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REVIEW ARTICLE

Environment, Genetics and Idiopathic Parkinson's Disease

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ABSTRACT: Since Idiopathic Parkinson's disease (IPD) was first described more than 170 years ago, there have been major advances in the understanding of the etiology of the disease as well as in its treatment. This article will review current knowledge concerning the role of the environment, genetic hypotheses and the aging factor in the etiology of IPD and proposes a complex interaction involving all these factors. Hypotheses regarding mitochondrial inhibition and free radical generation in IPD are discussed in relation to the mechanism of action of neurotoxins known to produce parkinsonian syndromes.

RÉSUMÉ: Depuis sa description initiale il y a un peu plus de 170 ans, de nombreuses découvertes fondamentales et cliniques ont permis de mieux compredre la maladie de Parkinson. Cet article détaille certaines observations récentes concernant le rôle possible de l'environnement, la génétique et le vieillissement dans l'étiologie de la maladie de Parkinson. Les hypothèses concernant l'inhibition mitochondriale et la génération de radicaux libres dans la maladie de Parkinson sont discutées en rapport avec les mécanismes d'action de neurotoxines connues comme produisant des syndromes parkinsoniens.

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Idiopathic Parkinson's disease (IPD) is characterized by major clinical disturbances caused by dopamine depletion in the corpus striatum, resulting from neuronal loss in the substantia nigra (SN).[1] The dopamine depletion is the result of a severe degeneration of the dopaminergic nigro-striatal pathway which normally inhibits the activity of a subpopulation of striatal neurons. While the physiological and neurochemical consequences of the dopamine depletion have been extensively documented, very little is known about the underlying cause of dopamine cell death or the mechanism by which dopamine cells degenerate. Numerous hypotheses have been presented including the following: a) IPD is the result of a random process.[2] This hypothesis assumes that the age-related loss of neurons can be accelerated by the cumulative action of repeated insults of ordinary life (viruses, toxins, head injury, etc.). A revised form of this concept is also known as the "accelerated aging hypothesis".[3] b) IPD is the result of a lack of neurotrophic hormone.[4] It is hypothesized that a dopaminergic-specific growth factor is synthesized and stored in the target striatal cells and that the inability of these cells to provide the required dopaminergic neurotrophic hormone, thereby causes impairment to SN neurons. c) IPD is caused by a defect in the DNA repair mechanism.[5] This hypothesis is derived from observations showing that cultured fibroblasts from parkinsonian patients are hypersensitive to the lethal effects of DNA-damaging agents such as x-rays. It postulates that somatic mutations occurring in early embryogenesis cause a lifelong accumulation of unrepaired DNA damage in neurons, resulting in lethal consequences on the SN. d) IPD is the result of a specific genetic defect(s) or genetic predisposition(s).[3,6,7] It is proposed that IPD is not a clear-cut genetic entity but that genetic factors play a role in its development. This model generally implies the presence of one or more inherited "susceptibilty factors" as a background to the effect of environmental agents. e) IPD has a viral etiology.[8,9] It is proposed that conventional viruses may produce IPD as a rare complication of systemic infection. Evidence for this hypothesis has been sought from studies on viral antibody titres. f) Finally, IPD is triggered by toxic compounds present in the environment (internal and external to the body) that have short- or long-term effects on neuronal function. [10-12] The case of the N-methyl-4-phenyl,1,2,3,6-tetrahydropyridine (MPTP) is one of the best examples of the environmental trigger factor hypothesis. This compound, a

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meperidine derivative, produces a chronic, irreversible parkinsonism in human subjects receiving the compound by injection.[10] This condition can be symptomatically treated with the standard L-DOPA therapy. It is thus postulated that people exposed to products with similar chemical conformations found in industrial compounds or in nature could develop IPD.

While all these hypotheses account for some of the well defined abnormalities related to IPD, none of them fully explain the cascade of events responsible for the initiation of IPD or identify its cause. We will endeavour to review old and new emerging concepts, while trying to integrate the neuropathological, the neurochemical and the genetic information related to the etiology of IPD.

Epidemiology

Prevalence of IPD in various countries shows a variation from 59 per 100,000 in China[13] to 353 per 100,000 in USA[14] with intermediate values for Sweden, Japan, Italy, Finland and many other countries.[15-22] This epidemiological data also indicates that incidence and prevalence rates of IPD are dependent on age, with cases below the age of 40 rarely encountered. The differences in methodology, case-ascertainment and age-structure of the varied populations, may explain the variation in the observed prevalence. In North America, the age-adjusted prevalence is the same for men as for women aged 40+ and is also the same for both black (338/10,000) and white 353/100,000) populations.[14]

Aging, Neuromelanin and Iron

The incidence of IPD increases at a steady rate until the sixth or seventh decade of life. Accordingly, the aging factor itself appears to play an important role in the etiology of IPD as a key susceptibility factor. The loss of striatal dopamine occurring normally as aging progresses[23] and age-related accumulation of neuromelanin in the SN are both believed to be trigger factors involved in IPD.[24-25] Recently, a direct relationship between the distribution of neuromelanin-bearing neurons in the SN and the pattern of cell loss in IPD has been elucidated.[26] These results suggest that the neuromelanin which plays a role during normal aging may also be involved in IPD and that the trigger factor(s) responsible may cause an acceleration of the normal aging processes.

Because increased iron concentration has been heralded as a potential catalyst of free radical reactions, it has been carefully examined in relation to IPD. Post-mortem studies reveal that the parkinsonian brain contains significantly more iron than that of age-matched controls,[27-30] particularly in the SN and striatal areas. Clinical studies[31] using desferoxamine (an iron chelator) indicate that parkinsonian (IPD) patients excrete significantly more iron in urine during a 6 hour period than age- and sex-matched control subjects, suggesting that parkinsonian (IPD) patients have a larger reserve of mobilizable iron. Interestingly, the age-related accumulation of iron in the SN and striatum appear to plateau around 50-60 years of age, the same age range at which IPD is usually clinically detectable. As a consequence, it has been proposed that the pathological accumulation of iron in IPD may result in increased local oxidative stress with toxic accumulation of free radical species and lipid peroxidation.[32-33]

Pattern of Neuronal Loss

The loss of pigmented neurons in the SN is one of the key features of IPD. As the nigral neuron numbers decrease in IPD, the severity of the symptoms increase. While not directly involved in the clinical course of the disease, other brain regions such as the thalamus, hypothalamus, neo-cortex, locus coeruleus, ventral tegmental area, dorsal nucleus of the vagus and nucleus basalis of Meynert show moderate to severe neuronal loss.[34] Moreover, the regional pathophysiology of IPD is sometimes accompanied by features characteristic of Alzheimer's disease such as dementia in the more advanced stages.[35]

Genetic Factors

A subgroup of IPD patients with early age-of-onset were shown to have classical autosomal dominant inheritance in their families.[6] There is also a juvenile form of parkinsonism which shows a clear autosomal recessive mode of transmission.[36] More recently, two large kindreds were reported[7] with evidence of a single gene action modulated by environmental factors affecting duration of the disease. All these studies suggest some degree of genetic influence in one or more pedigrees.

Studies employing the technique of twin analysis have also been used to further evaluate the genetic component of IPD. Amongst a total of 116 twin pairs, 5 siblings developed IPD (Table 1). This value lies within the predicted prevalence for the general population.[37] Authors concluded that **a**) genetic factors do not play an essential role in the etiology of IPD and **b**) since twins generally grow up in the same environment until adulthood, environmental risk factors which may be involved in the disease, should be sought after adolescence. These twin studies were recently reappraised by Johnson and co-workers based on clinical and statistical grounds.[38] A careful examination of the data from the American Study[39] reveals that the monozygotic twin concordance rate significantly differs from the dizygotic twin concordance rate, suggesting a genetic component to the disease, since dizygotic twin pairs share non genetic factors to the same extent as do the monozygotic twins. A second problem with that particular study is the lack of data assessing the risk of future development of symptoms in the unaffected co-twins. Since the duration of the disease varies from 1.5 to 29 years, it is conceivable that co-twins, unaffected at the time of the study, subsequently developed clinical manifestations. The addition of only a few cases would have considerable impact on the genetic

Table 1. Parkinson's Disease in Twins

Study Group	MZ/Concordant	DZ/Concordant
Ward et al, 1983[39]	43/1	19/1
Marsden, 1987[89]	11/1	11/1
Martilla et al, 1988[37]	18/0	14/1
Subtotal	72/2	44/3
TOTAL	116/5	

MZ: Monozygotic
DZ: Dizygotic

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analysis. The authors concluded that while very informative, the twin studies to date do not rule out a genetic component for IPD.

Furthermore, recent findings on xenobiotic metabolism, which leads to a deficiency in detoxification pathways involving sulfur metabolism,[40] have led to a different interpretation of the genetic data. Barbeau et al[41] initially proposed the ecogenetic hypothesis stating that the susceptibility to IPD is caused by an interplay of environmental toxins and the endogenous capacity of the body to detoxify itself, the latter being dependent on the genetic background. It is well known that pesticides and specific drugs normally induce specific liver enzymes (P-450s) which are responsible for the metabolism of these toxic compounds.[42] Accordingly, it was postulated that parkinsonian patients may be genetically deficient in specific detoxifying enzymes and thus incur the risk of overexposure to normally inoffensive levels of environmental toxins. Studies of sulfoxidation of S-carboxymethyl-L-cysteine by the cysteine dioxygenase revealed that 35% of the IPD patients (n=68) produced no S-oxide metabolite compared to 2.5% in the general population.[40] Similarly, 29% of the parkinsonian patients examined excreted more than 5% of the sulfated-acetaminophen compared to 84% of the general population. Authors pointed out that the increased incidence of IPD with age could be explained by a toxin encountered over the years of exposure by susceptible individuals. These results are suggestive of a deficiency in the detoxification pathways involving sulfur metabolism in IPD that could have a major impact with respect to a genetic predisposition to environmental toxins.

Endogenous and Exogenous Toxicity

Infection-related Toxicity

Many years ago, it was revealed that viral infections like poliomyelitis selectively affects areas of the central nervous system (CNS) in humans. In the early 1920's, a pandemic of von Economo's encephalitis produced clinical symptoms similar to IPD with the classic striatal dopamine loss.[43] However, the presence of certain symptoms and pathological changes in the infected individuals clearly differentiate this disease from IPD. Different strains of viruses have also been examined in IPD (measles, varicella, poliovirus, herpes, influenza), none of which have been able to link IPD to a specific viral infection.[8,9,43] Injections of parkinsonian brain tissue extracts to primates failed to induce IPD in these animals, ruling out possible infections caused by slow viruses.[44] However, it is quite possible that viruses have different effects in different primates.

Free Radical Toxicity

One of the basic mechanisms initially proposed to explain brain cell loss in IPD was the production of a toxic free radical species. Free radicals will damage a cell if their concentration at any given moment exceeds its endogenous antioxidant capacity. Enzymes such as catalase, glutathione peroxidase and superoxide dismutase represent the first line of defense against these toxic species (Figure 1). It is thus of interest that their nigral concentration has been found to be decreased[30,45-48] or unchanged[49] in the CNS of patients with IPD. Similarly, antioxidant blood levels were shown to be decreased[51] or unaffected[49,51] in living IPD patients.

As we will see later, toxins such as 6-hydroxydopamine, manganese and MPTP which are used to produce parkinsonian syndromes in animal, have been shown to generate and/or catalyse the formation of toxic free radicals.[52-55]

While there is no firm evidence to support or contradict the free radical hypothesis of the SN cell death, this concept has led to the development of new therapeutic approaches towards IPD.

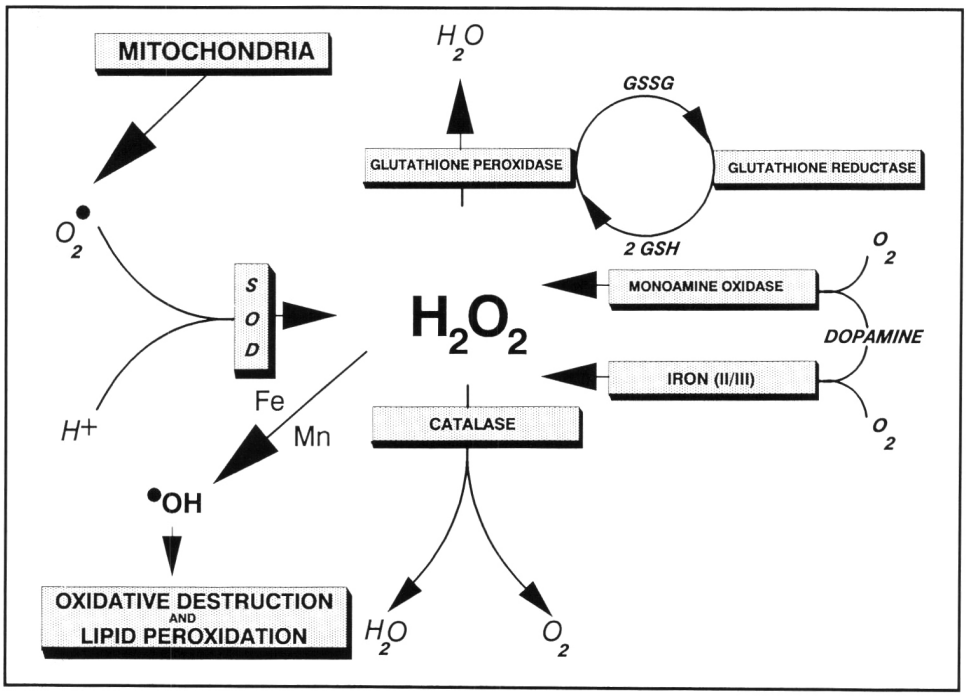

Figure 1 — Schematic representation of the key antioxidant systems presumably involved in the protection of dopaminergic neurons. $O_2^{\bullet}$: superoxide radical, $OH^{\bullet}$: hydroxyl radical, GSH and GSSG: reduced and oxidized form of glutathione.

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Vitamin E, a free radical scavenger, is currently being tested for its ability to modify the progression of IPD[56] while deprenyl, a monoamine oxidase B inhibitor known to slow down dopamine degradation and hydrogen peroxide production, is successfully being used to reduce the progression of IPD and delay the administration of L-DOPA to IPD patients.[57,58] Research on the role of free radical toxicity in the CNS remains one of the most active fields of investigation in IPD.

Neurotoxins

Several drugs and chemicals have been shown to induce parkinsonian syndromes causing effects ranging from transient to permanent, particularly agents that alter dopaminergic transmission in the CNS. Some of these compounds induce parkinsonism by affecting neurotransmitter synthesis, storage and release, or receptor sites. These include reserpine[59] and dopamine antagonists such as haloperidol. Other compounds act differently and damage or even kill neuronal cells. Toxins such as manganese, carbon disulfide, 6-hydroxydopamine and MPTP will selectively destroy specific subsets of neurons in the CNS. This group of toxins is of great concern since it irreversibly destroys specific neurons and produces irreversible parkinsonian symptoms with similar pathophysiological changes reported in IPD.

Manganese

Manganese (Mn) intoxication was first reported in mining workers, involved in the extraction and processing of Mn.[60] Chronic Mn toxicity was shown to produce an irreversible syndrome closely resembling IPD.[61,62] However, neuropathological investigations revealed that, in addition to the nigral cell loss that characterizes IPD, Mn intoxication also produced significant cell loss in the striatal and pallidal areas. Lewy bodies which are considered to be reliable pathological markers of IPD have never been found in Mn-induced parkinsonism. The mechanism by which Mn destroys neuronal cell remains unknown at this time. However, evidence of the oxidative properties of Mn suggest the possible involvement of a free radical species. Mn was shown to potentiate dopamine oxidation *in vitro* and produces oxygen-derived free radicals.[54] It can also catalyse the formation of toxic hydroxyl radicals from hydrogen peroxide (Figure 1) synthesized during dopamine oxidation by monoamine oxidase or Mn^{+2}. The accumulation of a black pigment in the cells of the caudate nucleus (which do not normally contain melanin) of patients with Mn-intoxication is rather suggestive of an accelerated dopamine oxidation.[63] Despite significant parallels between Mn intoxication and IPD, the concentration of Mn in the general environment is not important enough (as it is in mining operation) to be considered a good candidate in the etiology of IPD. However, the postulated mechanism by which it seems to destroy cells may also apply to the SN cell loss observed in IPD.

6-hydroxydopamine

Originally studied for its toxic effect of peripheral noradrenergic neurons,[64] 6-hydroxydopamine (6-OHDA) became well known for its effect in the CNS. Since it does not cross the blood brain barrier (BBB), it has to be administered intraventricularly, intracisternally or directly into the brain regions of interest. It was shown to selectively destroy central dopaminergic and noradrenergic neurons.[65] Although exogenous 6-OHDA cannot be easily implicated as an etiological agent in IPD, it has been widely used to mimic selective aspects of the disorder. In primates, intranigral administration of 6-OHDA was shown to produce a behavioral syndrome similar to parkinsonism.[66] Two of the most commonly offered mechanisms used to explain 6-OHDA toxicity involve the production of a free radical species (superoxide and hydrogen peroxide) and quinone oxidation products which also involve unstable free radical intermediates.[55,67] It has been postulated although not demonstrated that the free radical species produced during 6-OHDA oxidation in neurons react with several cellular components and destroy the cellular organization of the neurons.

N-methyl-4-phenyl-1,2,3,6-tetrahydropyridine

1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) has been shown to cause destruction of dopaminergic neurons of the SN in humans,[10] monkeys[68] and mice.[69] Inhibition of monoamine oxidase B (but not MAO A) which converts MPTP to its metabolite N-methyl-4-phenyl pyridinium ion (MPP+) was shown to effectively prevent most of the SN cell loss. Other

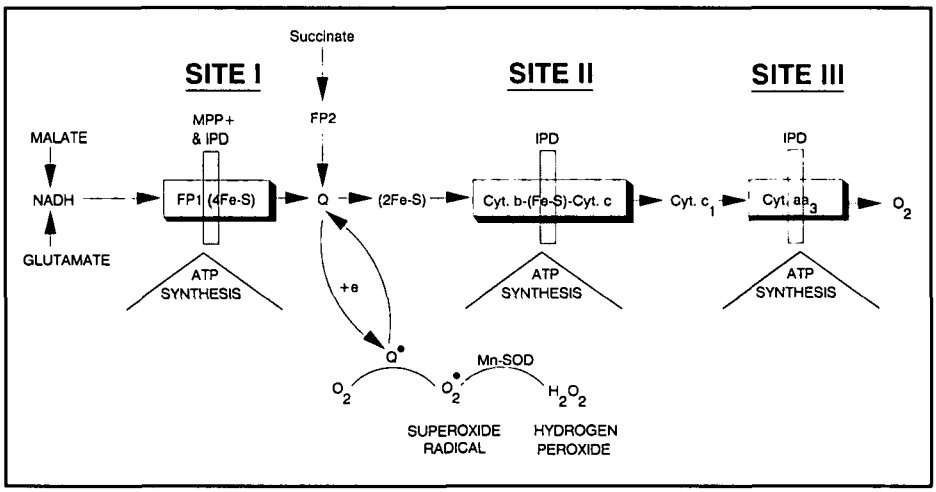

Figure 2 — Schematic representation of the mitochondrial respiratory chain and the sites affected by MPP+ neurotoxicity, and in patients with Parkinson's disease.

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minor pathways of transformation involving free radical intermediates have been described elsewhere.[53]

Although there are many conflicting reports regarding the effects of antioxidant administration on MPTP-induced dopamine loss,[70-75] high doses of antioxidants like vitamin E and ascorbic acid appear to dampen the reduction of catecholamine levels but do not prevent cell loss.[70] On the other hand, MPTP failed to affect the survival of hepatocytes exposed to diethylmethylmaleate, an agent that decreases the intracellular level of reduced glutathione by more than 80%,[74] suggesting that the toxicity is not caused by an alteration of the antioxidant capacity of the cells.

It is now believed that MPTP exerts toxic activity through the formation of MPP+ which in turn accumulates in the mitochondria[76] and inhibits the respiratory chain at Site I (Figure 2).[77,78,79] As a consequence, there is a rapid depletion of ATP levels,[73] an accumulation of NADH and lactate[78] and a significant alteration in the cellular calcium content.[80]

Because of the strong similarities between the MPTP syndrome and IPD at the neurochemical and neuroanatomical level as well as in response to L-Dopa therapy, it is now suspected that a compound similar to MPTP may be involved as a cause of IPD. Accordingly, the mechanisms by which MPTP appears to mediate its toxicity have been thoroughly examined in postmortem tissues as well as in living IPD patients which enabled the analogies between both entities to undergo further analysis.

Altered Mitochondrial Function in IPD

Several articles concerning the fate of mitochondrial respiration have recently been published. Inspired by the effect of MPP+ on the mitochondrial respiration, defects in the respiratory chain function in cells of parkinsonian patients have been identified.[81-84] A significant and specific reduction of complex I activity was found in IPD patients (Figure 2) at the level of the SN[81,84] and striatum.[83] However, at the moment, it is not possible to determine the extent of the glial cell contribution to this effect. Interestingly, the loss of Complex I activity was also found in skeletal muscles of living IPD patients (in addition to a loss of Complex II and IV activities).[82]

The similarity of the findings in the MPTP model and the loss of mitochondrial function in IPD suggests that Complex I deficiency may be directly related to the primary disease process, responsible for the nigral cell loss in IPD. Impaired electron transport combined to an abnormal oxidative phosphorylation could account for the cell loss.

Idiopathic Parkinson's disease: A Working Hypothesis

As one theory alone does not explain the etiology of IPD, evidence suggests several components may be involved, including a genetic predisposition. It is quite *possible* that IPD is the result of an interplay of environmental toxins and the aging factor combined with a specific genetic predisposition favouring the development of the disease process. Thus, IPD could be caused by an MPTP-like environmental toxin(s) of unknown origin which is(are) taken up by the body and accumulate over the years. Predisposed individuals who are slow metabolizers of such compound(s) are exposed longer and more intensely than fast metabolizers. The(se) compound(s) can cross the BBB, and accumulate in specific areas of the brain, providing that there are specific uptake systems and neuromelanin bearing cells which could permit a long-term trapping of the compound(s). The loss of dopaminergic neurons associated with age, leads to the release of the compound(s) which are then taken up by remaining neurons, increasing the cell concentration but not the region concentration. Iron which normally accumulates during aging in the SN and striatum[85] adds to the local oxidative stress. Finally, as is the case of the MPP+ mechanism, the compound gradually inhibits (weakly at first) the neuronal respiratory function[81-84] until it finally destroys the cellular integrity in IPD. As neurons die, glutathione levels drop,[30,46,48] iron and other transition metals accumulate[28-30] in the remaining cells which are metabolically more active (compensatory phase), lipid peroxidation increases[86] and the energy level gradually diminishes. The antioxidant system becomes less efficient[45-47] or, is no longer able to cope with the oxidative stress produced by the increased activity of the superoxide dismutase.[49,87,88]

This cascade of events, while being relatively complex, can be easily investigated using the MPTP model shortly after the injection of MPTP, when the accumulation MPP+ reaches its peak. Alternatively, animals could be exposed to moderate levels of MPTP for long periods of time. In both cases, one should take advantage of the fact that aged animals are more susceptible to MPTP than young ones.

Conclusions

We propose that neurological damages characterizing IPD are determined by many factors including individual response (presumably genetically predisposed) to toxin(s), the chemical nature of the toxin (which would require crossing of the BBB), the presence of specific uptake systems (catecholamine-related), age-related accumulation of neuromelanin, iron and other transition metals, and the access to a mitochondrial transporter.

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