ELSEVIER

Journal of Chromatography A, 827 (1998) 193–196

JOURNAL OF CHROMATOGRAPHY A

Short communication

Unique dynamic axial compression packing system

Carlos Orihuela*, Randy Fronek, Larry Miller, Dorothy Honda, James Murphy

Searle, 4901 Searle Parkway, Skokie IL 60077, USA

Abstract

A new dynamic axial compression system (MultiPacker; MODcol, St. Louis, MO, USA) was tested for packing small particle stationary phases into 5.08 cm I.D. preparative columns. The unique design of this system allows a column to be removed and the hydraulic system be used to pack other columns. Several stationary phases were packed and theoretical plate numbers were measured. Initial experiments showed that this system can pack stationary phases with suitable efficiencies for more difficult purifications.

Keywords: Column packing; Dynamic axial compression column packing; Stationary phases, LC; Preparative chromatography; MultiPacker column packing system

1. Introduction

In the past several years, there has been an increase in the use of small particle stationary phases for preparative chromatography purifications in the pharmaceutical industry [1–4]. To have columns with efficient media, a laboratory may either purchase prepacked columns or a slurry packing system. One type of slurry packing system available uses dynamic axial compression (DAC) technology [5–8]. The drawback of many DAC systems is that, once packed, the column must remain attached to the piston apparatus. In this way, pressure is continuously applied to the column bed to accommodate any bed disturbances that may result during purification. The difficulty arises when a laboratory has a need for another stationary phase. The first column must be unpacked to allow a different stationary phase to be packed.

A new alternative for slurry packing systems is now available. The MultiPacker system (MODcol, St. Louis, MO, USA) can pack a 25×5.08 cm column using an axial compression system. Once packed, the column can be removed, and the system used to pack another column. Pressure is maintained on the bed by installing a spring apparatus.

2. Experimental

The purity of the solvents was spectroscopic grade and obtained from a variety of sources. Standards used for measuring chiral column performance were *trans*-stilbene oxide for efficiency measurements, and 1,3,5-tri-*tert*.-butylbenzene as a void volume marker. Standards used for measuring achiral column performance were dipropylphthalate and ethylphthalate for efficiency measurements, and toluene as a void volume marker. All standards were supplied by Aldrich (Milwaukee, WI, USA).

Chromatographic bulk stationary phases were obtained from the following vendors:

1. Chiralcel OD, Chiralpak AD and Chiralpak AS, 20 μm packings, were from Chiral Technologies, Exton, PA, USA.

*Corresponding author.

0021-9673/98/$ – see front matter
PII: S0021-9673(98)00704-3

2. LiChrosorb Si 60, 15–25 μm, was from EM Separations, Gibbstown, NJ, USA.
3. Zorbax Pro10, 10 μm silica, was from BTR Separations, Wilmington, DE, USA.
4. Whelk-O (R,R), 10 μm, was from Regis, Morton Grove, IL, USA.

3. Equipment

Various preparative chromatographic systems complete with chart recorders were used along with a mechanical stirrer equipped with a glass rod and a PTFE paddle.

4. Methods

For column performance measurements, peak widths were measured at half height, and theoretical plate numbers were calculated using the following formula: $N=5.54\ (t_r/t_w)^2$ [9]. Peak widths (t_w) and retention times (t_r) were measured by hand from recorder traces.

Slurry preparation procedure for Chiralcel OD, Chiralpak AD, Chiralpak AS, Zorbax Pro10 Si and LiChrosorb Si 60:

1. The stationary phase was dried in a vacuum oven at 50°C and 635 mmHg for a minimum of 1.25 h (1 mmHg=133.322 Pa).
2. The dry stationary phase was then wetted, by mixing 250–300 g of packing and 800–900 ml of organic solvent (see Table 1). The mixture was blended with a mechanical stirrer set at 300 rpm in a sonicator for approximately 30 min.
3. The slurry was allowed to settle for 1 h and the supernatant liquid was decanted to remove fines.
4. The original volume was restored with fresh solvent and the mixture was reslurried for 15–20 min.
5. The slurry was poured into the column and the stationary phase was compressed according to the procedure used by the MultiPacker manufacturer [10,11].

Slurry preparation procedure for the Whelk-O stationary phase:

1. The stationary phase was dried in a vacuum oven at 50°C and 635 mmHg for a minimum of 1.25 h.
2. The dry stationary phase was then wetted, by mixing 300 g of packing and 775 ml of chloroform, and stirred by hand.
3. The solution was sonicated for 10 min with visual inspection for homogeneity.
4. The slurry was poured into the column and the stationary phase was compressed according to the procedure used by the MultiPacker manufacturer [10,11].

Packings were conditioned using media vendor-recommended procedures. For all column tests, three to five injections were made and the average plate number was reported. The column was removed from the hydraulic system, the spring assembly apparatus was installed and the column was retested. All plate numbers reported for chiral packings are for

Table 1
Stationary phases packed and resulting plate numbers

Stationary phase	Piston packing pressure (MPa)	Plates/m on MultiPacker[a] (h^b)	Plates/meter with spring installed[a] (h^b)	Expected plates/m on MultiPacker[a] (h^b)	Solvent used for slurry preparation
Chiralcel OD	4.14	6800 (7.4)	5100 (9.8)	4000–5000 (11.3)	Ethanol
Chiralcel OD	6.89	4600 (10.9)	4500 (11.1)	4000–5000 (11.3)	Ethanol
Chiralpak AD	3.45	4900 (10.2)	4700 (10.6)	4000–5000 (11.3)	Ethanol
Chiralpak AD	6.89	5000 (10.0)	4000 (12.5)	4000–5000 (11.3)	Ethanol
Chiralpak AS	6.89	8100 (6.2)	8600 (5.8)	4000–5000 (11.3)	Ethanol
Whelk-O (R,R)	10.34	12 400 (8.0)	12 200 (8.2)	15 000 (6.7)	Chloroform
Zorbax Pro10 Si	6.89	30 300 (3.3)	30 300 (3.3)	40 000–50 000 (2.3)	Acetone
LiChrosorb Si 60	6.89	12 800 (3.9)	Spring not installed	25 000–30 000 (1.9)	Heptane

[a] Flow-rate for column testing was 120 ml/min.
[b] Reduced plate height using $h=L/N(d_p)$; L=bed length, N=plates, d_p=particle diameter.

the first-eluting peak in *trans*-stilbene oxide and, for the achiral packings, the plate numbers for dipropylphthalate are reported.

5. Results and discussion

Although the range of values obtained for the individual column efficiency measurements were larger than expected, the average numbers reported are sufficient to determine the usefulness of each column packed (Table 1). The focus of the discussion is to comment on the speed, versatility, practicality and ease of use of the MultiPacker system.

The simple piston head design of the MultiPacker allows for quick and easy cleaning. However, once the slurry is poured into the column, the installation of the piston head requires two people. This involves tightening three bolts to ensure a proper piston seal. Since the slurry settles during this time, the column was removed from the MultiPacker system and the slurry reconstituted by physically shaking the column back and forth five to ten times. Though not a formal step in the procedure, the manufacturer suggested this step to ensure slurry integrity. Once the piston was installed, full bed compression typically took less than 45 s.

Testing of the columns was performed using a flow-rate of 120 ml/min. This flow-rate is the linear velocity scale up from a 1.0 ml/min flow on a 4.6-mm I.D. column. Some column manufacturers measure plate numbers using a flow-rate of 50 ml/min. This is a linear velocity scale up from approximately 0.4 ml/min on a 4.6-mm I.D. column. The flow-rate used to test the column is critical to the number of theoretical plates observed. Since the authors tested at faster flow rates, plate numbers may have been reduced by as much as 50% from those typically seen by the manufacturer.

One difficulty in packing small particle preparative columns is finding the ideal packing pressure. The 6.89 MPa packing pressure was the vendor-recommended packing pressure for all stationary phases except Whelk-O. In the case of the Chiralpak AS stationary phase, 8600 plates/m was achieved (Table 1). However, 4000–5000 plates/m was observed with the other Chiral Technologies stationary phases. These reduced values were originally thought to be due to the packing pressure chosen for a particular stationary phase. Second attempts were made to repack the Chiralpak AD and Chiralcel OD stationary phases. Using a packing pressure of 4.14 MPa to repack the Chiralcel OD stationary phase, higher theoretical plate numbers were obtained, but were still below the Chiralpak AS value. A packing pressure of 3.45 MPa was used to repack the Chiralpak AD stationary phase. In this case, a lower theoretical plate number was obtained when compared to the packing with a piston pressure of 6.89 MPa. Plate numbers obtained for the Whelk-O, LiChrosorb and Zorbax stationary phases were lower than expected. No second attempts to repack these stationary phases were made.

Once packed, the column can be run indefinitely while connected to the MultiPacker. The main advantage lies in this system's ability to maintain packing integrity when the piston rod is removed and a spring apparatus is installed. In order to minimize disturbing the bed during the installation of the spring, the piston rod must be retracted slowly, and the spring installed quickly. The installation of the spring was demonstrated by the manufacturer, and reproduced during the experiments.

Installation of the spring apparatus resulted in a plate number reduction that was typically less than 25%. In many instances, the number of theoretical plates was approximately the same, ($\pm$5%), as the original value. Since plate loss due to spring installation can be a problem, the manufacturer has recently developed an alternative means of retaining column integrity when the column is removed from the system. With the new design, the spring is installed during initial column packing; eliminating the retraction and installation step. This new apparatus was not tested in our laboratories.

The advantages of being able to pack a column and remove it from the MultiPacker, was demonstrated with the Chiralcel OD and Zorbax Pro 10 stationary phases. The Chiralcel OD column packed with a packing pressure of 4.14 MPa was used in two purifications, while the MultiPacker system was used to pack other columns. After a total of 22 preparative injections, the Chiralcel OD column was retested. No significant change in plate number was observed (Table 2).

The Zorbax Pro 10 column was used to purify 35

Table 2
Plate numbers after column use

Stationary phases	Plates/m after column use[a] (h^b)
Chiralcel OD	5600 (9.0)
Zorbax Pro 10 Si	24 500 (4.1)

[a] Flow-rate for column testing was 120 ml/min.
[b] Reduced plate height using $h=L/N(d_p)$; L=bed length; N=plates; d_p=particle diameter.

g of a sample that required 100 injections. The purification was run over a two-week period and subjected to operating pressures of 2.76 MPa. After the purification was complete, the column remained idle for two months and was then retested. Retesting of this column showed a 20% loss in theoretical plate number (Table 2).

6. Conclusion

The MultiPacker allows a preparative laboratory to pack a diverse number of stationary phases with efficiencies that are suitable for preparative separations. Preliminary experiments show that columns packed with the MultiPacker can retain their performance over time, and be a viable alternative to prepacked columns.

References

[1] L. Miller, D. Honda, R. Fronek, K. Howe, J. Chromatogr. A 658 (1994) 429–435.
[2] R.M. Ladd, A. Taylor, LC·GC 7 (1989) 585.
[3] A.M. Katti, Chromatographia 33 (1992) 5.
[4] E.P. Kroeff, R.A. Owens, E.L. Campbell, R.D. Johnson, H.I. Marks, J. Chromatogr. 45 (1989) 461.
[5] H. Colin, P. Hilaireau, J. de Tournemire, LC·GC 8 (1990) 302.
[6] M. Sarker, G. Guiochon, J. Chromatogr. A (1995) 27.
[7] D.R. Wu, K. Lohse, J. Chromatogr. A 658 (1994) 381.
[8] E. Godbille, P. Devaux, J. Chromatogr. 122 (1976) 317.
[9] L.R. Synder, J.J. Kirkland, Introduction to Modern Liquid Chromatography, Wiley, New York, 1979, p. 31.
[10] Y. Shalon, Am. Lab., September (1997) 20N.
[11] C.W. Martin, Y. Shalon, LC Mag., 3 (1985).