

Why do I have higher pressure at a lower flow rate in my HPLC runs - FAQ

All other factors being equal, the pressure should not increase when the flow rate is lower. Therefore there must be something else causing the increased pressure. Without more detail on the situation, it is difficult to determine with certainty.

Some possible explanations for the higher pressure are:

- **Blockages in the frits.** If this is the case, use a disposable pre-column, *Cogent Column Filter*. Replace it as soon as you see an increase in Pressure.
- **Blockages in the HPLC system.** These often occur in the pump. To determine if this is the case, run the pump without the column attached. If you see significant pressure without the column, it's a good indication that there is a blockage in the HPLC system.
- **Differences in columns.** Are you using the same column in both cases? Smaller ID, longer length, or smaller particle size columns will all produce higher pressures. Even a C18 phase from one column manufacturer may produce a different pressure from a C18 phase from a different manufacturer.
- **Particulates in the sample.** Be sure that your sample has been properly filtered to ensure that no particulates are being injected into the system. The same goes for your mobile phase as well. All mobile phase solvents should be vacuum filtered, especially if they contain a buffer such as ammonium acetate.
- **Different mobile phases.** Does the low flow rate method use the same mobile phase as the higher flow rate method? Every solvent system will have a different viscosity, and hence will produce a different pressure.
- **Temperature fluctuations.** Is the column thermostatted? If not, changes in lab temperature can affect the pressure. Lower temperatures can result in higher pressure.
- **Differences in instruments.** Is the same Instrument being used in both cases? The system plumbing, such as the tubing ID, can have a significant effect on pressure.
- **Pump qualification.** Are you sure that the pump is really delivering 0.3 mL/min, for example? We have an HSO Kit™ used for ensuring your instrument is calibrated and qualified properly, not just in terms of pump accuracy but also for other pertinent instrument parameters.