

# Filtration in HPLC

One of the crucial points for the longevity of columns and HPLC systems is the filtration of whatever we insert into an HPLC system, whether samples or eluents.

It is essential not to forget that a typical HPLC column uses 1.7 $\mu$  to 10 $\mu$  particles as stationary phase, so the interstitial space is quite small and susceptible to be filled by particles in suspension, either in the sample or in the eluent.

On the other hand, to generate high pressures and maintain their tightness, the pumps and injection valves of an HPLC system use seals, retainers and valves that can be damaged if subjected to particles from the eluents and, in the case of sample injectors, of the samples.

As such, it is essential that all samples and all eluents are filtered with suitable filters.

**The choice of filters depends on several factors:**

- The sample solvent or mobile phase eluent (aqueous, organic or mixture)
- Column particle size (greater than or less than 3 $\mu$ m)
- Sample volume to filter

If the solvent is 100% organic, the filter membrane to be used must be made of PTFE, as it is virtually inert to most organic solvents. However, it should not be used for aqueous solvent filters or mixtures with aqueous solvents, since being hydrophobic, the pressure to filter can lead to the rupture of the membrane.

For aqueous solvents, the best option is cellulose acetate (CA), compatible with most salts and buffers used in HPLC, being a cost-effective membrane.

For solvent mixtures, for example in isocratic methods, the most economical option is Nylon membranes, which can also be used to filter mixture of aqueous solvents / buffers and organic solvents such as Methanol and Acetonitrile.

Another option available are "universal" membranes: this category includes regenerated cellulose (RC) and hydrophilic PTFE membranes, excellent for most solvents and mixtures but whose cost is high – about twice as their equivalent in CA, PTFE and Nylon.

Another factor to take into account is the pore size, which is directly related to the particle size of the column. The ideal is to always use 0.22 $\mu$  filters, but sometimes, due to the viscosity of the solvents, filtration is slow. Thus, if the methods use columns of 4 $\mu$  particles or bigger, there is no inconvenience in using 0.45 $\mu$  filters.

With regard to samples, they must always be filtered through syringe filters. The choice of their diameter depends on the sample volume to be filtered:

- if your sample is less than 3ml, use 4mm syringe filters.
- if your sample is between 2 to 10ml, use 13mm syringe filters.
- if your sample is between 10 to 100ml, use 25mm syringe filters.

## Points to remember:

- never use any solvent that is not filtered by you or a colleague: do not rely on the solvent bottles that say they are filtered or in the water that comes out of the purification systems, as the filters can be 0.8 $\mu$ . You may also add a buffer to the water or phase modifier, so your water must be filtered after this addition.
- never inject a sample without first being filtered through a syringe filter.
- see our Filter Selection Guide for detailed information on the chemical compatibility of filtration membranes.