

# Flowcell Care and Cleaning

for the CombiFlash® Companion®

**Chromatography Technical Note**  
**TN01**

## Overview

For optimum performance, the CombiFlash Companion detector flowcell must be clear of any obstructions or build-up of residual sample material. Such problems primarily result from low solvent polarity. When solvent polarity is too low, compounds may precipitate or sample material may not be flushed from the system before the next purification.

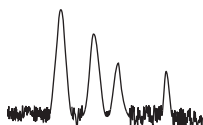
If the flowcell is obstructed, or performs poorly as a result of residual compounds, the Companion displays an error message. The error message identifies the fault and prompts the user for a response before continuing with the run.

This technical note describes the error messages and corrective actions, and advises of measures that can prevent the fault conditions.

## Flowcell Error Messages

Flowcell cleaning is recommended. Continued operation without cleaning may result in a noisy trace.

If the flowcell is partially obstructed the above message is displayed. Continuing to operate the system without servicing the flow cell may cause excessive noise in the trace (see Figure 1) or reduce amplitude of absorbing species.



**Figure 1: Noisy trace**

The detector lamp is not on. You can continue the separation without detection, or cancel. 280@81

When the above message occurs at the beginning of a run, either the lamp is not working at all, or the flow cell is blocked to a degree that the detector is not receiving adequate light. The number preceding the “@” symbol at the end of the message indicates which of the two faults is likely.

- ≤ 200: Contact Isco Technical Service. The lamp may not be working at all.
- > 200: The lamp is working but the flow cell requires cleaning.

In the example the number is 280, indicating that the flow cell should be cleaned.

## Flowcell Cleaning Solvents

The flowcell can be cleaned by pumping a solvent through the system. Generally, the recommended solvent is the highest polarity solvent you have recently used. Therefore, ethyl acetate (often solvent B) would be used. See Table 1 for alternative solvents. Recent synthesis solvents should also be considered.

**Table 1: Suggested Flow Cell Cleaning Solvents<sup>a</sup>**

Solvent	Description
Methanol	Miscible with most LC solvents. It will force other solvents through the system so that they will not interfere with the next sample. Breaks down the tertiary structure of silica and allows it to flush through.
Acetone	Common organic solvent with high dissolving properties.
Dichloromethane (DCM)	Effective at solubilizing organic molecules.
Dimethylsulfoxide (DMSO)	Strong solvent that will clear organic sample.
Dimethylformamide (DMF)	Strong solvent if other solvents do not produce the desired results.
Tetrahydrofuran (THF)	Strong solvent.
Mildly acidic aqueous solution	0.5M acetic acid in distilled water can be used to clear samples that are not removed by organic solvents.
Mildly basic aqueous solution	A dish-type detergent solution can wash through particulates not picked up by the acidic solution. The emulsification effect of detergent is an effective tool as well to help wash through oily samples. Follow the detergent with several minutes of clean, distilled water.

a. Always handle solvents in accordance with the manufacturer's or supplier's MSDS.

## Quick Flowcell Cleaning

*Quick cleaning* is advantageous because very little solvent is required and repriming is not necessary. To quick clean the system:

1. Remove the column from the system.
2. With a syringe, inject the selected solvent into the lower column mount.
3. Insert a bypass tube in place of the column.
4. From the PeakTrak menu select the Tools>Manual Control... command and pump 100 mL of Solvent A at 100 mL/min.
5. Replace the bypass tube with a column and start a run. The Companion will attempt to calibrate the flow cell.

- If the calibration is successful, the system is ready for use.
- If the alert message persists, refer to the thorough flowcell cleaning solvents and techniques.

## Thorough Flowcell Cleaning

To thoroughly clean the flowcell:

1. Remove the column from the system and insert a bypass tube.
2. If using an alternative solvent, place the solvent inlet line into a container of the selected solvent.
3. Pump 100 ml of solvent through the system at 100 mL/min.
4. If an alternative solvent was used, return the solvent inlet line to the appropriate solvent container.
5. Select the Tools>Auto Prime menu command to prime the system.
6. Replace the bypass tube with a column and start a run. The Companion will attempt to calibrate the flow cell.
  - If the calibration is successful, the system is ready for use.
  - If the alert message persists, refer to the alternative flow cell cleaning solvents and techniques.

The solvent strength and high flow rate will typically clear the obstruction and return the flow cell to optimum performance.

## Alternative Cleaning Techniques

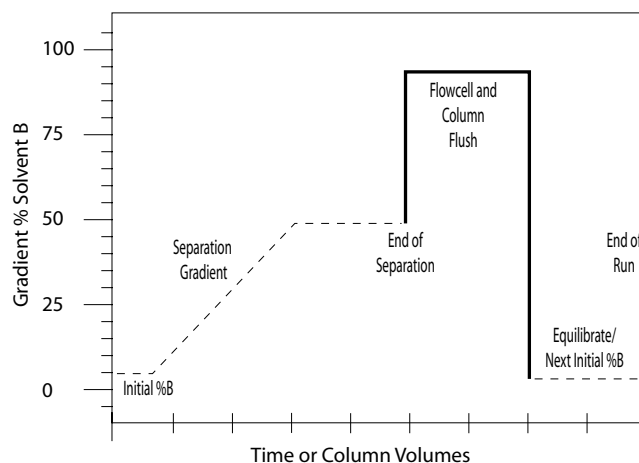
If the thorough flow cell cleaning instructions do not clear the obstruction, the following alternative techniques should be considered.

- Pump water/DMF/methanol (50/25/25) for 3–4 minutes followed by pumping straight methanol (or ethanol) for 2–3 minutes.
- Pump 5–10 mL DMSO followed by methanol, ethyl acetate, then hexane.
- After the lines have been filled with solvent, reduce the flow rate or pause up to 18 hours. Doing so will allow the solvent to remain longer to dissolve the obstruction.

## Preventive care

Often the only maintenance required is to operate the instrument with a bypass tube in place at a high flow rate with a high polarity solvent.

With typical gradient configurations under normal phase conditions, the B solvent has the highest polarity. Adding a high percentage of B solvent (much higher than is necessary to elute the sample) at the end of the separation ensures that all of the sample material is flushed through the flowcell. This can be added to the Companion's default methods so that flowcell cleaning occurs every run (see Figure 2).



**Figure 2: Flowcell flushing at end of run**

## Summary

A clean flowcell ensures optimum peak detection performance.

Pumping a high polarity solvent through the system at the end of each run may prevent flow cell problems.

If the Companion does detect a flow cell problem, determine the cause according to the displayed error message, then follow through with the recommended corrective action as soon as possible.

*Last modified July 15, 2005*

### Teledyne Isco, Inc.

P.O. Box 82531, Lincoln, Nebraska, 68501 USA  
 Toll-free: (800) 775-2965 • Phone: (402) 464-0231 • Fax: (402) 465-3001  
 E-mail: [service@isco.com](mailto:service@isco.com)