

Science and Technology Group Annual Report FY2021

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1 Introduction

Eukaryotic cilia/flagella are highly conserved organelles that have various functions, including motility and cell signaling. They have an internal cytoskeletal structure called an axoneme, which contains nine doublet microtubules surrounding two central microtubules and their associated structures.

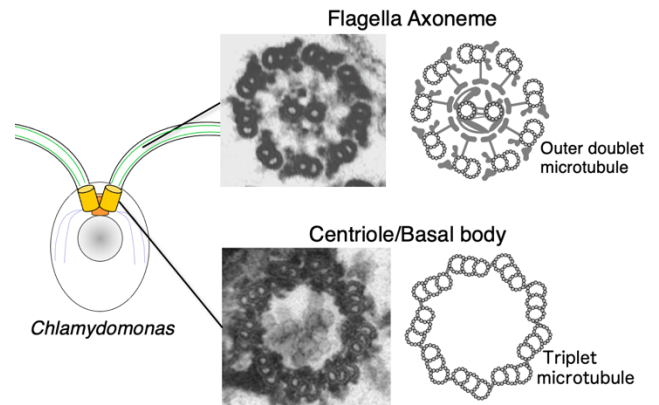
This characteristic “9+2” pattern is determined by the base structure, centriole/basal body.

Centrioles/basal bodies are also highly conserved organelles that have two main functions: first as templates for cilia/flagella assembly, and second as a core of the centrosome.

Their structures consist of with nine triplet microtubules arranged in rotational symmetry. The inner two of the triplets are continuous to become the axonemal outer doublets.

Recently, many proteomic studies have identified flagellar and centriole proteins and super-resolution imaging studies have revealed flagellar and centriole structures precisely. However, it is still unclear how their characteristic structures assemble and how their components function.

For understanding it, I have used mutants of a green alga *Chlamydomonas reinhardtii*. At OIST, I have continued the studies with several newly isolated mutants with flagella/centriole defects.



2 Activities and Findings

• Lab set-ups

I moved to OIST in April 2020. From the culture room became available (October 2020), basic systems for experiments using cells, including observation under the light microscopes, genetic crossing, etc. were established.

In FY2021, as the second step, an electroporator for genetic recombination was introduced and full-scale immunostaining was just started.

The lab setup will be almost completed in FY2022.

• About the mutant *bld13*

I have continued to get electron microscopic images of the centrioles in *bld13-1* and *bld13-2* enough for statistics. They suggested that 1) the defects were more severe in the proximal part than in the distal part, and 2) *bld13-1* and *bld13-2* have different defects in the triplets.

And also, I isolated diploids (*WT/bld13-1*, *WT/bld13-2*, and *bld13-1/bld13-2*) and confirmed dominant negative effects of the *bld13* mutations.

• About a suppressor mutant of *bld10*

I have again started analyses of an extragenic suppressor mutant of an allele of *bld10* that I previously isolated. Electron microscopy revealed its centriole and flagella defects precisely. I just started trying 3D-EM for them.

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3 Collaborations

- Hosei University (Dr. Masafumi Hirono, Dr. Hiroko Toyooka)
- Tokyo Institute of Technology (Dr. Ken-ichi Wakabayashi)
- University of Tokyo (Dr. Mikito Owa, Dr. Akira Noga)
- Paul Scherrer Institute (Dr. Manuel Hilbert, Dr. Michel O. Steinmetz)

4 Publications and other output

Presentations

- **Yuki Nakazawa**, Madoka Hiraki, and Masafumi Hirono. The 59th Annual Meeting of the Biophysical Society of Japan (Nov. 2021).

Grants

- Kakenhi KibanC 19K06749 (2019-2022(extended)).