

P-11 In Vitro Neural Cell Differentiation of Genetically Modified Human Embryonic Stem Cells Expressing Tyrosine Hydroxylase

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Background & Objectives: This study was to examine in vitro neural cell differentiation pattern of the genetically modified human embryonic stem cells expressing tyrosine hydroxylase (TH).

Method: Human embryonic stem (hES, MB03) cell was transfected with cDNAs coding for TH. Successful transfection was confirmed by western immunoblotting. Newly transfected cell line (TH#2/MB03) was induced to differentiate by two neurogenic factors retinoic acid (RA) and b-FGF. Exp. I) Upon differentiation using RA, embryoid bodies (EB, for 4 days) derived from TH#2/MB03 cells were exposed to RA (10^{-6} M)/AA (5×10^{-2} mM) for 4 days, and were allowed to differentiate in N2 medium for 7, 14 or 21 days. Exp. II) When b-FGF was used, neuronal precursor cells were expanded at the presence of b-FGF (10 ng/ml) for 6 days followed by a final differentiation in N2 medium for 7, 14 or 21 days. Neuron differentiation was examined by indirect immunocytochemistry using neuron markers (NF160 & NF200).

Results: After 7 days in N2 medium, approximately 80% and 20% of the RA or b-FGF induced Th#2/MB03 cells were immunoreactive to anti-NF160 and anti-NF200 antibodies, respectively. As differentiation continued, NF200 in RA treated cells significantly increased to 73.0% on 14 days compared to that in b-FGF treated cells (53.0%, $p < 0.05$), while the proportion of cells expressing NF160 was similarly decreased between two groups. However, throughout the differentiation, expression of TH was maintained (90%). HPLC analyses indicated the increased levels of L-DOPA in RA treated genetically modified hES cells with longer differentiation time.

Conclusions: These results suggested that a genetically modified hES cells (TH#2/MB03) could be efficiently differentiated in vitro into mature neurons by RA induction method.

P-12 전뇌 허혈성 뇌졸중 (Global Ischemia) 동물 모델 Mongolian Gerbil에 대한 인간 배아줄기세포 이식효과

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Background & Objectives: 전뇌 허혈성 뇌졸중 동물모델에서 중풍 치료제로서 인간배아줄기 세포의 인지 및 기억력 장애에 대한 기능 회복의 효능 및 신경세포 보호효과를 검토하고자 실시하였다.

Method: 전뇌 허혈성 동물 모델은 웅성 모래쥐 (Mongolian gerbil, 70~80 g) 경부를 절개하여 흉골허근과 흉골저작근 사이의 양쪽 총경동맥을 5분 동안 폐색시켜 만들었다. 본 연구에 사용된 인간배아줄