Cellular Transplantation as the Treatment of Alzheimer’s Disease in Mouse Models

Noboru Suzuki*, Jun Shimizu, Naruyoshi Fujiwara and Nagisa Arimitsu

Department of Immunology and Medicine and Department of Regenerative Medicine, St. Marianna University Graduate School of Medicine, Kawasaki, Japan

Abstract

Acetylcholine (Ach) and N-methyl-D-aspartate (NMDA) have been two major therapeutic targets of Alzheimer’s disease (AD) for decades. However, truly effective remedy for AD has not been successfully developed.

We previously transplanted neurons derived from human induced pluripotent stem (hiPS) cells into the hippocampus of human amyloid precursor protein transgenic AD model mice.

The cell transplantation significantly improved cognitive dysfunction in the dementia model mice. Human choline acetyl transferase (ChAT) positive cholinergic neurons located throughout the cortex of the grafted mice. Human and mouse ChAT positive neurons and alpha7 nicotinic acetylcholine receptor (α7nAChRs) positive neurons significantly increased in the cortex and hippocampus of the grafted dementia mice compared with the vehicle injected dementia mice.

Human and mouse vesicular GABA transporter (VGAT) positive neurons distributed mainly in the hippocampus and, though the number was small, human VGAT positive neurons located in the cortex. In the grafted mouse cortex, the number of GABA receptor (GABAR) positive neurons of both hiPS origin and mouse origin increased significantly compared with those in the vehicle injected mouse cortex.

We suggested that positive feedback loops of neurotransmitter secretion of the cortex and hippocampus induced the characteristic distribution of the transplanted neurons. In this review, we summarized current advances in stem cell therapy for dementia model mice, especially to highlight the relationships between major neurotransmitters and host/transplanted neurons.

Keywords: Alzheimer’s disease; Human iPS cells; Transplantation, Acetylcholine, GABA, Hippocampus

Introduction

Cholinergic neurons which secrete acetylcholine (Ach) play important roles in learning and memory functions. Cholinergic neuron activity and their acetylcholine production are down regulated in patients with Alzheimer’s disease (AD) [1], and down modulation of alpha7 nicotinic acetylcholine receptors (α7nAChRs) has been reported as one of the hallmarks of AD [2].

Japanese government is trying to handle the rapidly aging society with several new technology using, for example, robotics and regenerative medicine. In Japan, people older than 64 years increased to one fourth of the whole population and the cost of treating people with dementia was estimated to be around US$120 billion in 2014 [3].

We hardly modified the disease progression of AD using a single conventional medication [4]. Besides Ach and N-methyl-D-aspartate (NMDA) targeted conventional treatments, researchers tried to develop remedy using blocking molecules for production and accumulation of amyloid β (Aβ) [4]. Despite the increasing number of studies, no new drugs have been approved since 2003 for AD treatment. It was suggested that synaptic loss of brain by the accumulating Aβ oligomers was strongly associated with AD cognitive dysfunction compared with Aβ pathology, in which Aβ plaques were thought to be causally implicated [5].

Human induced pluripotent stem (hiPS) cell transplantation therapy for a degenerative eye disease has been reported in Japan [6]. Thereafter, they have become to receive much attention for their potential to rescue the impaired cellular functions of various diseased conditions, including cognitive function in patients with AD.

This review focused on the usefulness of stem cell transplantation for the treatment of dementia models and we summarized their effects on the disease pathology and cognitive function in AD models.

Neural Induction from Pluripotent Stem Cells

In the first step of vertebrate development, cells which are committed to differentiate into neural cells are suggested to differentiate into anterior/rostral fate (forebrain cells) of central nervous system (CNS), mainly by bone morphogenetic protein (BMP) inhibitors [7].

After the cell fate acquisition, an appropriate regional patterning of CNS is provided by several organizing centers of the neural tube [8]. BMP and sonic hedgehog (SHH) are important molecules for the dorsoventralization and ventralization of neurons within the neural tube, respectively. Retinoic acid (RA) is a key molecule for the caudalization of the neurons towards midbrain, hindbrain and spinal cord neuron fate [7]. Because of the inherent limitation of experimental studies, underlying molecular mechanisms of human brain development remain largely unexplored.

*Corresponding author: Noboru Suzuki, Department of Immunology and Medicine and Department of Regenerative Medicine, St. Marianna University Graduate School of Medicine, 2-16-1 Sugao, Miyamae-ku, Kawasaki, Kanagawa 216-8511, Japan, Tel: +81-44-977-8111; E-mail: n3suzuki@marianna-u.ac.jp

Received March 01, 2016; Accepted March 07, 2016; Published March 14, 2016

Citation: Suzuki N, Shimizu J, Fujiwara N, Arimitsu N (2016) Cellular Transplantation as the Treatment of Alzheimer’s Disease in Mouse Models. J Alzheimers Dis Parkinsonism 6: 219. doi: 10.4172/2161-0460.1000219

Copyright: © 2016 Suzuki N, et al. This is an open-access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.
Recent advances in cell biology of pluripotent stem cells, including embryonic stem (ES) and iPS cells, provided us new approaches for the analysis of brain development with human cells. Similar to in vivo assays, mouse [8] and human [9] ES cells showed forebrain identity during early differentiation period in default condition without any morphogens. RA promotes caudate fates of the ES cell forebrain identity and ventralization of the cells is induced by SHH supplementation [10]. We previously demonstrated that mouse [11] and monkey [12,13] ES cells differentiated into motor neurons with RA and the neuron transplantation ameliorated motor dysfunction of hemiplegic model mice. We observed that addition of Noggin, a BMP inhibitor, promoted forebrain identity of culturing mouse ES cells and, on the contrary, basic fibroblast growth factor (bFGF) promoted caudal fates of the cells [14].

Using a combination of RA, SHH and Noggin, we induced rapid neurogenesis in hiPS cells and the neuron progenitor cells were applicable for the treatment of AD [15,16] and spinal cord injury model mice [17].

MASH1 is a basic helix-loop-helix transcription factor and essential for the neuron development [18]. We found that MASH1 gene transfection of mouse ES cells induced motor neuron differentiation and the cell transplantation improved the motor function of hemiplegic [19] and spinal cord injury models [20]. Furthermore, we observed that brain derived neurotrophic factor (BDNF) significantly enhanced neural differentiation of mouse ES cells [21]. It seems that we are able to observe brain development process in pluripotent stem cells, especially using several morphogens.

Humoral Factors in Cell Transplantation of AD models

Neurotransmitters

One of the major histopathological changes of human AD is massive neural degeneration of nucleus basalis [22]. The degeneration leads to ACh deficits in the cortex and hippocampus. The ACh expression was negatively correlated with AD disease severity [23]. Available medicines approved for AD treatment are used to inhibit acetylcholinesterase and increase cholinergic neurotransmission of the brain. Certainly, the molecules improved cognition, behavior and functional and global clinical state of patients with AD at the mild to moderate stage [4]. In addition, there have been little advances of cholinergic medications during this decade [4].

Neural stem cell derived cholinergic neurons [24] and choline acetyltransferase (ChAT) overexpressing neural stem cells [25] significantly shorten the escape latency of Morris water maze (MWM) test after the transplantation, suggesting that upsurplus of cholinergic neurotransmitter improved cognitive function of AD models. It was recently reported that down regulation of vesicular GABA transporter (VGAT) [26] and GABA receptor (GABAR) [27] was more severe than previously thought in AD patients. Decreases of GABA expression were observed in PDAPP mice [28], tau protein transgenic mice, and apolipoprotein (apo) E4 knock-in/App mice [29], all of which exhibited an immature hippocampus with GABAergic neuronal dysfunction and memory deficits. Carrying apoE4 gene is a strong risk factor of AD and apoE4 directly impair the GABAergic inhibitory neuron function [30]. GABAergic interneuron progenitors transplanted into the hippocampal hilus were functionally integrated into the host hippocampus and improved learning and memory function in apoE4 knock-in/App mice [31].

Growth and neurotrophic factors

Simultaneous supplementation of BDNF enhanced therapeutic effects of stem cell transplantation on the learning and memory ability of AD model mice [32]. When genetically modified stem cells which produced nerve growth factor (NGF) were transplanted into the hippocampus of AD model mice, the learning and memory ability of the grafted mice were improved [33]. Transplantation of the vascular endothelial growth factor (VEGF) gene transfected cells ameliorated cognitive dysfunction of AD model mice, as well [34]. It has been shown that human neural stem cells expressed growth and trophic factors including BDNF, NGF and VEGF in the brain of AD model mice upon transplantation [35].

Cytokines, chemokines and toll-like receptors (TLR)

Aβ binds CD36, TLR4 and TLR6 and activates microglia [36]. Activated microglia secretes proinflammatory cytokines, such as interleukin (IL)1β, IL6 and TNFα and the process is thought to be the beginning of neuroinflammation of AD [36]. Thus, it was suggested that immune system has close relationship with integrity of neural network in the brain and therefore, AD progression.

Bone marrow stem cells and adipose tissue derived stem cells produced anti-inflammatory cytokines, such as IL4 and IL10, in the brain after the transplantation [37,38]. The cell transplantation significantly reduced Aβ plaque formation of the mice and improved the cognitive dysfunction. These stem cells may be useful to reduce local inflammation of the brain when applied to those with pre symptomatic stages of AD.

Cellular factors in cell transplantation of AD models

Recently, several researchers observed that intrinsic factors in the brain induced neural differentiation of transplanted stem cells. Human adipose tissue derived mesenchymal stem cells were transplanted by the two approaches; intravenous administration and injection into the cerebral ventricles of aging mice [39]. Both of the cell transplantation significantly improved the cognitive function. The transplanted cells effectively differentiated into ChAT positive cells and brain concentration of Ach significantly increased by both of the transplantation protocols. The transplanted mouse neural stem cells differentiated into cholinergic neurons and exhibited similar positive effects on the Ach concentration of brain and cognitive function in AD model mice [24].

SHH and BMP9 successfully generated ChAT, ISL1 LIM homeobox (ISL)1 and NK2 homeobox1 (NKX2.1) positive basal forebrain cholinergic neurons from mouse and human ES cells. ISL1 and NKX2.1 expressions are essential for the maturation and maintenance of the cell type in the development [40,41]. The transplantation into basal forebrain restored the cholinergic projection system and cognitive function in two AD mouse models [41].

Furthermore, GABAergic neuron progenitors differentiated into mature cells with inhibitory interneuron phenotype after the transplantation and migrated into hippocampus of AD model mice. The cell transplantation restored learning and memory function in the AD mice [31]. The transplanted mouse neuron stem cells promoted synaptogenesis of the brain and significantly improved cognitive function in two AD mouse models without alteration of Aβ or tau pathology [42]. Intracerebral micro environment as well as host cell-to-grafted cell interactions seem to play a crucial role in histological restoration and improvement of cognitive dysfunction in AD model mice.
As mentioned above, we previously transplanted hiPS cell derived neural cells into the hippocampus of AD model mice [15,16]. The cell transplantation significantly improved cognitive dysfunction of the model mice. Human and mouse ChAT positive neurons and α7nAChR positive neurons significantly increased in the cortex and hippocampus of the grafted mice compared with the vehicle injected mice [16].

In addition, human and mouse VGAT positive neurons were distributed mainly in the hippocampus and, though the number was small, human VGAT positive neurons were observed in the cortex [16]. In the grafted mouse cortex, the number of GABAR positive neurons of both human origin and mouse origin were significantly increased compared with those in the vehicle injected mouse cortex [16]. The α7nAChR positive and GABAR positive neurons expressed phosphorylated Akt and c-Fos in the cortex, suggesting that these receptor expressing neurons were possibly activated by the neurotransmitters secreted from the grafted neurons [16].

We suggested that positive feedback loops of neurotransmitter secretion of the cortex and hippocampus induced the characteristic distribution of the transplanted neurons. Each neuron distribution may form distinctive neuron networks using various humoral and cellular factors.

Conclusions

Transplantation of hiPS cell derived neuron is a promising candidate for the treatment of advanced AD. The graft autonomous effects on the regeneration of damaged neuron circuits are attractive mechanisms for the clinical application. Further studies are needed to elucidate how the grafts select the migration routes, which signals are important to decide the cell destination, whether long axonal projection of grafts exist, and how long the grafts survive and prolong the host neuron survival. These themes are suggested to be equally important for the practical application.

Funding

This study was partly supported by Grants-in-Aid for Scientific Research of Japan Society for the Promotion of Science. The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

References

1. Davis KL, Mohs RC, Marin D, Purohit DP, Perl DP, et al. (1999) Cholinergic markers in elderly patients with early signs of Alzheimer disease. JAMA 281: 1401-1406.
2. Court J, Martin-Ruiz C, Piggott M, Spurden D, Griffiths M, et al. (2001) Nicotinic receptor abnormalities in Alzheimer’s disease. Biol Psychiatry 49: 175-184.
3. McCurry J (2015) Japan will be model for future super-ageing societies. Lancet 386: 1523.
4. Mangialasche F, Solomon A, Winblad B, Mecocci P, Kivipelto M (2010) Apolipoprotein E ε4 and GABAR immunostaining in human brain and apparent GAD65 loss in Alzheimer’s disease. J Alzheimers Dis Parkinsonism 6: 219. doi: 10.4172/2161-0460.1000219
5. Selkoe DJ (2002) Alzheimer’s disease is a synaptic failure. Science 296: 789-791.
6. Kamio H, Manda M, Okamoto S, Sakai N, Sugia A, et al. (2014) Characterization of human induced pluripotent stem cell-derived retinal pigment epithelium cell sheets aiming for clinical application. Stem Cell Reports 2: 205-218.
7. Levine AJ, Brivanlou AH (2007) Proposal of a model of mammalian neural induction. Dev Biol 308: 247-256.
8. Gaspar N, Bocchot C, Houriez R, Dimidschatstein J, Naeije G, et al. (2008) An intrinsic mechanism of corticogenesis from embryonic stem cells. Nature 455: 351-357.
9. Chambers SM, Fasano CA, Papapetrou EP, Tomishima M, Sadelaan M, et al. (2009) Highly efficient neural conversion of human ES and iPS cells by dual inhibition of SMAD signaling. Nat Biotechnol 27: 275-280.
10. Wichterle H, Lieberam I, Porter JA, Jessell TM (2002) Directed differentiation of embryonic stem cells into motor neurons. Cell 110: 385-397.
11. Chiba S, Ikeda R, Kurokawa MS, Yoshikawa H,Takeno M, et al. (2004) Anatomical and functional recovery by embryonic stem cell-derived neural tissue of a mouse model of brain damage. J Neurol Sci 219: 107-117.
12. Ikeda R, Kurokawa MS, Chiba S, Yoshikawa H, Ide M, et al. (2005) Transplantation of neural cells derived from retinoic acid-treated cyonolus monkey embryonic stem cells successfully improved motor function of hemiplegic mice with experimental brain injury. Neurobiol Dis 20: 38-48.
13. Hazama Y, Kurokawa MS, Chiba S, Tsukada M, Imai T, et al. (2010) SDF1/CXCR4 Contributes to Neural Regeneration in Hemiplegic Mice with a Monkey ES-cell-derived Neural Graft. Inflamm Res 59: 203-205.
14. Chiba S, Kurokawa MS, Yoshikawa H, Ikeda R, Takeno M, et al. (2005) Noggin and basic FGF were implicated in forebrain fate and caudal fate, respectively, of the neural tube-like structures emerging in mouse ES cell culture. Exp Brain Res 163: 86-99.
15. Fujiwara N, Shimizu J, Takai K, Arimitsu N, Saito A, et al. (2013) Restoration of spatial memory dysfunction of human APP transgenic mice by transplantation of neuronal precursors derived from human iPS cells. Neurosci Lett 557: 129-134.
16. Fujiwara N, Shimizu J, Takai K, Arimitsu N, Ueda Y, et al. (2015) Cellular and molecular mechanisms of the restoration of human APP transgenic mouse cognitive dysfunction after transplant of human iPS cell-derived neural cells. Exp Neurol 271: 423-431.
17. Iinuma M, Umehara T, Arimitsu N, Shimizu J, Mitsawa H, et al. (2015) Induction of neural cells with spinal motoneuron phenotype from human iPS cells and the transplantation to totally transected spinal cords in mice. Inflamm Res 35: 154-163.
18. Casarosa S, Fode C, Guillemot F (1999) Mash1 regulates neurogenesis in the ventral telencephalon. Development 126: 525-534.
19. Ikeda R, Kurokawa MS, Chiba S, Yoshikawa H, Hashimoto T, et al. (2004) Transplantation of motoneurons derived from Mash1-transfected mouse mouse ES cells reconstitutes neural networks and improves motor function in hemiplegic mice. Exp Neurol 189: 280-292.
20. Hamada M, Yoshikawa H, Ueda Y, Kurokawa MS, Watanabe K, et al. (2006) Introduction of the Mash1 gene into mouse embryonic stem cells leads to differentiation of motoneuron precursors lacking Nogo receptor expression that can be applicable for transplantation to spinal cord injury. Neurobiol Dis 22: 509-522.
21. Kitagawa A, Nakayama T, Takenaga M, Matsumoto K, Tokura Y, et al. (2005) Lecithinized brain-derived neurotrophic factor promotes the differentiation of embryonic stem cells in vitro and in vivo. Biochem Biophys Res Commun 328: 1051-1057.
22. Cullen KM, Halliday GM (1998) Neurofibrillary degeneration and cell loss in the nucleus basalis in comparison to cortical Alzheimer pathology. Neurobiol Aging 19: 297-306.
23. Shinohoh H, Namba H, Fukushi K, Nagatsuka S, Tanaka N, et al. (2000) Progressive loss of cortical acetylcholinesterase activity in association with cognitive decline in Alzheimer’s disease: a positron emission tomography study. Ann Neurol 48: 194-200.
24. Gu G, Zhang W, Li M, Ni J, Wang P (2015) Transplantation of NSC-derived cholinergic neuron-like cells improves cognitive function in APP/PS1 transgenic mice. Neuroscience 291: 81-92.
25. Park D, Lee HJ, Joo SS, Bae DK, Yang G, et al. (2012) Human neural stem cells over-expressing choline acetyltransferase restore cognition in rat model of cognitive dysfunction. Exp Neurol 234: 521-526.
26. Schwab C, Yu S, Song W, McGeer EG, McGeer PL (2013) GAD65, GAD67, and GABAT immunostaining in human brain and apparent GAD65 loss in Alzheimer’s disease. J Alzheimers Dis 33: 1073-1088.
27. Rissman RA, Mobley WC (2011) Implications for treatment: GABAA receptors in aging, Down syndrome and Alzheimer’s disease. J Neurochem 117: 613-622.
28. Sun B, Halabiski B, Zhou Y, Palop JJ, Yu G, et al. (2009) Imbalance between GABAergic and Glutamatergic Transmission Impairs Adult Neurogenesis in an Animal Model of Alzheimer’s Disease. Cell Stem Cell 5: 624-633.
29. Li G, Bien-Ly N, Andrews-Zwilling Y, Xu Q, Bernardo A, et al. (2009) GABAergic interneuron dysfunction impairs hippocampal neurogenesis in adult apolipoprotein E4 knockin mice. Cell Stem Cell 5: 634-645.

30. Huang Y, Mucke L (2012) Alzheimer mechanisms and therapeutic strategies. Cell 148: 1204-1222.

31. Tong LM, Djukic B, Arnold C, Gillespie AK, Yoon SY, et al. (2014) Inhibitory interneuron progenitor transplantation restores normal learning and memory in ApoE4 knock-in mice without or with AβP accumulation. J Neurosci 34: 9506-9515.

32. Xuan AG, Long DH, Gu HG, Yang DD, Hong LP, et al. (2008) BDNF improves the effects of neural stem cells on the rat model of Alzheimer’s disease with unilateral lesion of fimbria-fornix. Neurosci Lett 440: 331-335.

33. Li LY, Li JT, Wu QY, Li J, Feng ZT, et al. (2008) Transplantation of NGF-gene-modified bone marrow stromal cells into a rat model of Alzheimer’ disease. J Mol Neurosci 34: 157-163.

34. Spuch C, Antequera D, Portero A, Orive G, Hernández RM, et al. (2010) The effect of encapsulated VEGF-secreting cells on brain amyloid load and behavioral impairment in a mouse model of Alzheimer’s disease. Biomaterials 31: 5608-5616.

35. Lee IS, Jung K, Kim IS, Lee H, Kim M, et al. (2015) Human neural stem cells alleviate Alzheimer-like pathology in a mouse model. Mol Neurodegener 10: 38.

36. Heneka MT, Carson MJ, Khoury EJ, Landreth GE, Brosseron F, et al. (2015) Neuroinflammation in Alzheimer’s disease. Lancet Neurol 14: 388-405.

37. Kim S, Chang KA, Kim Ja, Park HG, Ra JC, et al. (2012) The preventive and therapeutic effects of intravenous human adipose-dedired stem cells in Alzheimer’s disease mice. PLoS One 7: e45757.

38. Lee JK, Jin HK, Endo S, Schuchman EH, Carter JE, et al. (2010) Intracerebral transplantation of bone marrow-derived mesenchymal stem cells reduces amyloid-beta deposition and rescues memory deficits in Alzheimer’s disease mice by modulation of immune responses. Stem Cells 28: 329-343.

39. Park D, Yang G, Bae DK, Lee SH, Yang YH, et al. (2013) Human adipose tissue-derived mesenchymal stem cells improve cognitive function and physical activity in ageing mice. J Neurosci Res 91: 660-670.

40. Cho HH, Cargnin F, Kim Y, Lee B, Kwon RJ, et al. (2014) Isl1 directly controls a cholinergic neuronal identity in the developing forebrain and spinal cord by forming cell type-specific complexes. PLoS Genet 10: e1004280.

41. Yue W, Li Y, Zhang T, Jiang M, Qian Y, et al. (2015) ESC-Derived Basal Forebrain Cholinergic Neurons Ameliorate the Cognitive Symptoms Associated with Alzheimer’s Disease in Mouse Models. Stem Cell Reports 5: 776-790.

42. Ager RR, Davis JL, Agazaryan A, Benavente F, Poon WW, et al. (2015) Human neural stem cells improve cognition and promote synaptic growth in two complementary transgenic models of Alzheimer’s disease and neuronal loss. Hippocampus 25: 813-826.