

## 1. Description

Seplife®RP LXMS-30(300) is a medium and high-pressure chromatography resin independently developed by Sunresin. It is a porous high polymer based on polystyrene/divinylbenzene matrix, which has the characteristics of large specific surface area, excellent chemical and physical stability, etc.

Compared with the traditional reversed-phase silica gel which is not resistant to acid and alkali and has a narrow pH operating range, Seplife® RP LXMS-30(300) has the advantages of high flow rate and high dynamic capacity, which is very suitable for the application of fast and efficient chromatography.

While improving production efficiency, it further reduces the production cost for the users.

Seplife® RP LXMS-30(300) is widely used in reversed phase chromatography (RPC) separation of small molecular compounds, peptides, Oligonucleotides, low molecular weight proteins and other biomolecules.

### ADVANTAGES

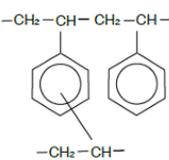
Highly hydrophobic

High chemical stability

Easy column packing

Efficient regeneration

## 2. Properties

Product	Seplife® RP LXMS-30(300)
Appearance	White spherical beads
Chemical structure	
Matrix	Polystyrene/DVB
Particle size (µm)	30±3

Average pore size (Å)	250~350
Bulk density (g/ml)	0.85-0.95
Recommended working flow rate (cm/h)	120-900
pH stability	2-12 (Operational), 1-14 (CIP)
Chemical stability	Stable to common aqueous buffers, 1M HCl, 1M HCl/90% methanol, 90% acetic acid, 6M GuHCl, 100% n-propanol, 100% ethanol, 100% methanol, 100% acetone, 0.45M NaOH/40% 2- Propanol, 1.0 M NaOH, 0.1% TFA in water, 0.1% TFA in acetonitrile, 100% isopropanol, 100% tetrahydrofuran
Dynamic capacity (mg/ml)	22~28 (Vitamin B12); 45~55 (Insolin)
Recommended column Packing pressure (bar)	5~8
Typical efficiency at 180 cm/h (Plates/meter)	8000
Max pressure resistance (bar)	50
Operating temperature (°C)	4-30 °C
Shipped as	Shipped in 20% ethanol slurry

### 3. Instructions

#### 3.1 DAC Column Packing

To ensure the RP LXMS-30(300) material is fully dispersed and free of lumps, the packing slurry can

be shaken, bottled rolled or ultrasonicated for approximately five minutes. And use the following methods:

1) The column volume (V) of the chromatographic column,  $V=Ac \times L$   $Ac=\pi \times r^2$ .

Ac: Cross-sectional area of the column; L: column height; r: column radius

2) Agitate the resins to form a homogenate and measure the desired mass or volume, about 1.1~1.15 times the column volume to prevent shrinkage.

3) Replace 20% ethanol with 100% methanol or 80% acetonitrile solution and equilibrate overnight.

4) Before loading the column, adjust the homogenate concentration to 50-70% with 100% methanol or 80% acetonitrile solution, and pour the homogenate into the DAC chromatography column at one time.

5) Complete the assembly of the column and operate the packing station according to the instructions supplied. A piston packing pressure of approximately 0.8~2.0 Mpa is recommended. Make sure that the packing pump pressure has been calculated using the correct ratio for the column ID/packing station being used to give a piston pressure.

6) Once column packing is complete, the flow of packing solvent has ceased and the pump has stopped, allow the column to stand/equilibrate for 10 minutes.

7) The column plunger should be locked in the compressed position so that the column can be operated in the Static Axial Compression (SAC) mode.

8) The packed column is now ready for use. It can be used while still assembled on the packing station or it can be undocked for use in a purification facility.

### 3.2 Column Efficiency Evaluation

After packing, clean the chromatographic column with 3-5 CV of 100% methanol or 80% acetonitrile solution. The flow rate should be controlled at 120-180cm/h to balance and perform column efficiency test.

The test method for column efficiency of RP chromatography columns is as follows:

Sample: 1:9(V:V) Acetone :100%MeOH or 80% acetonitrile

Loading volume: 1 % of column volume;

Eluent: 100%MeOH or 80% acetonitrile solution, two column volumes;

Linear flow rate: 120~180 cm/h;

Detection: UV @ 254 or 280 nm;

The prep- HPLC system geometry, including dead volume, will significantly affect the plate count determination.

### 3.4 Equilibration

After loading the column, equilibrate with the mobile phase for 3-4CV, and control the flow rate at 120-180cm/h until the conductance and pH of the flow-through remain unchanged before loading the sample.

### 3.5 Sample feeding

The solid sample can be prepared by dissolving in the equilibrium solution. Low-concentration sample solutions should be concentrated in advance as much as possible. High concentration sample solution can be diluted by the equilibrium solution. To avoid clogging of the column, samples should be processed by centrifugation or membrane filtration. The feed amount is calculated according to the capacity of the resin and the content of the target protein in the feed solution. Before loading, make sure that the sample buffer should be as consistent as possible with the equilibration solution. A small amount of sample feed can be used for the first experiment, then the amount of sample feed can be increased according to the retention time and peak shape of the target.

### 3.6 Elution

Use 2-10 CV of methanol, ethanol, acetonitrile, acetone, etc. (aqueous) solution to dissolve; use acid, alkali, buffer, etc. to adjust the pH; or use both to elute the active ingredient.

### 3.7 Regeneration and CIP

First use **acetonitrile, methanol**, ethanol, acetone, alkali + ethanol and other solvents to wash (3-4 CV) according to the operating flow rate, and then use mobile phase to balance and wash (3-4 CV).

### 3.8 Case study

Seplife® RP LXMS-30(300) has similar chromatographic performance with RPC SOURCE 30. Figure 1 shows the elution curves of albumin, lysozyme, insulin using Seplife® RP LXMS-30(300) and Cytiva RPC SOURCE 30 using the same column packing and chromatographic conditions

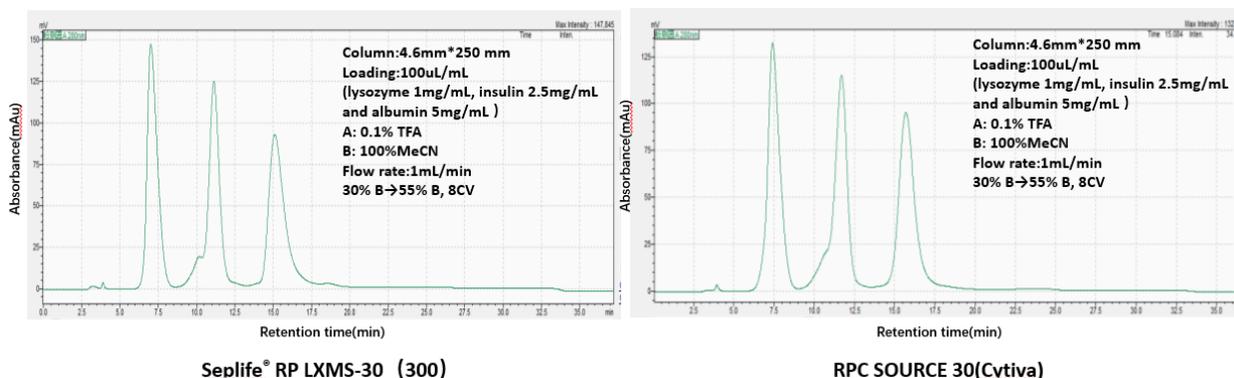


Figure 1. Elution curves of Seplife® RP LXMS-30 and Cytiva RPC SOURCE 30 for albumin, lysozyme, insulin

## 4. Storage

Seplife® RP LXMS Chromatography resins that are not for immediate use should be stored in **70-80% acetonitrile, 90-100% methanol** or 20-25% ethanol at 4-30 °C.

## 5. Transportation

Avoid sunlight, rain, and heavy pressure during transportation. It is strictly forbidden to transport with toxic and hazardous materials.

**6. Ordering information**

Product Name	References	Pack Size
Seplife® RP LXMS-30(300)	PS00042X(30)2-1	25ml
	PS00042X(30)2-2	100ml
	PS00042X(30)2-3	500ml
	PS00042X(30)2-4	1L
	PS00042X(30)2-5	5L
	PS00042X(30)2-6	10L

Production date: See label

Service life: 5 years, under proper storage conditions

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