Best practices for monitoring PFAS contamination in a routine shared-space commercial laboratory

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ABSTRACT
Per- and polyfluoroalkyl substances (PFAS) are synthetic chemicals that persist in the environment and have become ubiquitous contaminants. They have a negative impact on human health and are a threat to the environment, therefore many countries worldwide recommend they be monitored at some level.

Detection requirements for PFAS have been getting more challenging as advisory and regulatory limits are constantly being updated. For example, the recent Commission Regulation (EU) 2022/2388 set µg/kg maximum levels for PFOS, PFOA, PFNA and PFHxS in certain foodstuffs.1 Meanwhile, the US Environmental Protection Agency (EPA) recently tightened its lifetime health advisory levels (HALs) in drinking water for perfluorooctanoic acid (PFOA, 0.004 ng/L) and perfluorooctanesulfonic acid (PFOS, 0.02 ng/L).2 To develop and validate methods that can reach these extremely low levels, both high sensitivity instrumentation and good sample and system blanks (sub-ng/L levels) are required. Moreover, testing for PFAS should ideally be managed by commercial laboratories without the use of expensive or dedicated environments as there is no guarantee that those areas are PFAS-free. Unfortunately, due to the presence of PFAS in many lab supplies and consumer products, it is very difficult to obtain true sample blanks. This makes testing for PFAS to meet the regulatory levels an extremely challenging task.

On the other hand, effectively controlling the contamination allows PFAS testing to be conducted more universally. In this document we present a protocol to achieve and surpass EPA HALs, at the time of writing the lowest recommended levels, in a typical routine lab without a clean room or any special equipment. The full analytical method, including sample preparation procedure and validation data has been reported in the App Note 720007855en.3 Nevertheless, the information and guidelines provided in this document can serve as a reference for labs dedicated to PFAS testing, regardless of matrices and analytical scope.

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A. INTRODUCING THE ISSUE

1. What are PFAS?
PFAS (per- and polyfluoroalkyl substances) are a diverse group of highly stable chemicals that, due to persistence in the environment, represent a significant threat to ecosystems and human health. They occur in a wide range of consumer products, including aqueous film forming foams (AFFF), stain resistant fabrics, carpet and paper treatments, and chrome plating. Although regulations concerning acceptable exposure limits of PFAS are still being created and certain PFAS have been phased out of use, their stability still makes these compounds potentially prominent contaminants in the laboratory.

2. Why are we testing for PFAS?
Because exposure to PFAS is linked to human health and environmental impact, it is important to accurately and precisely measure these compounds in various matrices. Many regulations and exposure advisory limits require detection of various PFAS in the low ng/L (parts per trillion, ppt) or even pg/L (parts per quadrillion, ppq) ranges. The potential impact of reporting a result above a threshold can have a detrimental impact on the source of the sample - ex: manufacturing plant discharge, local water utility, landfill leaching, firefighting foam discharge, even though the ultimate goal is to rectify, clean up, and/or eliminate the source of PFAS entry into the environment. Therefore, we must have confidence in any reported result and ensure it is representative of the sample and not inadvertent contamination.

3. Why is controlling PFAS contamination important?
One of the challenges faced during PFAS analysis is the prominent occurrence of PFAS in everyday items that enter and are used in laboratory environments. For example, PFAS compounds are well known to be used in the manufacturing process of non-stick coatings like Teflon®. These types of coating are very valuable in laboratory environments, and therefore quite frequently present. The introduction of PFAS contamination into samples from laboratory sources can detrimentally impact the reported results of the analysis, especially when considering that PFAS methods strive to reach low or sub-ng/L concentrations. Cases such as this make potential PFAS contamination issues important to monitor and address. Teflon is certainly not the only source of PFAS contamination a sample is exposed to, there are various other sources which will be discussed. Ultimately, PFAS is everywhere, in every laboratory environment, regardless of whether it is a dedicated PFAS space or a shared location. Therefore, providing guidance on controlling contamination in the laboratory is helpful for all PFAS analysts to be successful.

B. INVESTIGATING PFAS CONTAMINATION
Based on recent toxicological data, the EPA has assigned HALs to four PFAS (PFOA, PFOS, PFBS and GenX) which have been investigated in this study, however, the advice given in this document are applicable to a wider range of PFAS.

The sample preparation procedure includes extracting the water sample using mixed mode Solid Phase Extraction (SPE) containing both Weak Anion eXchange (WAX) and reversed-phase chemistries, provides a 500x enrichment factor. The procedure is illustrated in Figure 1, and it is further detailed in App Note 720007855.3

**Figure 1. Scheme of the sample preparation procedure.**

- **Step 1.** Measure out water sample into a HDPE bottle and spike with extraction internal standards (EIS)
- **Step 2.** Pass 4 mL 2% (v/v) NH₄OH in MeOH, 4 mL MeOH and 4 mL H₂O through SPE
- **Step 3.** Load sample onto the cartridge (using a sample reservoir)
- **Step 4.** Wash with 4 mL 25 mM NH₄CH₃COO buffer (pH 4)
- **Step 5.** Dry the cartridge with positive pressure using a pre-washed plastic syringe
- **Step 6.** Wash with 4 mL MeOH
- **Step 7.** Elute with 2 x 4 mL 2% (v/v) NH₄OH in MeOH into previously washed plastic Falcon tubes
- **Step 8.** Dry samples under nitrogen at 50 °C to near dryness (only one drop remains)
- **Step 9.** Reconstitute with 0.25 mL MeOH + 0.25 mL of 4 mM NH₄CH₃COO solution containing injection internal standards (IIS), called “reconstitution solution”
- **Step 10.** Transfer samples to polypropylene vials and cap with pre-slit PTFE caps
There are materials and laboratory consumables that are commonly known to have the ability to contribute PFAS contamination to samples if used during sample preparation and analysis. These include, but are not limited to, those listed in Table 1. Additionally, this document will cover how to identify and manage potential PFAS contamination in consumables, reagents, and solvents that are necessary for the preparation and analysis of PFAS.

Table 1. Common sources that can contribute PFAS contamination to samples

<table>
<thead>
<tr>
<th>Common Sources of PFAS Contamination to Avoid</th>
<th>External Sources</th>
<th>Direct Sources</th>
</tr>
</thead>
<tbody>
<tr>
<td>Clothing/Lab coats treated with waterproofing materials</td>
<td>PTFE (Teflon) containers lined caps, and tubing</td>
<td></td>
</tr>
<tr>
<td>Waterproof papers, notebooks, binders</td>
<td>Aluminum foil</td>
<td></td>
</tr>
<tr>
<td>Cosmetics and personal care products (sanitizers, lotions, etc.)</td>
<td>Pipette tips branded as being &quot;low retention&quot;</td>
<td></td>
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<tr>
<td>Teflon tape</td>
<td>Permanent markers</td>
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<tr>
<td>Latex gloves</td>
<td>Vacuum grease</td>
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<tr>
<td>Antifog eyewear wipes and sprays</td>
<td>Glass transfer pipettes</td>
<td></td>
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<tr>
<td>Soaps and dishwashing detergents</td>
<td>PTFE filters</td>
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</tbody>
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C. MANAGING CONTAMINATION

In PFAS testing it is vitally important to screen existing lab equipment, consumables, solvents, and reagents for contamination, and to develop a methodical process to not only reduce this contamination, but also to identify its potential origins. In the diagram below (see Figure 2) the key principles of our approach are illustrated, in which these lab supplies are washed, tested/checked and selected where appropriate prior to use.

Figure 2. Diagram illustrating the backbone of the protocol used to control PFAS contamination in lab equipment.
The following steps are a result of many attempts to develop an effective and practical cleaning approach in our own lab, however the initial working conditions in other labs may vary. Therefore, the proposed procedure should be optimized for each case.

1. Laboratory environment
Before beginning testing it is important to make sure that the working environment is as clean as possible. Therefore, we recommend a worksurface away from the regular operation of the lab, with little chance of contamination by any day-to-day operation. There should be no obvious contaminants in the immediate vicinity that could potentially interfere with the setup. In this study we used a corner of the lab with no apparent sources of contamination nearby. Once a suitable location has been found, clean all surfaces and surroundings thoroughly with methanol.

2. LC-MS/MS system, sample vials, mobile phase bottles and reconstitution solution
The analytical system used was an ACQUITY™ Premier BSM with FTN and Xevo™ TQ Absolute Mass Spectrometer (detailed experimental conditions are found in App Note 720007855). Any LC used for PFAS analysis needs to be modified with a PFAS kit (p/n 176004548), consisting of PEEK mobile phase tubing (to reduce potential contamination from Teflon coated tubing) and an isolator column (to delay any contaminants from the LC pumps) in order to prevent PFAS contaminants from the mobile phases and LC pump from interfering with the sample peaks. Failure to use the PFAS kit or misuse of the PFAS kit has a high likelihood of introducing bias and inaccuracies into results.

It is also important to make sure that the sample vials are clean by running a 10-µL injection of a solvent blank, in this case a 1:1 methanol:water mixture. For PFAS analysis, it is recommended to avoid glass vials as PFAS can adsorb to glass. Therefore, it is recommended that sample vials consist of high-density polypropylene and to utilize a vial cap that also does not contain materials known to be sources of PFAS contaminants. After injection of the solvent blank, if there are no PFAS peaks present in the chromatogram, the LC-MS/MS system and the vials can be considered “clean” and appropriate for use and the peaks present in the previous sample (injection of 10 µL of the solvent blank sample) are a result of the sample blank itself being contaminated, either due to the vial or the solvents used. If the PFAS peaks are present in the zero-volume injection chromatogram, this indicates that the LC-MS/MS system is a source of contamination and should be thoroughly cleaned/flushed prior to further use.

When preparing, storing, or installing mobile phase on the system, it is highly advisable to use a new, clean set of clear glass mobile phase bottles (avoid fluoropolymer coatings) that will be dedicated to PFAS analysis only. This will avoid any accidental contamination of solvent bottles used for trace level PFAS analysis. The same is true of bottles used to contain reagents and solvents for sample preparation steps. Any bottles that contain a Teflon lined cap should be avoided. Additionally, bottle neck seals on mobile phase and reagent bottles should be removed for use for PFAS analysis (Figure 3).

In order to accurately test the sample preparation components, once it has been verified that the vials and the system are clean from contamination, the reconstitution solution should also be tested by injecting 10 µL of 4 mM ammonium acetate solution containing IIS (the so called “reconstitution solution” used in Step 9 of the sample prep procedure, see Figure 1).

![Figure 3. (A) Examples of the Teflon bottle neck seal typically found on the bottle (left) and removed (right) to use the bottle for PFAS analysis (B) Examples of the inside of solvent bottle caps using Teflon liners.](image-url)
3. Collection tubes and nitrogen evaporator

The suggested procedure of testing solvents, described in the following steps, involves blowing down 8 mL of a solvent or reagent using the nitrogen evaporator intended for use during all PFAS sample preparation, followed by reconstitution in 0.25 mL methanol + 0.25 mL of reconstitution solution. The 8-mL volume has been chosen as the SPE procedure involves using 2x 4 mL to elute analytes from the cartridge (see Step 7 in Figure 1) and therefore it is representative of the amount of solvent related contamination that would be present during sample preparation. Thus, it is important to first screen both polypropylene collection tubes and the nitrogen evaporator.

All 15-mL polypropylene collection tubes intended for use when preparing PFAS samples were previously washed with 3x 5 mL aliquots of methanol by hand shaking for 5 seconds and on a Vortex mixer for 20 seconds. The methanol was discarded, and the tube was left to dry prior to use.

The collection tubes and nitrogen evaporator were tested by placing empty collection tubes into multiple positions of the nitrogen evaporator with the heat bath and nitrogen flow turned on for 1 hour. Following the 1-hour period, 0.25 mL methanol + 0.25 mL of reconstitution solution were pipetted into the tube, vortexed, transferred to a sample vial, and analyzed using the LC-MS/MS system to assess the baseline levels of contamination.

There are various types of nitrogen evaporators that can be used during sample preparation of PFAS samples. It is important to ensure that the evaporator being used contains the least amount of Teflon components as possible for operation of the unit. In addition, evaporators that utilize needle valves should only be used with stainless-steel needle valves, and not those coated in PTFE (Figure 4). When using the nitrogen evaporator, it is best practice to pass nitrogen through for 30 min prior to the introduction of a sample.

Once collection tubes and the nitrogen evaporator have been tested and determined to introduce no additional contamination, testing can proceed to the following step.

Figure 4. (A) Example of a nitrogen evaporator that utilizes needle valves for the evaporation process. (B) Examples of different types of needle valves.
4. Methanol

Methanol is important not only for the preparation of some solvents used in the sample extraction protocol, but it is also used as the cleaning and washing agent for consumables used throughout the experimental procedure. Therefore, it is imperative that methanol does not contain detectable levels of PFAS. Manufacturer, lot number and even different bottles of the same lot can show wide variations of contamination level. Therefore, each bottle of methanol should be tested prior to use by blowing down 8 mL in a clean collection tube and reconstituted with 0.25 mL methanol + 0.25 mL of reconstitution solution, as described above. Contamination can be introduced into methanol during the manufacturing and/or bottling process as well as from fluoropolymer coatings used in glass solvent bottles and Teflon bottle cap liners used on glass solvent bottles.

Figure 5 illustrates an example of very variable contamination across different brands of methanol (A, B and C), with each trace compared to a low-level standard injection (equivalent to a sample concentration of 0.001 ng/L for PFOA, PFOS and PFBS, and 0.004 ng/L for GenX).

Once a suitable bottle of methanol has been found, it is possible to proceed and accurately test contamination in the labware.

5. Labware and pipette tips

All glassware, including solvent bottles, beakers, and graduated cylinders, as well as high density polyethylene (HDPE) plastic sample collection bottles were washed four times using approx. 10% of their respective volume in methanol prior to use. In the case of HDPE bottles, an additional step of rinsing the bottles with the same volume of clean water prior to use is recommended to aid further in cleaning and to wash away any residual methanol. Glassware should be tested by adding 8 mL of clean methanol to each glass container, which is then agitated and poured into a clean 15-mL collection tube, blown down, and reconstituted as described in the previous steps (see Step 9 of the sample prep procedure illustrated in Figure 1) prior to use in sample preparation and analysis. Additionally, as described in section 2, any glass solvent bottles being used to prepare reagents should have any Teflon bottle neck seals removed prior to use.

Once appropriate glassware was identified, it was kept segregated for this work and was not cleaned using a dishwasher at any point. Rinsing of glassware between use should only be done by washing thoroughly with methanol.

Plastic pipette tips were tested for presence of PFAS by taking replicate aliquots for a total of 8 mL of methanol directly into a clean 15-mL collection tube, blown down and reconstituted as stated prior to analysis. Even after verifying that the brand of pipette tips used in the laboratory were free from PFAS, all pipette tips were still washed prior to use to eliminate concern about lot-to-lot or tip-to-tip variation. Pipette tips were washed by pipetting up methanol and then dispensing to waste the required pipetting volume six times. Pipette tips that contain any coating to enhance performance, such as those marketed as “low retention” style tips, were avoided for PFAS analysis as the composition of these coatings is unknown.

Figure 5. Overlaid chromatographic traces of a standard injection (0.001 ng/L for PFOA, PFOS and PFBS; 0.004 ng/L for GenX), and different brands of methanol (A, B and C).
Both positive displacement and air displacement style pipettes have been used to prepare PFAS samples without excessive PFAS contamination attributed from either style of pipette. This indicates that the style of pipette does not have an impact on level of PFAS contamination.

Once the cleanliness of the labware has been verified, it is possible to accurately test solvents and respective components.

6. Sample preparation solvents

All solvents and reagents used during the SPE sample preparation procedure (Figure 1) should be both individually tested, as well as tested in their final use. For example, the elution solvent is a 2% ammonium hydroxide in methanol solution. Therefore, the ammonium hydroxide and methanol being used to make that solution should be tested individually (using the same procedures described throughout this paper of blowing down a portion in a 15 mL polypropylene tube, reconstituting, and analyzing), but the final 2% ammonium hydroxide in methanol solution that is prepared from those individual components should also be tested in the same manner.

To ensure the required reagents from the SPE procedure did not introduce PFAS contamination, both the ammonium hydroxide methanolic solution used as an elution solvent (see Step 2 and 7 of Figure 1) and ammonium acetate buffer used as a washing solvent (Step 4 of Figure 1) should be tested by blowing down 8 mL of each solution into a clean 15-mL collection tube and reconstituted as stated prior to analysis.

While completing this testing in different applications laboratories, we experienced repeated issues with contamination in the ammonium hydroxide solution, although no issues were found from ammonium acetate buffers.

Examples of PFOA contamination levels in three different brands of ammonium hydroxide is shown in Figure 6-A. It should be noted that Brand A was a shared bottle of ammonium hydroxide that was used in the laboratory for various applications work and not a bottle exclusively dedicated to only use for PFAS applications. The large PFOA contamination from Brand A could have been introduced into the bottle after exposure in the laboratory, and not present from the manufacturing or bottling process. This emphasizes the need to dedicate consumables, reagents, and solvents for PFAS use only.

Based on the results of Figure 6-A, it was determined that Brand C ammonium hydroxide was the most suitable to use for PFAS analysis. After making this decision, a solution of 2% ammonium hydroxide was prepared for use during an SPE extraction. During this procedure, it was determined that although each component (ammonium hydroxide and methanol) tested clean, that upon combining them, a fairly significant amount of PFOA contamination became present (Figure 6-B). This was true regardless of how the solution was prepared (i.e. in a glass bottle, an HDPE bottle, or a polypropylene 50 mL tube). It has not been determined how or why this occurs.

Figure 6. (A) Overlay of PFOA contamination in three different brands of ammonium hydroxide. (B) Demonstration of individual components of ammonium hydroxide and methanol containing no PFOA contamination, but PFOA contamination present in the mixture of the components into one solution.
7. Manifold and SPE cartridge
A brand-new SPE manifold was dedicated to PFAS testing. The manifold was completely washed twice with methanol prior to use. The inside of the glass manifold, as well as the top and underside of the lid was washed twice with 200 mL methanol. A clean glass beaker was used to pour approximately 10 mL of methanol through each hole on the chamber lid, with this last step being repeated twice. A clean methanol solvent squeeze bottle can be used to perform the cleaning steps if it has been verified to not contain PFAS contamination.

If it is not practical to purchase a new manifold, a previously used manifold can still be used for PFAS analysis. The same cleaning procedure outlined above is recommended to wash the manifold more rigorously. At a minimum, the sample needle valves should be replaced with new valves when introducing an SPE manifold used previously for other applications.

The sample needle valves should be thoroughly cleaned before and after use with the following procedure. Ensure the valves are in the open position, place in clean glass beaker, fill with enough methanol to cover the valves. Sonicate for approximately 15 minutes. Once placed into the SPE manifold lid for use, rinse methanol through each valve.

SPE reservoirs were slightly more challenging to clean. Batches of 4 reservoirs were washed in clean 1L glass beakers. Reservoirs were sonicated as described above, with the difference being inverted midway to submerge all parts of the reservoir in the cleaning solvent. When handling reservoirs, ensure contact only with the outside of the reservoir. Finally, each reservoir was connected to an adaptor and was rinsed with 10 mL clean methanol.

Oasis™ WAX for PFAS Analysis 6 cc Vac Cartridges (p/n 186009345) are designed specifically to perform low level quantification of PFAS. It is still recommended to include the conditioning steps (Step 2 in Figure 1) prior to loading any sample onto the cartridges to remove any PFAS contamination that could enter the cartridges after they are removed from their pouch prior to use.

It is possible for a sample highly contaminated with PFAS to permanently contaminate an SPE manifold once extracted. If possible, a dedicated SPE manifold set up(s) should be used for highly contaminated samples to isolate them from manifolds used for trace PFAS samples. In this case, samples would need to be pre-screened (see direct injection approach in App Note 720006764 as a potential approach for this) before directing them to the appropriate SPE manifold.

Figure 7 illustrates that, after appropriate washing, all the tested materials and solvents presented no detectable levels of PFOA, PFOS and GenX, while PFBS was found at levels below the 0.001 ng/L with the highest signal recorded after testing the ammonium hydroxide solution. It should be noted that the main source of PFBS contamination was the methanol used for preparing this solution and for testing the various items.
8. Water
In order to validate a PFAS testing method for water samples it is required to determine method performance parameters such as trueness and limits of quantitation (LOQ). In the absence of a QC sample or a standard reference material, it is possible to perform recovery experiments by spiking a blank water sample with known concentrations of PFAS and calculate the bias between the theoretical concentration and the calculated concentration. To correctly calculate the trueness and LOQ of the method, a clean water sample must be available in the laboratory. The guidelines state that the relative percentage of the blank water sample should be below 33% of that of the LOQ level. Therefore, water samples used as blank must be tested to ensure PFAS levels are below such thresholds. However, it is very difficult to obtain/procure a truly “blank” water sample that contains PFAS below 0.004 ng/L. As a prerequisite, to avoid overestimating the concentration of PFAS in the water tested, it is important to ensure that all lab equipment and solvents are clean by following the previous steps detailed in this document.

In this study we have tested both Milli-Q® water (resistivity at 25 ºC >18.18 MQ-cm) and a brand of LCMS grade bottled water by loading different volumes of blank water samples onto the SPE cartridge and following the sample preparation procedure illustrated in Figure 1 and in the App Note 720007855. While PFOS and GenX were not detected (S/N < 3), PFOA and PFBS were detected below 0.004 ng/L. Furthermore, the concentration of PFOA in the LCMS grade water was higher compared to Milli-Q® water, and increased by increasing the sample volume. By contrast, different types of water presented comparable amounts of PFBS, which, in our case, was coming from the ammonium hydroxide methanolic solution and methanol, but were below the LOQ of the method (see Figure 8). While this example shows that the water obtained from the Milli-Q system was more suitable in this particular case, this may not be the case in every laboratory. The contamination level of the supply source of the water purified by the purification system can vary drastically by laboratory location. Also, the type of the water purification system may include different internal components containing Teflon or other coatings that could contain PFAS. It is crucial to perform testing of water sources intended to be used as a blank sample and/or a spiked recovery or QC sample before use.

In extreme circumstances, where it is not possible to find a blank sample, and assuming the other items and solvents are clean, it is possible to use the described procedure as a purification step by running water through the SPE process and collecting it, thus obtaining cleaned-up water that can be potentially used as “blank”.

Once all of the consumables, reagents, and solvents for sample preparation have been verified to be “PFAS free” or at least contain the minimum amount of PFAS acceptable, the sample preparation can be run. While preparing samples, it is still important to include quality control samples, especially sample blanks, to ensure that contamination still is not being introduced during the procedure from an external source. Samples blanks that should be included in each batch of samples include a method/extraction blank, a reagent blank, and a solvent blank. The method or extraction blank is a blank water sample that is taken through the full sample preparation procedure. The reagent blank is an aliquot of the elution solvent used (equivalent to the portion used to elute samples) transferred directly into a 15 mL sample tube, dried, and reconstituted. The solvent blank is an aliquot of the reconstitution solution transferred directly into a sample injection vial. All of these blank samples should be analyzed on the LC-MS/MS in the same sample batch as the remaining field samples. Specific regulatory methods may also require additional quality control samples, so these should be included as required as well.

![Figure 8. Chromatographic traces of PFOA (m/z 413->169) in Milli-Q® water (A) and a brand of LCMS water (B) at different volumes of water sample loaded on the SPE cartridge. The methanol used to prepare sample prep solvents is also displayed for comparison purposes](image)
D. SUMMARY

To minimize the contamination of PFAS in the lab and reduce cross-contamination it is important to follow a rigorous wash-and-test protocol. In this white paper we propose a stepwise approach that was found effective in controlling PFAS contamination in a typical shared-space laboratory.

In summary, once a clean working environment has been identified, the first step is to make sure that the LC-MS/MS system presents no detectable levels of PFAS, using the Waters PFAS kit and Waters-branded Polypropylene LC vials. Then, the solution with injection internal standards used to reconstitute the sample after evaporation should also be tested. The next step is to test plastic collection tubes and the nitrogen evaporator. Subsequently, methanol should be tested and the methanol brand/lot with lowest level of PFAS should be used to wash all labware and the other items employed in the sample prep, including pipette tips, sample reservoirs, manifold and its components. The cleanest methanol will also be used in the SPE procedure and to prepare the elution solvent with ammonium hydroxide. Finally, it is advisable to continue using those consumable items found “clean” during the testing phase as well as repeating the full wash procedure of those more permanent items (e.g. glassware) between sample batches.

E. TIPS AND TRICKS

**Methanol and glassware** — Once you have found clean methanol, stick with it. Once you have tested your solvents and glassware and these are clean, continue to use them for PFAS batches in the future. To be more efficient with resources, triage slightly contaminated methanol bottles within a batch and use in the first washing step of the cleaning procedure. Otherwise put methanol back in the lab for general use.

**Pipettes** — To reduce contamination from pipette tips, best practise is to use two beakers for cleaning the pipette tips (i.e. wash each pipette tip three times with methanol in beaker #1 and further three times with methanol in beaker #2 before use).

**General** — After each washing step, when drying, it is advised to avoid evaporating residual solvents with compressed air or nitrogen, and allow items to dry naturally on a clean work surface.

**Reduce, Reuse** — All lab equipment was rinsed and reused between batches.

**Water** — In the unfortunate circumstances that you cannot find clean water to use as a sample blank, run the cleanest water you have through an SPE cartridge, collect, and use "cleaned-up" water as blank.

**Concentration step** — During this step, avoid the complete dryness of the eluate, stop the nitrogen evaporator when approx. one drop remains in the collection tube.

**Manifold** — To avoid leaks from the junctions and fittings between the reservoirs and cartridges, fill approx. ¾ of the cartridge with sample, apply the reservoir and fill the reservoir with more sample.

**Manifold** — Ensure sufficient spacing of the cartridges on the manifold to avoid unwanted cross contamination.

**Gloves and personal hygiene** — Before work, wash hands with water, avoid using soap. Avoid use of cosmetics and personal care products (e.g. hand lotions and sanitizers) when working in the preparation laboratory. Do not wear any clothing items known to be sprayed or inclusive of waterproofing chemicals. Do not use anti-fog sprays or wipes on safety glasses.

**Cleaning the laboratory** — Be mindful of what cleaners are being utilized in the laboratory to clean benchtops and floors, including floor waxes.

**Sample labelling** — Permanent markers can introduce PFAS contamination. Label tape from a label making printer has been used successfully in our laboratory without introducing extra PFAS contamination.
F. REFERENCES


