

ADVANCED CYTOMETRY & SORTING FACILITY AT SOUTH CAMPUS

BD CELESTA

QUICK GUIDE: START UP, QUALITY CONTROL, CLEANING AND SHUT DOWN

Making Cancer History®

PROCEDURE

1.0 Start Up

1.1. Check that the sheath cube is full, the waste tank is empty, and the FACS Flow Cart is on. Add 250 ml of bleach to the empty waste tank. If the sheath cube is low, remove the probe, replace the cube, replace the probe, and press "Restart" on the FACS Flow Cart.



1.2. Turn on the computer. Log in to Windows as the Operator. The password is "BDIS". Double-click the DIVA icon to open the instrument acquisition software. Log in to the software.

| DIVA icon | Login to the FACSDiva Software |] | |
|-------------------------|--|---|--|
| BD FACSDiva Software | Log In Control Control Contro | | Click the scroll down icon and select your name and login |

1.3. Turn on the instrument system power by pressing the green button on the right side of the instrument.



1.4. Ensure that there is a tube with approximately 2 ml of diH₂O on the SIP. Press the PRIME button to prime the instrument. Repeat priming again after the instrument has returned to STANDBY.

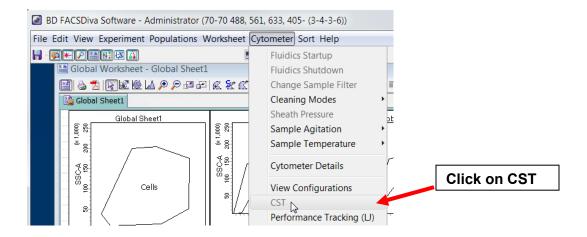


2.0 QualityControl

- NOTE: QC is run every weekday morning by Core staff, you need to run the QC only if Core Staff have not run it for the day. If you run the QC and if CST fails please contact Core staff via email or a note on the white board.
- 2.1. Open the CST experiment template from the Flow Lab folder. Press the RUN and LOW buttons on the instrument. Click "Acquire Data" on the Acquisition Dashboard and verify that events are accumulating on the plot. Remove the diH₂O from the SIP and replace it with the CST beads.

| Click on Experiment - New Experiment - Flow core - CST Template - OK | | | | | | | | | | |
|--|---|--|--|--|--|--|--|--|--|--|
| Image of the sector of the | eter Sort Help 日日和 N 画 勝 相 年 曲 供 刻 回 画面 Breet @ Experiment Templates Wang wang Watawah Wu Yee Zal 月 | | | | | | | | | |
| Experiment Layout | Sood SunX TCH Tonietti Ulirch Marcei Tonipati Soodal Reserver Romovali Overvilki Ozgotali Ral Reserver Tonipati Morriso Numerie Ontowali Overvilki Ozgotali Ral Reserver Tonipati Morriso SociA Maria Molfenti Molfenii Morriso Level Andrewilki Tonipati Morriso SociA Maria Molfenii Morriso Level Reserver Morriso Level Reserver Gallo Gallo Gallo Caporti Level Indemain IMPforms Gallo Gallo Gallo Caporti Chen Tonienti Morriso Divis Dention Doren Creft Level Indemain Morriso Level New Hitti Divis Dention Doren Creft Level Indemain Formation Level Level Indemain Divis Dention Doren Creft Level Indemain Level Level Indemain Divis Dention Doren Creft Level Indemain Level Indemain Divis Dention Doren Creft Level Indemain Level Indemain Gallo Gallo | | | | | | | | | |
| T SPP + | Name Date 8 Peak 9/26/18/8/5 AM Accordrep toropolelay 10/13/15/9-512 Calibriant drtt experiment 2017/222 22/81/12/80 Calibriant drtt experiment 2017/222 22/81/12/80 CST_and Accordry CC Template 4/9/18/19/9.40 CST_Drift 2/1/17/9.15 AM | | | | | | | | | |
| " | Name: CST and Accudrop QC Template Copies: 1 00 OK Cancel | | | | | | | | | |

- 2.2. Continue acquiring data for long enough to verify that two scatter populations and three fluorescent populations are discernable. Click "Stop Acquisition".
- 2.3. On the main toolbar select Cytometer > CST and wait until the cytometer connects to the CST software.



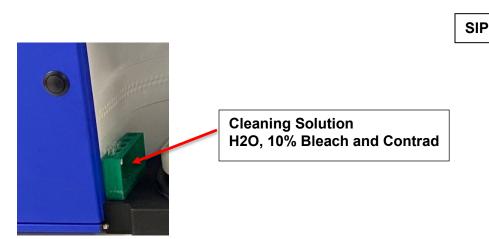
2.4. Ensure that the correct CST bead lot number is selected then click "Run". Review the Performance Tracking Report, and make a note of any problems or errors on the whiteboard.

| | | C | ytometer | Performa | ince Repo | ort | | |
|---|---------------------------------------|---------------------------------|-----------------|------------------------|--|----------------------------|---|--------|
| Cytometer: FACSAriaIII Cytometer Name: FACSAriaIII Serial Number: P658919000001 Input Device: Manual Cytometer Configuration: 70-70 488, 561, 633, 405- | | 5- (3-4-3-6) | | | User: Institution: Software: Date: Cytometer P/F: | Baseline: | Administrator N/A BD FACSDiva 8.0 02/21/2020 09:06 AM 02/11/2020 11:18 AM Pass | |
| Setup Bead | 5 | | | | | | | |
| Bead Product: Lot ID: Bead Lot Infor Detector Se | ST Exp 9 30 mation: A vai | rt #: 914 Jiration Date: 11/ | 3858 30/2021 | | | | | |
| Laser | Detector | Parameter | Target Value | Actual Target Value | % Difference Target Value | Bright Bead % Robust CV | Mid Bea Median Cha | |
| Blue | FSC | FSC | 125000 | 124585 | -1 | 3.38 | 124378 | 3 3.44 |
| Blue | с | SSC | 125000 | 125463 | 0 | 6.25 | 125987 | 7 6.46 |
| Blue | В | FITC | 5910 | 6173 | 4 | 3.86 | 128 | 13.37 |
| Blue | A | PerCP-Cy5-5 | 14073 | 13832 | -2 | 4.91 | 407 | 14.9 |
| Red | с | APC | 14043 | 13646 | -3 | 3.3 | 462 | 7.44 |
| Red | В | Alexa Fluor 700 | 8973 | 8637 | -4 | 4.33 | 317 | 7.56 |
| Red | A | APC-Cy7 | 8970 | 8605 | -5 | 4.66 | 300 | 7.64 |
| Violet | F | DAPI | 2586 | 2572 | -1 | 4.46 | 187 | 10.33 |
| Violet | E | AmCyan | 11594 | 11553 | -1 | 4.05 | 276 | 8.21 |
| Violet | D | BV605 | 18066 | 18210 | 0 | 5.19 | 407 | 23.97 |
| | с | BV650 | 19044 | 18915 | -1 | 4.38 | 975 | 12.69 |
| Violet | В | BV711 | 9781 | 9467 | -4 | 5.36 | 409 | 12.22 |
| Violet Violet | | BV786 | 14435 | 13928 | -4 | 6.92 | 332 | 16.06 |
| | A | | | | -4 | 4.89 | 828 | 7.66 |
| Violet | A D | PE | 13241 | 12836 | | | | |
| Violet Violet 561 Yellow- | | PE PE-Texas Red | 13241 10951 | 12836 | -5 | 5.24 | 231 | 13.9 |
| Violet Violet 561 Yellow- Green 561 Yellow- | D | | | | -5 | 5.24 | 231 271 | 13.9 |

- 2.5. Remove the CST beads from the SIP, replace the diH₂O, and place the instrument in STANDBY.
- 2.6. Exit the CST software and allow the instrument to reconnect to DIVA.

3.0 Cleaning

- 3.1 Place a tube with approximately 3 ml of Contrad on the SIP and leave the sample arm open. Press RUN and HIGH and leave it for one minute. Close the sample arm and continue running for one minute.
- 3.2 Repeat this procedure using a tube of 10% bleach.
- 3.3 Repeat this procedure using a tube of diH2O.
- 3.4 Leave the diH2O on the SIP and place the instrument in STANDBY.



4.0 Shut Down

- 4.1 You must clean the instrument before shutdown.
- 4.2 Turn off the instrument system power by pressing the green button on the right side of the instrument.
- 4.3 Exit DIVA and shut down the computer.



RELATED PROCEDURES

This handout is related to ACSF SOP IN012. Please see the full SOP for further information.

Handout: IN012-01

Version 1.1

2020-04-06