CARE AND USE MANUAL

Xtimate Lactose-NH2 HPLC Column Care and Use Manual

Welch

Distributed by AKRIBIS



Xtimate Lactose-NH_2 HPLC Column Care and Use Manual

Dear User:

Thank you for choosing our liquid chromatography column products! This manual is designed to help you use our products under optimal conditions and maximize column lifespan. Please read the usage instructions carefully before operation.

Xtimate Lactose-NH_2 column uses a special bonding process to make the retention of lactose on the stationary phase more stable, resulting in an extremely low RSD value for the lactose peak area. It is a dedicated column for lactose as specified in the Chinese Pharmacopoeia, Volume IV.

Preparation Before Use

Column Identification

To ensure the quality of every column, each column sold by Welch Materials is assigned a unique identification code. This code allows us to trace quality-related issues. Upon receiving your column, please follow these steps:

1. Verify that the packaging is intact and matches the ordered column model.
2. Ensure the package contains a COA report with the inspector's signature.
3. Check the column surface for any damage and confirm that the protective plugs on both ends are intact.
4. Verify that the column body bears a Welch Materials identification label. Confirm that the model and serial number on the box match those on the column label.

Note:

The packing material inside the column is composed of fine particles. Do not attempt to open the column arbitrarily, as the material may become airborne and pose a health risk. If opening is necessary, use appropriate protective measures.

Installation

To achieve optimal column efficiency, use narrow-diameter tubing for connections. The connections to the column inlet and outlet must be tightly sealed, with the tubing ends polished and free of burrs or bevels. Use professional tools to cut and trim tubing.

1. Remove the column from its packaging. Check the direction of the eluent flow indicated by the arrow on the column body. Unscrew the protective plugs at both ends of the column. Position the column so that the eluent flow aligns with the direction indicated, as shown in Figure 1.

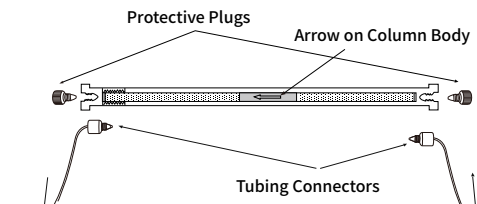


Figure 1. Column Installation

2. Carefully screw stainless steel or other compatible material fittings into both ends of the column. Ensure tight, zero-dead-volume connections to prevent leakage during operation.

Product Specifications

Column	Xtimate Lactose-NH_2
USP Code	L8
Pore Size	120 Å
Specific Surface Area	450 m^2/g
pH Range	2.0-8.0
Max. Temperature	75 °C
Max. Pressure Tolerance	40 MPa

Note:

When using silica-based columns at pH ＞7, use buffers with concentrations not exceeding 20 mM and temperatures not exceeding 40 °C to maximize column lifespan. Extreme pH or temperature conditions can significantly reduce column durability.

Column Activation

(Recommended for new columns or those unused for over a month)

Xtimate Lactose-NH_2 columns are shipped in pure acetonitrile. Activation should follow these steps:

Step 1: Activation

Activation Procedure	
Specification	4.6×300 mm, 5 μm
Flow Rate	0.5 mL/min
Mobile Phase	100% Acetonitrile
Time	5 hours
Temperature	30 °C

Routine Column Washing

Washing Mobile Phase	70% → 100% Acetonitrile
Flow Rate	Method Flow Rate
Time	30 mins each (Column Length ⩽100mm)
	40 mins each (Column Length ＞100mm)
Storage	Store the column in 100% acetonitrile. Place in a cool, dry place.

Column Regeneration:

If the column exhibits increased pressure, abnormal peak shapes, reduced efficiency, or decreased resolution, flush as follows:

Flow Rate	Method Flow Rate
Mobile Phase	100% Methanol → 100% Acetonitrile → 100% Isopropanol → 100% Acetonitrile
Time	3 hours each
Temperature	30 °C
Note	Isopropanol's high viscosity increases pressure; adjust flow rate within range.

Precautions:

The Refractive Index Detector (RID) used in the Lactose detection is highly sensitive to temperature fluctuations, mobile phase composition changes, and bubbles in the mobile phase, which may lead to baseline drift, increased noise, poor resolution or insufficient column efficiency. To ensure optimal performance, adhere to the following guidelines:

1. Mobile Phase Preparation

• Premix the mobile phase components thoroughly by manual shaking.

• Perform vacuum filtration and sonication degassing for 10 minutes.

• Use a single channel for mobile phase delivery; avoid in-line mixing.

2. Instrument Configuration

• Install an in-line filter to minimize particulate interference.

3. Injection Volume

• Strictly follow pharmacopeial requirements: 10 µL injection volume.

• Replace the sample loop with a 10 µL loop to reduce extra-column dead volume effects.

Precautions (continued):

4. Column Temperature Control

• Ensure the column reaches the set temperature before analysis.

• Most column ovens heat air, which indirectly transfers heat to the column. Since the mobile phase cannot be preheated to the target temperature, achieving stable column temperature is challenging.

Recommendation: Fill the column oven compartment with dry towels or cotton to improve heat transfer efficiency, enhancing baseline stability and chromatographic performance.

Glossary of Terms:

pH Range: The pH range tolerable for the column's mobile phase and sample solutions.

Specific Surface Area: The surface area per gram of silica.

Pore Size: The diameter of pores within silica particles, selected based on analyte molecular weight.

Carbon Loading: The percentage of carbon content in bonded functional groups relative to the total silica mass.

USP Code: Universal code for bonded phases in the USP chromatography column database.

Transition Procedure: A procedure to ensure miscibility between activation mobile phase and method mobile phases using transition mobile phase.

Maximum Pressure Tolerance: The upper pressure limit the column can withstand. Long-term use is recommended at pressures below 30 MPa. Monitor the instrument's pressure for safe operation.

Storage Solvent: Solvent used during storage or transport to maintain the activity of packing material. Handle with appropriate protective measures.