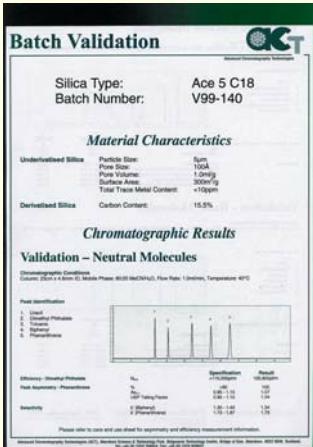


Reproducibility and Validation

Subtle changes in silanol activity are one of the primary causes of column to column selectivity changes. Base deactivated columns generally have better reproducibility than other column types due to fewer interactions

between silanols and polar compounds. ACE columns, by virtually eliminating silanol interactions, provide an outstanding level of column to column reproducibility for polar compounds.

Complete Validation



SILICA MANUFACTURE

Ultra pure reagents and strict control of the manufacturing process result in a high purity silica with uniform surface characteristics.

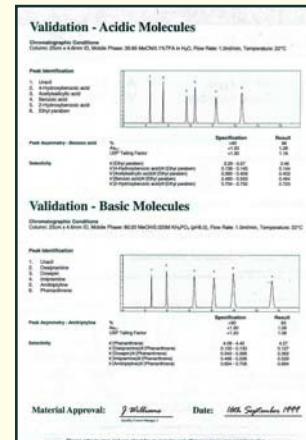
Advanced bonding techniques are then employed, resulting in a range of highly base deactivated phases that combine superb reproducibility with excellent robustness.



BATCH VALIDATION

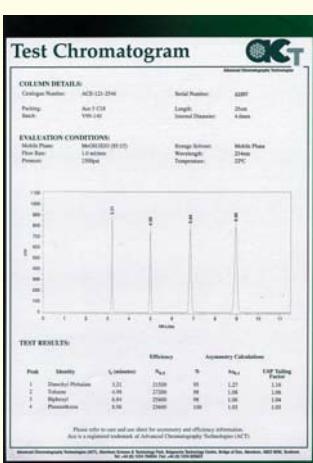
Every batch is extensively tested for selectivity and surface activity with a range of acidic, basic and neutral molecules.

ACE HPLC columns have the most stringent batch validation specification of any reversed-phase material.



COLUMN VALIDATION

All columns are tested with a multi-component mixture to ensure excellent performance and peak shape are obtained.



ACE 5 C18 - Basic Molecule Validation Parameters:

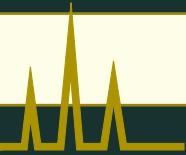
Peak Asymmetry - Amitriptyline

| | Specification | Result |
|--------------------|---------------|--------|
| % | >90 | 97 |
| As _{0.1} | <1.30 | 1.02 |
| USP Tailing Factor | <1.20 | 1.05 |

Selectivity

| | | |
|-------------------------------------|---------------|-------|
| k'(Phenanthrene) | 4.08 - 4.42 | 4.35 |
| k'(Desipramine)/ k'(Phenanthrene) | 0.120 - 0.130 | 0.125 |
| k'(Doxepin)/ k'(Phenanthrene) | 0.340 - 0.368 | 0.352 |
| k'(Imipramine)/ k'(Phenanthrene) | 0.496 - 0.538 | 0.517 |
| k'(Amitriptyline)/ k'(Phenanthrene) | 0.654 - 0.708 | 0.678 |

Sample: 1) Uracil 2) Desipramine 3) Doxepin 4) Imipramine 5) Amitriptyline 6) Phenanthrene, Mobile Phase: 80:20 MeOH/25mM KH₂PO₄ (pH6.0)
Column Dimensions: 250 x 4.6mm i.d., Flow Rate: 1.0ml/min Temperature: 22°C, Wavelength: 215nm



Benefits of Validated Columns

Method development and validation in regulated industries, including pharmaceutical and environmental, rely on the use of multiple columns and stationary phase batches. Production changes or batch differences in the HPLC column will have an adverse effect on method ruggedness. The use of fully validated ACE HPLC columns will ensure excellent column performance is maintained over the lifetime of an analytical method.

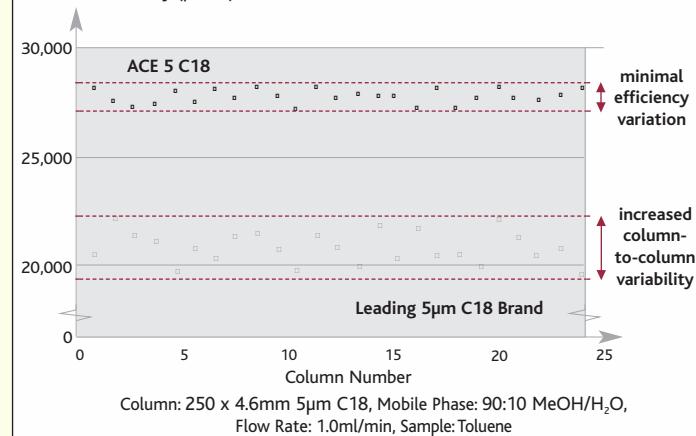
#1 Improved Column-to-Column Reproducibility

ACE ultra-inert columns consistently exhibit higher efficiencies and greater column-to-column reproducibility than leading competitor columns characterized by single validation procedures. Figure 7a demonstrates the increased performance and reduced column-to-column variation obtained with ACE columns compared to a leading brand. This indicates a higher degree of packing integrity with the ACE columns, which can in turn lead to increased column lifetime.

**SUPERIOR PERFORMANCE
AND PEAK SHAPE
- GUARANTEED**

Figure 7a. Comparison of Column-to-Column Reproducibility

Column Efficiency (plates)



#2 Improved Batch-to-Batch Reproducibility

Variations between different batches of stationary phase are the most common cause of customer concern. ACE stationary phases virtually eliminate the unpredictable negative effects of silanols on HPLC separations, by maintaining a rigid control of the manufacturing process and establishing tight specifications for purity, selectivity, retention, efficiency and asymmetry. Therefore, absolute batch-to-batch reproducibility is guaranteed with ACE ultra-inert HPLC columns. Figure 7b demonstrates the excellent batch reproducibility for both basic and acidic molecules.

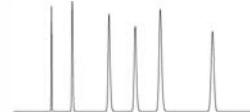
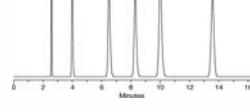
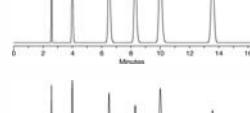
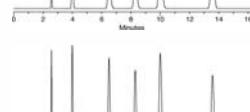
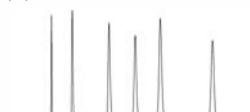
**SUPERIOR
REPRODUCIBILITY
- GUARANTEED**



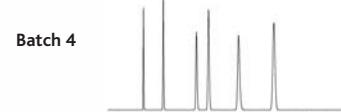
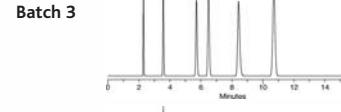
ACE® Stationary Phases
Virtually Eliminate the Negative
Effects of Silanols on HPLC
Separations

Figure 7b. ACE Batch-to-Batch Reproducibility

(a) Basic molecules



(b) Acidic molecules



Column: ACE 5 C18, 250 x 4.6mm
Mobile Phase: 80:20 MeOH/0.025 KH₂PO₄(pH 6.0)
Flow Rate: 1.0ml/min Sample: 1.) Uracil
2.) Desipramine 3.) Doxepin 4.) Imipramine
5.) Amitriptyline 6.) Phenanthrene

Column: ACE 5 C18, 250 x 4.6mm
Mobile Phase: 35:65 MeCN/0.1% TFA in H₂O
Flow Rate: 1.0ml/min Sample: 1.) Uracil
2.) 4-Hydroxybenzoic acid 3.) Acetylsalicylic acid
4.) Benzolic acid 5.) 2-Hydroxybenzoic acid 6.) Ethyl paraben