

Agilent VariTide RPC MediaData Sheet

General Description

VariTide RPC Media is chemically and physically stable across the complete pH range. This guide is intended to help with column packing, performance and lifetime.

Introduction

The following are guidelines for packing VariTide RPC 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

Personnel

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

Safety

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

Preparation of the Packing Slurry and Column Packing

The density of the VariTide RPC 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 VariTide RPC material required to pack a 10 cm bed lengh 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 which is comparable to the VariTide RPC pre-packed columns.

- VariTide RPC material requires no de-fining or conditioning prior to use, this is done as part of the proprietary production process for this material.
- Based on the required column volume, column id and length to be packed, calculate and weigh the appropriate amount of dry material.

- 3. Disperse the material in packing solvent, acetonitrile:water (80:20v/v), to give a final slurry concentration of approximately 0.23 g dry VariTide RPC/mL of packing solvent.
- 4. To ensure the VariTide RPC material is fully dispersed and free of lumps the packing slurry can be shaken, bottle rolled or ultrasonicated for approximately five min. As with 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 $\mu m/150$ mesh sieve, but care should be taken when sieving not to use excessive force which could cause the particles to fragment.
- The packing slurry is now ready for use and may be used immediately or stored for a period of up to one month. If stored, the VariTide RPC material will settle so will need to be dispersed by gently shaking prior to column packing.
- Take the homogenous, free flowing slurry and pour quickly into the assembled column in one continuous action.
- 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 (45 bar) 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 pistion pressure of 650 psi.
- 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 min.
- 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 VariTide columns.
- The packed column is now ready for use. It can be used whilst assembled on the packing station or it can be undocked for use in a purification facility.
- Connect the VariTide RPC column to the HPLC pumping system and flush the column into 7:1 w/w acetonitrile: water, one to two column volumes, at a linear velocity of 90-180 cm/h.
- 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 overleaf.



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. The typical efficiency of a VariTide RPC column would be in the order of 45,000 plates/metre at a linear velocity of 180 cm/h. The prep-HPLC system geometry, including dead volume, will significantly affect the plate county determination.

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.

	Load & Lock columns		
	1 in (2.5 cm id)	2 in (5.0 cm id)	3 in (7.5 cm id)
Column volume	57 mL	196 mL	442 mL
Weight of dry VariTide RPC	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

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