Coulter Counter Z2

* Remove Coulter Clenz (blue detergent) from platform and rinse aperture with isotone II
* Submerge aperture in 20mL of isotone II in a fresh 25mL Accuvette vial
* Prime Aperture by selecting “Prime Aperture” in Functions
* Check settings in Set-Up
* If the Kd, gain, or current is changed, you will need to measure aperture and optimize settings by pressing start on screen S2 in Set-Up
* Once all settings are correct, run a blank of 20mL isotone II by pressing start on screen S1 in Set-up or on screen A1 in Output
* You want a background ~300 counts or less
* If the background hasn’t dropped after a couple blank runs, you may need to prime in isotone II again and then recheck background
* For Yeast:
  + 100ul of 1 OD/mL culture into the bottom of the Accuvette vial
  + Pump in 10mL of isotone II to mix
  + Gently swirl
  + Place on platform and press start
  + Watch for blockage of the aperture while it its running. If blocked, stop run and use Kimwipe to gently rub away blockage
  + Gently swirl sample and try again
  + Want around 1 x105 events
* Once sample is counted:
  + open Coulter Z2 AccuComp on the computer
  + Select “Preferences” – “Preference files” – “Load Preference” and select saved preference
  + Acquire by either selecting “Acquire” – “Acquire from Z2” or click on the shortcut on the taskbar
  + Name and save sample data before running the next sample on the Z2
* In between samples, submerge aperture in 20mL isotone II to wash off the outside
* Every ~10 samples, run a blank with clean isotone II in a fresh Accuvette vial a couple times until the background drops to ~300 counts
* Once you are done with your runs, place 20mL Coulter Clenz in fresh Accuvette vial and Prime the machine
* Leave Coulter Counter Z2 turned on

Coulter Z2 AccuComp

* To overlay and export data
  + Select “File” – “Overlay”
    - Select data’s folder in Recent Directories, if folder is not listed select a folder with the subdirectory you need and toggle back using the [..] symbol in Directories then work your way forward selecting the Directories listed to create the path to your folder
    - Change file name from Multisizer (\*.#\*) to All Files (\*.\*)
    - Select files and click “add” to transfer it to “Selected Files”
      * The order they are selected in the order they will appear in the Overlay
    - Click “OK” when finished selecting files
    - In graph image, select region and click “View”- “Zoom In”
    - Change graph to “Number”
* To export:
* Select ”RunFile” – “Export”
* Select “Statistics”, “Tab delimited”, and “All runs”
* Click “export”, will export excel file to the folder you retrieved the data
* Open excel file and click ”yes” that it is a trusted file
* Adjust font size in “Home” and Margins and Orientation in “Page layout”
* Try to adjust and move data to fit on one page
* Return to Coulter Z2 AccuComp and change graph back to “Number %”
* Select “Edit” – “Copy Window”
* Return to Excel file and “Paste”
* With graph image selected, click on “Format” in the Picture Tools of the Toolbar
* Select the “Size and Properties” extension from the “size” section
* Scale the height and width equally to fit your excel page
* Unselect the graph image, click “File” and “Save”, click “No” so you do not save in a Text (Tab delimited) format , save in a .xlsx format
* Click “File” – “Print”