Kinetex™
Core-Shell Technology

Kinetex HPLC/UHPLC Columns
Tips for Care and Use

General Information

Thank you for selecting a Phenomenex Kinetex column. Every Phenomenex-manufactured column is individually QC tested and every column is supplied with a Certificate of Quality Assurance (CQA) which indicates testing conditions, operating parameters, and column details. The column details, including specifications and performance test results, should be entered into your information management system for easy tracking and reference. CQAs are available online at www.phenomenex.com/QD. You will need your column serial number to download the CQA.

If you have questions about your column's quality documentation contact: www.phenomenex.com/chat. The values mentioned in this column care are for reference and can differ significantly dependent upon LC system, running parameters, and sample analytes/matrix.

Inspection

Upon receipt of column:

1. Verify that the column is the one you ordered (i.e. dimension, particle size, media).
2. Check the column for any physical damage potentially caused during shipment.
3. Test the column immediately with a QC standard to verify performance.
4. Record the results of your test in your column information management system.

Column Characteristics

Kinetex Phases	Description	Shipping Solvent†	Particle Sizes (µm)	Pore Size (Å)	Surface Area (m2/g)	Carbon-Load (%)	pH Stability	Reversed Phase	Normal Phase	HILIC	100% Aqueous Stable
PS C18	Core-shell with bonded C18 and positive surface charge	Acetonitrile/Water (50:50)	2.6	100	200	9	1.5-8.5*	•			•
Polar C18	Core-shell with polymerically bonded C18 phase	Acetonitrile/Water (50:50)	2.6	100	200	9	1.5-8.5*	•			•
C18	Core-shell with bonded C18 phase	Acetonitrile/Water (50:50)	1.3, 1.7, 2.6, 5	100	200	12	1.5-8.5*	•			
EVO C18	Organo-silica core-shell with bonded C18 phase, resistant to extreme pHs	Acetonitrile/Water (45:55)	1.7, 2.6, 5	100	200	11	1-12	•			•
XB-C18	Core-shell with bonded C18 phase protective butyl side chains.	Acetonitrile/Water (50:50)	1.7, 2.6, 3.5, 5	100	200	10	1.5-8.5*	•			
C8	Core-shell with bonded C8 phase	Acetonitrile/Water (45:55)	1.7, 2.6, 5	100	200	8	1.5-8.5*	•			
Biphenyl	Core-shell with bonded biphenyl phase	Acetonitrile/Water (45:55)	1.7, 2.6, 5	100	200	11	1.5-8.5*	•			•
Phenyl-Hexyl	Core-shell with bonded phenyl-hexyl phase	Acetonitrile/Water (45:55)	1.7, 2.6, 5	100	200	11	1.5-8.5*	•			
F5	Core-shell with bonded pentafluorophenylpropyl phase	Acetonitrile/Water (40:60)	1.7, 2.6, 5	100	200	9	1.5-8.5*	•		•	•
HILIC	Unbonded core-shell silica	Acetonitrile/100 mM Ammonium Formate (93:7)	1.7, 2.6, 5	100	200	0	2.0-7.5		•	•	

† The shipping solvent for Kinetex columns is the same as the column QC test solvent which is listed in the individual columns CQA (Certificate of Quality Assurance) provided online.
* pH stability under gradient conditions. pH stability is 1.5-10 under isocratic conditions.

Typical Flow Rate, Backpressure, Temperature:

Here are some typical values for common dimensions of Kinetex HPLC and UHPLC columns. These numbers are not absolute values and can differ based on the LC system, running parameters, and sample analytes/matrix. The values below have been created using a solvent system of Acetonitrile and Water.

Particle Size (µm)	Internal Diameter (mm)	Typical Flow (mL/min)	Typical Pressure (PSI)		
			50 mm	150 mm	250 mm
1.3	2.1	0.5	10500	—	—
1.7	1.0	0.1	2600	—	—
1.7	2.1	0.5	5300	7300	—
1.7	3.0	0.8	7000	—	—
2.6	1.0	0.1	1700	4300	—
2.6	2.1	0.5	3000	6800	—
2.6	3.0	0.8	2300	5900	—
2.6	4.6	1.8	2800	5300	6900
3.5	4.6	1.5	—	2600	—
5	2.1	0.3	580	1200	—
5	3.0	0.5	620	1050	—
5	4.6	1.3	1100	1600	1700
5	10	5	—	2100	3000
5	21.2	25	630	1300	2300
5	30	50	670	1600	2300
5	50	80	—	1000	—

Note: Backpressures from H2O/Methanol mixture for same column dimension and flow rate will be approximately 40% higher than H2O/Acetonitrile mixture.

Maximum Backpressure:

- All analytical columns IDs 4.6 mm ID and under, pressure > 15,000 psi (1,034 bar) may compromise column longevity
- For 10 mm ID columns, pressure > 6000 psi (413 bar) may compromise column longevity.
- For 21.2, 30, and 50 mm ID columns, pressure > 3500 psi (241 bar) may compromise column longevity.

Maximum Temperature:

- Suggested max temperature for Kinetex LC columns is 60 °C, however temperature limits are dependent on your running parameters. Running at a pH greater than 8 at 60 °C will compromise column lifetime.
- Continuous use of Kinetex columns at the maximum temperature limit may compromise column longevity.

Mobile Phase Compatibility

When using any HPLC/UHPLC column, be sure to only use HPLC grade solvents and materials while also avoiding immiscible solvent/buffer combinations. Additionally, the use of solvent filtration is highly recommended to remove trace impurities from your mobile phase of choice. The following Kinetex™ phases are stable in 100 % aqueous conditions: Polar C18, EVO C18, Biphenyl, and F5. With all Kinetex columns please ensure that mobile phase pH does not exceed individual stationary phase limits. See chart in column characteristics section for individual Kinetex stationary phase pH limits.

Column Installation

Initial setup of your LC system is very important to ensure column performance:

Ensure that your LC system is ready:

1. Seals, lines, injector clean
2. Lines primed (no dry lines or bubbles)
3. Steady baseline
4. Consistent pressures

Flush LC system pump and line with mobile phase (HPLC grade and miscible with solvents that column is shipped in).

Mobile phase starting conditions checklist:

1. Ensure that the HPLC grade mobile phase is well mixed, filtered, and degassed prior to use.
2. Ensure that column shipping solvent, remaining solvent in LC system, and mobile phase solvents are miscible.

Set flow rate to 0.1 mL/min (for 2.1-4.6 mm ID) and install the column making sure that the arrow is in the direction of flow. Then increase the flow rate to 0.2 mL/min (2.1 mm ID) or 1.0 mL/min (4.6 mm ID) for 5-10 minutes. Collect solvent in a small beaker.

Stop flow and wipe the outlet end of column to remove any particulates before connecting to a detector.

Install fitting/tubing into outlet end and run minimum 10 column volumes at low flow (~0.2 mL/min) while monitoring the backpressure.

1. A steady pressure should indicate a constant flow while pressure fluctuation will indicate air in the system.
2. Wide fluctuations in pressure may shock and damage the column so it's important to monitor the pressure.

Monitor pressure as well as the signal from the detector, when both are steady, the column is ready for use.

Testing Column Performance

When testing column performance, please use the manufacturer approved test mix.

Reversed Phase	
Name:	Reversed Phase 2 Test Mix
Part No.:	AL0-3045
Contents:	Uracil, Acetophenone, Toluene, Naphthalene
Solvent:	Acetonitrile / Water (65:35 v/v)
Detection:	UV @ 254 nm
Injection Vol.:	Depends on dimensions

HILIC	
Name:	HILIC Phase Test Mix
Part No.:	AL0-8317
Contents:	Toluene, Uracil, Cytosine
Solvent:	Acetonitrile (containing toluene) / Water (85:15)
Detection:	UV @ 254 nm
Injection Vol.:	Depends on dimensions

Column Cleaning

Reversed Phase:

- Clean with a gradient that is closest to the last solvent system on the system:
- For example, if the last injection ended with Buffer/Acetonitrile (75:25), it's more appropriate to start with 95:5 Water/ Acetonitrile and then move step by step as needed to increase organic content (i.e. 75:25 Water/Acetonitrileà 50:50 Water/ Acetonitrileà5:95 Water/Acetonitrile).
- For hydrophobic or oily materials, try flushing with Isopropyl Alcohol (IPA), after the column has been flushed with Acetonitrile. When using IPA, use a low flow to prevent higher backpressures due to higher solvent viscosity.
- For materials that are very hydrophobic, try Tetrahydrofuran (THF) instead.

HILIC:

- To remove buffer, rinse with at least 10 column volumes of 95:5 Water/Acetonitrile. Repeat with 95:5 100 mM Ammonium Acetate (pH 5.8)/Acetonitrile. Then finish cleaning by flushing the column with 95:5 Water/Acetonitrile.

Tips:

- When cleaning, set your flow rate lower than that of your method flow rate, especially when attempting to clean using methanol or IPA.
- Cleaning for a longer period of time is more often beneficial than adding more cycles.
- Working with very high amounts of THF is not recommended especially if the system has PEEK tubing. Cleaning with THF is permissible if the tubing is metal.
- Reverse flushing the column (against the direction of the arrow on the column) is fine, but please use low flow. Below are suggested flow rates based on column ID for reverse flushing:
  0.1 mL/min (2.1 mm ID)
  0.3 mL/min (3.0 mm ID)
  0.5 mL/min (4.6 mm ID)

Column Regeneration

Reversed Phase

- Apply the same gradient flush as in the cleaning above, overnight at low flow.
- Reverse flushing is acceptable.

Normal Phase

- For water removal, flush with:

1. 30 mL 2.5 % 2,2 Dimethoxypropane and 2.5 % Glacial Acetic Acid in Hexane.
2. Then flush the column with cleaning method from above, overnight at a low flow rate.

Column Storage

It is very important to make sure that your column is clean before storage. This includes the removal of buffers, salts, sample, and ion-pairing agents. The recommended storage conditions are:

- Reversed phase: Acetonitrile/Water (65:35 v/v), Methanol can be used in place of acetonitrile.
- Normal phase: 100 % Hexane or IPA.
- HILIC: Acetonitrile/Water (80:20 v/v).
- Please store with HPLC grade or above solvents only.

Tips for Extending Column Lifetime

- Utilize sample preparation techniques such as solid phase extraction (Strata™-X SPE products) or accessories (Phenex™ Syringe Filters) to minimize the injection of unwanted contaminants onto your system and column.
- Use the correct guard column or guard cartridge system (SecurityGuard™) to help remove particulates before they foul your column.
- Do not overload your column. Inject suitable sample concentrations and volumes.
- Work in the appropriate separation mode for the column. Please see Column Characteristics chart for typical modes each stationary phase is used for.
- Store your column in appropriate solvent(s).
- Solvent switch correctly by slowly acclimating the phase from one miscible solvent to the other at a low flow: 0.1 mL/min for 2.1 mm ID and 0.5 mL/min for 4.6 mm ID.

Column Warranties

Phenomenex HPLC columns are warranted to meet the stated performance and quality and to be free of defects in material and workmanship. If you are unsatisfied for any reason, please give your Phenomenex Technical Representative a call. We'll do our best to solve the problem to your satisfaction. Should it become necessary to return the column, a Return Authorization Number must be obtained from Phenomenex first.

Disclaimers

New columns should be tested with the manufacturers recommended test mix, and previously used columns should be tested with the same or a suitable test mix for the analysis. Remember to re-equilibrate the system when changing solvents. Never change from one solvent to another which is immiscible, without going through an intermediate solvent which is miscible with both. This will damage the column. Never change to (or from) a buffer/salt solution where the buffer/salt is not soluble in the second solvent. Again this will damage the column. Never attempt to remove the column end fittings. This will void the warranty.

Column Shock

Handle columns with care. Do not drop or create physical shock. Do not start pump at high flow rates, instead ramp up gradually over a few minutes. Set your pump pressure limit to protect the column in event of blockage. This can create voids which will detrimentally affect the column's performance.

Column Questions and Support

For any additional questions visit:
www.phenomenex.com/chat

For more information on Kinetex UHPLC, HPLC, and Preparative columns, please visit
www.phenomenex.com/Kinetex

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