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PERSPECTIVE

Localized striatal delivery of GDNF as a treatment for Parkinson disease

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Ten years ago, a glial cell line–derived neurotrophic factor (GDNF) that has prominent actions on nigral dopaminergic neurons, both in vitro and in animal models of Parkinson disease (PD), was discovered. A recently published open-label clinical trial now reports that long-term intracerebral delivery of GDNF may also markedly improve symptoms in patients with PD. Here we review the experimental data underlying the current clinical trial and discuss the types of structural and functional changes induced by GDNF that may provide symptomatic benefit in PD patients. Data obtained in rodent and primate models of PD highlight the importance of how and where the factor is administered, supporting the view that GDNF has to be delivered locally in the brain parenchyma, at the receptive target site, to provide therapeutic benefit in PD patients.

The cardinal symptoms of PD, including a difficulty in initiating movement, slowness of movement and stiffness and shaking at rest, are to a large extent caused by the progressive degeneration of the dopamine-producing neurons in the substantia nigra. Nigral cell loss proceeds over many years, during the early symptomatic stage, during manifest PD and during severe, end-stage disease. At the onset of disease, about 50% of dopaminergic neurons have been lost, and there is on average a further loss of 45% within the first decade, accompanied by a profound striatal dopaminergic denervation. It is this slow and protracted degenerative process that creates opportunities for disease intervention, such as blocking nigral cell loss and promoting recovery by improved function—and possibly by inducing regeneration and sprouting—of the surviving nigral dopaminergic neurons. Results obtained in animal models of PD indicate that GDNF may possess the desired properties to be used as a disease-modifying therapeutic factor for PD.

Neurotrophic factors, by virtue of their neuroprotective properties, have attracted considerable interest as potential therapeutic agents in neurodegenerative diseases. Attempts to apply these factors clinically, however, have so far been disappointing because of their poor efficacy and induction of troublesome side effects. In these clinical trials, the recombinant protein was delivered either systemically or into the cerebrospinal fluid (intraventricularly or intrathecally) in patients suffering from amyotrophic lateral sclerosis, peripheral neuropathy, PD or Alzheimer disease[1,2]. Results from these studies indicate that the neurotrophic factors, whose receptors are widely distributed, are prone to inducing pronounced side effects when delivered by these routes. The poor penetration across the blood–brain barrier, as well as the limited passage of proteins from the cerebrospinal fluid into the brain tissue, has made it necessary to administer the factors at doses that are likely to induce side effects. These effects may not be so evident in small-sized experimental animals. For this reason, they may have gone unnoticed in the preclinical studies and may have become apparent in some cases only at the phase II/III stage of the clinical trails when larger numbers of patients were included.

The therapeutic value of neurotrophic-factor delivery, therefore, may not be possible to achieve unless the factors are delivered locally at the receptive target sites within the central nervous system. Steven Gill and collaborators[3] have, for the first time, tested this mode of delivery in patients with advanced PD using continuous intracerebral infusion of GDNF. Although quite promising, the results of this initial open-label trial should be interpreted cautiously because the study was based on a small number of patients who were monitored over a relatively short follow-up period. Nevertheless, the data reported indicate that pronounced clinical benefit, in the absence of any serious side effects, may be possible to obtain by GDNF using intrastriatal delivery.

Neuroprotective effects of GDNF in animal models of PD

In rodents, where dopaminergic neuron degeneration is induced by either MPTP (in mice) or 6-hydroxydopamine (6-OHDA; in rats), it is clear that GDNF can be both neuroprotective and restorative. Depending on the design of the experiment, both effects can contribute to the final outcome. GDNF can protect quite effectively against an acute insult induced by 6-OHDA, provided that the factor is given around the time of toxin injection and that it is administered at the site of the toxic insult, that is in the substantia nigra in animals given 6-OHDA in or near the substantia nigra[4,5] and in the striatum in animals with 6-OHDA injections in the striatum[6,7].

GDNF that is delivered into the substantia nigra is unable to protect the axons and axon terminals in the striatum against a 6-OHDA injection in the striatum but is highly efficient in protecting the nigral cell bodies from the retrograde cell death induced by the intrastriatal lesion[8]. Intrastriatal 6-OHDA injections cause a loss of about 50–80% of the nigral dopamine neurons, depending on the dose and the number of injection sites within the striatum. The toxin induces an acute lesion of the axon terminal followed by retrograde degeneration with atrophy of the cell body and cell death[9,10]. The cellular degeneration in the nigra is progressive and takes place within the first 3–4 weeks after the injection of the toxin (**Fig. 1b**)[9]. In this model, GDNF is able to promote survival of the nigral dopamine cells after repeated injections into the midbrain (over the cell bodies; **Fig. 1c**), at the level of

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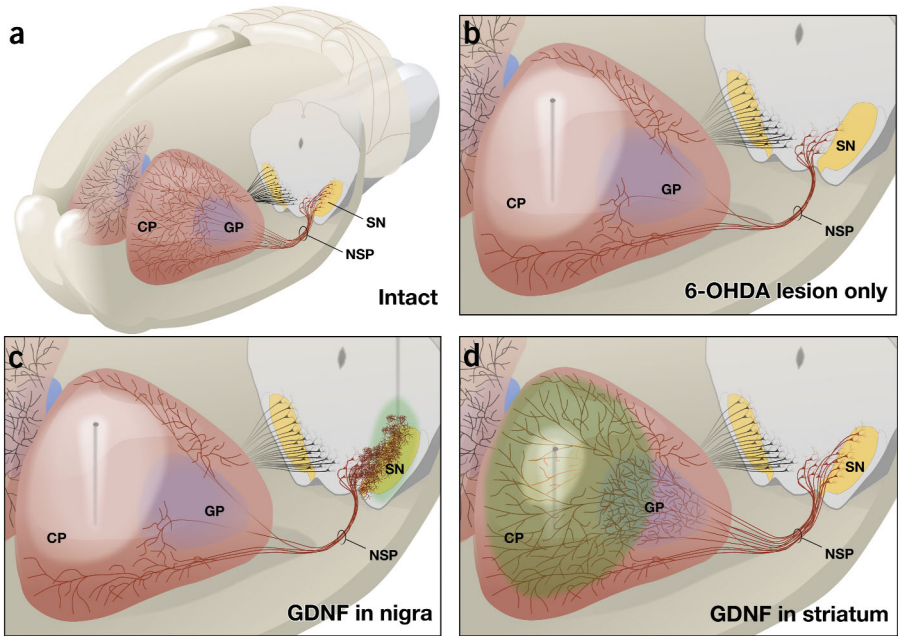

Figure 1 Protective effects of GDNF in the intrastriatal 6-OHDA model. (**a**) In the intact brain, the dopamine-producing cells reside in the substantia nigra (SN), which is located in the ventral midbrain, and send projections (the nigrostriatal projection, NSP) to the striatum, the input structure to the basal ganglia, which is situated beneath the cortex in the forebrain. The pre-terminal axons course in the medial forebrain bundle and pass through the globus pallidus (GP) before entering the target nuclei, where terminal arborizations and synaptic contacts with striatal neurons are seen. CP, caudate putamen. (**b**) Injection of 6-OHDA in the striatum, as indicated by the cannula track, leads to partial depletion of the terminals followed by retrograde degeneration of the neurons in the substantia nigra. (**c**) If GDNF is applied close to the cell bodies (shown as green halo around the cannula tip), the injured neurons can be protected from degeneration very efficiently. The degeneration of the terminals, however, is not prevented. Instead, fiber sprouting occurs mainly in the substantia nigra, the site of GDNF delivery. (**d**) Striatal delivery of GDNF, on the other hand, not only provides protection of the tyrosine hydroxylase–positive cell bodies but also preserves the fibers in the striatum. In this case, fiber sprouting occurs mainly within the striatum and globus pallidus, that is, within the area of high GDNF expression.

the axon terminals in the striatum (**Fig. 1d**) or through the lateral ventricular route. Once rescued, the nigral cells survive for months after the withdrawal of GDNF, indicating that they may not be dependent on GDNF for further survival[8].

Tests of spontaneous and drug-induced motor behavior in rats that were subjected to intrastriatal 6-OHDA lesions stress the importance of the GDNF application. Protection of the nigral dopamine cell bodies alone, in the absence of axons projecting to the striatum, as seen after injection of GDNF into the substantia nigra or the lateral ventricle, is insufficient to provide either protection against or recovery from lesion-induced motor deficits[11]. When GDNF is applied in the striatum at the time of lesion, by contrast, the entire nigrostriatal pathway is preserved and the animals retain their performance in both drug-induced and spontaneous motor tests[7]. Other studies, performed *in vitro*, indicate that neurotrophic factors may elicit different responses with respect to axonal growth and survival when applied locally at the axon or cell body[12]. It seems possible, therefore, that the nigral dopaminergic neurons may respond differently to GDNF when it is applied at their cell bodies or close to their axons within the striatum.

Restoration of function

The functional effects of GDNF have been explored after single or repeated bolus injections or continuous infusions of the recombinant protein, given either intracerebroventricularly (ICV) or directly into the brain parenchyma. In intact young or aged animals, single injections of GDNF into either substantia nigra or striatum (in doses of 1–10 μg) stimulate the function of dopaminergic neurons, as determined by an increased dopamine turnover in substantia nigra and/or striatum and by an increased release of dopamine and its metabolites in the striatum[13–18]. Of note, the stimulatory effect of intranigral GDNF is observed in both substantia nigra and striatum, whereas the effect of intrastriatal GDNF seems to be largely restricted to the striatum with no or limited effects on nigral dopamine turnover[16,18]. Single bolus injections of GDNF into the ventricular space may induce similar effects on dopamine turnover, but they are seen only at considerably higher doses (30–100 μg) and seem to be confined mostly to the substantia nigra[19].

These neurochemical changes are fully developed by 1–3 weeks after GDNF administration and may still be detectable after 6 weeks[16,19]. They are accompanied by small but significant increases in both spontaneous and amphetamine-induced locomotor activity. The changes in motor behavior, however, seem to be earlier in onset—they can be detected as early as 3 h after GDNF injection[20]—and do not last beyond the first week[13,16,20]. The results are different when GDNF is administered by continuous infusion. In a series of recent studies in primates, Gerhardt, Gash and colleagues[21–23] have studied the effect of GDNF that was infused over a 2-month period by programmable subcutaneous pumps. The factor was delivered ICV or directly into the putamen (unilaterally) at a dose of 7.5–22.5 μg/day in aged monkeys that showed an age-dependent decline in motor function (**Table 1**). Delivered in this way, GDNF induced an improvement in motor function that developed gradually over 2–6 weeks and was maintained during the 2-month wash-out period when GDNF was replaced with vehicle. Increased baseline and evoked dopamine release, as monitored by microdialysis, was observed bilaterally in substantia nigra, putamen and caudate nucleus in the ICV group[22]. Increases in dopamine and dopamine metabolites in tissues were observed, most prominently on the injected side, in putamen, caudate nucleus and globus pallidus in the intraputaminal group (substantia nigra was not included in this analysis)[21], indicating that a sustained, general upregulation of dopaminergic neuron function may be induced by either route of administration.

Similar functional changes, at both the neurochemical and behavioral level, have been observed in MPTP-lesioned monkeys in which GDNF was administered with a delay of 1–3 months, at a time when neurodegeneration is largely complete and the animals have reached a stable symptomatic stage (**Table 1**). In these experiments, GDNF was delivered either as single or repeated bolus doses of up to 1,000 μg intraventricularly[24–28] or by continuous ICV or by intraputaminal delivery at 7.5–22.5 μg/day[29]. In these severely lesioned animals, however, where less than 20% of the nigral dopaminergic neurons remain

Table 1 Effects of GDNF in intact or MPTP-lesioned primates

	GDNF delivery				Histological quantification[b]			Biochemical analysis				
	Refs.	Mode	Site	Dose[c]	SN cell number	SN cell size	Fiber density		DA	DOPAC	HVA	Functional assessment
MPTP lesion	24–28, 49	Bolus injection	ICV	10–1,000 μg	↑	↑	↑ SN	SN	↑	↑	↑	Stimulus-induced DA release in SN
								GP	↑		↑	Improvement in primate PD rating scale
								Put	↑		↑	Reduction in L-DOPA-induced dyskinesias
	29	Long-term protein infusion	ICV Put	7.5–22.5 μg/day	↑		↑ CN & Put	CN	↑	↑	↑	Improvement in primate PD rating scale
								GP	↑	↑	↑	
	45	rLV-GDNF gene delivery	Put & SN	2.3–3.5 ng/mg protein	↑	↑	↑					Protection of ^{18}F-DOPA uptake Improvement in primate PD rating scale and hand-reaching task
Young intact	14	Bolus injection	SN	150 μg		↑	↑ SN	SN	↑		↑	Small increases in daytime activity
								CN				
Aged intact	21–23	Long-term protein infusion	ICV Put	7.5– 22.5 μg/day	↑	↑	↑ GP, CN & Put	CN	↑	↑	↑	Baseline DA release in SN
								Put			↑	Stimulus-induced DA release in SN, CN and Put Improvement in general motor performance (only at 22.5 μg/day)
								GP	↑		↑	Increased hand movement speed
	45	rLV-GDNF gene delivery	Put & SN	2.3–3.5 ng/mg protein	↑	↑	↑ CN & Put	Put	↑		↑	Increased ^{18}F-DOPA uptake

Abbreviations: CN, caudate nucleus; DA, dopamine; DOPAC, dihydroxyphenylacetic acid; GP, globus pallidus; HVA, homovanillic acid; ICV, intracerebroventricular; Put, putamen; SN, substantia nigra.

[a]Arrows indicate increases in various measures of dopaminergic parameters in animals treated with GDNF as compared with control groups. [b]Histological evaluations were performed on sections stained with antibodies against tyrosine hydroxylase, the rate-limiting enzyme in dopamine synthesis. [c]ng/mg protein refers to the GDNF levels expressed from the rLV-GDNF vector in the caudate-putamen, as determined by ELISA on tissue samples.

and striatal dopamine amounts are reduced by more than 97%[24,27,29], the most prominent neurochemical changes were observed in downstream striatal targets, such as the globus pallidus[29] or the nigra itself[24,27]. Similar results have been obtained in rats with near-complete lesions of the nigrostriatal pathway that were induced by 6-OHDA[30–32]. Thus, GDNF-induced functional changes obtained in animals with advanced parkinsonism that was induced by MPTP or 6-OHDA are primarily mediated by increased dopamine turnover and/or release in striatal output structures. This occurs, above all, in substantia nigra and globus pallidus, and the sparse remaining striatal dopaminergic innervation has a minor effect in this case. In animals with partial lesions of the nigrostriatal pathway, where part of the striatal dopaminergic innervation is spared, significant recovery of motor function was obtained not only when GDNF was delivered intraventricularly but also after it was administered directly into the partly denervated striatum[33]. In less advanced cases, therefore, it seems likely that a functional GDNF response may be mediated by the spared terminals in the striatum.

The ability of GDNF to stimulate nigrostriatal function in intact and lesioned animals may, at least in part, reflect a direct action of GDNF on the function of dopaminergic neurons. These effects, as observed in *in vitro* studies, include increases in the spontaneous firing rate and the quantal size of terminal dopamine release[34], as well as an increased excitability of the dopaminergic neurons that is mediated by A-type K^+ channels[35] and high voltage–activated Ca^{2+} channels[36].

GDNF-induced axonal sprouting

The extent to which regeneration or axonal sprouting contributes to the functional recovery seen after infusion of recombinant GDNF protein in animals lesioned with MPTP or 6-OHDA is still unresolved. In rats with partial lesions from 6-OHDA, the recovered motor function persists for at least 6 weeks after a 4-week ICV GDNF infusion[3]. Nevertheless, no signs of sprouting or increased striatal tyrosine hydroxylase innervation density were observed in these animals. Grondin and colleagues[29], by contrast, observed an increased density of tyrosine hydroxylase–positive fibers in the lesioned striatum, close to the ventricle, after infusion of GDNF into the putamen for 12 weeks in MPTP-lesioned monkeys. Rosenblad and colleagues[10,37] have observed similar signs of axonal sprouting in the striatum in rats with partial lesions of the nigrostriatal pathway induced by 6-OHDA. In these experiments, GDNF was administered by repeated injections into the striatum for 3 weeks, starting 3–4 weeks after toxin injection. In one of these studies[37], this apparent sprouting was prominent and was associated with improved motor function in the GDNF-treated animals. Whether this was due to growth of new axons and axon terminals, rather than functional upregulation in spared but dysfunctional fibers, is unclear.

The most clear-cut evidence for growth of new axons comes from the observation of aberrant sprouting induced by repeated injections or continuous infusions of GDNF in rats lesioned with 6-OHDA (**Fig. 1**). This occurs most prominently near the site of GDNF administration, that is, in globus pallidus in rats given GDNF in the striatum (**Fig. 1d**) and in and close to the substantia nigra in animals where GDNF is injected close to the nigra (**Fig. 1c**)[7,11]. Whether this growth response is beneficial or detrimental to functional recovery is not clear. Because nigra and pallidum are both normal sites for dopamine release and contain functional dopamine receptors (**Fig. 1a**), there is every reason to believe that dopamine release from these aberrant terminals may be functional and thus may be involved in the functional response induced by continuous local infusions of GDNF.

Long-term delivery of GDNF

Continuous delivery of recombinant GDNF protein has potential problems, including complications associated with the chronically implanted infusion device[3,38]. This has prompted researchers to seek alternative ways of protein delivery (see **Box 1**). Among the alternatives pursued today, replication-deficient viral vectors (*in vivo* gene transfer) is one of the most likely strategies to be applied clinically in the near

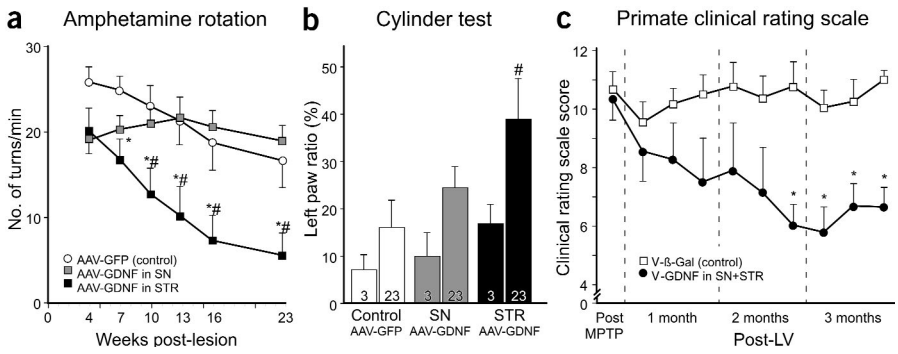

Figure 2 Summary of behavioral recovery documented in two experiments using the recombinant AAV and LV vectors in rodent and primate models of PD, respectively. (**a**,**b**) In the study of Kirik and colleagues[42], the rAAV-GDNF or rAAV-GFP (control group) vectors were injected into the striatum (STR) or the substantia nigra (SN) before an intrastriatal 6-OHDA injection. After the lesion, all animals were initially found to be severely impaired in the drug-induced and spontaneous behavioral tests at 1–4 weeks, indicating that the initial impact of the 6-OHDA lesion was the same in all experimental groups. Over the following weeks, significant recovery was observed in the striatal GDNF injection group leading to a progressive decline in the amphetamine rotation test between 7 and 16 weeks after the lesion, as shown in **a**, whereas the nigral GDNF group did not differ from the lesion controls. In addition, the animals were evaluated in the cylinder test at 3 and 23 weeks, as shown in **b**, providing a lateralized measure of individual forelimb use in a simple motor task before and after recovery of amphetamine rotation. This second independent observation supported the findings using the amphetamine test as a measure of striatal dopamine release and showed that striatal but not nigral delivery of GDNF provided significant recovery in the cylinder test performance. (**c**) In the study of Kordower and colleagues[45], the motor disability of the MPTP-treated monkeys was assessed using a modified primate clinical rating scale. After the baseline evaluation, the animals were divided into two groups to receive either the LV-GDNF or the LV-β-Gal control vector and were followed with repeated testing for an additional 3 months. During the course of the follow-up period, a reduction in the total disability was observed in the monkeys injected with LV-GDNF but not in those injected with the control vector. $*P < 0.05$ compared with control; $\#P < 0.05$ compared with the 3-week time point.

future. Three vector systems, adenovirus[39,40], adeno-associated virus (rAAV)[41,42] and lentivirus (rLV)[43–46], have been successfully applied for this purpose. The rAAV and rLV vectors are particularly promising in that they allow long-term expression of GDNF in the brain in the absence of any detectable cellular pathology or immune reactions. GDNF that is secreted locally from the transduced brain cells has been shown to provide near-complete protection of nigral dopamine cells from toxic insults[47], suggesting that viral vector delivery is as good as, if not better than, direct protein injections. This protection occurs even though the amount of GDNF that is produced locally from the transduced cells is at least three orders of magnitude less than the amount of protein commonly used for intracerebral injections[47].

The rAAV-GDNF vector has been used to overexpress GDNF in the striatum or substantia nigra in rats before an intrastriatal lesion was created with 6-OHDA[42]. Both sites of delivery were equally effective in protecting the nigral tyrosine hydroxylase–positive cell bodies from the toxic damage. Partial sparing of the nigrostriatal axons and regeneration of tyrosine hydroxylase–positive axons and terminals were seen, however, only after striatal delivery. Substantial functional recovery both in drug-induced and spontaneous motor behavior was observed in the striatal, but not the nigral, delivery group (**Fig. 2a,b**), indicating that maintenance of a functional nigrostriatal projection may be critical. The slow and protracted development of the functional effect (**Fig. 2a,b**) strongly indicates that GDNF-induced axonal sprouting may have been an important contributor to the functional recovery in this case.

In the study of Kordower and colleagues[45], rLV-GDNF vector was injected into striatum and substantia nigra either in aged monkeys or in monkeys subjected to an MPTP lesion 1 week before vector injection. In the aged animals, overexpression of GDNF induced not only an enhancement of ^{18}F-DOPA uptake in positron-emission tomography (PET) analysis (+27% as compared with the contralateral side) but also an increase in tyrosine hydroxylase–positive fiber density and tissue dopamine levels in the striatum and in tyrosine hydroxylase mRNA and tyrosine hydroxylase–positive cell counts in the substantia nigra. In the MPTP-lesioned monkeys, those injected with rLV-GDNF recovered gradually over the 3-month observation period, whereas the control group remained severely impaired (**Fig. 2c**). The profound reductions in nigral tyrosine hydroxylase–positive cell numbers (–90%), tyrosine hydroxylase–positive striatal fiber density (–70 to –75%) and striatal ^{18}F-DOPA uptake (–80%) were completely blocked or markedly attenuated in the monkeys treated with rLV-GDNF. In a similar study that used the rLV-GDNF vector to overexpress GDNF in the striatum in the rat 6-OHDA lesion model, we have obtained marked protection of the nigral dopaminergic neurons, as well as preservation of the nigrostriatal projection, accompanied by extensive sprouting of the tyrosine hydroxylase–positive fibers in the globus pallidus, entopeduncular nucleus and substantia nigra[46]. In this case, however, the GDNF-treated animals remained functionally impaired and tyrosine hydroxylase was depleted from the striatal terminals. The lack of functional recovery in these animals may be explained by the fact that the level of GDNF expression was five- to tenfold higher than in the Kordower[45] and Kirik[42] studies (1–2 ng/mg, as compared with ~0.2 ng/mg in the previous studies). It seems possible, therefore, that downregulation of tyrosine hydroxylase in lesioned nigrostriatal dopaminergic terminals, and/or extensive aberrant fiber sprouting in downstream striatal targets, that is induced by high local GDNF amounts, may be detrimental to functional recovery in the 6-OHDA lesion model. Although the mechanism underlying this effect remains unclear, these data indicate that vector-mediated GDNF delivery may have to be maintained within a defined dose range to avoid possible overdosing effects.

Clinical perspective

The potential of GDNF as a therapeutic agent in PD stems from its ability not only to provide symptomatic relief but also to modify the disease state. This aspect is indeed a feature of GDNF therapy that is different from other current therapeutic strategies for PD, such as deep-brain stimulation and dopamine replacement therapy. Yet, it is possible that a combined approach using, for example, fetal dopamine cell grafts and GDNF may prove to be even more potent in reversing the parkinsonian symptoms in patients. Experimentally, GDNF is both neuroprotective and able to induce a prominent functional upregulation in intact and lesioned nigral dopaminergic neurons; in some cases, it can also induce a pronounced regenerative growth response. The viral vector experiments, in particular, indicate that the most pronounced functional effects may result from a combined action involving all three mechanisms. To what extent all the

BOX 1 Intracerebral GDNF therapy: pros and cons of different delivery systems

Intracerebral delivery of neurotrophic factors may be achieved either by direct protein infusion or by gene transfer techniques. As discussed in recent reviews[1,2] each of these modes of delivery has its advantages and limitations.

Infusions of the recombinant protein are advantageous in that the dose can be well controlled and that the infusion can be stopped in case of unwanted side effects. The disadvantage is that the protein is delivered from a point source, which creates steep concentration gradients and restricts the access of the factor to the tissue that is close to the infusion cannula. Long-term protein delivery, moreover, may be complicated by maintenance problems associated with the infusion device. The observations of Gill and colleagus[3] indicate that the total daily dose of protein that can be infused directly into the brain may be restricted by the development of tissue changes and edema at the infusion site.

Cell-based delivery systems can, at least in theory, be used as "biological minipumps" at the implantation site. Cells enclosed in a semipermeable membrane provide an additional level of safety in that they are separated from the brain parenchyma and can be removed in case of unwanted side effects[2]. The limitation of this "*ex vivo*" gene transfer technique, at its current state of development, is that transgene expression may not be stable over time. With encapsulated cells, stable protein secretion has, however, been obtained for several months from devices implanted in the brain parenchyma[2].

Viral vectors have been developed into highly efficient tools for protein delivery to the central nervous system. rAAV and rLV vectors, in particular, allow stable, long-term intracerebral delivery of therapeutic factors in the absence of any detectable pathology or immune reactions, making them highly attractive for clinical use. The use of viral vectors in patients, however, raises important safety issues related to their potential immunogenicity, their risk of mutagenesis by insertion into the genome of the host cells and the possible side effects induced by the expressed transgene. For safety reasons, therefore, it may be necessary to use vector constructs, such as the tetracycline-regulated promoter, that allow the expression of the transgene to be externally regulated or shut off.

effects of GDNF can be obtained in patients with idiopathic PD remains to be documented. In the study of Gill and colleagues[3], the early onset of symptomatic improvement seems compatible with a functional upregulation in residual dopaminergic neurons, rather than a regenerative response. The small increase in ^{18}F-DOPA uptake, which was limited to the area immediately surrounding the infusion cannula, points in the same direction. Animal experiments indicate that the symptomatic effects of GDNF infusion may last for at least 6–8 weeks after the infusion is stopped[22,33]. Nevertheless, in the absence of structural protection or regeneration, it seems likely that these effects will subside over time if the infusion is terminated. In the study of Gill and colleagues[3], the observation time (12 months) was too short to allow assessment of disease progression or neuroprotection, and the PET scans, which were performed at 12 months, suggested at most a very limited regenerative response in these patients.

So, how should GDNF be delivered for optimal and sustained therapeutic effects? Continuously, yes, but where and how should the factor be administered? It is clear that functional upregulation in intact or lesioned nigral dopaminergic neurons can be obtained by the ICV, intranigral or intrastriatal routes, although the ICV route requires considerably higher doses of GDNF, at amounts that are prone to induce unwanted side effects[48]. Only intrastriatal GDNF, however, is capable of protecting degenerating or damaged nigrostriatal axons and terminals and inducing any substantial regenerative growth response. This clearly speaks in favor of the intrastriatal delivery route. From the animal experimental data, however, it seems clear that intrastriatal GDNF is effective in cases only when a significant portion of the nigrostriatal projection remains intact, and thus the efficacy is diminished in animals with advanced parkinsonism. In such cases, as suggested by the primate data[14,24,27], only intranigral delivery, acting through increased dopamine transmission in downstream striatal targets (substantia nigra and/or globus pallidus) may provide symptomatic relief. In PD patients, the optimal site of GDNF delivery may also depend on the site of the primary insult, whether acting on the axon terminals in the striatum or at the level of, or intrinsic to, the nigral cell bodies. In the latter case, one may predict that combined GDNF delivery to both substantia nigra and striatum, as was done in the primate study of Kordower and colleagues[45], may be the optimal choice.

Aberrant sprouting, as seen in substantia nigra and globus pallidus after intraparenchymal protein delivery or viral vector gene transfer, is an issue that remains to be resolved. Although the functional impact of this phenomenon is unclear, it is a concern because functional dopamine receptors are known to be present in both of these structures. The magnitude of aberrant sprouting, however, is clearly dose dependent. In future clinical trials, it will be essential to determine the lowest effective therapeutic dose to avoid overdosing and hence to minimize this effect. In cases of viral vector–mediated gene transfer, available primate data[45] indicate that it may be possible to obtain substantial neuroprotection and functional recovery in the absence of any major aberrant sprouting response. The issue of optimal dosing and the possible negative effects associated with sustained high-level GDNF expression, as well as the safety and efficacy of regulatable GDNF expression from viral vectors, need to be carefully assessed in animal models before any clinical trials in PD patients are carried out.

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COMPETING INTERESTS STATEMENT

The authors declare that they have no competing financial interests.

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