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12 www.ace-hplc.com The solvents used as mobile phases in HILIC are similar to those used in RPLC. As discussed in Section 2.0, the mobile phase conditions are key contributors to the various HILIC retention mechanisms. By modifying the proportions of the organic and aqueous components in the eluent, analyte retention can be altered.

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ACE. Step-by-Step HILIC Method Development Protocol. Step-by-Step Rational Method Development. Introduction: HILIC stationary phase and mobile . phase pH are two of the most powerful parameters for altering HILIC selectivity. Assessing these two critical parameters is therefore the optimum starting point for method development. The recommended

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ACE HILIC Method Development Kit For Developing Reproducible UHPLC / HPLC HILIC Separations Hydrophilic Interaction Liquid Chromatography (HILIC) uses similar eluents to Reversed-Phase Liquid Chromatography (RPLC) and provides a suitable retention and separation alternative for polar to very polar compounds.

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For 1/32" Microbore HPLC column connections up to 6000 psi, PEEK™ 1/32" (6-40 thread) fingertight fittings (part number ACE-MC3210, 10 pack) are recommended. For 1/16" UHPLC column connections up to 25000 psi, reusable 1/16" fittings (part number EXL-CC10, 10 pack) are recommended.

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The high stability of ACE C18, C8, C4, CN and Phenyl phases was demonstrated: after three months usage at pH 1.8, no retention loss was observed with any ACE phase. Even cyano bonded columns (regarded to be most vulnerable to hydrolysis) showed excellent resistance. pH 11.0 Robustness

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For more information contact your local ACE distributor or visit [www.ace-hplc.com](#) or email: [info@ace-hplc.com](#) Introduction In order to obtain stable and reproducible retention times, it is essential to fully equilibrate HILIC columns with mobile phase prior to analysis to ensure a stable adsorbed water layer exists at the stationary phase surface.

[Column Equilibration in HILIC Mode - HPLC](#)

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Hydrophilic Interaction Liquid Chromatography (HILIC) is less well understood practically and mechanistically, meaning that the method development process tends to be less systematic and well defined. This ACE Knowledge Note details a simple, rationally designed protocol for HILIC method development using the three ACE HILIC phases.

[A Simple Step-by-Step Protocol for HILIC Method ... - HPLC](#)

by Alan P McKeown, Advanced Chromatography Technologies Ltd, 1 Berry Street, Aberdeen, Scotland AB25 1HF, UK amckeown@ace-hplc.com Hydrophilic interaction chromatography (HILIC) separations are gaining popularity across many industry sectors and application areas.

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Originally, HILIC was applied to carbohydrate and peptide analysis in proteomics and glycomics (Zhu et al., 1991, Feste and Khan, 1992, Churms, 1996, Lin and Lee, 1998, Stregle, 1998).Later, HILIC has gradually been used for separations of drugs, toxins, plant extracts, and other small polar compounds in clinical, food, and pharmaceutical analysis (Tolstikov and Fiehn, 2002, Garbis et al., 2001).