

Eliminating shoulder peaks in an HPLC method using Cogent RP C18 columns - Tips & Suggestions

With regards to your question about the shoulder peaks observed using the Cogent C18 HPLC column.

First I would like to ask you about the **shoulder peaks**. Have you determined whether these are due to distortion of the peaks of interest or elution of contaminants? There are several ways you can tell the difference. One is to spike your sample with a standard of the peak of interest. Assuming the standard is not contaminated, the results will tell you which of the two cases it is. If it is a peak shape distortion issue, both the main peak and the shoulder peak will increase in area. If it is a contaminant elution issue, only the main peak will increase in area while the shoulder peak will stay the same. You could also try running a blank and see if you observe the contaminant peaks. If you are using Photo Diode Array detection, you can also use the "Peak Purity" feature if your software has it to check whether your main peak does not have any coelution issues.

The reason why I mention both possibilities is even though you may not have observed the peaks in your data before, it could still be a peak shape issue if damage has occurred to the packing over time. Has the column been exposed to any aggressive mobile phases (such as high pH)? This for example could partially dissolve the packing bed, creating voids which will show up as distorted peak shapes.

If it is a **contaminant issue**, reversing the column direction and using the 50/50 MeOH/H2O washing like you have been doing would be recommended. The buildup of contaminants occurs mostly at the head of the column, so back flushing would be the recommended way of cleaning the column. You could also try 50/50 IPA/H2O as well. IPA works very well for cleaning contaminants from the column. Also, contaminants are not necessarily from the column and could be from the injector, vials, or other parts of the HPLC system.

Click <u>HERE</u> for ordering information and pictures of the Cogent RP C18 columns.



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