Persistent Immune Effects of Wildfire PM Exposure During Childhood Development

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OVERVIEW

- California National Primate Center program
- Nonhuman primates as a model for early childhood development
- Rationale for investigation of wildfire smoke PM
- Effects of wildfire smoke PM on immunity
- Effects of wildfire smoke PM on lung function
CNPRC Vital Statistics

- 300 acre site, occupy 85 acres
- 20 core scientists, ~70 affiliate scientists
- ~400 employees
- ~40 graduate students and postdoctoral fellows
- ~60 undergraduate students
- ~5,000 monkeys
- Current NIH P51 base operating grant = $11 M total cost
  - Administration of P51 Core Grant Activities
  - Breeding Colony and Unassigned Animals
  - Support for Core Scientists and Research Units
  - Pilot Research Program
  - Consortium Activities
CNPRC Nonhuman Primates: Rhesus Macaque and Titi Monkey
- California National Primate Center program

- Nonhuman primates as a model for early childhood development

- Rationale for investigation of wildfire smoke PM

- Effects of wildfire smoke PM on immunity

- Effects of wildfire smoke PM on lung function
Overall lung architecture of nonhuman primates is most similar to human (monopodial vs. dichotomous growth)
Stages of Human Lung Development
Comparative Lung Anatomy in the Rhesus Macaque

Right Middle Lobe

32 days  90 days  6 months  Adult
Age-Dependent Changes in Innate Immune Function

Kollman, et. al. Immunity 2012
Postnatal Lung and Immune Systems Develop in a Synchronized Fashion

PROLIFERATION DIFFERENTIATION

Birth 1 Year

INSTRUCTION

Environment

Innate Immunity Adaptive Immunity Lung Function
The Monkey as a Model for Early Childhood Development

- Similar postnatal maturation of lung development
- Similar postnatal maturation of immune cell development
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Persistent Effects of Ozone Exposure During Lung Development: Impact on Innate Immunity

Airway Lumen
- LPS
- TLR4

Airway epithelium
- DC lymphocytes
- Macrophage

Airway Interstitium
- DC
- lymphocytes
- Lung Vasculature

Basement membrane

Peripheral Blood
- Eosinophil
- Neutrophil
- WBC

Ozone

LPS
Persistent Effects of Ozone Exposure During Lung Development: Impact on Innate Immunity

Receptors on cells of the innate immune system recognize PAMPS

PAMPS=Pathogen associated molecular patterns
Monkeys exposed to 0.5 ppm/8 hr/day ozone until 6 months of age

Exposed animals remain in filtered air until 12 months of age

Animals challenged with a single dose of endotoxin (LPS) at 12 months of age
Persistent Effect of Ozone: Lung (In Vivo LPS)

- LPS challenge = ↑BAL cells, ↑PMN
- LPS challenge in animals with prior ozone = ↓BAL cells, ↓PMN
Persistent Effect of Ozone: Peripheral Blood (In Vivo LPS)

- LPS challenge = ↑ WBC ↑ PMN
- LPS challenge in animals with prior ozone = Variable
Persistent Effect of Ozone: Peripheral Blood (In Vitro LPS)

Peripheral Blood from Ozone Exposed Animals: LPS Challenge In Vitro

Filtered air = ↑ IL-6 ↑ IL-8

Ozone = ↓ IL-6 ↓ IL-8
Summary

- Postnatal ozone exposure results in a persistent attenuation of the inflammatory response to LPS
- Both lung and systemic (peripheral blood) compartments are affected by prior exposure to ozone
- TLR4 signaling pathway is likely to be an important target for environmental persistence, but may have some overlap with other TLR pathways
CNPRC Monkey Model of Ambient Air Pollutant Exposure

A short bike ride to the Primate Center…
Spring is Peak Birthing Season at the CNPRC
Summer 2008 California Wildfires

On June 21-22 2008, a dry low pressure system produced dry lightning which ignited approximately 2000 forest fires across Northern California (Humboldt County)
Summer 2008 California Wildfires: PM 2.5

- From June 21-27 much of Northern California was covered in a thick blanket of smoke.
- Air quality in the Sacramento valley improved after June 28 due to onshore winds and Delta breeze.
- With calm winds, hazy conditions returned on July 7.

Daily 24 hour average concentration of PM 2.5 from June 1-July 31 on UC Davis campus.
NAAQS standard 35 ug/m³ per 24 hour period.
Summer 2008 California Wildfires: Ozone

Daily 8 hour average concentration of ozone from June 1-July 31 on UC Davis campus. June 13 and July 7 exceed the current NAAQS standard of 0.075 ppm/8 hours.
Can we detect persistent immune effects under ambient exposure conditions in animals that were housed outdoors as infants?

Can we detect persistent lung function effects under ambient exposure conditions in animals that were housed outdoors as infants?
Study Design

- Select animals from outdoor colony born in spring of 2008 (1-3 months of age, males n=25, females n=25)

- Select animals from outdoor colony born in spring of 2009 (1-3 months of age, males n=24, females n=25)

- Collect peripheral blood and measure pulmonary physiology
Study Methods

- LPS $\rightarrow$ TLR4 $\rightarrow$ IL-6, IL-8
- Flagellin $\rightarrow$ TLR5 $\rightarrow$ IL-6, IL-8
- Compare measures of innate immunity ligand stimulation in peripheral blood samples with pulmonary function measures
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- Nonhuman primates as a model for early childhood development
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Persistent Effects of Wildfire Smoke Exposures: LPS Stimulation of Peripheral Blood Cells from 3 Year Old Animals

Wildfire Smoke $\Rightarrow$ IL-6
Persistent Effects of Wildfire Smoke Exposures: Flagellin Stimulation of Peripheral Blood Cells from 3 Year Old Animals

Wildfire Smoke = $\downarrow$ IL-8
Persistent Effects of Wildfire Smoke Exposures: Comparison of 3 Year Old Animals with Adult Animals
Persistent Effects of Wildfire Smoke Exposures:

Changes in Toll-like Receptor Pathway Gene Expression

Blood samples from female monkeys born in 2008 show:

↑ c-Rel, Rel B
↑ MyD88 (LPS)
↑ IKK alpha (Flagellin)
Persistent Effects of Wildfire Smoke Exposures: Airways Responsiveness and Compliance in 3 Year Old Animals

Airways Responsiveness

Compliance
Correlation of Flagellin Induced Cytokine Synthesis with Lung Function

Airways Responsiveness

Compliance
Summary

CNPRC monkeys exposed to ambient wildfire smoke at infancy:

- exhibit persistent down regulation of LPS and flagellin-induced cytokine responses in peripheral blood cell cultures
- show gender dependent effects of exposure on parameters of innate immunity and lung function
- demonstrate a persistent change in molecular programming of peripheral blood cells
Effect of Early Life Exposures on Innate Immunity and Lung Function
Respiratory Disease Center

- Opening February 27, 2014
- 19,000 sq ft
- Inhalation exposure facility
- Pulmonary function laboratory
- Open bay laboratory space
- Office/conference space
QUESTIONS?