

Normal phase HPLC column regeneration

Here are some general procedures to follow to regenerate normal phase columns:

- 1. Pass between 20 to 30 ml of the weakest solvent in your mobile phase, such as hexane or chloroform.
- 2. Then, perform a gradient of chloroform or hexane and isopropanol from 100:0 to 0:100 in about 10 minutes and hold this phase for about 1 hour.
- 3. Perform the reverse process and the column will now be ready to use.

Some notes:

- If the column allows it (like most modern columns), you can perform the above process with the column inverted (don't forget to disconnect the detector!), to remove particles.
- If water contamination is suspected, you can wash with hexane/dimethoxypropane/glacial acetic acid at 95: 2.5: 2.5 for about 1 hour at 1 ml/min, which will restore the original selectivity.