



# Clarity HPLC/UHPLC Columns

## Tips for Care and Use

### General Information

Each Clarity column manufactured by Phenomenex is individually prepared and tested. 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. Electronic copies of your column's quality documentation can also be acquired at: [www.phenomenex.com/mysupport](http://www.phenomenex.com/mysupport).

### Inspection

Upon receipt of column, please verify that the column you received is the one you ordered (i.e. dimension, particle size, media). Additionally, please check the column for any physical damage potentially caused during shipment. Test the column immediately to verify performance and record the result of your test in your column information management system.

### Column Characteristics and Operating Recommendations

Phase	Particle Size (um)	Base Material	Pore Size (A)	Surface Area (m <sup>2</sup> /g)	Carbon Load (%)	pH Range	Temperature Range/ Limit (deg.C)	Typical Pressure (psi)	Pressure Limit (psi)	Typical Flow Rate (mL/min)	Max Flow Rate (mL/min)	Shipping Solvent	Storage Conditions
Oligo-RP	3, 5, 10	Organo Silica Fully Porous	110	375	14	1-12	60	800 (10 um)	5000 (10 um)	0.5 (2.0 mm ID)	1.0 (2.0 mm ID)	ACN:Water 65:35 *ratios may vary for different parts	(50:50) v/v ACN: HPLC Water Or (50:50) v/v MeOH: HPLC grade Water
								2000 (5.0 um)	5000 (5.0 um)	1.0 (4.6 mm ID)	2.0 (4.6 mm ID)		
								3500 (3.0 um)	5000 (3.0 um)	5.0 (10.0 mm ID)	20.0 (10.0 mm ID)		
Oligo-MS	1.7, 2.6, 5	Core-Shell	100	200	12	1.5-10	60	2000 (5.0 um)	5000 (5.0 um)	0.5 (2.0 mm ID)	1.0 (2.0 mm ID)	ACN:Water 50:50	(50:50) v/v ACN: HPLC Water Or (50:50) v/v MeOH: HPLC grade Water
								4000 (2.6 um)	8700 (2.6 um)	1.0 (4.6 mm ID)	2.0 (4.6 mm ID)		
								6000 (1.7 um)	15000 (1.7 um)	1.0 (4.6 mm ID)	2.0 (4.6 mm ID)		
Oligo-XT	1.7, 2.6, 5	Organo Silica Core-Shell	100	200	11	1-12	60	2000 (5.0 um)	5000 (5.0 um)	0.5 (2.0 mm ID)	1.0 (2.0 mm ID)	ACN:Water 60:40	(50:50) v/v ACN: HPLC Water Or (50:50) v/v MeOH: HPLC grade Water
								4000 (2.6 um)	8700 (2.6 um)	1.0 (4.6 mm ID)	2.0 (4.6 mm ID)		
								6000 (1.7 um)	15000 (1.7 um)	5.0 (10.0 mm ID)	20.0 (10.0 mm ID)		
Oligo-WAX	10	Fully Porous	360	-	-	1-11	60	3500	5000	1.0 (4.6 mm ID)	2.0 (4.6 mm ID)	1 % ACN:Hexane	(50:50) v/v ACN: HPLC Water Or (50:50) v/v MeOH: HPLC grade Water
										5.0 (10.0 mm ID)	20.0 (10.0 mm ID)		
										10.0 (21.2 mm ID)	200.0 (21.2 mm ID)		
Oligo-SAX	5	Non-Porous Polymer	-	-	-	2.5-12.4	5-85 At >pH 11, 45	2000	5000	1.0-1.6	2.0	20 mM Tris pH 8.0 + 1.25 M NaCl (54:46)	20 mM Tris + 1.0 M NaCl, pH8

\*Typical pressure is taken under acetonitrile and water conditions; backpressure will depend on method running conditions.



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[www.phenomenex.com/mysupport](http://www.phenomenex.com/mysupport)

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## 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 the LC system (pump and line) with HPLC grade mobile phase, making sure all solvents in the system and column are miscible.

Mobile phase starting conditions check list:

1. Ensure that 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.

Column Installation:

1. Set the flow rate to 0.1 mL/min, oven temperature ambient
2. Install the column in the direction of flow and condition the column for 10 minutes going to 1.0 mL/min.
3. Stop flow and wipe outlet end of column to remove any particulates before connecting to detector.
4. Install the column fully and flush for at least 10 column volumes.
5. Ramp flow to method flow and set oven temperature.
6. Monitor for pressure and baseline. When both are steady, the column is ready for use.
  - a. A steady pressure should indicate a constant flow while pressure fluctuation will indicate air in the system.
  - b. Wide fluctuations in pressure may shock and damage the column so it's important to observe the pressure.

### Tips:

Always recondition the column when solvent changing.

When working with ion exchange columns, which use a high salt mobile phase, never change to (or from) a buffer/salt solution where the buffer/salt is soluble in the second solvent.

## Column Cleaning

### Clarity Oligo-RP, Oligo-MS and Oligo-XT

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/Acetonitrile 50:50 Water/Acetonitrile 5:95 Water/Acetonitrile).

» For hydrophobic or oily materials, try flushing with Methylene Chloride or IPA. When using IPA, ensure use of a low flow to prevent higher backpressures due to higher solvent viscosity.

» For materials that are very hydrophobic, try THF instead.

### Tips:

If an increase in pressure is observed, you can try reverse flushing the column with the reduced flow rates indicated below:

- 0.1 mL/min (2.1 mm diameter column)
- 0.3 mL/min (3.0 mm diameter column)
- 0.5 mL/min (4.6 mm diameter column)

Flushing with THF requires stainless steel lines.

Removing any endfitting or frits will void the warranty.

### Clarity Oligo-WAX

Rinse the column with 10 column volumes of water before rinsing with 10 column volumes of:

1. Isopropyl Alcohol
2. Methylene Chloride
3. Isopropyl Alcohol
4. Mobile Phase

### Tips:

If an increase in pressure is observed, reverse flushing is fine but to prevent shocking the column, reduce the flow rate to half the method flow.

Do not remove any end fitting and/or frits as this will create a void.

### Clarity Oligo-SAX

To clean the column of ionic components, rinse with 15-20 column volumes of the following:

1. Deionized Water
2. 0.4 M NaClO<sub>4</sub>, 1 mM HCl and 30 % acetonitrile
3. Deionized Water
4. Mobile Phase

To clean the column of hydrophobic (non-ionic) components, rinse the column with 15-20 columns of the following:

1. Deionized Water (note this is critical to rid the column and system of salts)
2. 100 % ACN
3. Deionized Water
4. Mobile Phase

**Tips:**

If an increase in pressure is observed, reverse flushing is fine but to prevent shocking the column, reduce the flow rate to half the method flow.

Do not remove any end fitting and/or frits as this will create a void.

## Column Regeneration

### Clarity Oligo-RP, Oligo-MS and Oligo-XT

Rinse with:

1. 95:5 Acetonitrile/Water
2. Isopropanol
3. 95:5 Acetonitrile/Water

### Clarity Oligo-WAX

Flush with 5 column volumes of:

1. HPLC grade water
2. 95:5 Water/Acetonitrile
3. 85:15 Water/Acetonitrile

Flush with 10 column volumes of 10mM Ammonium Acetate pH 7.0.

Check column performance with manufacturer's standards

### Clarity Oligo-SAX

Run 2-3 column volumes of the highest concentration of eluent.

## Column Storage

- » Before storage, make sure the column has been flushed with HPLC grade solvents.
- » Store in HPLC grade or above solvents only. See Column Characteristics and Operating Recommendations Table on **page 1** for details of typical storage conditions.
- » Avoid jostling and dropping the column as this might cause column shock.

## Typical Loading Capacities

Column Type	ID (mm)	Approx. Dead Volume (mL)	Typical Flow Rate (mL)	Typical and (Max.) Injection Masses (mg)	Typical and (Max.) Injection Volumes (µL)
Capillary (Fused Silica)	0.32	0.0075	0.001 - 0.02	0.001 (0.01)	1 (10)
Microbore	1.0	0.07	0.02 - 0.1	0.01 (0.1)	5 (25)
Analytical	4.6	1.5	0.5 - 2.0	0.1 (2.5)	10 (200)
Semi-Prep	10.0	7.3	5.0 - 20	1.0 (25)	50 (1000)
Preparative	20.0	29.2	10 - 200	5.0 (500)	200 (5000)

## Tips for Extending Column Lifetime

- » Utilize sample preparation techniques such as solid phase extraction (Strata<sup>®</sup>-X SPE products and Clarity Sample Preparation Products) or accessories<sup>™</sup> (Phenex<sup>™</sup> Syringe Filters) to minimize the injection of unwanted contaminants onto your system and column.
- » Use the correct guard column or guard cartridge system (SecurityGuard<sup>™</sup>) to help remove particulates before they foul your column.
- » Do not overload your column. Inject suitable sample concentrations and volumes. See chart above: Typical Loading Capacities
- » Check for sample solubility in mobile phase. Use mobile phase as diluent where possible. Trace impurities can dramatically degrade column life. Filter all samples using a 0.45 µm or 0.2 µm porosity filter prior to injection.
- » Work in the appropriate separation mode for the column. Please see column characteristic 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 LC 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

If you have any additional questions, please reach out to our amazing technical team through:

Email: [support@phxtechnical.zendesk.com](mailto:support@phxtechnical.zendesk.com)

Live Chat: [www.phenomenex.com/chat](http://www.phenomenex.com/chat)

For more information on Clarity UHPLC, HPLC, and Preparative columns, please visit [www.phenomenex.com/Clarity](http://www.phenomenex.com/Clarity)

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