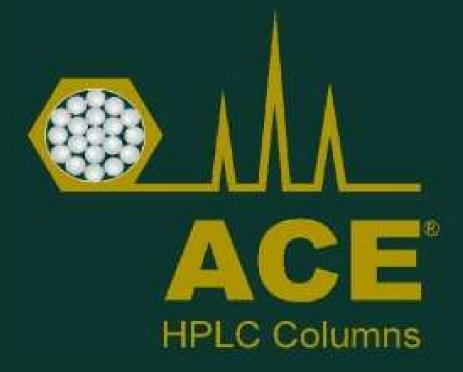


Exploring and Leveraging Multi-Mode Interactions To Maximise Chromatographic Selectivity With Uniquely Designed HPLC / UHPLC Stationary Phases



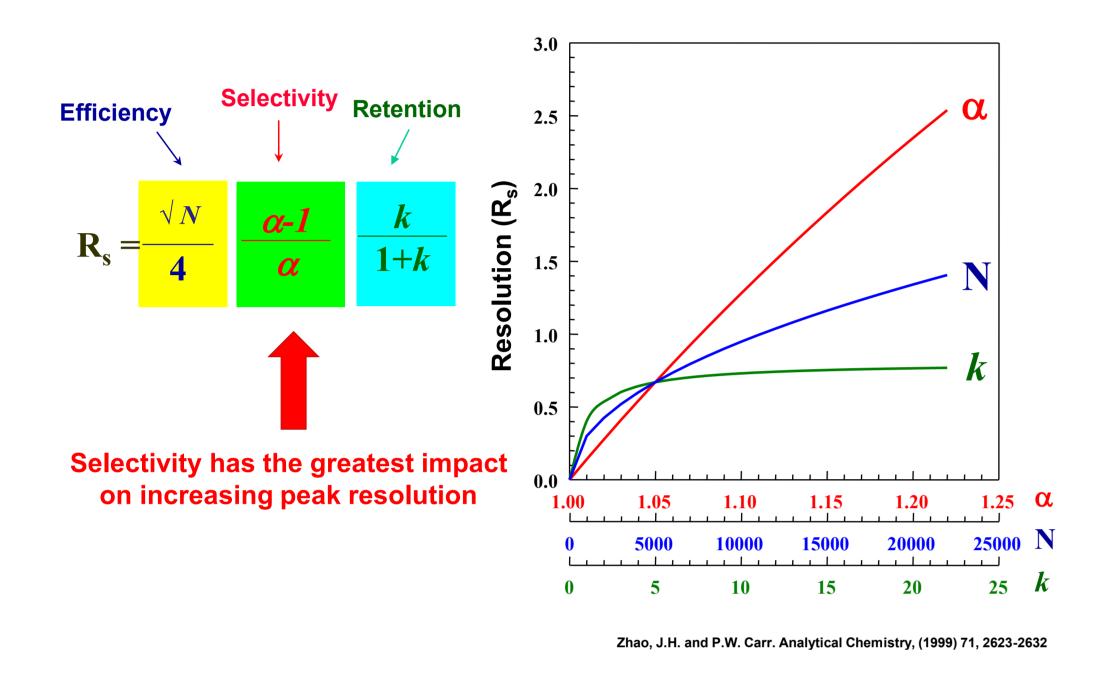
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THE CHALLENGE

Engineer new HPLC / UHPLC phases based upon the robust C18 ligand with alternative selectivities that are reproducibile, robust, exhibit low bleed and give efficient peaks

1. CHROMATOGRAPHIC PEAK RESOLUTION

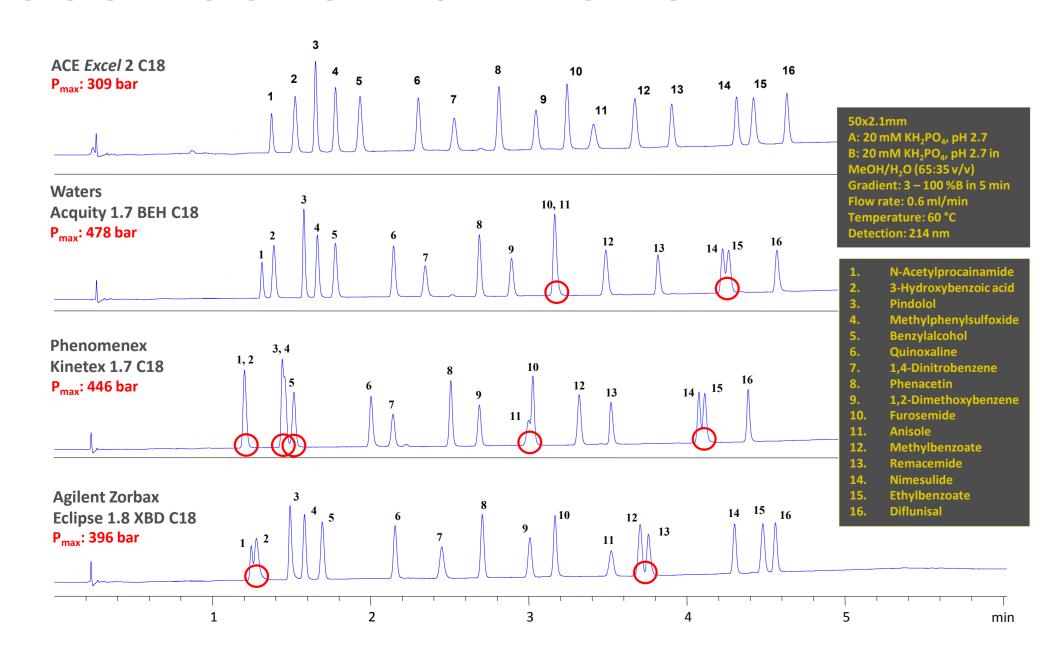


2. HPLC SURVEYS^a ...LISTENING TO THE ANALYST

- Column reproducibility and column lifetime are major factors for analysts
- Have been the top 2 feedback points since 2007
- Critical in pharmaceutical and other major industries for method transfers / consistency and long term performance
- Reversed-phase is the dominant separation mode
 - C18 & C8 = 60%; phenyl = 16%; CN = 9.5%; fluorinated = 5.9% - 92% analysts use C18 at some time in their work...they typically meet the above criteria
 - BUT limited selectivity

All trademarks are recognised...comparative separations may not be representative of all applications

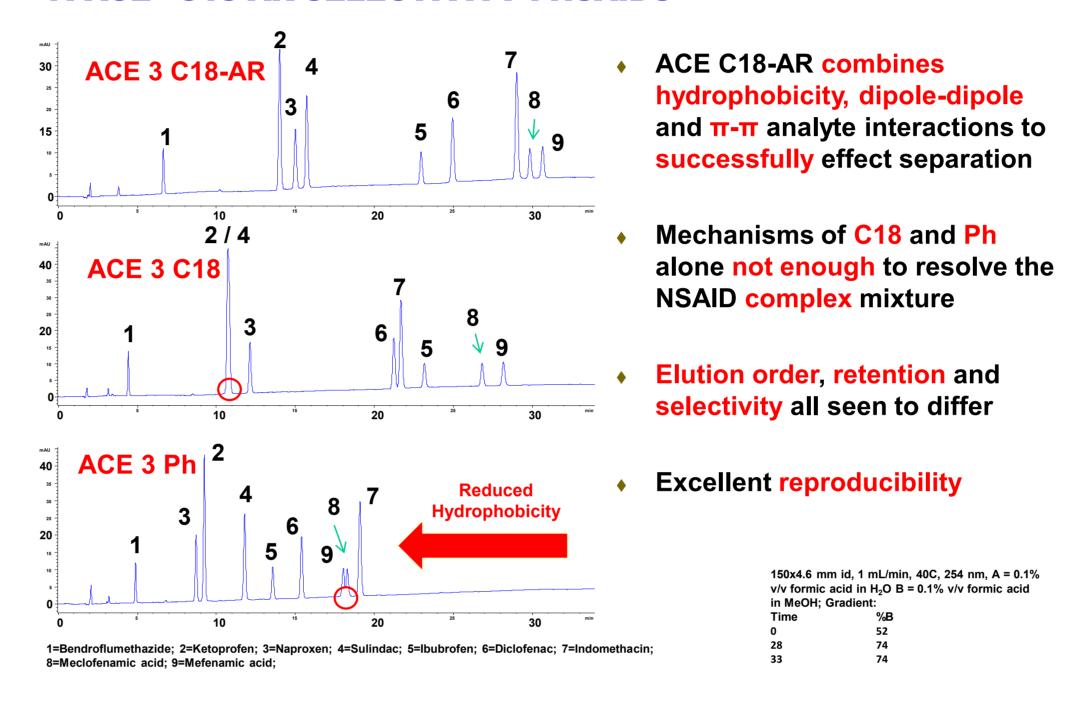
3. C18 PHASES PROVIDE SIMILAR SELECTIVITY



4. THE BENEFITS OF AROMATIC FUNCTIONALITY

- Phases with aromatic functionality include phenyl and pentafluorophenyl (PFP) based ligands
- Advantages
- Aromatic functionality potentially offer unique interactions with analytes (c.f. C18) giving alternative selectivity
- Provides enhanced retention of polar compounds
- Many aromatic functionality-based phases can be used in 100% aqueous eluents
- Disadvantages
 - Phenyl / PFP phases may suffer phase bleed
 - Batch-to-batch reproducibility & robustness traditionally weak

7. ACE® C18-AR SELECTIVITY: NSAIDS



10. ACE® SELECTIVITY RANKING: MEOH & MECN

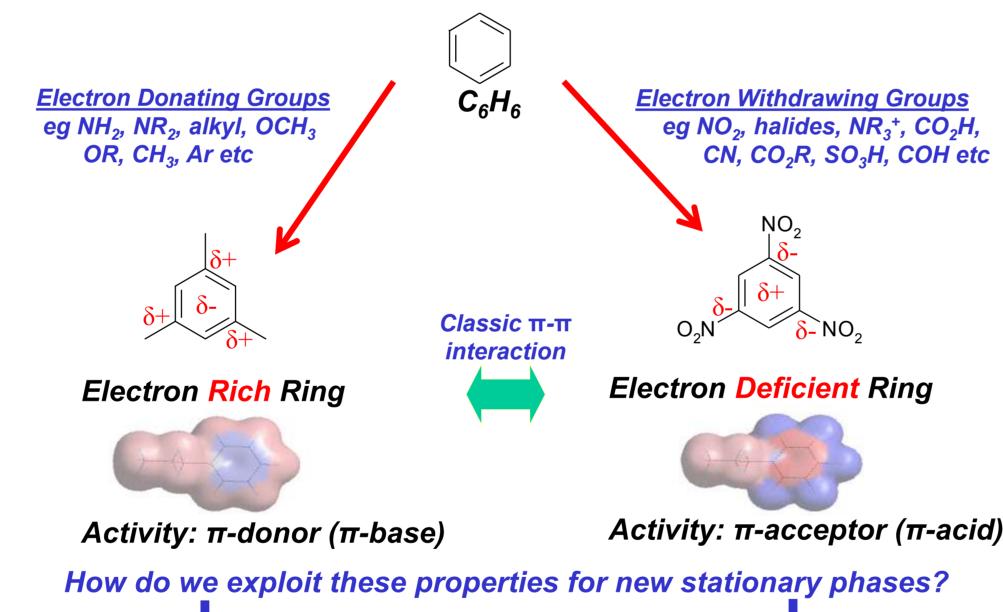
For >100 acidic, basic and neutral analytes assessed

WIEOH				MeCN
Column 1	Column 2	Selectivity 'S'	Column 1	Column 2
C18	C18-AR	12	C18	C18-AR
C18	C18-PFP	11	C18-AR	C18-PFP
C18-AR	C18-PFP	10	C18	C18-PFP

MeOH	MeCN	Selectivity Value
C18-PFP	C18	19
C18-AR	C18	18
C18-AR	C18-PFP	18
C18-PFP	C18-AR	18
C18-PFP	C18-PFP	18
C18	C18-AR	17
C18	C18-PFP	17
C18	C18	15
C18-AR	C18-AR	15



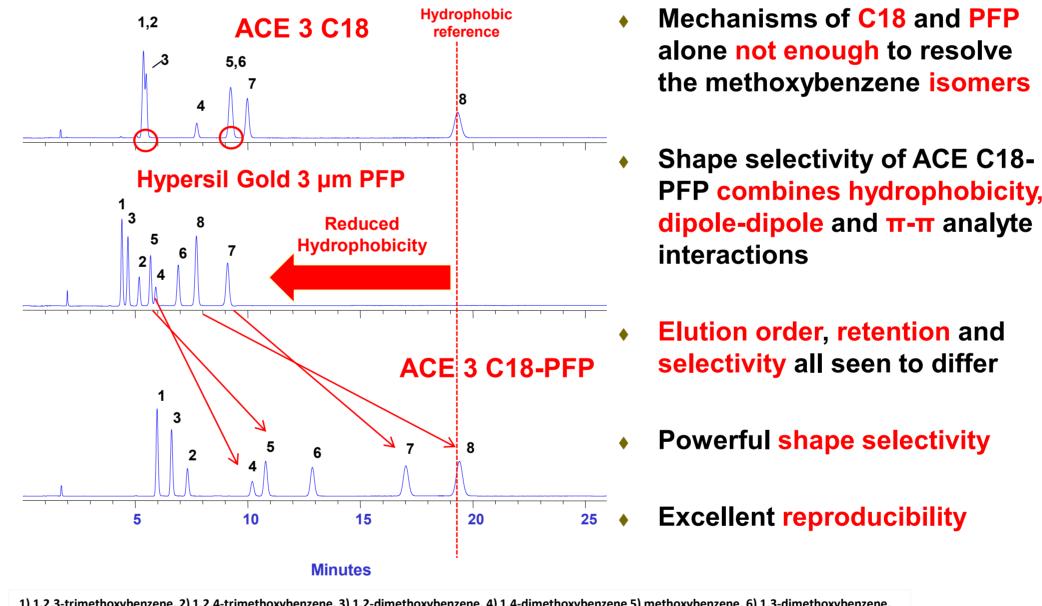
5. THE POWER OF TT...SCIENTIFIC LED PHASE DESIGN



C18+Phenyl = ACE® C18-AR

 $C18+PFP = ACE^{\otimes} C18-PFP$

8. ACE® C18-PFP SELECTIVITY: STRUCTURAL ISOMERS



11. WHAT DO I USE THESE NOVEL PHASES FOR?

- ◆ ACE® C18-AR
 - Useful for analytes that contain electron withdrawing moieties eg -NO₂, -halides, -NR₃⁺, -SO₂ -CO₂H, -SO₃H, -CO₂R, -CHO etc
 - Moderate shape selectivity
- ◆ ACE[®] C18-PFP
 - Useful for analytes that contain electron donating moieites eg -NH₂, -NR₂, -OCH₃, -OH₃, -alkyl, -Ar etc
 - Excellent shape selectivity...good for regioisomers

6. ACE® PHASES: MULTI-MODE SEPARATION MECHANISMS

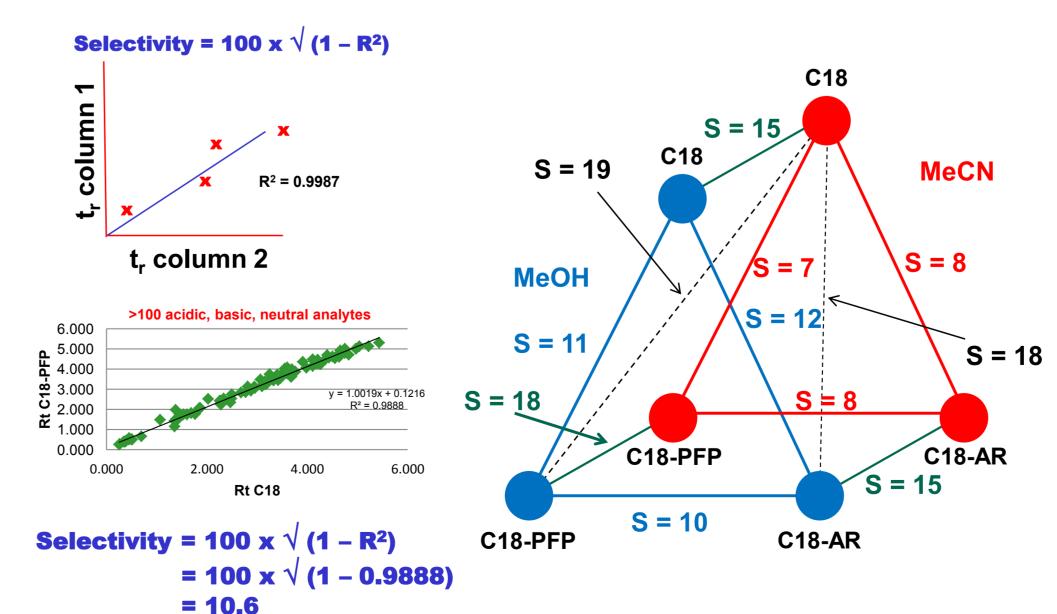
Combining the character of C18+phenyl and C18+PFP into single phases harnesses the best of each ligand type for unique selectivity

AC	E® C18	-AR		ACE® C18-PFP			
Separation mechanism	Typical C18	Typical Phenyl	ACE® C18-AR	Separation mechanism	Typical C18	Typical PFP	ACE® C18-PFP
Hydrophobicity	++++	+ / ++	++++	Hydrophobicity	++++	+/++	++++
π-π Interaction	-	+++	+++	π- $π$ Interaction	-	+++	+++
Dipole - Dipole	-	+	+	Dipole - Dipole	-	++++	++++
Hydrogen Bonding	-	++	++	Hydrogen Bonding	-	+++	+++
Shape Selectivity	++	++	++ / +++	Shape Selectivity	++	+++	++++

The predominance of each retention mechanism will be dictated by the analyte's physicochemical properties, its structure and the chromatographic conditions applied

Reproducible Multi-Mode Interactions Offer the Chromatographer More

9. ACE® PHASE COMPARISONS WITH SELECTIVITY 'S' VALUES'



Neue, O'Gara, Méndez "Selectivity in Reversed-Phase Separations: Influence of the Stationary Phase", J. Chromatogr. A 1127 (2006), 161-174

12. CONCLUSIONS

- ◆ The unique ACE® C18-AR and ACE® C18-PFP phases have been engineered based upon aromatic functionality
- These phases give complementary, yet alternative selectivity and offer the chromatographer new choices for method development
- The phases offer high peak efficiency, reproducibility, robustness and with low UV & MS bleed
- The phases are available for HPLC (ACE® 3, 5, 10μm) and for UHPLC (ACE[®] Excel ™ 2μm)

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