alkaline phosphatase activity, detected high telomerase activity, expressed Oct-4, SSEA-3, SSEA-4, Tra-1-60 and Tra-1-81 and formed embryoid bodies (EBs).

Conclusions: HAF cell supported undifferentiated growth of HES cell and therefore these results may help to provide a clinically practicable method to expand HES cells for cell therapies. This research was supported by a grant (SC11011) from stem cell research center of the 21st. century Frontier research program funded by the ministry of Science & Technology, Republic of Korea.

P-46 Transforming Growth Factor-α Increases the Yield of Functional Dopaminergic Neurons from in vitro Differentiated Human Embryonic Stem Cells Induced by Basic Fibroblast Growth Factor

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Background & Objectives: In this study, we examined the in vitro neural cell differentiation patterns of hES cells (MB03), following induction by basic fibroblast growth factor (bFGF) or retinoic acid (RA). The effects of neurotrophic factors, such as brain derived neurotrophic factor (BDNF) or transforming growth factor (TGF-α), on differentiating hES cells were additionally investigated.

Methods: Exp. I) Embryoid bodies (EB) were derived from hES cells for 4 days. When bFGF was used, neuronal precursor cells were selected for 8 days in ITSFn medium after EB formation. After selection, cells were expanded at the presence of bFGF for another 6 days followed by a final differentiation in N2 medium for 7, 14, 21 days. Exp. II) EBs derived from hES cells were exposed of RA for 4 days, and were allowed to differentiate in N2 medium for 7, 14, 21 days. Exp. III) In addition, to examine the effects neurotrophic factors in the production of mature neurons, groups of cells were exposed to either BDNF or TGF-α during the 21 days of final differentiation.

Results: bFGF or RA treated hES cells were resulted in similar neural cell differentiation patterns at the terminal differentiation stage, specifically, 75% neurons and 11% glial cells. Additionally, treatment of hES cells with BDNF or TGF-α during the terminal differentiation stage led to significantly increased tyrosine hydroxylase (TH) expression, compared to control ($p<0.05$). In contrast, no effect was observed on the rate of mature or glutamic acid decarboxylase-positive neurons. Immunostaining and HPLC analyses revealed the higher levels of TH (20.3%) and dopamine in bFGF and TGF-α treated hES cells than in RA or BDNF treated hES cells.

Conclusion: The results indicate that TGF-α may be successfully used in the bFGF induction protocol to yield higher numbers of functional dopaminergic neurons from hES cells.