Environmental Risk Factors for Autism Spectrum Disorders

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What is the evidence that environmental factors contribute to ASD risk? continued

1. Rapid increase in ASD prevalence
2. Genetic studies
   a. Incomplete monozygotic concordance
   b. Most genes associated with ASD are not major effect genes but rather create modest vulnerabilities
   c. In some cases, genes create major vulnerabilities but even in genetic syndromes highly associated with ASD, a significant percentage of carriers do NOT have ASD
   d. De novo gene mutations
   e. Some gene variants confer altered vulnerability to environmental stressors and environmental exposures
      i. Redox or methylation
      ii. Heavy metal metabolism
      iii. Metabolism of organophosphorus pesticides (OPs)

What is the evidence that environmental factors contribute to ASD risk? continued

Increased awareness, improved detection and broadening of diagnostic criteria for ASD likely contribute to increased prevalence

e.g., Diagnostic substitution – labeling people autistic who previously would have been diagnosed with something else

However, Hertz-Picciotto and Delwiche (2009) Epidemiology 20: 84-90:

600% increase in cases:
24% due to earlier diagnosis
56% due to inclusion of milder cases
120% due to changes in diagnostic criteria

400% of increased cases cannot be attributed to diagnostic distribution

What is the evidence that environmental factors contribute to ASD risk? continued

1. Rapid increase in ASD prevalence
2. Genetic studies
3. Clinical heterogeneity of ASD
   - Systemic and CNS pathophysiology
     - Immune dysfunction (including neuroinflammation)
     - Mitochondrial dysfunction

These pathophysiological outcomes known to be exacerbated by environmental factors
air pollution, organophosphorus pesticides, heavy metals

Environmental risk factors for ASD

- Rubella infection during the first trimester of pregnancy
- In utero exposure to thalidomide or valproic acid
- Paternal age
- Environmental chemicals (?)

However, efforts to identify specific environmental risk factors for ASD have produced a number of candidates but few definitive hits

- Heavy metals (lead, methylmercury)
- Pesticides
  - Organophosphorus pesticides (OPs), e.g., chlorpyrifos, diazinon
  - Organochlorine pesticides (OCs), e.g., DDT, dieldrin, lindane
- Persistent organic pollutants (POPs)
  - Polychlorinated biphenyls (PCBs)
  - Polybrominated diphenyl ethers (PBDEs)
  - Polycyclic aromatic hydrocarbons (PAHs)

The Challenge of Identifying Environmental Risk Factors for ASD

- The Challenge of Identifying Environmental Risk Factors for ASD, continued
  - Status of Developmental Toxicity Testing for the 2,863 Chemicals Produced Above 1 million pounds/year

- The Challenge of Identifying Environmental Risk Factors for ASD, continued
  - Genetic susceptibility
  - Environmental Factors
  - Timing
  - ASD risk, severity and treatment outcome
The Challenge of Identifying Environmental Risk Factors for ASD, continued

A significant challenge, particularly for epidemiological studies:

The complexity of heritable factors contributing to ASD susceptibility creates a range of sensitivities to environmental factors

Mechanistic approach for identifying environmental factors that influence ASD risk

Alternative mechanism by which genes interact with environment to influence ASD risk, severity and/or treatment outcome:

Heritable genetic vulnerabilities amplify adverse effects triggered by environmental exposures if genes and environment converge to dysregulate the same signaling system at critical times of neural development.

How do environmental chemicals interact with genetic mechanisms to increase ASD risk?

Largely unknown. Possibilities include:

- De novo mutations
- Epigenetic changes
- Some gene variants confer altered vulnerability to environmental stressors and environmental exposures
- Endocrine disruption

Genes associated with ASD susceptibility: Neuronal connectivity

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<td>ASD Pathology</td>
<td>Autism reflects altered patterns of neuronal connectivity within the developing brain</td>
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<td>Autism may also involve altered neuronal connectivity of the autonomic and sensory nervous system</td>
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ASD Pathology

Autism reflects altered tone of the autonomic nervous system
Neurodevelopmental processes that determine neuronal connectivity

and are thus likely to be altered in ASD:

- Neuronal migration
- Interneuron development
- Neuronal programmed cell death
- Axonal growth and branching
- Dendritic growth and plasticity
- Synaptogenesis and synaptic plasticity

Case Study: Mercury

Why suspect mercury as an environmental risk factor for ASD?

- Methylmercury is a known developmental neurotoxicant in humans
  - Minamata Bay, Japan
  - Iraq
- Vaccinations of young children with vaccines containing thimerosal
  - Ethylmercury poisonings in rural Iraq
  - Timing of vaccinations to clinical diagnosis of ASD
  - Estimated amount of mercury delivered to child as a result of vaccine schedule > estimated safe level

Experimental evidence does not support thimerosal as cause of ASD:

- Clinical symptoms associated with mercury poisoning different from those of ASD
- Multiple case-control, cohort and clinical trials have failed to find an association between vaccinations with thimerosal and incidence of ASD
  - See summaries from Institutes of Medicine

Experimental evidence does not support thimerosal as cause of ASD:

Animal Studies

- The majority of animal studies have failed to show adverse neurodevelopmental outcomes of relevance to ASD following exposure to thimerosal
- Studies in rodents and non-human primates identified significant differences in brain uptake and half-life between mercury species
  - Ethyl mercury << methylmercury

Do these data rule out mercury as environmental risk factor for ASD?

- Genetic susceptibility
  - What genes?
- Contributions to other ASD risk factors
  - Inflammation
  - Oxidative stress
Case Study: Polychlorinated Biphenyls (PCBs)

Non-dioxin-like congeners

1, 2, 2', 3', 4'-pentachlorobiphenyl (PCB 95)

Dioxin-like congeners

2, 3, 4, 5, 6-pentachlorobiphenyl (PCB 126)

Developmental Neurotoxicity

++

++

Carcinogenic

+/-

+++.

Arylhydrocarbon Receptor (AhR)

Low to no affinity

High affinity

Ca\(^{2+}\)-dependent signaling is a critical determinant of dendritic growth in the developing brain

A significant proportion of genes linked to ASD risk encode for proteins that regulate Ca\(^{2+}\)

PCB 95 increases Ca\(^{2+}\) in primary cultured hippocampal neurons

PCB 95 enhances dendritic growth in cultured hippocampal neurons via RyR-dependent mechanisms

Experimental approaches for investigating Ca\(^{2+}\)-dependent signaling pathways in PCB-induced dendritic growth
Exposure of rat pups to PCBs in the maternal diet throughout gestation and lactation interferes with normal patterns of dendritic growth in the hippocampus of weanling rats

Wayman et al. (2012) Environmental Health Perspectives 120:997-1002.

Developmental exposure to PCB 95 in the maternal diet interferes with the topographic organization of the auditory cortex in rats

Fig. 1. Exposure to PCB95 alters A1 maps. (Upper left) Tonotopic map from a typical control rat pup. (Upper right, lower left, and lower right) Examples of maps from PCB95-exposed rat pups. ✗ indicates an unresponsive site. Color bar, CF (kilohertz).

Kenet et al. (2007) PNAS 104:7646-7651

Relevance of these findings to ASD?

- Animal studies
  - Perinatal exposure to a mixture of the non-dioxin-like PCB 47 and dioxin-like PCB 77 shown to alter social behaviors in rats
  - PCB 95 found in significantly higher levels in postmortem brains of children with a syndromic form of autism (maternal 15q11-q13 duplication or Dup15q), but not idiopathic autism as compared to neartypical controls
    [Mitchell et al. (2012) Environmental and Molecular Mutagenesis 58:589-98]

- Human exposure studies

Environmental Risk Factors for ASD

Environmental exposures x Genetic susceptibility x Timing
(non-dioxin-like PCBs) (heritable defects in Ca** signaling)

↓

ASD risk, severity and treatment outcome

What do these findings mean to parents and clinicians?

- Chemical exposure both pre- and postnatal can influence clinical outcome (types and severity of behaviors, co-morbidities)
- Chemical exposures are more readily controlled than genetic factors to prevent or mitigate the expression of ASD-related traits


What do these findings mean to parents and clinicians?

- Minimizing or preventing exposure to chemical contaminants during pregnancy or early childhood may improve clinical outcome
  - Do not use OPs in the home/yard
  - Consume organically grown produce
  - Work with local agencies to minimize use of OPs in public places and/or increase notice to the public of OP spray schedules/locations
  - Keep dust levels as low as possible; wash stuffed toys routinely
  - Limit dietary consumption of fatty fish, meats (PCBs)
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