

## CONTROL OF CELL RECOGNITION AND ATTACHMENT OF HEPATOCYTES BY GLYCOPOLYMERS

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**INTRODUCTION:** Tissue engineering and regenerative medicine are now very attractive field of biomaterials. New polymers design for cell recognition and attachment is growing important. We paid attention to asialoglycoprotein receptor (ASGP-R) which appears in hepatocyte surface and E-cadherin that mediates the intercellular adhesion between hepatocytes, with the aim of the molecular design of new extracellular matrices (ECM) alternatives for cell and tissue engineering. On the former topics, the amphipathic polymer, PVLA, was designed by the combination of  $\beta$ -galactose into the styrene monomer and the functional expressions of hepatocytes that are phagocytosis, adhesion, drug metabolism (P-450) and bile acid secretion, etc., were examined by the adjustment of the coating condition of PVLA polymeric micelle. On the latter, the fusion chimeric molecule (E-cad-Fc) between extracellular domain of E-cadherin and fragment crystallisable region (Fc) of antibody molecule was designed by the genetic-engineering technique. The model system of attachment of hepatocyte onto polystyrene substrate coated with E-cad-Fc molecule which mimics the formation of cell-cell adhesion mediated by E-cadherin molecules between hepatocytes was established. Using this model system, we aimed at the elucidation of intercellular adhesion in comparison with hepatocyte-adhesions to PVLA and collagen.

### RESULTS & DISCUSSION:

*Control of drug metabolic activity in hepatocytes on collagen and PVLA.*

Aminopyrine N-demethylase (AMND) activity which is one of the drug metabolic activity of the hepatocyte showed the oscillational change with the cultivation time on the collagen-coated dish. This period of oscillation is synchronized with bile acid quantity in culture medium. And synchronization phenomenon between AMND activity and bile acid quantity was also observed on PVLA-coated dish (coating conc.: 1  $\mu$ g/ml). However, the period of oscillation on PVLA has slowed down than the oscillation period on

collagen and was partially perturbed. This time, the expression of mRNA of various proteins in hepatocyte is greatly different. That is, it synchronized to the oscillatory phenomenon of AMND activity and bile acid concentration, and the heat shock protein has appeared on collagen, and transcriptional regulatory factors appeared on PVLA.

*Biomimetics of Cell-Cell Adhesion mediated by E-cadherins:*

In order to clarify the cell adhesion mechanism through E-cadherins, the analysis in the model system was made. For the surface coated with the chimeric protein (E-cad-Fc), the hepatocyte remarkably adhered to the surface and cell adhesion was inhibited by addition of anti-E-cadherin antibody. Generally, the hepatocyte keeps the differentiated capacity by forming spheroid, and the DNA synthetic ability of spheroid lowers in comparison with the spreading form of hepatocytes. When cultured on E-cad-Fc, low DNA synthetic ability was also observed. In addition, the expression of tryptophan oxygenase, which is a special differentiative marker of hepatocyte, was maintained as same as spheroid. From these facts, the possibility of inducing the differentiation of hepatocyte by the cell adhesion through the E-cadherin was indicated. Chimeric protein (EGF-Fc) was also designed as new EMC analogues and examined in the viewpoint of cell adhesion and signal transduction