

Solvent Changing

on CombiFlash Systems

Chromatography Application Note

AN28

Abstract

Users need to change solvents on *CombiFlash* systems to convert between normal phase and reverse phase. Methods that automatically flush the system, including the solid load cartridge lines, allow an easy and complete change in solvents for various *CombiFlash* systems.

Overview

Incomplete solvent changes cause unpredictable purification of compounds (Figure 1).

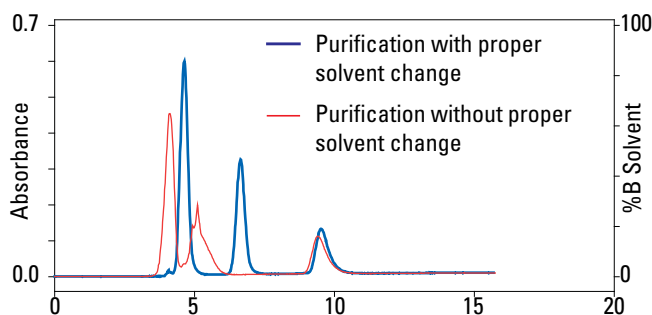


Figure 1: Normal phase purification after reverse phase purification with and without proper solvent change.

When changing between normal phase and reverse phase, the system needs to be flushed with an intermediate solvent that is miscible with all solvents being used. Solvents such as 2-propanol or acetone are miscible with all normal phase and reverse phase solvents. The system needs to be flushed a second time with the solvents to be used in the purification. Therefore, a total of two flushes are required when changing between normal and reverse phase.

The various methods for flushing solvents are described for various *CombiFlash* systems. You may find that the method is already installed on your system. If so, you only need to follow the “Using the Method” instructions for your system.

CombiFlash Rf-200 and Rf-75

Creating the “ChangeSolvent” Method

1. If still installed on the system, remove the column and solid load cartridge.
2. From the *PeakTrak* menu, select **File>New** to open a new default method for a 4 g silica column.
3. Select the **Method Editor** menu option to open the advanced settings.

4. Toggle the **Time to CV/CV to Time** run units so that it reads **Time to CV**.
5. Set the **Flow Rate** to 100 mL/minute.
6. Set the **Equilibration Volume** to 0.0 mL.
7. Set the **Air Purge** to 0.5 minutes.
8. Set the **Peak Collection** option to **NONE**.
9. Touch the **Edit Gradient** button and define a gradient to match Table 1.

Table 1: Solvent Changing Gradient for CombiFlash Rf 200 and Rf 75

Segment Length (minutes)	%B
Initial	0
0.3	0
2.9	100
0.5	100
0.0	0
0.3	0

10. Touch the **Save As** button and name the method **ChangeSolvent.mtd**.

Using the Method

1. Install a priming tube (PN 209-0165-46) in place of a column.
2. **Rf 200**—Install an empty (no media and sample) solid load cartridge on the Rf 200 system. Push the plunger all the way down into the cartridge. This cartridge may be reused for this purpose for future solvent changes.¹
Rf 75—Connect the solvent delivery tubing to the injection port on the upper column mount.
3. Place a rack in the fraction collector.
4. From the menu, select **File>Open** and open the **ChangeSolvent.mtd** method file.
5. Name the run with a generic, temporary name such as “Purge.” Use of such a name avoids saving a number of files with default file names.

1. Alternatively, an adapter that replaces a solid load cartridge and cap for this application can be made from the following parts available from Idex Health and Science (formerly Upchurch Scientific): P-135 (1/4-28 to 5/16-24 flat bottom fitting adapter) and P-683 (1/4-28 to Luer adapter). These fittings are connected together to create an adapter for connecting the solid load cartridge line to the inlet at the top of the Rf-200 injection valve.

- Place the solvent inlet lines you want to flush into the new solvent containers. You will need about 300 mL of each solvent. Make sure the correct solvents (A1, A2, B1, B2) are chosen to match the solvent lines and containers (either the intermediate solvent or the final solvents).
- Touch the Play button to open the Run Requirements window.
- Rf 200**—Select the Solid Load Sample Loading Technique.
Rf 75—Select the No Equilibration Sample Loading Technique.
- Start the run. When it finishes, the system has been flushed with the new solvent(s).
- Select the File>Delete menu command and delete the run file just generated.
- If using intermediate solvents, repeat steps 6 through 10 with the final solvents.
- From the menu, select Tools>Configuration and enter the new solvent names.
- If using a Foxy R2 fraction collector, install a rack. If the system is configured for manual fraction collection, ensure the outlet is diverted to a waste container.
- From the menu, select File>Open and open the ChangeSolvent.mtd method file.
- Name the run with a generic, temporary name such as “Purge.” Use of such a name avoids saving a number of files with default file names.
- Place the solvent inlet lines you want to flush into the new solvent containers. You will need about 500 mL of each solvent.
- Touch the Play button to open the Run Requirements window.
- Select the Solid Load Sample Loading Technique.
- Start the run. When it finishes, the system has been flushed with the new solvent(s).
- Select the File>Delete menu command and delete the run file just generated.
- If using intermediate solvents, repeat steps 6 through 10 with the final solvents.
- From the menu, select Tools>Configuration and enter the new solvent names if necessary.

CombiFlash Torrent

Creating the Torrent “ChangeSolvent” Method

- Remove the column and solid load cartridge if still installed on the system.
- From the menu, select File>New to open a new default method for an 80 g silica column.
- Select the Method Editor menu option to open the advanced settings.
- Toggle the Time to CV/CV to Time run units so that it reads Time to CV.
- Set the Flow Rate to 250 mL/minute.
- Set the Equilibration Volume to 0.0 mL.
- Set the Air Purge to 1.0 minute.
- Set the Peak Collection option to NONE. If the system is not configured with a Foxy R2 fraction collector or fractionation valve, select Manual.
- Define a gradient to match Table 2.
- Touch the Save As button and name the method ChangeSolvent.mtd.

Using the Method

- Install a priming tube (PN 209-0165-76) in place of a column.
- Connect the solid load cartridge tubing to the sample injection port.

Table 2: Solvent Changing Gradient for CombiFlash Torrent

Segment Length (minutes)	%B
Initial	0
0.5	0
1.0	100
0.5	100
0.0	0
0.5	0

Conclusion

Running solvent change methods are a convenient way to change solvents. The steps described in this application note effectively purge the fluid path to prepare the CombiFlash system for runs using new solvents.

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