Application of Transient Gene Expression Using Lipid Nanoparticle During Early Embryo Development.

Chihiro Emori[1], Hiroki Tanaka[2], Hidetaka Akita[2], Masahito Ikawa[1,3]

1 Research Institute for Microbial Diseases, Osaka University, Osaka, Japan

2 Graduate School of Pharmaceutical Sciences, Tohoku University, Miyagi, Japan

3 The Institute of Medical Science, The University of Tokyo, Tokyo, Japan

Zygotes and preimplantation embryos have unique features compared to other cell types. For example, they have zona pellucida (ZP), which protects zygotes/embryos against physical damage and microbial infections. Due to these features, microinjection has been a first choice for gene delivery into zygotes. Alternative methods are electroporation or use of viral vectors. In addition, it has been reported that lentivirus vector can be used for gene introduction in blastocyst embryos and this showed gene expression only in trophectoderm (TE) which will become placenta in the future. However, it requires to remove ZP for lentivirus vector infection to blastocyst. Since each method have advantages and disadvantages, we aim to find new methods to introduce gene into zygotes or embryos. We focused on lipid nanoparticle (LNP) which has been used for mRNA delivery. We have showed that LNP can be used for transient gene expression in zygotes in the previous meeting. In this study we examined whether LNP can be used for gene introduction in blastocyst embryos. To observe mRNA delivery and subsequent expression, we used mTmG reporter mice, which express tdTomato but switch to EGFP after Cre recombinase-mediated excision of the tdTomato cassette. We treated mTmG blastocyst with cre mRNA encapsulated LNP overnight and checked their fluorescence. We observed that there were embryos that showed green fluorescence without removing ZP. Interestingly we observed green fluorescence only in TE but not in inner cell mass (ICM) suggesting that LNP can use for placenta specific gene transduction. This may help us to study the function of placenta and embryo developments.