

Diesel Exhaust Toxicology: In vitro approaches

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Advantages of in vitro exposure studies

- “Rapid screening” for bioactivity
- “Inexpensive”
- Discern mechanisms of responses; extrapolation to in vivo responses [validation of in vitro system]

In Vitro Studies Design

Exposure



Biological
Responses

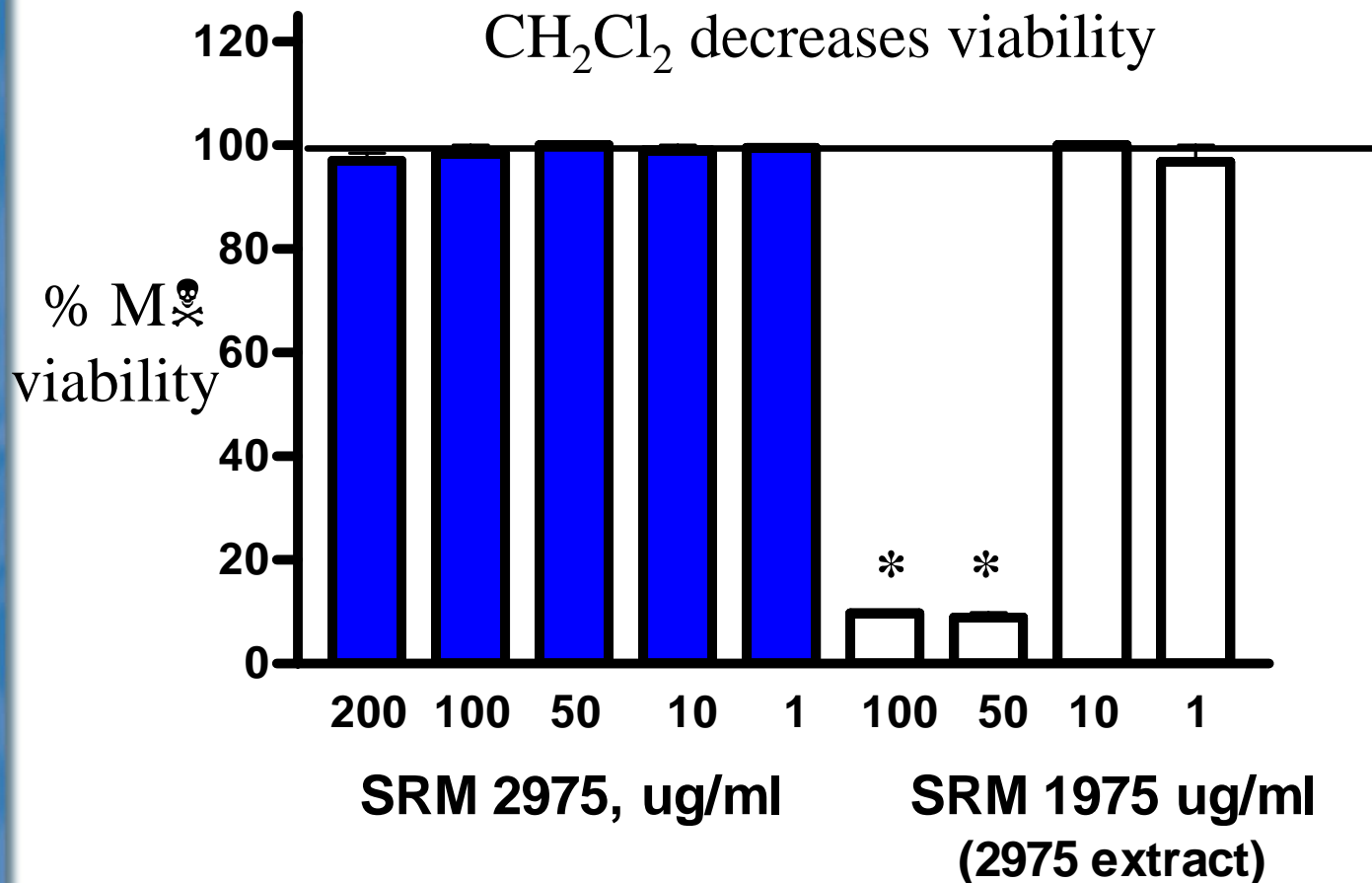
- Emission
(engine/fuel, etc)
- Whole exhaust
- PM
- Gases

- "Fresh" Exhaust
- Filter Collection
- Liquid Collection

- Health Endpoints
- Tissues/Cells &
Endpoints

Exposure

Filter Collection: How does one get PM off ?
 -scraping → loss of PM size, endotoxin introduction; suspension in aqueous media
 -extractions (organics) → solvent effects

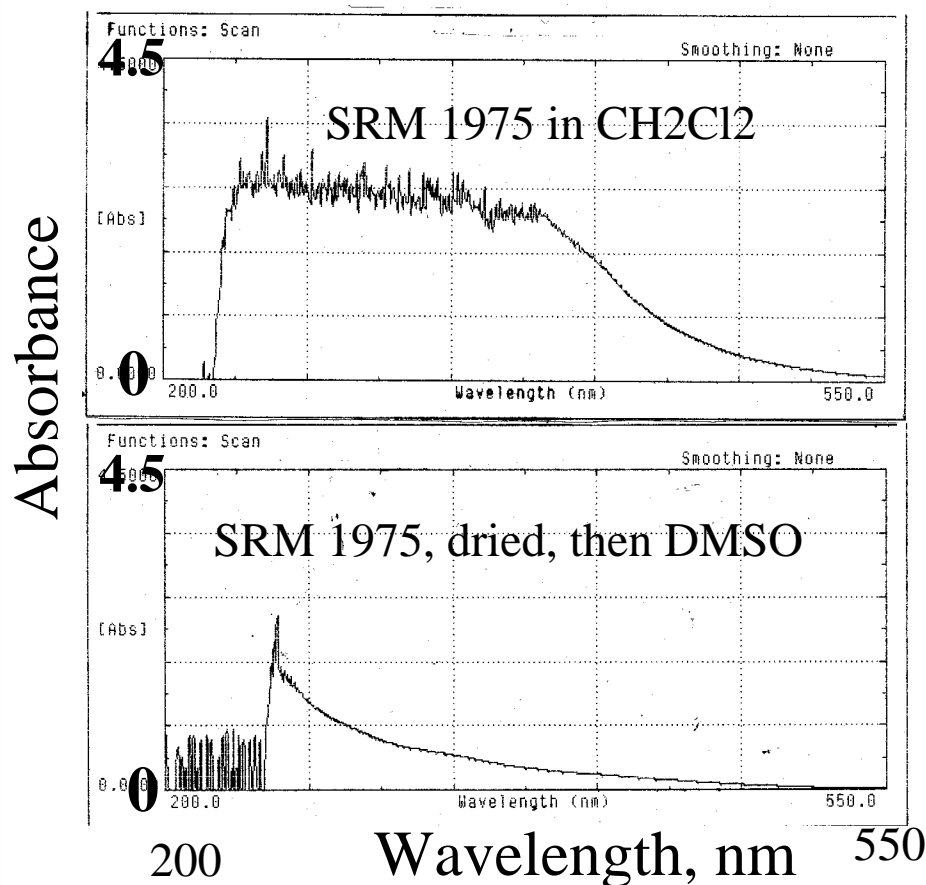


Exposure

Filter: How does one get PM off ?

-scraping → loss of PM size

-extractions (organics) → solvent effects



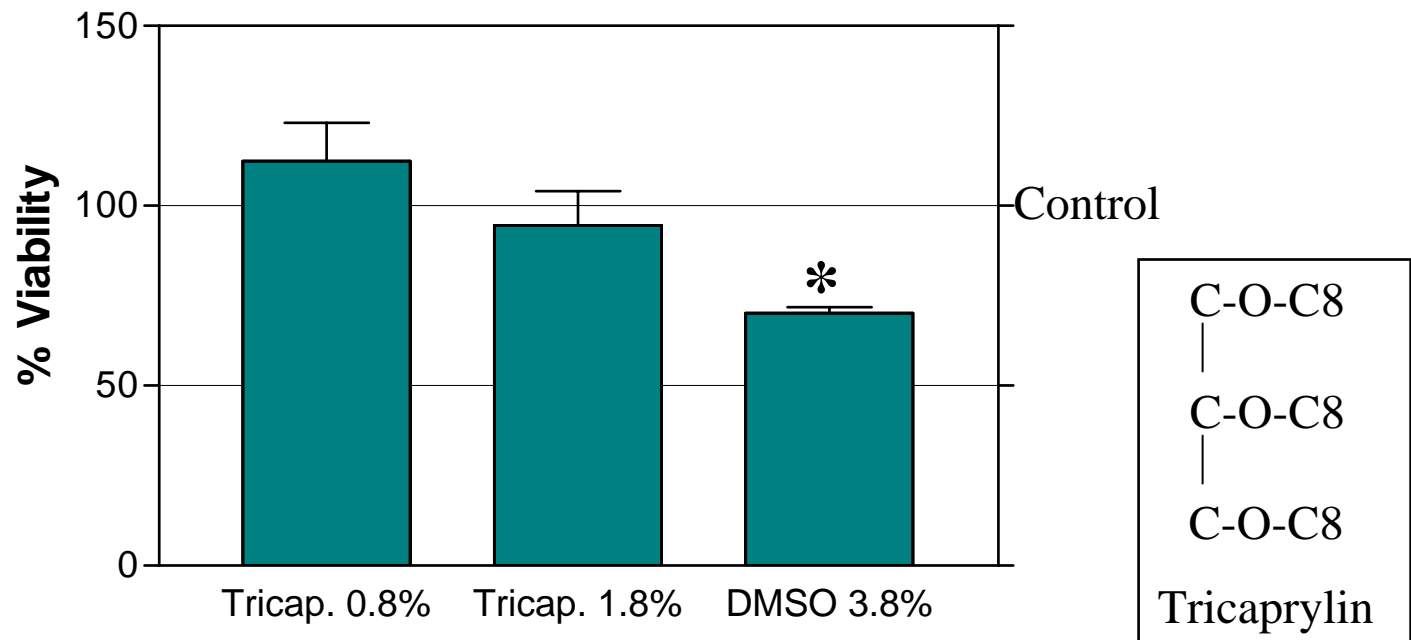
Solvent exchange
loses some
components

Exposure

-Filter Collection: How does one get PM off ?

-scraping → loss of PM size

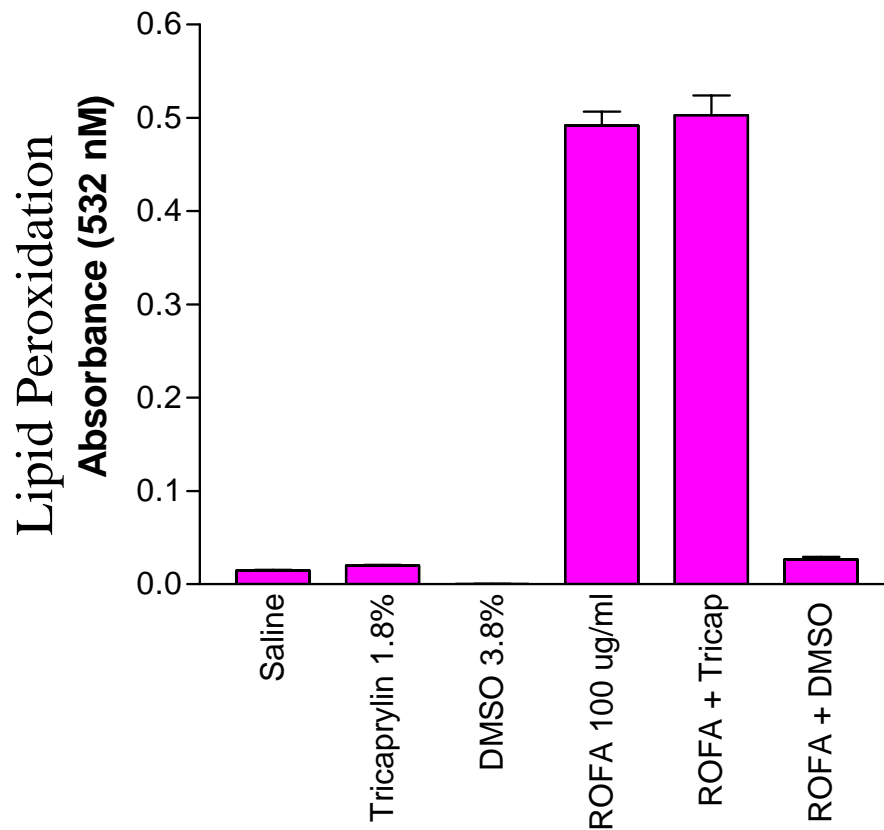
-extractions (organics) → solvent effects



BEAS cells 24 hr culture; neutral red uptake

Exposure

- Filter Collection:** How does one get PM off ?
 - scraping → loss of PM size
 - extractions (organics) → solvent effects**



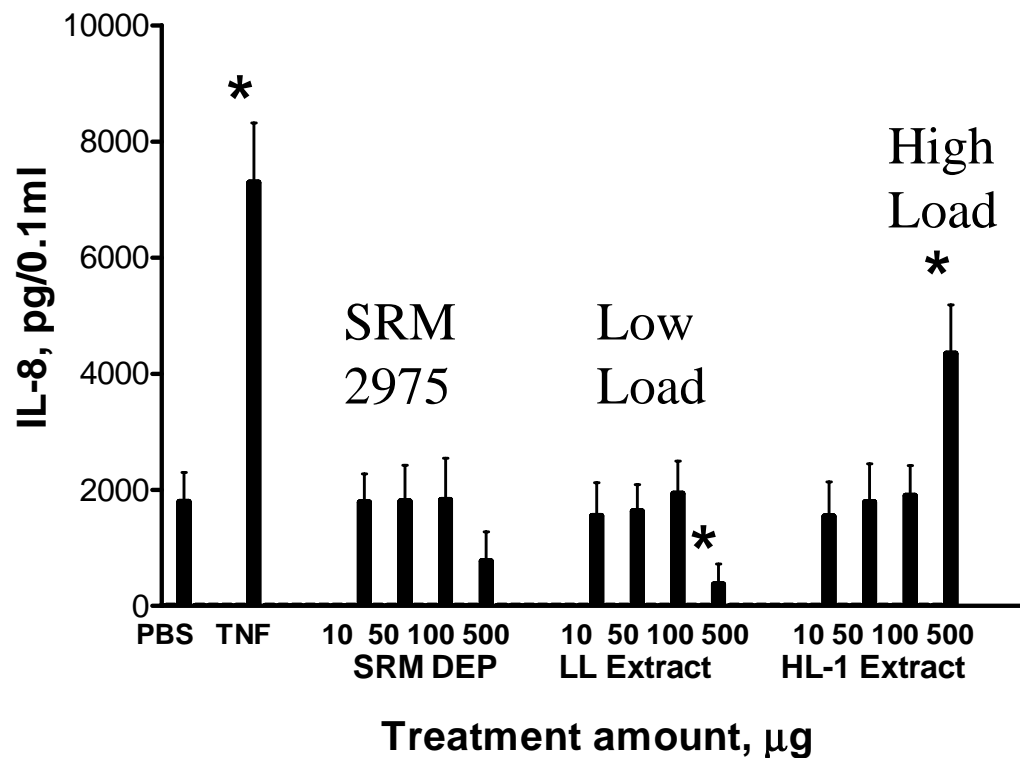
DMSO inhibited ROFA
oxidative stress

Exposure

-Liquid Collections

Collect exhaust into chilled liquid in impingers (Method 5)

No extraction step/ add directly to cultures



Catepillar model 3304

0 kw and 85 kw

Biological Responses

-Health Endpoints

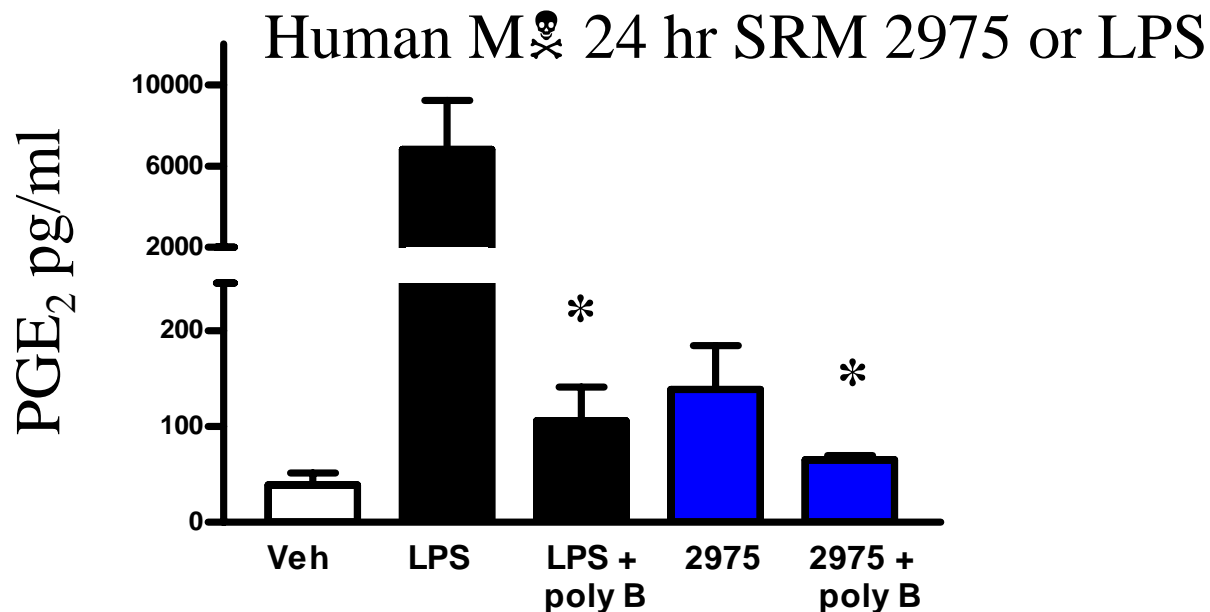
- Cancer (1° Lung, bladder): repeated exposures
- Fibrosis: repeated exposures
- Allergy & Asthma Induction: acute-repeated exposures
- PM-induced endpoints-acute exposures
 - Cardiopulmonary Mortality
 - Hospitalizations (lung infections, asthma symptoms)

Biological Responses

-Tissues/Cells & Endpoints

Lung:

- macrophages- inflammation, immune function, oxidative stress
- airway epithelial cells- inflammation, DNA damage, oxidative stress
- fibroblasts- fibrosis



Biological Responses

Cardiac Cells:

Myocytes- human rare; neonatal animal usually used
gap junction, calcium flux, cytokines

Perfused organ- electrical properties (eg, depolarizations)

Biological Responses

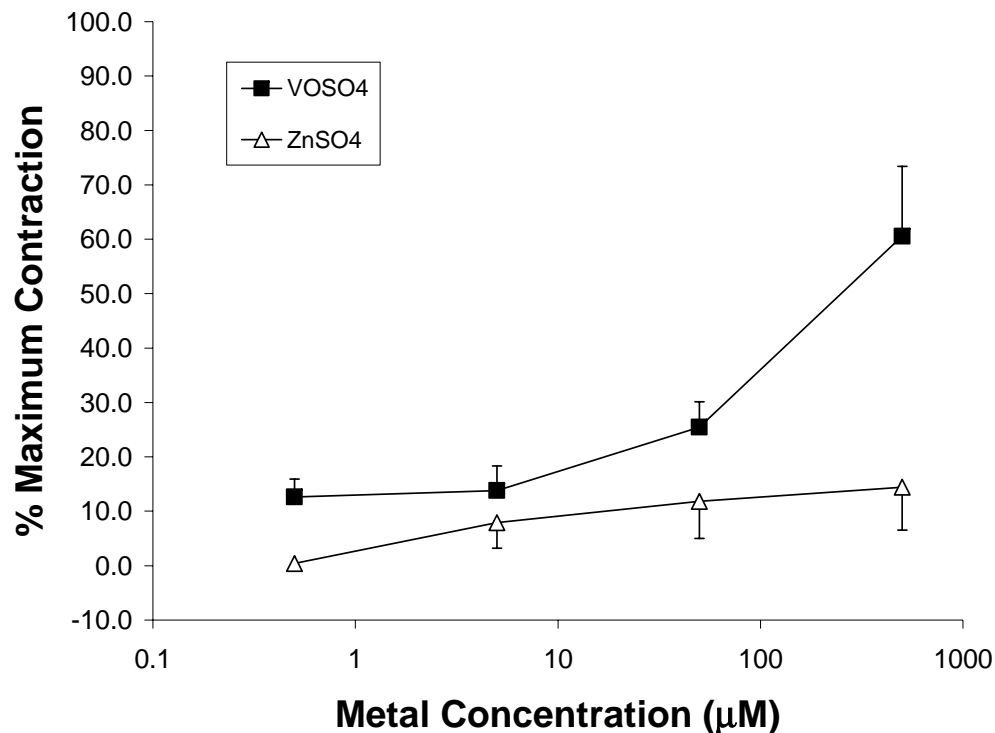
Vascular:

Endothelial- clotting, oxidative stress (Hirano et al, 2003 Toxicology)

Platelets- clotting

Whole blood- clotting, soluble mediator production

Rat pulmonary artery ring- constriction/dilation



Huang et al, EHP
In press

Biological Responses

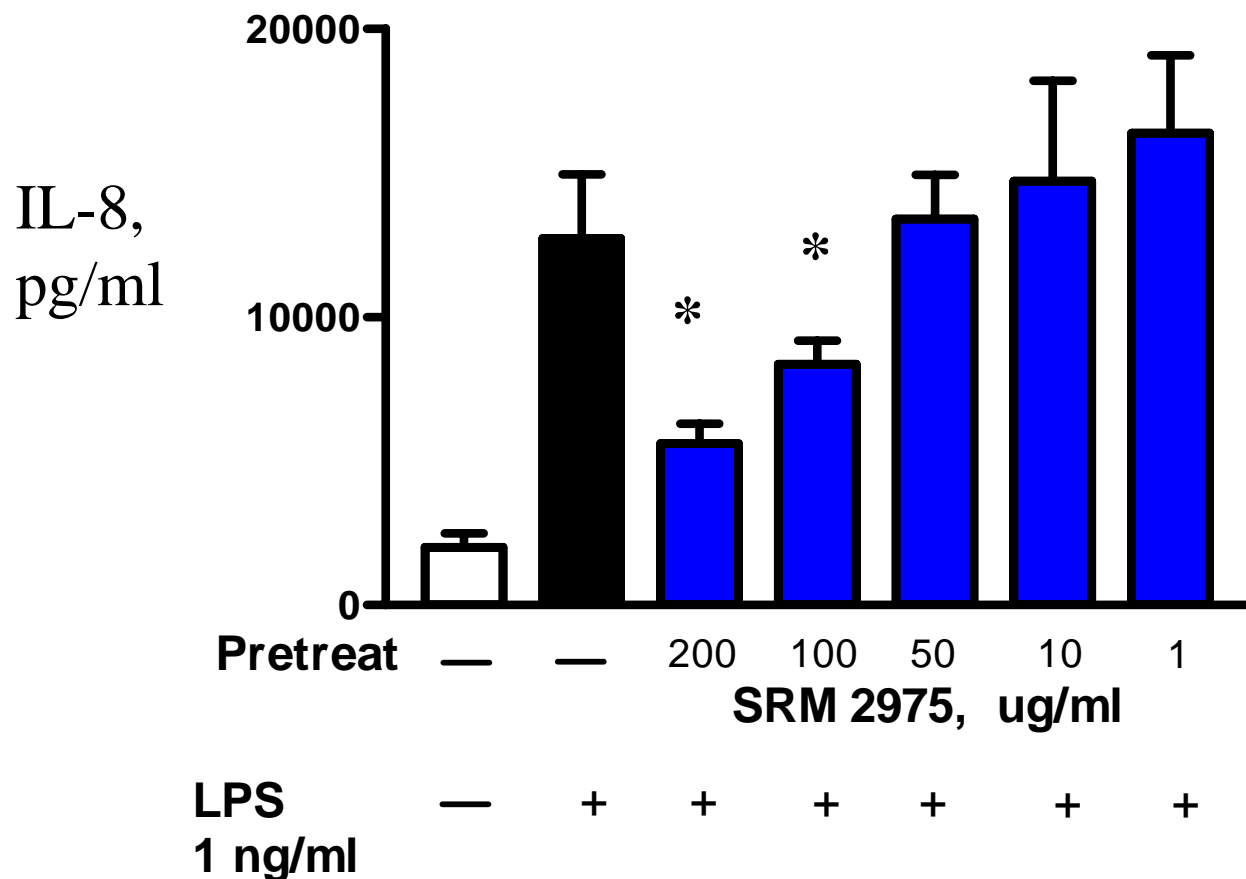
Allergy & Asthma:

-B lymphocyte transformed cell line- IgE induction, cytokine expression, other immune-related processes (Tsien et al, 1997 TAP)

-Human blood basophils- Particles & PAHs enhance mediator release (Devouassoux et al, 2002 J Allergy Clin Immunol; Kepley et al, 2003 Clin Immunol.)

Diesel Exhaust: Interactions with other PM or PM components

Decreased M α response to LPS



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