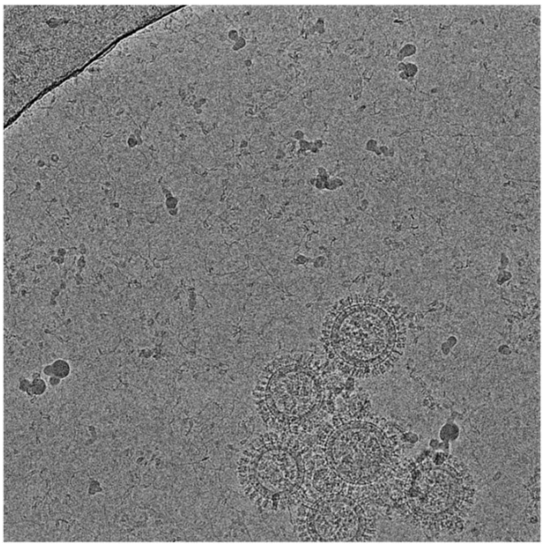
Quantum Wave Microscopy Unit
Professor Tsumoru Shintake



Cryo-EM image of Influenza A Virus

Abstract

Electron Microscopy Group

A cryo-electron microscope equipped with a tilted electron beam and a ring slit was utilized for the structural analysis of microcrystals of proteins and organic-inorganic hybrid materials. This research is being conducted in collaboration with the Namba Laboratory at Osaka University. Perovskite thin film crystals, a material that is attracting attention in photovoltaic power generation, were provided by the Qi Unit at OIST. We are engaged in the process of refining the analytical and crystallization methodologies, with the objective of presenting the findings in a public forum.

Collaborative research agreement (CRA) with Siemens Healthineers and Acrorad, for studies on high-Z compound semiconductor crystals for medical imaging applications, using electron holography and other specialized electron microscopy techniques.

Applied Microscopy Research Project aims to develop applied microscopy research using our in-house developed techniques for electron microscopy and using enveloped viruses as samples. Previously, we have shown that the ethanol sensitivity of influenza viruses increases with temperature and demonstrated (using both in vitro and in vivo (mouse) systems) that influenza virus inactivation can occur at ethanol concentrations as low as 20% (v/v) at human body temperature (Tamai et al., 2023). We are now trying to understand the molecular mechanism of this inactivation process, using

cryo-electron microscopy and our microvolume ethanol processing device as core techniques. Various other biophysical characterization methods will also be used in this project. In FY2023, our research focused on 1) establishing purification system for enveloped viruses and their surface proteins and 2) optimizing experimental conditions for cryo-EM and other biophysical measurements for characterizing the purified samples. Work is also underway to establish a technique for observing the mouse lung using X-ray micro-CT technology, with the aim of further developing our 2023 study.

Wave Energy Converter (WEC) Project

The project has been carrying out R&Ds to harness electricity from the moving water in the ocean waves in Seragaki Seashore since 2018. A generator, fitted with a propulsion unit supplied by Yamaha Motor (hereinafter referred to as "Y-generator") had been installed in May 2022 with a 102cmφFalcon three-blades turbine and after the support structure was modified, the generator had been reinstalled in March 2023.

1. Staff

- Dr. Cathal Cassidy, Staff Scientist
- Dr. Masao Yamashita, Staff Scientist
- Dr. Ryo Kanno, Staff Scientist
- Dr. Takeshi Mise, Staff Scientist
- Seita Taba, Research Unit Technician
- Jun Fujita, Research Unit Technician
- Shuji Misumi, Research Unit Technician
- Ayumi Maegawa, Research Unit Technician
- Hideki Takebe, Specialist

2. Collaborations

2.1 Imaging magnetic monopoles in spin ice

- Type of Collaboration: Joint Research
- Researchers :
    - Dr. Ankur Dhar, SLAC, Stanford University
    - Dr. Ludovic Jaubert, CNRS Bordeaux
    - Prof. Nic Shannon
    - Dr. Cathal Cassidy

2.2 Application of electron holography and related analysis techniques to investigate electrostatic potential distributions at metal-CdTe interfaces

- Type of Collaboration: Collaborative Research Agreement (CRA)

- Researchers :
    - Dr. Cathal Cassidy
    - Tamami Unten, Acrorad
    - Dr. Miguel Labayen de Inza, Siemens Healthineers

2.3 Protein Crystal Imaging with Precession TEM

- Type of collaboration : Joint Research
- Researchers :
    - Prof. Tsumoru Shintake
    - Dr. Masao Yamashita
    - Dr. Ryo Kanno
    - Ayumi Maegawa
    - Seita Taba
    - Prof. Keiichi Namba, Osaka University
    - Dr. Fumiaki Makino, Osaka University
    - Prof. Yabin Qi
    - Dr. Katsuya Ono

2.4 Applied Microscopy Research on enveloped virus

- Type of collaboration: Joint research with Ishikawa Unit
- Researchers :
    - Prof. Tsumoru Shintake
    - Prof. Hiroki Ishikawa
    - Dr. Miho Tamai
    - Dr. Masao Yamashita
    - Dr. Ryo Kanno
    - Dr. Takeshi Mise
    - Seita Taba
    - Jun Fujita
    - Prof. Takemasa Sakaguchi, Hiroshima University (sample provided)

2.5 OIST Wave Energy Converter (WEC) Project

- Type of collaboration: Contracted Research
- Researchers :
    - Prof. Tsumoru Shintake
    - Shuji Misumi
    - Jun Fujita
    - Hideki Takebe
    - Koma Ariga, YAMAHA MOTOR CO.,LTD.

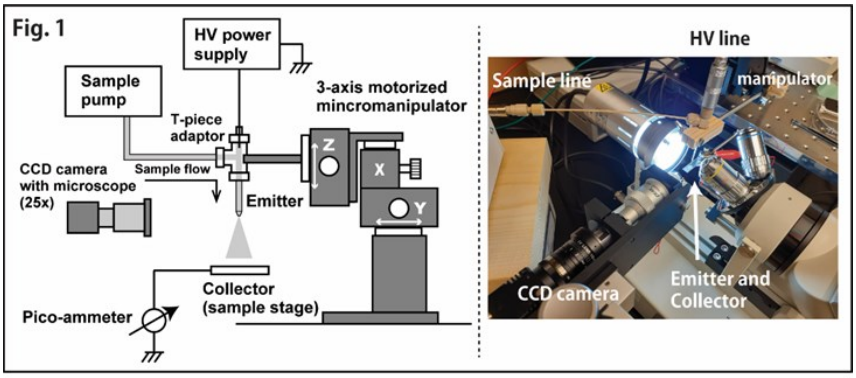
- Atsushi Murashima, YAMAHA MOTOR CO.,LTD.
- Shintaro Futagawa, YAMAHA MOTOR CO.,LTD.

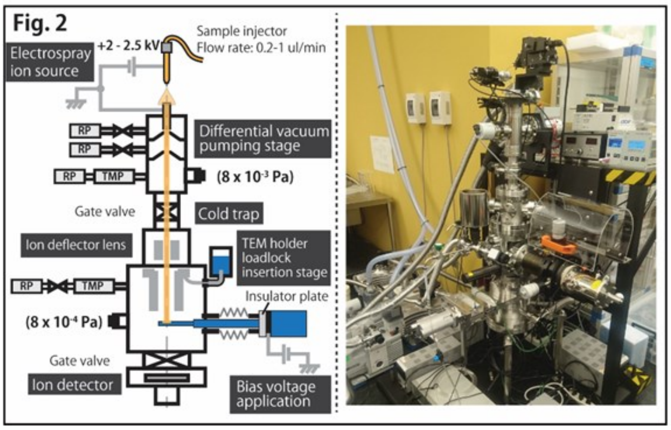
3. Activities and Findings

3.1 Electron Microscopy Group

3.1.a Development of electrospray sample deposition system (Dr. Masao Yamashita)

Our unit has been working on developing a new biospecimen preparation system for EM based on electrospraying (ES) technology. We have long been focusing on such application of ES since it can potentially be used to generate desolvated "naked" biomolecules, which is logically the most ideal form for specimens to be imaged by TEM/STEM at low voltages. However, this kind of application of ES is not common as it is only applicable to those samples that can meet very specific and stand with rather harsh conditions. For this reason, ES has rarely been applied to large biological macromolecules and, as a result, how well biomolecular structures may be preserved after the solvent removal by ES is still not well understood. To address this problem and explore the potential of ES for EM specimen preparation, we have built two experimental devices (Fig. 1 and 2). The device in Fig. 1 is a custom made electrosprayer currently being optimized for spraying various biomolecular solutions including those of protein complexes, DNAs, and viruses. Fig. 2 is an electrospray molecular deposition device designed to be an optimization platform for making TEM specimens on a one-atom-thick graphene sample carrier film. Although challenging, we believe that the realization of this technique should greatly contribute to a field of single (bio)molecular imaging. Further development is underway.





3.1.b Electron holography studies of CdTe crystals and devices (Dr. Cathal Cassidy)

We renewed our CRA (Collaborative Research Agreement) with Acrorad and Siemens Healthineers. This year we continued our work on holography, 4D-STEM, and also initiated some high-resolution EELS studies of interfacial alloy materials. A major focus of this year was travel and presentation at conferences – Forchheim, Germany (INAM/IKTS), Edmonton, Canada (Japan-Canada Microscopy Symposium), and Melbourne, Australia (IUCr Congress).

3.1.c Observing magnetic monopoles in spin ice (Dr. Ankur Dhar, Dr. Cathal Cassidy)

Based upon the thesis work of Dr. Ankur Dhar, in collaboration with Profs. Shannon & Jaubert, we prepared and submitted a manuscript on electron holography studies of magnetic monopoles. Our manuscript is available to view on arXiv, and publication activities are ongoing: https://arxiv.org/abs/2112.01362

3.1.d Protein Crystal Imaging with Procession TEM (Prof. Tsumoru Shintake, Ayumi Maegawa et al.)

Based on a new type of electron microscope theory developed by Professor Shintake in 2021, a unique ring slit was incorporated into the JEOL CRYO ARM300 at the Namba Laboratory of Osaka University in 2022. Imaging using this system eliminates the spherical aberration of the focusing lens in the electron microscope, making it possible to capture molecules inside crystals that have been difficult to observe directly in high resolution. In particular, it is thought that this system will be highly effective in directly visualizing proteins, which are biopolymers, and analyzing crystals of organic-inorganic hybrid materials and drug crystals that are easily affected by electron beam damage.

In fiscal year 2023, as a demonstration of this microscope and analysis system, crystallization of multiple samples, imaging with an electron microscope, and analysis by image processing were performed. The first image of a lysozyme crystal was taken with a tilted beam from one direction without beam precession, but by changing the tilt direction of the beam and imaging from three or four directions, the loss of diffraction information was suppressed. Various crystallization and shooting methods were considered to adjust the orientation of the crystal. For example, this includes changing the shape of the crystal and adjusting the angle using electron beam diffraction images. Some results of the samples analyzed are shown below.
(Figure 1: Lysozyme space group P43212, Figure 2: Lysozyme space group P21212)

In fiscal year 2024, we plan to develop a unique image processing program and improve the crystallization method, including a method for synthesizing images with changed beam directions, and to make external presentations and submit papers.

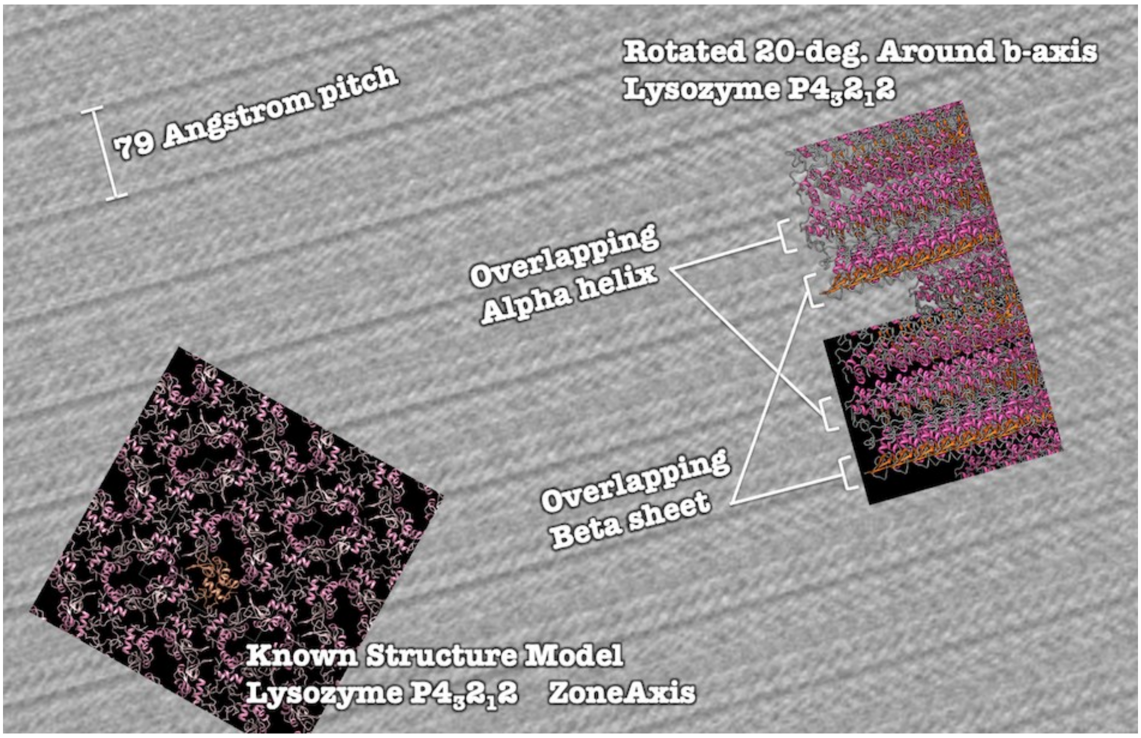


Figure 1: Lysozyme space group $P4_32_12$ crystal structure
A model diagram generated from a known crystal structure (PDB ID: 4LYZ) was superimposed on the

electron microscope image, from which noise had been removed using an FFT filter.

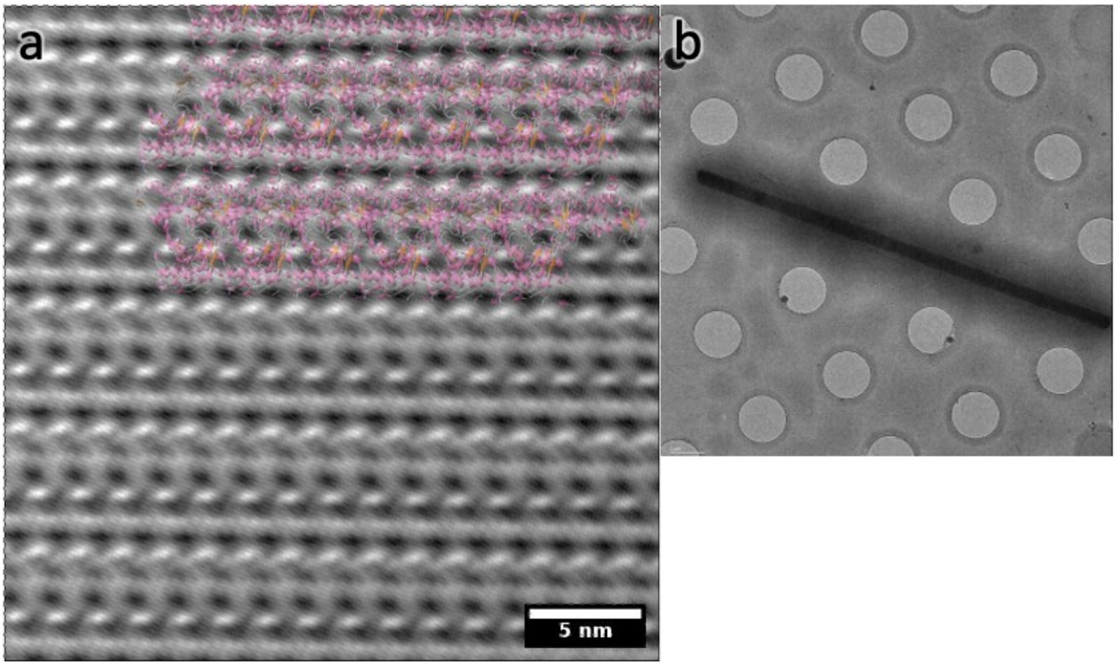


Figure 2: Lysozyme space group $P2_12_12$ crystal structure

a) The microscope image following FFT filtering exhibits the same structural characteristics as a known crystal (PDB ID: 5OCV) belonging to a different space group than that depicted in Fig. 1.
b) It has been reported that lysozyme has the capacity to form predominantly tetragonal and orthorhombic crystals. Fig2a image, derived from needle-like crystals, appears consistent with the orthorhombic crystal structure. Low magnification electron microscope image of crystals obtained under the same conditions.

3.1.e Applied Microscopy Research Project

a) Progress in purified virus sample preparation and their imaging

An experimental system for mass amplification of enveloped viruses using eggs has been established in our laboratory in collaboration with Prof Sakaguchi of Hiroshima University. This allowed us to use highly concentrated and purified viral samples for cryo-EM, ensuring that even at high imaging magnification there is enough virus in the field of view to make a statistically valid assessment of viral structure in the viral population (Fig. 1). Building on this improvement, a detailed and systematic study of how ethanol-induced structural changes in the virus proceed at different temperatures will be carried out in our future research. In the development of the virus amplification system mentioned above, we introduced a new rapid virus quantification method (Fig. 2), which was confirmed to work very well.

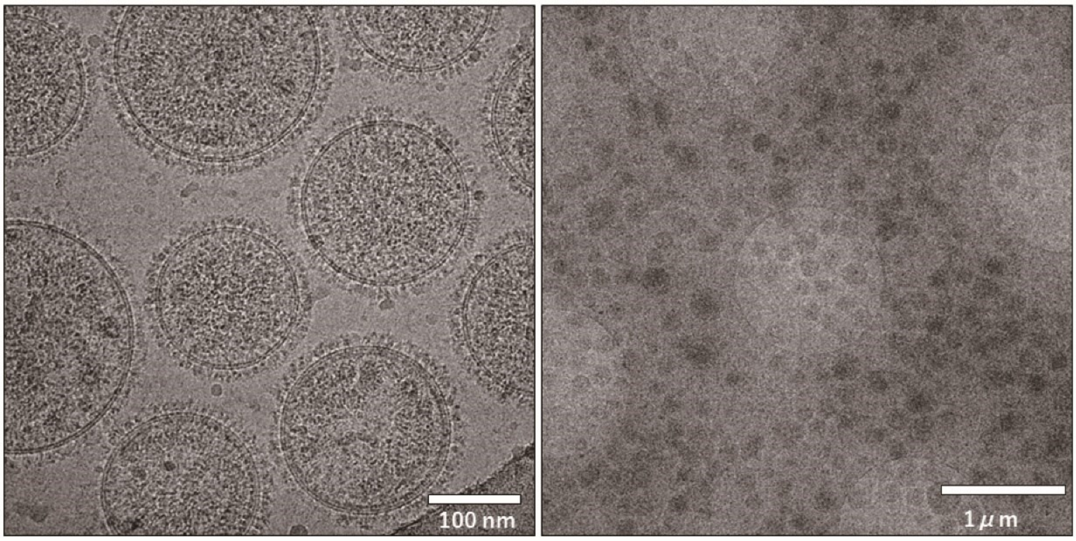


Figure 1 : The images of Newcastle Disease Virus, acquired by 300 kV cryo-TEM.

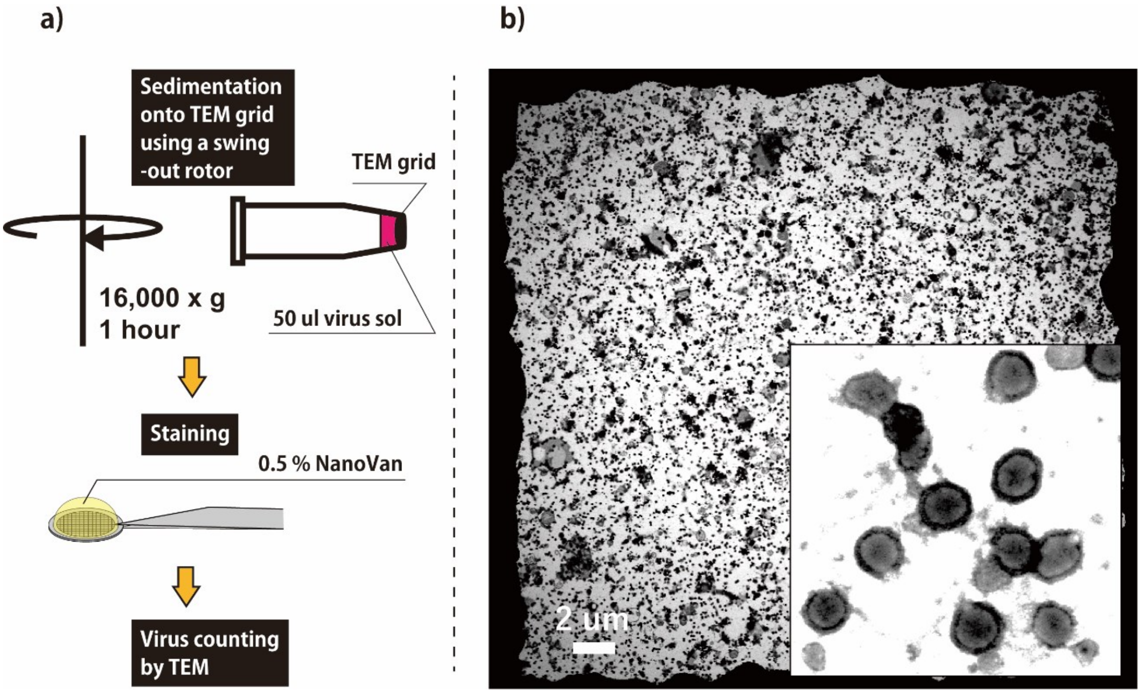


Figure 2: Virus counting method used in this study. a) Experimental design. b) Virus deposition result. One grid square of 400-mesh TEM grid is shown. Majority of the black dots represent virus particles. (inset) Higher magnification view of this TEM grid.

b) Progress in the development of microvolume ethanol processor

As mentioned above, the microvolume ethanol processor is one of the key technologies in this project. Based on the vapor diffusion approach, this device allows several microliters of highly concentrated virus samples to be treated with ethanol without mixing with a solution of high concentration of ethanol. The importance of this technique is that it effectively avoids both sample dilution and the introduction of uncontrolled damage (artefacts) to the viral sample due to exposure to high concentrations of ethanol during mixing. The use of this technique for cryo-EM sample preparation is therefore essential for achieving the correct assessment of the effects of ethanol on the structure of the virus. We have continued to improve the unit since the last annual report and the current version (ver. 4) has improved temperature control functionality and can operate in a temperature range of 10°C to 50°C (Fig. 3).

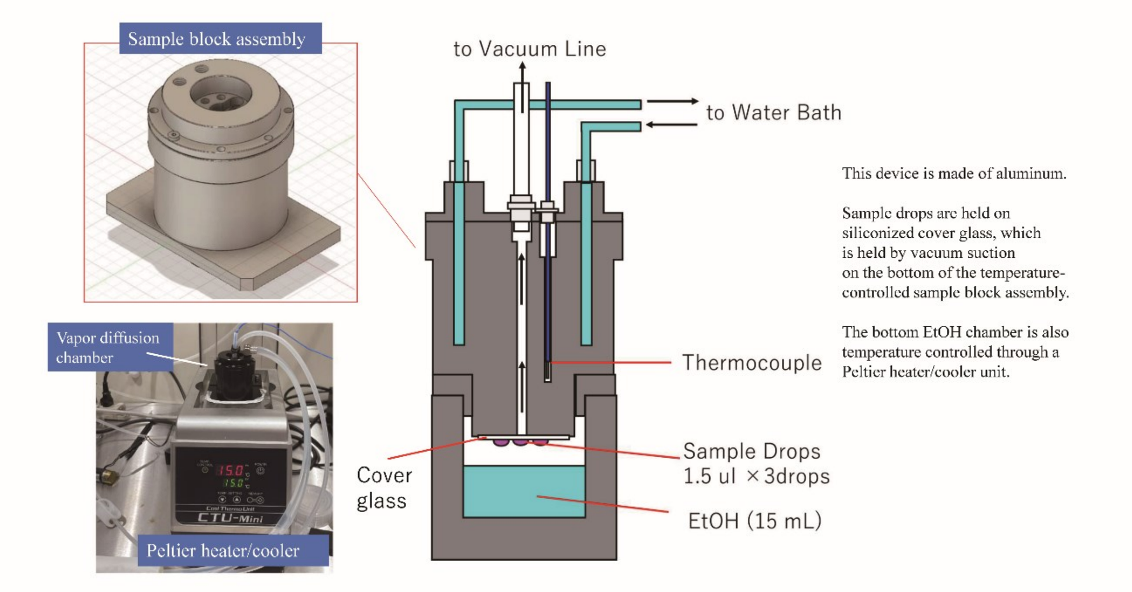


Figure 3: Microvolume ethanol processor (version 4)

c) Progress in viral surface protein purification and their analysis

Our previous experiments suggest that low concentrations of ethanol may affect the viral surface proteins and alter its conformation (Fig. 4). To investigate this further, we are attempting to establish a purification system for the envelope viruses' surface proteins. So far, we have been successful with influenza haemagglutinin, yielding approximately 0.8 mg/ml of purified protein in the peak fraction of the final gel filtration step (Fig. 5). Purification of other surface proteins are currently underway. To investigate how the function and structure of these proteins are affected by ethanol, we are pursuing a variety of characterization approaches, including cryo single particle analysis, Biacore and fluorescence measurements. These works are still at a preliminary stage. Some highlights from the current work are presented below (Figs 6, 7).

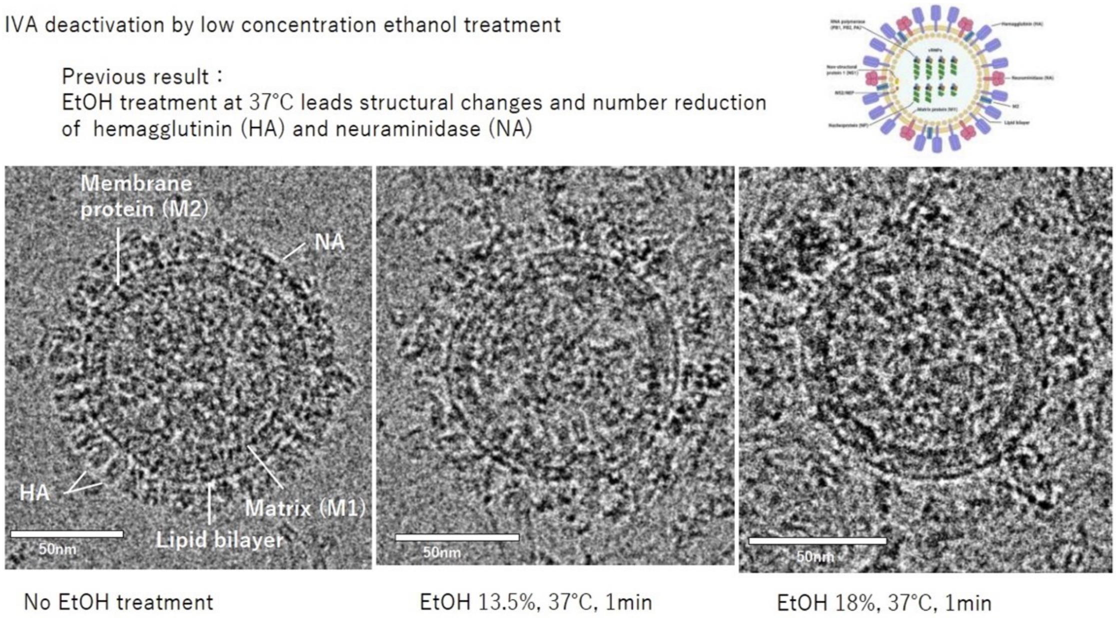


Figure 4: Conformational change on surface proteins on influenza-A virus after 1 minute exposure to EtOH.

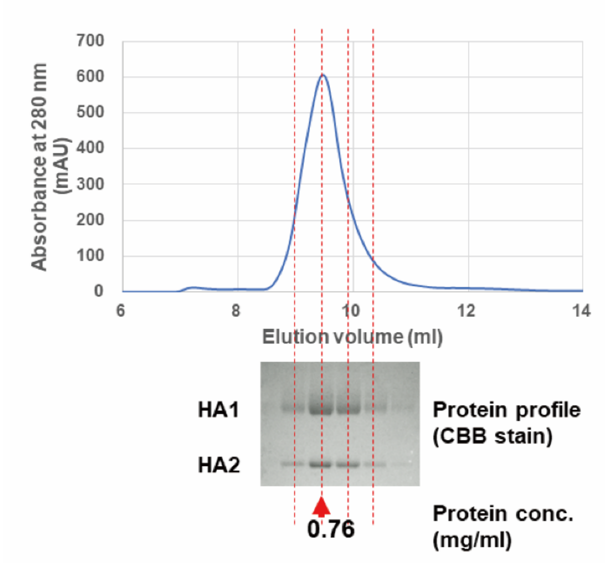


Figure 5: Purification of hemagglutinin protein from influenza A. Result of the final gel filtration step is shown together with the protein gel image. Protein concentration was determined by the BCA method.

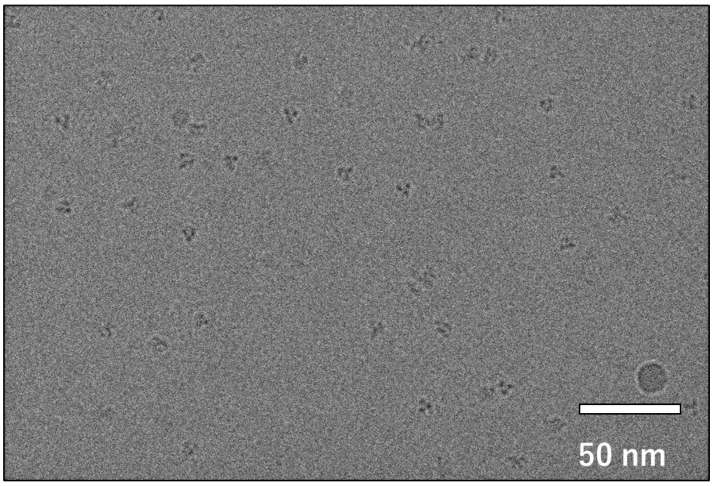


Figure 6: Cryo-TEM analysis of purified Hemagglutinin from influenza-A virus. Dispersed particle images of Hemagglutinin, acquired by 300kV Cryo-TEM (Titan Krios).

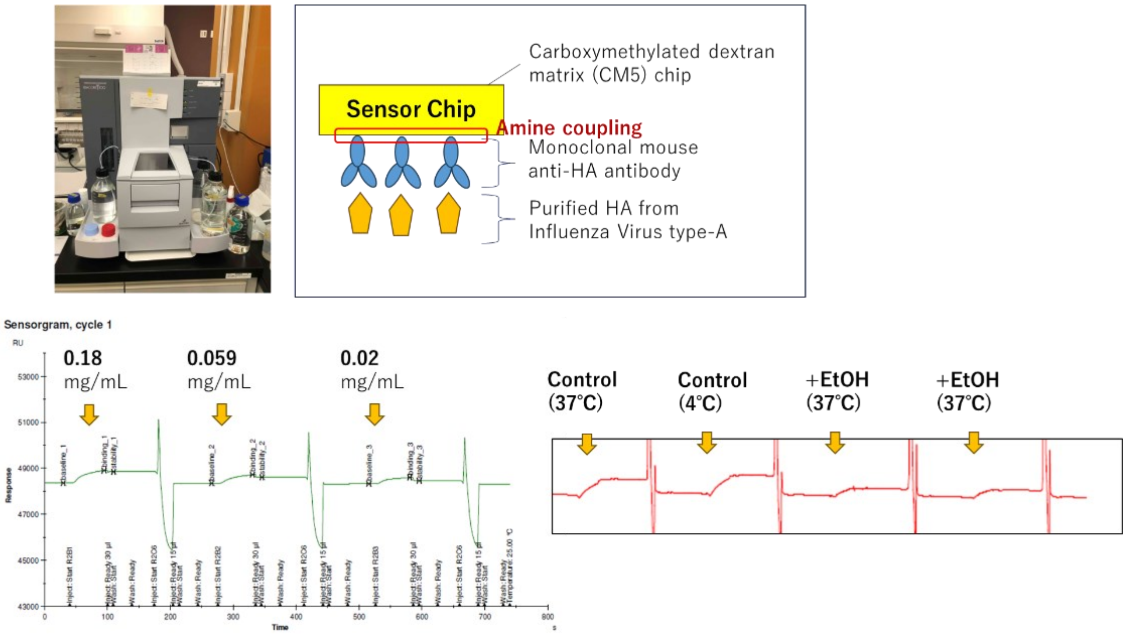


Figure 7: Design of surface plasmon resonance SPR experiment for influenza HA (top) and preliminary result (bottom). The immobilized antibody captures the purified Hemagglutinin, whereas the signal gets significantly low after the ethanol treatment for 7 minutes at 37°C.

d) Progress in X-ray micro-CT experiment

We aim to use micro-CT to identify mice infected with the influenza A virus (IAV) lung condition and the effect of ethanol vapor inhalation. In our previous work, we observed the lungs of uninfected mice, as demonstrated in the following images (A-F), and established a micro-CT method.

This year, our precise micro-CT method was applied to the lungs of a mouse infected with the IAV (G-I) and the control mouse(J-L). Table M presents the weight changes of the mice used in the experiment. The mouse infected with the IAV was immediately used for micro-CT as it lost about 10 % weight on day 3. Our micro-CT analysis detected shadows in addition to weight loss. As a next step, we will investigate whether these white arrows are the ground-glass opacity (GGO) or not. The lungs of humans infected with IAV show ground-glass opacity (GGO). If the lung of an infected mouse showed the ground-glass opacity (GGO) was confirmed, this similarity underscores the relevance of our findings to human health.

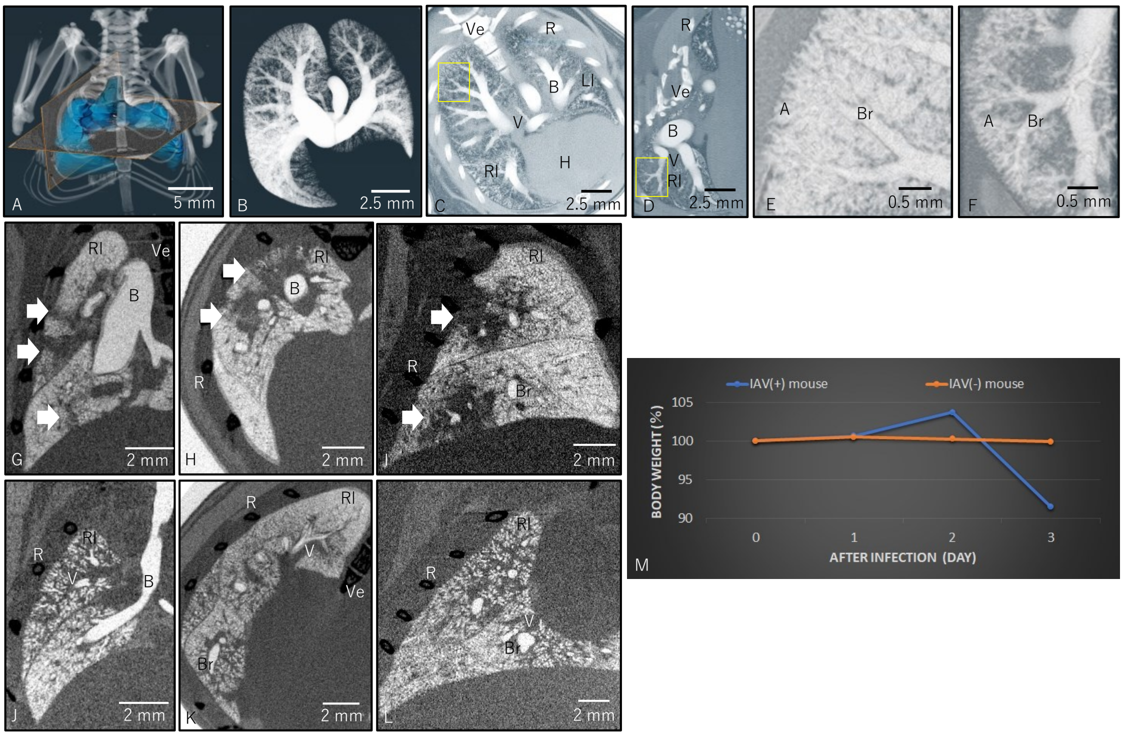


Figure 8: The micro-CT image of lungs from an uninfected mouse (A-F), an infected mouse (G-I), and a control mouse (J-L). A: Model drawing of lungs (blue) and mouse skeleton. B: The bronchi and alveoli of a mouse. C: Section image (0.7 mm thick) of Fig. A in the transverse plane (xy-plane). D: Section image (0.7 mm thick) of Fig. A in the longitudinal plane (yz-plane). E: Enlarged view of the yellow box in Fig. C. F: Enlarged view of the yellow box in Fig. D. G-I: The micro-CT results of IVA-infected mice. G, H, and I are the images of the sections xz-plane, xy-plane, and yz-plane, respectively. As a next step, we will investigate whether these shadow white arrow showings are ground-glass opacity (GGO) or not. J-L: The micro-CT results of the control mouse lung of IVA uninfected mouse.

J, K, and L are the images of the sections xz-plane, xy-plane, and yz-plane, respectively. M: The table shows the mouse weight used in the above G-L experiment. H: Heart, Rl: Right lung, Ll: Left lung, V: Vessel, R: Rib, B: Bronchi, Ve: Vertebra, Br: Bronchiole, A: Alveolar sac.

3.2 Wave Energy Converter (WEC) Project

3.2.a Experiment with Y-Generator Test Equipment

A generator, fitted with a propulsion unit supplied by Yamaha Motor (hereinafter referred to as “Y-generator”) had been installed in May 2022 with a 102cmφFalcon three-blades turbine and it’s support structure was modified in March 2023 (Fig.1)

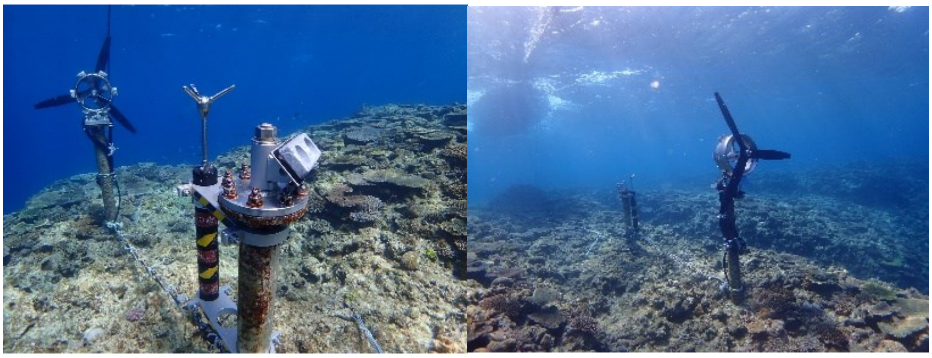


Figure 1: Simplified support structure

As no significant improvement in measurement of wave power was seen, the height position was changed in June 2023 by extending the prop by 50cm length on the testing machine side and by 100 cm on the water flow speed measurement device support (Fig.2).

Figure 2: Changed height position of props

In early August, slow-moving massive typhoon No.6 hit Okinawa directly (Fig.3) and it had tremendous impact. The power generator and the backup data logger (output voltage observation) had stopped due to power failure in OIST MSS facility.

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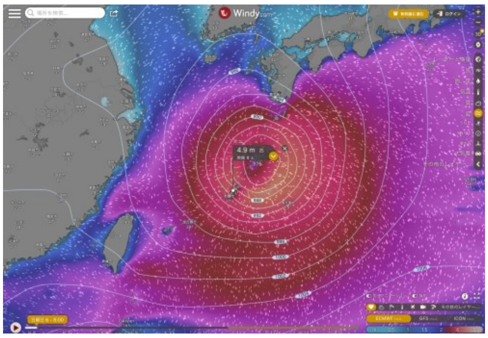


Figure 3: Adapted from “Windy”.

Huge damage on most devices and equipment by the typhoon were discovered (Fig.4).
The testing machine prop repeatedly hit by strong wave swell was toppled down, the Y-generator fell off its pedestal and the cable were broken in several places. One turbine blade was also damaged. Another prop somehow held its own, but the excavation hole had been expanded and it was not stable. The sensor rod of the measurement device attached to the prop was heavily bent and there were some broken sections in undersea cable.

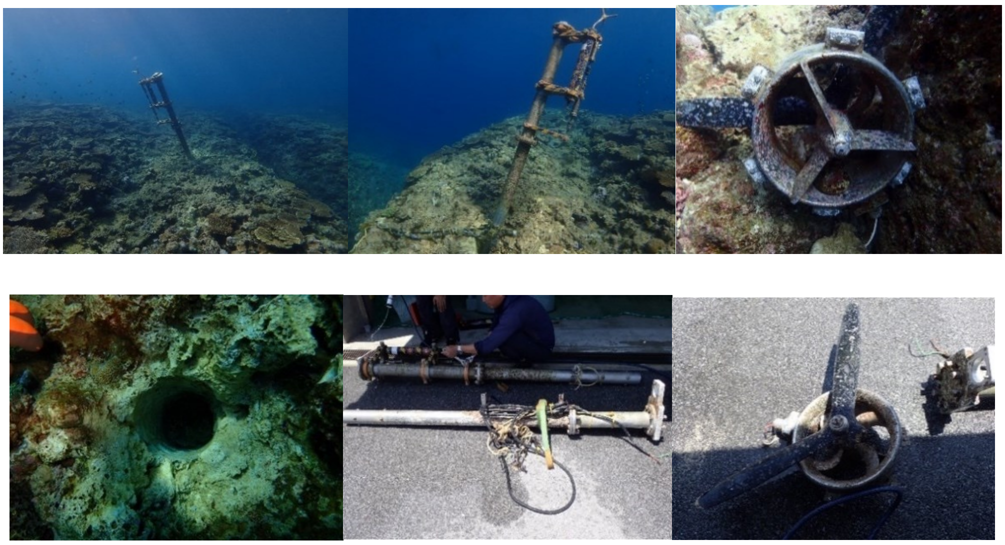


Figure 4: Damaged devices

Necessary repair works for the Y-Generator, sensor, turbine and cable had been provided and some were restored but no specific design and method for better structure could be found, therefore, Y-Generator was decided not to be reinstalled given this circumstance.

3.2.b Summary of Experiment with Y-Generator Test Equipment

The following table summarized the results of the measurements with theoretical energy efficiencies. The kinetic energy of the water flow was proportional to turbine diameter, the specific gravity of the seawater, and the cube of the flow velocity (equation at the lower right of this table) and could be calculated from the efficiency η of the turbine and the generator.

P1 was the theoretical kinetic energy of the water flow multiplied by the efficiency η (product of the efficiency ηt of the turbine and the efficiency ηg of the generator) and P2 was the measured power. The efficiency ηt of each turbine was not measured this time but it was estimated from the results of Falcon turbine towing tests at Col. 2 in the table.

The efficiency of the generator, ηg, was assumed to be about 0.9 based on previous experimental results. The first two Cols. on the left were obtained by the previous OIST-Half-Scale Generator, and the from Col.3 to 7 were by the Y-Generator. Col.4 was Y-generator + Falcon turbine (78 cm diameter), but the resistance of the Hexagon (hexagonal) support frame was estimated to be about 5%.

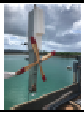
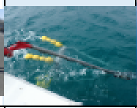
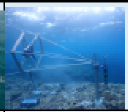
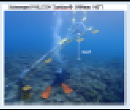
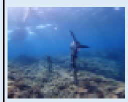
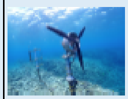
Col.5 was installed on the seafloor, but the flow velocity was not commensurate with the wave height. Col.6 was raised in height from the seafloor (near the water surface), but the power was still weak because the flexibility of the support frame rather prevented it from fully absorbing the force of the waves. In 2023, the structure changed from the Hexagon-Frame to a fixed seabed structure, then the power generation efficiency of Col.6 and 7 were improved.

The measured wave power was in any case, lower than the theoretical value.

One of possible reasons was non-uniform flow around turbine and in contrast with sharp flow velocity in breaking wave zone, the flow velocity spread over a long period of time, power was not transmitted to the power converter.

Estimated power generation P1 and measured power P2 of Seragaki-WEC

(HSG = Half Scale Generator, Y-Generator = Generator made by YAMAHA Motor Machinery)

	1	2	3	4	5	6	7
	HSG+Wooden Turbine (110cmϕ)	HSG+Falcon Turbine (78cm Φ)	Y-Generator +Falcon78cmΦ +Hexagon frame	Y-Generator +Falcon78cmΦ +Hexagon frame	Y-Generator +Falcon110 cmΦ +Floating Hexagon frame	Y-Generator+ Falcon 110cmΦ	Y-Generator+ Falcon 110cmΦ
Photo							
ρ (kg/m3)	1025	1025	1025	1025	1025	1025	1025
r (m)	0.55	0.36	0.36	0.36	0.51	0.51	0.51
S(m3)	0.950	0.407	0.407	0.407	0.817	0.817	0.817
1／2	0.5	0.5	0.5	0.5	0.5	0.5	0.5
v (m/s)	1.4	1.5	2	3	4	2	2
η	0.15	0.3	0.24	0.24	0.24	0.3	0.3
P1(W) calc	200	211	400	1351	6429	1005	1005
date	29-Jul-21	29-Oct-21	16-Feb-22	16-Apr-22	30-Nov-22	13-Mar-23	1-Jun-23
Experimental Method	Towing Tests	Towing Tests	Hexagon, Towing Tests	Hexagon, Power Conditioner (Peak)	Hexagon, Power Conditioner (Peak)	Power Conditioner (Peak)	Power Conditioner (Peak)
Installation	Towing	Towing	Towing	2M under the sea	2~3M under the sea	0.5M under the sea	1M under the sea
P 2 (W) Peak	80	200	360	38	175	125	175
Ratio (P2/P1)	0.40	0.95	0.90	0.03	0.03	0.12	0.17
*P3(W) Average				5.0	5.0	6.8	7.5
Comment	Wooden turbine 4 blades, shape not optimized?	Falcon-Turbine 3 blades. Optimized shape.	Uniform current velocity for towing test. Is there a Hexagon-Frame and efficiency decrease?	Current velocity is estimated from Hs in Naha. Hexagon-Frame is fixed	Current velocity is estimated from Hs in Naha. Close to the sea surface, the current velocity was large, but the power was small because it was not fixed.	Current velocity is measured by Vector. The undertow was large.	Extend the trestle by 50 cm, 1 m from the bottom of the sea.

Falcon nominal 78cmΦ --> Actual 72cmΦ.

Falcon nominal 110cmΦ --> Actual 102cmΦ

$$P = \frac{1}{2}\rho S v^3$$

η is the assumed efficiency (product of turbine ηt and generator ηg). ηt includes the effect of the Hexagon-Frame.

Estimated power generation P1 (equation on the right) and measured P2 (peak value) of the WEC estimated from the flow velocity.

P2 was set to P(Conditioner)x2.5 in the case of high cable loss.

*P3(W) Average is the estimated average value from 1 to 20 minutes assuming a wave height value of 2 m.

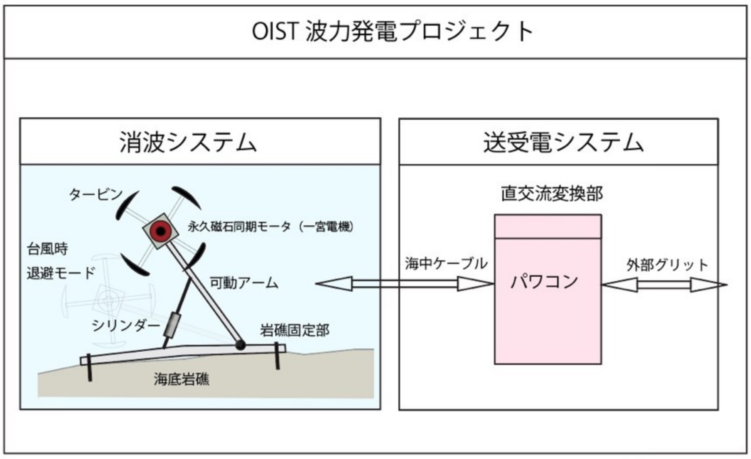
In conclusion, it seemed the current system was not feasible to obtain the expected power generation efficiently with following reasons:

- Seragaki's topography makes it difficult to obtain wave swells with a period of 8 seconds or more, which is necessary for power generation.

-There were limitations in converting the propulsion unit, originally developed for boat maneuvering system, into wave power generator with little or no modification.

3.2.c Darius turbine

Considering the results of past experiments, a new generator with Darius turbine has been devised. Appointed Mitsubishi Heavy Industries Systems and Ichinomiya Denki have produced a turbine and a motor of scale-down prototypes respectively and with their dedicated cooperation, performance tests are planned in 2024. In the future, based on the results of the performance test, the development of a generator with even higher efficiency will be pursued.



3.3. Laser Interferometric Flow Cytometry (Shuji Misumi)

Conventional flow cytometry is a device that irradiates laser light onto particles and analyzes them from the scattered light, making it suitable for the analysis of bio-cells.

The particle size that can be measured with this device is limited to 300 nm or larger.

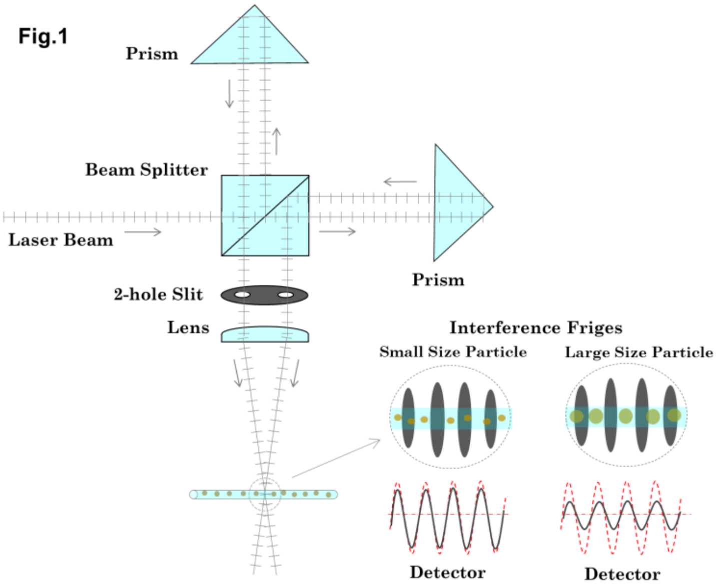
This invention is characterized by its ability to analyze particle sizes as small as 300 nm or less by using laser interference.

Figure 1 is a conceptual diagram of the invention.

By splitting the laser beam and crossing the two laser beams, interference occurs, and particles are analyzed within the interference fringes. The particle size to be measured can be changed by adjusting the angle at which the laser beams cross.

We will build this device and continue our research so that we can initially measure large particle sizes and then progressively measure smaller particle sizes.

Fig.1



4. Publications

4.1 Journals

1. Tani K, **Kanno R**, Harada A, Kobayashi Y, Minamino A, Takenaka S, Nakamura N, Ji XC, Purba ER, Hall M, Yu LJ, Madigan MT, Mizoguchi A, Iwasaki K, Humbel BM, Kimura Y, Wang-Otomo ZY, "*High-resolution structure and biochemical properties of the LH1-RC photocomplex from the model purple sulfur bacterium, Allochromatium vinosum*", February 2024 Commun Biol. 12;7(1):176.

2. S. Yuan, L. Dai, Y. Sun, F. Auras, Y.-H. Zhou, R.-Z. An, Y. Liu, C. Ding, **C. Cassidy**, X. Tang, S.-C. Dong, H.-B. Kang, K. Chen, X. Liu, Z.-F. Ye, Y. Zhao, C. Adachi, L.-S. Liao, N. Greenham, Y. Qi, S. Stranks, L.-S. Cui, R. H Friend, "*Efficient blue electroluminescence from reduced-dimensional perovskites*", January 2024 Nature Photonics. https://doi.org/10.1038/s41566-024-01382-6

3. M.T. Schreiber, **C. Cassidy**, "*Quantification of Gas-based Charge Compensation by Off-Axis Electron Holography in Open-Cell Environmental TEM*", July 2023 (Minneapolis). Microscopy & Microanalysis 29 1575-1576.

4. C. Ding, M. Niu, **C. Cassidy**, H.B. Kang, L.K. Ono, H. Wang, G. Tong, C. Zhang, Y. Liu, J. Zhang, S. Mariotti, T. Wu, Y. Qi, "*Local Built-In Field at the Sub-nanometric Heterointerface*

Mediates Cascade Electrochemical Conversion of Lithium–sulfur Batteries", May 2023, Small 19, 2301755.
https://doi.org/10.1002/smll.202301755

5. H.Adaniya, M. Cheung, M. Yamashita, S. Taba, C. Cassidy, T. Shintake, "Low energy scanning transmission electron microscoy applied to ice-embedded biological macromolecules " June 2022, Microscopy 72 – selected to Editor's Choice in 2023
https://doi.org/10.1093/jmicro/dfac056

4.2 Books and other one-time publications

Nothing to report

4.3 Oral and Poster Presentations

1. **Tsumoru Shintake**, *"Inspired Chain: From X-ray Crystallography to Electron Microscopy and EUV lithography"*, OIST-Kyudai Joint Symposium Series 1: Bio-Inspired Wonders and Energy Innovations, 29 February 2024 (OIST)

2. **Ayumi Maegawa**, "*Revolutionizing Molecular Imaging with Advanced Cryo-EM Technology"*, BioJapan 2023, 11-13 October 2023 (Yokohama)

3. **Ryo Kanno**, *"Cryo-EM study without single particle analysis"*, Data Analysis Hands-on Seminar, Cryo-EM Course (INGEM), Tohoku University, 04-06 October 2023 (Sendai)

4. **Cathal Cassidy**, A. Dhar, T. Unten, N. Kishi, B.C. Wolz, M. Labayen de Inza, T. Shintake *"Nanobeam electron diffraction & holography studies of functional metal/semiconductor junctions"* International Union of Crystallography Congress (IUCr), Session: Coherent Scattering with Electrons and X-rays, August 2023 (Melbourne).

5. **Cathal Cassidy**, M.T. Schreiber, *"Gas-based charge compensation measured by off-axis electron holography in environmental TEM",* Japan Society Microscopy-Microscopy Society Canada Joint Symposium, June 2023 (Edmonton)

6. **Cathal Cassidy**, *"CdTe research activities at OIST",* Joint Workshop with Fraunhofer IKTS and Siemens Healthcare, May 2023 (Forchheim)

5. Meetings and Events

Nothing to report

6. Other

6.1 Visitors

- 17 August : Mr. Satoshi Kawakami, Director, Industry-University Collaboration Office, Ministry of Economy, Trade and Industry
- 11 September : Delegation from Okinawa Electric Power
- 13 October : Deputy Mayer of Naha city

6.2 Media

Nothing to report