

Penn Cytomics & Cell Sorting SRL Policies

Access: All instruments located within the centralized laboratory (206/231/297 John Morgan Building) are available 24 hours/day, 365 days/year. Rooms housing these instruments are unlocked Monday-Friday, 8am-6pm. For after-hours access to these rooms, 3 requirements must be met: you must have a Penn ID for door access, you must have completed the analyzer training module, and you must have emailed william.murphy@pennmedicine.upenn.edu to request after-hours access. All Penn Cytomics satellites are accessible Monday-Friday from 9am-5pm. Satellite facilities are only available after hours for users within that building/department, or for users with authorization. Because of long standing support from the Abramson Cancer Center (ACC) project grant, highest priority is given to ACC members with peer-reviewed funded projects, followed by ACC members with other funded projects and non-ACC members with peer-reviewed funded projects, non-ACC members with funded projects, and finally users from outside the institution. There is no discount to ACC members vs. non-members.

Biosafety: Flow cytometric analysis of human and other potentially biohazardous samples is permitted at all of our locations, so long as appropriate precautions are taken. All lab materials must be disposed of correctly, even if they are not contaminated. Each satellite facility has dedicated receptacles for gloves and sharps; FACS tubes and pipette tips are considered to be sharps. Cell sorting of biohazardous material is only allowed on certain instruments, pending staff approval of a designated biosafety form. Sorting of BSL-2⁺ materials is only permitted on a dedicated instrument in a biohazardous suite (Aria D) and may only be performed by appropriately trained staff equipped with a PAPR. (Note: The SRL does not support radioactive samples/materials for analyzers and sorting).

Lab Coats/PPE: As mandated by the University of Pennsylvania Environmental Health and Radiation Safety (EHRS) office, lab coats must be worn at all times while using any Penn Cytomics instruments. Penn Cytomics supplies gloves to users in all locations. Eating/drinking in lab areas is strictly prohibited.

Cytometer User Etiquette:

All users should:

- Fill the sheath and empty the waste container before running their experiments.
- Clean the cytometer after their session.*
- Make sure the cytometer is in STANDBY mode before leaving.
- Take their tubes, tube racks, and waste/trash back with them.
- Shut the cytometer off if they are the last scheduled user of the day.

*Run on High Speed

1. 2% Contrad for 30 seconds with the arm to the side
2. 2% Contrad for 5 minutes with the arm under
3. Water for 30 seconds with the arm to the side
4. Water for 2 minutes with the arm under

5. FACSClean Bleach for 30 seconds with the arm to the side
6. FACSClean Bleach for 5 minutes with the arm under
7. Water for 30 seconds with the arm to the side
8. Water for 2 minutes with the arm under

Note: Always run FACSClean Bleach when running human samples

FACS tubes: Penn Cytomics recommends the use of Corning Falcon brand 5mL round-bottom polystyrene (Cat. 352054) or polypropylene tubes (Cat. 352063). Other brands of tubes may not fit the cytometers appropriately. As of January 1, 2022, the use of tube inserts (e.g., cluster tubes, bullet tubes) are not allowed on any Penn Cytomics instruments, as they lead to damage of the BAL seals.

[Fisher Polystyrene Tubes](#) (All Analyzers, Melody, Aria II and AriaFusion)

[Fisher Polypropylene Tubes](#) (Influx, Aria II, and AriaFusion)

Entrée: Entrée is our usage and data backup software and identifies when a session is taking place. If the previous user forgot to log out of Entrée, **DO NOT USE THEIR SESSION**. Please log them out and log in with your credentials. Failure to do so will result in your data going to the previous user, and you will also be billed as a no-show. Please leave an Entrée message at the end of your session to notify the core of any log in/log out issues. Of note, you are not required to use the same project in Entrée that you chose when you initially scheduled your instrument booking.

Billing: A billing session is initiated the instant a user logs into an instrument using their Penn credentials, to the moment the user logs out of the cytometer's acquisition software. The SRL usage and billing software, termed Entrée, tracks instrument usage to the second, and generates a billable line item to be invoiced monthly. Users are able to select an appropriate funding source to bill when they initiate a session. In order to encourage maximum usage, and to prevent overestimation of time needed on an instrument, SRL policy is to bill a minimum of 75% of scheduled time for underused sessions, and to bill for actual usage for sessions lasting longer than the reserved time. For example, a user who reserves 4 hours of instrument time and uses 1.5 hours will be charged for 3 hours of usage. Users who fail to use reserved instrument time (no-shows) likewise trigger a minimum charge of 75% of scheduled time. Principal Investigators and Departmental BAs will receive a separate monthly invoice notifying them of these charges.

Scheduling: The SRL online scheduling calendar is available for trained users to reserve instruments up to 30 days in advance. Users may only schedule up to 3 hours of consecutive time during normal operational hours (Monday-Friday, 9am-5pm). Longer sessions are acceptable during off-peak evening hours and weekends (see Access Policy). High-parameter analyzers (A5/X-50/SE Symphonies) are excluded from this policy, as it is recognized that these experiments generally require a significantly longer time to complete.

The following policies have been implemented in order to maximize instrument usage:

In an effort to minimize placeholder reservations:

For cell sorter reservations (trained users only) and *most* analyzers: The scheduling calendar prevents adding, modifying, or deleting instrument bookings within a six-hour time window before a user's scheduled time.

For A5/X-50/SE Symphonies: The scheduling calendar prevents adding, modifying, or deleting instrument bookings within a 36-hour time window before a user's scheduled time.

If a user needs to schedule, modify, or delete a reservation within these time windows, they will have to call 215-898-3528 and speak to a staff member.

The SRL enforces a 72-hour cancellation policy for staff-performed cell sorting reservations; users are billed for the entirety of the scheduled time, unless the SRL can fill that time interval with another reservation.

Users may not "share" scheduled time with fellow lab members, as this will result in a double charge within the SRL billing system (User 1 is billed as a no-show, User 2 is billed for actual instrument usage). In this instance, User 1 should cancel their reserved time, and User 2 should reserve the instrument under their own credentials.

Data and Nomenclature: User data are stored on the local computer for 4 weeks after acquisition for analyzers and 3 months after acquisition for sorters. When users export data from the acquisition software, and upon completion of the usage session, Entrée automatically uploads data to two independent locations. Please use the following naming convention: PennKey_YYYYMMDD_Descriptor (ex: jonesder_20221103_Data). Do NOT use special characters, except for underscore (_) and dash (-). This convention allows staff to identify the owner of the data, indicates when data are eligible for deletion from the computer, and assists in future data retrieval requests.

Computers: The computers are for data acquisition and analysis only. Internet browsing is not allowed, as the computers are on a local network. In addition, USB flash drives are forbidden in any Penn Cytomics workstation computer, as to prevent the transmission of viruses and other malware.

Consumables Provided: Penn Cytomics supplies sheath fluid, 2% contrad, FACSClean, bleach, detergent, Kimwipes, and gloves. Labware/reagents, such as tubes, filters, transfer pipets, pipette tips, buffer, etc. are to be supplied by the user. Lab coats and other PPE are the responsibility of the user. In satellite locations (areas other than JMB), the department is responsible for disposing of its own sharps containers, biohazardous waste, and trash. In satellite locations, chairs/furniture are also the responsibility of the department to replace.

SRL Policy Violations: The SRL employs a 3-tiered process to address failures to adhere to written policies. After a first-time offense, SRL staff will reach out to the responsible party to

specify which policy was broken. If this user becomes a repeat offender despite staff intervention, their Principal Investigator will be notified. If the user still fails to adhere to policy, they risk suspension of privileges. Staff members use their judgment based on the severity of the offense (e.g., failure to wear a lab coat is much less severe than sorting biohazardous material on a non-approved cell sorter).

Acknowledgements: All publications that include results or products generated in the Penn Cytomics and Cell Sorting Resource Laboratory must include the following acknowledgement: “The data for this (manuscript or presentation) were generated in the Penn Cytomics and Cell Sorting Shared Resource Laboratory at the University of Pennsylvania and is partially supported by the Abramson Cancer Center NCI Grant (P30 016520). The research identifier number is RRID:SCR_022376.”