

Ragon Institute Flow Cytometry Facility

General Usage Guidelines

Visit our website!

http://www2.massgeneral.org/aids/flow_cytometry.html

Or www.ragoninstitute.org and follow the link to “flow cytometry” on the right side of the page.

It contains information about our instruments, where we are located, links to sign up calendars and flow resources, and many of the documents used by the facility, as well as a flow cytometry syllabus of resources to expand your flow knowledge.

We offer access to our instruments to all members of The Ragon Institute and MGH. To use our instruments, you must first create an account. ALL NEW USERS (including new technicians) MUST BE TRAINED BY FACILITY PERSONNEL before using the instruments independently. Please email Michael Waring (mwaring@partners.org) and include the following information:

- your name
- a grant number for billing, along with the TITLE of the grant.
- the FULL NAME of the PI on the grant
- If the grant is and NIH fund, provide the NIH number.
- what group/department you are a part of
- your telephone number
- Describe any previous flow cytometry experience (instruments used, etc)
- information about the samples you are interested in running (type of sample, number of parameters you wish to measure, etc)

You will also be required to attend an Intro to Flow Cytometry class before your account will be activated, followed by an orientation session on the instrument you wish to use. Please visit our training page for more information (follow the menu on the left side of our main page linked above).

The following services are provided to members of the CFAR and MGH communities. Please visit specific instrument pages on the website for applicable costs/rates:

- User-run analysis
- Operator-run cell sorting (PLEASE NOTE: There is no user-run sorting.)
- Flow Cytometry classes are held roughly every 6 weeks. See the Training page for more information.
- Advice and assistance in planning your flow experiments.

--All Users are on an email notification list for updates, problems, etc. Please let me know when you are leaving MGH so I can remove you from my list and delete your accounts.

- Please let me know if anything is not working properly or you have any problems with the instruments, website, or the facility in general-- email mwaring@partners.org or parcfacs@partners.org. Positive comments are welcomed as well!
- Information about each of our instruments can be found on our website. There are additional user documents found on each instrument page and on the Core Documents page, including user guides for each instrument. User guides are provided in hard copy near each instrument.
- Instrument reservations are booked online through www.schedulebook.com, a user account is required for access. A guest account is available for viewing the calendars only, username flowguest password guest. Email mwaring@partners.org to request access to reserving instruments. The sorting calendar and Mike's schedule are only available for viewing, users are not permitted to make their own reservations and are required to email their requests.
- Researchers that are not members of the Ragon Institute may not schedule reservations more than 1 week in advance.
- There is a 2 hour limit during peak usage hours (12 pm to 9 pm), and no more than 2 reservations per week can be between 2 and 6 pm.
- For sorting, Tuesday and Thursday afternoons from 2:30 to 4:30 are reserved for members of the Ragon Institute. Any unreserved time is released to all users on the day before the event.
- The LSR II's are reserved from 3-7pm on Tuesday, Wednesday, and Thursday every week for members of the Ragon Institute, and unreserved time is released to all users the day before the event.
- If you do not arrive for your reservation within 20 minutes of the start time, anyone else can use the instrument and you forfeit your reservation.
- Delete/cancel unwanted reservations 1 hour before start time or you may be billed 1 hour of usage time. Please update your reservations as soon as you know they are different from the scheduled time.

ALL HUMAN SAMPLES MUST BE FIXED!!

MGH considers all materials of human or non-human primate origin to be BL2. All such samples run on the analysis instruments must be fixed as below. It is possible to run viable cell lines of human origin on the 4 Laser LSR, but this requires additional precautions and training; contact Mike if you have questions about it. Unfixed primary cells or blood samples should NEVER be run on our instruments by users; if it is necessary to acquire data from such samples, this can be arranged on our FACS Aria sorter which is housed in a biosafety cabinet. Uninfected cell lines prepared for cell cycle staining can be fixed in 70% EtOH for a minimum of 30 minutes.

Please remember that it is very important to use fresh paraformaldehyde to be sure that any viable/potentially infectious organisms in your FACS samples are inactivated.

- Stock solution (i.e. 10%) of PFA can be frozen.
- We ask that you fix for 30 minutes in 2% PFA.
- Make sure your sample is resuspended before fixation to help reduce clumping.
- Working solutions should not be more than 3 weeks old, ideally it should be made fresh every week and stored at 4C in the dark (wrap in foil).