Institute of DNA Medicine
Department of Gene Therapy

Yoshikatsu Eto, *Professor and Director* Toya Ohashi, *Professor*

General Summary

Our research focuses on efforts to develop methods of molecular therapy and cell therapy for genetic diseases, diabetes, kidney diseases, and cancer. Members of the laboratory staff were recruited from various departments of The Jikei University, including the departments of internal medicine, surgery, and gynecology. All members are working to develop novel therapies for intractable diseases.

Research Activities

Molecular therapy for genetic diseases

Fabry disease is a lysosomal storage disease characterized by deficient alpha-galactosidase A activity. This deficiency results in progressive accumulation of globotriaosylceremide (GL-3), mainly in vascular endothelial cells, leading to renal and cardiac failure. Enzyme replacement therapy is now available. However, antibodies form against infused enzyme in some male patients. Our studies this year revealed that these antibodies neutralize the enzyme and inhibit its cellular uptake. Although normalization of urinary GL-3 levels was achieved more efficiently in seronegative patients than in seropositive patients, the clinical outcome was not affected by antibody formation. Finally, our experiment showed that neonatal intravenous infusion of the enzyme induced immune tolerance in a murine model of Fabry disease.

Phosphorylation of shugoshin by aurora B

The conserved protein shugoshin plays a role in the maintenance of centromeric cohesion in mitosis and meiosis. In a previous study, we demonstrated that shugoshin accelerates kinetochore-driven formation of kinetochore microtubules for spindle assembly.

This year, we investigated the mechanism of bipolar attachment via shugoshin. Aurora B is a mitotic kinase that regulates bipolar attachment. Thus, we examined the phosphorylation of shugoshin by aurora B. We found that two highly conserved serine residues of shugoshin, located at the mid-portion and the C-terminal, were phosphorylated by aurora B.

Gene transfer to pancreas for the regeneration of islet cells

In our study of regeneration therapy for the endocrine pancreas in diabetes mellitus, we have developed a novel gene-delivery system: *in vivo* direct injection to the murine pancreas using an adeno-associated virus (AAV) vector. When the gene for cyclin-dependent kinase 4, a gene crucial for cell-cycle regulation and beta-cell proliferation,

was transduced to the remaining beta cells by the direct injection method, we observed improvement of metabolism in mice with diabetes. We are now investigating the precise mechanism for the therapeutic effects so that clinical application will be possible in the future.

Generation of an erythropoietin-producing organoid derived from human mesenchymal stem cells

Differentiation of autologous stem cells into functional tissue for organ regeneration is a promising regenerative therapeutic approach for many human diseases, including renal failure. Yet to be accomplished, however, is differentiation into tissue capable of producing erythropoietin. We report the generation of a stem-cell-derived organoid capable of producing erythropoietin and sensitive to regulation by anemia, indicating a function in erythropoiesis.

Mesenchymal-to-epithelial transition during the inclusion cyst formation from human ovarian surface epithelium

Most surface epithelial-stromal tumors of the ovary are thought to arise from epithelial inclusion cysts. Thus, these cysts are the precursor lesion of ovarian carcinoma. On the basis of this hypothesis, we aimed to characterize human ovarian surface epithelium in which the mesenchymal-to-epithelial transition occurs during inclusion cyst formation. We used specimens from 9 patients with endometrial cancer who underwent hysterectomy and bilateral salpingo-oophorectomy. Immuohistochemical study was performed with 10 normal ovaries containing 92 inclusion cysts and 4 normal tubes to examine the expression of antigen markers, including calretinin, podoplanin, D2-40, thrombomodulin, human bone marrow endothelial (HBME)-1, vimentin, epithelial membrane antigen (EMA) WT1, CA125, MOC31, TAG-72, Ber-EP4, and E-cadherin. We found that the staining rates for mesothelial markers in normal ovarian surface epithelium were 100% (10 of 10) for calretinin, 80% (8 of 10) for podoplanin, 80% (8 of 10) for D2-40, 70% (7 of 10) with thrombomodulin, 100% (10 of 10) for HBME-1, and 100% (10 of 10) for vimentin. In tubal epithelium the staining rates for epithelial markers were 100% (4 of 4) for HBME-1, 100% (4 of 4) for vimentin, 100% (4 of 4) for EMA, 75% (3 of 4) for TAG-72, and 100% (4/4) for Ber-EP4. Inclusion cysts showed staining for both markers with an incidence of 51.1% (47 of 92) for HBME-1, 44.6% (41 of 92) for vimentin, 65.2% (60 of 92) for TAG-72, and 88.0% (81 of 92) for Ber-EP4. Ovarian surface epithelium has both mesencyhmal and epithelial characteristics. In contrast, inclusion cysts gain epithelial characteristics and lose mesencyhmal characteristics. These findings support the observation of a mesenchymal-to-epithelial transition during inclusion cyst formation from the ovarian surface epithelium.

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