Human idiopathic Parkinson's disease (PD) is a progressive neurodegenerative disorder that is primarily characterized by degeneration of the dopaminergic neurons of the nigrostriatal pathway. Different 6-OHDA rat models of PD have been developed in which this toxin has been injected into different parts of the nigrostriatal pathway: (a) the medial forebrain bundle which leads to extensive dopamine (DA) depletion; (b) the substantia nigra pars compacta, which leads to more specific and moderate DA depletions; and (c) subregions of the caudate–putamen complex (CPu), which also leads to specific DA depletions. In this article we review the dopaminergic depletion and behavioral consequences of 6-OHDA lesions in the rat. It was examined whether the relation between DA depletion and behavioral deficits mimic idiopathic PD. In addition, it was evaluated which model most closely approximates the human situation, especially in relation to the stage of this progressive disease. It was concluded that with respect to the site of the lesion, rats with partial lesions of the ventrolateral CPu are the most appropriate models to study early and late stages of PD. The choice of the behavioral parameters determines the use of unilateral or bilateral lesions, although it is obvious that the bilateral model mimics the human situation more closely.

Key Words: 6-hydroxydopamine; rat; animal model; Parkinson's disease; striatum; substantia nigra; medial forebrain bundle; dopamine; behavior; motor function.

INTRODUCTION

Idiopathic Parkinson's disease (PD) is a neurodegenerative disease that affects approximately 1% of the U.S. population over the age of 55 (6). In 1817 James Parkinson described the motor disorder that now bears his name (46). PD or paralysis agitans is a progressive degenerative disorder. The types of symptoms present and their severity depend significantly on the length of time since onset, the rapidity of functional decline, and whether the patient received medication. The movement disturbances can be separated into positive symptoms (behaviors that do not likely occur in healthy people) and negative symptoms (deficits in or loss of a normal behavioral capacity) (35). The positive motor symptoms of PD are a tremor at rest, muscular rigidity, and involuntary movements due to L-DOPA treatment; the negative motor symptoms are bradykinesia (poverty or slowing of movement) and postural disturbances. Apart from the motor deficits patients with PD often exhibit cognitive dysfunction as well. In the most extreme cases, the individual suffers from dementia (a severe impairment of memory, abstract thinking, language, and other cognitive processes). Also, in approximately 40% of PD patients depressive symptomatology has been found (18).

Although PD has an unknown etiology, postmortem studies have established degeneration of nigrostriatal dopamine (DA) as the hallmark of idiopathic PD (34). However, the exact threshold of nigrostriatal DA dysfunction for the clinical expression of parkinsonism is not known. Before symptoms of PD become apparent, 50–60% of the neurons in the substantia nigra (SN) and about 20% of the DA innervation in the putamen can still be found (33). Because of compensatory phenomena, this substantial (approximately 80%) loss of DA levels in the striatum is thought to be necessary before symptoms become obviously manifest (28). Therefore, the diagnosis of PD can only be made when the individual has already had the disease for a certain time. The nigrostriatal system presumably has considerable reserve capacity to endure deficits of over 50% without symptomatic manifestation. Compensatory re-
sponses by the surviving dopaminergic neurons and also by the postsynaptic cells in the striatum help mitigate the progressive loss of DA innervation. Compensatory responses by afferents to the dendrites of dopaminergic neurons in the substantia nigra (SN) have been reported as well (3). An increased metabolic turnover and hence heightened activity of the remaining dopaminergic cells is one type of compensatory response. A second type of compensatory response is an increased postsynaptic DA receptor density and/or sensitivity. Postmortem brain studies generally indicate modest but significant increases in D₁ and D₂ receptor binding in the putamen of PD patients (59). Based on numerous animal studies, such changes are thought to be a type of denervation supersensitivity. The severe akinesia observed at later stages of the disease is commonly associated with an average loss of neurons in the SN in the range of 60–80%. In addition, DA levels are reduced by over 95% in putamen, but only by 60–90% in the caudate nucleus (8).

The nigrostriatal dopaminergic pathway consists of the A9 cell group, which is located in the substantia nigra pars compacta (SNC). The axons of these neurons run along the medial forebrain bundle (MFB) and terminate in the dorsal striatum. The striatal complex can be divided into a dorsal part (nucleus caudatus and putamen) and a ventral part (including the nucleus accumbens). In the human brain, the nucleus caudatus and the putamen are segregated anatomically by the internal capsule. In contrast, the rat brain lacks such an anatomical segregation and therefore the brain structure is referred to as the caudate–putamen complex (CPu). Because of extensive loss of dopaminergic neurons of the A9 cell group in PD, there is a dramatic decline in striatal DA, leading to the motor impairments. In addition to the SNC, there is also significant degeneration within the midbrain A8 (retrorubral area projecting to the ventrocaudal putamen) and A10 (ventral tegmental area, VTA, projecting to the nucleus accumbens) dopaminergic cell groups (30). Because of the topographic projections of the A8, A9, and A10 cell groups, the dopaminergic pathway to the putamen is more heavily damaged than the corresponding pathway to the nucleus caudatus and the nucleus accumbens. PD is often thought of as a DA-specific disorder. However, numerous histological studies have demonstrated loss of cells in a number of nondopaminergic cell populations, including noradrenergic neurons of the locus coeruleus (LC) and dorsal vaginal nucleus, serotonergic neurons of the dorsal raphe, and cholinergic neurons within the substantia innominata (particularly in the nucleus basalis of Meynert) and in the pedunculopontine nucleus (30, 31). Damage to these important neuronal systems may play a significant role in some of the non-movement-related aspects of PD (e.g., cognition and depression).

**THE 6-OHDA RAT MODEL OF PARKINSON’S DISEASE**

Experimental models of PD are needed to gain insights into the possible pathological mechanisms of the disease. In addition to this function, they are essential in the development and testing of new therapeutic strategies, whether pharmacological or otherwise. In modeling PD a major advance came with the introduction of the catecholamine neurotoxin 6-hydroxydopamine (6-OHDA) (27). This molecule is transported into the cell bodies and fibers of both dopaminergic and noradrenergic neurons. It causes degeneration of nerve terminals and can also affect cell bodies, particularly when administered to the cell body regions. 6-OHDA neurotoxicity is based on its potent inhibitory effect on the mitochondrial respiratory enzymes (chain complexes I and IV) (24). Due to metabolic deficits of the blockage of these enzymes, the neurons can no longer exert their normal physiological functions and consequently they die (for a recent overview see Ref. 10). Since in PD it is mainly the dopaminergic nigrostriatal pathway that is subject to degeneration, animal models have been developed in which 6-OHDA lesions of the dopaminergic system were made. Reasonable selectivity for DA is achieved by pretreating the subjects with desimipramine, a noradrenalin transporter blocker that inhibits 6-OHDA uptake into the noradrenergic neurons. Further selectivity for the nigrostriatal tract can be achieved by injecting the toxin directly into distinct parts of this ascending pathway.

In the preclinical research of PD, rat models have been widely used in which 6-OHDA was injected into either one of three target sites. 6-OHDA was injected into the SNC, MFB, or the CPu. It remains obscure, however, which of these models is most appropriate in modeling PD. To model PD the animal model must mimic both the dopaminergic cell loss and the behavioral deficits associated with idiopathic PD. When a model is obtained that meets this standard, insights into the possible pathological mechanisms of the disease might be obtained and neuroactive agents can be tested that might alleviate PD symptomatology.

**Injection of 6-OHDA into the MFB**

The animal model of PD that to this day undoubtedly has contributed the most in preclinical PD research is the rat with a unilateral 6-OHDA lesion of the MFB (62). Injection of 6-OHDA into the MFB unilaterally can cause a total destruction of A9 and A10 cell groups (48), resulting in the following well-described syndrome: (a) near total depletion of DA in the ipsilateral CPu, (b) denervation supersensitivity of the postsynaptic DA receptors in the ipsilateral CPu, and (c) a characteristic turning behavior in response to both d-amphetamine and apomorphine (see Fig. 1). The completely lesioned (A9 and A10) hemiparkinsonian (i.e., parkinsonianlike syndrome induced in one hemisphere)
rat exhibits more extensive neurodegeneration than is seen in human idiopathic PD (see Ref. 48). In contrast, a hemiparkinsonian rat model that is characterized by unilateral destruction of only the nigrostriatal (A9) pathway while leaving the mesolimbic (A10) pathway intact parallels more closely the extent of neurodegeneration seen in human idiopathic PD (48). This was investigated by Perese et al. (48) using a selective lesion of the A9 pathway which was almost complete, i.e., tissue DA content in the lesioned CPu was reduced with more than 99%. The selective (A9) versus total (A9 and A10) lesion was achieved by a different location of toxin injection and a different concentration.

Unilaterally lesioning the MFB causes a striking asymmetry in the motor behavior of the rats. Following unilateral MFB lesioning, rats initially tend to turn preferentially toward the side of the lesion, a postural motor asymmetry of behavior that may recover only slightly if the depletion is near total. When challenged with drugs acting on the DA system they will display active rotational behavior (63). An imbalance in DA activity between the two striata causes the rotational asymmetry. Thus, the animal rotates away from the side of greater activity (65). Subcutaneous administration of the DA-releasing agent d-amphetamine creates a DA imbalance that favors the nonlesioned nigrostriatal projection and thus produces ipsilateral rotations (see Fig. 1). This imbalance can be detected even in rats with 50% sparing of nigral dopaminergic neurons, as measured with tyrosine hydroxylase (TH) immunocytochemistry (26). The postsynaptic agonist apomorphine induces rotation contralateral to the lesioned side (see Fig. 1) because of a stimulation of denervation-induced upregulated D2 receptors in the denervated striatum (65). Postsynaptic supersensitivity occurs only after most of the dopaminergic neurons in the SNC (approximately 90%) have been eliminated (26). At lower levels of cell loss compensatory increases in DA synthesis by spared dopaminergic cells in the SNC (1) or high levels of endogenous DA in the extracellular space of the denervated CPu (13) could avoid the development of supersensitivity. Rats with more restricted lesions of the SNC show no rotational asymmetry in response to apomorphine (26). Consequently, in many studies the two rotation-inducing agents d-amphetamine and apomorphine have been used to behaviorally assay the extent of neuronal loss following lesions of the SNC (e.g. Ref. 12). It has been demonstrated that rats, which are moderately lesioned (with a 75–90% reduction in dopaminergic fiber density in the CPu), rotated on d-amphetamine but not on apomorphine (29). However, it has to be noted that control rats which received no lesions were often seen to rotate extensively on d-amphetamine. Although it has not been reported, this might be due to a natural imbalance in DA between the two CPu complexes (cf. Ref. 19). However, such an explanation was not discussed (29). More extensive lesions of the CPu (>90% reduction in CPu dopaminergic fiber density) and concomitantly SN (>50% depletion of dopaminergic neurons) are required to generate rotations demonstrable with a low dose of apomorphine but not with d-amphetamine. It was therefore concluded that apomorphine, rather than d-amphetamine, is a better predictor of extensive lesions of the CPu produced by 6-OHDA (29).

It has been shown that a unilateral injection of 6-OHDA into the MFB resulted in rats that either rotated after the administration of apomorphine, Apo (+), or did not rotate after the administration of apomorphine, Apo (−) (6). In the Apo (+) rats a 99.8% depletion of DA tissue content in the CPu and an 85% depletion of DA tissue content in the SN were demonstrated. For the Apo (−) rats the striatal and nigral DA tissue contents were about 75 and 55%, respectively. These results confirm the conclusion that apomorphine-induced rotational behavior is a good indicator of extensive CPu lesions by 6-OHDA. These complete and partial unilateral depletions of the MFB impaired the hierarchic phases of paw reaching differently as measured in a skilled motor task in which animals have to reach for a food reward using fine, controlled movements of the forepaw (staircase test, see Table 1). Barnéoud et al. (6) reported that a complete DA depletion, but not a partial one, decreased the number of attempts made with the contralateral paw and induced a bias toward the ipsilateral paw, i.e., the ipsilateral paw was preferentially used by these rats as has also been found recently by Dunnett et al. (32). A partial DA lesion impaired the sensorimotor coordination of both paws.
Besides impairments in using the contralateral limbs for skilled movements in tests of reaching, impairments in movements of spontaneous food handling have been reported after unilateral lesioning the MFB (67). Rats with lesions similar as the latter had a unilateral DA depletion that exceeded 99% in the ipsilateral CPu. It has also been shown that MFB lesions affected place (cognitive) and cue (sensorimotor) orientation in the Morris water escape task (see Table 1) (68). Rats with unilateral MFB lesions that caused

### TABLE 1
Overview of Several Behavioral Tests with a Brief Description

<table>
<thead>
<tr>
<th>Test</th>
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<tr>
<td>Paw retraction test</td>
<td>Procedure as described by Ellenbroek et al. (22). This is a test for akinesia. The test apparatus consists of a polyvinylchloride (PVC) frame with four holes. Two holes are for the forelimbs of the rat and the other two are for the hindlimbs. As soon as the rat is placed with all four limbs in the holes the rat is released. The latency to retract the contralateral fore- and hindlimbs from the holes is registered.</td>
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<tr>
<td>Adjusting steps test</td>
<td>Procedure as described by Olsson et al. (45). This is a test for akinesia. The rat is held with one hand by the experimenter fixing the hindlimbs (slightly raising the torso) and with the other hand fixing the forelimb that is not to be monitored. In this way the other forepaw has to bear the weight. The rat is moved slowly sideways in both forehand and backhand positions. This is done for both the contralateral and ipsilateral forepaw. The number of adjusting steps for both directions and both paws are counted.</td>
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<tr>
<td>Staircase test</td>
<td>Procedure as described by Montoya et al. (42). This is a test for fine motor control. The test apparatus consists of a clear Perspex chamber with a hinged lid. To this chamber a narrower compartment with a central raised platform running along its length is connected, creating a trough on either side. Due to the narrowness of the side chambers, the rats can use only their left paw for reaching into the left trough and their right paw for the right trough. A removable double staircase is inserted into the end of the box, sliding into the troughs on either side of the central platform. Each of the eight steps of the staircase contains a small well, and two saccharin-flavored pellets are placed in each well. The number of pellets eaten during the test period indicates the rat’s success in grasping and retrieving the pellets. The number of steps from which pellets have been removed provides an index of the attempts to reach the food and how far the rat can reach. The number of missed pellets remaining at the end of the test on the floor of the side compartment indicates a lack of sensorimotor coordination in grasping and retrieving pellets.</td>
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<tr>
<td>Apomorphine or d-amphetamine-induced rotations</td>
<td>Procedure as described by Ungerstedt (65). This is a test to ascertain maximal lesions. Drug-induced rotations are measured using an automated rotometer consisting of a rotation bowl and a tether attached to the torso of the rat.</td>
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<tr>
<td>Locomotor activity</td>
<td>Procedure as described by Cools et al. (15). The rats are placed on a 160 × 160 cm horizontal flat glass table serving as openfield, 95 cm high and surrounded by a neutral white background. Behavior is recorded with a computerized and automated tracking system.</td>
</tr>
<tr>
<td>Reaction-time (RT) task</td>
<td>Procedure as described by Amalric et al. (2). This is a test for motor initiation. Food-deprived rats are placed in operant boxes, each with a retractable lever and a stimulus light located above the lever. A food pellet magazine is located to the right of the lever. Animals are trained to hold down the lever. After the presentation of the visual cue stimulus at a randomly variable time interval the rats have to release the lever within 700 ms to get a reward (food pellet). The performance of the rats is measured as the number of correct and incorrect (premature lever release or lever release after the 700 ms time interval) trials.</td>
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<tr>
<td>Fixed ratio (FR) bar-pressing task</td>
<td>This is a test for skilled motor control. Food-deprived rats are placed in operant boxes, each with a retractable lever. During the experiment the rats have to make five lever presses [fixed ratio 5 schedule, Cousins et al. (16)] or 10 lever presses [fixed ratio 10 schedule, Barnéoud et al. (5)] to receive one food pellet as a reward.</td>
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<tr>
<td>Morris water escape test</td>
<td>Procedure as originally described in Morris et al. ([43] see also [68]). A black tank with a diameter of about 1.5 m is filled with water. The rats are placed in the (aversive) water and have to swim toward an escape platform (about 15 cm in diameter). Parameters as swimming distance and swimming velocity are measured. A place version of this test (hidden platform below water surface) measures spatial learning (cognition). In contrast, a cue version in which the platform is visible above the water surface assesses sensorimotor function.</td>
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<tr>
<td>Forelimb use asymmetry test</td>
<td>Procedure as described in Schallert et al. (56). The rat is placed in a transparent cylinder. During a time period of 5 min the rearing behavior of the rat is scored. The behavior is analyzed during rearing and landing. The percentage of simultaneous and asymmetric use of the paws during these movements is determined.</td>
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over 95% tissue DA content depletion in the CPu were impaired in the rate of acquisition in both versions of the Morris water task, although they were eventually able to learn both versions, i.e., to locate the escape platform. These results suggest that unilateral destruction of the MFB affects sensorimotor and cognitive skills.

The MFB was also lesioned unilaterally and it was demonstrated that depletion of DA levels in the CPu by over 80% resulted in dramatic reductions in the ability of rats to make adjusting steps (see Table 1), whereas rats with DA level reductions in the CPu of less than 80% showed no detectable deficit (14). Originally, this test was conducted by letting the animal bear weight on a forepaw and monitoring the time until initiation of movement (55). Subsequently, a variety of stepping parameters were studied in rats with a unilateral 6-OHDA lesion of the MFB (45). They reported that adjusting steps were reduced consistently after the unilateral MFB 6-OHDA lesion. In contrast to apomorphine-induced rotations, the deficit in adjusting steps was evident at milder DA depletion, i.e., at 80% reduction in CPu DA levels (14). Furthermore, a depletion of CPu DA levels of about 80% resulted in forelimb use over 80% showed no detectable depletion of rats with DA level reductions in the CPu of less than 80% resulted in dramatic reductions in the ability of rats to make adjusting steps (see Table 1), whereas rats with DA level reductions in the CPu of less than 80% showed no detectable deficit (14). Originally, this test was conducted by letting the animal bear weight on a forepaw and monitoring the time until initiation of movement (55). Subsequently, a variety of stepping parameters were studied in rats with a unilateral 6-OHDA lesion of the MFB (45). They reported that adjusting steps were reduced consistently after the unilateral MFB 6-OHDA lesion. In contrast to apomorphine-induced rotations, the deficit in adjusting steps was evident at milder DA depletion, i.e., at 80% reduction in CPu DA levels (14). Furthermore, a depletion of CPu DA levels of about 80% resulted in forelimb use asymmetry in a cylinder (see Table 1) (61). The behavioral deficit was highly correlated with the DA depletion.

Spirduso et al. (60) showed that unilateral lesioning of the MFB caused deficits in high-speed motor initiation, comparable to reaction time responding. The movement initiation deficits of the contralateral paw were found to be linearly related to the DA loss, measured as [3H]DA uptake by the CPu. The behavioral deficits in this task were found with a DA depletion of about 40–100%. Thus, this task is sensitive to detect a weak to severe DA depletion. In addition, small yet substantial deficits were also seen in ipsilateral paw performance following more severe lesions, which may be related to DA depletions found in the nonlesioned CPu (60).

When modeling PD with 6-OHDA lesions of the MFB in rats, it is also possible to make bilateral lesions. Using a bilateral model of PD has several advantages. Since idiopathic PD is bilateral, the real pathological situation will be more closely approximated. Another advantage is that sprouting of axons from an intact side of the brain to the contralateral is avoided (66, but see 9). This implies that the model will be more stable with respect to compensation after the lesion. Rats with extensive bilateral MFB lesions were reported to manifest a severe motor symptomatology with akinesia reminiscent of that seen in advanced PD (64). Unfortunately, the extensive bilateral lesion also causes aphagia (deficit in swallowing) and adipsia (deficit in drinking) in the rats (64), requiring them to be tube-fed, which has somewhat limited the use of the bilateral 6-OHDA lesion model. The MFB has also been lesioned bilaterally by the use of a bilateral 6-OHDA intracerebroventricular injection (68). These rats displayed a tissue DA depletion in the CPu of about 99% and were unable to acquire either the place or the cue version of the Morris water escape task. These findings further indicate cognitive impairments in addition to motor impairments in the MFB lesion model.

Injection of 6-OHDA into the SNC

In order to make a more selective animal model of PD with more moderate DA depletions, the SN has been targeted for 6-OHDA injection. Animals that received a medial and a lateral injection or a single lateral injection into the SNC had moderate sparing of dopaminergic neurons (12). In these rats, the dopaminergic cell loss in the SNC as measured with TH immunocytochemistry was 88%. The percentage of surviving dopaminergic cells in the VTA on the lesioned side was 69% when compared to the unlesioned side. This means that the toxic effects of 6-OHDA are not restrained to the SNC, but that the VTA is affected partially as well. Interestingly, rats that received either a single lateral injection into the rostral SNC or a medial and lateral injection into the rostral SNC had greater sparing of dopaminergic cells in the medial SNC than in the lateral SNC (12, 20). This relative sparing of dopaminergic cells in the medial SNC reflects the pattern of cell loss in brains of patients with PD, in which the DA depletion is mainly lateral (23, 25). Also very interesting is the fact that the number of TH-immunoreactive neurons remaining in the lesioned SNC was correlated with the number of rotations induced by apomorphine, but not with d-amphetamine-induced rotations (12). Consistent with other studies (e.g., Ref. 26) typically only animals in which the lesioned SNC contained 10% or fewer TH-immunoreactive neurons than the intact side rotated after administration of apomorphine. Furthermore, it was demonstrated that TH-immunoreactive fibers in the CPu showed a distribution that paralleled the pattern of nigral cell sparing: The lateral CPu on the side of the lesion was less densely innervated than the lateral CPu (12). In addition, the percentage of CPu area that was innervated by TH-immunoreactive axons was strongly correlated with the number of TH-immunoreactive cells in the SNC. Finally, the percentage of CPu area that was innervated by TH-immunoreactive fibers was also correlated with the number of rotations in response to apomorphine, but not to d-amphetamine. Thus, this animal model with partial lesions of the SNC can predict the extent of cell loss in SNC and axonal loss in the CPu, using the apomorphine-induced rotational behavior as an indication of cell loss.

Since PD affects the brain bilaterally, it is obvious that a bilateral model can be used for preclinical research. Van Oosten and Cools (66) reported about a bilateral 6-OHDA rat model of PD in which the neurotoxin was injected into the SNC. They mentioned sev-
eral rationales for their decision. Among these was the fact that especially the striatal region that is innervated by the nigrostriatal (A9) fibers, and not so much by the mesolimbic (A10) fibers, is depleted in PD. One of the rationales for using a bilateral model was that unilateral injection of 6-OHDA affects the nonlesioned hemisphere as well, which makes a unanimous interpretation of drug-induced effects (e.g., rotational data) difficult. In their study it was shown that relatively small bilateral 6-OHDA lesions, especially of the SNC, and to a minor degree the VTA, produced changes in parameters known to be CPu-specific (66). A unilateral lesion of the SNC was found to deplete DA levels in the CPu by more than 95% on one side of the brain but caused bilateral impairments in skilled paw use, as measured in the staircase test (69). Apparently, unilateral lesions of the SNC do not guarantee behavioral impairments to be restricted to one side of the body.

Injection of 6-OHDA into the CPu

In order to make more selective destructions of the nigrostriatal dopaminergic pathway, the CPu has been targeted as the site of toxin injection in many recent studies. In the majority of these studies discrete subregions in the CPu were selected as a target for the lesion. Terms like ventrolateral and dorsomedial striatum predominate these studies. Using this terminology, there is an important aspect to keep in mind. All the regions named in the following are regions within the dorsal striatum (i.e., the caudate–putamen complex) and not within the ventral striatum (including the nucleus accumbens). Hence, the dorsomedial part of the striatum implies the dorsomedial part of the dorsal striatum or CPu. Figure 2 gives an indication of the regions that are referred to in a rostrocaudal direction. A subdivision is made into a ventral, dorsal, medial, and lateral part. All the other terms are derived from this (e.g., the ventrolateral part of the CPu comprises both the ventral part and the lateral part of the CPu and the dorsomedial part of the CPu comprises both the dorsal and medial part of the CPu).

The ventrolateral sector of the CPu, which receives intensive input from motor and sensorimotor areas of the neocortex and receives its DA innervation exclusively from the SN (33), may be equivalent to the putamen in primates and humans. The dorsomedial CPu, on the other hand, has a mixed DA innervation from both SN (A9 cell group) and VTA (A10 cell group) and receives inputs from frontal cortical areas and the limbic system; therefore this CPu subregion may represent an equivalent of the nucleus caudatus in humans. Lesions involving the dorsomedial parts of the CPu have more general effects on locomotion and drug-induced turning behavior, whereas lesions involving the ventrolateral parts of the CPu have pronounced effects on movement initiation, sensorimotor orientation, and skilled motor behavior (11, 16, 17, 21, 53). The putamen is the striatal subregion that presents the most profound DA depletions in patients with PD (e.g., Ref. 44). Therefore, partial lesions that are focused on the ventrolateral CPu in rats are likely the most relevant models of PD (40).

Kirik et al. (33) tried to establish the optimal parameters for a stable unilateral 6-OHDA lesion of the ventrolateral CPu of sufficient magnitude with respect to both the extent of denervation in the CPu and DA cell loss in SN. This was done to induce consistent long-lasting contralateral behavioral deficits in the adjusting steps test (rigidity and akinesia) and the staircase test (fine motor control). They found that this indeed possible when the neurotoxin was distributed over multiple injection sites along the rostrocaudal axis of the ventrolateral CPu. The functional effects induced by intrastriatal 6-OHDA lesions depended not only on the total dose of the toxin injected, but also on the site of toxin injection. The dose of toxin administered in a single injection had an effect on the extent of CPu DA depletion. When a dose of 6-OHDA was distributed over three or four sites along the rostro-caudal axis of the lateral CPu or when injections were made close to the junction of the globus pallidus, pronounced behavioral deficits occurred. Impairment in
FIG. 3. Left rat hemisphere with a CPu subdivision in six areas by Chang et al. (14). The shaded areas indicate CPu subregions. Abbreviations: DM, dorsomedial; DC, dorsocentral; DL, dorsolateral; VM, ventromedial; VC, ventrocentral; VL, ventrolateral [adapted from (47)].

in the staircase test was obtained only with a four-site lesion in which an 80–95% reduction in TH-immunoreactive fiber density throughout the rostrocaudal axis of the lateral CPu and a 75% loss of TH-immunoreactive neurons in SN. In addition, a 60–70% reduction in TH-immunoreactive fiber density in the lateral CPu, accompanied by a 50–60% reduction in TH-immunoreactive neurons in SN, was shown to be already sufficient for the induction of impairment in initiation of stepping in the adjusting steps test.

Chang et al. (14) reported that a reduction in CPu DA levels of 80% or more resulted in impairment in the adjusting steps task. Also, discrete unilateral CPu lesions were used to localize the subregions in the CPu that mediate adjusting steps (14). However, they used a CPu subdivision that differs from the one in Fig. 2 (see Fig. 3). Adjusting steps were shown to be reduced after lesions of dorsolateral, ventrolateral, or ventrocentral CPu (Chang’s terminology), but not after lesions of dorsomedial, dorsocentral, or ventromedial CPu (Chang’s terminology). Furthermore, none of the discrete CPu lesions resulted in rotation after apomorphine administration, which is consistent with the consideration that apomorphine-induced rotations require the lesions to be maximal.

Bilateral lesions in the CPu would probably serve a closer parallel to the human disease than unilateral lesions because both striata are affected in human PD. Amalric et al. (2) investigated the effect of bilateral 6-OHDA CPu lesions on a reaction-time (RT) task which measures motor initiations (see Table 1). In this RT task the rats are trained to respond to a visual stimulus when holding down a lever. The rats have to release the lever within a certain time interval after the visual cue. An anticipated release of the lever (before the visual stimulus) and a delayed release of the lever (after the predetermined poststimulus interval) are not rewarded by a reinforcer (food pellet). All correct responses are rewarded. The overall decline of DA tissue content in the two lesioned CPu was about 75% of control values. However, lesioned animals differed in the extent of DA depletion, as was demonstrated with TH immunocytochemistry. Thus, the group of lesioned animals could be subdivided into a group with extensive lesions, particularly of the medial CPu, and a group with less extensive lesions that appeared to be restricted to dorsal and lateral parts of the CPu.

Also, animals with asymmetric lesions (one side partly unaffected by 6-OHDA) were observed. These latter lesions had no effect on reaction-time performance. Therefore, it can be concluded that a bilateral DA-depleting lesion has to be symmetrical in both CPu for behavioral deficits to become apparent in this RT task. Animals with the most extensive medial lesions had deficits in both anticipated responses and delayed responses, whereas the animals that were less severely lesioned (dorsally and laterally) showed only an increase in delayed responses (2). Delayed responses and increased anticipated were associated with motor and cognitive deficits, respectively. On basis of these data it was concluded that a large medial CPu DA depletion may provide both motor and cognitive deficits. The dorsolateral CPu is known to receive a dense innervation from cortical sensorimotor areas (41). DA activity in the dorsolateral CPu thus appears to be critical for motor function, i.e., the initiation of movements with temporal constraints. It is mainly the lateral aspect of the CPu that has the emphasis in mediating the function of motor initiation (e.g., Ref. 11). This is confirmed by Cousins et al. (16), who reported that mild DA level depletions (about 30% of control levels) in the ventrolateral CPu caused increased initiation times in a skilled motor control task of lever pressing. In this task the rats had to make five presses [in a fixed ratio (FR) five-lever-pressing task] to receive one food pellet as a reward (see Table 1). These motor deficits observed in the rats with a DA depletion were argued to show similarities with the motor deficits observed in patients with PD (16).

Besides the motor effects (see also Ref. 52), cognitive deficits could be observed as well after bilateral lesions of the ventrolateral CPu (37). Spatial cognitive deficits, as measured in the (place) Morris water escape task, were reported in two groups of rats with either a 64 or a 60% reduction in nigral neurons. The tissue DA content in the CPu of these two groups of rats was about 55 and 80%, respectively. The differences in depletion could be attributed to age, as the latter rats were middle-aged rats (12 months old), whereas the former
were young adult rats (2 months old). Both age groups were impaired in locating the platform and this cognitive deficit was related to nigrostriatal DA depletion and not to a decrease in dopaminergic transmission in the prefrontal cortex, since DA levels in this latter brain area remained stable after lesioning (37). Furthermore, the cognitive deficits were not likely due to an asymmetry in motor activity, since it has been reported that rats with almost total unilateral CPu DA depletions due to a MFB lesion, eventually learned to locate the platform in the (place) Morris water maze (68).

Besides correlating behavioral deficits to DA depletion in the CPu, it is also important to accurately evaluate compensation following partial dopaminergic lesions. Barnéoud et al. (5) recently reported bilaterally lesioned rats (dopaminergic lesion in both the ventrolateral and dorsomedial CPus, which mounted up to a 70% decrease in CPu DA levels) that were able to compensate for some of their behavioral deficits (FR 10 schedule of reinforcement) although initiation of lever pressing after a reward and sustained action were still impaired. This study demonstrated that it is necessary to use several parameters to accurately evaluate compensation following partial dopaminergic lesions (5).

At advanced stages of PD, DA levels are reduced by over 95% in putamen, but by only 60–90% in the nucleus caudatus (34, 44). For investigation in animal models of advanced PD, an animal model providing lasting dopaminergic depletion of 80–100% in the CPu of the rats is needed. Ben et al. (7) compared the effects of a single and a double 6-OHDA injection bilaterally into the CPu using the same dose (16 μg/CPu) on DA and 3,4-dihydroxyphenylacetic acid (DOPAC, a DA metabolite) levels in the CPu. Injections of 6-OHDA at two different sites of the CPu (i.e., in the caudal and rostral CPus) induced a relatively stable DA level decrease of about 90% compared with controls. Although the single bilateral 6-OHDA lesioned rats showed a more pronounced loss at week 2 postoperatively than at week 8, which might be indicative of a compensatory phenomenon. This was not observed in double bilateral 6-OHDA lesioned rats. The DOPAC/DA ratio, which might provide a good index of DA release and turnover, was increased in both the single and double bilaterally lesioned groups.

**DISCUSSION**

Animal models of human idiopathic PD are needed to gain insight into the etiology of the disease itself and to test therapeutic strategies. During the past decades several rat models of PD have been used, but there is a lack of consensus about the location of the lesion, the percentage of DA depletion in the CPu, and the behavioral tests to use to relate the extent of the lesion to the PD-like symptoms. This review shows that the 6-OHDA rat model of PD closely mimics the human disease. However, as human PD is a progressive disease, it is subdivided into different stages. Subsequent stages of the disease are characterized by a progressive degeneration of the nigrostriatal pathway and a corresponding progressive decline in striatal DA levels. Therefore, to have a reliable and good animal model of PD, an important question has to be met first. In what stage of human PD lies the interest? When PD symptoms start to emerge, about 50% of the dopaminergic neurons in the substantia nigra are lost and striatal DA levels have decreased by about 80% (33, 39). In 6-OHDA lesioned rats similar data have been obtained. Compensatory responses to 6-OHDA lesions in the rat have been studied extensively in both unilaterally and bilaterally lesioned animals (13, 70). It was shown that extracellular DA levels in the CPu do not decrease until CPu tissue DA depletion exceeds 80%. At lower levels of DA depletion, neuronal compensation mechanisms are able to compensate for the depletion. These mechanisms involve increased release of DA from remaining dopaminergic terminals as well as an upregulation of DA receptors and supersensitisation of DA receptors. Beyond 80% depletion of striatal DA levels, these compensation mechanisms are insufficient in neutralizing the DA depletion. This is confirmed by the finding of a modest and a marked drop in extracellular DA with an 80–95% and >95% depletion of tissue DA concentration in the CPu of rats (13). As a consequence of the inadequate compensatory mechanisms at such high levels of DA depletion, “clinical” symptoms become manifest. Thus, to mimic idiopathic PD, a rat model is needed in which the DA depletion is 80% at least.

In preclinical research, rat models have been developed focusing on the nigrostriatal pathway. This dopaminergic pathway is lesioned at different levels to mimic PD. There is a lack of consensus among researchers about the site for toxin injection. The targets of injection of the neurotoxin 6-OHDA are (a) the site of origin of the nigrostriatal pathway, i.e., the SNC; (b) the axon bundle that partly projects toward the CPu, i.e., the MFB; and (c) the terminal site of the nigrostriatal pathway, i.e., the CPu. In addition, the lesions can be unilateral or bilateral. To mimic idiopathic PD it is important that the DA depletion in the CPu of the rat resembles the situation in the diseased human brain. In addition, the behavioral deficits that can be measured after the lesion should validate the model even more.

**Location of 6-OHDA Lesion Site**

Medial Forebrain Bundle. The most widely used 6-OHDA rat model for studying PD uses animals with unilateral lesions of the MFB (27). These lesions are almost complete and very few dopaminergic neurons in the SNC survive. This model is valuable as a model of advanced stages of PD. On the other hand, a disadvantage of this model is that not only dopaminergic axons
from the A9 cell group in the SNC are running along the MFB. Axons from the A10 cell group, which terminate in the nucleus accumbens (of the ventral striatum) comprise the MFB as well. Thus, when lesioning the MFB these latter axons are damaged as well. Therefore, MFB lesion models have been developed that are both selective and complete for the nigrostriatal (A9) pathway, i.e., only the A9 cell group is affected and at a maximal (about 100%) level (e.g., Ref. 48). However, a less extensive DA depletion after lesioning the MFB has also been reported. Bannéoud et al. (6) reported about moderate DA depletions (about 75% CPu tissue DA depletion) after selective MFB lesions. The accompanying mild paw reaching impairments (staircase test) in these animals were proposed as a model of the early symptoms of PD (6). Also, at relatively low levels of DA depletion (a reduction of about 80% in CPu tissue DA content), deficits in the adjusting steps task (14) and in a reaction time task (60) were observed. In addition to unilateral MFB lesion models, models were developed in which the MFB was lesioned bilaterally. These models have been limited in use, as it appeared that the rats were suffering from adipsia and aphagia (64). Taken together, MFB lesion models mostly have total elimination of dopaminergic cells in the ipsilateral SNC, but also extensive loss of cells in the VTA. Since the extent of this degeneration exceeds the situation in PD, more selective animal models have been developed in which the neurotoxin was injected into either the SNC or into the CPu.

Substantia nigra pars compacta. When using SNC as the target site for toxin injection, PD is mimicked more closely with respect to dopaminergic cell loss. Unilaterally lesioning the SNC was shown to deplete dopaminergic neurons by about 90% in the SNC (12). When compared to MFB lesions [97% loss of TH immunostaining in the SNC (12)], the TH-staining loss in the SNC is somewhat less extensive after SNC lesioning (12). Thus, the dopaminergic cell loss in the SNC model advances an intermediate stage of the human pathological situation. Thus, the pattern of DA depletion was similar to that observed in brains of PD patients, in which the DA depletion in the SNC is mainly lateral (23, 25). Furthermore, in rats that received unilateral 6-OHDA injections into the SNC, the DA depletion in the CPu was also more lateral than medial (12). Individual animals with unilateral DA depletions of 90% or more in the SNC rotated after the administration of apomorphine. In addition to unilateral lesions of the SNC, bilateral models with the SNC as the target site for 6-OHDA injection were used as well. In this model it was shown that small bilateral lesions produced changes in behavioral parameters that are CPu-specific, e.g., via the paw retraction test (see Table 1) (66). A difficulty of the SNC as the target for 6-OHDA injection is the small size of the structure. It is very difficult to inject the 6-OHDA into this structure without lesioning adjoining structures (e.g., the VTA). This is reflected by the reduction in DA neurons in the VTA by about 30% after SNC lesioning, which was found by Carman et al. (12).

Caudate-putamen complex. Partial lesions in the CPu are probably of more value to future preclinical PD research since it can be easily done and the behavioral and biochemical data collected closely approach the human situation. Moreover, rather selective lesions of the CPu can be achieved. But also in modeling PD with CPu 6-OHDA lesions there is no consensus about the lesion site. Some researchers used the dorsomedial CPu (e.g., Ref. 51), whereas others used the ventrolateral CPu (e.g., Ref. 37) and still others used another CPu area for toxin injection (e.g., Ref. 14). Since the putamen represents the most profound DA depletions in the brain of PD patients (e.g., Ref. 44) and the putamen in humans is equivalent to the ventrolateral section of the rat CPu (33), partial lesions aimed at the ventrolateral part of the CPu are probably best. Lesions of this target site showed impairments in behavioral parameters that are associated with PD, like movement initiation, sensorimotor orientation, and skilled motor behavior (11, 16, 17, 21, 53). Furthermore, since PD is a progressive disease, there should be separate models for both early stages of the disease and manifest stages of the disease. The two models are thus dependent on the extent of the lesion and the severity of behavioral deficits.

Partial DA depletion (reduction of 60–80% of CPu DA levels) by local unilateral injection of 6-OHDA in the medial CPu has recent been claimed to be a good model of early and moderate stages of PD in which to examine or study the effects of neurotrophic therapies (e.g., Ref. 36). In this model paw-reaching deficits (staircase test) were observed when CPu DA levels were reduced by about 80%. This is in agreement with a study in which paw-reaching impairments were also obtained with an 80–95% reduction in dopaminergic fibers in the lateral CPu [and a 50–60% reduction in dopaminergic neurons in the SN (33)]. In this latter model, the remaining intact nigrostriatal projection is thought to have a role in the regeneration and functional recovery in response to growth promoting factors (33). Deficits in paw reaching have been suggested to be similar to the motor deficits seen in patients with PD (16). In one study with a DA depletion of 80% in the CPu rotational behavior after the administration of apomorphine was observed (36). This finding contradicts with most other studies in which apomorphine-induced rotations were only observed in animals with a DA depletion in the CPu of 90% or more (e.g., Ref. 6). The discrepancy of this finding with the other studies can be due to the lesion site, i.e., lesioning the CPu (36) instead of the MFB (6).

Chang et al. (14) found that discrete single lesions in
the dorsolateral, ventrolateral, or ventrocentral CPu (Chang's terminology) reduced adjusting steps. The adjusting steps test allows the characterization of non-drug-induced deficits in forepaw movement as a model of akinesia and gait problems, as observed in PD patients (55). Taken together, a discrete lesion in the ventrolateral CPus would be preferable because this part is thought of as equivalent to the putamen in humans. Not only the location of toxin injection, but also the number of injection sites and the concentration of the neurotoxin, is of importance to the model as was shown by Kirik et al. (33). Also, the injection volume is important because of differences in diffusion of the neurotoxin from the injection site to the surrounding brain tissue.

Models in which the ventrolateral CPu was lesioned bilaterally mimicked PD dosely as well. A depletion of CPu tissue DA content of about 75% induced both motor and cognitive deficits as measured in a RT task (2). The use of a RT task may therefore provide a good index of motor and cognitive abnormalities present in the early stages of PD, as stated by Amalric et al. (2). Lindner et al. (37) showed that many clinical symptoms could be observed in rats that were bilaterally lesioned in the ventrolateral part of the CPu. In this study, young adult rats had a depletion of nigral neurons of about 65% accompanied with a reduction in CPu DA levels of about 80%. In addition, these rats showed deficits concerning akinesia (FR10 test), rigidity (adjusting steps task), tremor (observation of vacuous or tremulous jaw movements), and cognition (Morris water maze) (37). In the same study, it was attempted to determine whether older rats exhibited more robust parkinsonian deficits than younger rats due to a hypothesized age-related decline in compensatory mechanisms and neuroplasticity. This was hypothesized since the human disease affects people in later stages of their lives. However, the difference between the two age groups was not dramatic. This was probably due to the age of the older rat, which was in fact just middle-aged (12 months), i.e., they were still relatively young to have more CPu damage. Thus, this age is not suitable to test the hypothesis for age-related changes in recovery (38).

Since the dorsoventral and the mediolateral axis of the rat CPu is very important in modeling PD, the question arises whether the rostrocaudal aspect is of importance as well. For instance, it appears that the caudal aspect of the CPu is more damaged by 6-OHDA than its rostral aspect (2, 52). In the studies equated in this review, behavioral deficits in relation to the rostrocaudal axis are not unambiguous. Most of these studies used anterior-posterior (cf. rostrocaudal) coordinates between 1.5 and −1.5 mm from bregma. It remains to be demonstrated, however, whether the behavioral deficits occur beyond this range as well.

Behavioral Aspects of 6-OHDA Lesions

Behavioral deficits in response to 6-OHDA lesions can give an indication of the extent of the lesion. Table 2 gives an overview of models with different target sites for the lesions, the accompanying depletion in the SNC and/or in the CPu, and the behavioral deficits in these models. Models with unilateral lesions often use drug-induced rotational behavior, which can be used as an indicator of nigrostriatal DA depletion. Extensive (maximal) lesions of the CPus (>90% loss of DA fiber density) and concomitantly SNC (>50% loss of dopaminergic neurons) are generally assumed to be required to generate rotations demonstrable with low doses of apomorphine, but not with amphetamine (29). Other investigators have argued against drug-induced rotational behavior as a reliable indicator of nigrostriatal DA depletion (e.g., Ref. 14). Chang et al. (14) used adjusting steps as an indicator of DA depletion. The stepping test is preferable to acquire an indication of the depletion in DA in experiments in which there is a possibility of damaging the CPu. For instance, grafts can disrupt apomorphine-induced rotation by damaging postsynaptic receptors in the CPu (4). Such damage would further enhance stepping deficits. In contrast, this damage would result in reductions in drug-induced rotation, an effect indistinguishable from any therapeutic effect of substances that are administered to alleviate the symptoms of the disease. Thus, there may be a discrepancy between the interpretation of the behavioral outcome and the actual neuronal damage. The adjusting steps task can also be used as an indicator of submaximal lesions since deficits are detected when DA levels in the CPu are decreased by about 60–80% (14, 37). This fits nicely with the fact that PD symptoms start to occur in the PD patients when DA levels in the CPu decrease beyond 80% (28). Behavioral tests also provide the possibility to assess the extent to which the behavior of the rats relates to clinical symptoms of PD patients (e.g., Ref. 37). Examples of these behavioral tests are the FR-bar-pressing task or the paw-retraction test for akinesia, the Morris water maze for cognitive deficits, the staircase test for fine motor control, and the adjusting steps task for rigidity/akinesia (see Table 1).

Unilateral and Bilateral 6-OHDA Lesions

Most rat models of PD involve unilateral lesions [for an excellent overview of unilateral lesions of the SN or MFB see (57, 58)]. These models have been invaluable in preclinical PD research. However, there are several good arguments for using a bilateral model of PD instead. One is that the human disease affects the brain bilaterally as well. Another is that there is no intact site which can partly compensate for the affected site (cf. Ref. 66). In rats with intrastriatal 6-OHDA lesions, it was demonstrated that bilateral 6-OHDA lesions caused impairments in more behavioral motor para-
**Table 2**

Overview of Dopaminergic Lesions in MFB, SNC, or CPu and the Accompanying Behavioral Deficits

<table>
<thead>
<tr>
<th>6-OHDA lesion site</th>
<th>Concentration 6-OHDA</th>
<th>Percentage reduction in:</th>
<th>Behavioral deficit</th>
<th>Ref.</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>SNC neurons</td>
<td>CPU DA levels</td>
<td></td>
</tr>
<tr>
<td>MFB</td>
<td>Unilateral 4 µg/1.5 µl</td>
<td>85</td>
<td>99.8</td>
<td>Apo (+): staircase test decrease in use of contralateral paw, bias toward ipsilateral paw</td>
</tr>
<tr>
<td></td>
<td>Unilateral 4 µg/1.5 µl</td>
<td>56</td>
<td>72</td>
<td>Apo (-): staircase test deficit in sensorimotor coordination</td>
</tr>
<tr>
<td></td>
<td>Unilateral 8 µg/2 µl</td>
<td>n.d.</td>
<td>&gt;80</td>
<td>Adjusting steps deficit</td>
</tr>
<tr>
<td></td>
<td>Unilateral 8 µg/2 µl</td>
<td>n.d.</td>
<td>&gt;95</td>
<td>Apomorphine-induced rotations</td>
</tr>
<tr>
<td></td>
<td>Unilateral 8 µg/4 µl</td>
<td>n.d.</td>
<td>96</td>
<td>Deficit in (place and cue) Morris water maze</td>
</tr>
<tr>
<td>SNC</td>
<td>Unilateral 3 µg/1.5 µl</td>
<td>88</td>
<td>n.d.</td>
<td>Only when individual DA depletions were &gt;96% then apomorphine-induced rotations were observed</td>
</tr>
<tr>
<td>or L</td>
<td>Unilateral 4 µg/2 µl</td>
<td>n.d.</td>
<td>80-100</td>
<td>Deficit in paw retraction test, adjusting steps, and locomotor activity</td>
</tr>
<tr>
<td></td>
<td>Bilateral 4 µg/1 µl</td>
<td>n.d.</td>
<td></td>
<td></td>
</tr>
<tr>
<td>CPu</td>
<td>C+L</td>
<td>27 (SN+VTA)</td>
<td>40</td>
<td>No effect on staircase test or apomorphine-induced rotations</td>
</tr>
<tr>
<td></td>
<td>C+L</td>
<td>47 (SN+VTA)</td>
<td>54</td>
<td>No effect on staircase test or apomorphine-induced rotations</td>
</tr>
<tr>
<td></td>
<td>C+L</td>
<td>62 (SN+VTA)</td>
<td>82</td>
<td>Staircase test deficit and apomorphine-induced rotations</td>
</tr>
<tr>
<td></td>
<td>L</td>
<td>75</td>
<td>80-95 in lateral CPu</td>
<td>Adjusting steps deficit and staircase test deficit</td>
</tr>
<tr>
<td></td>
<td>M</td>
<td>53 in rostral and 74 in caudal CPu</td>
<td>Motor initiation and response inhibition deficits (RT task)</td>
<td>2</td>
</tr>
<tr>
<td></td>
<td>VC</td>
<td>n.d.</td>
<td>n.d.</td>
<td>Adjusting steps deficit</td>
</tr>
<tr>
<td></td>
<td>DL</td>
<td>n.d.</td>
<td>n.d.</td>
<td>Adjusting steps deficit</td>
</tr>
<tr>
<td></td>
<td>VL</td>
<td>n.d.</td>
<td>n.d.</td>
<td>Motor initiation deficits (RT task)</td>
</tr>
<tr>
<td></td>
<td>VL</td>
<td>60-64</td>
<td>53-77</td>
<td>Deficit in FR10, adjusting steps, and (place) Morris water maze</td>
</tr>
<tr>
<td></td>
<td>C</td>
<td>80</td>
<td>20-40 in rostral and 60-70 in caudal CPu</td>
<td>No effect on apomorphine-induced rotations, locomotor activity, and staircase test. Deficit in adjusting steps</td>
</tr>
<tr>
<td></td>
<td>C</td>
<td>80</td>
<td>20-40 in rostral and 60-70 in caudal CPu</td>
<td>No effect on apomorphine-induced rotations. Deficit in locomotor activity, staircase test, and adjusting steps</td>
</tr>
</tbody>
</table>

Abbreviations: 6-OHDA, 6-hydroxydopamine; DA, dopamine; MFB, medial forebrain bundle; SNC, substantia nigra pars compacta; Apo (+), rats rotating after apomorphine administration; Apo (-), rats not rotating after apomorphine administration; RT, reaction time; FR, fixed ratio; M, medial; L, lateral; D, dorsal; V, ventral; C, central; n.d., not determined; CPu, caudate putamen complex; SN, substantia nigra; VTA, ventral tegmental area.

digms than unilateral 6-OHDA lesions (52). Thus, a bilateral model would be preferable in regard to compensatory mechanisms. In a rat model of PD compensation during the experiment complicates the interpretation of the data and therefore it is necessary to control for functional recovery or compensation after the lesion (see “Compensation” below). In models with bilateral lesions, compensation during an experiment can occur as well, but it is excluded that this is mainly due to sprouting of axons from the other side of the brain. There are, however, some limitations. An obvious one is that in a bilateral model administration of drugs acting on the dopaminergic neurotransmission will not lead to rotational behavior since there is no DA imbalance between the two brain sides. In addition to the lack of rotational behavior, the forelimb use asym-
Compensation

Compensation or recovery of functions can be achieved via regeneration of dopaminergic projections from remaining dopaminergic tissue. For instance, it has been shown that unilaterally lesioning the SNC led to a sprouting of dopaminergic fibers in the ventrolateral part of the CPu (9). In the other areas of the CPu, the TH immunostaining of fibers on the lesioned side seemed to be the comparable to the control side. The sprouting of dopaminergic fibers in the ventrolateral CPu was only observed 4 and 7 months after lesioning, but not after 10 days. Since it appeared that the TH-immunoreactive fibers in the ventrolateral CPu were less numerous at 4 months than at 7 months after lesioning, it was suggested that there exists an ongoing process of regrowth of dopaminergic fibers in this part of the CPu up to 7 months (9). This regrowth of dopaminergic fibers is likely to be a compensatory response to a diminished nigrostriatal DA innervation. Assuming a homogenous dopaminergic innervation of the CPu by the nigrostriatal pathway, these latter data would also indicate a differential regrowth of DA fibers in CPu subregions. Moreover, besides compensation that is mediated by sprouting from remaining intact DA fibers of the lesioned side, compensation on the behavioral level can also be mediated by sprouting from DA fibers from the ipsilateral nonlesioned side. This is supported by the recent finding that bilaterally lesioning of the CPu caused additional behavioral deficits besides those observed after unilaterally lesioning the CPu (52). The emergence of these additional behavioral deficits can be explained by the fact that a bilateral lesion reduces the possibility of compensation by sprouting from the intact brain side.

Barneyoud et al. (5) reported recovery of function in a fixed-ratio-bar-pressing task after a partial dopaminergic lesion (70% reduction in CPu DA levels). This recovery of function did not apply to all behavioral parameters studied. This recovery of function is possibly due to compensatory mechanisms like sprouting from remaining dopaminergic fibers. Since different behavioral functions are mediated by different parts of the CPu, the recovery of function of some behavioral parameters in time might be due to its mediation via the part of the CPu that is subject to sprouting. It should be noted, however, that other compensatory responses (e.g., elevated DA biosynthesis, metabolism, and release by the remaining dopaminergic neurons) could also contribute to the recovery of function. It has, for instance, been reported that normalization of extracellular DA levels seems to be sufficient to account for recovery of function (49, 50). This was suggested since the time course of increases in extracellular DA levels and behavioral recovery were similar, in contrast to those for DA biosynthesis, metabolism, and DA release (49). In addition to various compensatory responses, even other brain structures (outside the CPu) can be involved in the functional recovery of some of the behavioral parameters. Thus, it should be noted that regenerative processes occur in the 6-OHDA model whenever the lesion is not complete (9).

When inducing a DA lesion in rats, the rats will go from a state of having no parkinsonian symptoms (before the lesion) to a state of displaying severe parkinsonian symptoms. From this moment on compensatory mechanisms will come into action to antagonize the neurobiological deficits. This means that PD symptoms in the rat will be alleviated to some extent in the course of time. In contrast, human idiopathic PD is a progressive disease with a clear reverse development: PD symptoms will worsen in the course of time. This contradiction between the idiopathic situation and the situation in the animal model is illustrated in Fig. 4. It is important to keep in mind that when behaviorally testing the lesioned rat at time point A, time point A’ in the human disease is mimicked (see Fig. 4). In addition, it is also relevant to notice that not all behavioral parameters are subject to compensatory mechanisms as was shown by Barnéoud et al. (5).

Variation Within the 6-OHDA Model

A further important point to note is that when 6-OHDA is injected into the MFB, SNC or CPu, there will always be some variability among the lesioned animals. This was shown by several investigators (e.g., Refs. 2, 6, 60). Although the target of toxin injection is exactly the same among the animals, the regional dis-
The distribution of TH immunoreactivity in the CPU will vary among the rats. This seems to be a drawback of the 6-OHDA model of PD. Furthermore, lesions do not only vary among animals in regard to the extent of the lesion, but in the bilateral model the lateral distribution of TH immunocytochemistry is subject to variation as well (2). Figure 5 shows two cross sections of a rat brain that was bilaterally lesioned in the CPU. The two sides of the brain show different intensities in TH-immunoreactivity staining. This means that although the lesion was aimed to be symmetrical between the two brain sides, the DA depletion differed.

In conclusion, independent of the site of injection, the 6-OHDA-induced DA depletion appears to be a valuable model to investigate PD symptomatology and to gain more insight into the possible pathological mechanisms of this neurodegenerative disease. However, the choice of using a bilateral model or a unilateral model depends on the aim of the experiment and one should be aware of the consequences of the choice for a certain 6-OHDA model.

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