

Report for Joint/Usage Research Program for Endocrine/Metabolism (Fiscal Year 2019)

Date: 2020/4/30

To Director of Institute for Molecular and Cellular Regulation, Gunma University

| Principal Applicant | |
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| Institution | Wellcome Trust / CRUK Gurdon Institute |
| Position | Group Leader |
| Name | Fengzhu Xiong |

We report on the results of joint research in fiscal 2019 as below.

(Program No. 19005)

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|--|---|--------------------|--|--------------|-------------------------------------|
| 1. Research Title | Interplay between cellular metabolism and tissue mechanics during vertebrate morphogenesis | | | | |
| 2. Purpose and Significance of the research project | The role of cellular metabolism in tissue patterning and homeostasis is increasingly being recognized. Cell metabolic activities were found to regulate cell signaling and movements that drive tissue morphogenesis. However, it is still unclear how metabolic activity regulates tissue mechanics (which directly controls morphogenesis) or vice versa. In this study, we propose to perform and correlate mechanical and metabolic measurements in the forming zebrafish embryonic body axis. This study will reveal the molecular and cellular mechanisms of morphogenesis through the regulation of cell metabolism. | | | | |
| 3. Period of The Program | April 1, 2019 ~ March 31, 2020 | | | | |
| 4. Project Members | | | | | |
| Name | Age | Gender | Institution/Department | Position | Role |
| (Principal Applicant) Fengzhu Xiong | 33 | M | Wellcome Trust / CRUK Gurdon Institute | Group Leader | Investigator |
| (Research Collaborators) | | | | | |
| | | | | | |
| ※If additional space is required, attach a separate sheet. | | | | | |
| 5. Collaborative Researcher of IMCR | Name of the Laboratory | Integrated Systems | Signaling | Name | Tohru Ishitani; Masayuki Oginuma |



6. Research Plans

1) Establishment of transgenic zebrafish lines to monitor metabolic activity

To quantify metabolic activity of the metabolic activity during zebrafish embryogenesis, Dr. Masayuki Oginuma will make transgenic zebrafish lines to monitor metabolite amounts for the main metabolic pathways by using fluorescent proteins (**ATP**: Queen2, Yaginuma et al. 2014. **H2O2**: Hyper., Belousov et al., 2006. **Glycolysis**: Peredox., Hung et al. 2011. **pH**: phluorin, MJ Mahon et al, 1998). In addition, chemical assays for metabolic measurements will be used for additional metabolites.

2) Implementation of tissue mechanical measurements

To quantify and modify the tissue forces and mechanical properties (elasticity and viscosity), Dr. Fengzhu Xiong will implement soft gel implants and magnetic pins (Xiong et al., 2018), ferro fluid droplets (Serwane et al., 2017), and atomic force microscopy (AFM) cantilevers (Chan and Xiong et al., unpublished) on the live zebrafish embryo undergoing anterior-posterior extension. These methods enable measurement of mechanical properties in live, intact embryos.

3) Integrated analysis of the interplay between metabolism and mechanics

To discover the mechanisms linking metabolic activities and tissue mechanics, we will measure one when we perturb the other. For example, we will image the zebrafish embryo embedded in soft gels of different stiffness (applying differential stress on the embryo), and examine effects for metabolism using metabolic reporter lines. We will also modify metabolic activity of the zebrafish embryo by chemical inhibitors and genetic knockdowns / overexpression, and examine the changes of mechanical properties

7. Research results:

In this year, we established transgenic zebrafish lines to monitor ATP, Glycolysis and pH by using fluorescent proteins (Queen2, Peredox, and phluorin), and found metabolic pathway show gradient patterns in early zebrafish embryo. Dr. Fengzhu Xiong visited IMCR and advised to set up systems to quantify mechanical properties (elasticity and viscosity) and modify the tissue forces. Now we start to modify the forces of zebrafish embryo and measure metabolic activity using the above method.

8. Publications and/or Presentations resulting from Joint Research Program with IMCR.

①Please describe a list of publications in which the name of the collaborative researcher of IMCR appears and send one paper reprints of each publication to IMCR.

②Please describe a list of publications which include the description that the research is supported by Joint Research Program with IMCR and send one copy of each publication to IMCR.