# Redi*Sep*<sup>®</sup> Reversed-Phase C18, C18Aq, and C8 Columns

Use, Cleaning, and Storage Instructions

Chromatography Application Note P/N 60-2203-053 Revision H, AN02

## **Application Overview**

Proper preconditioning, cleaning, and storage of reusable Teledyne ISCO Redi*Sep* Silver reversed-phase C18 and Redi*Sep* Gold® high performance C18, C18Aq, and C8 columns will help to extend column life and improve column consistency and reliability.

### **General Method**

## Initial Preconditioning

Flush the column with six column volumes (CV) of 1:1 water:acetonitrile prior to immediate use. Alternatively, pump 2 CV of 100% acetonitrile through the column, then cap and store it for one hour before use. Refer to Table 1 for column volume data.

**Table 1: Column Data** 

| Column Media<br>Weight                    | Column Volume (mL) | Optimal Flow Rate (mL/min) |
|---|--------------------|----------------------------|
| RediSep C18 Columns                       |                    |                            |
| 4.3                                       | 4.1                | 18                         |
| 13  | 12.9               | 30                         |
| 26  | 26.4               | 35                         |
| 43  | 42.4               | 40                         |
| 86  | 87.7               | 60                         |
| 130                                       | 132.6              | 85                         |
| 240                                       | 242.8              | 150                        |
| 360                                       | 361.1              | 150                        |
| RediSep Gold High Performance C18 Columns |                    |                            |
| 5.5                                       | 4.6                | 18                         |
| 15.5                                      | 13.5               | 30                         |
| 30  | 26.4               | 35                         |
| 50  | 42.6               | 40                         |
| 100                                       | 87.7               | 60                         |
| 150                                       | 129.7              | 85                         |
| 275                                       | 243.4              | 150                        |
| 415                                       | 363.5              | 150                        |
| 950                                       | 825                | 250                        |
| 1900                                      | 1650               | 300                        |
| 8600                                      | 7500               | 850                        |

# Sample Loading Capacity

The sample load on the column should be 0.1 to 2 percent of the column media weight.

#### Post-run Care

With proper care, Redi*Sep* C18 columns may be used thirty times or more. Reversed-phase methods should be written so that at the end of the separation, the column is washed with 2–4 CV of a high percentage (100%) of the organic (B) solvent. After 2–4 CV of wash, the percent organic can be returned to the starting condition for 2–3 CV to anticipate the next run column equilibration conditions (see Figure 1). Alternatively, the percent organic can be changed to 80% in anticipation of column storage.

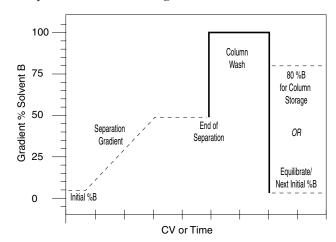


Figure 1. Post-run column flushing.

## Storage

It is possible to store C18 columns for long periods with no loss in performance. Following are some general rules for storage of reversed-phase C18 columns:

- Once wetted, never dry out a reversed-phase column.
   This will induce channeling due to expansion and contraction of the stationary phase. The air purge feature (if available on the Flash chromatography system) should be turned off.
- Store columns wet and well-capped in 10–30% water and 70–90% organic solvent such as acetonitrile, methanol, or ethanol.

# **Summary**

Precondition columns before initial use. Clean columns with high (100%) organic solvent. Never dry the column and store wet and capped with 70–90% organic solvent.

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