

MWCO Filters Supplemental Information

Protocol Details / Instructions

- Before starting, please refer to the manual for details found at <u>www.analytical-sales.com</u> > Resources > Manuals & Instructions Sheets > <u>MWCO Filters Info/Instruction Sheet</u>.
 - *Note:* It is written for microspin centrifuge style MWCO filters with 800uL capacity so volumes need to be adjusted for *400uL filter plates*.
- Pre-rinsing is required (see literature/manual). PES membranes ship with a preservative on it that needs to be rinsed off for any protocol.
- To prevent membrane dry-out during the "pre-rinsing" step, run tests on unused wells/filter membranes to determine an appropriate centrifuge/pull vacuum duration.

General Parameters

- · Selecting the appropriate MWCO filter
 - Typically 3 times smaller than the molecular weight of solute being filtered.
- Roughly **3 to 6 times smaller** than the molecular weight if filtering proteins.
- Conforms to SLAS format.
- Our PES MWCO filters do not have additional dead volume retention.

Sample Considerations

- Preform other sampler preparation/cleanup steps before using MWCO filters.
 The "dirtier" your sample the more likely the filter will clog.
- The optimal sample concentration, centrifugation time and rcf (spin force/speed) are variables that need to be determined for your specific sample.
- Some volume should remain above the filter (PES) membrane which would have your higher weight molecules (e.g. proteins, etc.) that you would then transfer into a collection plate.
- Filtrate collection (for 96 well filter plates):
 - If keeping, make use of any collection plate (e.g. # 17P687Z)
 - If discarding, either a reservoir or collection plate can be used.

Note: If centrifuging, a 96 well collection plate will provide the most stable platform. Often they are taped together for additional stability.

Usage Considerations

- Start with the most dilute form of your sample.
- Methods of pulling your sample through filter membrane:

• Centrifugal force / Centrifuge

- Pros: Samples are pulled through each membrane independent of each other.
- Cons: Not all centrifuges are capable of accommodating the taller stack of a collection plate with a filter plate on top.
- Determining G-Force/RCF & Duration
 - · G-Force/RCF are used, not rpm. They are independent of your centrifuge's design.
 - Do not spin at more than 2000 xg
 - Use a low rcf (<1000 xg) in a first trial for 10 minutes.
 - If the filtration is incomplete, increase the duration by estimation to drive to completion and /or increase the rcf incrementally to a satisfactory filtration rate.

Negative Pressure / Vacuum manifold (<u># 96844</u>)

- Pros: Can be automated on liquid handlers. Lower cost than a centrifuge that can hold a collection plate w/filter plate on top.

Breakthrough of your sample can happen.

- Cons: Negative pressure works across the entire plate. If there is an empty well then the rest will no longer experience the negative pressure.

MWCO filters (1k Da & 3k Da) Additional Considerations

• Consider having additional sample cleanup prior to MWCO filtration.

• Take extra care determining your parameters for pulling the sample through

• The lower the MWCO filter the more important it is to start with the cleanest sample possible.

- Having a wide range/variability of how "dirty" the samples are can lead to difficulties as each well will pull through at different rates.
- Unused wells/rows need to be sealed with a film. Please see our patterned adhesive film (# 961801)







For your convenience, we offer both 96-Well MWCO filter plates and MWCO micro-spin filters