Flow cytometer using instructions

2021.12 Revised

Equipment Introduction:

1.Model:

BECTON DICKINSON FACSAriaII $_u$ (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 cytoplasma 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, FACSAriaII_u only provides SORTING services in principle, and ANALYSIS service using the Attune NxT or BD LSR II or FACSCalibur. Each instrument is also open for users to board by themselves. Those who wish can apply to the center. After training, they can board the machine by themselves after passing the qualification examination. (For more information, please contact Center technician)

Place:

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

Opening hours:

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

Please use your personal student ID or identification card to enter and exit by yourself, and do not lend it to others. At the end of the experiment, please make sure to turn off the light and the sensor door.

If you have not set up access control card , please come to center get the backup access card before PM 17 : 00 \circ Please return the card at 10 o'clock the next day \circ 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 \circ

Application method:

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

- 1. Online appointments are fully open for entrusting or self-boarding:
 - URL: https://insreservation.cgu.edu.tw/labs/4
- 2. How to make an appointment: Please refer to the webpage for details
 - (a) From Monday at AM 9:00, appointments will be open until next week
 - (b) It is not possible to change or delete the appointment by yourself on the day, please contact the administrator for processing

attention:

- I. The operation instruction is made by the users who wish to apply. Flow center will hold irregularities depending on the number and actual needs °
- II. 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
 - \circ Troubleshooting \circ

III. 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 ! \lceil Note: to prove information by yourself, then the technician audit \rfloor \circ

Model BD FACSAriaIIu

- 1. Online appointments are fully open for entrusting or self-boarding:
 - URL: https://insreservation.cgu.edu.tw/labs/4
- 2. How to make an appointment: Please refer to the webpage for details
 - (a) From Monday at AM 9:00, appointments will be open until next week
 - (b) It is not possible to change or delete the appointment by yourself on the day, please contact the administrator for processing
 - (c) If you entrust the machine, please contact the technician in advance to confirm the content of the experiment in order to facilitate the experiment

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 \circ
- 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 FACSAriaII $_u$ Analysis Qualification: who need to have any Analyzer license and do FACS analysis for more than 50 hours , or who already have license of FACSAriaII $_u$ sorter , And then, IF new FACSAriaII $_u$ 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 there is no record of using the computer within one month, you must re-authenticate before you can use it again. 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	Itaara	Charges fee (NT dollars / per hour)	
FACSCalilbur	Item	Within institution	other
Configuration:	Fully commissioned services	500	1000
488nm laser 633 nm laser	technician to accompany	500	1000
software: CellQusest	Use by yourself (Only for certified users)	60	Not open
Model	Item	Charges fee (NT dollars / per hour)	
BD LSR II	nem	Within institution	other
Configuration:	Fully commissioned services	500	1000
488 and 633nm laser 405 nm laser	technician to accompany	500	1000
software: FACSDiva	Use by yourself (Only for certified users)	60	Not open
Model	Charges fee (NT dollars / pontage		-
Thermo Attune NxT	nem	Within institution	other
Configuration:	Fully commissioned services	500	1000
488 and 633nm laser 405 and 561 nm laser	technician to accompany	500	1000
software: Attune NxT	Use by yourself (Only for certified users)	60	Not open

(b) sorting service fee

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

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 project funding, you will have to pay the bill at the General Services Department.

Sample preparation:

1.carrying by yourself:

- (1) $41 \sim 53$ um nylon mesh
- (2) FALCON #2052 12x75 mm plastic tube
- (3) pipets $(200 \sim 1000 \text{ul})$ & tips
- (4) Flash drive
- 2. Sample final concentration : $5x10^5 \sim 1x10^6$ cells/ml \circ 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.

(Note: It takes $1 \sim 2$ hours to clean the pipelines and warm-up before getting on the machine. To avoid wasting time, standard beads and laser power, please do not cancel the experiment temporarily)

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.

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 \circ

- 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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