

Solving baseline problems in Aqueous Normal Phase ANP methods – Tips & Suggestions

When using a gradient method with the Cogent Diamond Hydride™ HPLC column and an inconsistent baseline may be observed...

The following are some possible causes of the issue along with suggestions to fix it.

1. *Is the mobile phase filtered before use on the instrument?* A 0.45µm nylon membrane filter with vacuum filtration is recommended such as our [SOLVFil easy apparatus](#). This is especially important for mobile phases that contain dissolved solids such as ammonium acetate. It is good practice to filter any mobile phase though.
2. *How many runs were tried?* When ammonium formate / acetate is involved in the mobile phase, it sometimes can take three runs or so to get a reproducible baseline.
3. *How much post time is used in the gradient?* Please allow a sufficient post time to insure the mobile phase is fully equilibrated with the starting gradient solvent conditions before each injection. We suggest at least 5 column volumes.
4. Try injecting multiple blanks and see if the baseline becomes more consistent. The inconsistent baseline may be due to carryover of strongly retained material in the sample that slowly elute in subsequent runs. If this is the case, your method will need to incorporate stronger eluting conditions at the end of the gradient program to wash these compounds out so that they do not carry over into the next run.



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