

FY2022 Annual Report

Molecular Cryo-Electron Microscopy Unit

Professor Matthias Wolf



Abstract

FY2022 was in the middle of the ongoing COVID-19 pandemic. Our projects were determined by establishing an ELISA for quantitative COVID-19 antibody detection, and the testing of 7,500 patients. Furthermore, the team came together in an interdisciplinary collaboration with other units at OIST and Tottori University to develop our own COVID-19 vaccine. We also started successful research about the structure of bacteriophages using cryo-EM.

1. Staff

- Dr. Matthias Wolf, Professor
- Dr. Hideyuki Matsunami, Staff Scientist
- Dr. Vladimir Meshcheryakov, Staff Scientist
- Dr. Takahide Kono, Staff Scientist
- Dr. Melissa Matthews, Postdoctoral Scholar
- Dr. Chloe Minnai, Postdoctoral Scholar

- Dr. Nadishka Jayawardena, Postdoctoral Scholar
- Dr. Rafael Ayala Hernandez, Postdoctoral Scholar
- Dr. Higor Alves Iha, Research Unit Technician
- Mr. Makoto Tokoro Schreiber, Graduate Student
- Mr. Keon Young Kim, Graduate Student
- Ms. Ting-Hua Chen, Graduate Student
- Ms. Rika Yoshizawa, Research Unit Administrator

2. Collaborations

2.1 COVID Vaccine antigen development and effect of antigen clustering

- Description: This project started during the COVID-19 pandemic. Wolf Unit expressed and purified spike protein and apoferritin. We made several antigens suitable for vaccination studies, including Spike, RBD, ferritin-spike, ferritin-RBD, liposome-spike and spike-rosettes. Antigenicity was evaluated after mice vaccination (in collaboration with Ishikawa Unit) using ELISA and ELISpot. Hamsters were challenged after vaccination to measure efficacy (collaboration with Tottori University). Antigen engineering was performed with input from Laurino Unit.
- Type of collaboration: Joint research
- Researchers:
 - Dr. Melissa M. Matthews, Dr. Tae Gyun Kim, Keon Young Kim, Dr. Higor Iha, Dr. Vladimir Meshcheryakov, Wolf Unit, OIST
 - Professor Hiroki Ishikawa, Dr. Miho Tamai, Dr. Daiki Sasaki, Immune Signaling Unit, OIST, Prof. Paola Laurino, OIST
 - Dr. Fumiko Obata, Prof. Jun Fujii, Prof. Satoshi Shibata, Noriko Shibata, Tottori University
 - Dr. Ofey Hsieh, Academia Sinica, Taipei, Taiwan, Wolf Lab at Institute of Biological Chemistry

2.2 3D Structure of Bacteriophages

- Description: Structure determination of several key bacteriophages by cryo-EM. This work was funded by MAFF and RSF with a bilateral grant. A main interest lies

on adopting bacteriophages to marine pathogens such as *Aeromonas* for biocontrol.

- Type of collaboration: Joint research
- Researchers:
 - Prof. Olga S. Sokolova, LMU Moskow University
 - Prof. Andrey V. Letarov, LMU Moskow University
 - Dr. Andrey Moiseenko, LMU Moskow University
 - Dr. Rafael Ayala, OIST Wolf Unit
 - Biological Chemistry

2.3 SARS-CoV-2 IgG seroprevalence in the Okinawa Islands

- Description: We determined the SARS-CoV2 anti-S antibody titers from ~6,500 samples provided by the Okinawan Prefectural Government (OPG), using ELISA. Prof. Mizumoto et al at OPG and Kyoto University evaluated and summarized our results statistically.
- Type of collaboration: Joint research
- Researchers:
 - Prof. Mizumoto et al, Kyoto University

3. Activities and Findings

3.1 COVID-19 Vaccine antigen development and effect of antigen clustering

With the ongoing pandemic, the team at Wolf Unit came together to develop our own vaccine candidate in collaboration with Ishikawa Unit and Laurino Unit. Because competitor were quicker to publish the idea of spike protein attached to apoferritin, we focused on investigating the effect of scaffolding itself. Several designs were explored, expressed, purified, and tested in animals. The two best candidates were tested in a challenge with real virus using golden Syrian hamsters in the BSL-3 facility at Tottori University and proved efficacious.

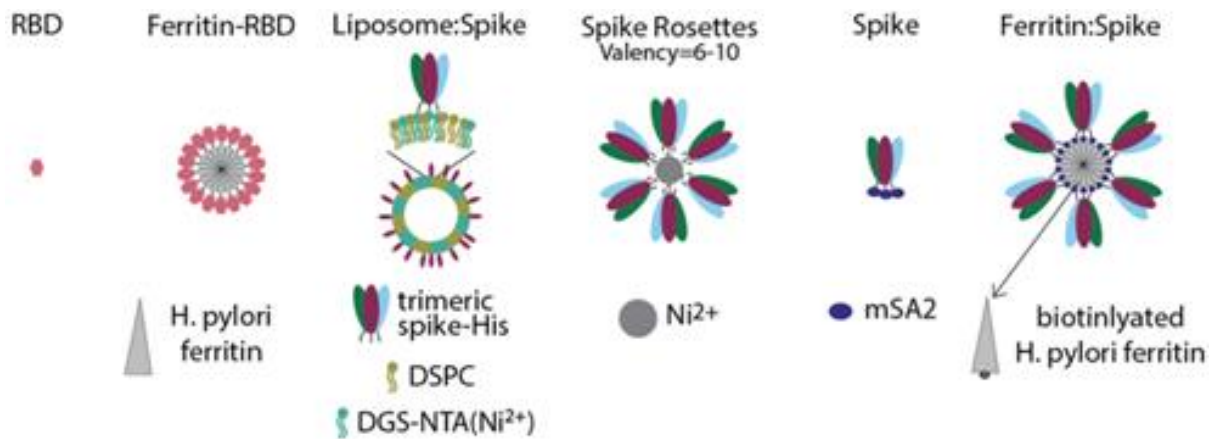


Figure 1: Six types of antigens were investigated with and without clustering or adjuvant.

3.2 Structure of bacteriophages

In collaboration with a group at Moscow University funded by a bilateral grant between MEXT (Japan) and the RSF (Russia), we determined the structures of two bacteriophages – a siphophage and a myophage – by cryo-EM. The phages were purified by the Moscow group.

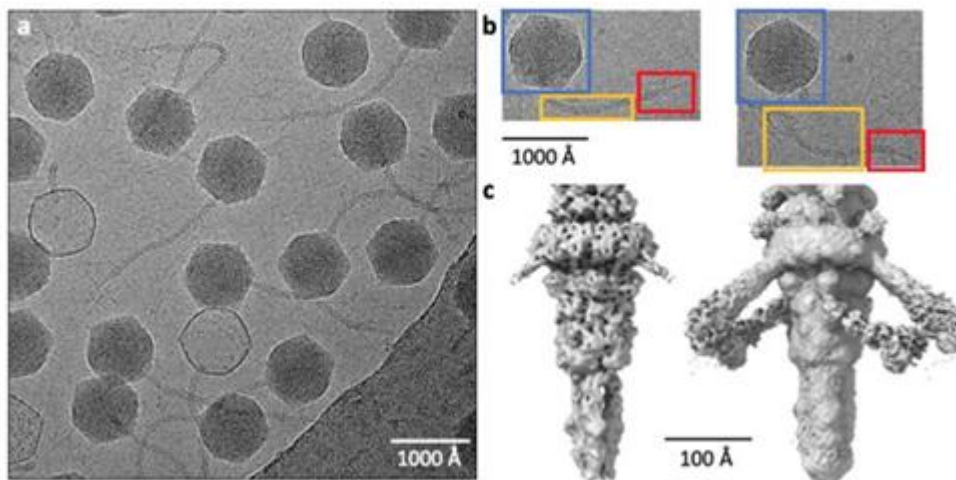


Figure 2: (left) DT57C phages in amorphous ice. (right) Structure of the tail tip complex.

4. Publications

4.1 Journals

1. Efghijk, S., Uxyaabc, E., Defgh, K., Cdefg, HI. Article Title Here as It Appears in the Publication. *Publication Title in Italics*, doi:doi: 10.1177/1059712310397633 (2011).

4.2 Books and other one-time publications

Nothing to report

4.3 Oral and Poster Presentations

1. 3D Structure of the Tail Complex of E. coli Bacteriophage DT57C
R Ayala, E Kulikov, A Golomidova, A Moiseenko, M Wolf, AV Letarov, ... M Wolf
Microscopy and Microanalysis 28 (S1), 1156-1158, 2022
2. Cryo-EM SPA for Structural Understanding of A-to-I RNA Editing: Human Adenosine Deaminase Acting on RNA 2 (ADAR2) Complexed with dsRNA
M Matthews, A Thuy-Boun, P Beal, A Fisher, M Wolf
Acta Crystallographica Section A: Foundations and Advances 78, a314-a314, 2022
3. Crystallization and X-ray crystallographic analysis of ribose 5-phosphate isomerase B
TH Kwon, K Min, M Wolf, C Ban, TG Kim
BioDesign 10 (2), 23-28 ,2022
4. Assembly mechanism of the pleomorphic immature poxvirus scaffold
J Hyun, H Matsunami, TG Kim, M Wolf
Nature Communications 13 (1), 1704 6, 2022
5. SARS-CoV-2 IgG seroprevalence in the Okinawa Main Island and remote islands in Okinawa, Japan, 2020-2021
K Mizumoto, Y Shimakawa, Y Aizawa, C Butcher, N Chibana, M Collins, ...M Wolf
medRxiv, 2022.03. 02.22271759, 2022
6. Author Correction: Cryo-EM structure of the Ebola virus nucleoprotein–RNA complex at 3.6 Å resolution (vol 563, pg 137, 2018)
Y Sugita, H Matsunami, Y Kawaoka, T Noda, M Wolf
NATURE 601 (7893), E11-E11, 2022
7. Growth of size-matched nanoalloys – a comparison of AuAg and PtPd
E. y. El koraychy, D. Nelli, C. Roncaglia, C. Minnai and R. Ferrando
8. Growth of out-of-equilibrium PtPd nanoalloys
C. Minnai
9. Neuromorphic behaviour in discontinuous metal films
S. K. Bose, J. B. Mallinson, E. Galli, S. K. Acharya, C. Minnai, P. J. Bones, et al.

5. Intellectual Property Rights and Other Specific Achievements

Patent

A convex-concave toroidal coil charged-particle lens using the magnetic vector potential

PCT/IB2022/059206, 2022/09/27

6. Meetings and Events

6.1 A Capsid with a twist: Assembly mechanism of the pleomorphic immature poxvirus scaffold

- Date: Nov 9, 2022
- Venue: ICCB IOCB Taipei
- Speaker: Prof. Matthias Wolf

6.2 A Capsid with a twist: Assembly mechanism of the pleomorphic immature poxvirus scaffold

- Date: November 2-4, 2022
- Venue: OIST - Kyoto University Joint Workshop, OIST,
- Prof. Matthias Wolf

6.3 Molecular insights into a T4-like coliphage using WALC, a novel method for iterative segmented cryo-EM reconstruction

- Date: August 16, 2022
- Venue: IBC, Academia Sinica, Taipei, Taiwan
- Prof. Matthias Wolf

6.4 A Capsid with a twist: Assembly mechanism of the pleomorphic immature poxvirus scaffold

- Date: July 7, 2022
- Venue: ISDD & KSSB Joint Symposium, POSTECH, Pohang, July 7, 2022
- Prof. Matthias Wolf

6.5 Temporal Dynamics of Charge Buildup in Cryo-Electron Microscopy

- Date: May 10, 2022
- Venue: PICO 2022, Kasteel Vaalsbroek, The Netherlands
- Prof. Matthias Wolf

6.6 A Capsid with a twist: Assembly mechanism of the pleomorphic immature poxvirus scaffold

- Date: March 23, 2022
- Venue: Taiwan Cryo-EM Symposium 2022, Academia Sinica IBC
- Prof. Matthias Wolf

6.7 A Capsid with a twist: Assembly mechanism of the pleomorphic immature poxvirus scaffold

- Date: Feb. 28, 2022
- Venue: OIST AMED BINDS Workshop, Feb. 28, 2022
- Prof. Matthias Wolf

7. Other

Nothing to report.