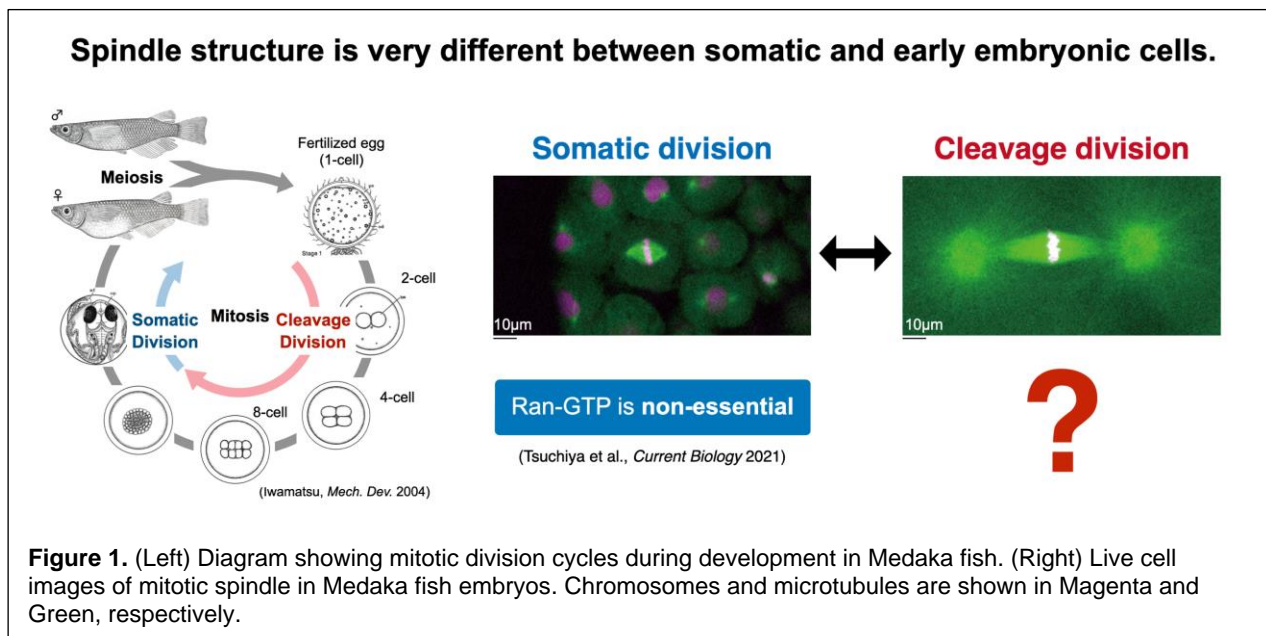


# Science and Technology Group Annual Report FY2022

Ai Kiyomitsu  
Science and Technology Associate

## 1 Introduction

During cell division, a microtubule-based structure called spindle segregates duplicated chromosomes to daughter cells to maintain genomic information. In animal mitosis, the spindle is assembled by multiple pathways including centrosomes and a chromosome-derived Ran-GTP gradient. Prior studies demonstrated that the Ran-GTP gradient is critical for acentrosomal spindle assembly in female meiosis, but dispensable for bipolar spindle formation in somatic human cell line with centrosomes (Tsuchiya et al., *Current Biology* 2021). Although spindle assembly mechanisms have been extensively studied in oocytes and somatic cells, mechanisms for centrosomal spindle assembly in large vertebrate embryos remain poorly understood.

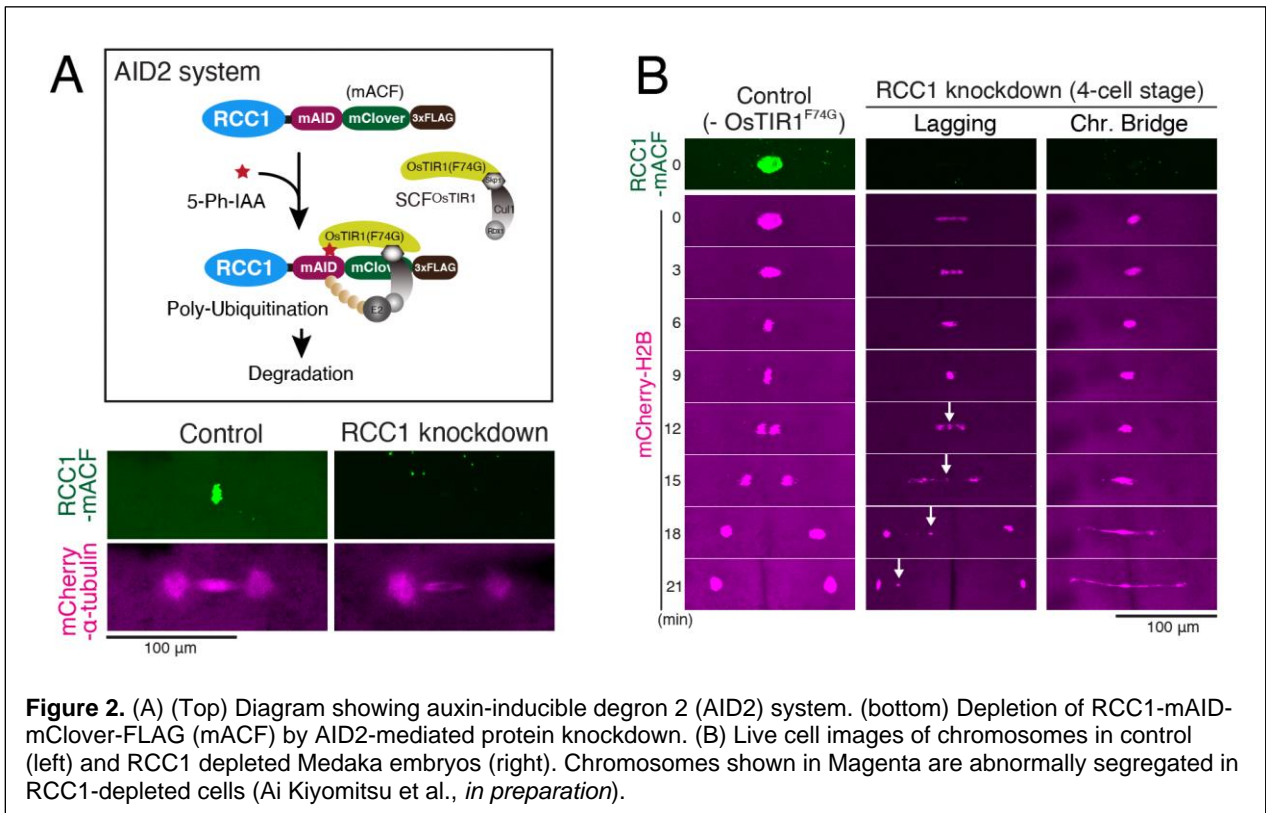


## 2 Activities and Findings

To understand the mechanisms of spindle assembly in Medaka early embryos, I analyzed requirement of Ran-GTP using CRISPR knock-in, live imaging, a dominant negative Ran mutant, and an auxin inducible degron2 (AID2)-based protein knockdown system. Most importantly, I could establish AID2 system (Figure 2A) in Medaka embryos in this fiscal year. Using this technique, I found that depletion of RCC1, a guanine-nucleotide exchange factor for Ran, diminishes the microtubule-dense region around chromosomes in zygotic spindles (Figure 2A), which results in abnormal chromosome segregation including anaphase lagging, chromosome bridge, and chromosome non-disjunction (Figure 2B). Based on these and other results, we are currently preparing a manuscript.

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## 3 Collaborations

Kiyomitsu Unit (OIST)  
Prof. Minoru Tanaka (Nagoya University)  
Dr. Toshiya Nishimura (Hokkaido University)  
Dr. Satoshi Ansai (Tohoku University)

## 4 Publications and other output

<Poster presentation>

Ai Kiyomitsu, *Ran-GTP promotes zygotic spindle assembly for accurate chromosome segregation in Medaka *Oryzias latipes**. OIST STG forum, March 28, 2023.

<Grant acquisition>

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JST FOREST grant (7 years from 2023-2029), Representative: Tomomi Kiyomitsu, Collaborator: Ai Kiyomitsu