

Using RediSep® Columns Everywhere!

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 technical 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 (LCA), and how to use Teledyne ISCO solid load cartridges.

Columns

For the purposes of this technical note, there are two types of flash systems. The most common type holds the column 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™ inlet fitting, and a Luer-slip outlet, which 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™ 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 Luer) works well.

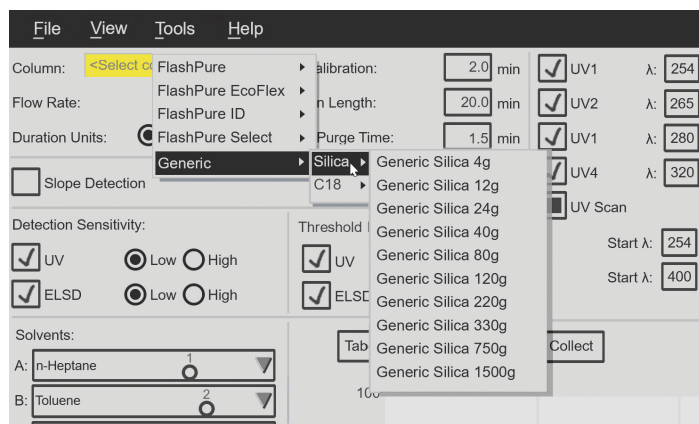


RediSep column installed on a system with an injection port shuttle

RediSep column installed on a flash system without a column shuttle

Flash system software

RediSep 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.

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

Solid Sample Loading

Note

The solid load cartridge should be attached to the system after equilibration is complete.

Solid Sample Loading with a Column Shuttle

Additional parts are needed to run a RediSep solid load cartridge on a column shuttle system:

1. To the inlet line, a female Luer to 5/16-24 male adapter (IDEX™ PN P-661) and a male Luer to 1/4-28 female adapter (IDEX PN P-655) are needed.
2. The adapters can be hand tightened to each other.
3. The top of the P-655 adapter can then be hand tightened to the inlet line.
4. A female Luer to 1/4-28 male adapter (IDEX PN P-604) is also needed at the bottom of the solid load cartridge.
5. The bottom of the P-604 adapter will be hand tightened to the top of the column holder.



RediSep Solid Load Cartridge setup on system with a column shuttle

Solid Sample Loading with Tubing Column Connections

Additional parts are needed to perform solid loading on a tubing connection system with a RediSep solid load cartridge:

1. The female ending on the inlet line needs a female Luer to 5/16-24 male adapter (IDEX PN P-661). This adapter can be hand tightened to the end of the inlet line.
2. Once in place the adapter can be hand tightened to the solid load cartridge cap.
3. The bottom of the solid load cartridge will attach to the top of the column. A 1/4 turn is sufficient to secure the cartridge to the column.



RediSep Solid Load Cartridge setup on system with tubing connections.

Large Column Adapter

The large column adapter (LCA, PN 60-5394-563) is used to run columns larger than 415 grams on Teledyne ISCO's CombiFlash NextGen systems or non-Teledyne ISCO flash systems.

Large Column Adapter in a Column Shuttle

In column shuttle systems, the LCA can attach to the system just as a column would. Pull up on the column block (NextGen) or the clamping plate and insert the male ending of the LCA into the base of the column shuttle. When lowering the top of the column shuttle, ensure the top of the LCA is attached to the inlet line.



Large column attached connected to a column shuttle on a CombiFlash NextGen 300+

Large Column Adapter with Tubing Connection

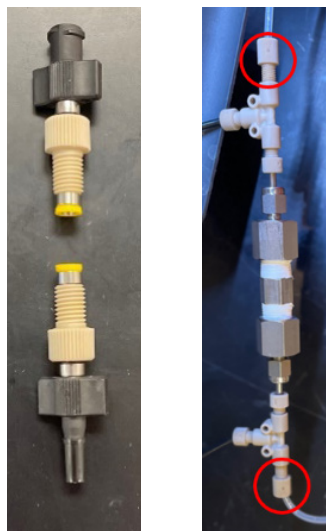
Some flash chromatography systems like the Biotage Selekt will connect to the column with only tubing. The LCA needs to be modified for these systems. Remove the black endings of the LCA and replace them with the inlet and return line. Make sure that the inlet line replaces the female Luer ending and that the return line replaces the male ending. 1/4 - 28 connections should be used to connect the LCA.

Large Column Stand Setup

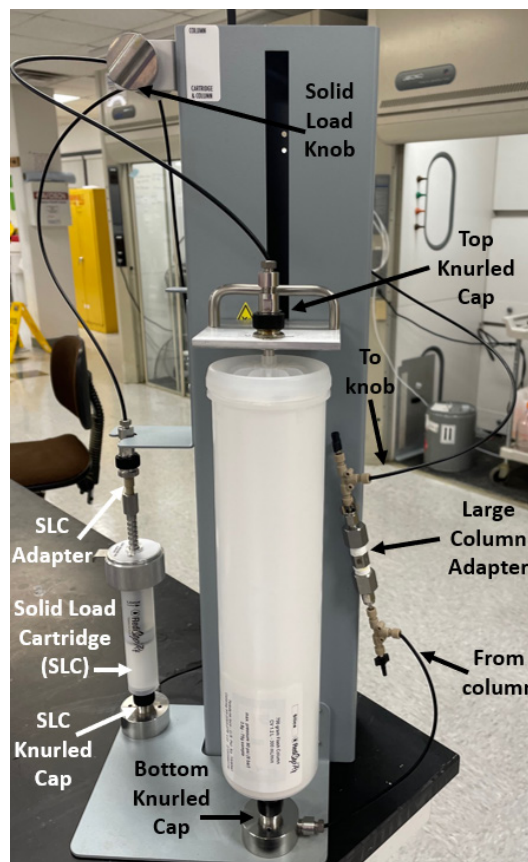
The large column stand (PN 60-5394-551) is used to hold the larger columns and attaches to the LCA. The tee unions on each side of the LCA are hand tightened to lines that lead to and from the column. The tee union nearest the female fitting on the LCA should connect to the "To knob" line, and the other tee union should connect to the "From column" line.

The column will be placed between the top and bottom knurled caps. Different Luer adapters are needed to run different size columns. There are two different male Luer adapters for the top knurled cap: PN 60-5243-0075 for columns $\leq 330\text{g}$, and PN 60-5243-074 for columns $\geq 750\text{g}$. The bottom knurled cap has three adapters for different size columns: PN 60-5244-116 $\leq 330\text{g}$, PN 60-5244-126 750g to 1.5kg, and PN 60-5244-118 for 3kg columns. The knurled cap must be unscrewed to place the adapters into their ports.

Some of the bottom knurled cap adapters can also be used for the solid load knurled cap. PN 60-5244-116 can be used for solid load cartridges (SLC) $\leq 260\text{g}$ and PN 60-5244-126 $\geq 375\text{g}$. The top of SLC knurled cap needs a PN 60-5244-058 adapter when using 25 and 65g SLC caps.



Left: Removed black endings.
Right: The red circles indicate the 1/4 - 28 fittings attached to LCA.



Large column adapter and solid load cartridge set up to the large column stand.

Solid Load Knob

The solid load knob makes it possible to perform solid loading by switching the flow path. Four lines are attached to the back of the solid load knob. By switching the knob from COLUMN to CARTRIDGE & COLUMN, the user switches the flow path from blue to red as shown in Figure 4.

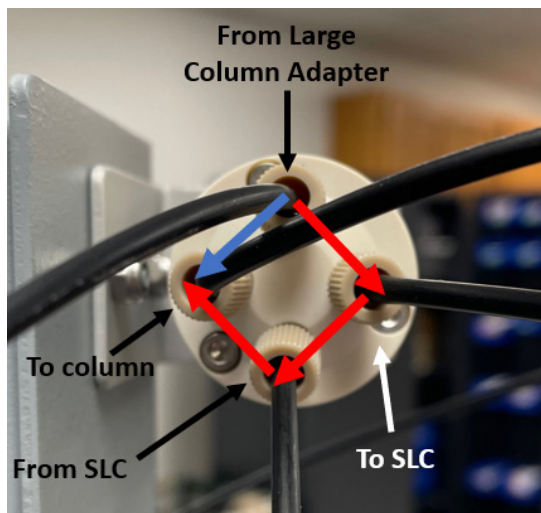
To perform solid loading while using the large column adapter, the user should select "Liquid load" when setting up the flash system. The solid load knob should be in the COLUMN position, and equilibration can begin. Once equilibration is over, the knob can be switched to CARTRIDGE & COLUMN. The flash system can then be told the sample has been injected.

Liquid Load

Liquid loading can be performed as usual. For column shuttles, the column block can be lifted, and the sample can be added directly to the top of the LCA. For systems that only use tubing, the top of the LCA can be disconnected to the inlet line, and then the sample can be loaded.

Conclusion

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



Back of the solid load knob with tubing attached. Blue arrow indicates flow path when the knob is set to COLUMN. Red flow path indicates flow path when the knob is set to CARTRIDGE & COLUMN.

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