Sepax Hydrophilic Interaction Chromatography (HILIC) Separation – More Choices for Your Compounds

Innovative Surface Chemistry

Column Information

To solve the challenges of more and more highly polar pharmaceuticals and small biological molecules, Sepax developed a series of chemistries of weak acidic, neutral, and basic stationary phases for separating basic, neutral and acidic compounds of high polarity. Utilizing highest purity and enhanced mechanical stability silica and pure bonding reagents, Sepax HILIC bonded phases have been innovatively and specially designed to ensure maximum surface coverage, resulting in high stability of the stationary phases. The chemistry of monolayer formation is completely controlled that results in very reliable lot-to-lot and column-to-column reproducibility. The uniform, spherical Sepax HILIC particles have a nominal surface area of 300 m^2/g with a controlled pore size of 120 Å. Sepax HILIC columns are packed with a proprietary slurry technique to achieve uniform and stable packing bed density for maximum column efficiency.

Structure of HILIC Stationary Phases

With the chemical structures shown in Figure 1, Polar-100 and Polar-Diol are neutral, polar phases, while Polar-Diol is more polar than Polar-100. Polar-Silica is a weak acidic phase. Polar-Pyridine and Polar-Imidazole are basic phases, while Imidazole phase is more basic than Pyridine phase.

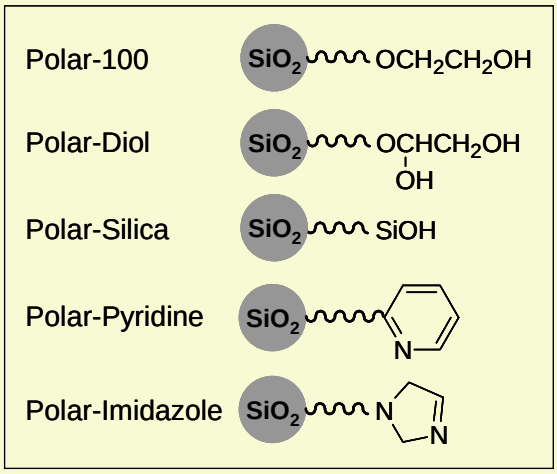

Figure 1. Chemical structures of HILIC phases.

Characteristics

- HILIC phases with unique chemistries of acidic, neutral, and basic surfaces
- Ultra-pure silica particles with controlled pore sizes
- Available in particle size of 1.8, 2.2, 3, 5, and 10 µm
- High chemical stability for low leaching
- Available columns with ID in the range of 75 µm to 30 mm and length from 1 to 30 cm
- Available packings from grams to multi-Kilogram
- pH stability: 1.5 - 8.0
- Suitable for separations of polar pharmaceuticals, peptides, amino acids, and other compounds

Separation of Nucleosides

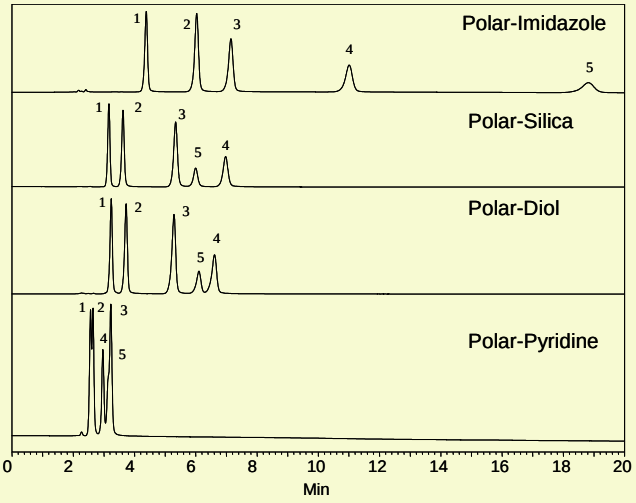

Figure 2. Separation of nucleosides by various HILIC columns (5µm, 4.6x150mm). Mobile phase: 90/10(V/V) = ACN/10mM ammonium acetate in H_2O. Flow rate: 1.0 mL/min. Ambient Temperature. UV 254 nm. Sample: 1. Uracil, 2. Adenosine, 3. Uridine, 4. Cytidine, 5.Guanosine.

Selection of Sepax HILIC columns

Phases	Standard particle size (µm)	Standard pore size (Å)	Other available particle size (µm)	Other available pore size* (Å)
Polar-100	3, 5	120	1.7, 2.2, 10	200, 300
Polar-Diol	3, 5	120	1.7, 2.2, 10	200, 300
Polar-Silica	3, 5	120	1.7, 2.2, 10	200, 300
Polar-Pyridine	3, 5	120	1.7, 2.2, 10	200, 300
Polar-Imidazole	3, 5	120	1.7, 2.2, 10	200, 300

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Sepax Polar HILIC 色谱柱使用和维护注意事项

请在色谱柱使用前仔细阅读本说明，并按要求进行操作，以保证色谱柱良好的重现性和耐用性。

色谱柱安装：

1. 色谱柱安装时，确认液路流向与色谱柱标签所示箭头方向一致。
2. 色谱柱接入仪器系统，接头松紧适中，系统开启后，请注意压力变化，确认与管路接头处无液体渗漏。

色谱柱使用和维护：

1. 请首先按照色谱柱出厂 QC 方法对色谱柱进行检测，理论塔板数和拖尾因子等应与 QC 报告相符。（因为仪器和实验条件的差异，实际检测结果与 QC 报告可能存在偏差，如偏差超过±20%请及时与厂家或色谱柱供应商联系）。
2. 请务必在说明书要求的柱温、压力和 pH 值范围内使用色谱柱，任何超出范围的色谱条件都可能导致色谱柱不可修复的损伤。
3. 当所用流动相中含有缓冲盐时，要保证缓冲盐在高比例的有机相中有较好的溶解性，且样品溶剂或初始洗脱条件中有机相的比例建议不低于 60%。
4. 建议采用流动相溶解样品，以避免溶剂效应的产生。此外，要保证待测样品与流动相有很好的溶解性，以免样品在流动相中析出而导致柱压升高和系统污染，若出现此情况，可对色谱柱进行低流速反向冲洗，以除去堵塞柱头的杂质。
5. 使用过程中，压力突然增加预示色谱柱入口端的筛板发生了堵塞。在这种情况下，建议将色谱柱反接后用适宜的溶剂进行冲洗。 如果柱压升高，请使用乙腈/水（50 ：50，v/v）来清洗去除极性污染物。若该冲洗不能解决问题，则使用乙腈/水（95:5，v/v）进行清洗。

色谱柱保存：

1. 如无特殊说明，每支色谱柱出厂时均保存在该色谱柱 QC 测试报告所述的溶剂中（报告底部）。建议的保存方法是该色谱柱存放的最佳方法。
2. 如长期不用，请将色谱柱从仪器系统中卸下，塞上堵头，以免柱头干涸，影响下次使用。一段时间后使用色谱柱如出现峰形异常，可用保存溶剂低流速冲洗色谱柱活化过夜。