chymal stem cells in various aspects. They retained a normal diploid karyotype and growth characteristics over the successive culture. We have isolated hFLDSCs from low density mononuclear liver cells. To support the hypothesis that multipotent hFLDSC exist in most human tissues, we isolated and cultured a clonogenic and highly proliferative cell population from fetal liver and studies these cells biologic characteristics and differentiation potential. The results suggest that these cells, which we have termed hFLDSC, have all the properties of MSCs. Thus, our goal of the present study was to investigate whether hFLDSCs isolated 10-week old human abortus can support the growth of hES cells in a culture medium that is known to favor their proliferation while retaining the undifferentiated state.

Method: Human Fetal Liver Derived Stem Cells seperated by Ficoll centrifugation were cultivated at 37℃ with 5% CO_2 for 5 days in the medium made with Dulbecco's modified Eagle's medium 1,000 mg/L low glucose (DMEM-LG; GIBCO) and 10% fetal bovine serum and 100 U/ml penicillin, 100 μg/ml streptomycin for proliferation and expansion.

Results: We examined the effect of newly established hFLDSCs originated human abortus compared with conventional feeder cells, MEF or STO cells from mouse for maintaining human embryonic stem cells in vitro.

Conclusions: The established hFLDSCs were fully supported the growth and maintenance of characteristics of hES cell comparable to the mouse feeder system. Our hFLDSCs were maintained for upto 70 passages and have characteristics of mesenchymal stem cells. Also, these cells were maintain chromosomal stability. We are more detailed studies undertaken such as multicellular lineaged differentiation and capability of genetic modification.

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P-19 Effective Endothelial Differentiation from Human Embryonic Stem Cells using Growth Factor

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Background & Objectives: 전분화능을 가진 배아줄기세포는 3가지 germ layer leanage로 분화가 가능하며 무한대로의 증식이 가능하다. 이러한 배아줄기세포의 능력을 이용하여 세포치료, 독성연구, 발생학 연구 등에 널리 이용되고 있다. 본 연구에서는 이러한 인간 배아줄기세포를 이용하여 혈관생성 전구세포로의 효율적인 분화유도를 시도하였다.

Method: 인간배아줄기세포는 CHA-hES 3를 이용하였으며 배양액의 조성은 DMEM/12에 20% SR과 4 ng/ml의 bFGF를 첨가하여 배양하였다. 분화유도는 미분화된 인간배아줄기세포를 5일 동안 suspension culture하여 EB formation을 만든 후 VEGF, EGF, BFGF, EPO를 첨가한 분화유도 media를 이용하여 분

화유도하였다.

Results: RT-PCR과 Immunostaing 결과 여러 가지 growth factor가 첨가된 분화유도 조건에서 endothelial marker의 발현임 증가됨을 확인하였으며, FACS analysis 결과 endtothlial marker의 발현을 확인하였다.

Conclusions: 실험결과 VEGF, EGF, bFGF, EPO를 이용하여 혈관형성세포로의 분화를 유도한 결과 혈관세포의 표식인자인 PE-CAM, VE-CADherin, vWF의 발현을 확인하였으며 RT-PCR 결과 혈관세포 특이 유전자인 PE-CAM, Flk-1, Tie-1, 2의 발현이 시간이 지남에 따라서 증가함을 확인하였다. 따라서 인간배아줄기세포에서 혈관형성세포로의 분화유도가 더 효과적으로 이루어 짐을 확인하였다.

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P-20 In Vitro Differentiation of Insulin Producing Cells from Human Embryonic Stem Cells

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Background & Objectives: Human embryonic stem (ES) cells are derived from the inner cell mass of blastocyst and are capable of differentiating into a variety of cell lineages. Human ES cells can be new potential sources for cell therapy to cure degenerative diseases, such as diabetes mellitus. The objective of this study is to improve the culture conditions for differentiation of insulin-producing pancreatic beta cells using human ES cells.

Method: Insulin -producing islet like clusters were induced from human ES cells in vitro by 5-step protocol, as in mouse system. Undifferentiated human ES cells were cultivated on feeder layer of STO cells for 6~7 days (stage 1). Embryoid bodies (EB) were generated in suspension culture for 7 days (stage 2), and transferred to plates with coated poly-L-ornithine and fibronectin in ITSFn medium for 10 days (stage 3). The selected nestin positive cells were expanded in N_2 medium contained B27 supplement and 10 ng/ml bFGF for 6~7 days (stage 4). The pancreatic differentiation and maturation of these cells was carried out by addition of two factors, 10 mM nicotinamide and 10 μM LY294002 (inhibitor of phosphoinoside 3-kinase) into and withdrawal bFGF out of medium for 20 days (stage 5).

Results: Distinct morphology cell clusters were observed after differentiation of final maturation process. Immunocytochemical analysis demonstrated that insulin and glucagon expressed in the all clusters. Cytokeratin19, pancreatic ductal cell marker, is expressed in the cluster. Most cells in the clusters expressed insulin and C-peptide. Expression of glucagon was localized in the some parts of cell clusters. Pancreatic polypeptide (PP) is expressed partially in the cluster. RT-PCR analysis showed the insulin was expressed in