

# User guidelines for the Flow Cytometers at the Institute of Microbiology, Infectious Diseases and Immunology

Before using our equipment, make sure that you are familiar with our user guidelines and billings structure. By accepting these guidelines, you confirm that you have read and understood all chapters and will act as intended within this document. Any misuse can lead to an exclusion from the user list and so to the denial of all granted permissions.

These guidelines contain following chapters:

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# User guidelines for the Flow Cytometers at the Institute of Microbiology, Infectious Diseases and Immunology

## General Remarks

The flow cytometers of the Institute of Microbiology, Infectious Diseases and Immunology are placed in room E906 of the Campus Benjamin Franklin and are for general use. They were expensive, so treat them with care, in order to maintain the instruments in a good condition.

The entire procedure of registration and booking the instruments is carried out via openIRIS, an online booking tool implemented in the Charité domain environment. A prior registration in openIRIS is necessary for the registration and usage of the flow cytometers. Contact responsible if you have problem with the registration. The URL for openIRIS is <https://charite.science-it.ch/>.

It is not allowed to make reservations for anyone other than yourself.

It is forbidden to use the computers for any other purpose than data acquisition and analysis.

The flow cytometry Lab, H. 5011 Room E906, is a S2-laboratory. Users must wear personal protection equipment, lab coat, gloves, at any time

Any user who violates any of the rules described in this document will be obligated to pay for any repairs or instrument downtime.

## BD LSRFortessa X-20

For working on the BD LSRFortessa X-20 analyzer, an instrument training is required and can be scheduled individually. To do so, go to openIRIS, change to tab 'Service' and submit a request for 'Training request BD LSRFortessa X-20'. The training will ensure that user can self-operate the instrument and know all lab specific rules.

## Access to the flow cytometry lab

The LSRFortessa X-20 is self-operated and available at any time if access to the Institute of Microbiology itself is possible. To grant access to the Institute of Microbiology and so to the flow cytometry lab, you have to provide a transponder number and the name of its holder. If you do not have a transponder, a filled-out key request ("Schlüssel Antrag") of the Charité is needed.

## Scheduling

After the training has been completed, the instrument can be booked via the tab 'Scheduler' in openIRIS. The advance-booking period is 4 weeks for members of the Institute of Microbiology and for all other scientists it is 5 days.

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For every booking of the LSRFortessa X-20 the user has to provide information about the species and the biological safety level of the sample(s) going to be measured. With completing the booking the user confirms that no other sample as defined will be processed in the laboratories of the Institute of Microbiology.

If additional time is required on an analyzer, please ask the next user for permission. In the worst case, you have to suspend your measurement and wait until the instrument is available again. In any case, you are obliged to book the additional time used on the instrument in the booking calendar.

If an existing booking slot is cancelled or shortened within 24h before the start of that event, it will still be charged completely unless the initial booked time will be used by anybody else. Cancellation or shortening of bookings is generally allowed at any time.

## Responsibilities

The Flow Cytometers are high tech instruments and need careful handling to maintain their day-to-day performance, so all users are responsible for taking care of the instruments. Firstly that is the usage in accordance with regulation and as learned within the training.

If the instrument usage exceeds the booked time, users are obliged to book the additionally used time in the booking calendar.

Filtering the cells is recommended to avoid clogging of the instrument (35µm mesh, directly prior acquisition).

The last user of the day is responsible for shutting down the instrument. Check the calendar at the end of your measurement.

## Data management

In general, the responsibility for data created on the LSRFortessa X-20 lies with the user and not with the Institute of Microbiology.

The computers connected to the instruments are for data acquisition only and not for long-time data storage. The DiVa database is very sensitive to size, so users are not allowed to keep data in it and must export their data at the end of their appointment. In order to maintain the data integrity, it is recommended that users should first export the data out of the DiVa software to a local drive. A folder structure was generated for this purpose: D:\users\

If any data found in the database, it will be deleted without further notice.

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To protect the acquisition computer from viruses, trojans, etc. it is strictly not allowed to plug in any external harddrives.

## BD FACSAria Fusion

The work on the BD FACSAria Fusion cell sorter requires a project in openIRIS. To create it, go to openIRIS, open the tab 'Services' and submit the request 'FACS Aria Fusion Cell Sorting Project at CBF'. The default project duration is 12 month and after this period the project details should be revised before prolonging it. A meeting is scheduled after creation to discuss experimental and technical needs and possibilities, e.g. cell preparation, staining panel, analysis, etc.. This is necessary to avoid misunderstandings what may lead to time consuming and cost intensive experimental mistakes.

Cell sorting is offered as a service what means that only designated operators run the FACSAria Fusion. This is at the moment by Jens Hartwig only. The service hours for cell sorting are:

Mo-Thu 9.30 – 18.00

Fr 9.30 – 14.30

Outside of these times sorting cells is only possible in exceptional cases. One of these exceptions are so-called power users. These are users who got an intensive training on the Aria Fusion and so, are able to do cell sorting on their own.

## Scheduling

If a project was created in openIRIS and a meeting was held, user can now book the FACSAria Fusion via the 'Scheduler' tab of openIRIS. Bookings are requests for the cell sorting service of the Institute of Microbiology and must contain a fully filled out request form what will automatically open. By submitting the request, the user confirms that only the samples specified in the project and in the request will be brought for sorting. The request is accepted if cell sorting is possible, if not, an alternative date is proposed.

Booking request can be made 2 weeks in advance. In urgent cases or for exceptions concerning service hours, direct contact is appreciated.

Cancellations and sort time reductions can be made at any time but if that happens within 48h before the start of the sort appointment the cancelled time frame will be charged anyway unless it will be used by others.

## Responsibilities

For each staining panel, appropriate control samples are needed: fully unstained and single stains cells or compensation beads.

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Each samples will be filtered through a 35µm mesh directly before the sort to avoid clogging of the instrument.

## Data management

The data created during cell sorting have to be exported and stored in a network folder specified by the user. Data consists of fcs files, the DiVa experiment and a pdf file including sort reports and worksheets of samples and reanalyses. The safety of the data on the network folder can not be guarenteed by the Institute of Microbiology.

After each cell sorting appointment the entire experimental data is deleted out of the DiVa database.

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## Billing procedure

We charge for every 30 minutes started. You will be charged for the entire duration of all bookings made in the online calendar for every quarter of a year irrespective if you really have used the instrument or not.

Actual billing rates for instrument usage and training per half hour are:

| Instrument                       | Price |
|----------------------------------|-------|
| BD FACSAria Fusion, cell sorter  | 40€   |
| BD LSRFortessa X-20, analyzer    | 20€   |
| Training for BD LSRFortessa X-20 | €     |

## Acknowledgment

If publications or any other formal presentations were supported through the work of flow cytometers of the Institute of Microbiology, Infectious Diseases and Immunology this should be acknowledged in a way representing the effort of the staff.

## Closing remarks

As already mentioned the flow cytometers of the Institute of Microbiology are expensive high-tech machines and are operated by many users. So, please follow these rules to prevent unnecessary downtimes and expenses due to Deliberate infringement or careless behaviour will lead to a forbiddance of using our instruments.

If you have any questions, feel free to ask.