

FEI Vitrobot

Site: Rm. A10, B2, Interdisciplinary Research Building of Science and Technology

Contact: Dr. Yuan-Chih Chang / Ms. Hui-Ju Huang

Service: Preliminary cryo-EM/cryo-TOMO studies, liposome & polymer, etc

Instructions

The cryo-grid preparation by Vitrobot is the most critical step to obtain a high-resolution cryoEM structure. However, every bio-sample has unique properties and is preserved in different buffer solutions, resulting in different optimal conditions for cryo-grid preparation. Moreover, many factors can influence the final outcome of cryo-grid preparation, so it is best that users who know the sample well prepare/test the cryo-grid preparation. In addition, due to the limited manpower, it is indeed difficult for ASCEM to provide customized cryo-grid service for every user. For Vitrobot applicants, ASCEM provides basic training and certification for users from Academia Sinica. A certified user can prepare or test their own cryo-grid (cryo-sample) with various experimental conditions.

Q1. In what condition can ASCEM assist to operate Vitrobot for users from Academia Sinica?

A1. If the need is occasional and sample is relatively simple (e.g. liposome); or when a project is urgent and the user cannot be certified in time, ASCEM can assist to prepare cryo-grid.

Q2. Alternatives for non-Academia Sinica Vitrobot applicants

A2. Some other cryoEM institutes in Taiwan, e.g. Institute of Physics of NTU, NCKU, NSRRC, and Genomics Research Center of AS, are also equipped with Vitrobot or equivalent sample preparation instruments for cryoEM structure determination. Non-AS users are recommended to do cryo-grid preparation there. Alternatively, non-AS user may collaborate with a certified member of an AS lab. If a direct use of ASCEM Vitrobot is still a necessary, the certified user may directly book online, the Vitrobot open time for non-AS user is 09:00-16:00; uncertified user may apply for staff operation.

Booking & Review DAAIS Online [Facility Reservation System](#)



(1) **Account Register:** Both User and Lab Supervisor/PI need to create an account first on the **DAAIS Online Facility Reservation System**.

(2) **Service Request:** User submits a “Service Request” on the **System** for the machine time of the next two months. When the capacity of an instrument is nearing full, booking by a lab should not exceed 2 days every week. The

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booking procedures need to be done before the machine time, an afterward make up application is not accepted.

- (3) **Authority Apply:** For Vitrobot, User needs to apply for the “**Authority of Usage**” before submitting a “**Service Request**”. Also, PI needs to approve the application online first, then ASCEM will receive it.
- (4) **Case Review:** Each “**Service Request**” will be auto-emailed by the **System** to the PI for an online approval first, then the “**Request**” will be forwarded to ASCEM for a further review and arrangement, or the case will be considered as “cancelled”.
- For viruses, even inactivated viruses, or other biohazardous samples, [the Requirements and Affidavit](#) should be attached and reviewed.

Online Booking Time Slots

- AS user 09:00 – 20:00, non-AS user 09:00-16:00 each session is 2 hours.
- Self-operated by certified users from Academia Sinica only (need to complete training courses first).
- All Vitrobot users need to sign for [the Agreement for the Regulations of Vitrobot](#), and then bring the original document to Vitrobot Manager/Staff (see Agreement No. 7) on site at the first time of operation.
- Before the first booking, please initiatively contact staff for guide of the booking system.
- **AS user User Training:** Submit a “**Service Request**” on the **System** for a “**ASCEM Training Course**”, then choose “**Vitrobot**”. 4.1. A training course is offered once in two weeks and is limited to 3 trainees each time. Please contact Facility Manager for details. Users need to re-do certification and re-sign the Agreement when not using Vitrobot for over three consecutive months.
- **Staff operate** User needs to contact and fully discuss with the facility managers before submitting a “**Service Request**” for **Vitrobot2** on the **System**. The available time slots are 14:00-16:00, Monday through Friday.

Academia Sinica Cryo-EM Center (ASCEM) 108.01.01起	
Facility/Equipment	
Apply	ASCEM Training Course
Apply	CEM 1, JEM1400 Cryo-EM
Apply	CEM 2, FEI Tecnai F20 TEM-1
Apply	CEM 3, Talos Arctica
Apply	CEM 4, Titan Krios
Apply	CryoEM software support 冷凍電顯軟體服務建置
Apply	FEI Vitrobot-2
Apply	FEI Vitrobot-3

Deadline for booking and cancellation

- Vitrobot 2: can be booked and canceled 24 hours in advance. Contact facility manager for cancellation.
- Vitrobot 3: can be booked and canceled 2 hours in advance. Contact facility manager for cancellation.
- User must be the applicant. 4 hours is the maximum for each booking (2 hours/session).
- Contact the manager within the specified period if any adjustment is needed. Once the manager clicks “approve” for the machine time on the **System**, cancellation for refund will not be accepted.
- Manager retains the right to adjust the machine time depending on the machine condition as well as to arrange time for routine maintenance of machines as needed (2 to 3 days every two weeks).

Notes for Sample Delivery

- **Sample property:** bio-solution. Note: Magnetic samples and samples that do not meet bio-safety standards are

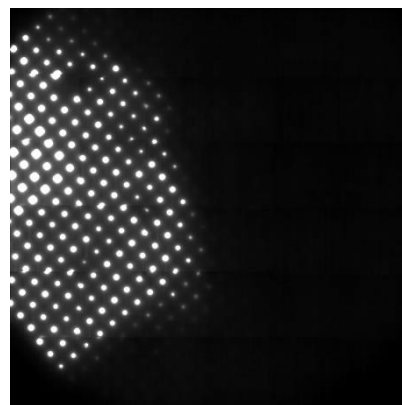
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unacceptable. For viruses, even inactivated viruses, or other biohazardous samples, [the Requirements and Affidavit](#) should be attached and reviewed.

- **Reminders for cryo-sample preparation:** It is recommended to use hydrophilic buffer that most suitable for sample, low-salt concentration, better not to add surfactant, and better no sucrose or glycerol. Moreover, the buffer viscosity will affect the thickness of the ice layer, the more viscous the buffer when making the sample, the ice layer will be too thick or be empty.
- The samples need to be delivered to the staff before the machine time. If the samples are not delivered on time, no make-up will be offered and the fee will still be charged according to the regulations.
- Users are recommended to be present during the machine time of experiment for discussing sample conditions.
- **Staff Operate**
 1. A maximum of 4 cryo-grid samples will be made by the staff each time. If the 4 cryo-grids are all failed (see images), the staff will re-produce one more time without charging the staff-operation fee for the reproduction, but the user still needs to pay for Vitrobot using and consumables.
 2. User needs to communicate with the staff about the sample state and the conditions for making cryo-grid samples, otherwise the common equipment settings will be adopted for the procedures.
 3. The conditions for preparing cryo-grid samples are the consensus between the staff and the user, user needs to understand that the ice thickness of the cryo-grid samples will vary depending on the sample state and also, there is no guarantee that the ice thickness will meet the experimental requirements.
 4. The pre-treatment of the cryo-grids is not covered by the staff, such as grid cleaning and various coating.
 5. Samples with biosafety concerns need to be approved first. Please submit your application via the Google form (including Vitrobot) and the Requirements and Affidavit should be attached.



Failed cryo-grid production



Not failed cryo-grid production

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Case Closure

The applicant must complete the **“Time and Charge Confirmation”** for closing each individual case on the **DAAIS Facility Reservation System**. The **System** will email a link to guide the next steps, the applicant and the Supervisor/PI can complete the procedures via the link or directly log in the **System** to **close** the **“Pending Cases”**. Then, ASCEM will compile all the **closed** cases monthly and email a **payment notice** to the Supervisor/PI.

Case ([New cases](#) [Disposed & Pending case](#) [Not-closed cases](#) , [Closed cases](#) , [Integrated query](#))

[Service Request - Disposed & Pending case](#)

No.	Date of application	Applicant	PI	G
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Charge & Pay

(Start from 2023.08.01, Currency: NTD)

Charge for Data Collection Details (Extra fee will be charged if the facility consumables are used.)	Regular Time (Mon - Fri 8:30-17:30)	Off Time (Mon - Sun)
FEI Vitrobot	600 / session, for academia users	
	2,400 / session, for industry users	
	1,200 / session, for staff operation	

- To promote the academic research and the application of cryoEM, and under the recommendations of DAAIS, all academia users, no matter it is AS or non-AS budgets being used to pay, share the same charge scheme and discount from August 1, 2023 in accordance with the announcement in the ASCEM User Committee Meeting held on July 18, 2023.
- Each case will be charged according to the record on the **DAAIS Facility Reservation System**. Course/Practice/Operation - NTD 600/session (for academic users), each session is two hours; consumables - copper grid NTD 390/piece.
- If the charge has not been paid for over than six months after receiving **the payment notice** due to user reasons, the booking authority of the whole lab members will be suspended temporarily until the charge is paid.

Pay by AS budgets - It will be charged via AS intramural budget accounts and will be charged on the 7th day after sending out the payment notice.

Pay by non-AS budgets - including MOST or NSTC projects. Please provide the payment notice to the accountant of your institute for assistance and pay the charge as soon as possible by one of the three methods below.

Method 1: 【User-DAAIS】 Directly go to DAAIS office and **pay cash or check** (payee: Academia Sinica). The receipt will be immediately issued on site.

Contact: Mr. Eric Wen-Chih Chiu, DAAIS, Rm. A102, Interdisciplinary Building of Science and Technology, Academia Sinica.

Provide information to DAAIS: facility name used, receipt title, service statements/lists.



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Method 2: 【User-DAAIS】 Send a check by registered mail (payee: Academia Sinica). The receipt will be issued by DAAIS after receiving the check.

Mailing address: Mr. Eric Wen-Chih Chiu, DAAIS, Rm. A102, Interdisciplinary Sci. & Tech. Building, No. 28, Ln. 70, Sec. 2, Academia Rd., Nankang, Taipei 11574, Taiwan (R.O.C.)

Enclosed information for DAAIS: facility name used, receipt title, service statements/lists.

Method 3: 【User-Facility】 Telegraphic or ATM transfer. The receipt will be issued by DAAIS after receiving the proof of the payment.

BENE NAME: ACADEMIA SINICA **BANK A/C:** 004056030013

BENEFICIARY ADDRESS: NO. 128, SEC. 2, ACADEMIA ROAD, NANGANG DIST., TAIPEI 115, TAIWAN, R.O.C.

BANK NAME: LAND BANK OF TAIWAN NAN KANG BRANCH (Swift Code: LBOTTWTP004)

BANK ADDRESS: NO. 364, NANKANG ROAD SEC.1, TAIPEI 115, TAIWAN, R.O.C.

Provide information to Facility (contact: Ms. Hsin-Hua Lin): proof of payment, receipt title, and number of receipts to be issued.