BECKMAN COULTER
Life Sciences

Optimizing the Beckman Coulter Multisizer 4e for use with small apertures

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Introduction

The purpose of this application note is to provide some guidelines to facilitate successful testing with the Multisizer 4e when employing 30 µm and smaller. Additional information for changing and working with small size apertures is given in Chapter 4 of the User's manual.

Using aperture tubes below 30 µm on a Multisizer instrument can be challenging due to the small size of the aperture which becomes easily clogged by larger particles. Many of these particles may come from random dust and other contaminating materials in the instrument environment.

It is necessary to take some proactive steps to ensure a smooth and accurate testing process when working with smaller apertures. The steps outlined below, while fundamental in nature, become increasingly important when employing the smaller apertures.

Locating the instrument in a controlled environment such as a laminar flow hood (low airborne particulate levels and reduced electrical noise) is highly recommended. In addition the instrument should be thoroughly cleaned as recommended in chapter 4 of the User's manual.

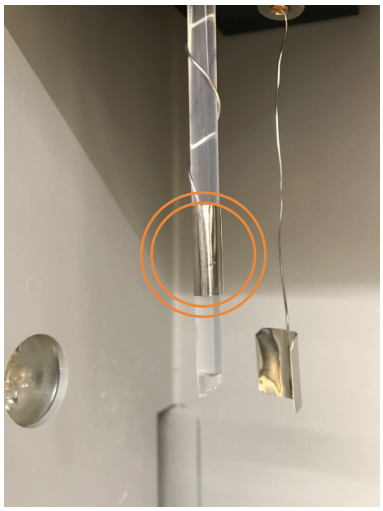

Figure 1. Electrode positioning

Guidance for filtering Isoton and electrolyte solutions

We recommend the use of a graduated dispenser, pn 8320309, which fits different bottles to dispense the isoton II electrolyte solution. (For convenience a 2 L bottle of Isoflow pn 8547008 can be used).

The dispenser has a nozzle to which a syringe filter or series of syringe filters can be fitted to remove any stray particles and ensure the cleanest possible electrolyte solution for use with the sample

BECKMAN PART NUMBER	DESCRIPTION	QUANTITY
8320309	Graduated 10 mL dispenser	1
8547008	IsoFlow (twice filtered Isoton II)	1 quad pack
8457008	2 L Isoton Bottle	1
PALL Acrodisc® Syringe Filters		
4611	0.1 µm, 25 mm	(50/pkg)
4612	0.2 µm, 25 mm	(50/pkg)
4614	0.45 µm, 25 mm	(50/pkg)
4618	0.8 µm, 25 mm	(50/pkg)

Figure 2. 8320309 Graduated 10 mL dispenser

Figure 3. PALL Acrodisc Syringe Filters

Process steps for Instrument setup and preparation

1. Remove the Electrolyte Jar from the MS-4e and perform a thorough cleaning of the jar and ensure the final rinse is done with a laboratory grade DI water.
2. Fill the Electrolyte Jar with fresh ISOTON II that has been filtered through at least a 0.2 µm filter.
3. Ensure that the internal electrode is ~1/2" above the tip of the filling Teflon tubing. If not, carefully wind its wire around the filling tubing until the electrode is at ~1/2 above the tip of the tubing as shown in figure 2.

4. Following the “Change Aperture Tube Wizard” function located in the Run menu to install the new aperture. Before installation of the new aperture ensure it is clean and the orifice inlet is clear of debris. Do not touch the orifice and essential to use particle free gloves.

5. Using a 200 mL beaker of clean, 0.2 µm filtered DI water rinse the outside of the aperture tube and the electrode by completely immersing them in the water, gently swirling the fluid as you rinse down the aperture tube.

6. The Beckman Coulter “Accuvette ST” is recommended for use when performing small particle sampling. Ensure the new accuvettes and caps are rinsed thoroughly with a lab grade filtered DI water. Preferably filtered down to minimum of 0.1 µm.

7. If using a separate precision fluid dispenser ensure that a minimum of a 0.1 µm disc filter is used when dispensing new ISOTON II into the rinsed accuvette. Fill accuvette to either the 10 ml or 20 ml level depending on the application or material requirements.

8. After the “Change Aperture Tube Wizard” steps have been completed, add the suitable particle size control standard to the accuvette. Attach the cap and roll the accuvette gently between your hands to adequately suspend the particles and prevent bubbles in the sample.

9. Perform a Calibration if the aperture was not previously calibrated or perform a Verify if the aperture was previously calibrated.

10. After calibration/verification is complete, repeat step 5 only to thoroughly rinse the aperture tube and electrode prior to running samples.

Process steps for running samples

1. Install cleaned accuvette with filtered electrolyte into the MS-4e and perform preview, flush and preview again to ensure zero counts in the progress bar. When reviewing the real time result the graph will indicate if the blank fluid and containers are clean and your process is sound.

2. When satisfied with Preview, cancel it and run three blanks using the same Control Mode that will be used in your SOM when formally running the samples, i.e. Volumetric or Time mode; flush before run, etc. See Figure 4 below.

3. Ensure that the SOM is setup for only 1 run at a time, not multiple runs, to prevent blockages. Set the pre-flow stabilizing time to 10 seconds and total stabilizing time to 15 seconds respectively for best accuracy as shown in the Figure 5 below.

4. Perform “Unblock” at end of each run completion to ensure there is no buildup of material at the aperture inlet. “Unblock” will disperse the particles that gathered at the aperture by inertia after a run.

5. Some samples, such as proteins may require to perform unblock between runs with a 5% solution of Micro 90 because the tendency of protein to stick to the aperture surface and create noise. Have a squirt bottle of DI water to spray the aperture after the Micro 90 unblock.

6. In general, it is good practice to do unblock with Micro 90 solution between samples and at the end of the last sample to have the aperture always in optimal condition.

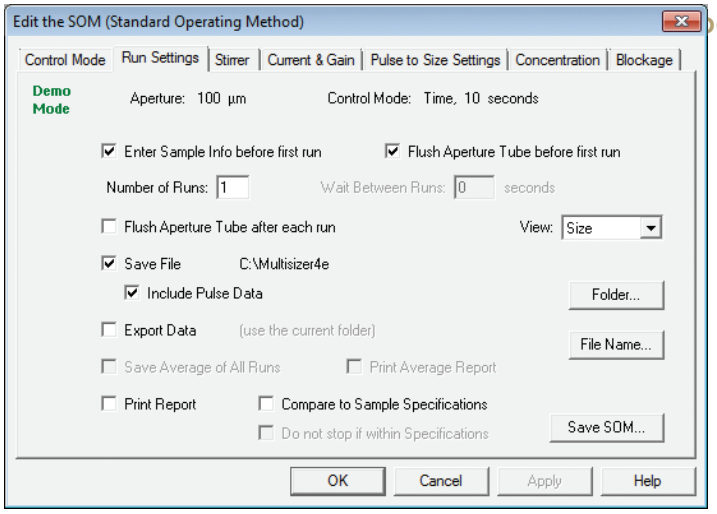

Figure 4

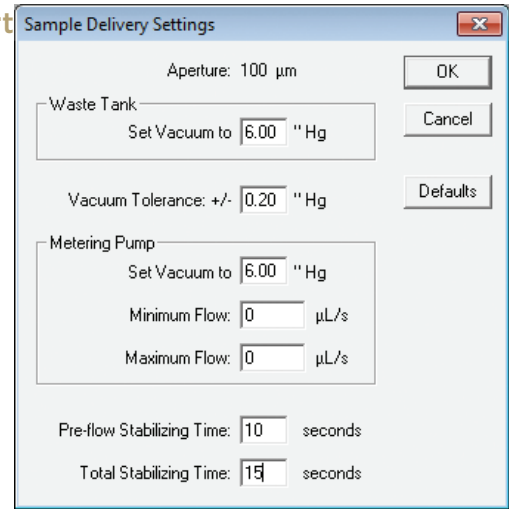

Figure 5

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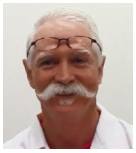

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The purpose of this application note is to provide some guidelines for successful operation of the Beckman Coulter's Multisizer 4e for use with small Apertures (30 µm and smaller.)

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