560e Induction of Embryonic Stem Cells into Endoderm Hepatic Lineage

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Embryonic stem (ES) cells are considered as a potential reliable source of cells for cellular therapies for liver diseases, toxicology screens, and the development of bioartificial liver devices. Although recent studies suggested that ES cells can differentiate into the endodermal hepatocyte lineage, their application has been hampered by the lack of efficient differentiation methods. In this study, we hypothesized that adult hepatocytes can induce the ES cells into hepatic differentiation. ES-D3 cells were used to assess the differentiation of ES cells into the hepatic endoderm lineage and to observe the effect of dose-response in co-culture systems. In order to monitor ES cell differentiation, we used Oct-4 promoter driven green fluorescent protein (GFP) ES cells, which are the marker for primitive ES cells. Oct-4/GFP gene expression was monitored by fluorescence microscopy. Embryonic bodies (EBs) and single ES cells were tested to investigate the endoderm hepatic lineage differentiation. EBs were formed by hanging drop methods. ES cell-derived cells were identified by morphologic, phenotypic, functional analysis, and gene expression. Immunofluorescence was employed to characterize the ES cell-derived cells using the hepatic lineage markers. Reverse transcriptase polymerase chain reaction (RT-PCR) was performed to analyze the gene expression of endoderm hepatic lineage. We describe here that ES cells in hepatocyte co-culture systems activate the differentiation of ES cells and the formation of a homogeneous population. Several markers for endoderm hepatic lineage of ES cells are being investigated. Further characterization of ES cell-derived hepatic cells is underway. This culture system will be useful in vitro model to induce and direct ES cells into hepatic differentiation and for application in cellular therapies for liver disease or bioartificial liver devices.