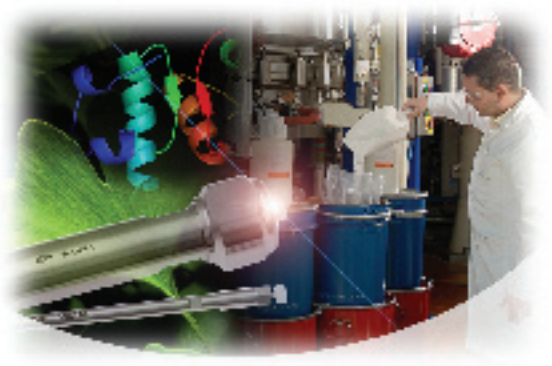
PLRP-S Polymeric Reverse Phase Media



Packing Information

PLRP-S media is chemically and physically stable across the complete pH range. This guide is intended to help with column packing, performance and lifetime.

1.0. INTRODUCTION

The following are guidelines for packing PLRP-S material into the laboratory Load & Lock™ columns using the recommended packing station. The packing pressure used must NOT exceed the safe operating pressure of the packing station/column. The operational instructions supplied with the Load & Lock packing station and columns must be adhered to at all times.

2.0. PERSONNEL

These guidelines have been written for personnel having a good knowledge of the methodologies used for packing laboratory Dynamic Axial Compression (DAC) columns.

3.0. SAFETY

Please read the MSDS provided with the PLRP-S material before opening the bottle. The person or persons using the PLRP-S material must comply with the Health and Safety Regulations in force in the Country and Establishment where the material is being used.

4.0. PREPARATION OF THE PACKING SLURRY AND COLUMN PACKING

The density of the PLRP-S material is less than conventional RP silica-based media and therefore less weight of material is required to pack the same column volume. The recommended weight of dry PLRP-S material required to pack a 10 cm bed length of the three diameters of laboratory Load & Lock columns is given in Table 1. The recommended weights equate to a packed bed density of 0.33 g of dry material per mL of packed column bed; this is comparable to the PLRP-S pre-packed columns.

4.1 PLRP-S material requires no de-fining or conditioning prior to use, this is done as part of the proprietary production process for this material. PLRP-S is supplied as a dry powder ready for use.

4.2 Based on the required column volume, column ID and length to be packed, calculate and weigh the appropriate amount of dry material.

4.3 Disperse the material in packing solvent, acetonitrile:water (80:20v/v), to give a final slurry concentration of approximately 0.23 g dry PLRP-S /mL of packing solvent.

4.4 To ensure the PLRP-S material is fully dispersed and free of lumps, the packing slurry can be shaken, bottled rolled or ultrasonicated for approximately five minutes. As with all HPLC media, do not use a magnetic stirring bar as this will grind the particles and produce fines. The slurry preparation may be assisted by sieving through a 106 µm/150 mesh sieve (212 µm/72 mesh for 50 µm particle), but care should be taken when sieving not to use excessive force which could cause the particles to fragment.

4.5 The packing slurry is now ready for use and maybe used immediately or stored for a period of up to one month. If stored, the PLRP-S material will settle so will need to be dispersed by gently shaking prior to column packing.

4.6 Take the homogenous, free flowing slurry and pour quickly into the assembled column in one continuous action.

4.7 Complete the assembly of the column and operate the packing station according to the instructions supplied. A piston packing pressure of approximately 650 psi (45bar) 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 of 650 psi.

PLRP-S Polymeric Reverse Phase Media

4.8 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.

4.9 The column plunger should be locked in the compressed position so that the column can be operated in the Static Axial Compression (SAC) mode, the optimum for the PLRP-S columns.

4.10 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.

4.11 To test the column efficiency connect the PLRP-S column to the HPLC pumping system and flush the column into 7:1 w/w acetontrile:water, one to two column volumes, at a linear velocity of 90-180 cm/h.

4.12 Connect the column outlet to the detector and continue flushing the column until a stable base line has been achieved. The efficiency of the column can be determined using the procedure below.

	Load & Lock Columns		
	1" (2.5 cm ID)*	2" (5.0 cm ID)	3" (7.5 cm ID)
Column volume	57 mL	196 mL	442 mL
Weight of dry PLRP-S	19 g	65 g	146 g
Packing solvent	85 mL	300 mL	650 mL
Flow rate equivalent to 180 cm/h	17 mL/min	59 mL/min	133 mL/min

* actual ID 2.7 cm

Table 1. Summary of the column packing/testing parameters for the three sizes of Load & Lock columns based on a 10 cm packed bed length.

5.0. COLUMN TESTING PROCEDURE

Test the column using a UV detector at 254 nm with acetone as the test probe and 7:1 w/w acetonitrile:water as the eluent. Typical efficiencies of PLRP-S columns are shown in the Table 2. The prep-HPLC system geometry, including dead volume, will significantly affect the plate count determination.

Column	Plates/meter
PLRP-S 8 µm	30,000
PLRP-S 10 µm	25,000
PLRP-S 10-15 µm	20,000
PLRP-S 15-20 µm	12,000
PLRP-S 30 µm	8,000
PLRP-S 50 µm	4,000

Table 2. Typical efficiency of PLRP-S columns at a linear velocity of 180 cm/hr.

VARIAN

Varian, Inc.
www.varianinc.com
North America: 800.926.3000, 925.939.2400
Europe The Netherlands: 31.118.67.1000
Asia Pacific Australia: 613.9560.7133
Latin America Brazil: 55.11.3238.0400

Other sales offices and dealers throughout the world–check our Web site.

SI-0893/C/3.10 Printed in the UK