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Chapter 14

Cell-Based Therapies for Parkinson's Disease: Preclinical and Clinical Perspectives

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Abstract

Parkinson's Disease (PD) is a highly prevalent neurodegenerative disease that affects millions of people globally and remains without definitive treatment. There have been many recent advances in cell-based therapy to replace lost neural circuitry and provide chronic biological sources of therapeutic agents to disease-affected brain regions. Early neural transplantation studies highlighted the challenges of immune rejection, graft integration, and the need for renewable, autologous graft sources. Neurotrophic factors (NTFs) offer a potential class of cytoprotective agents that may complement dopamine (DA) replacement and cell-based therapies in PD. In fact, chronic NTF delivery may be an integral goal of cell transplantation in PD, with ideal grafts consisting of autologous drug (e.g., DA, NTF)-producing cells capable of integration and function in the host brain. This chapter outlines the past and recent preclinical and clinical advances in cell-based and NTF therapies as promising and integrated approaches for the treatment of PD.

Keywords: transplantation, tissue graft, stem cells, pluripotent cells, autologous cells, dopamine replacement, neurotrophic factors

1. Introduction

Parkinson's disease (PD) is the second most common neurodegenerative disorder, following Alzheimer's disease. In the developed world, the prevalence of PD is approximately 0.3% of the population and 1% of those over 60 years of age [1]. Hallmarks of PD include degenera-

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tion of dopamine (DA) neurons in the substantia nigra (SN) and of dopaminergic nerve terminals in the striatum, as well as the formation of Lewy bodies containing alpha-synuclein [2–4]. However, PD also has widespread effects on neurons and nonneuronal cells throughout the nervous system [4].

Motor symptoms of PD include bradykinesia (i.e. slowness of movement), rigidity, and rest tremor. These motor symptoms are often seen with postural and gait instability, sleep disorders, sensory dysfunction, neuropsychiatric conditions, and dysautonomia [5]. Nonmotor symptoms of PD include dementia, depression, gastrointestinal, or sexual dysfunction and are managed accordingly. Current therapies for PD aim to improve symptomology, but unfortunately there are no disease modifying treatments. Recent preclinical studies have provided promising leads for the development of potential new therapies to restore or preserve neurological function in patients with PD. As the pathophysiology of PD has become better understood, efforts are expanding to augment or replace the degenerated neural circuitry using cell-based therapies. The goal of this chapter is to discuss past and current approaches to cell-based therapies in PD, including studies to replace lost dopaminergic cells through neural grafting, and the potential of neurotrophic factors (NTFs) to promote DA neuron survival.

2. Current therapies

2.1. Medical therapies

The use of levodopa is the mainstay of PD treatment, and it is usually administered together with a decarboxylase inhibitor, carbidopa. Levodopa can cross the blood-brain barrier, whereas DA and carbidopa cannot. Carbidopa therefore prevents the peripheral conversion of levodopa to DA, allowing for higher doses in the central nervous system. Identified in the 1960s, levodopa was the first medication demonstrated to provide a significant clinical and mortality benefit in the treatment of PD [6]. However, long-term use of levodopa can lead to loss of therapeutic effect, dyskinesia, and neuropsychiatric complications, likely due to the progressive loss of DA neurons and increasing off-target effects of DA ([7], for review see [8]). Levodopa is converted by catechol-O-methyl transferase (COMT) to an inert metabolite [9]; as such, COMT inhibitors may be administered to prevent peripheral metabolism and increase levodopa availability to the brain. The use of selective monoamine oxidase B (MAO-B) inhibitors was initially thought to be neuroprotective and has since been used in symptom control [10] as monotherapy in early PD, as well as an adjunct treatment to levodopa [11]. Cholinergic, adrenergic, glutamatergic, and serotonergic drugs are also being used for treating PD symptoms that do not respond to DA treatment or for treating levodopa-induced side effects. All medical therapies only provide partial and temporal relief of symptoms and are not disease modifying [8].

2.2. Standard surgical therapies

Prior to the advent of levodopa therapy, ablative therapies were used in the control of motor symptoms. Pallidotomy and thalamotomy were used in the symptomatic control of rigidity

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and tremor, respectively [12,13]. Pallidotomy has been demonstrated to provide sustained improvement for tremor, rigidity, bradykinesia, and drug-induced dyskinesias, compared with medical therapy [14]. In the past decades, deep brain stimulation (DBS) has become the standard of surgical care for PD owing to the versatility of stimulator programming, and the avoidance of creating a permanent surgical lesion [15]. The two primary targets of DBS are the internal globus pallidus (GPi) and subthalamic nucleus (STN). DBS improves motor symptoms and often permits a reduction of medication dose and associated side effects, but does not slow or halt progression of the disease [16].

3. Burgeoning therapies for PD

3.1. Cell-based therapies

As PD is characterized by the loss of dopaminergic nigrostriatal neurons, cell-based therapies initially focused on the potential to replace these neurons and replenish DA supply in the striatum using fetal mesencephalic neural grafts. More recently, studies have included the transplantation of induced pluripotent stem cells (iPSCs), reprogrammed somatic cells or induced neural progenitor cells (iNPCs).

3.2. Fetal transplantation

3.2.1. Preclinical studies

Early PD transplantation studies involved grafting of fetal ventral mesencephalic (fVM) tissue into the anterior chamber of the rat eye [17]. These studies identified the optimal developmental stage of the neural tissue to be used to promote DA neuron survival and outgrowth [17]. The first graft transplantation studies in a unilaterally lesioned 6-hydroxydopamine (6-OHDA) PD rat model examined the effects of solid grafts of fetal adrenal medullary or fVM tissue implanted into the lateral ventricle or preformed cavities adjacent to the striatum and reported reduced amphetamine-induced rotation behavior [18]. Subsequent studies showed that grafting cell suspensions of fVM tissue from 14- to 15-day-old rat fetuses into the striatum of 6-OHDA rats also reduced amphetamine-induced rotation behavior [19]. Follow-up studies used fVM tissue from 9- to 19-week-old human fetuses. These implants reduced and even reversed motor asymmetry in unilaterally lesioned 6-OHDA rats [20].

3.2.2. Clinical studies

In 1987, solid graft adrenal medullary transplants were implanted in the head of the caudate in two patients and produced significant clinical improvement, including reduced tremor [21]. Unfortunately, follow-up studies showed only modest clinical effect, with concerns regarding efficacy and safety of this technique. Many patients suffered major postsurgical complications and psychiatric problems, thus this transplant approach was abandoned. Subsequent open label studies in six human patients utilized human fVM tissue from 6- to 8-week-old fetuses grafted into the caudate and putamen, demonstrating overall clinical improvement

and normal DA signaling seen by ^{18}F-Fluorodopa (^{18}F-FDOPA) uptake in Positron Emission Tomography (PET) imaging [22–24]. Two patients in this study continued to demonstrate clinical improvement 20 years later [25]. In a subsequent open label study nigral grafts from 6- to 7-week-old embryos were implanted into the caudate and putamen of seven PD patients [26,27]. Significant improvement in the activities of daily living was noted after 12 months, in both "on" and "off" states. The dose of levodopa could be reduced by an average of 39%. Four patients reported an "important difference in their daily lives," two patients reported improvement in "some respects," and one patient did not improve. Other open label studies with a small number of patients also showed mostly beneficial effects ([28–31], reviewed in [32]).

A randomized double-blind controlled trial (RDBCT) enlisted 34 patients that underwent transplantation of fetal mesencephalic tissue into the putamen or sham surgery. The patients showed limited clinical improvement, despite graft survival and significant reinnervation of the striatum as confirmed by PET and at autopsy. Interestingly, patients with less severe motor dysfunction showed significant clinical improvement, suggesting this technique may have produced some degree of neuroprotection. Furthermore, graft-induced dyskinesias were observed in over half of the patients [33]. Interestingly, these patients underwent a 2- and 4-year follow-up RDBCT that demonstrated clinical improvement regardless of the age of the patient, which was accompanied by significantly increased ^{18}F-FDOPA uptake in the putamen [34]. Another RDBCT had 40 patients with advanced PD undergo transplantation. When results were normalized according to age, patients under the age of 60 showed significant clinical improvement, as measured with the Unified Parkinson's Disease Rating Scale (UPDRS) and Schwab and England score, while those over the age of 60 did not [35]. Clinical improvement was correlated with increased ^{18}F-FDOPA uptake in 85% of patients with a transplant and postmortem examination confirmed dopaminergic cell survival and fibre outgrowth; however, 15% of patients developed graft-induced dyskinesias or dystonia [35].

To more accurately assess the potential of fetal grafts, a new European study has been designed to optimize and control for patient selection, tissue composition, tissue placement, and trial design. TRANSEURO is an open label multicenter trial to define the feasibility and efficacy of human fetal ventral mesencephalic grafts in patients with PD (https://clinicaltrials.gov/ct2/show/NCT01898390). The primary outcome measure of this study is the change in motor UPDRS scores in the absence of PD medications at 3 years posttransplantation. It is hoped that this new trial will shed light on the true potential of dopaminergic allografts for PD treatment.

The use of human fVM tissue, however, is complicated by ethical issues and difficulty in obtaining human tissue. Strategies are being developed that involve expansion of fVM tissue and its dopaminergic neuroblasts [36], and other cell sources are also being investigated for cell-based treatment of PD.

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3.3. Native human stem cells

3.3.1. Preclinical studies

Human embryonic stem cells (hESCs) were first isolated from the inner cell mass of blastocysts. These cells demonstrate pluripotency and have been shown to differentiate into neural cells, including neurons, astrocytes, and oligodendrocytes [37,38]. hESCs may prove useful to avoid the technical concerns associated with the use of fetal tissue. hESCs have been shown to differentiate into midbrain DA neurons, and injection of these cells in 6-OHDA lesioned rats [39] and 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP)-lesioned monkeys [40] leads to significantly improved motor function in both models. However, the development of clinical applications using these cells has been slowed by various biological and social factors, including the potential for immune rejection and tumor formation, as well as ethical and political opposition [41].

Alternative stem cell sources have been investigated for differentiation into a neural lineage, in particular mesenchymal stem cells (MSCs) from bone marrow, umbilical cord blood, dental pulp, and adipose tissue [42]. Autologous MSCs are favorable due to their availability, potential for differentiation, and the absence of ethical issues associated with hESCs. In addition, MSCs have been demonstrated to exert regenerative and neuroprotective effects in a number of animal PD models potentially, due to endogenous NTF expression. MSCs have been shown to differentiate into dopaminergic cells that express tyrosine hydroxylase [43]. Implantation of these differentiated MSCs into the striatum of 6-OHDA mice led to significant behavioral improvement, with striatal graft cells confirmed at postmortem analysis [43].

3.3.2. Clinical studies

There is currently very limited clinical data on the efficacy of hESCs in PD. An initial clinical study in seven PD patients demonstrated that transplantation of autologous bone-marrow-derived MSCs was safe and feasible with no serious adverse side effects. Unfortunately, no clinical efficacy was observed, potentially due to the small number of patients and uncontrolled nature of the trial [44].

3.4. Induced pluripotent stem cells

3.4.1. Preclinical studies

With the discovery that somatic cells, such as fibroblasts, can be reprogrammed to a pluripotent state by viral delivery of four transcription factors, Oct4, Sox2, Klf4, and cMyc [45,46], studies have focused on the potential of these induced pluripotent stem cells (iPSCs) to improve current cell-based therapies for treatment of many degenerative medical conditions, including PD. Compared with fetal grafting and hESC cells, iPSCs provide increased accessibility as well as ethical advantages. These cells can differentiate into many different cell types including cardiomyocytes [47], hepatocytes [48], oligodendrocytes [49], glia, and neuronal subtypes [50]. Murine iPSCs have also been shown to be reprogrammed into dopaminergic

neurons that express the transcription factors Nurr1, Pitx3 and tyrosine hydroxylase and demonstrate electrophysiological properties of DA neurons [51]. Subsequent studies demonstrated successful differentiation of dopaminergic neurons from both established human iPSC lines and patient-derived somatic cells [52,53].

In all of these studies, iPSC-derived DA cells demonstrated expression of key dopaminergic markers and electrophysiological properties of DA neurons. Furthermore, these DA cells were successfully incorporated into a 6-OHDA rat model of PD, leading to significantly reduced motor asymmetry [52]. Most recently, primate-derived iPSCs were successfully transplanted back into the putamen of MPTP lesioned monkeys; these autografts led to significant motor improvements, and postmortem analysis showed graft survival and outgrowth into the transplanted putamen [54]. Despite promising results in preclinical studies, several factors have provided road blocks to utilization of these cells in humans, including use of viruses to modify cells and risk of tumorigenicity [55].

3.4.2. Clinical studies

The first pilot study in humans was performed in Japan in 2014, and utilized iPSC-derived autologous pigmented retinal epithelial cells for treatment of macular degeneration. Transplantation in the first patient was completed without adverse effects; however, long-term follow-up is necessary [56]. Unfortunately, iPSCs derived from fibroblasts of a second patient were discovered to have genetic mutations, including three single-nucleotide variations and three copy-number variants, prompting suspension of the trial [57]. Studies are now focusing on use of allogenic partially matched donor cells from the Center for iPS Cell Research and Application, an iPSC bank, for treatment of macular degeneration [57]. There is also potential to transform fibroblasts directly into neurons (iN cells) or dopamine cells (iDA cells) using specific transcription factors: Ascl1, Brn2, Mrt1l, without or with Lmx1a and FoxA2, respectively [58–60]; however, this technology still needs to be tested in preclinical models.

3.5. Autologous brain-derived progenitor cells (BDPCs)

Recently, the safety and feasibility of performing small volume brain biopsies has been demonstrated in PD patients undergoing DBS surgery [61]. These tissue specimens yield an expandable cell population that expresses several NTFs known to be highly protective against PD neurodegeneration, including glial-derived neurotrophic factor (GDNF), brain-derived neurotrophic factor (BDNF), and cerebral dopamine neurotrophic factor (CDNF) [61]. The cultures yield large numbers (i.e. 10^7) of cells with limited capacity for self-renewal. These cells, called BDPCs, also show expression of progenitor and neural markers including nestin, Olig1, and GalC. Colocalization of neural and oligodendroglial markers suggests these BDPCs may be grafted into the host brain to integrate as autologous glia [61]. A patient-derived cellular source of neuroprotective agents, reimplanted into the host brain, may confer long-lasting therapeutic benefit in PD patients. Preclinical studies on the potential for these BDPCs to be used as an autologous cell-based therapy for PD are currently underway.

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4. Neurotrophic factors

Neurotrophic factors (NTFs) are being intensively evaluated as therapeutic agents for PD owing to their known roles in neuronal survival, differentiation, and plasticity. Additionally, NTF deficiency has been associated with PD and replacement or enhancement of NTF signaling confers neuronal protection in both in vitro and in vivo preclinical PD models [62]. These secreted proteins regulate vital biological programs in the developing and adult nervous systems and are currently the most potent cytoprotective agents known against PD-related degeneration in the brain [63]. The NTF families include (1) glial-derived neurotrophic factor (GDNF) family of ligands (GFL), (2) neurotrophins, (3) neuropoietic cytokines (neurokines), and (4) cerebral DA neurotrophic factor (CDNF)/mesencephalic astrocyte-derived neurotrophic factor family (MANF). To date, GDNF and other GFL members have received the most attention in development of potential new clinical therapies for PD [64].

4.1. GDNF family of ligands

GDNF was the first member of the GDNF family of ligands (GFL) to be discovered. Other members include neurturin (NRTN), persephin (PSPN), and artemin (ARTN). GFLs are important for cell survival, neurite outgrowth, cell differentiation, and cell migration [65].

4.1.1. Preclinical studies

Application of GDNF to rat ventral mesencephalic cultures increased survival, neurite length, and differentiation of DA neurons [66]. GDNF also reduced apoptosis and enhanced cell survival in cultures derived from monkey, porcine, and human mesencephalic tissues [67]. These effects extend to promote differentiation and protection of dopaminergic neurons against 6-OHDA and MPTP neurotoxins [65]. Numerous in vivo studies have demonstrated therapeutic effects of GDNF in the 6-OHDA rat model of PD; the infusion of recombinant GDNF significantly increased DA neuron survival in both the SN and ventral tegmental area and improved parkinsonian symptomology, including motor impairments and amphetamine-induced rotational behavior [68–71]. GDNF has also been shown to be neuroprotective in the MPTP mouse model of PD [72,73]. Studies in nonhuman primates have demonstrated that intracerebral administration of GDNF in MPTP-treated rhesus monkeys results in significant improvements in bradykinesia, rigidity, and postural instability, as well as increased DA levels in the midbrain, globus pallidus, and SN [74].

4.1.2. Clinical studies

Based on the success of using GDNF in preclinical models of PD, GDNF has now been studied in four clinical trials via infusion into the ventricular system or putamen [75]. The first RDBCT compared effects of intracerebroventricular administration of recombinant methionyl human GDNF (r-metHuGDNF, Liatermin®; Amgen) in escalating doses or placebo in 50 PD patients over a period of 8 months. No significant improvement in "on" and "off" total and motor UPDRS was seen in patients treated with GDNF. Adverse effects included paresthesias,

nausea, and vomiting. A follow-up open label study in 16 of these patients for 20 months showed no additional improvement in PD symptomology. It was felt that the adverse effects resulted from off-target GDNF influence and the lack of therapeutic benefit from an inability of GDNF to diffuse into the parenchyma from the ventricular source [76].

A subsequent open label study that enrolled 5 PD patients investigated the effects of intraparenchymal delivery of GDNF via implanted catheters in the dorsal putamen (unilateral in one patient; bilaterally in four patients) and connected to an extracranial pump system [77]. After one year, there were no serious clinical side effects, a 39% improvement in the off-medication UPDRS motor scores and a 61% improvement in the activities of daily living (ADL) subscore. Medication-induced dyskinesias were considerably reduced and (PET) scans of ^{18}F-FDOPA uptake showed a significant 28% increase in putamen DA storage after 18 months [78]. In a follow-up report, the group described one of the patients with bilateral GDNF infusions who had received treatment for 39 months, then was followed clinically and with PET for another 36 months. The UPDRS motor and ADL scores "off" medication remained improved by 74% and 76%, respectively, levodopa usage ceased after a year, and at 36 months post-GDNF cessation, the ^{18}F-FDOPA uptake remained 29% higher in the posterior putamen [79]. Another group led a second open label study that enrolled 10 patients treated unilaterally with intraputamenal GDNF [80]. A significant increase in total and motor UPDRS scores was observed after 24 weeks, but benefit was lost with cessation of treatment. These positive outcomes spurred a second multicenter, placebo-controlled trial in which 34 PD patients were randomized to receive bilateral intraputamenal GDNF (15 μg/putamen/day; a dose lower than that of the previous studies) or placebo via continuous infusion. At 6 months, there was no significant treatment benefit reflected in the "off" UPDRS motor scores; however, a 32.5% increase in putamenal ^{18}F-FDOPA uptake was observed in the GDNF-treated cohort [81]. The disparate outcomes of these studies may reflect differences in study design, cohort size, drug dosage, and/or delivery systems. The r-metHuGDNF manufacturing company subsequently withdrew the agent on the grounds of safety concerns regarding production of neutralizing antibodies in several patients and related cerebellar injuries in animal studies, although no such injuries were reported in human trials. Efforts are now underway to evaluate adeno-associated virus (AAV)-mediated GDNF in an open label phase I for patients with advanced PD (https://www.clinicaltrials.gov/ct2/show/NCT01621581).

4.2. Neurturin

4.2.1. Preclinical studies

Neurturin (NTRN) shares 40% sequence homology with GDNF [82] and has been shown to promote survival of DA neurons in the nigrostriatal system [82–84]. In vitro, NTRN leads to neurite outgrowth in cultured spinal motor neurons and protects against glutamate toxicity. Early studies infusing NTRN directly into the SN was shown to be neuroprotective against 6-OHDA toxicity, while striatal infusion improved behavioral parameters of DA neuronal function in rats [83,85]. In MPTP-lesioned monkeys, intraputamenal infusion of NTRN led to significant improvement in parkinsonian deficits as well as increased DA metabolite levels in

the globus pallidus [86]. CERE-120, an (AAV) vector expressing NTRN, has also shown potential therapeutic benefit in preclinical studies [87]. When MPTP-lesioned monkeys were given CERE-120 into the striatum, motor symptoms were improved and loss of DA neurons was reduced [88]. After one year follow-up, no toxic adverse effects were observed [89].

4.2.2. Clinical studies

A Phase 1 open-label clinical trial demonstrated safety, tolerability, and potential therapeutic benefit in PD patients after one year [90]. A subsequent RDBCT enrolled 58 patients to receive AAV2-NTRN bilaterally into the putamen or sham surgery. The primary endpoint was change from baseline to 12 months in the UPDRS motor score in the off state, and no significant difference was found between patients treated with AAV2-NTRN compared with control individuals. Three of 38 patients in the AAV2-NTRN group and two of 20 in the sham surgery group developed tumors, with uncertain relations to the actual treatment [91]. Postmortem analysis of two patients revealed that, unlike the animal studies, putamenal AAV-NTRN injections did not confer adequate retrograde labeling of neurons in the SN [92]. This deficiency in axonal transport of AAV-NTRN to the SN was addressed in a phase 1 safety study that enrolled six patients who received bilateral dual injections into the putamen and SN [93]. Two-year follow-up suggested that the procedures were well-tolerated and no serious adverse effects were reported. A second phase 2 RDBCT was then conducted, enrolling 51 patients to receive bilateral putamen and SN AAV-NTRN (https://clinicaltrials.gov/ct2/show/NCT00985517). In 2013, it was announced that the trial did not demonstrate statistically significant improvement in patient UPDRS scores after 15–24 months of follow-up. However, a more robust response to CERE-120 was observed in PD patients treated within 5 years of diagnosis, and no safety concerns were raised. There was a marked placebo effect as the control patients and the CERE-120 treated patients both improved significantly following surgery. Long-term observational studies of the participants are planned to assess delayed clinical effect (http://www.prnewswire.com/news-releases/ceregene-reports-data-from-parkinsons-disease-phase-2b-study-203803541.html).

4.3. Preclinical studies with other neurotrophic factors

4.3.1. Persephin/artemin

Persephin (PSPN) shows approximately 40% sequence homology to GDNF and NTRN [94]. PSPN promotes survival of cultured ventral midbrain dopaminergic neurons as well as motor neurons and prevents their degeneration after 6-OHDA toxicity [94]. PSPN-overexpressing neural stem cells grafted into the striatum prevented loss of DA neurons and led to behavioral improvements in 6-OHDA lesioned rats [95]. Artemin (ARTN) promotes survival of DA neurons in culture [96] and also protects against DA neuron degeneration in the striatum following neurotoxic doses of methamphetamine [97]. Although early preclinical studies have shown therapeutic benefit of both PSPN and ARTN, more studies are necessary before these NTFs can be tested in a clinical setting.

4.3.2. BDNF

Brain-derived neurotrophic factor (BDNF) is an essential regulator of neuronal differentiation and plasticity. It has been suggested that alterations in BDNF expression may be responsible for the development of neurodegenerative disorders [98]. Postmortem studies of PD patients have demonstrated that BDNF levels are reduced in the substantia nigra pars compacta as a result of decreased transcription of the BDNF gene [99]. Another study reported that only 10% of melanized neurons in the substantia nigra of PD patients were immunoreactive for BDNF expression, compared with 65% in healthy controls [100]. Serum BDNF levels have also been shown to correlate with a loss of striatal DA transporter binding in PD patients, suggesting an influence on striatal neurodegeneration [101]. Animal studies have demonstrated that BDNF antisense oligonucleotide infusion in rats produces a Parkinsonian phenotype [102], and BDNF knockout mice have reduced dopaminergic neurons in the substantia nigra [103]. BDNF promotes *in vitro* survival and differentiation of human and rat embryonic dopaminergic neurons, and it has protective effects against various toxins including 6-OHDA and MPTP [99]. In a nonhuman primate model of PD, intrathecal BDNF infusion resulted in milder PD symptoms and less neuronal cell loss in the substantia nigra [104]. To date, there are no clinical studies evaluating the efficacy of BDNF therapy in human PD, likely due to the logistical challenges of CNS drug delivery and dosing, as BDNF has poor blood-brain barrier penetration if administered parenterally, and intrathecal or intraventricular delivery results in poor penetration of the brain parenchyma [105].

4.3.3. CDNF/MANF

Mesencephalic astrocyte-derived neurotrophic factor (MANF) was first discovered in 2003 and was shown to be selectively neuroprotective for dopaminergic neurons [106]. Later, cerebral DA neurotrophic factor (CDNF) was discovered as a homologue of MANF with 59% sequence homology [107]. CDNF has been shown to be neuroprotective to DA neurons and intrastriatal injection of CDNF or AAV-CDNF reduces degeneration of DA neurons and parkinsonian behavior in rats and increases TH levels in the striatum and SN [107–110]. Interestingly, intranigral infusion of a combination of both CDNF and MANF via lentiviral mediated delivery reduced amphetamine-induced rotational behavior and increased striatal TH-fibers and TH-positive neurons in the substantia nigra [111]. Intranigral CDNF alone also improved behavior and increased TH fibers in the striatum, but both to a lesser extent than with CDNF/MANF together, and did not protect against TH neuronal loss in the SN [111]. Intra-nigral MANF alone did not affect behavior or striatal TH fibers, but did protect against SN neuronal loss [111]. Results of these studies suggest that combined delivery of CDNF/MANF may be more effective than single NTFs and may be a more effective potential therapeutic treatment for PD, although neither NTF has been tested in a clinical setting.

5. Conclusions

There remains a critical need for new therapies to delay or prevent the progression of PD. As discussed in this chapter, cell-based therapies may provide a promising therapeutic benefit to

PD patients. NTFs offer a potential class of cytoprotective agents that complement DA replacement and cell-based therapies in PD, with ideal grafts consisting of immunologically inert cells that continuously produce and release these agents in the host brain. Further development and refinement of these potential therapies is essential to develop personalized care for PD patients.

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References

[1] de Lau, L.M. and M.M. Breteler, Epidemiology of Parkinson's disease. Lancet Neurol, 2006. 5(6): pp. 525–35.

[2] Spillantini, M.G., et al., Alpha-synuclein in Lewy bodies. Nature, 1997. 388(6645): pp. 839–40.

[3] Damier, P., New aspects in the pathophysiology of dyskinesia. Salpetriere Deep Brain Stimulation Group. Adv Neurol, 1999. 80: pp. 611–7.

[4] Dickson, D.W., Parkinson's disease and parkinsonism: neuropathology. Cold Spring Harb Perspect Med, 2012. 2(8).

[5] Massano, J. and K.P. Bhatia, Clinical approach to Parkinson's disease: features, diagnosis, and principles of management. Cold Spring Harb Perspect Med, 2012. 2(6): p. a008870.

[6] Cotzias, G.C., M.H. Van Woert, and L.M. Schiffer, Aromatic amino acids and modification of parkinsonism. N Engl J Med, 1967. 276(7): pp. 374–9.

[7] Marsden, C.D. and J.D. Parkes, "On-off" effects in patients with Parkinson's disease on chronic levodopa therapy. Lancet, 1976. 1(7954): pp. 292–6.

[8] Lotia, M. and J. Jankovic, New and emerging medical therapies in Parkinson's disease. Expert Opin Pharmacother, 2016: pp. 1–15.

[9] Nutt, J.G. and J.H. Fellman, Pharmacokinetics of levodopa. Clin Neuropharmacol, 1984. 7(1): pp. 35–49.

[10] Parkinson Study, G., Effects of tocopherol and deprenyl on the progression of disability in early Parkinson's disease. N Engl J Med, 1993. 328(3): pp. 176–83.

[11] Stern, M.B., et al., Double-blind, randomized, controlled trial of rasagiline as monotherapy in early Parkinson's disease patients. Mov Disord, 2004. 19(8): pp. 916–23.

[12] Spiegel, E.A., H.T. Wycis, and H.W. Baird, 3rd, Long-range effects of electropallidoansotomy in extrapyramidal and convulsive disorders. Neurology, 1958. 8(10): pp. 734–40.

[13] Hassler, R. and T. Riechert, [Indications and localization of stereotactic brain operations]. Nervenarzt, 1954. 25(11): pp. 441–7.

[14] Vitek, J.L., et al., Randomized trial of pallidotomy versus medical therapy for Parkinson's disease. Ann Neurol, 2003. 53(5): pp. 558–69.

[15] Munhoz, R.P., A. Cerasa, and M.S. Okun, Surgical treatment of dyskinesia in Parkinson's disease. Front Neurol, 2014. 5: pp. 65.

[16] Deep-Brain Stimulation for Parkinson's Disease Study, G., Deep-brain stimulation of the subthalamic nucleus or the pars interna of the globus pallidus in Parkinson's disease. N Engl J Med, 2001. 345(13): pp. 956–63.

[17] Olson, L. and A. Seiger, Development and growth of immature monoamine neurons in rat and man in situ and following intraocular transplantation in the rat. Brain Res, 1973. 62(2): pp. 353–60.

[18] Freed, W.J., et al., Transplanted adrenal chromaffin cells in rat brain reduce lesion-induced rotational behaviour. Nature, 1981. 292(5821): pp. 351–2.

[19] Brundin, P., et al., The rotating 6-hydroxydopamine-lesioned mouse as a model for assessing functional effects of neuronal grafting. Brain Res, 1986. 366(1–2): pp. 346–9.

[20] Brundin, P., et al., Behavioural effects of human fetal dopamine neurons grafted in a rat model of Parkinson's disease. Exp Brain Res, 1986. 65(1): pp. 235–40.

[21] Madrazo, I., et al., 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(14): pp. 831–4.

[22] Lindvall, O., et al., Grafts of fetal dopamine neurons survive and improve motor function in Parkinson's disease. Science, 1990. 247(4942): pp. 574–7.

[23] Lindvall, O., 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(6): pp. 615–31.

[24] Lindvall, O., 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(2): pp. 155–65.

[25] Kefalopoulou, Z., et al., Long-term clinical outcome of fetal cell transplantation for Parkinson disease: two case reports. JAMA Neurol, 2014. 71(1): pp. 83–7.

[26] Freed, C.R., 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(22): pp. 1549–55.

[27] Freed, C.R., et al., Transplantation of human fetal dopamine cells for Parkinson's disease. Results at 1 year. Arch Neurol, 1990. 47(5): pp. 505–12.

[28] Spencer, D.D., 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(22): pp. 1541–8.

[29] Peschanski, M., et al., Bilateral motor improvement and alteration of L-dopa effect in two patients with Parkinson's disease following intrastriatal transplantation of foetal ventral mesencephalon. Brain, 1994. 117 (Pt 3): pp. 487–99.

[30] Widner, H., 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(22): pp. 1556–63.

[31] Freeman, T.B., et al., Bilateral fetal nigral transplantation into the postcommissural putamen in Parkinson's disease. Ann Neurol, 1995. 38(3): pp. 379–88.

[32] Olanow, C.W., J.H. Kordower, and T.B. Freeman, Fetal nigral transplantation as a therapy for Parkinson's disease. Trends Neurosci, 1996. 19(3): pp. 102–9.

[33] Olanow, C.W., et al., A double-blind controlled trial of bilateral fetal nigral transplantation in Parkinson's disease. Ann Neurol, 2003. 54(3): pp. 403–14.

[34] Ma, Y., et al., Dopamine cell implantation in Parkinson's disease: long-term clinical and (18)F-FDOPA PET outcomes. J Nucl Med, 2010. 51(1): pp. 7–15.

[35] Freed, C.R., et al., Transplantation of embryonic dopamine neurons for severe Parkinson's disease. N Engl J Med, 2001. 344(10): pp. 710–9.

[36] Ribeiro, D., et al., Efficient expansion and dopaminergic differentiation of human fetal ventral midbrain neural stem cells by midbrain morphogens. Neurobiol Dis, 2013. 49: pp. 118–27.

[37] Thomson, J.A., et al., Embryonic stem cell lines derived from human blastocysts. Science, 1998. 282(5391): pp. 1145–7.

[38] Barberi, T., et al., Neural subtype specification of fertilization and nuclear transfer embryonic stem cells and application in parkinsonian mice. Nat Biotechnol, 2003. 21(10): pp. 1200–7.

[39] Roy, N.S., et al., Functional engraftment of human ES cell-derived dopaminergic neurons enriched by coculture with telomerase-immortalized midbrain astrocytes. Nat Med, 2006. 12(11): pp. 1259–68.

[40] Takagi, Y., et al., Dopaminergic neurons generated from monkey embryonic stem cells function in a Parkinson primate model. J Clin Invest, 2005. 115(1): pp. 102–9.

[41] Cooper, O., M. Parmar, and O. Isacson, Characterization and criteria of embryonic stem and induced pluripotent stem cells for a dopamine replacement therapy. Prog Brain Res, 2012. 200: pp. 265–76.

[42] Glavaski-Joksimovic, A. and M.C. Bohn, Mesenchymal stem cells and neuroregeneration in Parkinson's disease. Exp Neurol, 2013. 247: pp. 25–38.

[43] Offen, D., et al., Intrastriatal transplantation of mouse bone marrow-derived stem cells improves motor behavior in a mouse model of Parkinson's disease. J Neural Transm Suppl, 2007(72): pp. 133–43.

[44] Venkataramana, N.K., et al., Open-labeled study of unilateral autologous bone-marrow-derived mesenchymal stem cell transplantation in Parkinson's disease. Transl Res, 2010. 155(2): pp. 62–70.

[45] Takahashi, K. and S. Yamanaka, Induction of pluripotent stem cells from mouse embryonic and adult fibroblast cultures by defined factors. Cell, 2006. 126(4): pp. 663–76.

[46] Takahashi, K., et al., Induction of pluripotent stem cells from adult human fibroblasts by defined factors. Cell, 2007. 131(5): pp. 861–72.

[47] Kuzmenkin, A., et al., Functional characterization of cardiomyocytes derived from murine induced pluripotent stem cells in vitro. FASEB J, 2009. 23(12): pp. 4168–80.

[48] Espejel, S., et al., Induced pluripotent stem cell-derived hepatocytes have the functional and proliferative capabilities needed for liver regeneration in mice. J Clin Invest, 2010. 120(9): pp. 3120–6.

[49] Czepiel, M., et al., Differentiation of induced pluripotent stem cells into functional oligodendrocytes. Glia, 2011. 59(6): pp. 882–92.

[50] Hu, B.Y., et al., Neural differentiation of human induced pluripotent stem cells follows developmental principles but with variable potency. Proc Natl Acad Sci U S A, 2010. 107(9): pp. 4335–40.

[51] Wernig, M., et al., Neurons derived from reprogrammed fibroblasts functionally integrate into the fetal brain and improve symptoms of rats with Parkinson's disease. Proc Natl Acad Sci U S A, 2008. 105(15): pp. 5856–61.

[52] Hargus, G., et al., Differentiated Parkinson patient-derived induced pluripotent stem cells grow in the adult rodent brain and reduce motor asymmetry in Parkinsonian rats. Proc Natl Acad Sci U S A, 2010. 107(36): pp. 15921–6.

[53] Swistowski, A., et al., Efficient generation of functional dopaminergic neurons from human induced pluripotent stem cells under defined conditions. Stem Cells, 2010. 28(10): pp. 1893–904.

[54] Hallett, P.J., et al., Successful function of autologous iPSC-derived dopamine neurons following transplantation in a non-human primate model of Parkinson's disease. Cell Stem Cell, 2015. 16(3): pp. 269–74.

[55] Momcilovic, O., et al., Genome wide profiling of dopaminergic neurons derived from human embryonic and induced pluripotent stem cells. Stem Cells Dev, 2014. 23(4): pp. 406–20.

[56] Reardon, S. and D. Cyranoski, Japan stem-cell trial stirs envy. Nature, 2014. 513(7518): pp. 287–8.

[57] Garber, K., RIKEN suspends first clinical trial involving induced pluripotent stem cells. Nat Biotechnol, 2015. 33(9): pp. 890–1.

[58] Pang, Z.P., et al., Induction of human neuronal cells by defined transcription factors. Nature, 2011. 476(7359): pp. 220–3.

[59] Pfisterer, U., et al., Direct conversion of human fibroblasts to dopaminergic neurons. Proc Natl Acad Sci U S A, 2011. 108(25): pp. 10343–8.

[60] Chanda, S., et al., Generation of induced neuronal cells by the single reprogramming factor ASCL1. Stem Cell Rep, 2014. 3(2): pp. 282–96.

[61] Xu, H., et al., Neurotrophic factor expression in expandable cell populations from brain samples in living patients with Parkinson's disease. FASEB J, 2013. 27(10): pp. 4157–68.

[62] Rangasamy, S.B., et al., Neurotrophic factor therapy for Parkinson's disease. Prog Brain Res, 2010. 184: pp. 237–64.

[63] Kordower, J.H. and A. Bjorklund, Trophic factor gene therapy for Parkinson's disease. Mov Disord, 2013. 28(1): pp. 96–109.

[64] Rodrigues, T.M., et al., Challenges and promises in the development of neurotrophic factor-based therapies for Parkinson's disease. Drugs Aging, 2014. 31(4): pp. 239–61.

[65] Airaksinen, M.S. and M. Saarma, The GDNF family: signalling, biological functions and therapeutic value. Nat Rev Neurosci, 2002. 3(5): pp. 383–94.

[66] Lin, L.F., et al., GDNF: a glial cell line-derived neurotrophic factor for midbrain dopaminergic neurons. Science, 1993. 260(5111): pp. 1130–2.

[67] Meyer, M., et al., Improved survival of embryonic porcine dopaminergic neurons in coculture with a conditionally immortalized GDNF-producing hippocampal cell line. Exp Neurol, 2000. 164(1): pp. 82–93.

[68] Beck, K.D., et al., GDNF induces a dystonia-like state in neonatal rats and stimulates dopamine and serotonin synthesis. Neuron, 1996. 16(3): pp. 665–73.

[69] Bowenkamp, K.E., et al., Glial cell line-derived neurotrophic factor supports survival of injured midbrain dopaminergic neurons. J Comp Neurol, 1995. 355(4): pp. 479–89.

[70] Winkler, C., et al., Short-term GDNF treatment provides long-term rescue of lesioned nigral dopaminergic neurons in a rat model of Parkinson's disease. J Neurosci, 1996. 16(22): pp. 7206–15.

[71] Clarkson, E.D., W.M. Zawada, and C.R. Freed, GDNF improves survival and reduces apoptosis in human embryonic dopaminergic neurons in vitro. Cell Tissue Res, 1997. 289(2): pp. 207–10.

[72] Schober, A., et al., GDNF applied to the MPTP-lesioned nigrostriatal system requires TGF-beta for its neuroprotective action. Neurobiol Dis, 2007. 25(2): pp. 378–91.

[73] Chen, Y.H., et al., MPTP-induced deficits in striatal synaptic plasticity are prevented by glial cell line-derived neurotrophic factor expressed via an adeno-associated viral vector. FASEB J, 2008. 22(1): pp. 261–75.

[74] Gash, D.M., et al., Functional recovery in parkinsonian monkeys treated with GDNF. Nature, 1996. 380(6571): pp. 252–5.

[75] Aron, L. and R. Klein, Repairing the parkinsonian brain with neurotrophic factors. Trends Neurosci, 2011. 34(2): pp. 88–100.

[76] Nutt, J.G., et al., Randomized, double-blind trial of glial cell line-derived neurotrophic factor (GDNF) in PD. Neurology, 2003. 60(1): pp. 69–73.

[77] Gill, S.S., et al., Direct brain infusion of glial cell line-derived neurotrophic factor in Parkinson disease. Nat Med, 2003. 9(5): pp. 589–95.

[78] Patel, N.K., et al., Intraputamenal infusion of glial cell line-derived neurotrophic factor in PD: a two-year outcome study. Ann Neurol, 2005. 57(2): pp. 298–302.

[79] Patel, N.K., et al., Benefits of putaminal GDNF infusion in Parkinson disease are maintained after GDNF cessation. Neurology, 2013. 81(13): pp. 1176–8.

[80] Slevin, J.T., et al., Improvement of bilateral motor functions in patients with Parkinson disease through the unilateral intraputaminal infusion of glial cell line-derived neurotrophic factor. J Neurosurg, 2005. 102(2):pp. 216–22.

[81] Lang, A.E., et al., Randomized controlled trial of intraputamenal glial cell line-derived neurotrophic factor infusion in Parkinson disease. Ann Neurol, 2006. 59(3): pp. 459–66.

[82] Kotzbauer, P.T., et al., Neurturin, a relative of glial-cell-line-derived neurotrophic factor. Nature, 1996. 384(6608): pp. 467–70.

[83] Horger, B.A., et al., Neurturin exerts potent actions on survival and function of midbrain dopaminergic neurons. J Neurosci, 1998. 18(13): pp. 4929–37.

[84] Akerud, P., et al., Differential effects of glial cell line-derived neurotrophic factor and neurturin on developing and adult substantia nigra dopaminergic neurons. J Neurochem, 1999. 73(1): pp. 70–8.

[85] Oiwa, Y., et al., Dopaminergic neuroprotection and regeneration by neurturin assessed by using behavioral, biochemical and histochemical measurements in a model of progressive Parkinson's disease. Brain Res, 2002. 947(2): pp. 271–83.

[86] Grondin, R., et al., Intraputamenal infusion of exogenous neurturin protein restores motor and dopaminergic function in the globus pallidus of MPTP-lesioned rhesus monkeys. Cell Transplant, 2008. 17(4): pp. 373–81.

[87] Gasmi, M., et al., AAV2-mediated delivery of human neurturin to the rat nigrostriatal system: long-term efficacy and tolerability of CERE-120 for Parkinson's disease. Neurobiol Dis, 2007. 27(1): pp. 67–76.

[88] Kordower, J.H., et al., Delivery of neurturin by AAV2 (CERE-120)-mediated gene transfer provides structural and functional neuroprotection and neurorestoration in MPTP-treated monkeys. Ann Neurol, 2006. 60(6): pp. 706–15.

[89] Herzog, C.D., et al., Expression, bioactivity, and safety 1 year after adeno-associated viral vector type 2-mediated delivery of neurturin to the monkey nigrostriatal system support cere-120 for Parkinson's disease. Neurosurgery, 2009. 64(4): pp. 602–12; discussion 612–3.

[90] Marks, W.J., Jr., et al., Safety and tolerability of intraputaminal delivery of CERE-120 (adeno-associated virus serotype 2-neurturin) to patients with idiopathic Parkinson's disease: an open-label, phase I trial. Lancet Neurol, 2008. 7(5): pp. 400–8.

[91] Marks, W.J., Jr., et al., Gene delivery of AAV2-neurturin for Parkinson's disease: a double-blind, randomised, controlled trial. Lancet Neurol, 2010. 9(12): pp. 1164–72.

[92] Bartus, R.T., et al., Bioactivity of AAV2-neurturin gene therapy (CERE-120): differences between Parkinson's disease and nonhuman primate brains. Mov Disord, 2011. 26(1): pp. 27–36.

[93] Bartus, R.T., et al., Safety/feasibility of targeting the substantia nigra with AAV2-neurturin in Parkinson patients. Neurology, 2013. 80(18): pp. 1698–701.

[94] Milbrandt, J., et al., Persephin, a novel neurotrophic factor related to GDNF and neurturin. Neuron, 1998. 20(2): pp. 245–53.

[95] Akerud, P., et al., Persephin-overexpressing neural stem cells regulate the function of nigral dopaminergic neurons and prevent their degeneration in a model of Parkinson's disease. Mol Cell Neurosci, 2002. 21(2): pp. 205–22.

[96] Baloh, R.H., et al., Artemin, a novel member of the GDNF ligand family, supports peripheral and central neurons and signals through the GFRalpha3-RET receptor complex. Neuron, 1998. 21(6): pp. 1291–302.

[97] Cass, W.A., et al., Protection by GDNF and other trophic factors against the dopamine-depleting effects of neurotoxic doses of methamphetamine. Ann N Y Acad Sci, 2006. 1074: pp. 272–81.

[98] Zuccato, C. and E. Cattaneo, Brain-derived neurotrophic factor in neurodegenerative diseases. Nat Rev Neurol, 2009. 5(6): pp. 311–22.

[99] Murer, M.G., Q. Yan, and R. Raisman-Vozari, Brain-derived neurotrophic factor in the control human brain, and in Alzheimer's disease and Parkinson's disease. Prog Neurobiol, 2001. 63(1): pp. 71–124.

[100] Parain, K., et al., Reduced expression of brain-derived neurotrophic factor protein in Parkinson's disease substantia nigra. Neuroreport, 1999. 10(3): pp. 557–61.

[101] Ziebell, M., et al., Striatal dopamine transporter binding correlates with serum BDNF levels in patients with striatal dopaminergic neurodegeneration. Neurobiol Aging, 2012. 33(2): pp. 428 e1–5.

[102] Porritt, M.J., P.E. Batchelor, and D.W. Howells, Inhibiting BDNF expression by antisense oligonucleotide infusion causes loss of nigral dopaminergic neurons. Exp Neurol, 2005. 192(1): pp. 226–34.

[103] Baquet, Z.C., P.C. Bickford, and K.R. Jones, Brain-derived neurotrophic factor is required for the establishment of the proper number of dopaminergic neurons in the substantia nigra pars compacta. J Neurosci, 2005. 25(26): pp. 6251–9.

[104] Tsukahara, T., et al., Effects of brain-derived neurotrophic factor on 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine-induced parkinsonism in monkeys. Neurosurgery, 1995. 37(4): pp. 733–9; discussion 739–41.

[105] Nagahara, A.H. and M.H. Tuszynski, Potential therapeutic uses of BDNF in neurological and psychiatric disorders. Nat Rev Drug Discov, 2011. 10(3): pp. 209–19.

[106] Petrova, P., et al., MANF: a new mesencephalic, astrocyte-derived neurotrophic factor with selectivity for dopaminergic neurons. J Mol Neurosci, 2003. 20(2): pp. 173–88.

[107] Lindholm, P., et al., Novel neurotrophic factor CDNF protects and rescues midbrain dopamine neurons in vivo. Nature, 2007. 448(7149): pp. 73–7.

[108] Voutilainen, M.H., et al., Chronic infusion of CDNF prevents 6-OHDA-induced deficits in a rat model of Parkinson's disease. Exp Neurol, 2011. 228(1): pp. 99–108.

[109] Back, S., et al., Gene therapy with AAV2-CDNF provides functional benefits in a rat model of Parkinson's disease. Brain Behav, 2013. 3(2): pp. 75–88.

[110] Ren, X., et al., AAV2-mediated striatum delivery of human CDNF prevents the deterioration of midbrain dopamine neurons in a 6-hydroxydopamine induced parkinsonian rat model. Exp Neurol, 2013. 248: pp. 148–56.

[111] Cordero-Llana, O., et al., Enhanced efficacy of the CDNF/MANF family by combined intranigral overexpression in the 6-OHDA rat model of Parkinson's disease. Mol Ther, 2015. 23(2): pp. 244–54.

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