Differences Between HILIC Columns and ANP Columns - Tech Information

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Introduction: When separating highly polar analytes, two approaches are most commonly considered: Hydrophilic Interaction Liquid Chromatography (HILIC) and Aqueous Normal Phase (ANP).

While both often begin with high-organic mobile phases and use “inverse” gradients, they do not rely on the same retention mechanism and they behave very differently in day-to-day work. Understanding these differences helps you choose the right column and method—especially for LC-MS workflows.

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1) Retention Mechanism

- HILIC: Retention is dominated by partitioning into a semi-permanent, water-rich layer that forms on the surface of ordinary silica or polar phases. This hydration shell can vary with temperature, buffer strength, and recent gradient history, which is a frequent source of drift.
- ANP (on Cogent TYPE-C™ silica hydride): The silica-hydride surface does not support a persistent water layer, leading to a different retention process that behaves more like local solvent displacement rather than classic HILIC partitioning. This underpins faster mass transfer, less drift, and more reproducible retention.

  For a foundational perspective distinguishing HILIC vs. ANP mechanisms, see the comparative discussion by Pesek & Matyska.

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2) **Equilibration and Throughput**

- HILIC: Because the water layer must rebuild and stabilize after gradients, equilibration is slow. Labs often experience significant downtime between runs, which limits throughput—particularly in gradient methods.
- ANP: On TYPE-C™ columns, re-equilibration is rapid (commonly 3–5 column volumes), supporting fast sequences and even ballistic gradients (sub-minute to ~5-minute methods) with excellent run-to-run precision.

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3) **Salt Requirements & LC-MS Compatibility**

- HILIC: Many methods require high, sometimes 50–100 mM, salt to tune retention—effective for selectivity but problematic for LC-MS (ion suppression, fouling) and for preparative recovery.
- ANP: Typically operates with ≤15 mM salts (often 5–15 mM) and can avoid non-volatile buffers, which improves MS signal stability, reduces instrument maintenance, and speeds cleanup in prep workflows.

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4) **Precision, Robustness, and Column Lifetime**

- HILIC: The variable hydration shell can cause retention-time variability and reproducibility issues; some users report unexpected column failures during long sequences.
- ANP: TYPE-C™ silica hydride phases exhibit extraordinary retention-time precision and robustness, with markedly longer lifetimes reported vs. typical HILIC phases under comparable use.

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5) **Selectivity & Analyte Scope**

1. HILIC: Excellent for many polar/ionic analytes but can struggle with certain strong acids/bases because of interactions with ionized silanols on silica, especially as pH rises.
2. ANP: Retains polar compounds and can retain some non-polar species on the same column (dual-mode capability), expanding selectivity space and simplifying method development. Strong sulfonic acids and other difficult polar analytes are often retained better and more reproducibly than in HILIC.

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6) **Ballistic Gradients & High-Speed Methods**

If your workflow demands very fast LC-MS cycles, ANP on TYPE-C™ columns is typically the safer, more precise choice, because the stationary phase does not rely on a slowly regenerating water layer. This allows aggressive gradients and short columns while maintaining precision.

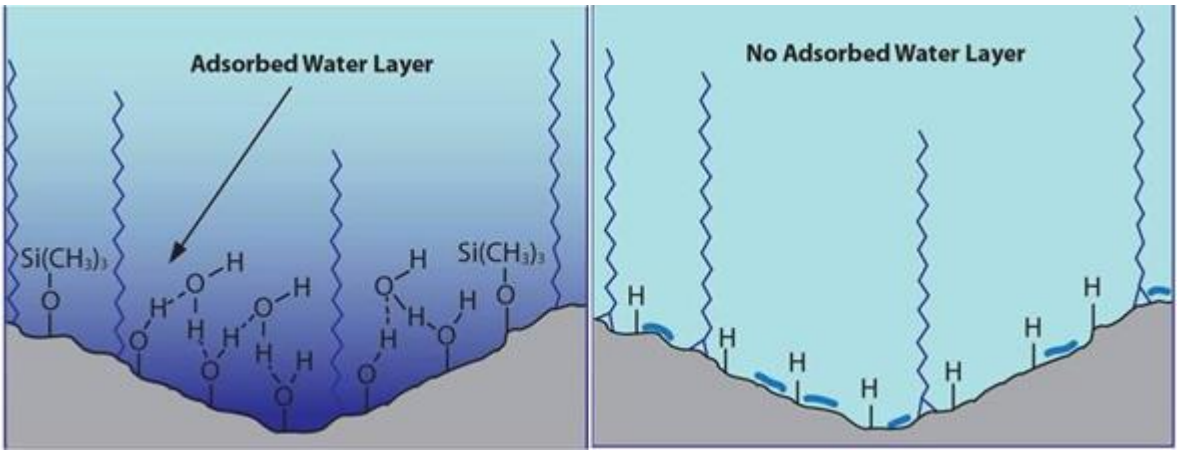
Practical Takeaways

- For high-throughput LC-MS with polar analytes: Start with ANP on Cogent TYPE-C™ columns; expect fast equilibration, low salt, and stable precision.
- If you must run HILIC: plan for longer equilibration, careful control of temperature and salts, and more frequent verification of retention-time stability.

🟢Summary

ANP vs. HILIC — what's the real difference? HILIC relies on a hydration shell on conventional silica that drives partitioning-based retention; this layer changes with conditions and slows equilibration, which can reduce throughput and precision—often with high salt levels that complicate LC-MS. ANP on Cogent TYPE-C™ silica hydride uses a different surface (no persistent water layer), enabling fast re-equilibration (3–5 CV), low-salt, LC-MS-friendly operation, excellent retention-time precision, and longer column life.

ANP can also retain polar and some non-polar analytes on the same column, streamlining method development and delivering reliable performance for modern regulated labs.



HILIC Phases have a Water Shell Cogent TYPE-C Silica has no Water Shell

See also: Comparison of the efficiency in ANP vs. HILIC.
See also: Wikipedia definition of ANP
See also: ANP v. HILIC Advantages

A very popular journal article on this subject:

Journal of Chromatography A, E. Barto, A. Felinger & P. Jandera Investigation of the temperature dependence of water adsorption on silica-based stationary phases in hydrophilic interaction liquid chromatography, 2017, Volume 1489 pages 143-149

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