

High Performance Packed Column for HPLC

Shim-pack

Bio-HIC column

INSTRUCTION MANUAL

■ Introduction

To maintain and maximize peak performance of Shim-pack Bio-HIC columns, and to ensure the long life and stability of columns, please read the following instructions before use.

■ Specifications

The product specifications of this product are as follows.

Item	Shim-pack Bio-HIC Butyl
Base material	Hydrophilic non-porous polymer
Chemical bonding group	Butyl group
Particle size (μm)	4
Column sizes Length × ID (mm)	100×4.6
Recommended Flow rate (mL/min)	0.5-1.0
Max. Flow rate (mL/min)	1.2
Max. pressure (MPa)	20
Range of pH	2.0 - 12.0
Temperature Range	10 - 60°C
Column material	SUS

■ Operating Precautions

Check if anything is missing or damaged. If there are any signs of damage, notify your local Shimadzu representative at once.

Each of the Shim-pack Bio-HIC columns is delivered with a Column Performance Report. The information supplied in the report include the column serial number, and chromatographic test conditions. Please keep the report for future reference.

■ Column performance

The Shim-pack Bio-HIC columns have stable quality products for customers by QC tests. Shim-pack Scepter series columns are shipped with the solvent used for the final QC test of the column, as detailed in the Column Performance Report delivered with the column.

When switching between solvents with significantly, please take care of different polarities, the miscibility and precipitation of salts.

■ Column Installation

The flow direction of the column is shown on the column (→). When installing the column, ensure that the flow direction matches the mobile phase flow direction.

The presence of voids in the tubing connect part may cause leakage and result in poor column performance (theoretical plate, peak symmetry). Take care of the ferrule tip length or cut surface of the tubing to avoid creating a void.

Use the shortest possible tubing connection from the injector to the column to minimize peak broadening.

The column should be connected with male nuts. Ensure that the fittings are connected properly to avoid creating dead volume between the tubing and the column interface. Male nuts can be ordered by referring to the part number below.

Item name	P/N	Remarks	Pressure
Male nut, PEEK	228-18565-84	5 pcs	20 MPa
Male nut 1.6 MN	228-16001	1 pc	130 MPa
Ferrule 1.6 F	228-16000-10	1 pc	130 MPa
UHPLC Fitting 2 S	228-56867-41	1 pc	130 MPa
Nexlock fittings	228-62544-90	1 pc	130 MPa

NOTE Stains or air in the flow line may deteriorate the column. Before connecting the column, be sure to flow the mobile phase to flush the flow line.

If peaks are tailing more on the early eluting compounds than later eluting compounds, there is a possibility that there is a dead volume. In such case, check that all column connections are properly connected.

Also, make sure to use appropriate internal diameter and length size of tubing at the injector and detector, especially when using semi-micro size columns, to avoid system dead volumes.

■ Operating Precautions

Use narrow internal diameter tubings leading to and from the column (tubing 0.15 mm or less, 0.006 inch I.D. is recommended) so that band spreading can be minimized.

Optimize the sampling rate and the response time, if needed.

■ Sample

Samples should be dissolved in an eluent or solvent weaker than the mobile phase, which helps avoid sample precipitation at column inlet/head and inconsistent retention values.

To ensure the proper binding of the target materials to the resin, dissolve the sample in the starting mobile phase. If the sample precipitates at this salt concentration, dissolve the sample in the doubling dilution of the starting mobile phase with appropriate buffer or water.

In order to prevent the precipitation of salts contained in sample or solvent, check the miscibility of these with mobile phase before injection.

■ Clogging of column

The most common cause of the increase of column back pressure or split peaks is blockage of the inlet filter by sample particulates, or large quantities of lipophilic compounds adsorbing to the head of the column.

- Filtrate the mobile phase using a 0.2-0.5 µm membrane filter before using the column.
- Filtrate the sample using a syringe filter before injecting to the column.

■ Column Handling Precautions

Do not drop or bump the columns, to avoid a deterioration of the column performance.

Recommended operating conditions are shown in the table in "Specifications". Sudden pressure surge may cause the degradation of column performance. Avoid using a column repeatedly near the pressure limit or sudden change in pressure, which may cause shortening of in the column life.

Column should be disconnected from the system after the pressure drop to "0".

Please note that operating the sample injection valve slowly or using an auto-sampler with slow valve switching speed will also generate a rapid pressure increase at the column inlet, which will cause premature column deterioration.

The columns are compatible with water-soluble organic solvents up to 50%. Make sure to confirm that there is no salt precipitation before using a mobile phase with organic solvent.

■ Column storage

Shim-pack Bio-HIC columns are shipped in 20% ethanol. The column can be stored in water or mobile phase for short term (overnight). For long term, store the column in 20% ethanol or methanol in water.

■ Washing the column

Most bound samples are eluted by washing the resin with salt-free buffer. However, when deterioration of the column such as increasing pressure is observed, more effective column cleaning procedure below is required.

1. Wash the column with water for 30 column volumes.
2. Inject 100-250 µL of 0.1-0.2 M NaOH several times.
3. Wash the column with water for 20 column volumes
4. If problem persists, inject 100-250 µL of 20% acetic acid aqueous solution several times.
5. Wash the column with water for 20 column volumes

If the issue still persists, replace with a new column.

■ Technical Support

Shim-pack Bio-HIC columns are manufactured, inspected, packaged and shipped under strict standards of quality control. Should you find any defect in performance, please contact your local Shimadzu representative, who will ensure your complete satisfaction.

We regret that we cannot guarantee the lifetime of columns, also that we cannot accept any claim when performance has deteriorated due to noncompliance with the operation procedures elucidated above, or as a result of normal aging.