

Don't Forget the Filter

Paying attention to the finer points of filtration in sample preparation ahead of HPLC analyses can help avoid simple mistakes that lead to unnecessary downtime and a loss of productivity.

The pressure to increase the number of samples processed is being felt across almost all lab environments. Sacrificing quality is not an option, so what to do? We all know that efficient analysis starts with solid sample prep. Though establishing a proper methodology and employing the right materials will produce reliable and accurate results, failure in even the most basic aspects of sample preparation can result in misleading outcomes, invalid results, the need to repeat HPLC analyses and, ultimately, a loss of time and productivity.

We spoke with Giles Barton, Global Lead Product Specialist-Lab Filtration at GE Healthcare Life Sciences about maximizing a key step in sample prep – filtration. With nearly three decades in the field, Giles is perfectly placed to help you avoid filtration fiascos and achieve smooth sample prep.

What's your background?

I've been involved in providing product and application support for the Whatman filter portfolio for the last 27 years. I originally trained as a biochemist and worked as a bench scientist in academia, using a variety of analytical techniques including HPLC, GC and TLC.

What are the biggest sample prep problems facing labs?

Laboratories are under increasing pressure in terms of time and cost, often dealing not only with high numbers of samples but also samples of varying natures.



Sample preparation is an area where subtle differences between sample types or the sample preparation methods and materials used can impact the results obtained. Ensuring compatibility between the materials in the filtration device and the solvent can minimize issues; for example, the levels of extractable compounds that might interfere with analyte detection.

What happens when sample prep goes wrong?

Prior to analytical high-performance liquid chromatography (HPLC) applications, samples are prepared by a variety of methods, all of which have the potential to influence analytical results by affecting the sample injected into the system. Ineffective sample preparation can lead to reduction in column life, increased service requirements for components, potential downtime of instrumentation, reduction in chromatographic separation efficiency and reproducibility, and many other problems.

At first glance, filtration seems like a simple process...

Although filtration is a seemingly small part of a large workflow, the impact of choosing the right filter for a given application can be

substantial. For example, methods that do not remove sufficient particulate matter can result in column blocking. Inappropriate choice of sample preparation devices could, under certain circumstances, release extractable compounds into the sample, bind analytes of interest or introduce errors through sample loss – all influencing the quality and reproducibility of the results observed. If you are confident that your filtration consumables are not contributing to errors that require rework or root-cause investigations, you will save money and give yourself peace of mind.

What are the most common sample prep 'fails'?

Incorrect filter membrane choice is one problem we see a lot. Issues can appear as extraneous peaks from extractables, which can interfere with detection of the analytes of interest. Incompatibility of filter media with a sample or solvent in terms of chemical resistance is not solely limited to dissolution effects on the filter media. Solvents can cause slight swelling of polymeric membrane materials when not fully compatible, which in turn can affect the filtration performance, leading to ineffective clean-up of samples. One way to address this issue is by choosing a membrane that



is broadly compatible with both aqueous and organic solvents. For typical HPLC sample preparation, syringe and syringeless filters containing regenerated cellulose membranes are a good option because of their broad solvent compatibility.

Use of a filter device that is larger than needed for the volume being prepared is another common pitfall. Use of too-large filter devices means that small amounts of sample will remain in the filter, even after precautions are taken to expel as much as possible.

What are the advantages of taking the filtration step seriously?

A fundamental goal of most laboratories engaged in analytical testing is to develop consistent processes that underpin consistent

results – and that requires consistent filtering. Selection of a filtration device with the right pore size and retention characteristics helps ensure effective particulate removal, while choice of appropriate size and design of filter unit can help minimize sample loss. Good filtration also helps to protect the column and potentially extends the lifetime of both the injector and the column.

What's your advice to readers looking to improve filtration?

Research data suggests that users should consider standardizing membranes across multiple encapsulated formats. Standardization reduces the variation associated with sample preparation on multiple membranes and increases lab process consistency.

Reader Survey – Sample Prep for HPLC*

We asked Giles to comment on some of the key findings in our HPLC Sample Prep Survey.

> A vast majority (94%) of the respondents considered proper sample preparation to be important in obtaining accurate results.

Recent conversations I've had with customers reinforce the importance of sample preparation. However, not all sample prep is created equal – a key factor is ensuring that the choice of method is appropriate for the analytical method being used.

>The most important reason for preparing samples properly prior to HPLC analysis was to ensure the quality

of results (67%), followed by simply good lab practice (12%), preserving chromatograph column life to reduce costs (10%), adherence to SOP or regulated method (5%), and minimizing chromatograph downtime (4%).

Allowing the chromatograph to operate at optimum speed was cited by only 2% of the respondents.

This bears out what I hear from customers – there are a variety of different goals in terms of the sample preparation step, and individual circumstances will have a bearing on the importance of each, but quality of results is paramount.

>Survey respondents felt that the areas with the greatest need for improvement in sample preparation were reproducibility (27%) and laboratory/workflow efficiency (24%).

Laboratories are under time and cost constraints to provide reproducible results in a timely manner and reduce the need for repeat analyses. Pay attention to optimizing

Flawless Filtration

1. Choose filter media type based on consideration of the solvent type.
2. Consider possible interactions between sample analytes of interest and filter media.
3. Ensure appropriate choice of pore size based on particle size of the analytical column
4. Do not use a filter device larger than required for the sample volume being prepared.
5. For difficult-to-filter samples, consider using a filter device with efficient pre-filters incorporated to increase sample volume handling, rather than using multiple filter units to prepare samples.
6. In general, it is best to filter the sample slowly and gently.

the sample preparation step, both in terms of sample cleanliness and streamlining the sample preparation workflow, and help provide optimal lab economy.

>46% of respondents agreed with the statement "I feel pressured for time on a daily basis."

Trying to streamline the workflow, either in sample preparation or in the analysis itself, is increasingly important for scientists in analytical laboratories. Choosing sample preparation solutions that reduce the number of steps or allow multiple samples to be processed in parallel can reduce some of the bottlenecks.

**Results come from a short survey of 303 readers. Most work in R&D, QA/QC, with the most common job titles being researcher/scientist and supervisor/manager.*