



Micro-Guard® Cartridges



Introduction

Guard cartridges have been an accepted part of the HPLC technology for a number of years because of the important role they play in protecting the analytical column and HPLC system. Micro-Guard systems not only extend the lifetime of the analytical column, but also provide a convenient method for in-line sample preparation. Contaminants which interfere with analytical separations, and compounds which foul the analytical columns, can be removed with these systems. Interferences caused by anions, cations, organics, salts, insolubles, and particulates can be reduced or eliminated using the Micro-Guard systems. The Micro-Guard systems are recommended for use with Bio-Rad's HPLC columns and columns from other manufacturers.

The Micro-Guard HPLC cartridge protection system consists of inexpensive, disposable cartridges packed with 9 or 12 μ m materials, and an appropriate holder. In effect, the cartridge provides a 3 cm long extension of the analytical column. When the HPLC system begins to lose resolution, simply replace the spent cartridges; a minimum of hardware is thrown away.

Specifications

Micro-Guard cartridges are 4.6 mm ID x 4 cm long (3 cm internal bed). Materials in contact with the mobile phase are stainless steel and Kel-F[®]. Maximum pressure is 4,000 psi for the complete Micro-Guard cartridge and holder system. Using standard test mixes these cartridges have a lifetime of over 300 injections. Maximum recommended temperature is 60 °C.

The cartridges contain materials specifically designed for use with the designated analytical columns. The Micro-Guard holder is made of stainless steel for high durability and corrosion resistance. Table 1 lists the cartridges and the Bio-Rad column used in conjunction with these cartridges.

Table 1. Cartridge and Column Compatibility Guide

Guard	Packing Material	Shipping Solvent	Compatible Columns
ODS-5S	5 µm spherical silica gel, C ₁₈	Methanol	ODS-5S reversed phase columns or any 5 μ m C ₁₈ column.
Disaccharide	5 µm spherical silica gel, NH ₃	Isopropanol	Disaccharide column or any 5 µm amino bonded phase silica column.
Hi-Pore® cartridge	5 μm spherical silica gel, C ₈ wide pore	Methanol	Hi-Pore RP-304 and RP-318 columns or any 5 µm C ₈ column.
Carbo-C	Aminex [®] cation exchange resin, calcium form	H ₂ O	Aminex HPX-87C and HPX-42C columns.
Cation H ⁺	Aminex cation exchange resin, hydrogen form	H ₂ SO ₄	Aminex HPX-87H, Fast Acid, Fruit Quality and Fer- mentation Monitor- ing columns.
Anion SA	Aminex anion exchange resin, sulfate form	(NH ₄) ₂ SO ₄	Aminex HPX-72S column.
Anion OH	Aminex anion exchange resin, hydroxide form	NaOH	Aminex HPX-72-O column
Carbo-P	Aminex cation exchange resin, lead form	H ₂ O	Aminex HPX-87P, Fast Carbohydrate columns.
Deashing	Aminex resin (1 cation and 1 anion ex- change resin)	H ₂ O	Aminex HPX-87P, 42C, 42A, 87C, and Fast Carbohydrate columns.
Anion CO ₃ ⁻	Aminex anion exchange resin, carbonate form	NaHCO ₃	Replacement anion cartridge in the deashing system.
Cation K ⁺	Aminex cation exchange resin, potassium form	KH ₂ PO ₄	Aminex HPX-87K column.
Cation Na ⁺	Aminex cation exchange resin, sodium form	Na ₂ HPO ₄	Aminex HPX-87N column.
Industrial Grade Cation H ⁺	Aminex cation exchange resin, large particle size, hydrogen form.	H ₂ SO ₄	Aminex HPX-87H, 85H, Fast Acid, Fruit Quality, Fer- mention Monitor- ing columns.
Industrial Grade Carbo-C	Aminex cation change resin, large particle size, calcium form	H ₂ O	Aminex HPX-87C, HPX-42C columns, and replacement in the industrial grade deashing system.

Cartridge Capacity

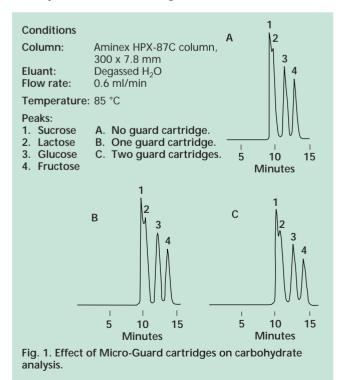
Every sample is unique, and the cartridge capacity and lifetime depends on sample contamination, running conditions, and amount of use. For silica and carbohydrate cartridges for example, back-pressure and loss of resolution are both good indicators that a cartridge is depleted and should be changed. Life expectancy of a cartridge can be evaluated by investigating its depletion in performance under controlled conditions.

Impact on Resolution

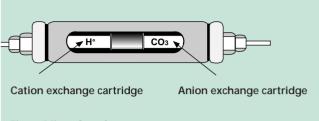
Figure 1 shows the same separation run on a resin-packed analytical column with no guard cartridge, one Micro-Guard cartridge, and two Micro-Guard cartridges in-line. The guard cartridges had no significant impact on resolution.

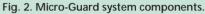
In-Line Carbohydrate Deashing Cartridges

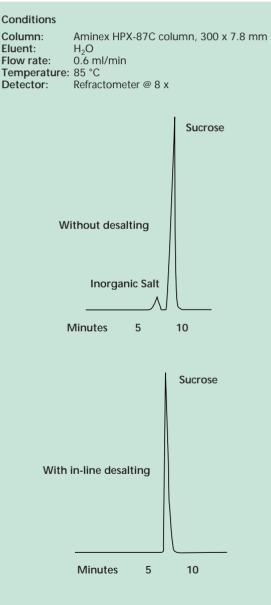
Resin-based carbohydrate analysis columns can be protected from contamination, and error in quantitation can be substantially reduced, by using a guard that combines cation and anion exchange resins to remove inorganic salts from the sample. Bio-Rad's in-line carbohydrate deashing (de-ionizing) columns consist of one cation exchange and one anion exchange Micro-Guard cartridge, and a single deashing holder which holds both cartridges (see Figure 2). The two cartridge deashing system offers a high capacity for salts, and is optimized to prevent loss of resolution due to band spreading. With the deashing column installed in front of the carbohydrate analysis column, only neutral compounds pass through the HPLC column (see Figure 3). The deashing column offers convenience along with highly efficient in-line purification of carbohydrate samples. The effectiveness of this system was demonstrated in the work done by L. E. Fitt [J. Chromatog., 152, 243 (1978)].

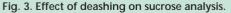


Samples that contain a high amount of contaminants require preliminary sample clean-up. For example, a sample containing a high concentration of salt should be passed through both AG[®] 2-X8 and AG 50W-X8 resin prior to HPLC. The deashing system should be used as the final sample clean-up to protect the analytical columns.









Industrial Guard Cartridges

Industrial guard cartridges are especially useful when large numbers of dirty samples are being injected. These cartridges allow higher through-put than the analytical Micro-Guard cartridges, and provide slightly lower efficiency. Packed with a larger size packing material than the analytical guard cartridges, the industrial guard cartridges are recommended for HPLC analysis in industrial settings.

Applications

Micro-Guard cartridges have been used together with analytical columns in a variety of applications. Table 2 lists some specific applications, and Table 3 lists some compounds that have been separated using each cartridge.

Procedures for Using Bio-Rad's Guard Columns

To install a cartridge, unscrew an end nut from one end of the holder. Carefully insert the new cartridge. Reseal the holder finger-tight using both end nuts. Further tightening is unnecessary and may damage the seal or holder. Never use tools to over-tighten this seal. Care must be taken to insure miscible and correct elution solvents when installing a new cartridge. Table 1 lists the solvents in which the cartridges were shipped. If necessary, allow at least three bed volumes of mobile phase to pass through the cartridge before connecting the analytical column.

Cartridge Storage

To store a cartridge out of its holder between uses, protect its Kel-F frit assemblies from dirt and scratches. Store it in a stoppered glass test tube, with a few drops of packing solvent (see Table 1) to protect the cartridge from drying out. Store at 4 °C for long-term storage of resin cartridges (do not freeze).

Leakage

If the cartridge leaks at less than its rated pressure limit, do not try to force a seal with tools. A leak indicates that the sealing surface on the seal is either dirty, scratched, or otherwise deformed. Replace this seal with a new seal, using the tools provided in the seal replacement kit.

Cartridge Care

Micro-Guard column lifetime can be increased by following the precautions taken with analytical columns. Table 4 gives some suggestions.

Table 2. Specific Applications Using Micro-Guard Columns

Cartridge	Sample	Application	Reference
Anion-OH	Yogurt and buttermilk	Removal of acids and milk salts	Richmond, M.L., J. of Diary Science, 65 (8), 1394 (1982).
Carbo-C	Syrup, wort, and beer	Determination of carbohydrates in brewing substrates	Dadic, M. and Belleau, G., American Society of Brewing Chemist Inc., 40, 141 (1982).
Cation H⁺	Wood degradation products	Analysis after acidic, hydrothermal, and enzymatic hydrolysis	Bonn, G., et al., <i>J. of Chrom.</i> , 287 , 215 (1984).
Cation H+	Urine	Determination of abnormal urinary organic acids	Bennett, M. J. and Bradley, C. E., <i>Clin. Chem.</i> , 30 (4), 542 (1984).
Cation H⁺	Algae	Determination of ascorbic acid in algae extracts and growth media	Grun, M. and Loewus, F. A., <i>Anal. Biochem.</i> , 130 , 191 (1983).
Cation H+	Food	Determination of lactic acid	Ashoor, S. H. and Welty, J., J. of Chrom., 287, 452 (1984).
Cation H+	Food	Determination of ascorbic acid	Ashoor, S. H., et al., J.A.O.A.C., 67, 78 (1984).
Cation H+	Food	Determination of acetic acid	Ashoor, S. H. and Welty, J., J.A.O.A.C., 67, 885 (1984).
Cation H⁺	Polar compounds	HPLC elution of short-chain organic acids, aliphatic aldehydes, ketones, alcohols, and carbohydrates	Pecina, R., et al., J. of Chrom., 287, 245 (1984).
Cation H+	Anaerobic bacteria culture media	Determination of short chain fatty acids	Guerrant, G. O. et al., J. Clin. Microbiol., 16, 355 (1982)
Deashing	Wood and wood pulp	Sugar analysis	Pettersen, R. C., et al., J. of Chrom. Sci., 22, 478 (1984).

Table 3. General Applications Using Micro-Guard Cartridges

Guard	Application
ODS-5S	Separation of herbicides; measurements of drug metabolites; nucleoside evaluation; enzyme removal; cosmetic analysis.
Disaccharide	Disaccharide separations; sugars from corn tissue; separating oligo and monosaccharides.
Hi-Pore cartridge	Protein separations.
Carbo-C	Sugar determination.
Cation H ⁺	Organic acid analysis; fermentation analysis; crude fermentation broth separations; carbohydrate analysis.
Anion SA	Organic base separations.
Anion OH	Separation of organic tertiary aminex; separation of labeled compounds.
Deashing	Deashing of corn syrup carbohydrates; deashing prior to sugar analysis.

Table 4. Cartridge Care

Micro-Guard	Acceptable	Avoid
ODS-5S, Disaccharide,and Hi-Pore cartridges	Organic solvents Aqueous mobile phases Buffer mobile phases pH 2.5-7	Back pressure greater then 4,000 psi Storing in aqueous solvent Precipitating buffer pH less than 2 or greater than 7 Temperature above 60 °C
Carbo-C	CH ₃ CN (30% max.) DI water Ethanol (less than 5%) Isopropanol (less than 5%) Average flow rate of 1.2 ml/min when oper- ated independently	Acids and bases Salts Other organic solvents Flow rate above 1.5 ml/min at room temper- ature
Cation H⁺ and Cation K⁺	CH ₃ CN (30% max.) DI water (pH 3 or lower) Acids (except HCI) Average flow rate of 1.2 ml/min when operated independently	Bases Salts Other organic solvents Flow rate above 1.5 ml/min
Carbo-P	DI water Average flow rate of 1.2 ml/min	Acids and bases Salts Organic solvents Flow rate above 1.3 ml/min
Anion SA, Anion OH, and Anion CO ⁻ 3	CH ₃ CN (30% max.) Aqueous buffers with SO ₄ pH 2-13 (recommended pH 4-10) Butanol (less than 5%) Isopropanol (less than 5%)	Acids Other organic solvents Flow rate above 1.2 ml/min. Temperature above 60 °C Salts

Note: All solvents and buffers should be filtered and degassed.

Regeneration

The Micro-Guard cartridges are designed to protect your column. When resolution does begin to deteriorate or back-pressure increases, the cartridge can simply be replaced. Premature death of a cartridge may indicate further sample clean-up or preparation is necessary. In most cases cartridges cannot be regenerated. If column contamination is not due to irreversibly bound material, regeneration may be possible by following the procedures used for the similar analytical columns.

Ordering Information

Catalog Number	Product Description	
Silica/Reversed Phase Micro-Guard Cartridges		
125-0124	ODS-5S Cartridges, 2	
125-0127	Disaccharide Cartridges, 2	
125-0134	Hi-Pore Cartridges, 2	
125-0131	Standard Cartridge Holder	
Aminex R	esin Micro-Guard Cartridges	
125-0128	Carbo-C Cartridges, 2	
125-0129	Cation H ⁺ Cartridges, 2	
125-0132	Anion SA Cartridges, 2	
125-0133	Anion OH Cartridges, 2	
125-0119	Carbo-P Cartridges, 2	
125-0506	Anion CO ₃ ⁻ , 2	
125-0507	Cation K ⁺ , 2	
125-0508	Cation N, 2	
125-0131	Standard Cartridge Holder	
Aminex Resin Industrial Grade Micro-Guard Cartridges		
125-0502	Industrial Grade Cation H ⁺ , 2	
125-0503	Industrial Grade Carbo C, 2	
125-0131	Standard Cartridge Holder	
Carbohyd	Irate Deashing Cartridges	
125-0118	Deashing - 1 Cation, 1 Anion Cartridge	
125-0139	Deashing Holder	
Micro-Gu	ard Accessories	
125-0028	1/16" Reverse Nuts with Ferrules, Parker type, 2	
125-0027	HPLC Connector, 1/16" LDV reverse nut union, 1	
125-0040	1/16" x .010" I.D. x 3 " long stainless steel tubing, 5	
125-0041	1/16" x .010" I.D. x 6" long stainless steel tubing, 5	
125-0147	Seal Replacement Kit	
125-0148	Seals	

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