Circulating Factors Induce Endothelial Cell Activation Following Exposure to Inhaled NO_2: Evidence From a Novel Translational *In Vitro* Model

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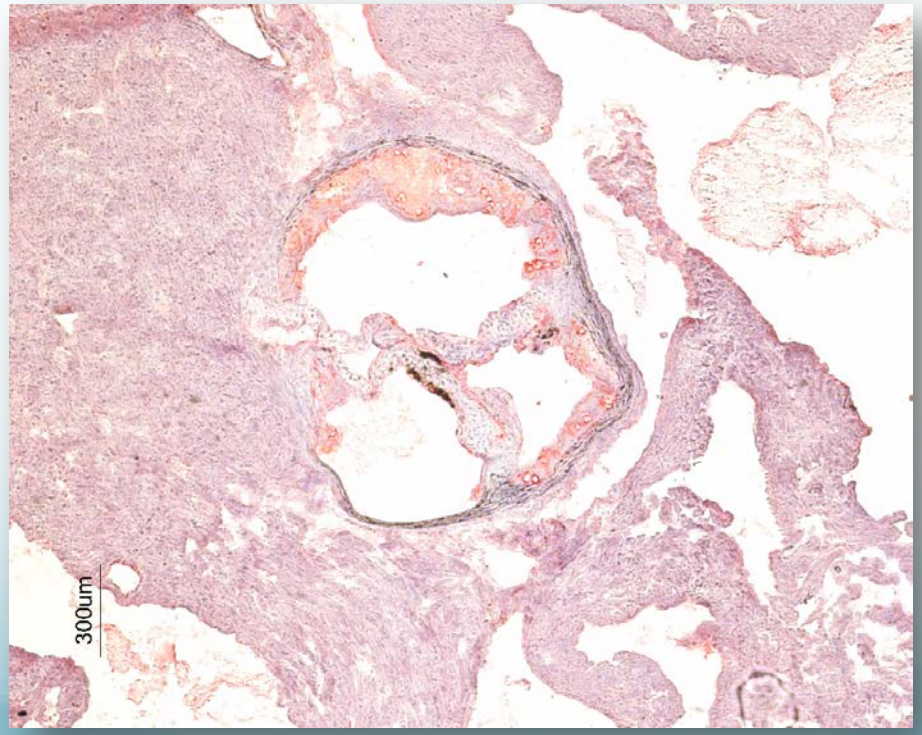
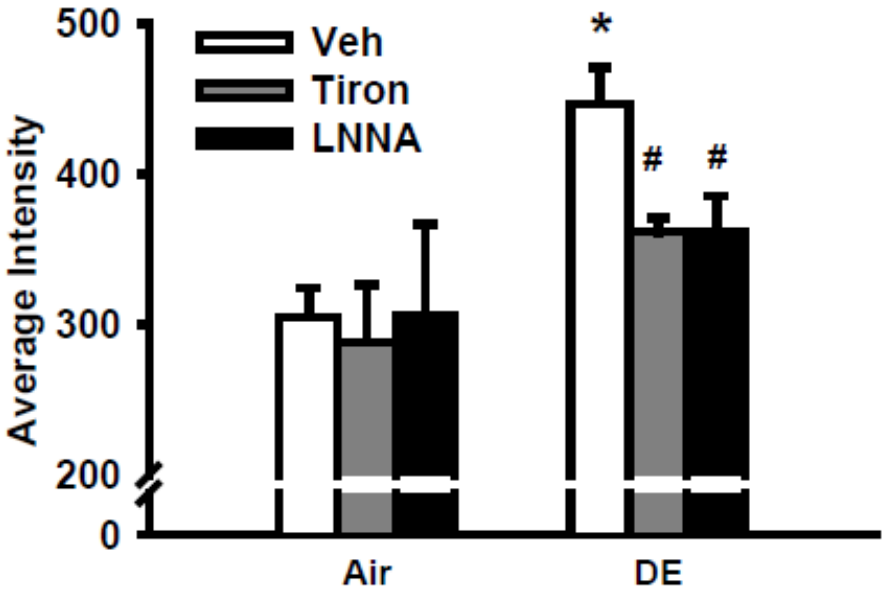
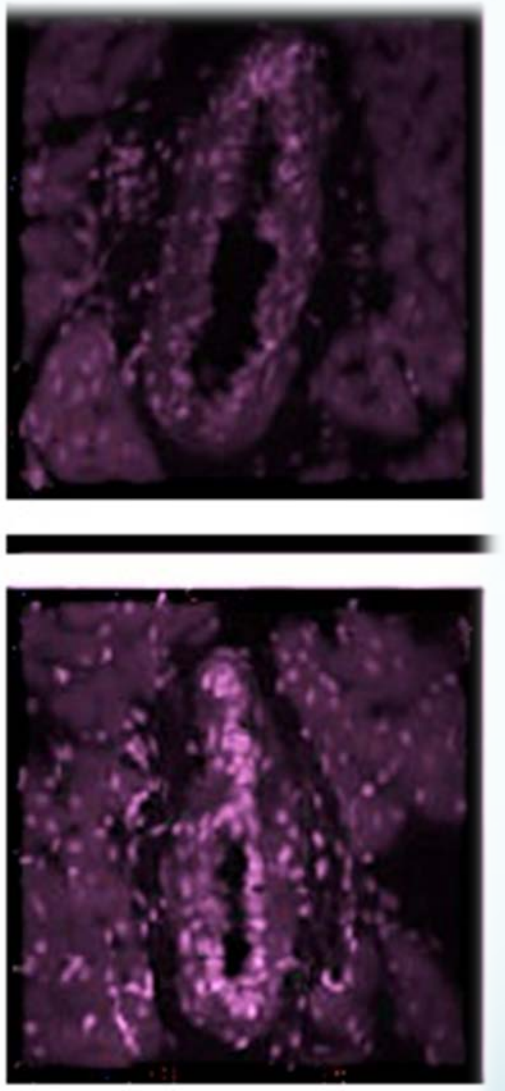
Early Events in Atherosclerosis

Endothelial Oxidative Injury → Activation of Adhesion Molecules → Recruitment of Inflammatory Cells → Plaque formation



Endothelial Oxidative Injury

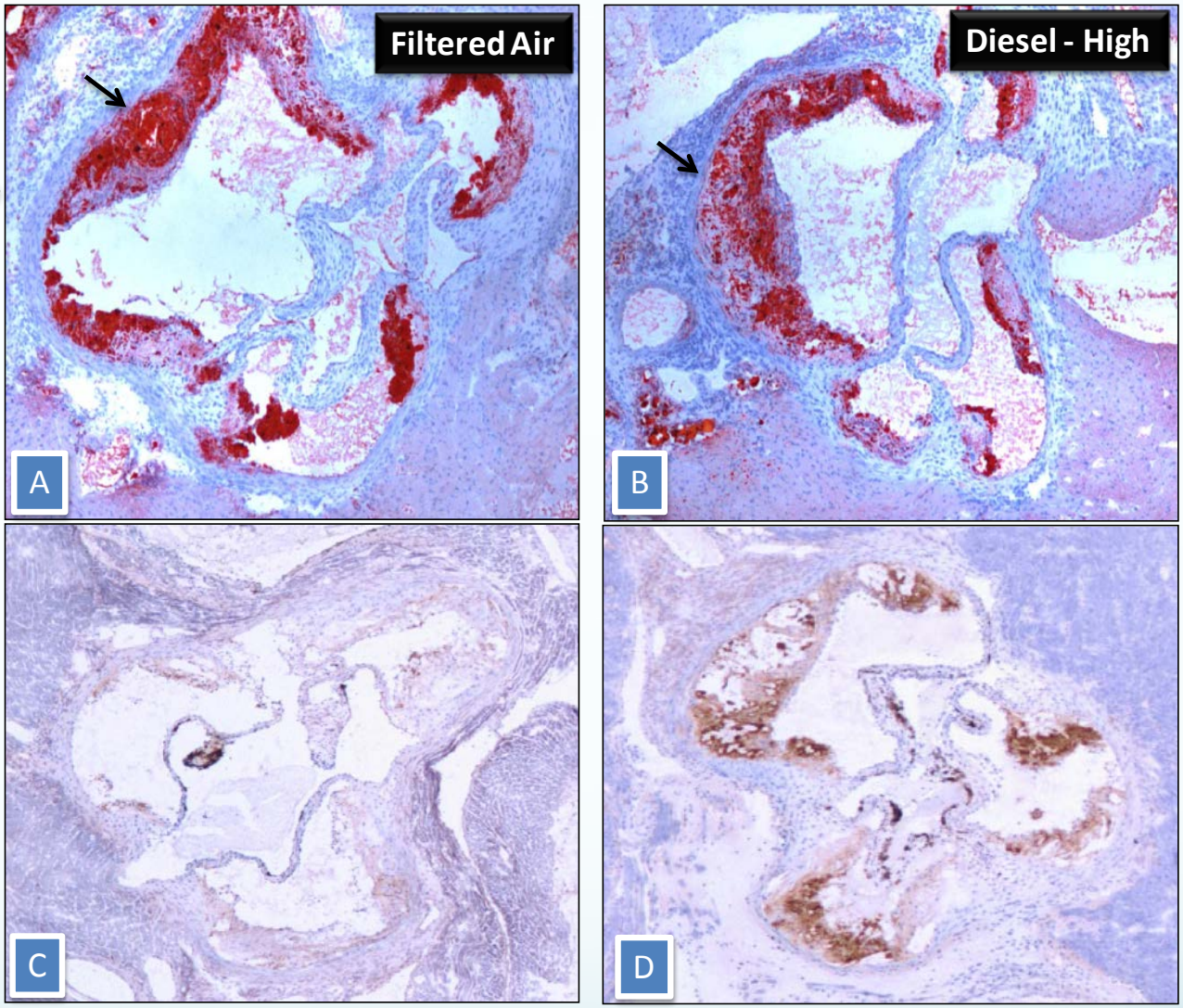




Diesel Inhalation Increases Superoxide Levels in the Coronary Vasculature of Rats

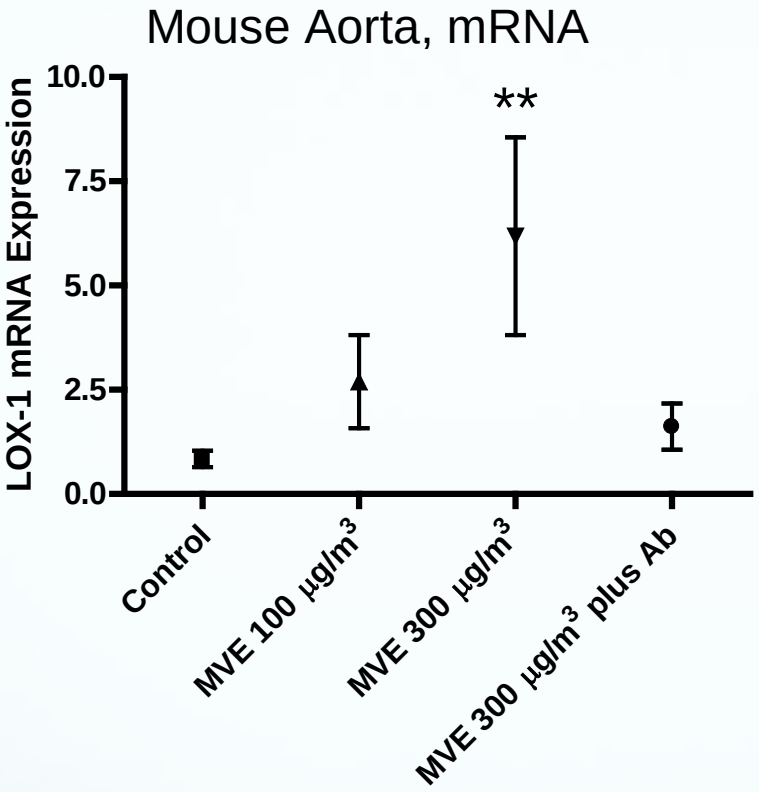
Cherng et al, EHP, 2010

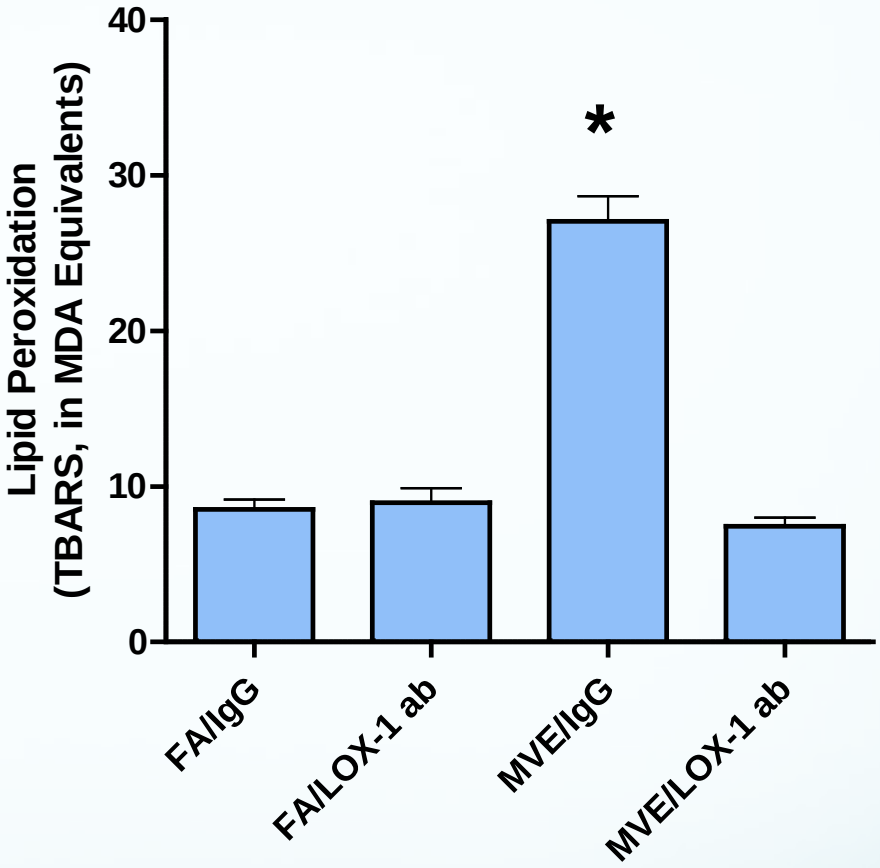
Recruitment of Inflammatory Cells



Diesel Inhalation Enhances Inflammation of Vascular Lesions in Mice

Campen et al., TAAP, 2010

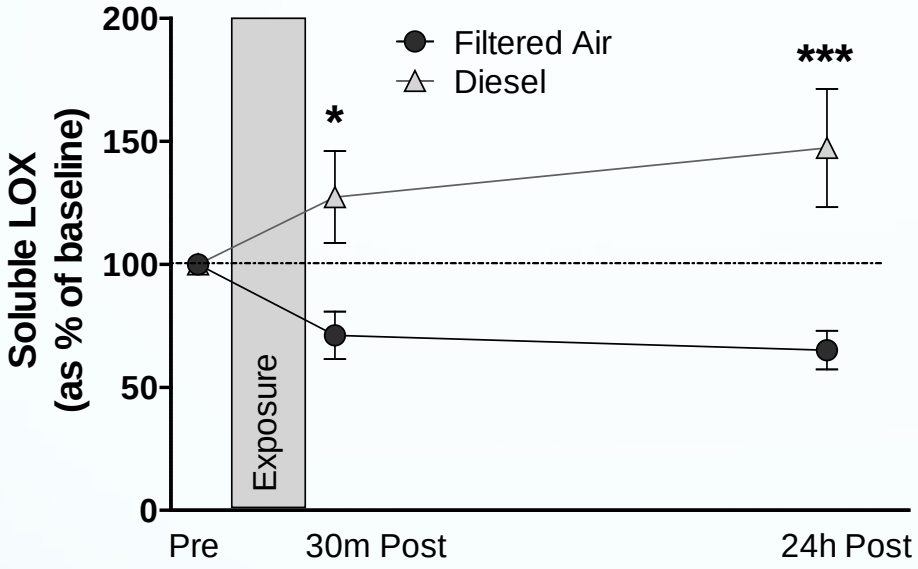


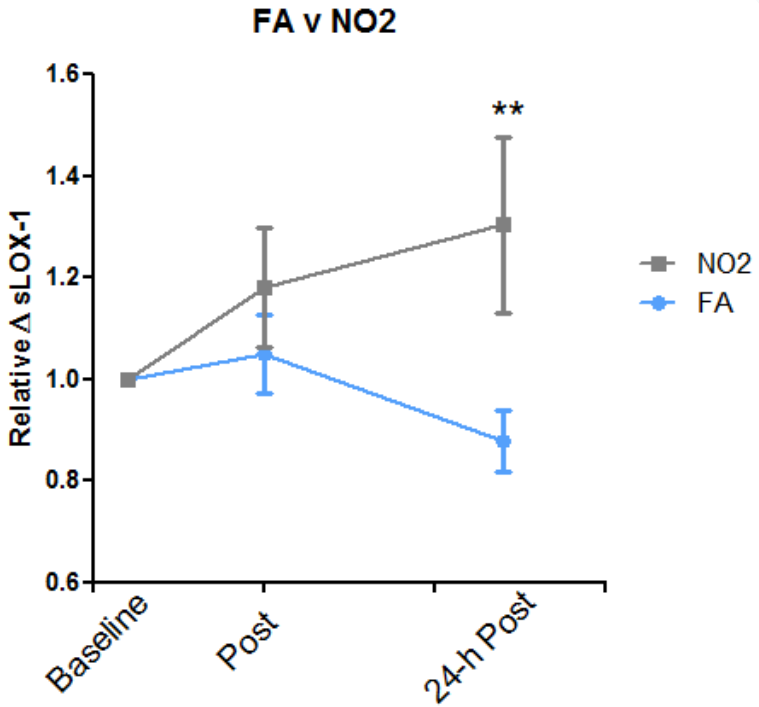


Mixed Emissions (Gasoline + Diesel) Upregulate Vascular Lipid Peroxidation and Inflammation via LOX-1-Dependent Pathway

How Pertinent are All These Findings to Human Responses?

- **Obtained samples from human diesel exposures at UNC/EPA in Chapel Hill**
- **9 volunteers exposed to diesel at 100 μg PM/m^3 or filtered air for 2 hours**
  - **Each individual was exposed to both scenarios, enabling pairwise statistics**
- **8 volunteers exposed to NO_2 at 500 ppb or filtered air for 2 hours**
- **Intermittent exercise on an ergometer/cycle**
- **Obtained plasma for pre, ~1-h post, and 24-h post**





Diesel and NO_2 Exposure in Humans Increases Soluble LOX1

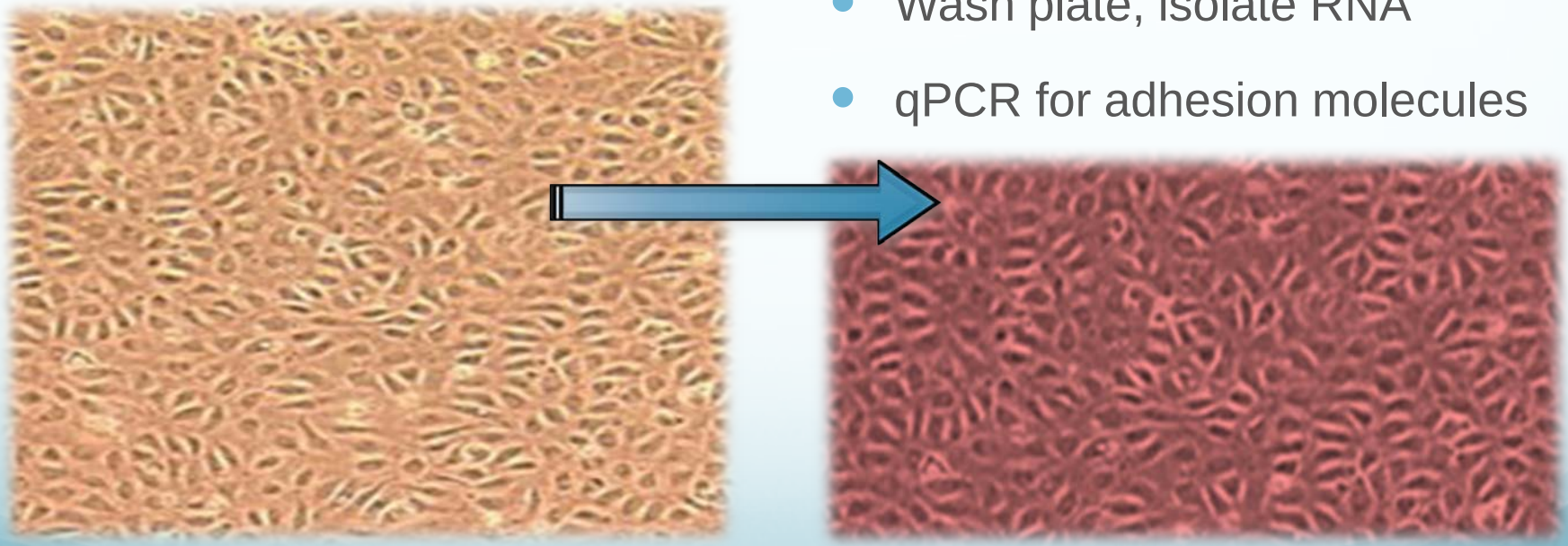
(also MMP9 and ET-1)

Complex Mixtures, Complex Responses

- **Identifying the components of blood that are altered by exposures is complicated**
  - **Many components**
  - **Concentration and form**
  - **Metabolism/dynamics**
  - **Receptor changes**
  - **Common pathways = cumulative effects!**
- **Functional outcome may have more meaningful interpretation**

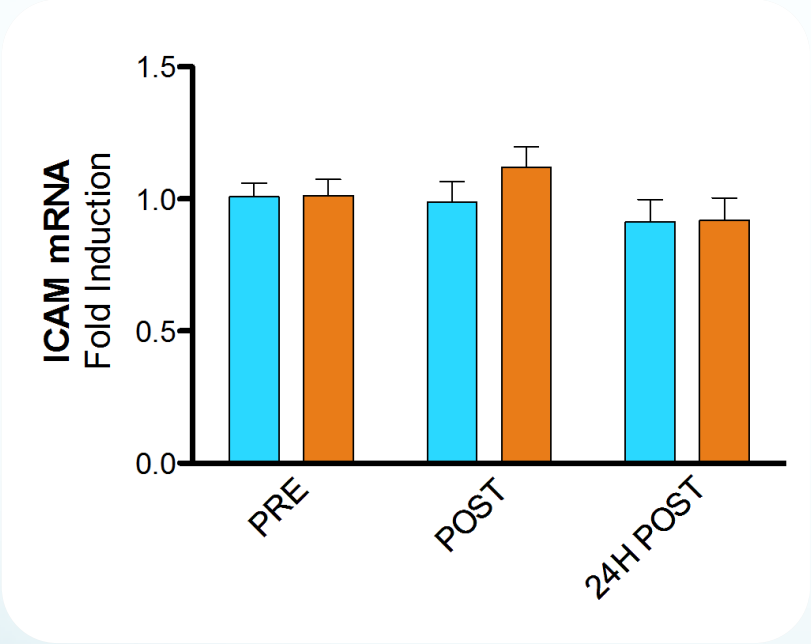
Inflammatory Potential Assay: General Technique

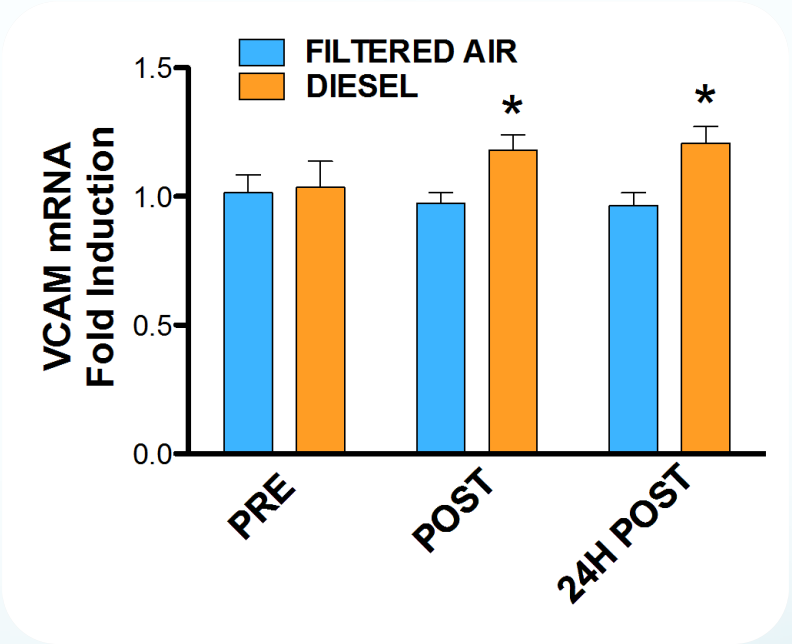
- Add plasma to media for coronary endothelial cells
- Incubate for 24 hours
- Wash plate, isolate RNA
- qPCR for adhesion molecules



Activation of Adhesion Molecules

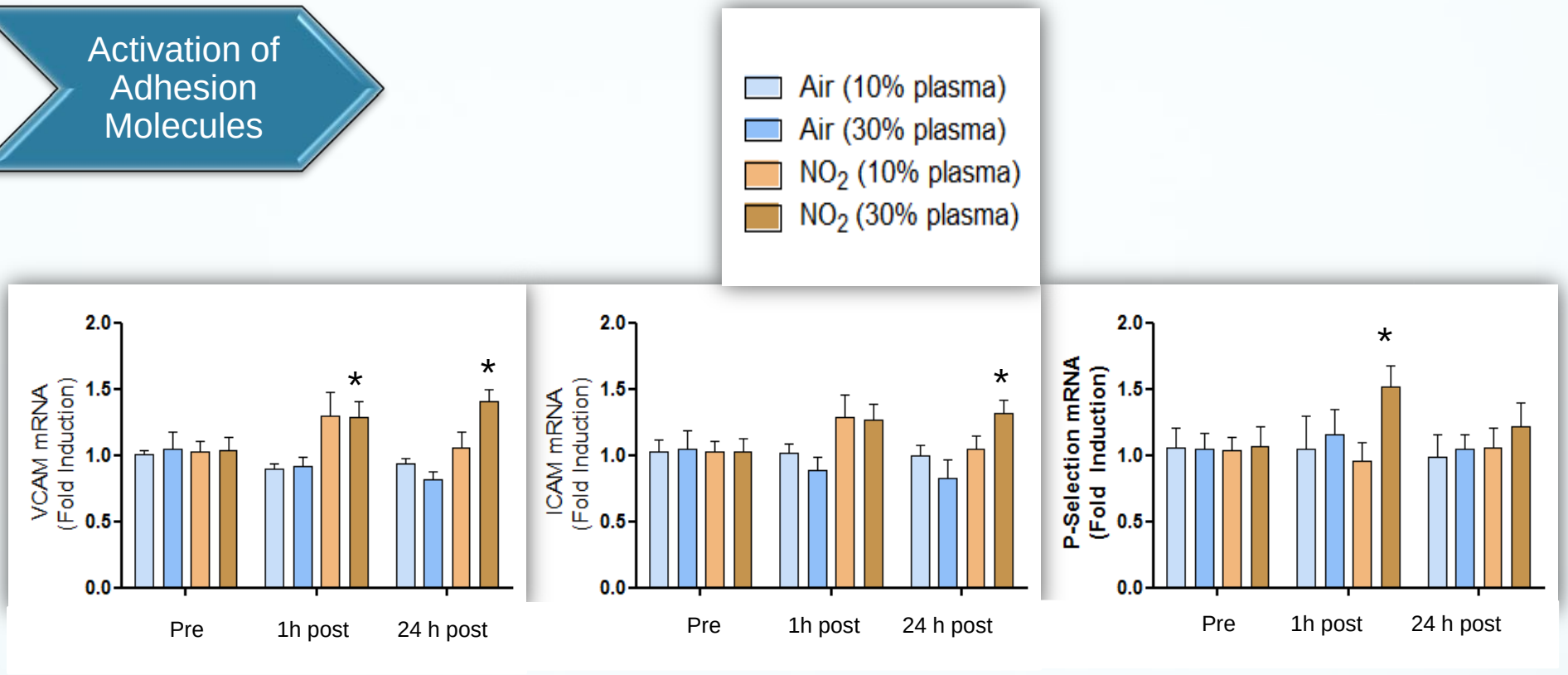
Treated primary human coronary endothelial cells for 24 with plasma (10% vol in media) from exposed subjects





Plasma Inflammatory Potential

Activation of Adhesion Molecules



In Vitro Inflammatory Potential in Human Plasma after NO_2 Exposure

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