL-1112-3546U	EXL-1112-0546U	EXL-1112-7546U	EXL-1112-1046U	EXL-1112-1246U	EXL-1112-1546U	EXL-1112-2546U

ACE Excel 5µm C18-Amide UHPLC/HPLC Columns (supplied in dual compatible UHPLC/HPLC "Excel" hardware format with 1000bar/15000psi pressure limit)

Column Diameter	Column Length								
	20mm	30mm	35mm	50mm	75mm	100mm	125mm	150mm	250mm
2.1mm	EXL-1212-0202U	EXL-1212-0302U	EXL-1212-3502U	EXL-1212-0502U	EXL-1212-7502U	EXL-1212-1002U	EXL-1212-1202U	EXL-1212-1502U	EXL-1212-2502U
3.0mm	EXL-1212-0203U	EXL-1212-0303U	EXL-1212-3503U	EXL-1212-0503U	EXL-1212-7503U	EXL-1212-1003U	EXL-1212-1203U	EXL-1212-1503U	EXL-1212-2503U
4.6mm	EXL-1212-0246U	EXL-1212-0346U	EXL-1212-3546U	EXL-1212-0546U	EXL-1212-7546U	EXL-1212-1046U	EXL-1212-1246U	EXL-1212-1546U	EXL-1212-2546U

ACE 5µm C18-Amide Semi-Prep and Preparative HPLC Columns

Column Diameter	Column Length					
	50mm	75mm	100mm	125mm	150mm	250mm
7.75mm	ACE-1212-0508	ACE-1212-7508	ACE-1212-1008	ACE-1212-1208	ACE-1212-1508	ACE-1212-2508
10.0mm	ACE-1212-0510	ACE-1212-7510	ACE-1212-1010	ACE-1212-1210	ACE-1212-1510	ACE-1212-2510
21.2mm	ACE-1212-0520	ACE-1212-7520	ACE-1212-1020	ACE-1212-1220	ACE-1212-1520	ACE-1212-2520

ACE 10µm C18-Amide Analytical, Semi-Prep and Preparative HPLC Columns Please enquire

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