Prenatal air pollution exposure induces sexually dimorphic fetal programming of metabolic outcomes in adult offspring

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Obesity Trends* Among U.S. Adults
BRFSS, 1990, 2000, 2010
(*BMI ≥30, or about 30 lbs. overweight for 5′4″ person)

Source: Behavioral Risk Factor Surveillance System, CDC.
Obesity is a complex disease....

Genetics

Social factors

Development

Environment
Developmental Programming

Early Life → Adult

Immune system

Microglia

Neuroendocrine Function

Disease Risk

Mental Health Outcomes:
- cognition
- mood
- addiction

Stress
Nutrition
Toxicants

Outcomes:
- cognition
- mood
- addiction

Photo credit S. Hutton and L. Pevny
Immune molecules play a ubiquitous role in neural development.

Microglial progenitor cells migrate from the yolk sac to the developing brain early in embryonic development.
Local self-renewal can sustain CNS microglia maintenance and function throughout adult life

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Infiltrating monocytes trigger EAE progression, but do not contribute to the resident microglia pool

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Air Pollution & Neuroinflammation

Direct mechanisms

- Adsorbed compounds reach the brain
- Particulate matter reaches the brain

Peripheral mechanisms

- Soluble compounds reach the brain

CNS pathology

- Neuroinflammation (iNOS, TNFα, IL-1β, COX2, & NFκB)
- Neuron damage/loss
- Microglia activation (HLA-DR & CD14) (ROS & cytokine production)
- Blood brain barrier damage/dysfunction (Changes in inflammatory, tight junction, & transport proteins)
- Aβ42 accumulation (Neuronal, vascular, & diffuse plaques)
- Aβ and α-Synuclein aggregation
- Lipid peroxidation
- DNA damage
- Astroglisis (GFAP)

Cardiovascular system
- Blood brain barrier damage/dysfunction
- Circulating monocytes

Circulating cytokines
- Liver

Lung

TRENDS in Neurosciences

Block and Calderón-Garcidueñas, 2009
Prenatal Air Pollution Exposures

Moms exposed E2-17: **Diesel Exhaust Particles (DEP)**: 50 μg in PBS/Tween every 3 days

- Inner city (LA, Mexico City, Beijing) conditions

**Control (Ctrl):** PBS/Tween solution

All offspring were weaned at P28, and housed in standard caging conditions until adulthood.
Vulnerable Populations?

Households receiving public assistance

children under six living in poverty

racial breakdown of owners and renters
Obesity is an Inflammatory Condition

• Elevated inflammatory markers in blood and tissues
• Changes within immune system directly implicated:
  – insulin resistance/metabolic syndrome
  – type II diabetes

Obesity causes inflammation, but can inflammation cause obesity?
Obesity is an Inflammatory Condition *within the brain as well*

- Peripheral inflammation
- Neuroinflammation
- Obesity affects behavior:
  - Anxiety
  - Cognitive deficits
  - Higher risk of Alzheimer’s
2nd Phase: Adult Offspring (Male & Female)

**HFD**
- 9 weeks of diet choice:
  - 45% saturated fat (high-fat)
  - and
  - 10% saturated fat (low-fat)

**LFD**
- 9 weeks of low-fat diet only
  - (10% saturated fat)

OR

Adult offspring (4 months old)

Pre-Diet Body Weights; Activity & Anxiety Levels in Open Field (1 Day Before Diet)

Monitor body weight gain and food consumption every 3 days

Measure Activity & Anxiety Levels in Open Field Weekly

Collect brain, fat, & blood for endpoint immune & metabolic marker analysis
Male offspring exposed to DEP in utero gain more weight on a HFD...

(Bolton, Auten, & Bilbo, *Brain, Behavior, & Immunity*, 2014)
...and are more insulin-resistant
Original Contribution

Association of Childhood Obesity With Maternal Exposure to Ambient Air Polycyclic Aromatic Hydrocarbons During Pregnancy

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DEP male offspring do not eat more, nor are they less active on HFD (Bolton, Auten, & Bilbo, Brain, Behavior, & Immunity, 2014)
HFD results in increased monocyte trafficking to the fat tissues, and these cells are more activated in DEP male offspring (Bolton, Auten, & Bilbo, Brain, Behavior, & Immunity, 2014).

(A) CCR2

(B) CD11b

(Bolton, Auten, & Bilbo, Brain, Behavior, & Immunity, 2014)
Saturated Fatty Acids Produce an Inflammatory Response Predominantly through the Activation of TLR4 Signaling in Hypothalamus: Implications for the Pathogenesis of Obesity

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Mice exposed prenatally to air pollution + adult high fat diet have worse outcomes and **increased microglial activation**.
Is there a recruitment of peripheral monocytes to the brain, as in the fat tissue?
Flow Cytometry Methods

Step 1:
Saline-perfuse animals at week 9 of diet

Step 2:
Dissection of specific brain regions

Step 3:
Tissue dissociation and myelin depletion

Step 4:
Immunostaining of single cell suspension
Animals on HFD show evidence of increased monocyte infiltration to the brain as well (Bolton, Auten, & Bilbo, Brain, Behavior, & Immunity, 2014)
DEP male offspring on a HFD express more activation markers on these infiltrating cells in the hypothalamus.

(Bolton, Auten, & Bilbo, Brain, Behavior, & Immunity, 2014)
Neurobiology of Disease

Stress-Induced Recruitment of Bone Marrow-Derived Monocytes to the Brain Promotes Anxiety-Like Behavior

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DEP male offspring are more anxious after only 4 weeks of HFD

(Bolton, Auten, & Bilbo, *Brain, Behavior, & Immunity*, 2014)
Conclusions & Future Directions

Why are there sex differences?

Which comes first: microglial activation or monocyte infiltration?

Cause or effect?

Obesity

Neuro-Inflammation
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