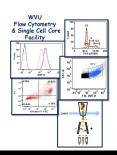
# WVU FLOW CYTOMETRY & SINGLE CELL CORE FACILITY



Newsletter Volume 4, issue 1

**July 2017** 

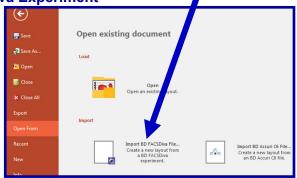
# Importing FACSDiva Experiments (including graphs and gates) into FCS Express 6 Analysis Software



Did you know that you can now import your FACSDiva experiment directly into the FCS Express 6 analysis software? With the current version of FCS Express you can import all the graphs and gates you made in FACSDiva directly into the FCS Express software. This means you don't have to start your analysis from scratch.

How do you import a FACSDiva experiment into FCS Express software? It is quite simple. Just remember to transfer the Exported Experiment File folder from the FACSDiva software to your memory stick along with the Exported FSC File folder. Below are step by step instructions on how to import your experiment into the FCS Express 6 software.

- After you running all your samples on the flow cytometer and collected the data using FACSDiva, export the experiment and the FCS files as you normally do
- 2. Login to the FCS Express 6 software using your user name and password provided to you by the facility
- 3. Click on the 'FILE" tab
- 4. A menu opens, click on "OPEN FROM"
- 5. Click on "Import BD FACSDiva File, create a new layout from BD FACSDiva Experiment"



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Facility Location:		

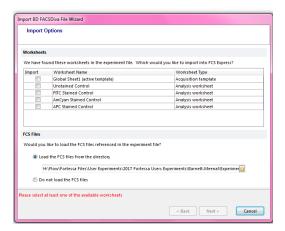
2160 HSCN
Phone:304-293-6273
e-mail: flowcore@hsc.wvu.edu

Hours of operation: 9:30 am to 5:00 pm, M-F

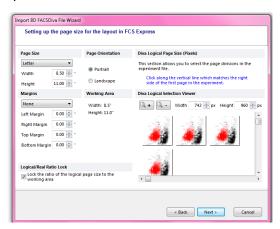
After hours access for experienced users with prior approval from Kathy Brundage

Contact Kathy at: kbrundage@hsc.wvu.edu

- 6. Locate and select the experiment from DIVA Experiment Folder. It has an XML extension.
- 7. A box opens up



- 8. From the choices listed in the box, select the type of worksheet you want to open
- 9. Select "Load the FCS Files from the Director"
- 10. Select the data files you exported in Diva.
- 11. Click "NEXT"
- 12. If you have a configuration file saved then select that option
- 13. If you don't have a saved configuration file or want to create a new configuration file select that option and another box opens



- 14. In this new box, select page size, layout in FCS Express and Save for future use
- 15. Select the data file you want to load initially in the template.

Now you can use the FCS Express software just like you did before. If you would like some help the first time stop by the facility, I will be glad to help you.

# **New User Guide**

Hands-on training for FACSCaliber, LSRFortessa, C1 Single Cell Auto Prep System, NanoSight NS300 and Zetasizer Nano Z is **mandatory** for all new users and must be scheduled by consultation with the facility director.

Training will initiate with user's first experiment. Due to the complexity of the instruments and software, facility staff will fully assist with the acquisition of the first dataset and will continue with additional assistance on a "needs" basis until users are comfortable operating the instrument on their own. Sorting on the FACSAria is by facility staff only.

# Flow Cytometers in the Facility

# **FACSAria III Cell Sorter**

**Operator: Facility Staff** 

#### Lasers:

488 nm solid state 561 nm solid state 633 nm solid state

407 nm solid state

### **Detection Parameters:**

**Forward Scatter** 

**Side Scatter** 

Simultaneous detection

of 13 fluorochromes

# **Applications:**

Cell Sorting (Aseptic)

**Cell phenotyping** 

**Cell Viability** 

FISH, FRET, SPA



# **LSR Fortessa**

**Operator: User of Facility Staff** 

### Lasers:

405 nm solid state

488 nm solid state

561 nm solid state

628 nm solid state

### **Detection Parameters:**

**Forward Scatter** 

**Side Scatter** 

Simultaneous detection

of 12 fluorochromes

### **Applications:**

**Cell phenotyping** 

**Cell Viability** 

**Cell Cycle analysis** 

FISH, FRET, SPA



# Other Instrumentation Available in the Facility

## **AutoMACS Magnetic Bead separator**

Operator: User Application:

Single extracellular marker cell sorting

Depletion/negative cell sorting

# gentleMACS Octo Dissociator with Heaters

Operator: User Application:

Dissociation of tissues into single cell suspension for culture or flow cytometry assays

Homogenizes tissues for downstream molecular biology applications

# C1 Single Cell Auto Prep System

**Operator: User or Staff** 

Application:

Uses microfluidics, to separate cells into individual compartments, isolate RNA from the single cells, and generate cDNA for downstream genomic applications.

**Downstream applications:** 

RNA seq DNA seq PCR

Format: 96 or 384 chambers per chip



# **MSD Multi-Array Platform**

Operator: User Applications:

Detection of cytokines, cell signaling proteins Multiplexed assay design: (1-10 analytes/plate)

Detection range: 1 – 10,000 pg/ml Sample volumes: 25 μl or less

Assay Time: 4—6 hours depending on analytes

being detected



### Nanosight NS 300

**Operator: User or Staff** 

**Application:** 

Determines the size and concentration of 10 nm to 2 microns particles in size

Equipped with 4 lasers (405 nm, 488 nm, 532 and 642) to detect fluorescently labeled particles



## **Zetasizer Nano Z**

**Operator: User or Staff** 

**Application:** 

Measures the zeta potential of particles in a solution using laser Doppler micro-electrophoresis



# Fee Schedule

Instrument	Operator	For WVU & NIOSH Users	For Non-WVU Users
AutoMACS	Facility Staff or User	\$14.70	\$22.05
C1 Single Cell Auto Prep System	Facility Staff	\$210/plate	\$315/plate
	User	No Cost	\$315/plate
FACSAria III	Analysis: Facility Staff	\$52.50/h	\$78.75/h
	Analysis: User	\$34.65/h	\$51/98/h
	Sorting	\$77.70/h	\$116.55/h
	Sorting Setup	\$19.43/sort	\$29.15/sort
gentleMACS	Facility Staff or User	\$10.50/sample	\$15.75/sample
LSRFortessa	Facility Staff	\$52.50/h	\$78.75/h
	User	\$34.65/h	\$51.98/h
MSD Sector Imager	Facility Staff or User	\$10.50/h	\$15.75/h
NanoSight NS300	Facility Staff	\$61.00/h	\$91.50/h
	User	\$42.50/h	\$63.75/h
Zetasizer Nano Z	Facility Staff	\$25/sample + \$52.50/h	\$25/sample + \$78.75/h
	User	\$25/sample	\$25/sample + \$78.75/h



#### Internal WVU user:

Click  $\underline{\text{here}}$  to login or register using your institute login and password.

### Not a WVU user?

Login using iLab credentials

If you don't have an account, please  $\underline{\text{register}}$  for an iLab account.

The facility uses iLAB scheduling/billing software from Agilent to manage the use of the facility's instrumentation. If you would like to use the instruments housed in the facility please use the URL shown below to register as a WVU User and to login to reserve an instrument.

https://wvu.corefacilities.org/account/login

# The New iLAB Scheduling/Billing Software

As of August 1st, the facility will be using a new scheduling/billing software called iLAB. Although there are a lot of similarities between the old CORES software and the new iLAB software there are some major differences.

- 1. All users whether or not they are at WVU must register in order to use the software. You can register at https://wvu.corefacilities.org/account/login by clicking on "register a new account".
  - 2. In order to make an appointment you must be linked to at least one funding string
- 3. All PIs must have a registered account and link their lab members to the appropriate funding string.
- 4. The first time you try to make an appointment you will need to contact me so that I can give you access to the equipment.
  - 5. Billing will be done on a bi-monthly basis. The 2nd and 4th Monday in a month.
  - 6. If you are not at WVU, a invoice will be sent out for payment using a check

As with all new systems, I am sure there will be bumps along the way. Please contact me immediately if you do encounter a problem so that it can be resolved quickly. I appreciate your patience as we undergo this transition.

Upcoming Holidays & Events				
September 4	Labor Day	Facility Closed		
October 16—30	Kathy out of lab	Facility open for experienced users only.  No Sorting		
November 22—23	Thanksgiving	Facility Closed		

### Note to users:

Please acknowledge the WVU Flow Cytometry & Single Cell Core Facility when reporting your data, using the appropriate phrase(s):

<u>LSRFortessa users:</u> Flow Cytometry experiments were performed in the West Virginia University Flow Cytometry & Single Cell Core Facility, which is supported by the following National Institutes of Health equipment grant S10OD016165 and the following Institutional Development Award (IDeA) grants U57GM104942 (WV CTSI), P30GM103488 (CoBRE) and P20GM103434 (INBRE).

**For all users of the facility:** Experiments were performed in the West Virginia University Flow Cytometry & Single Cell Core Facility, which is supported by the following National Institutes of Health and the Institutional Development Awards (IDeA) grants U57GM104942 (WV CTSI), P30GM103488 (CoBRE), P20GM109098 (Stroke CoBRE) and P20GM103434 (INBRE).