# Use Redi*Sep*<sup>®</sup> Columns Everywhere!



## **Chromatography Technical Note**

## **Abstract**

There are several reasons to use RediSep columns on non-Teledyne ISCO flash systems. Chemists who use RediSep columns enjoy increased loading capacity, higher resolution, lower cost, and possibly improved selectivity for their purification. This tech note explains how to install standard RediSep (up to 330 g) columns as well as larger RediSep (750 g and larger) columns using the Large Column Adapter, and how to use Teledyne ISCO solid load cartridges. There is also an explanation about how to convert RediSep column volumes to gradient segments into minutes or elution time expressed as volume of mobile phase pumped.

#### **Columns**

For the purposes of this technical note, there are two types of flash systems. The most common type is where the column is held on the system with a movable column shuttle, such as the CombiFlash® NextGen systems. Other systems simply connect the column to the system with tubing.

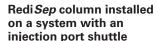
### Installation

RediSep columns have a Luer-Lok<sup>TM</sup> inlet fitting, and a Luer-slip outlet, and so are compatible with all flash systems. For systems that use a shuttle to hold the column and injection ports, RediSep columns can be installed like any other column.

For systems that connect the column only with tubing, the column is attached using the same fittings as for other columns. The fittings are usually included with system; otherwise use  $Idex^{\mathbb{T}} P-675$ , 1/4-28 Female to Male Luer Assembly. The column outlet is at low pressure so a Luer-slip fitting (Idex P-658, 1/4-28 Female to Female Leur, works well.











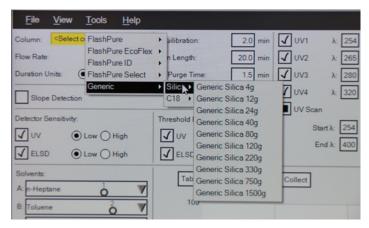
Redi*Sep* column installed on a flash system without a column shuttle

## Flash system software

Redi*Sep* columns are the standard that all other columns are based upon, so the column size you need is very likely listed in the system. In some cases you may need to choose a "generic column" to load a method for your flash system. The system will set an appropriate flow rate for the column.







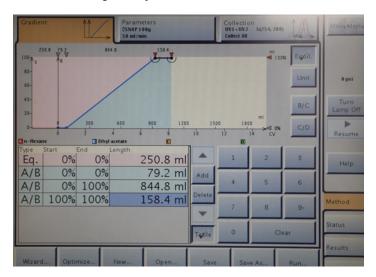
Selecting a generic flash column a on Buchi system

If a Redi*Sep* column size isn't listed, choose the size closest to the Redi*Sep* column you will be using.

The best way to program the method, or to scale-up, is to use Column Volumes (CV). Most columns are sufficiently similar in size that, except for selectivity differences, the method can be programmed in native column volumes. For systems where columns have significantly different column volumes than RediSep columns, the segments can be programmed in other units that relate to column volumes with a few simple equations:

$$Segment\ Length\ (minutes) = \frac{Number\ of\ CV\ x\ mL/CV}{Flow\ Rate\ (mL\ per\ min)}$$

Some systems allow the segments to be programmed by volume, so each segment is the number of column volumes multiplied by the column volume.



Control to set run time units on a Biotage system

## Solid load cartridges

Installation

The photographs below show systems with and without column shuttles. The captions list any additional parts that are needed.



Solid load cartridge connection with adapter made of P-661 (female Luer to 5/16-24 male) and P-665 (Male Luer to 1/4-28 female)



Solid load cartridge connection with adapter made of P-661 (female Luer to 5/16-24 male) and P-665 (Male Luer to 1/4-28 female) for the upper connection; the lower connection is P-604 (female Luer to 1/4-28 male)





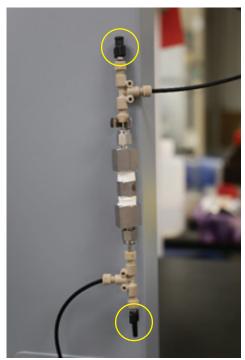
## **Large Columns**

The large column adapter (PN 60-5394-551) is used to run columns larger than 330 g silica on the Combi*Flash* NextGen systems, and can also be used on non-Teledyne ISCO flash chromatography systems. These columns have a large tapered fitting fitting that won't fit most flash chromatography systems. On systems with sliding column shuttles, you can install the adapter containing an over-pressure protection valve as if it were a smaller flash column. The gradient methods are run as described for the smaller columns. Although the columns might be rated to run at a faster flow rate than the flash system allows, the large columns were designed to run on a Combi*Flash* Torrent® that uses wider tubing to reduce back pressure. The tubing used in flash chromatography systems may limit the flow rate due to high backpressure, which may damage the column (the column pressure limit is noted on the column label).



Large column adapter for systems with column shuttle

For systems that can't fit the Large Column Adapter directly, the Luer fittings can be removed, and 1/4-28 connections can be attached in their place as shown in the image below.



Remove circled parts from pressure relief valve.



Connect ¼-28 fittings directly to pressure relief valve.

Modification of the pressure relief valve for Buchi or Biotage system.





### Solid load

The large column adapter also enables the use of Redi*Sep* solid load cartridges (25 g and larger). Although a solid load cartridge is being used on the large column adapter, the flash system is programmed to run as a liquid load so the pumps are stopped at the correct time.

- 1. Install the column and set the knob on the large column adapter to "Column."
- 2. Place the solid load cartridge in place with the proper fittings and connect the tubing.
- 3. Start the run, choosing "Liquid load." After equilibration ends, the pumps will stop.
- 4. Switch the valve on the large column adapter to "Cartridge and Column."
- 5. Tell the flash system the sample has been injected to restart the pumps.

## Liquid Load

For liquid injections, the run is started using "liquid injection". When the pumps stop, the column can be loaded using the usual injection port on the flash system or the liquid injection adaptor that comes in the Large Column Adaptor Accessory Kit.

#### Conclusion

It is easy to run Redi*Sep* columns on any flash chromatography system and have the advantages of cost, selectivity, loading capacity, and resolution. Redi*Sep* columns set the standard for column sizes, so they can be easily run using existing methods.



