Practical Problems in Neurotoxicology

Controversial topics in neurotoxicology

- Drug-Related
  - D-fenfluramine (Fen-Phen)
  - Ecstasy (MDMA)
- Diet-related
  - Aspartame
  - Food colors
- Environment-related
  - aluminum
  - lead
  - mercury
  - Neurodegenerative disorders
    - Parkinson’s disease
Parkinson’s Disease Background

- Idiopathic (sporadic) neurological condition
- Constellation of neurological signs:
  - tremor, rigidity, bradykinesia, inability to initiate movement
- Average age of onset = 55 yr
- Caused by the death of dopamine nerve cells in one specific brain region
- Marked by pathological and chemical changes
What does Parkinson’s disease look like?
Older Man – Severe PD

Younger Man - PD
Parkinson’s disease and dopamine systems

- Frontal Cortex
- Gyrus Cinguli
- Corpus Callosum
- Basal Ganglia
- Nuc. Accumbens
- Olfactory Tubercle
- Hypothalamus
- Pituitary
- Medial Forebrain Bundle
- Tegmentum
- Entorhinal Cortex
- Sub. Nigra
- Medial Forebrain Bundle
- Tuberoinfundibular System (Median eminence)
- Midbrain
- Nigrostriatal System
- Mesolimbic/cortical System
Parkinson’s Disease: Loss of Nigrostriatal Dopamine Neurons

Substantia Nigra

HO

HO

NH₂

Parkinson’s Disease: Pathogenesis

Control    PD
How loss of dopamine affects motor function

Notes:
• Direct pathway = D1 mediated
• Indirect pathway = D2 mediated
• Symptoms reported at >60% degeneration; degeneration is progressive.
• Other neuroreceptors play minor role (A2A; etc.)

Young PD Man (baseline)

[Image: Young PD Walk.avi]
Who gets and what causes Parkinson's Disease?
Age-Specific Incidence of Parkinson’s Disease

<table>
<thead>
<tr>
<th>Age (Years)</th>
<th>Female PD Incidence (per 100,000 patient years)</th>
<th>Male PD Incidence (per 100,000 patient years)</th>
</tr>
</thead>
<tbody>
<tr>
<td>30</td>
<td>50</td>
<td>50</td>
</tr>
<tr>
<td>40</td>
<td>100</td>
<td>150</td>
</tr>
<tr>
<td>50</td>
<td>200</td>
<td>250</td>
</tr>
<tr>
<td>60</td>
<td>250</td>
<td>300</td>
</tr>
<tr>
<td>70</td>
<td>300</td>
<td>350</td>
</tr>
<tr>
<td>80</td>
<td>350</td>
<td>400</td>
</tr>
</tbody>
</table>

Kaiser population (Van Den Eeden et al. 2003)

Possible Causes of Parkinson's Disease

- Genetic mechanisms
  - A few specific mutations can cause rare (familial) Parkinson’s.
  - Other genes may increase susceptibility (speculative).
- Biological insults
  - Influenzae (e.g., von Economos’ encephalitis re. Awakenings)
- Chemicals
  - Manganese
  - Carbon monoxide
  - Cycad fruit (Guam)
  - MPTP
Manganese Intoxication

- First noted with manganese miners more than 150 years ago.
- Symptoms of manganese are similar to those of Parkinson’s disease.
- Exposed individuals develop the clinical signs and symptoms of Parkinson’s much earlier than typical people with PD.

Parkinsonism and Environmental Exposure: the Cycad in Guam

Toxins
- BOAA (excitatory amino acid)
- MAM (cytotoxin)
Some other chemicals that can induce parkinsonism

- Carbon monoxide
- Cyanide
- Carbon disulfide
- Rotenone
- MPTP (The Case of the Frozen Addict)

- However, all proven toxicants account for less than 0.5% documented cases of PD.

Does all chemical exposure increase risk of PD?
Coffee consumption DECREASES Risk of PD!!

Since the 1960s, over 40 case-control studies have shown an inverse association of PD and smoking.

Smoking decreases risk of PD!!!

p < 0.0001

Duration of Smoking (years)
Risk Factors

Factors associated with increased risk:
- Age: >50 yr
- FH of PD
- Hx of trauma, emotional stress, personality (shy, depression)
- Hx of exposure to toxins such as herbicide, pesticides, solvents, MPTP and MPTP-like compounds
- Race: Caucasians > Asians > African Americans
- Low cholesterol

Factors associated with decreased risk:
- Diet: Vitamin E, MVI, cod liver oil
- Cigarette smoking
- Drinking alcohol
- Drinking coffee

Genetic Mechanisms

- α-synuclein
- DJ-1 (PARK7)
  - In knockout mice, no loss of nigral neurons or DA content; DA reuptake affected
- Parkin
  - large gene on Chr 6
  - ubiquitin ligase
  - In KO mice, reduced response to acoustic stimulus as mice age
- kinases
  - LRRK2
  - PINK1 (PARK6)
Many factors interact to make the “Parkinson's Pie”

- Single Gene
- Multiple Genes
- Single Chemical
- Single gene & chemical
- Multiple Chemicals
- Complex interactions

How does one investigate the potentially complex relationship between genes and environment in causing this disease?

Case story: MPTP (“The good, the bad, and the ugly”
MPTP & Parkinson’s disease

Story beginning: Medicinal Chemistry
(an easy to make opioid/narcotic)

- Meperidine (Demerol)
- 4-propyloxy-4-phenyl-N-methylpiperidine (meperidine analog)
- MPTP

Addicts accidentally synthesized MPTP, a meperidine analog, which caused Parkinson’s disease.
MPTP - Frozen Addict

Kitchen Chemistry & PD

Meperidine

Death of DA neurons: parkinsonism!!!

MPP⁺

MPTP

4-propyloxy-4-phenyl-N-methylpiperidine (meperidine analog)

addicts accidentally synthesized...
MPTP dramatically decreases tyrosine hydroxylase positive neurons in substantia nigra

---

A role for endogenous dopamine in PD?

- Two mechanisms for oxidative breakdown of dopamine create oxidative stress
  - Metabolism of dopamine with MAO-A or MAO-B results in formation of hydrogen peroxide, which is then converted to hydroxy radical by the Fenton reaction
  - Nonenzymatic auto-oxidation of dopamine to semiquinone produces superoxide radicals and hydrogen peroxide.
- Dopamine is neurotoxic to cells in culture
  - markers for lipid peroxidation and apoptosis are observed
- Deficiencies in mitochondrial electron transport have been observed in PD, but may represent a consequence, not a cause of the disease
Environmental agents as cause of PD

- The herbicide paraquat is very similar structurally to MPP⁺.
- MPTP data led to hypothesis that MPTP-like compounds might have been environmental cause of disease.

Class Discussion:

- Is paraquat neurotoxic, and what factors should you consider in formulating an hypothesis?
- What factors do you consider in testing the more global hypothesis related to causation by environmental agents?
- What factors do you consider in determining how or if to use MPTP as an animal model for elucidating either causative or preventive mechanisms in PD?