

CombiFlash® NextGen Quick Start Guide

Overview

These quick start instructions assume that:

- The system has been installed according to Section 2 of the User Manual.
- The operator is familiar with the safety warnings in Section 1 of the User Manual.
- The system will be operated from the touch screen display.

Quick Start Instructions

1. Perform Thin Layer Chromatography (TLC) on your sample to verify a suitable column media and solvent system, and the difficulty of the separation (ΔR_f of the compounds being separated).
2. Select a RediSep® Rf column size appropriate for your sample amount. If the separation is difficult ($\Delta R_f < 0.2$), your sample loading should be near the low end of the selected column's range.
3. Raise the column mount trolley from its lowest position and insert the RediSep Rf column on the CombiFlash NextGen system.
- If the column is not automatically recognized by the RFID reader (or the system does not have this option), you can manually select the column media and size from the drop menu in Figure 1.

Figure 1: Column Select menu

4. The system will load a universal method for the selected column size and media.
 - If using RediSep® Rf Gold columns you will be given the option of a Gold Speed or Gold Resolution method.

Figure 2: Gold Speed and Gold Resolution option upon RFID detection of RediSep Gold column

- If you want to optimize the loaded default method, you can do so by selecting METHOD EDITOR from the top menu.
5. Choose a solvent system from the menu shown in Figure 1.
 6. On the main window, select the PLAY button, bringing up the MINIMUM RUN REQUIREMENT screen.

Figure 3: MINIMUM RUN REQUIREMENT screen and loading options

7. From the MINIMUM RUN REQUIREMENT screen, you will be offered several loading options:
 - If the sample is soluble in the starting mobile phase and the sample will be injected choose LIQUID.
 - If the sample is not soluble in the starting mobile phase, use a solid load cartridge.
 - If the solid load cartridge is already prepared and loaded on the system, then choose SOLID.
 - If the solid load cartridge has not been loaded on the system yet, then choose SOLID (Pause) which will equilibrate the column while you prepare the cartridge and will pause until you have installed the loaded cartridge.
 - If the sample is already loaded directly onto the column media, then select NONE (on column).

Note

Systems without an injection valve will only show LIQUID, SOLID (Pause) or NONE (On Column) as loading options. This includes the NextGen and NextGen 300 systems without the optional injection valve.

8. Click OK to begin the run. If you selected:
 - LIQUID or SOLID (Pause) loading please continue to Step 9.
 - Solid loading please continue to Step 12.
 - None (on column), equilibration will be skipped and continue to Step 12.
9. The system will equilibrate the column. During this time, prepare the sample.
10. The NextGen will pause after performing the column equilibration. It will ask you to inject the sample or load the solid load cartridge.

Figure 4: Pause for solid load cartridge or liquid injection

11. Once the sample is loaded onto the system, click OK to begin the run.

Use and Disclosure of Data

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12. As the run begins and progresses, you will be able to monitor the UV absorbance (or UV-Vis or other enabled detector options depending on the system configuration) trace.
13. From the run screen you can:
 - Modify the gradient without stopping the separation.
 - Change the fraction collection settings to ALL, PEAKS, or NONE.
 - Open the METHOD EDITOR screen to change additional method parameters.
 - Insert an isocratic hold.
 - Advance to the next tube.
 - Halt the run.
 - Terminate the run.
14. Once the run is completed, an END OF RUN screen will appear, depending on whether or not you chose END OF RUN HOLD earlier in the MINIMUM RUN REQUIREMENT screen.
 - If you chose END OF RUN HOLD a message will appear asking if you want to continue with air purge or extend your run.

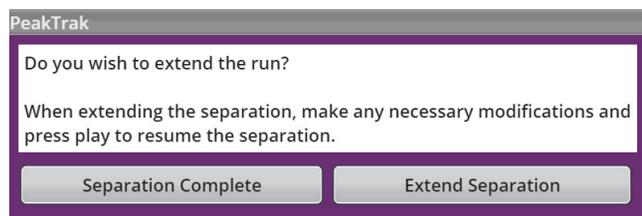


Figure 5: Dialog at end of original method if END OF RUN HOLD is enabled

- If you did not choose END OF RUN HOLD, then the system will automatically wash the injection valve and air purge the column and solid load cartridge (if used).
15. As the system undergoes its valve wash and air purge processes, a report screen will appear. Here you can edit your report and save or print the pdf of the report (if enabled). Additionally, you have the option to use the completed method as modified for another separation or scale sized column.

Solid Load Cartridge Cap Instructions

Ensure the empty cartridge has a bottom frit. Prepare the media and sample:

To prepare an empty solid sample cartridge.

1. Dissolve your sample in a minimal amount of a suitable, volatile solvent.
2. Place the media into the solvent. If using silica gel, a particle size of 40–60 μm (240–400 mesh) is recommended. The amount of silica required is about four to five times the mass of your sample.
3. Agitate the solvent for a moment to allow the sample to adsorb to the silica.
4. Remove the solvent with a suitable method, such as rotary evaporation. Alternatively, you can dry the prepared cartridge after step 5 by drawing the solvent out using a vacuum or using the NextGen system's air purge (if equipped).
5. Load the media and sample mixture into the cartridge. Tap the cartridge on the bench top to settle the mixture.
6. Place a frit on the top of the cartridge. Force the frit down against the mixture using a plunger suitable for the car-

tridge (a plunger is included with the solid load cartridge cap).

7. Wipe any residual powder inside the neck of the cartridge.

Note

You can remove solvent from a prepared cartridge by attaching the cartridge cap and manually purging (TOOLS > MANUAL CONTROL) the cartridge with air.

After you have prepared the pre-filled or empty cartridge, place the solid sample cartridge on the system:

8. Attach the desired adjustable cartridge cap:
 - 60-5237-047, fits 2.5 and 5 gram solid load cartridges.
 - 60-5237-048, fits 12 and 20 gram solid load cartridges.
 - 60-5237-044, fits 32 and 65 gram solid load cartridges.
9. Press the lever on the side of the cap and fully extend the plunger (Figure 6).

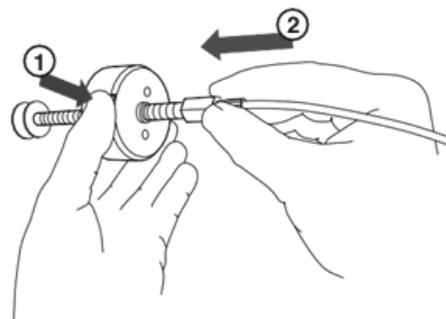


Figure 6: Press the lever and extend the plunger

10. Slide the plunger into the cartridge until it reaches the top frit (Figure 7).
11. Press the lever and push the cartridge into the cap. Align the cartridge so that it fits fully into the recess in the cap (Figure 7).
12. Load the solid sample cartridge with cap on the sample injection port.

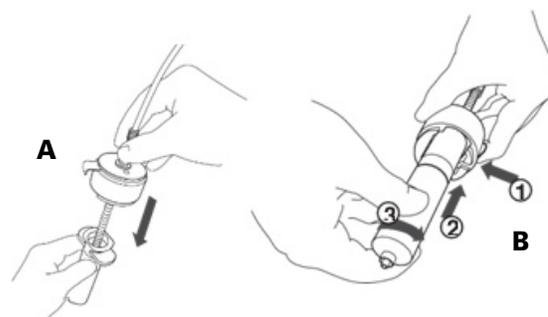


Figure 7: In 7a, Insert the plunger into the cap; and 7b Align and push cartridge into the cap, and then rotate the cartridge to secure it.

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