

July 11, 2024



Overview of Kirara service and its new results on material bioproduction

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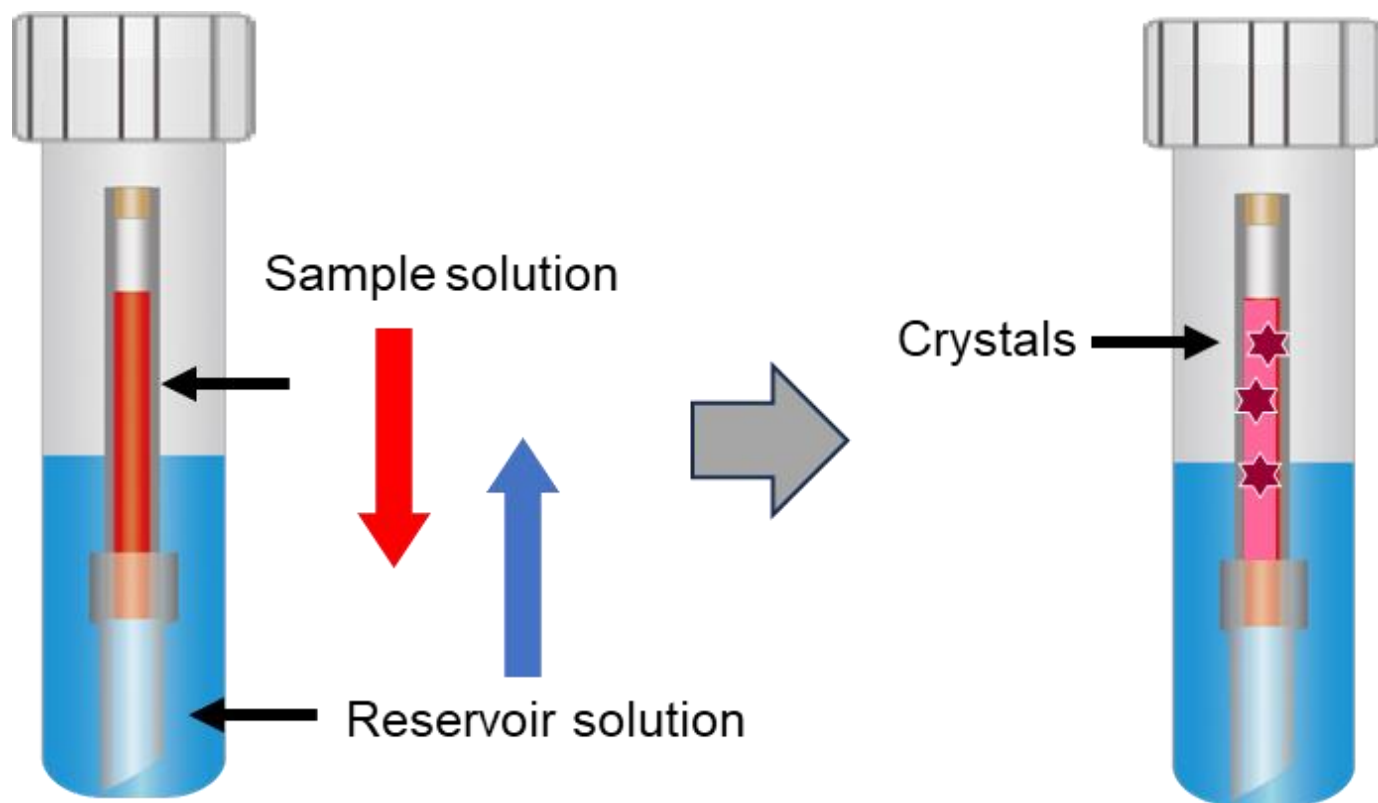
'Kirara' Service

- 'Kirara' service was launched in 2019.
- An initial target: high-quality protein crystal growth
 ⇒ challenging to other experimental field.
- A tiny incubator: ICE Cubes facility in the ISS Columbus module.
- **96 samples can be installed.**



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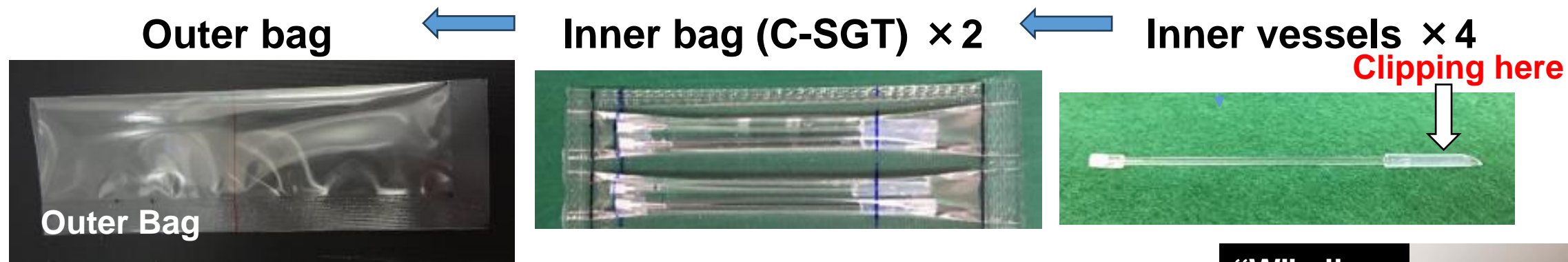
The principle of the counter diffusion



- **Glass capillary: A protein sample is loaded.**
- **Gel-Tube: A silicon-tube pre-loaded with agarose gel.**
- **The device is dipped in a reservoir solution.**
- **The reservoir solution diffuses in.**
- **The concentration increases and a protein crystal starts growing.**

Sample configuration for Kirara

- Double containment:



- Bags: non-gas-permeable soft plastic sheet, sealed by heat.
- Inner vessel: glass capillary with silicon tube
- Mixing two solutions:
 - One in the inner vessel and the other in the tube.



- Bags, vessels, experimental methods, and procedures can be customized.

Loading sample & starting reaction

- Loading sample at CS Lab. (2~4 weeks before launch)

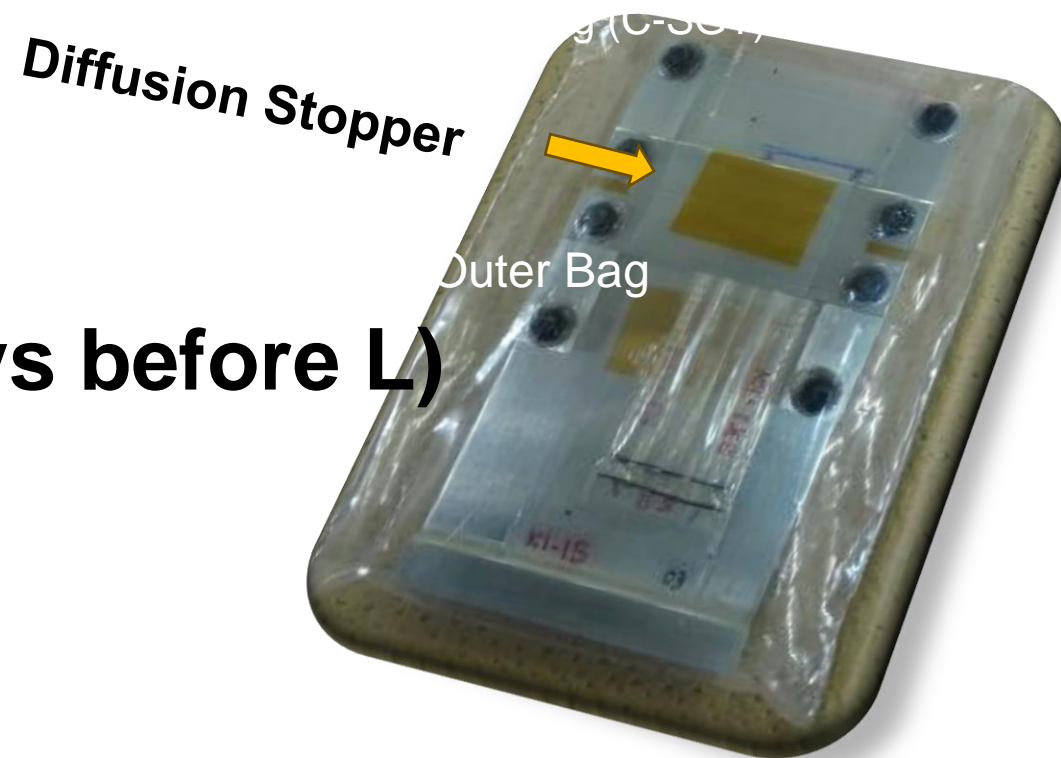
- **Suppress reaction**

- apply diffusion stopper: clip the bags
- store in 4 °C or in dry ice.

- **At Kennedy Space Center (~2 days before L)**

- Removing the stopper
- Heating to 20 °C

⇒ **Reaction Starts!**



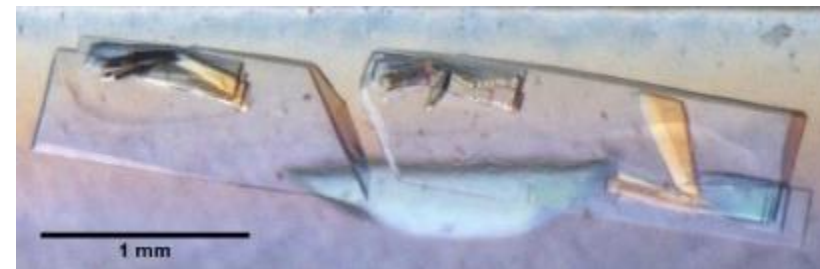
Time-Line of the service

- 4 months** **Application data sheet**
- 3 months** **Samples for the optimization (optional)**
- 3 weeks** **Sending samples (nominal)**
- 2 to 4 weeks** **Loading samples**
- 1 week** **Transporting samples**
- 2 days** **Install samples @ Kennedy Space Center(20 °C)**
- 0 day** **Launch to the ISS (20°C)**
- +1 month** **Recovery from ISS (20°C)**
- +1.5 months** **Delivery to users (20°C)**



Kirara launched samples (# of entry)

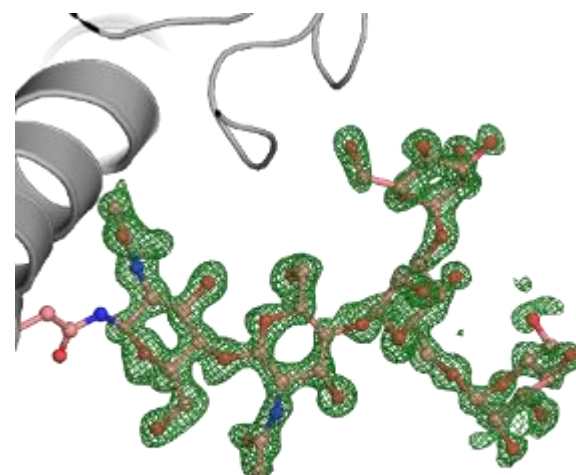
- **Protein crystallization**
 - For X-Ray diffraction (13)
 - For neutron diffraction (3)
 - For STEAM education (3)
- **Biomolecular Structure Formation (2)**
- **Colloid crystallization (3)**
- **Small molecular crystallization (4)**
- **Small molecular complex formation (1)**
- **Enzymatic cellulose synthesis (5)**
- **Bacterial cellulose production (2)**



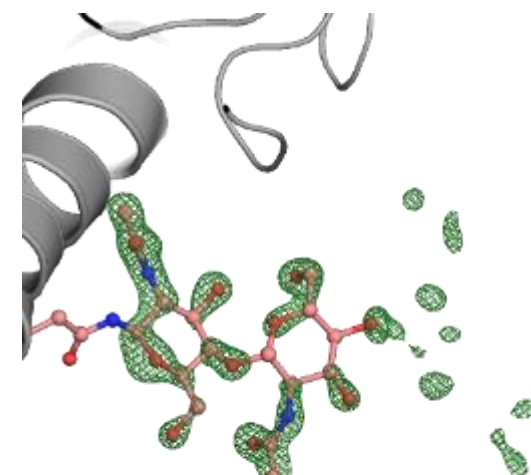
Benefits for the protein crystallization

- Cluster → Single crystal

	Space	Ground
Data Collection		
Space group	P21	P21
Unit cell dimensions		
a, b, c (Å)	50.26 127.49 63.04	50.55 127.94 63.68
α, β, γ (°)	90.00 91.44 90.00	90.00 91.15 90.00
Resolution (Å) ↓	46.74-1.06 (1.08-1.06)	63.97-1.67 (1.70-1.67)
Mosaicity (°) ↓	0.33-0.64	0.24-1.64
Completeness (%)	98.8 (94.6)	98.1 (95.8)
Mean I/ σ (I)	8.2 (1.6)	5.3 (3.3)
Rmeas	0.102 (0.984)	0.426 (2.210)
Refinement		
Rfree ↓	0.188	0.212
Twin fractions(%) ↓	N/A	7.5*
Precipitant solution	PEG 4000 / NaCl	PEG 4000 / NaCl



Space ©JAMSS



Ground ©JAMSS

Clear electron density map.
⇒ The atomic coordinates become more accurate.

Enzymatic cellulose synthesis

- CDP + cellobiose
(Capillary)
- α -G1P
(Reservoir)

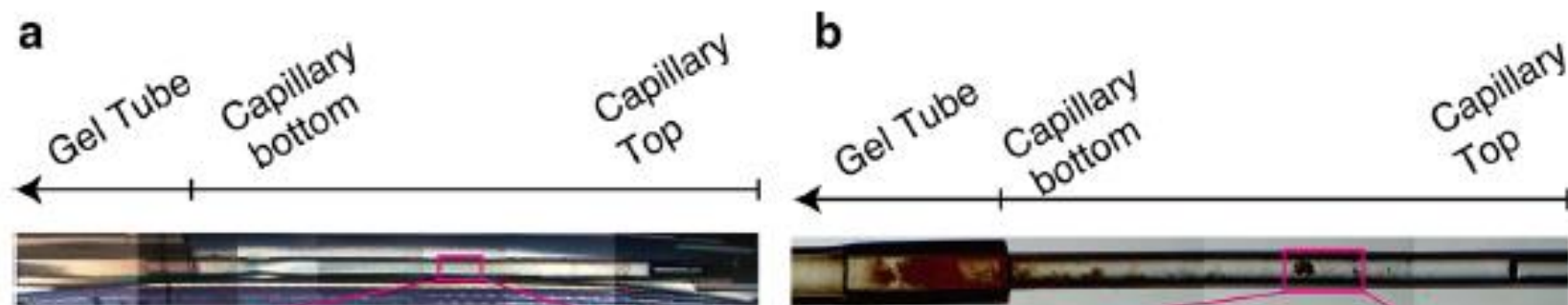


Cellulose

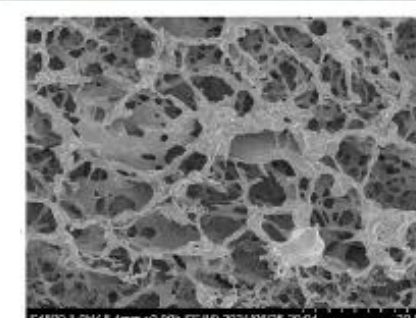
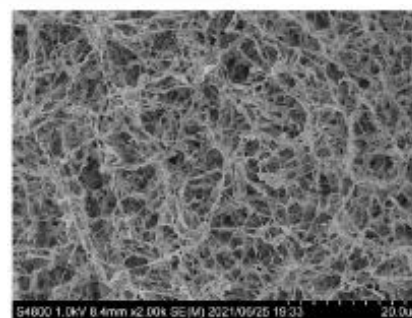
Uniform outlook.

Sparse network in SEM image.

- Launched in Kirara #1, 2019
- The enzymatic cellulose synthesis in space is the first in the world.



Optical Images



SEM Images

in space

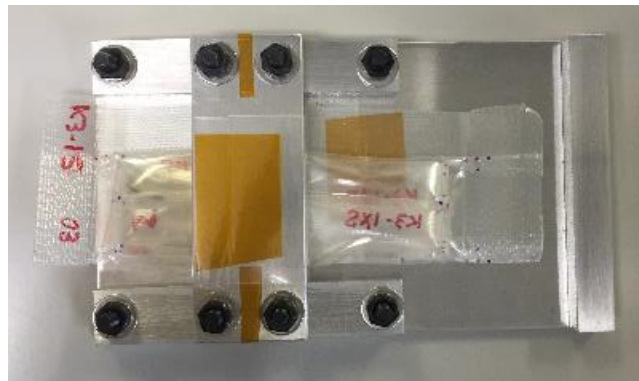
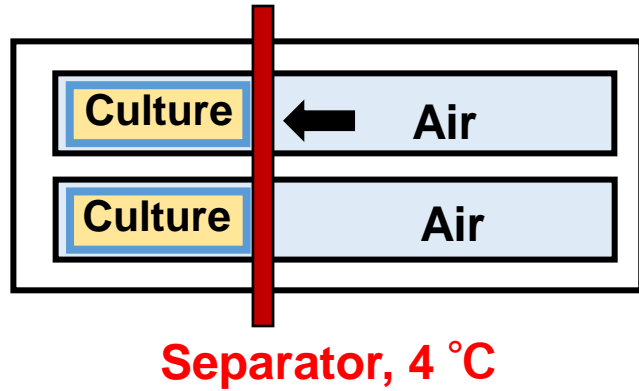
on ground

*T. Kuga, et.al., (2022) Cellulose,
<https://doi.org/10.1007/s10570-021-04399-0>*

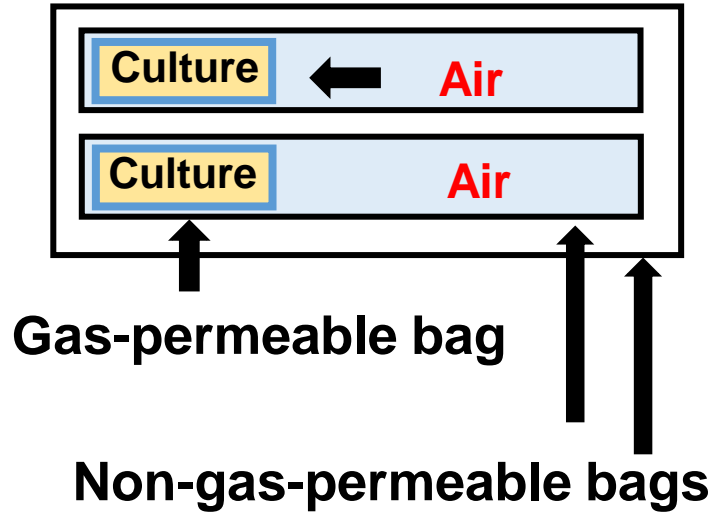


Bioproduction using Kirara: Type 1 (Kirara#4)

Sample preparation & transport to KSC



Cultivate at 20 °C on ISS



Bacterial cellulose



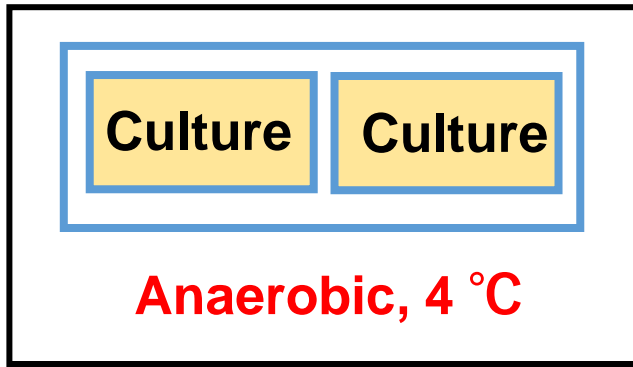
Biosafety level ≤ 1

**The first time in the world.
Cellulose production is possible in space.**

A. Tanaka, et.al., *Int. J. Microgravity Sci. Appl.* 41 (2024) 410302, <https://doi.org/10.15011/jasma.41.410302>

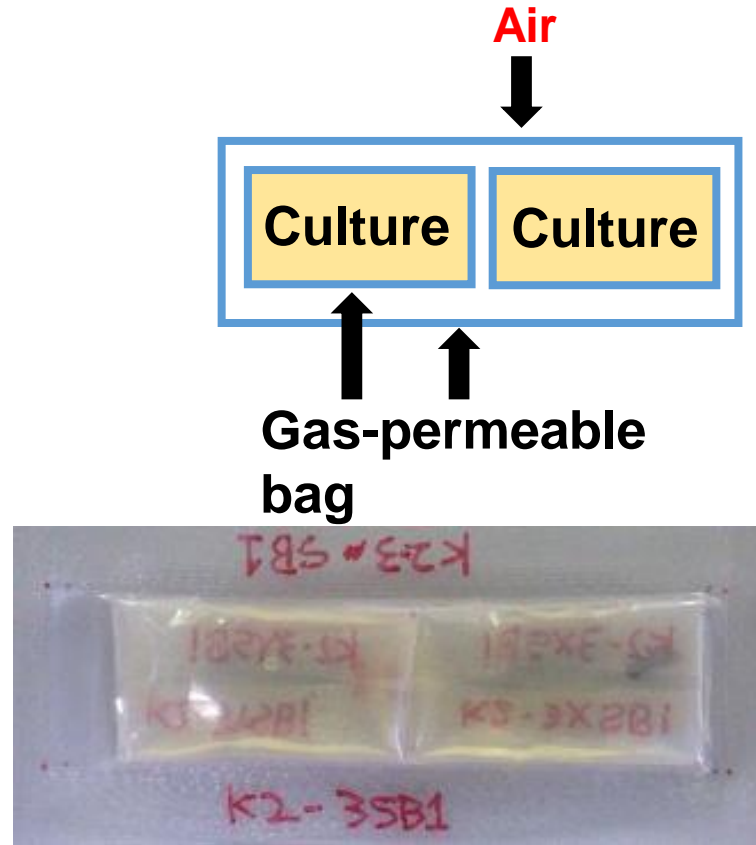
Bioproduction using Kirara: Type 2 (Kirara#5)

Sample preparation & transport to KSC



Non-gas-permeable bag with an oxygen absorber

Cultivate at 20 °C on ISS



Bacterial cellulose



Biosafety level ≤ 1

Large-scale production is possible in space.



A. Tanaka, et.al., *Int. J. Microgravity Sci. Appl.* 41 (2024) 410302, <https://doi.org/10.15011/jasma.41.410302>

Contact JAMSS

- Containers, experimental methods, and procedures can be customized according to user's request.

As earlier is preferable to contact us!!

- JAMSS Kirara:

https://www.jamss.co.jp/en/space_utilization/kirara/

- JAMSS Kirara Data Sheets etc:

https://www.jamss.co.jp/en/space_utilization/kirara/

- JAMSS inquiry form

https://www.jamss.co.jp/en/contact_service/

