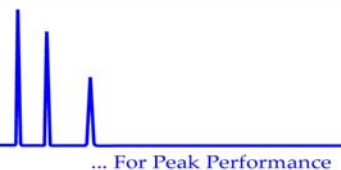




ZirChrom®



ZIRCHROM SEPARATIONS, INC. - Manufacturers of Zirconia- and Titania-Based HPLC Media.

Specialists in Ultra-Durable, High Efficiency HPLC Phases

RECOMMENDATIONS for USE, CLEANING and STORAGE of TITANIA-BASED HPLC COLUMNS

Sachtopore®-RP

Thank you for purchasing this titania-based reversed phase high performance liquid chromatographic column from ZirChrom Separations. We are sure you will be completely satisfied with its performance. In order to enjoy the tremendous benefits of its unique features compared to silica and polymer-based HPLC media, it is very important that you read the recommendations below. Please keep in mind that while this is a reversed-phase column the substrate is titania, not silica, and the non-polar stationary phase is cross-linked polyethylene deposited in the pores, not alkylsilanes bonded to the surface. If at any time you have a question about this product we invite you to visit our web site (<http://www.zirchrom.com>) where you will find additional information on the use of titania-based HPLC columns. In addition, our staff is always eager to help you with any aspect of using this column (1-866-STABLE-1).

Use:

1. Upon receipt, we suggest you duplicate the results on the enclosed chromatogram. You should be able to achieve a plate count of at least **100,000/meter** (*specification for 150 x 4.6 mm i.d. format*) for toluene under the operating conditions given on the chromatogram. Be sure to inject roughly the same amount of material as indicated in the chromatogram.
2. This column can be used with any common organic modifier (acetonitrile, methanol, tetrahydrofuran, isopropanol) but we find that **acetonitrile gives somewhat better plate counts**. Sachtopore®-RP is slightly less hydrophobic than common silica-based supports. For simple non-ionic compounds, we recommend that you use about **10-15% less organic modifier** to obtain roughly the same retention as you would on a typical C₈ or C₁₈ silica-based phase.
3. We very strongly advise that you use our columns at temperatures well above ambient. At a minimum, we urge you to set the column to **50 °C**, but preferably to the highest temperature consistent with the stability of your analyte. We routinely use Sachtopore®-RP columns at 75 °C and find them to be stable at 150 °C. We also recommend that you set the **flow rate to 3 ml/min**. at these super-ambient temperatures. **This will substantially increase the speed of analysis.** The backpressure of titania-based columns is remarkably low at room temperature and decreases substantially at 50 °C and higher.
4. When chromatographing basic compounds on any stationary phase a buffer must be used (**see ZirChrom Buffer Wizard at www.zirchrom.com**). We strongly advise the use of phosphate, and carboxylic acid (acetate, citrate, bicarbonate/carbonate) buffers not the amine buffers used with silica columns. Our first choice for cationic drugs is **10-25 mM pH 7.0 ammonium phosphate buffers**. However, this column is stable from pH 1-14 and you can use any buffer you like in this pH range. For LC-MS work, we strongly advise use of 10-100 mM ammonium hydroxide/ammonium fluoride buffers or ammonium hydroxide/ammonium formate buffers at pH 10-12.
5. When carboxylated molecules are chromatographed, we strongly advise the use of a buffer plus about 5 mM ammonium fluoride. Our favorite buffer for carboxylates is 10-25 mM ammonium phosphate plus 5 mM ammonium fluoride at pH 6-8. However, excellent results can be obtained at very low pH (pH < 1, without fluoride added due to HF formation) and up to quite high pH (>11).

6. To maximize the life of this ultra-durable column, we recommend the following precautions regarding day-to-day operation of the column.

- Always use a guard column.**
- Clean up samples before injection (either filtering to remove particulates or solid phase extraction techniques).
- Use HPLC grade solvents and filter all solutions before use.
- Minimize pressure surges.
- Use an in-line filter (0.5 micron) in front of column to catch large particulates.
- Always check the solubility of the buffer being used when mixing with organic mobile phases using an LC pump.

Caution:

Do not use PEEK tubing at temperatures above 100 °C, or with THF containing mobile phases.

Cleaning/Regeneration:

Carboxylic acids, fluoride and phosphate all adsorb strongly to titania-based columns. To fully remove these from the titania surface, or to remove any substance that may have fouled the column, use the following three-step cleaning protocol:

****IMPORTANT**** - During these steps you should remove your detector from the flow path to protect it from aggressive cleaning conditions.

1. Flush column with a mixture of 20/80 ACN/0.1 M sodium hydroxide or tetramethyl ammonium hydroxide for 50 column volumes at ambient temperature. Follow base wash with 10 column volumes of water at ambient temperature.
2. Flush column with a mixture of 20/80 ACN/0.1 M nitric acid for 50 column volumes at ambient temperature. Follow acid wash with 10 column volumes of water at ambient temperature.
3. Flush column with 100% organic solvent for 20 column volumes at ambient temperature. For Sachtopore®-RP, methanol, acetonitrile, isopropanol, and tetrahydrofuran are all adequate solvents.

Storage:

The Sachtopore®-RP column **should not be stored in phosphate buffer**. We strongly suggest flushing the column with 50/50 modifier-water prior to storage over night and with 0.1M ammonium hydroxide followed by 50/50 organic modifier-water before long-term storage.

A complete list of chromatography products offered by ZirChrom Separations:

HPLC Columns

Part #	Product Name	Chromatographic Mode and Uses
DB01	DiamondBond®-C18	C18 Modified Carbon Reversed-phase
EZ01	ZirChrom®-EZ	Deactivated Reversed-phase
MS01	ZirChrom®-MS	Deactivated Reversed-phase for LC/MS
ST01	ZirChrom®-SELECT	Deactivated Carbon Reversed-phase
TI01	Sachtopore-RP	Reversed-Phase (Titania)
TI02	Sachtopore-NP	Normal Phase (Titania)
ZR01	ZirChrom®-CARB	Carbon Reversed-phase
ZR02	ZirChrom®-PHASE	Normal Phase
ZR03	ZirChrom®-PBD	Reversed-Phase
ZR04	ZirChrom®-WCX	Weak Cation-Exchange
ZR05	ZirChrom®-WAX	Weak Anion-Exchange / Sugar Analysis
ZR06	ZirChrom®-SAX	Strong Anion-Exchange
ZR07	ZirChrom®-SHAX	Strong Hydrophilic Anion-Exchange
ZR08	ZirChrom®-PEZ	Cation-Exchange / Protein Analysis
ZR09	ZirChrom®-PS	Reversed-Phase

Specialty Products

Part #	Product Name	Description
AB01	Rhinophase®-AB	Pseudo-Affinity Phase for Antibodies
BW01	Advanced Buffer Wizard Software	Includes 50 Buffer Systems (CD Rom)
MK01	Ion-exchange Method Kit #1	Includes ZirChrom®-SAX, ZirChrom®-SHAX and ZirChrom®-WAX
MK02	Ion-exchange Method Kit #2	Includes ZirChrom®-SAX, ZirChrom®-WCX and ZirChrom®-PEZ
MK03	Reversed-phase Method Kit #1	Includes ZirChrom®-PBD, ZirChrom®-CARB and DiamondBond®-C18
MK04	Reversed-phase Method Kit #2	Includes ZirChrom®-EZ, ZirChrom®-CARB and ZirChrom®-PBD
NPZ	Nonporous Zirconia	0.5, 1, 2 or 3 Micron Particle Sizes

Note: All chromatography products are available in microbore, analytical, semi-preparative and preparative column formats.

