MICROSOLV

Tanaka Plots Do Not Apply to Columns Used with Aqueous Normal Phase Methods - Tech Information

DATE: 14-FEBRUARY-2017 Last Updated: 9-FEBRUARY-2026

Do Tanaka Plots Apply to Columns Used with Aqueous Normal Phase (ANP) Methods?

Short answer: No.

While Tanaka plots are widely used to compare and characterize reversed-phase (RP) columns, they do not translate to columns operated in Aqueous Normal Phase (ANP) or HILIC modes.

The underlying tests and derived parameters of the Tanaka protocol were designed and validated in RP conditions, so they cannot reliably describe—or predict—column behavior in ANP/HILIC mechanisms.

What is a Tanaka plot (and why it's RP-specific)?

The Tanaka test set, developed by Nobuo Tanaka et al., prescribes separations of a defined panel of solutes under specific conditions. Each pair or subset of analytes is mapped to a stationary-phase characteristic (e.g., shape selectivity is commonly inferred from the relative separation of triphenylene vs. o-terphenyl, which possess similar hydrophobicity but different molecular shapes). From these RP measurements, a multi-dimensional "plot" is constructed to compare different RP columns. Critically, all Tanaka tests are run in RP mode, making their interpretation inherently RP-centric.

Because the solvation environment, stationary-phase surface state, and dominant retention forces in ANP/HILIC are fundamentally different from RP, Tanaka outputs do not capture the variables that control retention and selectivity in these modes.

Why Tanaka metrics cannot be ported to ANP/HILIC

- Different retention mechanisms:
- RP retention is dominated by hydrophobic partitioning and residual secondary interactions; ANP/HILIC retention relies on high-organic, water-modulated surface interactions (including charge-mediated and adsorption phenomena). The RP-oriented Tanaka parameters therefore don't represent ANP/HILIC drivers of retention or elution order.
- Test solutes and conditions are RP-biased:
- The canonical pairs (e.g., triphenylene / o-terphenyl) probe shape selectivity under RP hydrophobicity constraints. In ANP/HILIC, polarity, ionization, buffer content, and water-layer dynamics dominate; RP-chosen probes cannot generalize these effects. **Method space is not comparable:**
- Tanaka protocols mandate fixed RP mobile phases and gradient/isocratic conditions. ANP/HILIC use high-organic starts, water ramps, and often volatile salts at low concentration;

comparing columns across such mechanistically different method spaces with RP metrics is not meaningful.

What to use instead when evaluating columns for ANP (and HILIC-like) work

1. ANP-focused scouting gradients
   Evaluate columns with high-organic starts (e.g., ACN-rich) and water increases, including low-level volatile modifiers as needed. Compare retention windows, peak shapes, and elution orders for your real analyte classes rather than relying on RP surrogates.

2. Mechanism-relevant probe panels
   Build small probe sets that span polarity and ionization states (acids/bases/zwitterions/neutral polyols). Track response to water %, buffer identity/concentration, and pH (where appropriate) to expose ANP-relevant selectivity differences among columns.

3. Reproducibility & equilibration checks under ANP conditions
   Since ANP methods emphasize fast re-equilibration and run-to-run precision, quantify equilibration volumes, carryover, and baseline stability in your ANP gradient design—metrics that matter operationally but are outside Tanaka scope.

4. Application-driven comparison
   Selectivity in ANP is analyte- and surface-dependent; prioritize real sample matrices and method-transfer robustness across days. Use practical endpoints (resolution of critical pairs, LOD/LOQ behavior, MS cleanliness) rather than RP plot coordinates.

Bottom line

Tanaka plots are excellent for RP column benchmarking—but they are not a universal descriptor of column performance.

For ANP (and HILIC-like) methods, use ANP-appropriate scouting strategies and probe panels to characterize selectivity, retention control, and equilibration behavior. This approach yields actionable insights that directly reflect ANP mechanisms rather than RP surrogates

MicroSolv Technology Corporation
9158 Industrial Blvd. NE, Leland, NC 28451
Tel: (732) 380-8900
Fax: (910) 769-9435
Email: customers@mtc-usa.com
Website: www.mtc-usa.com