Integrating Literature Analysis into the NTP Research Pipeline

Windy Boyd
NTP Board of Scientific Counselors Meeting
December 12, 2018
Evidence Mapping
- Inform Research
  - Data pockets
  - Data gaps

Literature Analysis

Define Hypotheses & Design a Testing Strategy
- Knowledge Integration
- Longer-term in vivo Tests
- Short-term in vivo Tests

Inform Public Health Decisions

Communication

Data Mining
- QSAR Profiling
- Bioactivity Screening

In vitro Studies
• Parkinson’s disease (PD) due to progressive neurodegeneration
  – Aggregation of $\alpha$-synuclein in Lewy bodies
  – Loss of dopaminergic neurons in substantia nigra
  – Signs include tremor, rigidity, and shuffling gait
• Highly prevalent but etiology of most PD cases unknown
  – Genetics only account for ~10% of cases
Exposures to pesticides linked to Parkinson’s in epidemiological studies

Need for better understanding of which environmental factors may be contributing and how they act

Neurodegeneration is not included in routine toxicological testing strategies

Lack of methods to rapidly identify environmental exposures
NTP Parkinson’s Disease Project

Strategy to identify potential chemical contributors

• Project team
  – Combined scientific expertise in neurotoxicology, *in vitro* screening, toxicoinformatics, and literature analysis

• Goals
  – Identify previously evaluated chemicals, genes and pathways, and model systems
  – Develop a battery of *in vitro* and alternate model organism assays to screen chemicals for potential effects
NTP Parkinson’s Disease Project

Strategy to identify potential chemical contributors

- Expert knowledge
- Published literature
- Tox21 HTS data
- Chemical and assay selection
- Toxicogenomic databases
• Questions: Which chemicals, genetic targets, and models have been reported in the scientific literature?
• PubMed search identified >90,000 records with mention of Parkinson’s disease
• Screened studies for environmental chemical exposure and categorized by study characteristics
Exposures Associated with Parkinson’s Disease

Automated Tagging of All Environmental Exposures

- Metals (1734)
- Smoking (1163)
- Alcohol (778)
- Occupational (442)
- Air Pollution (235)
- Miscellaneous (172)
- PAHs (35)
- Stress (4339)
- Drugs of Abuse (2654)
- Pesticides (2576)
- Nutrition (2490)
Manual categorization of 1,840 studies revealed similar trend as automated tagging and allows researchers to explore published literature.
## Parkinson’s Disease Evidence Map

### Most-reported Environmental Chemicals

<table>
<thead>
<tr>
<th>Chemical</th>
<th>Exposure</th>
<th>Positive control</th>
<th>Human</th>
<th>In vivo</th>
<th>In vitro</th>
</tr>
</thead>
<tbody>
<tr>
<td>Manganese</td>
<td></td>
<td></td>
<td>115</td>
<td>106</td>
<td>98</td>
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<td></td>
<td></td>
<td></td>
<td>6</td>
<td>8</td>
<td></td>
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<tr>
<td>Paraquat</td>
<td></td>
<td></td>
<td>22</td>
<td>135</td>
<td>97</td>
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<td></td>
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<td></td>
<td>37</td>
<td>36</td>
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<td>Rotenone</td>
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<td>10</td>
<td>137</td>
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<td></td>
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<td></td>
<td>204</td>
<td>274</td>
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<td>Nicotine</td>
<td></td>
<td></td>
<td>21</td>
<td>36</td>
<td>17</td>
</tr>
<tr>
<td></td>
<td>Treatment</td>
<td></td>
<td>16</td>
<td>61</td>
<td>18</td>
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</table>
### Environmental Chemicals in >10 Studies

<table>
<thead>
<tr>
<th>Chemical</th>
<th>Human</th>
<th>In vivo</th>
<th>In vitro</th>
</tr>
</thead>
<tbody>
<tr>
<td>Maneb</td>
<td>8</td>
<td>43</td>
<td>22</td>
</tr>
<tr>
<td>Aluminum</td>
<td>17</td>
<td>14</td>
<td>14</td>
</tr>
<tr>
<td>Iron</td>
<td>17</td>
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<td>19</td>
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<td>Dieldrin</td>
<td>10</td>
<td>6</td>
<td>23</td>
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<td>Mercury</td>
<td>19</td>
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<tr>
<td>Copper</td>
<td>10</td>
<td>6</td>
<td>12</td>
</tr>
<tr>
<td>Lead</td>
<td>20</td>
<td>2</td>
<td>4</td>
</tr>
<tr>
<td>Cadmium</td>
<td>5</td>
<td>2</td>
<td>10</td>
</tr>
<tr>
<td>PCBs</td>
<td>9</td>
<td>3</td>
<td>3</td>
</tr>
<tr>
<td>Zinc</td>
<td>8</td>
<td>5</td>
<td>2</td>
</tr>
<tr>
<td>Mancozeb</td>
<td>1</td>
<td>7</td>
<td>4</td>
</tr>
</tbody>
</table>

- Very few chemicals with multiple reports
- All metals and/or pesticides except PCBs
- Many chemicals with single study (not shown)
Candidate Chemical Library

- **Predicted actives**
  - Positive controls
    - MPTP, rotenone, paraquat
  - Metals and metal compounds
    - Manganese tricarbonyl (MMT), maneb, methyl mercury, ziram
  - Organochlorines
    - DDT, heptachlor, dieldrin, lindane, endosulfan, TCE, hexachlorobenzene
  - Organophosphates
    - Chlorpyrifos, diazinon
  - Other pesticides
    - Permethrin, benomyl, tributyltin methacrylate, quintozene

- **Unknowns**
  - Triphenyl phosphate
  - Isopropylated phenyl phosphate
  - Captan
  - Glyphosate
  - Pyridaben
  - Acetaminophen

- **Predicted Negatives**
  - Saccharin sodium
  - L-ascorbic acid
  - D-glucitol
  - Acetyl salicylic acid
Informing Assay Selection for Targeted Testing

Motor deficits

Related changes in gene expression

Neuroinflammation

Mamta Behl
Tox Branch
**Parkinson’s Disease Evidence Map**

*Some studies may have characterized multiple health effects or species and therefore may be represented multiple times. Row and column grand totals represent counts of distinct references.*

### in vitro Effects of Paraquat Exposure

<table>
<thead>
<tr>
<th>Effect</th>
<th>Human</th>
<th>Rat</th>
<th>Mouse</th>
<th>Rat x Mouse</th>
<th>Bovine</th>
<th>Grand Total</th>
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<tbody>
<tr>
<td>DA (TH+) neurons</td>
<td>2</td>
<td>8</td>
<td>9</td>
<td></td>
<td></td>
<td>16</td>
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<tr>
<td>Dopamine (DA and metabolite levels, DAT and receptor expression, TH immunoreactivity)</td>
<td>4</td>
<td>13</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>11</td>
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<tr>
<td>alpha synuclein, Tau phosphorylation, tubulin</td>
<td>10</td>
<td>3</td>
<td></td>
<td></td>
<td></td>
<td>13</td>
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<tr>
<td>Proteasome (Parkin, proteasomal activity)</td>
<td>22</td>
<td>13</td>
<td>2</td>
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<td>37</td>
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<tr>
<td>Mitochondrial effects</td>
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<td>28</td>
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<td>2</td>
<td>1</td>
<td>89</td>
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<tr>
<td>Other (general expression changes, etc.)</td>
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<td>60</td>
<td>19</td>
<td>2</td>
<td>1</td>
<td>137</td>
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* SPECIFICITY

**More**

**Less**
Reported *in vitro* Models

<table>
<thead>
<tr>
<th>Cell line</th>
<th>Category</th>
<th>Cell, tumor, subfraction type</th>
<th>Tissue origin</th>
<th>Count</th>
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<tbody>
<tr>
<td>SH-SYSY</td>
<td>tumor</td>
<td>neuroblastoma</td>
<td>brain, bone marrow metastasis</td>
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<tr>
<td>SK-N-SH</td>
<td>tumor</td>
<td>neuroblastoma</td>
<td>nerve, bone marrow metastasis</td>
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<td>primary mesencephalic cultures</td>
<td>primary</td>
<td>neurons, glia</td>
<td>mesencephalon</td>
<td>22</td>
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<tr>
<td></td>
<td></td>
<td>neurons</td>
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<td>2</td>
</tr>
<tr>
<td>PC12</td>
<td>tumor</td>
<td>pheochromocytoma</td>
<td>adrenal gland</td>
<td>20</td>
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<tr>
<td>N27</td>
<td>transformed</td>
<td>neurons</td>
<td>mesencephalon</td>
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<tr>
<td>primary cerebral cortex cultures</td>
<td>primary</td>
<td>glia</td>
<td>cerebral cortex</td>
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<tr>
<td></td>
<td></td>
<td>microglia</td>
<td>cerebral cortex</td>
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</table>

✓ majority of studies conducted in human and rat tumorigenic cell lines with fewer more relevant, complex models
Selected genes associated with Parkinson’s
- Illumina’s NextBio datamining software
- Comparative Toxicogenomics Database (CTD)

Grouped 233 genes into 15 disease-relevant pathways

Linked genes to studies included in literature analysis

Gene expression reported in 47% of relevant studies
- 57% of 233 genes evaluated in those studies
Identifying Chemical-Gene Combinations

Chemical Classes
- Pesticides
- Nicotine
- Metals
- Other

Biological Pathways
- Apoptosis
- Adenosine Receptor
- Axon guidance/synaptic
- Dopamine
- Dopamine Receptor
- Immune System
- α-synuclein/Lewy body
- Neuronal survival/activity
- Other
- Other receptors
- Mitochondria/ox phos
- Transcription factor
- Transporter
- Ubiquitin

Nisha Sipes
BSB

Paraquat
Rotenone
PCBs BPA
Nicotine
Manganese
SNCA
PARK7
TH
SLC6A3
CASP3
PCBs BPA
Nicotine
Manganese
SNCA
PARK7
TH
SLC6A3
CASP3

CytoScape
Identifying Chemical-Assay Combinations

ToxCast and Tox21 Data

Legend:
- Screened in Tox21
- Active in Tox21
- Published literature
Strategy to identify potential chemical contributors

Expert knowledge

Published literature

Tox21 HTS data

Chemical and assay selection

Toxicogenomic databases
DNTP Translational Toxicology Pipeline Plan

**Literature Analysis**

- Data Mining
- QSAR Profiling
- Bioactivity Screening
- In vitro Studies
- ADME/Chemistry

**Define Hypotheses & Design a Testing Strategy**

- Knowledge Integration
- Longer-term in vivo Tests
- Short-term in vivo Tests

**Communicate**

- Inform Public Health Decisions
- Systematic Review

- Evidence Map

Informed

Next

Future?
<table>
<thead>
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</tr>
<tr>
<td>• Nisha Sipes</td>
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<tr>
<td>• Mamta Behl</td>
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<tr>
<td>• Andy Rooney</td>
</tr>
<tr>
<td>• Vickie Walker</td>
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<tr>
<td>• Scott Auerbach</td>
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<tr>
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<tr>
<td>• Brandy Beverly</td>
</tr>
<tr>
<td>• Kembra Howdeshell</td>
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<tr>
<td>• Kyla Taylor</td>
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</table>
Thank you

Questions?

…on behalf of OHAT