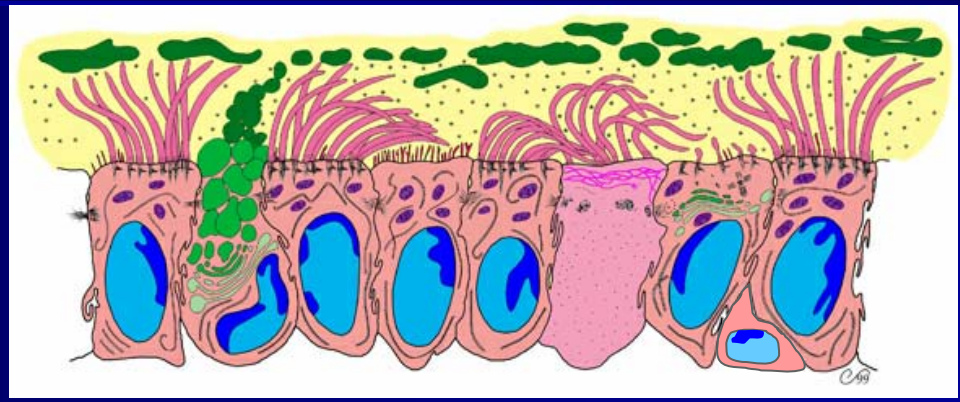
# Effects of Diesel Exhaust on Epithelial Cells: Potential Interaction with Viral Infections

ETH-Conference on Combustion Generated Nanoparticles
August 21st-23rd, 2006


Ilona Jaspers, Ph.D.
CEMALB
Pediatrics/Immunology & Infectious Diseases
University of North Carolina at Chapel Hill

# Respiratory Epithelium

Tracheal/bronchial Epithelium.



**Composition:**

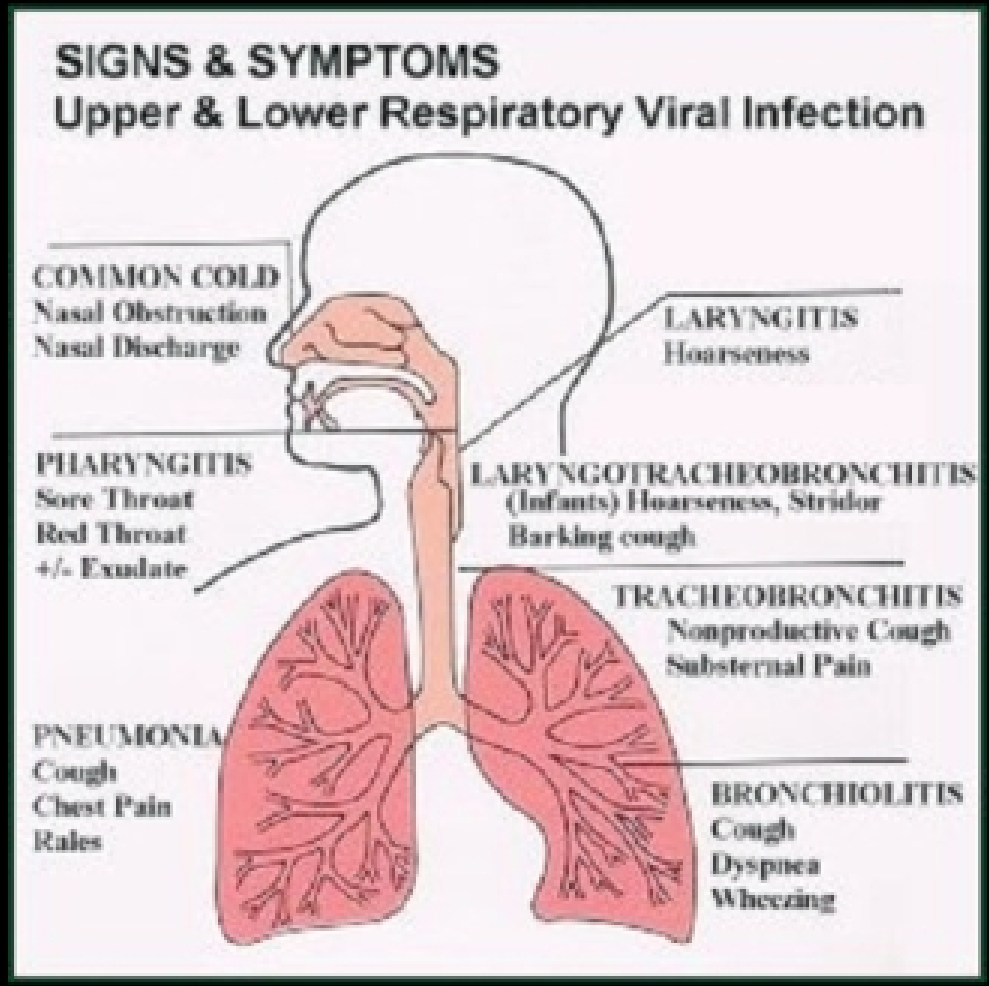
Trachea, bronchi, bronchioles:
Ciliated, non-ciliated, mucus/goblet, basal
Alveolar sacs:
type I and type II cells

**Function:**

mucociliary escalator
mucus production
barrier, defense
source of immune mediators

**The respiratory epithelium is the major site for infection and replication of respiratory viruses!**

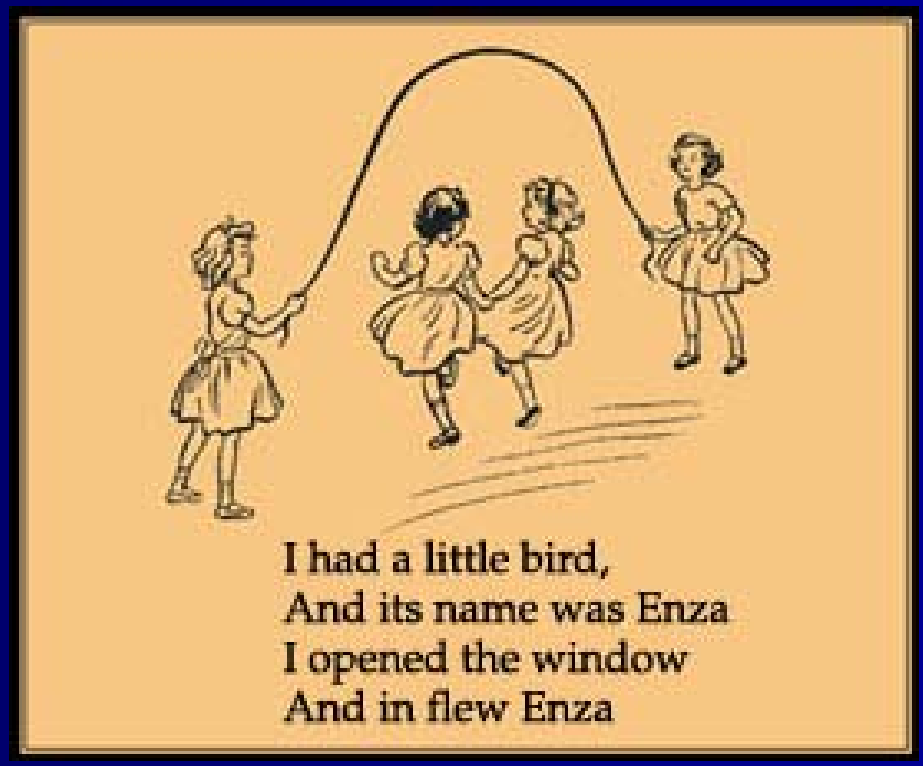
# *Respiratory Viruses*




**URI** -
Rhinovirus
Coronavirus
Enterovirus
Adenovirus
EBV

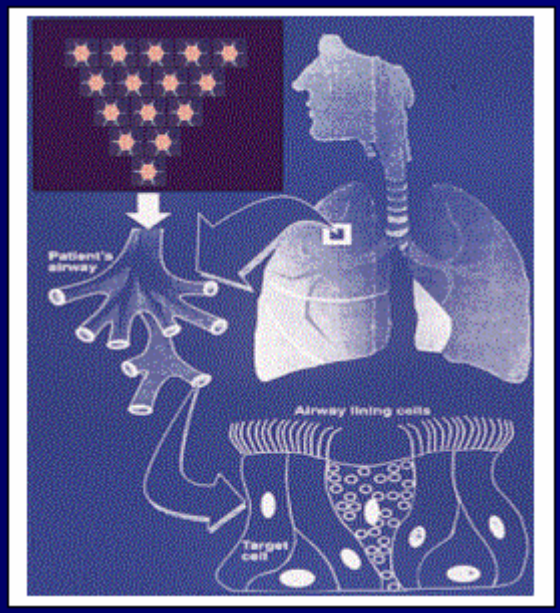
**LRI** -
Influenza
Parainfluenza
RSV

# The Spanish Flu of 1918 killed 20 Million People Worldwide

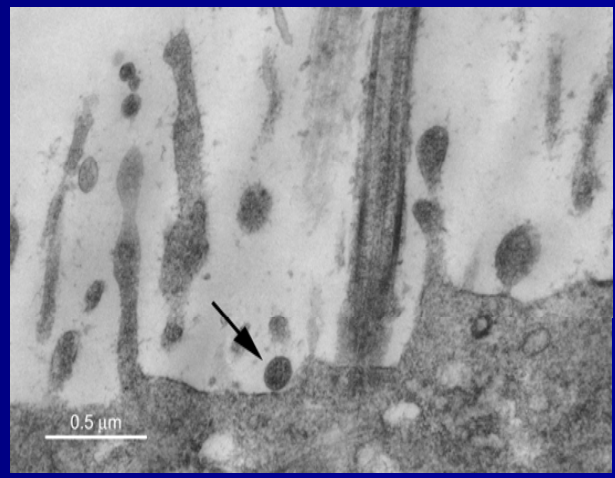


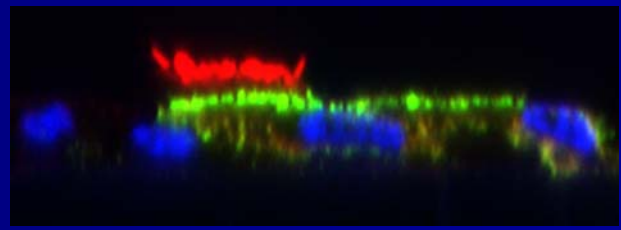

**“After the 1918 Influenza epidemic in the U.S., little girls jumped rope to a new rhyme”; Tom Dunne**

# Infection of the Human Respiratory Epithelium




R.J. Pickles, Ph.D.
Cystic Fibrosis Center
UNC-CH



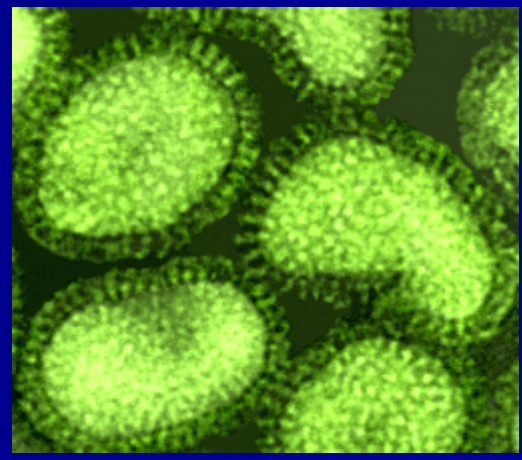



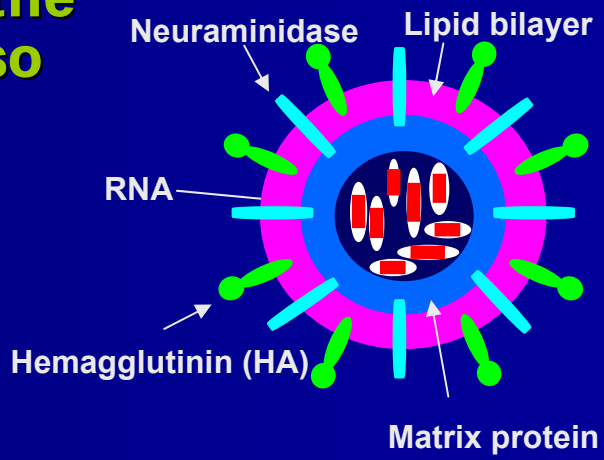
# Factors Affecting Susceptibility To Viral Infections

- Age
- Nutrition
- Pre-existing pulmonary disease
  - smoking
- Environmental Pollution

# Influenza

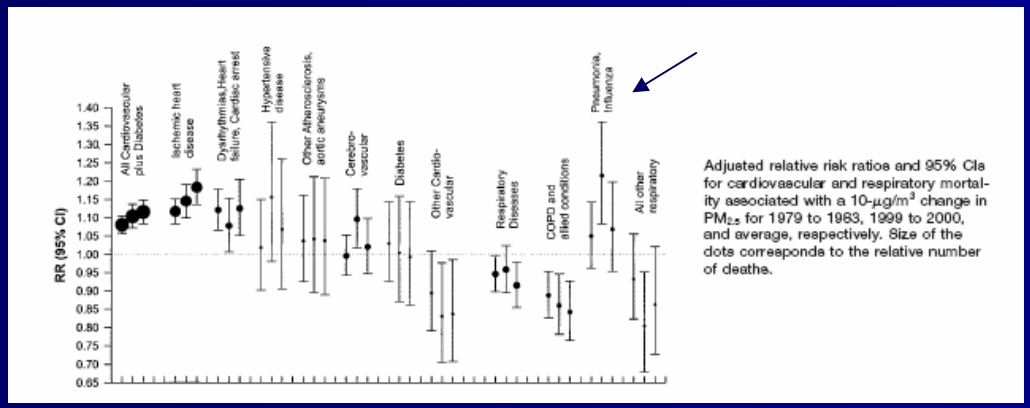
- Enveloped negative-stranded RNA virus
- Member of the Orthomyxoviridae family
- Classified as influenza A, B, and C, with influenza A being the most pathogenic one
- Replicate in epithelial cells of the upper respiratory tract, but also monocytes/ macrophages and leukocytes can be infected





# Air Pollution and Influenza

- Inhaled particles and influenza target the same tissue (respiratory epithelial cells)
- Some Epidemiological studies show association between particle exposure and virus infection.




C.A. Pope et al., *Circulation*, 2005

- Repeated exposures of mice to diesel exhaust (DE) enhances susceptibility to respiratory virus infections (Hahon et al., 1985; Harrod et al., 2003)

# Diesel Exhaust

- Diesel engines are used in trucks, buses (school buses), farm equipment, ships, etc.
- Inhalation of DE is associated with increased asthma and allergic diseases
- Combustion process of diesel fuel generates mixture of hundreds of organic and inorganic compounds in the gas and particle phase.
- Organic hydrocarbons (PAH) are adsorbed on diesel exhaust particles, which are very small (<1micron) and therefore respirable


# Effects of DE on Epithelial Cells

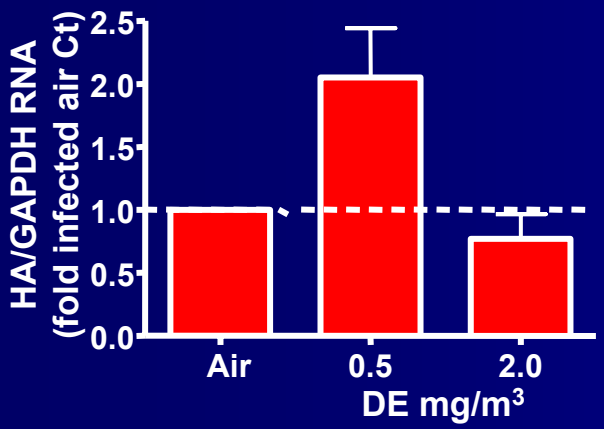
- Epithelial cells release inflammatory cytokines/chemokines in response to DE exposures.
- Mechanism of this is believed to involve DE-induced generation of oxidative stress and activation of oxidant-dependent signaling pathways (i.e. NF-κB).
- Mediators released by DE-exposed epithelial cells induce dendritic cell maturation (Bleck et al., 2006)
- DE-induced mediator release is polarized (i.e. IL-6 & GM-CSF => apical; IL-8 => basolateral) (Auger et al., 2006)

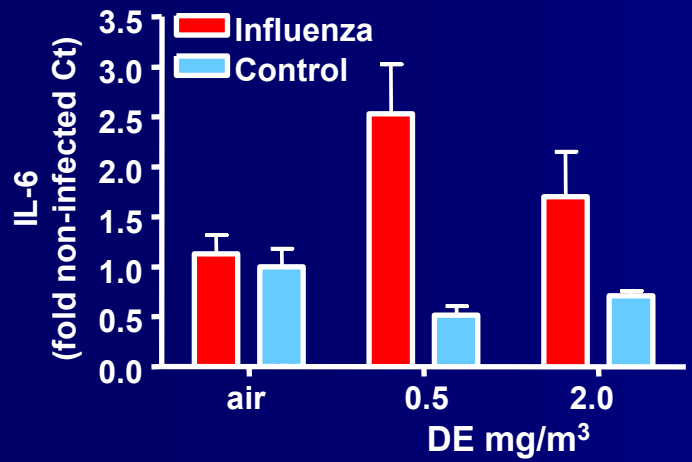
# Mouse in vivo DE Exposure

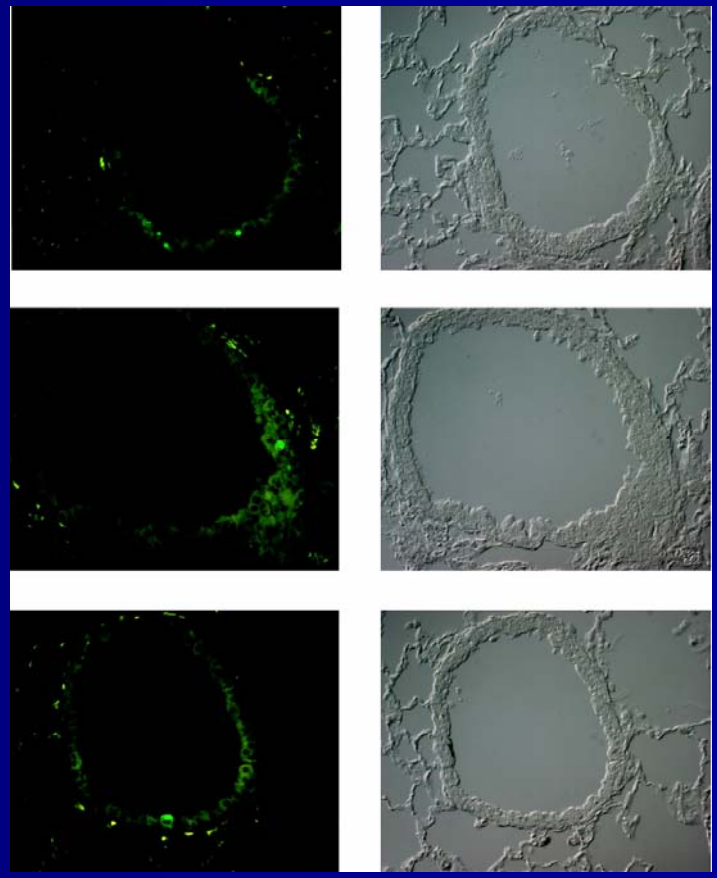
- 30 kW Deutz, 4 cylinder engine
- BFM1008 type
- Engine speed ~1725 rpm under load of a compressor (22.8 ratio)

- Hinner Chambers:
- Internal volume of 11 cubic ft.
- Chamber flow: ~2.5 cfm

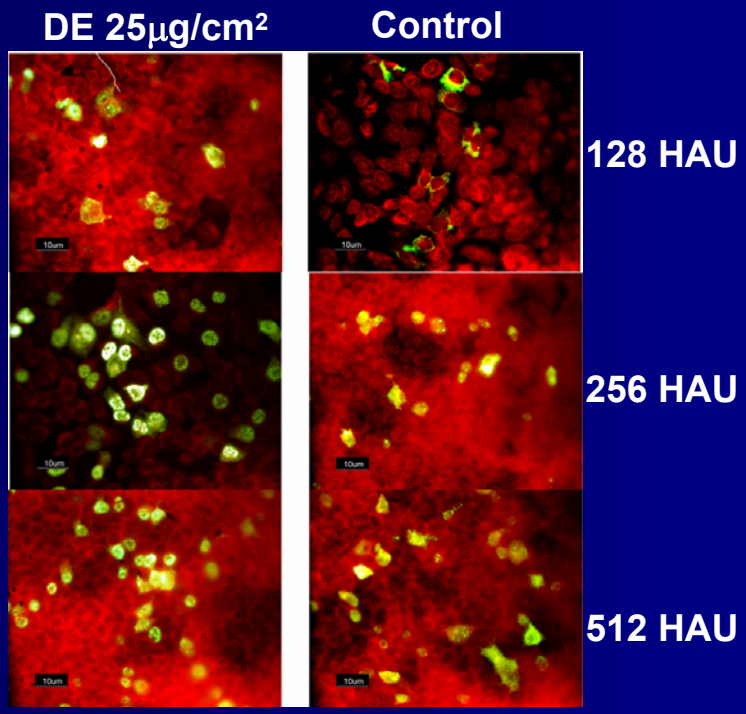
# DE Enhances Influenza Infections in Mice

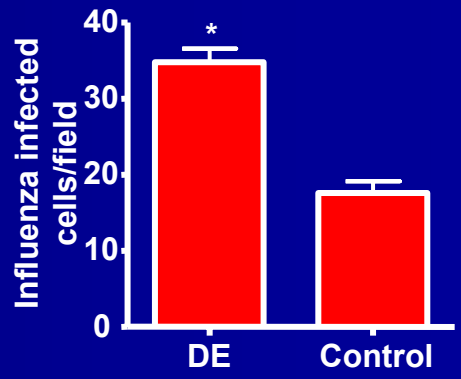


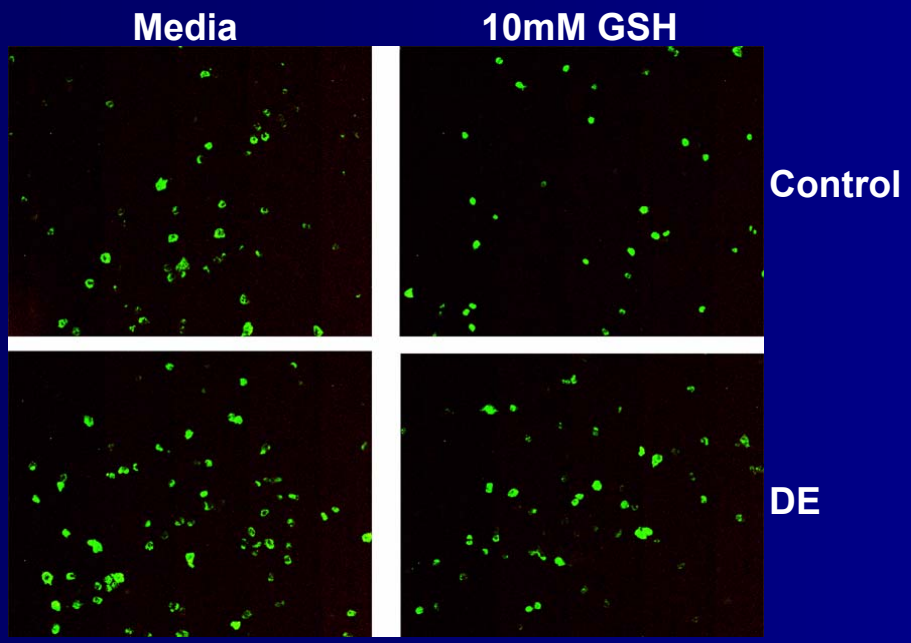
# DE Increases The Susceptibility to Influenza

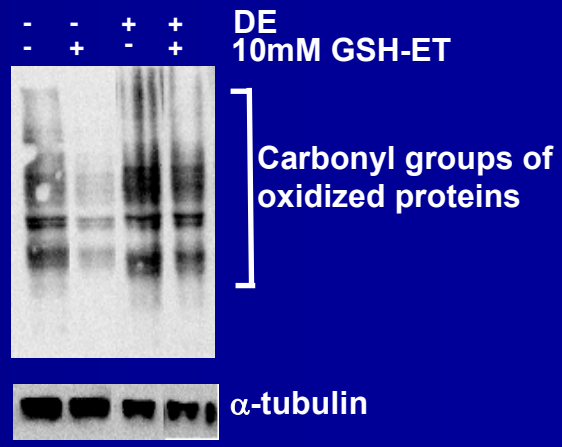


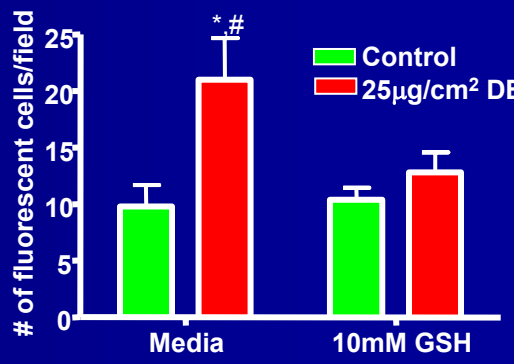




Jaspers et al., *Toxicol. Sci.,* 2005

# Antioxidants Reverse Effects of DE On The Number Of Infected Cells



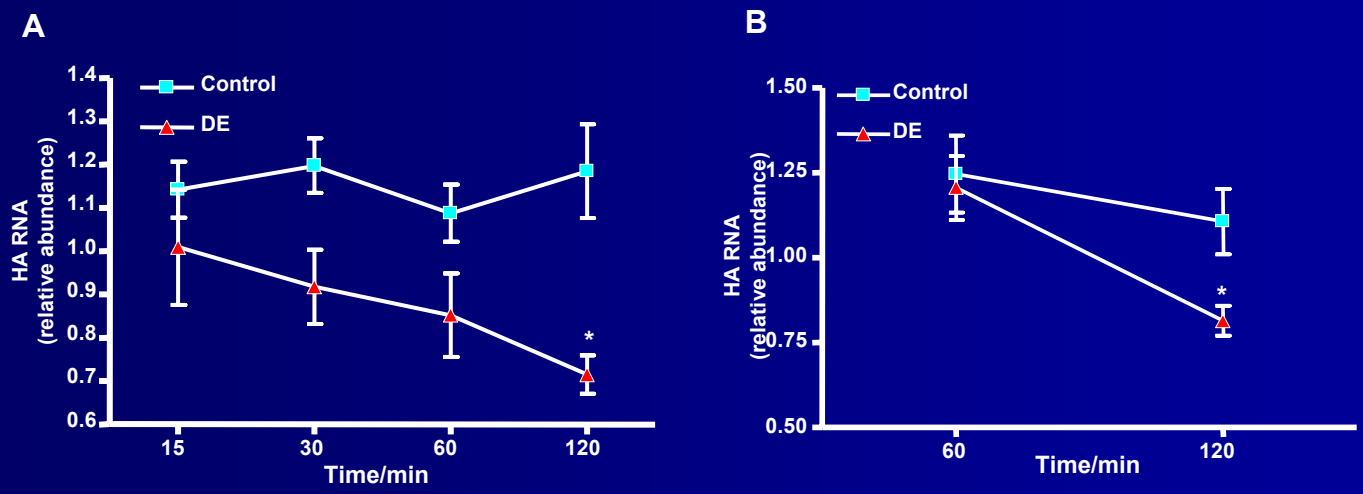

Jaspers et al., *Toxicol. Sci.,* 2005

# Potential Mechanisms Enhancing Viral Infections of Epithelial Cells

- Decreased antiviral defense response (i.e. interferon)
- Increased virus attachment
  - Increased expression of receptors (i.e. ICAM-1 for rhinovirus)
- Facilitated entry of virus (i.e. does virus "piggy-back" onto particles?)

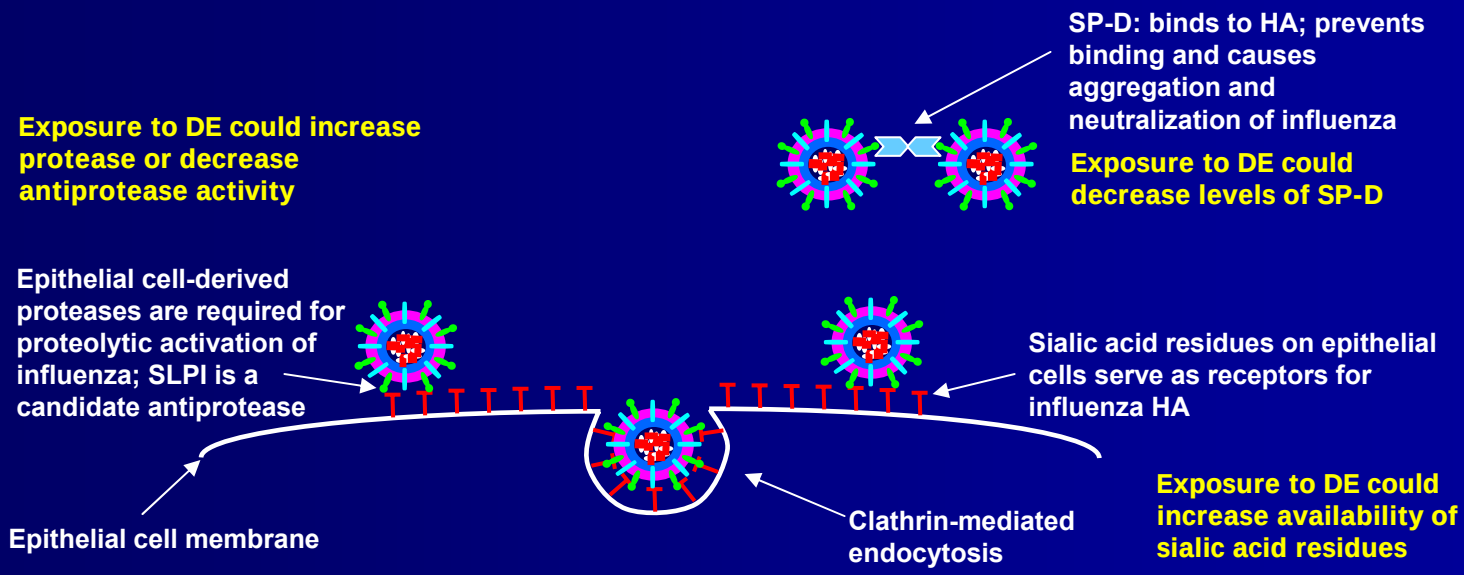
Exposure to DE does not decrease interferon-dependent antiviral defense responses!

# DE Enhances Influenza Virus Attachment/Entry



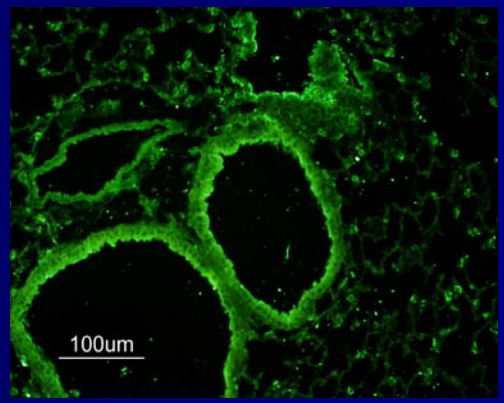

Jaspers et al., *Toxicol. Sci.,* 2005

# Enhancement of Influenza Attachment: Potential Mechanisms

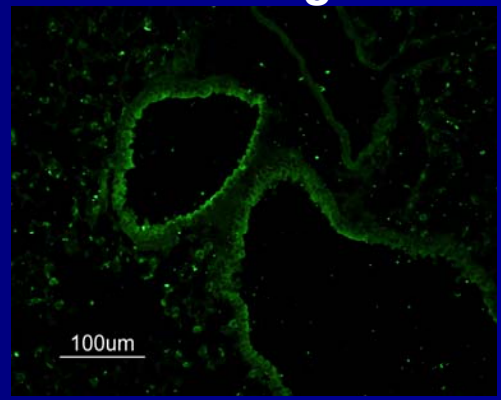


# Exposure To DE Decreases SP-D Levels

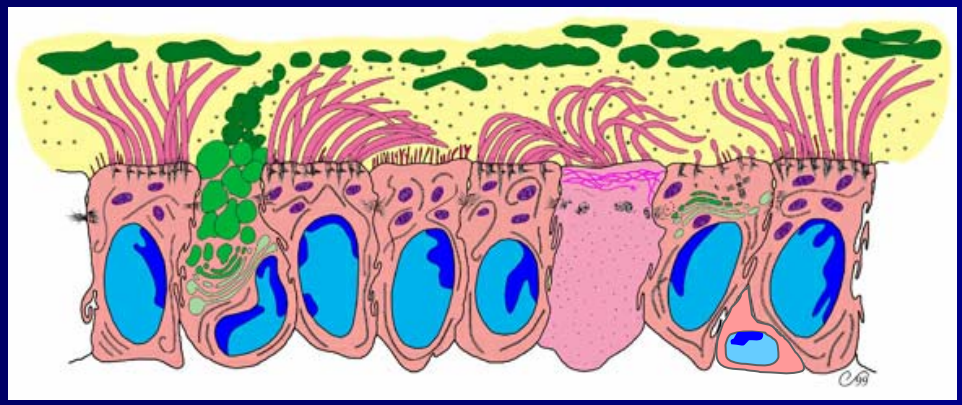
Air




DE 0.5 mg/m$^3$



# Polarization of Respiratory Epithelium



- The apical and basolateral membranes of airway epithelial cells have different biochemical/morphological characteristics and fulfill different functions.

- Junctional complexes between epithelial cells, such as tight junctions, maintain the polarized phenotype and function of epithelial cells as well as the vectorial release and activity of released mediators.

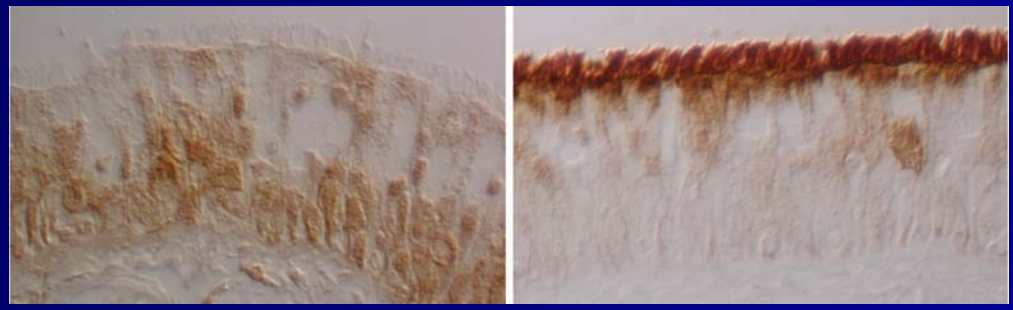
# Polarized Distribution Of Receptors On Epithelial Cells

**Segregation of receptor and ligand regulates activation of epithelial growth factor receptor**

Paola D. Vermeer1, Lisa A. Einwalter1, Thomas O. Moninger2, Tatiana Rokhlina1, Jeffrey A. Kern3, Joseph Zabner1 and Michael J. Welsh1,4

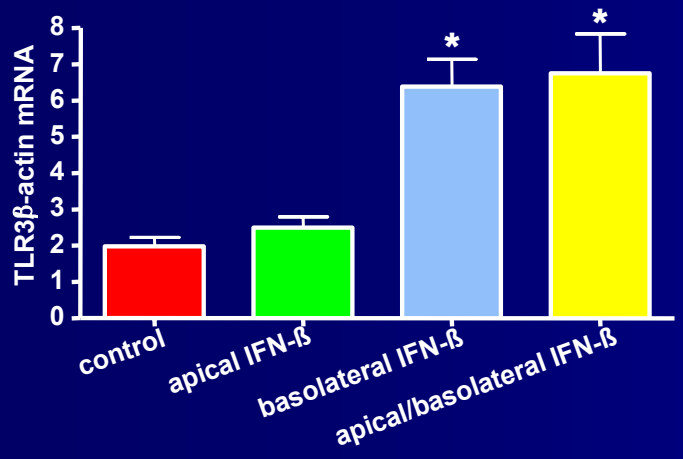
*Nature* **422**, 322-326 (20 March 2003 **)**

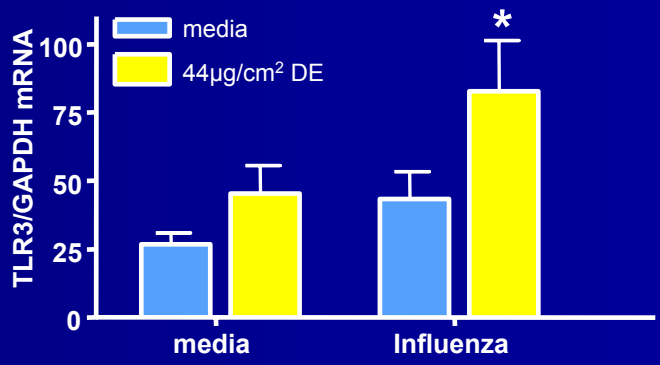
**Certain receptors (i.e. IFNAR, EGFR, etc.) are exclusively localized either on the apical or basolateral membrane, thus restricting access of potential ligands**



**IFNAR** **Cilia**

# IFNAR Activation Is Polarized And Enhanced By DE Exposure



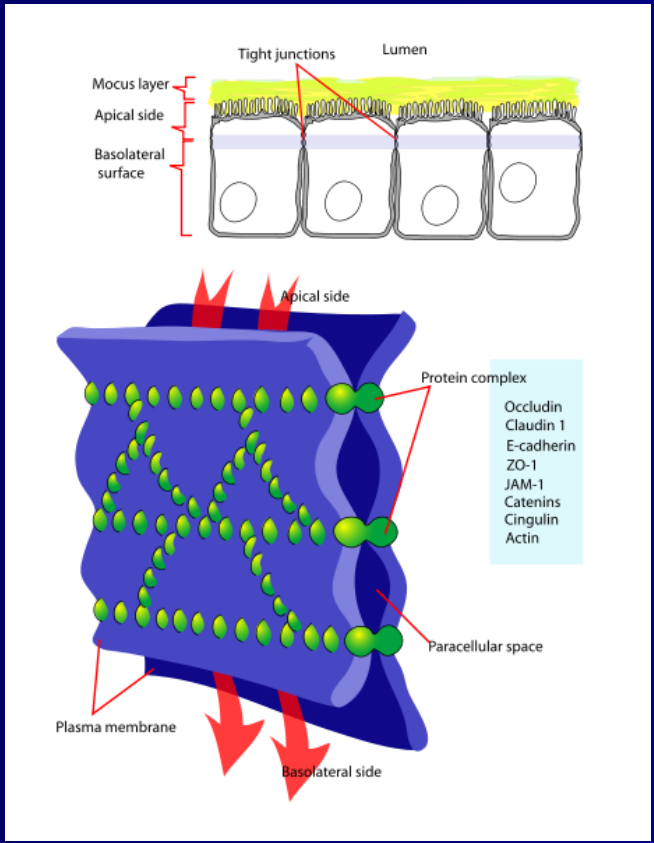



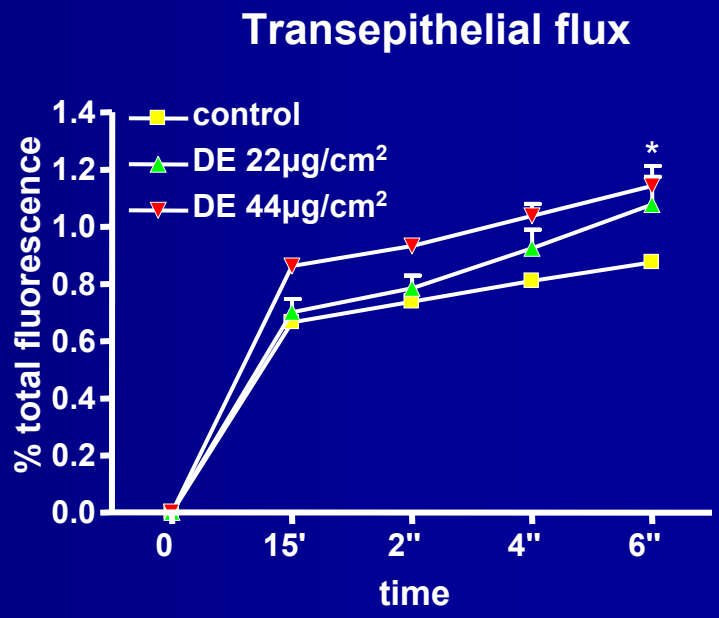

Ciencewicki et al., 2006

Disruption of junctional complexes enhances receptor activation.

Potential mechanism for pollutant-induced health effects?

# DE Enhances Trans-epithelial Flux



# Summary

- Exposure to DE enhances the susceptibility to respiratory virus infections
- Potential mechanisms mediating this effect include
  - Oxidative stress
  - Increased virus attachment/entry
  - Decreased levels of surfactant proteins (SP-A, SP-D)
  - Disruption of junctional complexes

# Future Direction

- Translate *in vitro* and mouse *in vivo* data into humans
  - Using LAIV vaccine and human diesel exposure chamber
- "Are all diesels created equal?"
  - What are the effects of emissions generated by newer diesel engines on viral infections?
- Discern the role of gas-phase and particulate phase components
  - Using *in vitro* exposure system

# Acknowledgements

**UNC-CEMALB**
Wenli Zhang
Missy Brighton
Johnny Carson, Ph.D.
Philip Bromberg, M.D.
Dave Peden, M.D.

**Curriculum of Toxicology**
Jon Ciencewicki

**American Chemistry Council-LRI**
Young Investigator Award

**NIEHS/NIH RO1 ES013611**

**US EPA**
Lisa Dailey
Michael Madden, Ph.D.
M. Ian Gilmour, Ph.D.
Kym Gowdy (NC State)

**UNC-Peds**
Melinda Beck, Ph.D.
Mirek Styblo, Ph.D.