



Technical Bulletin #A0100: Sample Filtration for Microvials

Overview and Objective

Protasis instrumentation employs standard laboratory sample formats, specifically microvials, and both 96- and 384-well plates. We recommend good laboratory practice and sample preparation methods that are already widely accepted in chromatography, mass spectrometry, and other analytical instrument platforms that employ microflow systems and technologies. These protocols specify the use of particulate-free samples and solvents that have been filtered and/or purified prior to analysis.

This Technical Bulletin describes one method of filtration for microvial-based samples with volumes ranging 5 - 50 μL , with a filtration recovery efficiency $> 95\%$. A companion technical bulletin (#A0101) describes our recommended method of filtration for well plate-based samples. Both are available for download in electronic format from the Knowledge Base of the Protasis technical support website, www.microNMR.com. All prices valid April 2007.

Microvial Filtration

Recommended Parts and Apparatus

1) Microfilter

10- μL Neptune Barrier Tip, Part #BT10¹

Cost: \$99.00 for a 10-pack of 96 tips per pack (960 tips)

Distributor: Continental Lab Products
1-800-456-7741

2) Microvial and Cap

300- μL polypropylene microvial (Part #09-1300-100)

Cost: \$23.00 per pack, 100 per pack

Vial cap with PTFE/Silicone septum (Part #09-0030N)

Cost: \$20.50 per pack, 100 per pack

Distributor (for both microvial and cap): MicroLiter Analytical Supplies
1-888-232-7840

3a) Benchtop Centrifuge²

Clay Adams Compact II Centrifuge with 6-PlaceRotor (Part #22-289446)

¹ The Neptune Barrier Tip is made from a polypropylene housing and the filtration media is a matrix of sintered HDPE spheres. Protasis can provide solvent compatibility charts for both materials upon request.

² Alternatively, a centrifuge that is compatible with a 54-microvial well-plate can be employed.

3200 RPM (1163 x g), 12.8 lb, Power:120 volts, 60 Hz
Cost: \$1072.95
Distributor: Fisher Scientific
1-800-766-7000

Accessories included with purchase of centrifuge:

- Clay Adams Shield Cushion Adapter - Pack of 6 (Part #05-100-112)
- Clay Adams Rubber Cushion - Pack of 6 (Part #05-100-92)
- Clay Adams Adapter - Pack of 6 (Part #05-100-93)

3b) Additional centrifuge accessories (Fisher Scientific) necessary to modify all metal shields for accommodation of the microvials described above:

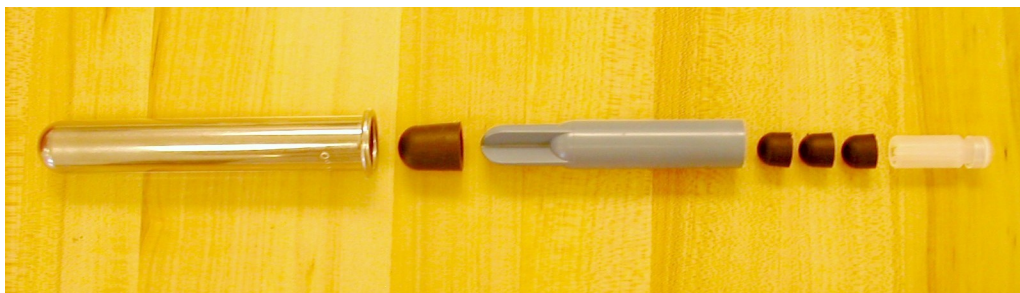
- Clay Adams Rubber Cushion (Part #05-100-92)
 - Cost: Pack of 12 for \$39.99

Note: The cost of the microvial with the filter is less than 53 cents each.

Overall Procedure

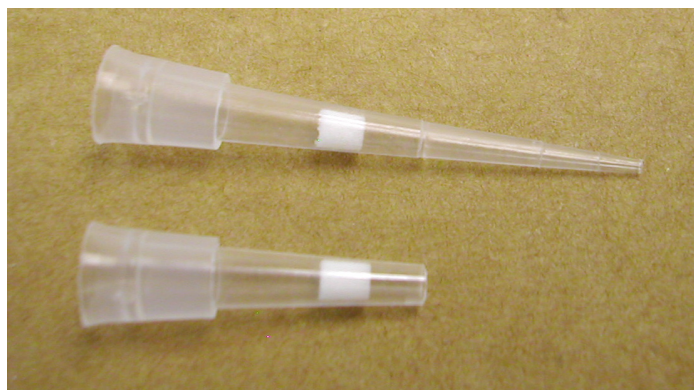
Step 1: Centrifuge Preparation. Accommodation of microvials into the metal tubes (called metal shields by the manufacturer) of the centrifuge requires use of the adapter and cushions specified above. These items mechanically support the specific microvials described in (2) above, and prevent the microvials from seating too deeply in the rotor during centrifugation. This in turn facilitates easy removal of the microvials following centrifugation.





A green cushion should already be in place at the bottom of each metal shield as received (not visible). Insert one of the large, black cushions (Clay Adams Shield Cushion Part #05-100-112) into each metal shield, and push it to the bottom. Subsequently insert one plastic adapter (Clay Adams Part #05-100-93) and push until it is snug against the large cushion. Finally, insert three small cushions (Clay Adams Rubber Cushions Part #05-100-92). This should ensure that the top of the microvial remains flush with the top of the metal shield for easy removal following centrifugation. Note that the combination of one large cushion, one adapter, and three small cushions is required for *each* of the six metal shields that go into the centrifuge spinner.

Step 2: Using a clean razor blade, trim the tip of the Neptune Barrier Tip (BT10) just below the filtration medium, at the indentation line, as shown below. Note that when cut correctly, the filtrate volume will fill the microvial below the tip, but without touching the bottom of the filtration medium (the white part).



Step 3: Insert the filter into the microvial (shown below), and subsequently add up to 50 μ L of fluid. Screw on the cap tightly.



Step 4: Place the microvial in the centrifuge and activate the centrifuge for a few minutes or longer depending upon the viscosity of your solution. Upon removal from the centrifuge, the microvial must be kept in an upright position. After centrifugation, unscrew the cap, remove and dispose of the Neptune Barrier Tip assembly, then screw the cap back on the microvial tightly.

User Note

Some users spin down every sample before automated analysis to be certain that any particulates present are confined to the very bottom of the sample container. In addition, it is always wise to not place the syringe needle at the very bottom of a well or vial being sampled. This prevents particulates that may have settled there from being drawn into the syringe needle.