

## SOP-E038

### Operation of the Beckman-Coulter Mutisizer 4 located in Bindley Room 25

**Objective:** To obtain the cell count of a sample using the Beckman-Coulter Mutlisizer 4.

**Procedure:**

1. Turn the power on, power switch is located on the back of the instrument.
2. Click on the Mutlisizer icon on the desktop, program > OK, the password is: beckman
3. Click on "SOP" tab at top
4. Select "load an SOP"
5. Choose one which matches the orifice size and or your sample (P.aeruginosa 20µM, for algae)
6. There should be a cuvette (Accuvette, Beckman-Coulter cat#A35471) on the stage with Coulter Clenz.
7. Remove this cuvette and place a clean cuvette with mH<sub>2</sub>O, or PBS, and rinse off the orifice by manually moving it up and down via the stage to rinse the Clenz off. **PLEASE NOTE TO BE CAUTIOUS WHEN PLACING THE CUVETTES ON THE STAGE, THE ELECTRODE MUST BE INSIDE THE CUP, AND IS EASILY DAMAGED.....DON'T SMASH IT.**
8. Put a clean cuvette with Isoton II diluent (Beckman-Coulter cat # 8546719) on the stage, this will be the "blank".
9. You should have enough volume such that the electrode is at least half covered.
10. Click on the "Fill" button located on the lower right of the screen
11. Click on "Start"
12. When it is finished counting the blank, a window will come up asking if you want to save, select "no", then go up and select the save button, look for your folder/ open the folder/ name the sample/ save the file.
13. Run and save the blank a total of three times.
14. Each time you finish a count you will have to select "no" when it asks if you want to save. (It wants to save in a default directory), find your directory and save.
15. Now you must load one of the blanks before running your sample. (Usually choose the most recent one run.), Go to sample > load blank run > select the blank you want to load.
16. Mix gently by inversion the cuvette with your sample to be counted in it. Place it on the stage.
17. If you have not run a sample before, you should select "Preview" first. The concentration should be under 8%. If it is not you will need to dilute your sample before trying again.
18. If it is OK, select "Start"
19. When it is finished counting save as above. Count and save each sample three times.
20. To export the data to a flash drive you need to first make an overlay. Click on file > overlay.
21. Select the three counts to overlaid.
22. Go to "runfile" > "export"
23. Select all the boxes and select "export all run"
24. Save in your directory
25. When finished put the Isoton II cuvette back on the stage and "Start".
26. **If you are running algae or something else rather nasty you will need to follow the Extended Cleaning Protocol discussed below.**
27. If your samples are not going to clog up the machine then,
28. Take the Isoton II cuvette off and place the Coulter Clenz cuvette back on the stage.
29. Exit out of the program
30. Turn the power switch off.

**Extended Cleaning Protocol – use whenever you count algae**

1. Replace your sample cuvette with the cuvette of Isoton II diluent. Run as a sample at least twice. Do not need to save this data.
2. In the fluids area behind the door on the left of the instrument, take out the Isoton II jar. Place a container with warm 10% bleach solution in its place. There is a container suitable for this near the sink area. The bleach is also near or under the sink.
3. Select the "Flush" tab located on the lower right corner of the screen. You will need to hold the lid down on the bleach container in order to place enough pressure that the instrument realizes there is a container there.
4. Do this "Flush" at least three times.
5. Remove the bleach container, dump out remaining bleach. Rinse this container with Millipore water to rinse all the bleach out. Add fresh Millipore water to the container and place it back on the Coulter Counter as before hold the lid down and "flush". Do this at least three times.
6. Remove the water container, and put the Isoton II jar back on the Coulter Counter. "Flush" at least three times.
7. Exit out of the software.
8. Turn off the power switch.
9. Clean up the area for the next user.

Note: If the concentration gets above 8% it can indicate a blockage, the counter should automatically try to clear the blockage.

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**Verified by:** \_\_\_\_\_ **Date:** 4/5/2013

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