

Flow cytometer using instructions

2019.12 Revised

Equipment Introduction :

1. Model :

BECTON DICKINSON FACSAriaIIu (Laser ex : 375 or 405 & 488 & 635nm)

BECTON DICKINSON FACSCalibur (Laser ex : 488 & 635nm)

BECTON DICKINSON BD LSR II (Laser ex : 405 & 488 & 635nm)

ThermoFisher Attune NxT (Laser ex : 405 & 488 & 561 & 635nm)

2. Function :

(1) ANALYSIS : including cell surface and cytoplasm analysis

Cell Marker Analysis (Ag-Ab)

Cell Cycle Analysis (DNA content & sub-G1)

Apoptosis and Necrosis

Free Radical (ROS)

Membrane Potential

pH value

[Ca²⁺] analysis etc

(2) SORTING

Ordinary Sorting

Sterile Sorting

ACDU (clony : cell/well)

Remarks :

1. Analysis : about pH value 、 [Ca²⁺] analysis , because of its special experimental conditions, involving experimental design, calibration curve and application software and other factors, it is not yet officially provide services, if necessary, please prior to communicate with the technician to discuss. The remaining projects have been entrusted to the machine.
2. Based on the requirements of laser life and accuracy, FACSAriaIIu only provides SORTING services in principle, and ANALYSIS service using the FACSCalibur or Attune NxT. FACSCalibur also open by the user on their own machines, those who wish to apply to the center, after training, pass the qualification examination, you can on the plane. (For more information, please contact Center technician)

Place :

The ninth floor of the First Medical Building , Instrumentation Center (Room number : 0910)

Opening hours :

1. Monday - Friday AM 8 : 30 ~ PM 17 : 00
2. Off-hours or holidays : (Only for certified users)

If you have not set up access control card , please come to center get the backup access card before PM 17 : 00 ◦ Please return the card at 10 o'clock the next day ◦ The responsibility of borrowing period belongs to the card user. If you lose card, you will have to pay a card making fee of 500 NT dollars ◦

Application method :

Model BD FACSCalibur or Model Thermo Attune NxT or Model BD LSR II

1. Fully commissioned services or technician to accompany the practice and analysis : Please fill in the application form one week ago and take it to Room 0910.(We do not accept phone reservations)

Note: open booking day to next week

2. Use by yourself (everyone can do) : Please fill in the weekly booking schedule from Monday AM9: 00 at Room0910 ◦ (Need to participate in operating class, and was identified by technician)

Note: only open the appointment this week, If want to book Monday please contact technician

attention :

- a. The operation instruction is made by the users who wish to apply. Flow center will hold irregularities depending on the number and actual needs ◦
- b. Technical certification standards :
 - The principle of the flow cytometer ◦
 - Preparation of starting ◦
(PBS buffer preparation ◦ View of the pipeline ◦ description of the sample, etc.)
 - Machine checking ◦ (pressure ◦ pump ◦ laser ◦ flow rate ◦ machine condition test)
 - The relation of the software and the machine ◦
 - The procedure of cleaning before shut down, and then record the condition of machine ◦
 - Troubleshooting ◦

- c. Those who pass the certification exam must use more than 10 times or more than 15 hours during technician working hours to be able to use during non-working hours. If within two months there is no record of the aircraft, you will need to reconsider before you re-enter the aircraft. However, user who is using over 100 hours, will be able to present proof that no further authentication is required ! 「 Note: to prove information by yourself, then the technician audit 」 ◦

Model BD FACS Aria II_u

1. Fully commissioned services ; or technician to accompany the practice and analysis : Please fill in the application form one week ago and take it to Room 0910 ◦ (We do not accept phone reservations)

Note: open booking day to next two week

2. Use by yourself (Only for certified users) : Please contact the center technician and check the schedule to make a reservation. (We do not accept phone reservations)

Note: open booking day to next two week

Notes :

- a. Based on reducing the instrument damage rate and enhancing the research energy, the center provides one to two times of operation training and certification every year, three to four people each time, and apply for eligibility criteria please contact the center technician.
- attention :**
1. The training time will be announced by the center about every summer vacation ◦
 2. Those who accept the operation of trainers, training standards (accudrop beads 20000 NT dollars / EA) must be purchased before training. And then apply for the certification exam to the center within three months. If you need a technician to accompany you on the machine during the training period, you will be charged for the attestation.
- b. Apply for FACS Aria II_u Analysis Qualification: : who need to have FACSCalibur license and do FACS analysis for more than 50 hours , or who already have license of FACS Aria II_u sorter ◦ And then , IF new FACS Aria II_u uesr do not using mechine for a month, need to re-certification ; But with continuous operation of more than 200 hours of users to count.
- c. Those who pass the certification exam must use more than 15 times during technician working hours to be able to use during non-working hours. If within one months there is no record of the aircraft, you will need to reconsider before you re-enter the aircraft. However, user who is using over 200 hours, will be able to present proof that no further authentication is required ! 「 Note: to prove information by yourself, then the technician audit 」

Fee principle:

1. Charges: :

(a) analysis service fee

Model	Item	Charges fee (NT dollars / per hour)		
		Within institution	Global Research & Industry Allianc	other
FACSCalibur				
Configuration : 488nm laser 633 nm laser software : CellQusest Pro	Fully commissioned services	500	750	1000
	technician to accompany	500	750	1000
	Use by yourself (Only for certified users)	60	Not open	Not open
Model	Item	Charges fee (NT dollars / per hour)		
		Within institution	Global Research & Industry Allianc	other
Thermo Attune NxT				
Configuration : 488 and 633nm laser 405 and 561 nm laser software : Attune NxT Software	Fully commissioned services	500	750	1000
	technician to accompany	500	750	1000
	Use by yourself (Only for certified users)	60	Not open	Not open
Model	Item	Charges fee (NT dollars / per hour)		
		Within institution	Global Research & Industry Allianc	other
BD LSR II				
Configuration : 488 and 633nm laser 405 nm laser software : FACSDiva	Fully commissioned services	500	750	1000
	technician to accompany	500	750	1000
	Use by yourself (Only for certified users)	60	Not open	Not open

Fee principle:

1. Charges: :

(b) sorting service fee

FACS Aria II _u Add general boot fee 500 NT/Each , sterile sorting 1000 NT/Each					
Model	Item	Charges fee (NT dollars / per hour)			
		Within institution	Global Research & Industry Allianc	other	
FACS Aria II _u	A : cell sorting				
	Configuration : 488nm laser	Fully commissioned services	500	750	1000
	633 nm laser	Use by yourself (Only for certified users)	100	Not open	Not open
	B : cell analysis				
	375 / 405 nm laser	Fully commissioned services	500	750	1000
	software : FACSDiva	Use by yourself (Only for certified users)	100	Not open	Not open

2. Charge method: :

In each of March, June, September and December, the total cost of every three months will be counted and a detailed list of fees will be set out. After being entrusted by the commission department, we will be transferred to the account.

If you do not pay for PI research funding, you will have to pay the bill at the General Services Department.

Sample preparation:

1. carrying by yourself :

- (1) 41~53um nylon mesh
- (2) FALCON #2052 12x75 mm plastic tube
- (3) pipets (200~1000ul) & tips
- (4) Flash drive

2. Sample final concentration : $5 \times 10^5 \sim 1 \times 10^6$ cells/ml . Please filter the sample before putting it on the machine to avoid clogging the pipe.

3. Sample processing is not good make the machine can not be normal operation, you will be asked to reprocess the sample or cancel the experiment .

Precautions:

1. Fully commissioned services :

Sorting case : If you need to cancel the reservation, please be sure to notify the technician 1 days in advance, otherwise it will charge the boot costs. ◦

Analysis case : If you need to cancel the reservation, please notify our center 2 hours in advance, otherwise the fee for the booking period will be charged. (Note: It takes 1 ~ 2 hours to clean the pipelines and warm-up before getting on the machine, even overtime . To avoid wasting time, standard beads and laser power, please do not cancel the experiment temporarily) ◦

2. Use by yourself (Only for certified users) :

If you be late more than half an hour appointment on the plane, the technician may have to arrange for this time to other people on the plane. If you find that the experimental problems can not be on the plane, please cancel the booking or call the administrator to cancel the booking, if you forget to cancel will be charged for the booking period ◦

3. You are responsible for the cost of repairs due to the machine malfunction ◦

4. Violation of the provisions of three times, you will be forbidden to use the machine, and you can not apply within one year.

5. When the machine is abnormal, please notify the center technician immediately. If it can not be processed immediately, please shut down the machine with the correct procedure and fill in the usage record in detail.

Contact:

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