

December 2023

Keywords or phrases:

Yield Measurement, Unknown Volume Measurement, Centrifugal Concentrators, Picus®2, Vivaspin®

Effective Use of Picus® 2 Pipettes with Vivaspin® Ultrafilters

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Introduction

Ultrafiltration and diafiltration are important techniques for sample concentration, buffer exchange and desalting in many life science purification workflows. To use centrifugal ultrafilters effectively, it is essential that they are handled correctly and paired with the correct pipette and pipette tips.

In this guide, we provide advice on optimal handling and selection of the best combination of Vivaspin® ultrafilter, Picus® 2 pipette and pipette tip to:

- Ensure complete retentate retrieval and accurate recovery measurement
- Avoid sample-to-pipette and pipette-to-sample contamination
- Prevent sample foaming and degradation with appropriate pipetting speeds
- Eliminate interference in downstream analysis
- Minimize non-specific adsorption of macromolecules on the ultrafilter

Use of Manual Mode

Knowing the final volume of a concentrated sample is critical to accurately determine macromolecule recovery. Picus® 2 electronic pipettes allow precise measurement of the retentate volume during aspiration from Vivaspin® ultrafilters, eliminating the need for additional process steps such as gravimetric methods. In manual mode, the user can easily control aspiration and dispensing using the adjustment wheel. When aspiration is visually controlled, Picus® 2 uses the movement of the piston to accurately measure the volume and display it on the screen.

The main factors affecting accuracy in manual mode are the attention of the user and the resolution of the pipette. Pipette resolution is the smallest increment of volume adjustment, e.g., 1 µL for Picus® 2 1,000 µL pipettes. Inaccuracy can be caused by accidental aspiration of air when the pipette tip is too close to the surface of the liquid and when residual sample is left inside the ultrafilter. A step-by-step protocol for manual mode is provided on page 3 of this guide.

Select Suitable Pipette Tips

The construction of centrifugal ultrafilters, especially those with dual membranes and low volume dead stop pockets, can pose a challenge for access to and retrieval of retentate samples. As such, using the correct pipette tips in combination with ultrafilters that feature sufficient spacing between the membranes and angular dead stops will enable convenient and complete retentate retrieval.

Furthermore, if the pipette shaft enters the ultrafilter, there is a risk of contamination. Extended length pipette tips are therefore recommended as their length eliminates the need for the pipette to be brought into the concentrator.

Finally, the use of low retention tips can help maximize sample recovery with low surface tension liquids, which tend to leave a film of liquid on the inner surface of standard tips.

Pipetting Speed

Sample foaming can be a problem when handling macromolecular samples, potentially leading to protein degradation. This can be avoided by utilizing the low surface tension of low retention pipette tips. Foam and bubble formation can also be prevented by adjusting the speed setting of the Picus® 2 pipettes (Figures 1 and 2).

Figure 1. Pipetting BSA solution (1 mg/ml) with fast speed (Picus® 2 setting 9) results in sample foaming.



Figure 2. Pipetting BSA solution (1 mg/ml) with slow speed (Picus® 2 setting 1) is gentler on the protein.



Vivaspin®, Picus® 2, and Pipette Tip Compatibility

Complete aspiration of retentate samples is critical to maximize target molecule recoveries. Table 1 lists the recommended pipettes and tips to use with various Vivaspin® ultrafilters to achieve this.

Table 1. Vivaspin® Compatibility with Picus® 2 pipettes and tips

| Vivaspin® | Picus® 2 | Tip |
|--------------------|---|---|
| Vivaspin® Turbo 15 | LH-747081 | LH-X781000 |
| | | LH-X781001 |
| | | LH-XF781001 |
| Vivaspin® Turbo 4 | LH-747081 | LH-X781000 |
| | | LH-X781001 |
| | | LH-XF781001 |
| | If concentrate volume is below 100 µL use: LH-747041 | LH-X780200 LH-X780201 LH-XF780201 |
| Vivaspin® 20 | LH-747081 | LH-X781000 |
| | | LH-X781001 |
| | | LH-XF781001 |
| Vivaspin® 6 | LH-747081 | LH-X781000 |
| | | LH-X781001 |
| | | LH-XF781001 |

Sample Concentration

1. Prepare Vivaspin® concentrator
 - a. For preservative removal: in pipetting mode, pipette buffer up and down three to five times to rinse the membrane
 - b. For passivation: pipette the passivation solution up and down three to five times over the membrane and ultrafilter housing, and remove the solution (rinsing the membrane and device with passivation solutions can reduce non-specific binding between molecule of interest and ultrafilter surfaces)
2. Perform concentration with centrifuge. Process time depends on the sample type, relative centrifugal force, temperature and required concentration factor.
3. Measure retentate volume with Picus® 2 manual mode

Retentate Volume Measurement

1. Set the pipette to manual mode
2. Adjust speed settings to 3 for aspiration and 3 for dispensing; accurate control of the aspiration is easier at a slow speed setting
3. Ensure the tip is below the liquid surface when aspirating while avoiding the tip being placed too close to the bottom as this may interfere with liquid flow and distort results
4. Use the adjustment wheel to aspirate liquid into the tip in a controlled manner; the force with which the wheel is turned influences the aspiration speed
5. As the liquid is drawn into the tip, ensure the end of the tip is close to the bottom of the ultrafilter; the angular dead stop of Vivaspin® Turbo assists with this
6. Release the wheel as soon as all liquid is aspirated into the tip and observe if any residual liquid gathers at the bottom of the ultrafilter; if so, continue aspiration by turning the wheel
7. Read the total volume aspirated from the pipette's display
8. Empty the tip completely by pressing QUIT and double-clicking the operating button

Summary

Sartorius Picus® 2 pipettes and extended length pipette tips enable collection of concentrated samples from Vivaspin® concentrators. Use of the Picus® 2 pipette manual mode simplifies retentate volume measurement and extended length pipette tips ensure that the entire sample can be retrieved while protecting the sample and pipette from contamination.




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