

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

ZirChrom®-MS

Thank you for purchasing this zirconia-based reversed phase high performance liquid chromatographic column from ZirChrom Separations. This product and/or its method of use is covered by one or more of the following patent(s): **US Patent No. 5,015,373, 5,108,597, 5,141,634, 5,205,929, 5,254,262, 7,897,798, Re: 34,910, 5,271,833, 5,346,619, 5,540,834, 6,846,410, 8,137,548** and foreign equivalents. Additional patents are pending in the United States. 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 zirconia, not silica, and the non-polar stationary phase is made by cross-linking polybutadiene over a surface that has been deactivated with a metal chelator deposited in the pores. 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 a complete list of 70+ technical articles in peer reviewed journals on zirconia-based HPLC. In addition, our staff is always eager to help you with any aspect of using this column (1-866-STABLE-1).

Use:

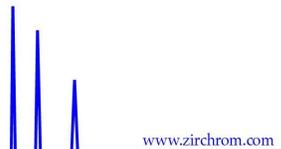
1. Upon receipt, we suggest you duplicate the results on the enclosed chromatogram. You should be able to achieve a plate count of at least **100,000 plates/meter** (*specification for 150 x 4.6 mm i.d. format*) for phenanthrene under the operating conditions given on the chromatogram. Be sure to inject 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). ZirChrom®-MS has almost the same hydrophobicity as common silica-based supports. For simple non-ionic compounds, we recommend that you use about **the same organic modifier proportion** 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 ZirChrom®-MS at temperatures below **50°C**.
4. When running ionizable compounds on any stationary phase a buffer must be used (**see ZirChrom Buffer Wizard at www.zirchrom.com**). Our first choice for both acidic and basic analytes is 10-25 mM ammonium acetate at pH 5.0. However, this column is stable from pH 1-10 and a variety of buffers may be employed in this pH range. At present, we **DO NOT** recommend the use of high concentrations (>100 mM) of fluoride, carboxylic acid (acetate, citrate, bicarbonate/carbonate) buffers, and salts. Also, please **DO NOT** exceed a concentration of 25 mM for phosphate buffers.
5. 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.

Cleaning/Regeneration:

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 acetonitrile/pH 10 ammonium hydroxide solution for 50 column volumes at ambient temperature. Follow base wash with 10 column volumes of water at ambient temperature.





- Flush column with a mixture of 20/80 acetonitrile/0.1 M nitric acid for 50 column volumes at ambient temperature. Follow acid wash with 10 column volumes of water at ambient temperature.
- Flush column with 100% organic solvent for 20 column volumes at ambient temperature. Methanol, acetonitrile, isopropanol, and tetrahydrofuran are all adequate solvents.

Storage:

The ZirChrom®-MS column **should not be stored in phosphate buffer**. We strongly suggest flushing the column with 10 column volumes of 50/50 organic modifier/water prior to storage over night and with 10 column volumes of pH 10 ammonium hydroxide solution followed by 10 column volumes of 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 |
|--------|------------------|--------------------------------------|
| DB01 | Diamondbond®-C18 | C18 Modified Carbon Reversed-phase |
| EZ01 | ZirChrom®-EZ | Deactivated Reversed-phase |
| MS01 | ZirChrom®-MS | Deactivated Reversed-phase for LC/MS |
| 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 |
| ZR06 | ZirChrom®-SAX | Strong Anion-exchange |
| ZR07 | ZirChrom®-SHAX | Strong Hydrophilic |
| ZR08 | ZirChrom®-PEZ | Cation-exchange |
| ZR09 | ZirChrom®-PS | Reversed-phase |

Specialty Products

| Part # | Product Name | Chromatographic Mode |
|--------|---------------------------------|--------------------------------------|
| AB01 | Rhinophase-AB | Pseudo-Affinity Phase for Antibodies |
| BW01 | Advanced Buffer Wizard Software | 50 buffer systems (CD-ROM) |
| MK01 | Ion-exchange Method Kit #1 | SAX, SHAX, WAX |
| MK02 | Ion-exchange Method Kit #2 | SAX, WCX, PEZ |
| MK03 | Reversed-phase Method Kit #1 | PBD, CARB, DB01 |
| MK04 | Reversed-phase Method Kit #2 | EZ, CARB, PBD |
| NPZ | Nonporous Zirconia | 0.5, 1, 2, or 3 µm |
| ZRC01 | ZirChrom®-Chiral(S)LEU | Pirkle Type chiral phase |
| ZRC02 | ZirChrom®-Chiral(R)NESA | Pirkle Type chiral phase |
| ZRC03 | ZirChrom®-Chiral(S)NESA | Pirkle Type chiral phase |
| ZRC04 | ZirChrom®-Chiral(S)PG | Pirkle Type chiral phase |
| ZRC05 | ZirChrom®-Chiral(R)PG | Pirkle Type chiral phase |
| ZRC06 | ZirChrom®-CelluloZe | Polysacchiride chiral phase |

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

