REVIEW

Cell Therapy for Parkinson's Disease: What Next?

Anders Bjorklund, MD, PhD,[1]* and Jeffrey H. Kordower, PhD[2]

[1]*Wallenberg Neuroscience Center, Lund University, Sweden*
[2]*Department of Neurological Sciences, Rush University Medical Center, Chicago, Illinois, USA*

ABSTRACT: The idea to use transplants of dopamine-producing cells to substitute for the lost midbrain dopamine neurons in Parkinson's disease (PD) goes back to the 1970s. In this review we give an overview of the history of cell transplantation in animal models of PD, and summarize the experience gained from the open-label and placebo-controlled clinical trials performed so far using intrastriatal transplants of human fetal dopamine neuroblasts. Further development of this therapeutic approach face numerous challenges, for example in the development of protocols that allow generation of fully functional and safe midbrain dopamine neurons from stem cells. Based on recent promising advancements, efforts are now being made to develop standardized and efficient protocols, and adapt these protocols to good laboratory practice (GLP)/good manufacturing practice (GMP) conditions, to move this technology closer to clinical translation.

Key Words: dopamine; transplantation; clinical trials

The idea to substitute lost dopamine neurons with new ones, through transplantation, was first explored in laboratories in the United States and Sweden in the 1970s. The first grafting studies were performed either in the anterior chamber of the eye,[1] in the cerebral ventricles,[2] or into the brain parenchyma.[3–5] This early work was based on 2 rather different concepts: either delivery of dopamine released from cells implanted into the ventricle, adjacent to the striatum,[6,7] or restoration of synaptic dopamine release from dopamine neurons implanted into the brain parenchyma with the goal of reinnervating the denervated striatum.[8,9] Rats with unilateral, 6-hydroxydopamine (6-OHDA)-induced lesions of the nigrostriatal pathway received transplants of tissue obtained from the developing ventral mesencephalon (VM), implanted either (1) as a solid piece in the lateral ventricle[6] or a cortical cavity[8] adjacent to the denervated caudate-putamen, or (2) as a crude cell suspension injected directly into the striatal parenchyma.[10] Reversal of the unilateral motor deficit induced by the 6-OHDA lesion was monitored by drug-induced rotation. Previous studies had shown that the survival of brain tissue (and midbrain dopamine neurons) after transplantation is critically dependent on the age of the donor.[4,11] In the case of nerve cells obtained from the developing central nervous system (CNS), the optimal age was shown to be at, or close to, the cell-cycle exit. Based on these observations, the dopamine neurons used for transplantation in these experiments were neuroblasts obtained from mid-trimester rat fetuses.

Subsequent work, carried out in collaboration with Steve Dunnett and Susan Iversen in Cambridge, UK, showed that the recovery of motor functions induced by the grafted fetal dopamine neurons was well correlated with the extent of graft-derived reinnervation of the host caudate-putamen. Moreover, the impact of the grafts on different aspects of motor behavior was found to be dependent on which part of the caudate-putamen was innervated by the outgrowing axons.[9,12–14] More complete behavioral recovery was obtained only with transplants whose axonal terminal network covered a large part of the denervated striatal territory. This could be achieved with intraparenchymal grafting of cell suspensions, but not with transplants placed in the ventricular space. For this reason, intraventricular transplantation was soon

*Correspondence to: Dr. Anders Bjorklund, Wallenberg Neuroscience Center, Division of Neurobiology, Lund University, BMC A11, S-221 84 Lund, Sweden; anders.bjorklund@med.lu.se

Relevant conflicts of interest/financial disclosures: Nothing to report. Full financial disclosures and author roles may be found in the online version of this article.

Received: 1 October 2012; **Revised:** 11 December 2012; **Accepted:** 13 December 2012
Published online in Wiley Online Library (wileyonlinelibrary.com).
DOI: 10.1002/mds.25343

abandoned in favor of the intraparenchymal grafting approach.

Adrenal Chromaffin Cells and the Early Clinical Trials

At this point the development took an unexpected and somewhat surprising turn. Studies performed by Olson et al.[15] and Olson[16] at the Karolinska Institute in Stockholm had shown that adrenal chromaffin cells survive and grow neurites in a neuron-like manner after transplantation into the anterior eye chamber of rats . In an article published 1981 in *Nature*, Freed et al.[17] reported the results of an experiment using intraventricular grafts of adrenal medullary tissue in rats with unilateral 6-OHDA lesions, showing a significant 40% to 50% reduction in apomorphine-induced rotation 2 months after transplantation. In the absence of any significant reinnervation of the striatum from the surviving chromaffin cells, it was suggested that the effect was due to the release of catecholamines and subsequent diffusion of the released amines into the adjacent striatum. Based on these observations chromaffin cells from the adrenal medulla were seen as an interesting alternative to fetal dopamine neurons. The use of fetal tissue for transplantation was widely viewed as ethically problematic at this time, and the possibility to use cells that could be obtained from the patients themselves was thus very attractive. Moreover, the use of autologous grafts would have the additional advantage of avoiding any immune reactions associated with the use of allogeneic fetal brain tissue. The Freed et al.[17] study suggested that chromaffin cells could be just as good as dopamine neurons, although they would work through a different mechanism, diffuse catecholamine release instead of reinnervation and restoration of synaptic dopamine neurotransmission.

Indeed, despite the limited experimental data available at the time, the Karolinska team decided that it was appropriate to test adrenal medullary transplants clinically in patients. The first patient was operated in March 1982, and over the subsequent 3 years a total of 4 advanced PD patients received injections of adrenal medullary tissue (from 1 of their own adrenals) unilaterally into either the head of the caudate nucleus (patients 1 and 2) or the putamen (patients 3 and 4). The results, reported in 1985 by Backlund et al.[18] and 1987 by Lindvall et al.,[19] were unimpressive, and in the meantime experimental studies performed by the Karolinska group[20,21] had reported that chromaffin cells survived poorly and only displayed very transient reductions in drug-induced rotations when implanted into the striatal parenchyma, probably due to their normal dependence on nerve growth factor (NGF) for their long-term survival.[22]

The results of these early trials were thus quite discouraging. However, the interest in adrenal medullary grafting was powerfully rekindled by a report by a Mexican team, led by the neurosurgeon Ignazio Madrazo, published in the *New England Journal of Medicine* in 1987 (Madrazo et al.[23]). They reported dramatically positive results in 2 patients that received pieces of adrenal medulla placed in a premade cavity in the head of the caudate nucleus on 1 side using an open surgical approach rather than stereotaxic surgery. In a review published 3 years later, Madrazo et al.[24] summarized the results obtained in 42 consecutively operated cases followed over 1 to 3 years. Four patients died and 4 were lost to follow-up. Of the remaining 34, 15 showed a good response (more than 30 points on the Unified Parkinson's Disease Rating Scale [UPDRS] scale in the on state) and 7 a moderate response (20–30 points). Other investigators, however, did not observe the same dramatic improvements (see review in Ref. 25). In a 2-year follow-up of 61 cases, comparable to those operated on by Madrazo et al.,[23,24] Goetz et al.[29] reported only modest long-term improvements in about 32% of the cases, while 22% had persistent psychiatric side effects. When global improvement was calculated for the entire group (including deaths), signs of improvement were seen in only 19% of the patients at 2 years. The mechanism of improvement remains largely unknown, and there is little evidence to suggest that the improvement is due to catecholamine release from the grafted cells. Indeed, no or very few surviving chromaffin cells have been found in those patients that have come to autopsy[25]; although at autopsy, some limited graft-mediated sprouting was observed in the caudate nucleus[26]; a response also seen in parkinsonian monkeys.[27,28]

Thirteen U.S. centers contributed to the database organized by the Rush Medical Center in Chicago. The summaries of the results from this database, published in 1990 and 1991,[29,30] played an important role in the further development of the cell transplantation field. Over a 3- to 5-year period, several hundred patients received this form of surgery, in the United States and elsewhere. Many of these cases were never followed or reported properly in the scientific literature. Overall, this upsurge in adrenal medullary grafting was quite damaging to the cell transplantation field. In retrospect, we can see that the approach was ill-conceived (relying on poor and very limited preclinical data), and the way the initial open-label clinical trials were conducted led to overly optimistic and poorly documented reports that paved the way for disappointment and gave the skeptics good reasons to question the credibility of the whole field.

The First Trials Using Fetal VM Tissue Grafts

The enthusiastic endorsement of adrenal medulla transplantation was mostly confined to non-European

sites. This probably reflected a resistance to the use of fetal tissue, especially in the United States, because the use of tissues obtained from abortions was, and is, a highly contentious and also much politicized issue. In Europe, particularly in the northern European countries, the situation was quite different. Use of fetal tissue was seen as a more interesting and a scientifically more valid approach. Furthermore, the attitudes to legal abortions were overall more liberal. The teams in Lund and Stockholm joined forces to obtain permission to use VM tissue from aborted fetuses for this purpose. In 1985, the Lund group received permission to use human fetal tissue for preclinical studies in the rat PD model, showing that cell suspensions prepared from VM tissue from 6.5- to 9-week-old aborted fetuses survived well in the striatum of 6-OHDA–lesioned rats (using daily injections of cyclosporine to prevent rejection), innervated the host striatum, released dopamine, and were able to reverse the lesion-induced motor deficits in the grafted animals.[31,32]

In March 1986, the Ethical Delegation of the Swedish Society for Medicine issued provisional guidelines for the use of tissue from dead aborted fetuses for transplantation purposes (later included in Swedish law), which paved the way for the first trials using fetal VM tissue, starting in 1987. The results obtained in these open trials were quite promising, showing significant and sustained improvement in a number of clinical parameters, accompanied by marked increases in [^{18}F]-dopa positron emission tomography (PET) uptake in the striatum.[33–39] The recovery in [^{18}F]-dopa uptake is a good indicator of survival and growth of the grafted dopamine neurons, which is supported by postmortem data showing robust graft survival and good reinnervation of the grafted putamen at various time points posttransplantation.[40] Further studies using [^{11}C]-raclopride and $H_2{}^{15}O$ PET have shown that transplant-derived reinnervation of the putamen is able to restore D_2 receptor occupancy, ie, dopamine release,[41] and that the transplant can restore to normal the activation of those motor cortical areas that are underactive in PD.[42]

The National Institutes of Health–Sponsored Placebo-Controlled Trials

Although promising, the observations made in these small open-label trails could not in any way control for investigator bias and placebo effects. Thus, to prove efficacy, proper placebo-controlled trials were needed. In the United States, a ban on federal funding for fetal tissue research had been introduced by the Reagan administration in 1988. In November 1992, 3 papers appeared in the *New England Journal of Medicine*.[36,43,44] The overall promising results reported in these 3 studies appeared at a critical time, just before the Clinton administration took over. These reports generated considerable interest and debate, and may have played a role in the new president's decision to lift this ban in January 1993 (the first made in his presidency). This opened the way for the National Institutes of Health (NIH) to provide funding for 2 placebo-controlled studies, the Colorado/Columbia trial and the Tampa/Mount Sinai/Rush trial, that were initiated in mid 1990s and reported in 2001 and 2003.[45,46]

Both of these trials failed to reach significance on their primary endpoints and thus, as defined by clinical trial procedures, were negative. Furthermore, a significant number of patients in each trial developed an unexpected side effect of the surgery, ie, the development of graft-induced dyskinesias, abnormal involuntary movements that persisted also after removal of levodopa medication.[45,46] In retrospect, however, it is clear that these trials may have been performed prematurely. The cell transplantation approach was in a very early stage of development and many technical issues and problems, such as cell preparation and storage, site(s) of implantation, patient selection and immunosuppressive treatment, had not been studied properly, and certainly not properly sorted out. Thus, the protocols used in the 2 trials were based on very limited or insufficient experience from human studies. Notably, in the Colorado/Columbia study the tissue used for transplantation was stored in culture for up to 4 weeks, and no immunosuppression was used, and the patients included in the Tampa/Mount Sinai/Rush trial were quite advanced, likely due to the emergence of another surgical treatment at that time, deep brain stimulation. It remains questionable as to whether such severe patients would benefit from this approach. It is interesting to note that the secondary endpoints in both trials revealed that younger patients (Colorado/Columbia study) and less severe patients (Tampa/Mount Sinai/Rush study) displayed statistically significant benefit following fetal nigral grafting. Furthermore, Ma et al.,[47] in a follow-up study, reported the longer-term outcome in 33 transplanted patients from the Colorado/Columbia trial, 14 of which were originally included in the sham control group and had opted to receive transplants after the end of the formal 12-month endpoint. This analysis, which was performed unblinded and without the original control group, showed significant improvements in UPDRS motor scores and [^{18}F]-dopa PET at 2 to 4 years posttransplantation, which is in line with what had been reported in the open label studies that preceded this trial. Moreover, the posttransplantation changes in [^{18}F]-dopa uptake in the grafted putamen were found

to be significantly correlated with the clinical outcome over the course of the study, while uptake in other, non-transplanted areas (caudate and ventro-rostral striatum) showed a progressive decline. These data suggest that the clinical improvement induced by the transplants develop slowly over time. This is in line with observations made in the Lund program showing that grafted patients continue to improve over at least 2 to 3 years.[42,48,49] The reason for this protracted development is that the grafted neurons need to become integrated into host circuitry in order to exert their maximal effect. This process may take months to years. Clinical trials using a short endpoint will thus runs the risk of missing the full impact of the transplanted cells.

What Next?

The overall negative outcome of the 2 NIH trials, and the observations of troublesome dyskinetic side effects in particular, have had a major impact in the field, and no further trials using fetal VM tissue have been performed since the publication of these results. For further development of the cell therapy approach we face 4 major challenges:

First, why is the clinical outcome so variable? It is true that the procedures used at different centers vary greatly, and there has so far been no attempt to standardize either tissue preparation, implantation technique or immunosuppressive treatment. All these technical parameters need to be optimized in future trials.

Second, the results obtained so far suggest that not all patients respond equally well to transplants of dopamine neurons implanted into the putamen. Indeed, there are interesting observations suggesting that good clinical outcome in grafted patients are obtained only in patients who have a significant sparing of the dopaminergic innervation outside the motor striatum, in ventral striatum (including nucleus accumbens) in particular.[47,50] Thus, selection of patients may be needed in order to obtain consistent results, at least as long as there is a need to limit transplantation to the striatum.

Third, graft-induced dyskinesias, as observed in the NIH trials, have subsequently been confirmed also in some of the patients in the open-label trials.[51] This is a major concern, and the underlying mechanisms, whether related to the severity of L-dopa–induced dyskinesias prior to surgery, to the development of hotspots of dopaminergic innervation derived from the grafts, or to the inclusion of other neuronal elements, particularly serotonergic neurons, in the graft preparation, need to be sorted out (see Ref. 51). Recent studies in three patients with graft-induced dyskinesia shown that silencing of serotonin neurons through activation of inhibitory serotonin 1A (5-HT1A) receptors (by buspirone) is efficient in blocking the involuntary movements,[48,49] thus suggesting that serotonin neurons included in the graft preparation may play a role. This points to the need to standardize and control the composition of the cell preparation used for transplantation.

Fourth, the use of fetal tissue for grafting is unsatisfactory and in many ways problematic. Although fetal dopamine neurons are the only kind of dopaminergic cell that is know to work well for cell replacement in PD, the fetal VM tissue used to obtain these cells is problematic in many respects. The cells are difficult to obtain in sufficient number, and the quality and viability of the tissue is highly variable and almost impossible to standardize. In addition, the use of tissues and cells from dead aborted fetuses is ethically problematic, and indeed not legally allowed in many countries. For further development of the cell transplantation approach, it will be essential to develop techniques to generate transplantable dopamine neuroblasts from stem cells. In this development, however, fetal dopamine neurons will serve as a reference and a standard against which stem cell–derived neurons will have to be compared.

Generation of Transplantable Dopamine Neurons from Stem Cells: How Far Have We Reached?

Cells with at least some of the characteristics of mesencephalic dopamine neurons have been produced from stem cells of widely different origins. So far, the most promising results have been obtained using embryonic stem (ES) cells.[51–62] Other types of stem cells, such as neural stem cells (NSCs) and progenitors from the embryonic ventral mesencephalon, adult NSCs from the subventricular zone, bone marrow stromal cells, fibroblast-derived induced pluripotent stem (iPS), and fibroblast-derived induced neural (iN cells) cells, have also been used for this purpose (for recent reviews, see Refs. [63] and [64]). Most of these protocols have been developed for mouse cells, and it is only recently that it has been possible to obtain efficient generation of authentic midbrain dopamine neurons from human ES cells. The protocols recently developed by Kriks et al.[65] and Kirkeby et al.[66] are particularly promising. The procedure used in these protocols involve stepwise exposure of the cells to extrinsic factors activating sonic hedgehog and wnt signaling (which act to generate ventral midbrain progenitors), in combination with neuronal differentiation and survival factors, such as brain-derived neurotrophic factor (BDNF), glial cell line–derived neurotrophic factor, ascorbic acid, and cyclic adenosine monophosphate (cAMP). The cells generated in these protocols express all markers of authentic midbrain dopamine neurons

and they have been shown to survive and function after transplantation to the striatum, in both rodent and primate PD models. Importantly, the human ES cell–derived dopamine neurons have been shown to survive well after grafting, without any sign of tumor formation.

So, Are These Cells Ready for Use in Humans?

No, not yet. Some properties which are fundamental for successful clinical translation have not yet been demonstrated for human ES cell–derived dopamine neurons, such as long-term stability of the grafted cells and long-lasting functional recovery, as assessed in tests of both spontaneous and drug-induced motor behaviors. Moreover, their ability to effectively reinnervate the striatum and restore striatal dopamine release in vivo have yet to be demonstrated; this feature, in particular, may be critical for mediating substantial clinical benefit. Further experimental studies, as well as generation of cells under good manufacturing practice (GMP) conditions, remains to be performed before human ES cell–derived dopamine neuroblasts can be selected as safe candidate cells for use in patients.

A major concern in the use of cells derived from ES cells is the risk of tumor growth. Because life expectancy is virtually normal in PD patients, even a minor risk of tumor formation associated with stem cell therapy is unacceptable in this disorder. Human ES cells can give rise to unlimited numbers of progeny, but are associated with a risk of tumor formation. The initial in vivo observations on the cells obtained in the Kriks et al.[65] and Kirkeby et al.[66] protocols, showing no signs of overgrowth over at least 4 to 6 months, are quite promising in this regard. Based on these findings, there are now efforts to improve and standardize the protocols, and adapt them to good laboratory practice (GLP)/GMP conditions, to move this technology closer to clinical translation.

References

1. Olson L, Seiger A. Brain tissue transplanted to the anterior chamber of the eye. 1. Fluorescence histochemistry of immature catecholamine and 5-hydroxytryptamine neurons reinnervating the rat iris. Z Zellforsch Mikrosk Anat 1972;135:175–194.
2. Rosenstein JM, Brightman MW. Intact cerebral ventricle as a site for tissue transplantation. Nature 1978;276:83–85.
3. Das GD, Altman J. Studies on the transplantation of developing neural tissue in the mammalian brain. I. Transplantation of cerebellar slabs into the cerebellum of neonate rats. Brain Res 1972;38:233–249.
4. Bjorklund A, Stenevi U, Svendgaard N. Growth of transplanted monoaminergic neurones into the adult hippocampus along the perforant path. Nature 1976;262:787–790.
5. Lund RD, Hauschka SD. Transplanted neural tissue develops connections with host rat brain. Science 1976;193:582–584.
6. Perlow MJ, Freed WJ, Hoffer BJ, Seiger A, Olson L, Wyatt RJ. Brain grafts reduce motor abnormalities produced by destruction of nigrostriatal dopamine system. Science 1979;204:643–647.
7. Freed WJ, Perlow MJ, Karoum F, et al. Restoration of dopaminergic function by grafting of fetal rat substantia nigra to the caudate nucleus: long-term behavioral, biochemical, and histochemical studies. Ann Neurol 1980;8:510–519.
8. Bjorklund A, Stenevi U. Reconstruction of the nigrostriatal dopamine pathway by intracerebral nigral transplants. Brain Res 1979;177:555–560.
9. Bjorklund A, Dunnett SB, Stenevi U, Lewis ME, Iversen SD. Reinnervation of the denervated striatum by substantia nigra transplants: functional consequences as revealed by pharmacological and sensorimotor testing. Brain Res 1980;199:307–333.
10. Bjorklund A, Schmidt RH, Stenevi U. Functional reinnervation of the neostriatum in the adult rat by use of intraparenchymal grafting of dissociated cell suspensions from the substantia nigra. Cell Tissue Res 1980;212:39–45.
11. Seiger A, Olson L. Quantitation of fiber growth in transplanted central monoamine neurons. Cell Tissue Res 1977;179:285–316.
12. Dunnett SB, Bjorklund A, Stenevi U, Iversen SD. Grafts of embryonic substantia nigra reinnervating the ventrolateral striatum ameliorate sensorimotor impairments and akinesia in rats with 6-OHDA lesions of the nigrostriatal pathway. Brain Res 1981;229:209–217.
13. Dunnett SB, Bjorklund A, Schmidt RH, Stenevi U, Iversen SD. Intracerebral grafting of neuronal cell suspensions. V. Behavioural recovery in rats with bilateral 6-OHDA lesions following implantation of nigral cell suspensions. Acta Physiol Scand Suppl 1983;522:39–47.
14. Dunnett SB, Bjorklund A, Schmidt RH, Stenevi U, Iversen SD. Intracerebral grafting of neuronal cell suspensions. IV. Behavioural recovery in rats with unilateral 6-OHDA lesions following implantation of nigral cell suspensions in different forebrain sites. Acta Physiol Scand Suppl 1983;522:29–37.
15. Olson L, Seiger A, Freedman R, Hoffer B. Chromaffine cells can innervate brain tissue: evidence from intraocular double grafts. Exp Neurol 1980;70:414–426.
16. Olson L. Fluorescence histochemical evidence for axonal growth and secretion from transplanted adrenal medullary tissue. Histochemie 1970;22:1–7.
17. Freed WJ, Morihisa JM, Spoor E, et al. Transplanted adrenal chromaffin cells in rat brain reduce lesion-induced rotational behaviour. Nature 1981;292:351–352.
18. Backlund EO, Granberg PO, Hamberger B, et al. Transplantation of adrenal medullary tissue to striatum in parkinsonism. First clinical trials. J Neurosurg 1985;62:169–173.
19. Lindvall O, Backlund EO, Farde L, et al. Transplantation in Parkinson's disease: two cases of adrenal medullary grafts to the putamen. Ann Neurol 1987;22:457–468.
20. Herrera-Marschitz M, Stromberg I, Olsson D, Ungerstedt U, Olson L. Adrenal medullary implants in the dopamine-denervated rat striatum. II. Acute behavior as a function of graft amount and location and its modulation by neuroleptics. Brain Res 1984;297:53–61.
21. Stromberg I, Herrera-Marschitz M, Hultgren L, Ungerstedt U, Olson L. Adrenal medullary implants in the dopamine-denervated rat striatum. I. Acute catecholamine levels in grafts and host caudate as determined by HPLC-electrochemistry and fluorescence histochemical image analysis. Brain Res 1984;297:41–51.
22. Stromberg I, Ebendal T, Seiger A, Olson L. Nerve fiber production by intraocular adrenal medullary grafts: stimulation by nerve growth factor or sympathetic denervation of the host iris. Cell Tissue Res 1985;241:241–249.
23. Madrazo I, Drucker-Colin R, Diaz V, Martinez-Mata J, Torres C, Becerril JJ. Open microsurgical autograft of adrenal medulla to the right caudate nucleus in two patients with intractable Parkinson's disease. N Engl J Med 1987;316:831–834.
24. Madrazo I, Franco-Bourland R, Ostrosky-Solis F, et al. Neural transplantation (auto-adrenal, fetal nigral and fetal adrenal) in Parkinson's disease: the Mexican experience. Prog Brain Res 1990;82:593–602.
25. Lindvall O. Neural transplantation in Parkinson's disease. In: Dunnett S, Bjorklund A, eds. Functional neural transplantation. New York: Raven Press; 1994:103–137.

26. Kordower JH, Cochran E, Penn RD, Goetz CG. Putative chromaffin cell survival and enhanced host-derived TH-fiber innervation following a functional adrenal medulla autograft for Parkinson's disease. Ann Neurol 1991;29:405–412.
27. Fiandaca MS, Kordower JH, Hansen JT, Jiao SS, Gash DM. Adrenal medullary autografts into the basal ganglia of Cebus monkeys: injury-induced regeneration. Exp Neurol 1988;102:76–91.
28. Plunkett RJ, Baniewicz KS, Cummings AC, Miletich RS, Schwartz JP, Oldfield EH. Long-term evaluation of hemiparkinsonian monkeys after adrenal autografting or cavitation alone. J Neurosurg 1990;73:918–926.
29. Goetz CG, Stebbins GT 3rd, Klawans HL, et al. United Parkinson Foundation Neurotransplantation Registry on adrenal medullary transplants: presurgical, and 1- and 2-year follow-up. Neurology 1991;41:1719–1722.
30. Goetz CG, Stebbins GT 3rd, Klawans HL, et al. United Parkinson Foundation Neurotransplantation Registry: multicenter US and Canadian data base, presurgical and 12 month follow-up. Prog Brain Res 1990;82:611–617.
31. Brundin P, Nilsson OG, Strecker RE, Lindvall O, Astedt B, Bjorklund A. Behavioural effects of human fetal dopamine neurons grafted in a rat model of Parkinson's disease. Exp Brain Res 1986;65:235–240.
32. Brundin P, Barbin G, Strecker RE, Isacson O, Prochiantz A, Bjorklund A. Survival and function of dissociated rat dopamine neurones grafted at different developmental stages or after being cultured in vitro. Brain Res 1988;467:233–243.
33. Lindvall O, Brundin P, Widner H, et al. Grafts of fetal dopamine neurons survive and improve motor function in Parkinson's disease. Science 1990;247:574–577.
34. Lindvall O, Rehncrona S, Brundin P, et al. Human fetal dopamine neurons grafted into the striatum in two patients with severe Parkinson's disease. A detailed account of methodology and a 6-month follow-up. Arch Neurol 1989;46:615–631.
35. Lindvall O, Widner H, Rehncrona S, et al. Transplantation of fetal dopamine neurons in Parkinson's disease: one-year clinical and neurophysiological observations in two patients with putaminal implants. Ann Neurol 1992;31:155–165.
36. Widner H, Tetrud J, Rehncrona S, et al. Bilateral fetal mesencephalic grafting in two patients with parkinsonism induced by 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP). N Engl J Med 1992;327:1556–1563.
37. Lindvall O, Sawle G, Widner H, et al. Evidence for long-term survival and function of dopaminergic grafts in progressive Parkinson's disease. Ann Neurol 1994;35:172–180.
38. Wenning GK, Odin P, Morrish P, et al. Short- and long-term survival and function of unilateral intrastriatal dopaminergic grafts in Parkinson's disease. Ann Neurol 1997;42:95–107.
39. Hagell P, Schrag A, Piccini P, et al. Sequential bilateral transplantation in Parkinson's disease: effects of the second graft. Brain 1999;122(Pt 6):1121–1132.
40. Kordower JH, Rosenstein JM, Collier TJ, et al. Functional fetal nigral grafts in a patient with Parkinson's disease: chemoanatomic, ultrastructural, and metabolic studies. J Comp Neurol 1996;370:203–230.
41. Piccini P, Brooks DJ, Bjorklund A, et al. Dopamine release from nigral transplants visualized in vivo in a Parkinson's patient. Nat Neurosci 1999;2:1137–1140.
42. Piccini P, Lindvall O, Björklund A, et al. Delayed recovery of movement-related cortical function in Parkinson's disease after striatal dopaminergic grafts. Ann Neurol 2000;48:689–695.
43. Freed CR, Breeze RE, Rosenberg NL, et al. Survival of implanted fetal dopamine cells and neurologic improvement 12 to 46 months after transplantation for Parkinson's disease. N Engl J Med 1992;327:1549–1555.
44. Spencer DD, Robbins RJ, Naftolin F, et al. Unilateral transplantation of human fetal mesencephalic tissue into the caudate nucleus of patients with Parkinson's disease. N Engl J Med 1992;327:1541–1548.
45. Freed CR, Greene PE, Breeze RE, et al. Transplantation of embryonic dopamine neurons for severe Parkinson's disease. N Engl J Med 2001;344:710–719.
46. Olanow CW, Goetz CG, Kordower JH, et al. A double-blind controlled trial of bilateral fetal nigral transplantation in Parkinson's disease. Ann Neurol 2003;54:403–414.
47. Ma Y, Tang C, Chaly T, et al. Dopamine cell implantation in Parkinson's disease: long-term clinical and (18)F-FDOPA PET outcomes. J Nucl Med 2010;51:7–15.
48. Politis M, Wu K, Loane C, et al. Serotonergic neurons mediate dyskinesia side effects in Parkinson's patients with neural transplants. Sci Transl Med 2010;2:38ra46.
49. Politis M, Oertel WH, Wu K, et al. Graft-induced dyskinesias in Parkinson's disease: high striatal serotonin/dopamine transporter ratio. Mov Disord 2011;26:1997–2003.
50. Piccini P, Pavese N, Hagell P, et al. Factors affecting the clinical outcome after neural transplantation in Parkinson's disease. Brain 2005;128(Pt 12):2977–2986.
51. Hagell P, Piccini P, Björklund A, et al. Dyskinesias following neural transplantation in Parkinson's disease. Nat Neurosci 2002;5:627–628.
52. Lee SH, Lumelsky N, Studer L, Auerbach JM, McKay RD. Efficient generation of midbrain and hindbrain neurons from mouse embryonic stem cells. Nat Biotechnol 2000;18:675–679.
53. Björklund LM, Sanchez-Pernaute R, Chung S, et al. Embryonic stem cells develop into functional dopaminergic neurons after transplantation in a Parkinson rat model. Proc Natl Acad Sci U S A 2002;99:2344–2349.
54. Cho MS, Lee YE, Kim JY, et al. Highly efficient and large-scale generation of functional dopamine neurons from human embryonic stem cells. Proc Natl Acad Sci U S A 2008;105:3392–3397.
55. Kawasaki H, Suemori H, Mizuseki K, et al. Generation of dopaminergic neurons and pigmented epithelia from primate ES cells by stromal cell-derived inducing activity. Proc Natl Acad Sci U S A 2002;99:1580–1585.
56. Kim JH, Auerbach JM, Rodriguez-Gomez JA, et al. Dopamine neurons derived from embryonic stem cells function in an animal model of Parkinson's disease. Nature 2002;418:50–56.
57. Rodriguez-Gomez JA, Lu JQ, Velasco I, et al. Persistent dopamine functions of neurons derived from embryonic stem cells in a rodent model of Parkinson disease. Stem Cells 2007;25:918–928.
58. Roy NS, Cleren C, Singh SK, Yang L, Beal MF, Goldman SA. Functional engraftment of human ES cell-derived dopaminergic neurons enriched by coculture with telomerase-immortalized midbrain astrocytes. Nat Med 2006;12:1259–1268.
59. Sanchez-Pernaute R, Lee H, Patterson M, et al. Parthenogenetic dopamine neurons from primate embryonic stem cells restore function in experimental Parkinson's disease. Brain 2008;131(Pt 8):2127–2139.
60. Takagi Y, Takahashi J, Saiki H, et al. Dopaminergic neurons generated from monkey embryonic stem cells function in a Parkinson primate model. J Clin Invest 2005;115:102–109.
61. Cooper O, Hargus G, Deleidi M, et al. Differentiation of human ES and Parkinson's disease iPS cells into ventral midbrain dopaminergic neurons requires a high activity form of SHH, FGF8a and specific regionalization by retinoic acid. Mol Cell Neurosci 2010;45:258–266.
62. Friling S, Andersson E, Thompson LH, et al. Efficient production of mesencephalic dopamine neurons by Lmx1a expression in embryonic stem cells. Proc Natl Acad Sci U S A 2009;106:7613–7618.
63. Lindvall O, Kokaia Z. Stem cells in human neurodegenerative disorders—time for clinical translation? J Clin Invest 2010;120:29–40.
64. Ming GL, Brüstle O, Muotri A, et al. Cellular reprogramming: recent advances in modeling neurological diseases. J Neurosci 2011;31:16070–16075.
65. Kriks S, Shim JW, Piao J, et al. Dopamine neurons derived from human ES cells efficiently engraft in animal models of Parkinson's disease. Nature 2011;480:547–551.
66. Kirkeby A, Grealish S, Wolf DA, et al. Generation of regionally specified neural progenitors and functional neurons from human embryonic stem cells under defined conditions. Cell Rep 2012;1:703–714.