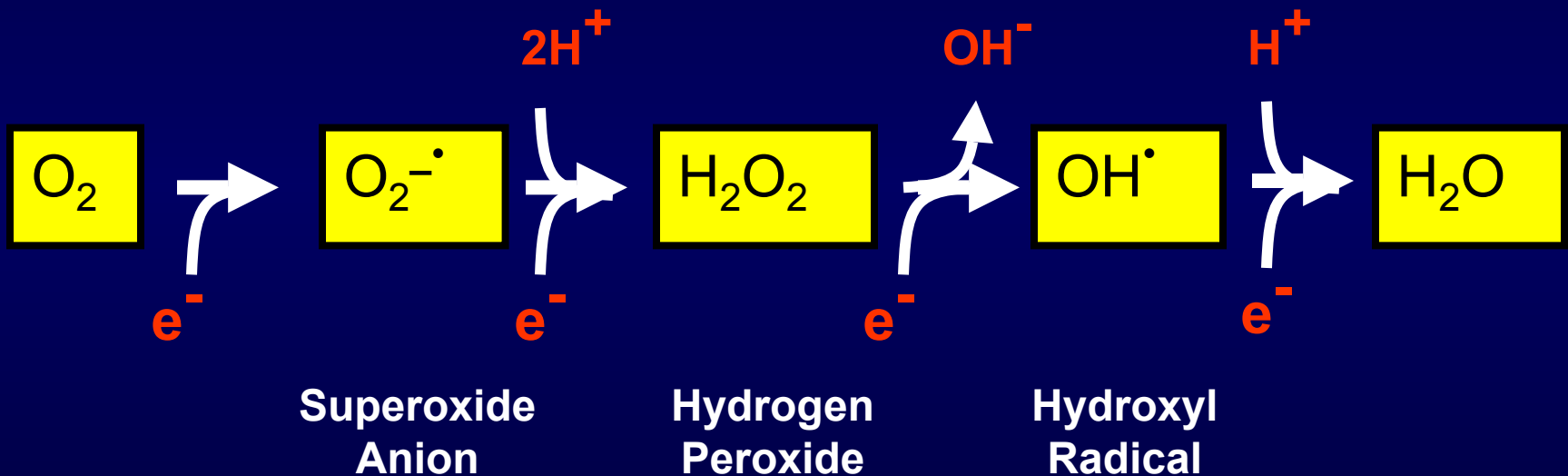


Three Tiers of Oxidative Stress in Response to Particulate Air Pollutants

Andre Nel MD/ PhD

Professor of Medicine at UCLA

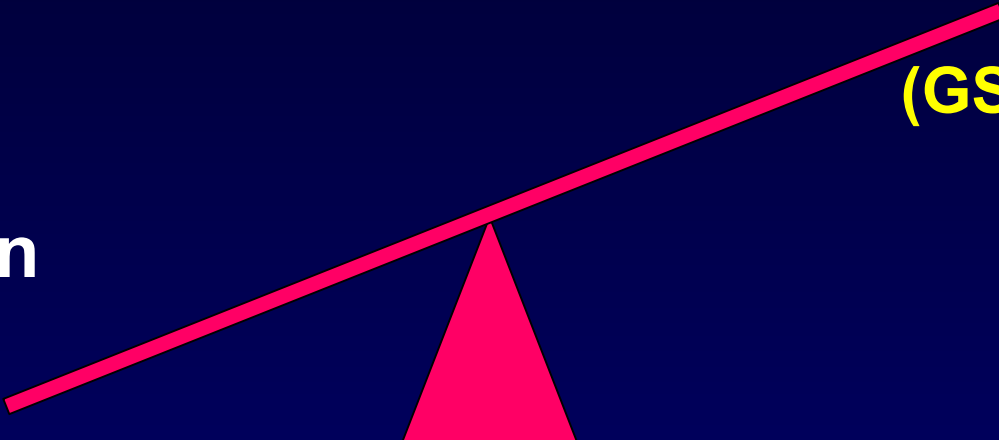
Reactive Oxygen Species (ROS)



Normal

**ROS
Inactivation**
(GSSG lo)

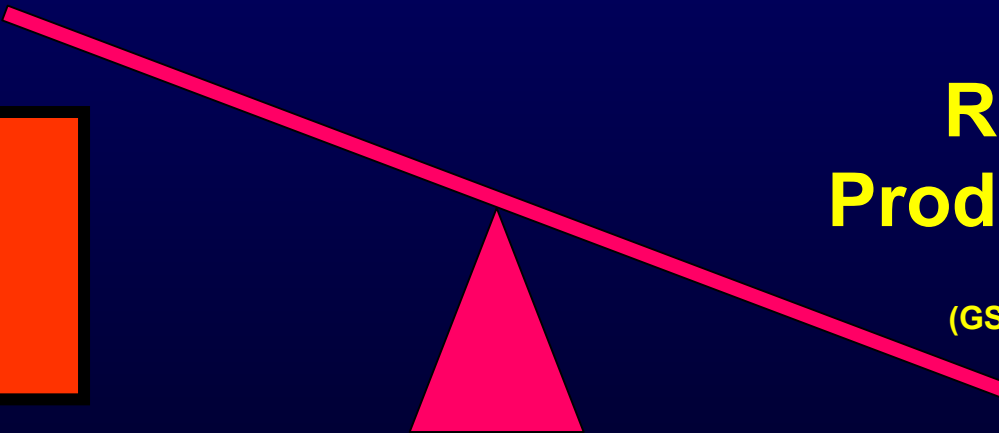
**ROS
Production**
(GSH hi)



**ROS
Inactivation**
(GSSG hi)

**ROS
Production**
(GSH lo)

**Oxidative
Stress**



Oxidative Stress Hypothesis

1. PM contains pro-oxidative chemicals
2. PM chemicals generate ROS → Oxidative stress
3. Oxidative stress → inflammation

Important Organic Chemicals

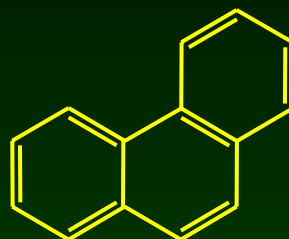
PAHs = Aromatic fraction



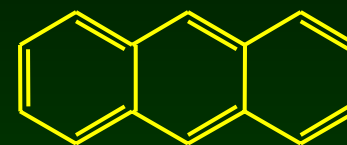
Naphthalene



Benzo(a)pyrene

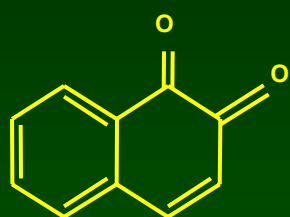


Phenanthrene

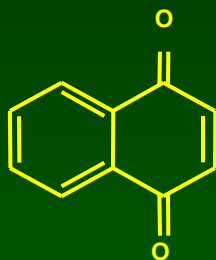


Anthracene

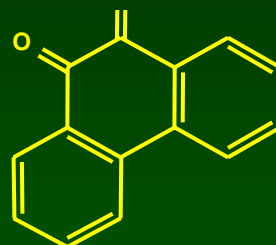
Quinones = Polar fraction



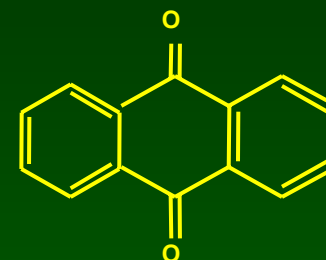
1,2-Naphthaquinone
(1,2-NQ)



1,4-Naphthaquinone
(1,4-NQ)

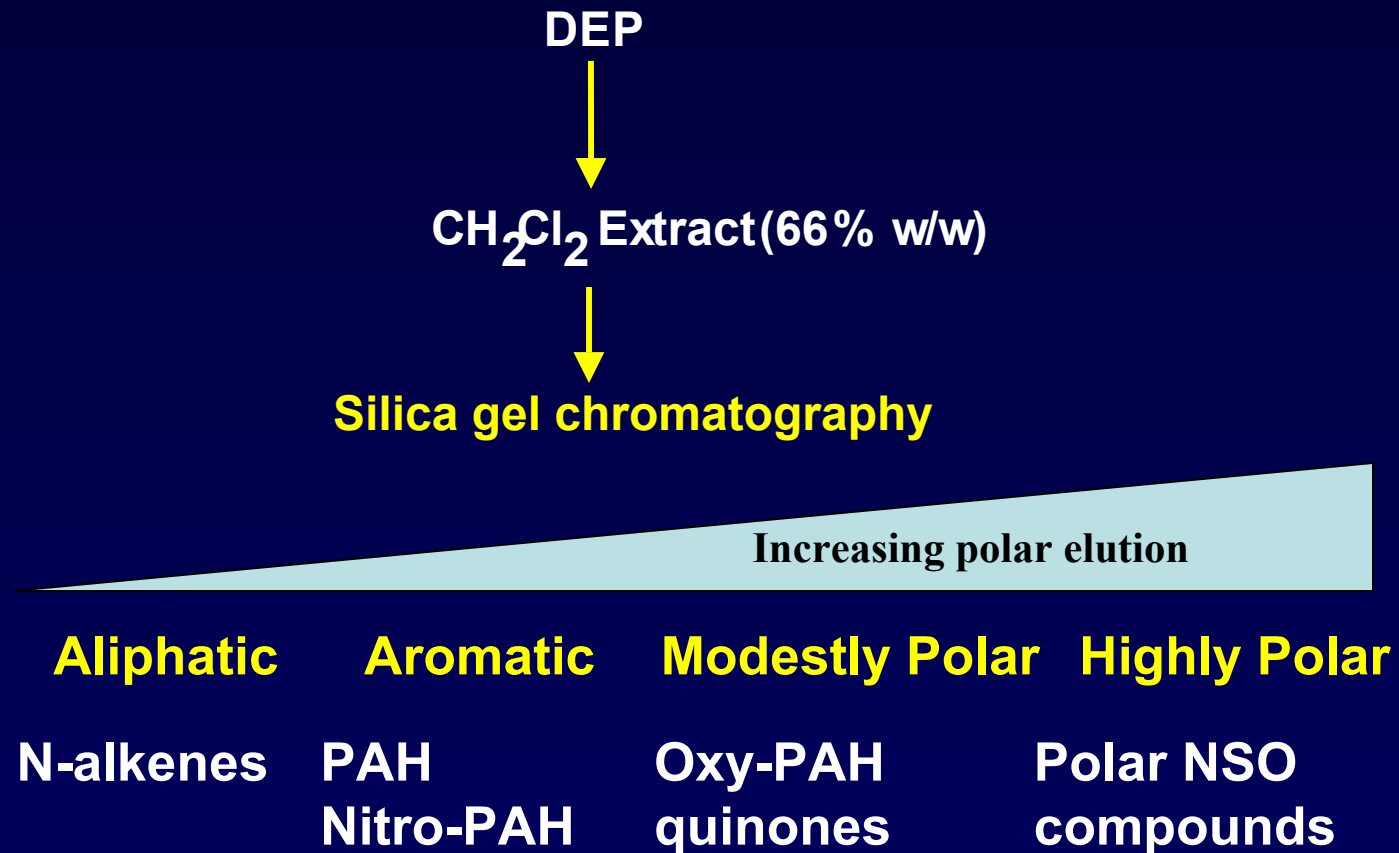


9,10-Phenanthraquinone
(9,10-PQ)

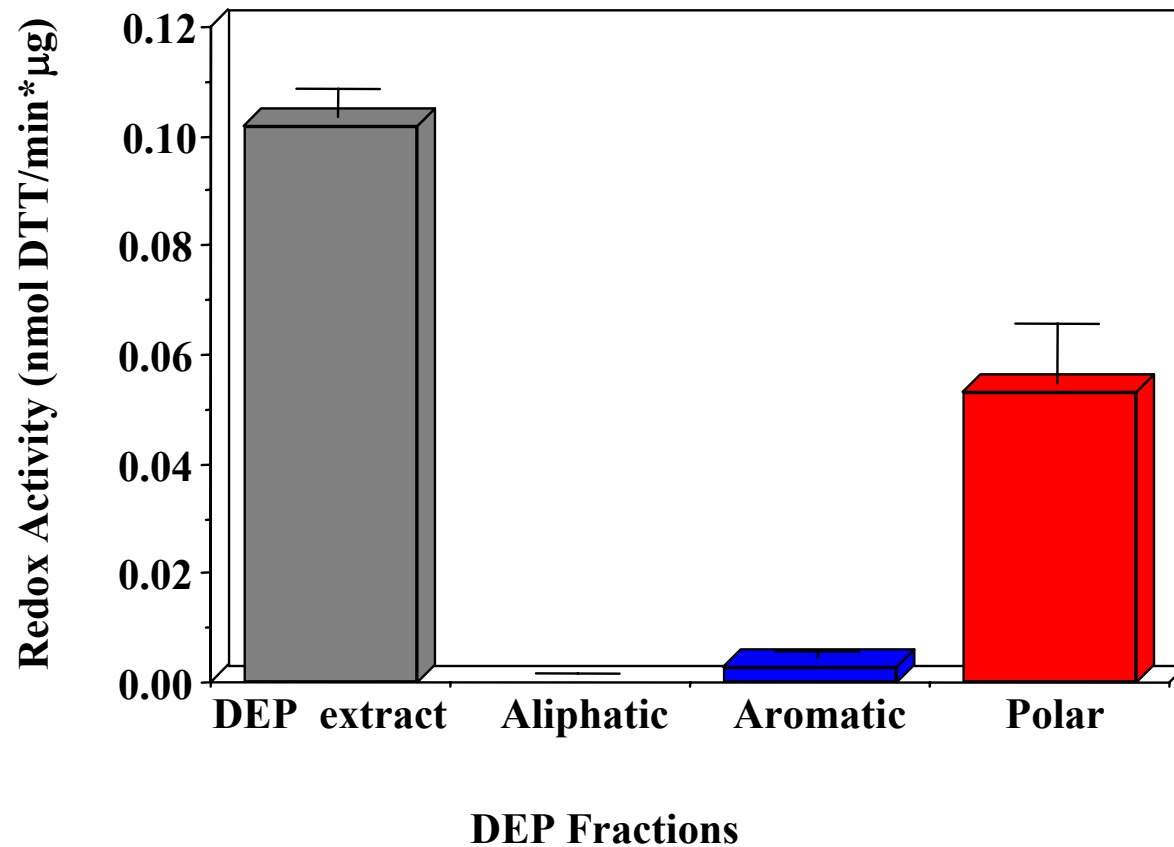


9,10-Anthraquinone
(9,10-AQ)

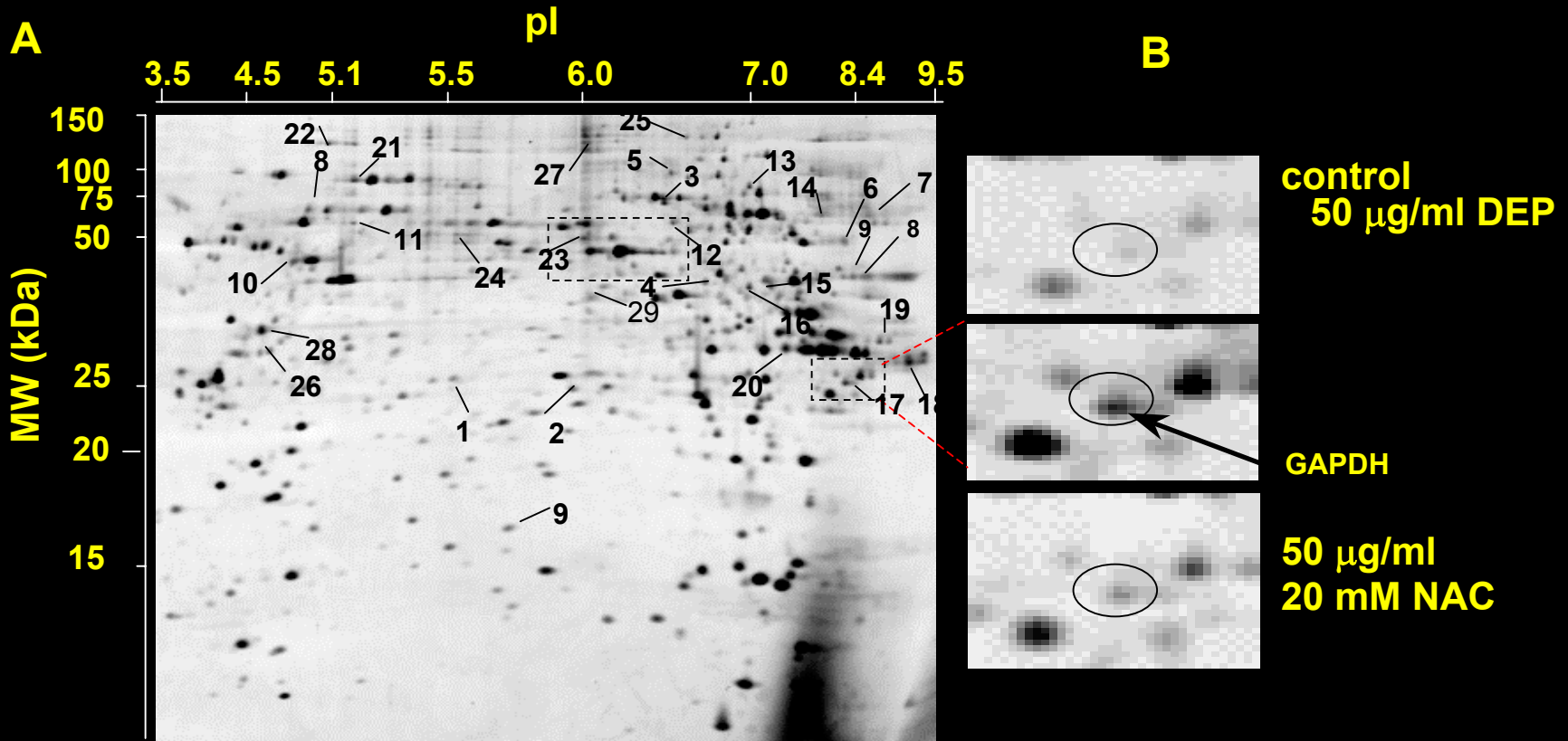
Organic chemical fractionation of DEP



Differential Redox Cycling ability of individual DEP chemical fractions

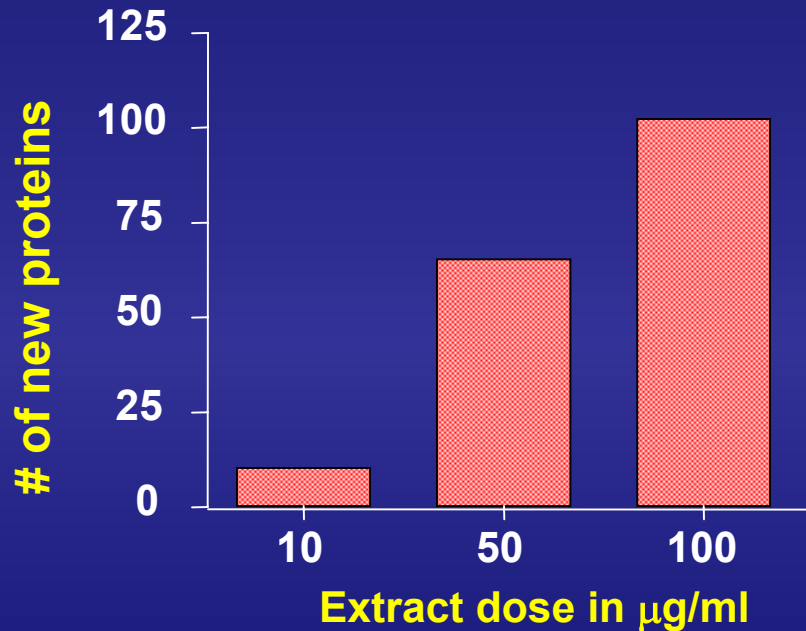
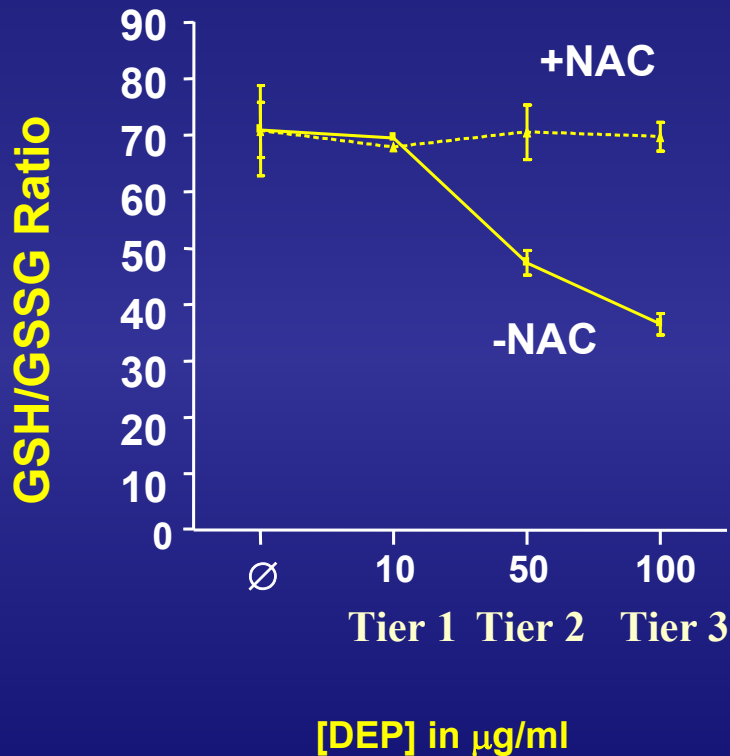


Exploration of the role of Oxidative Stress through the use of Proteomics

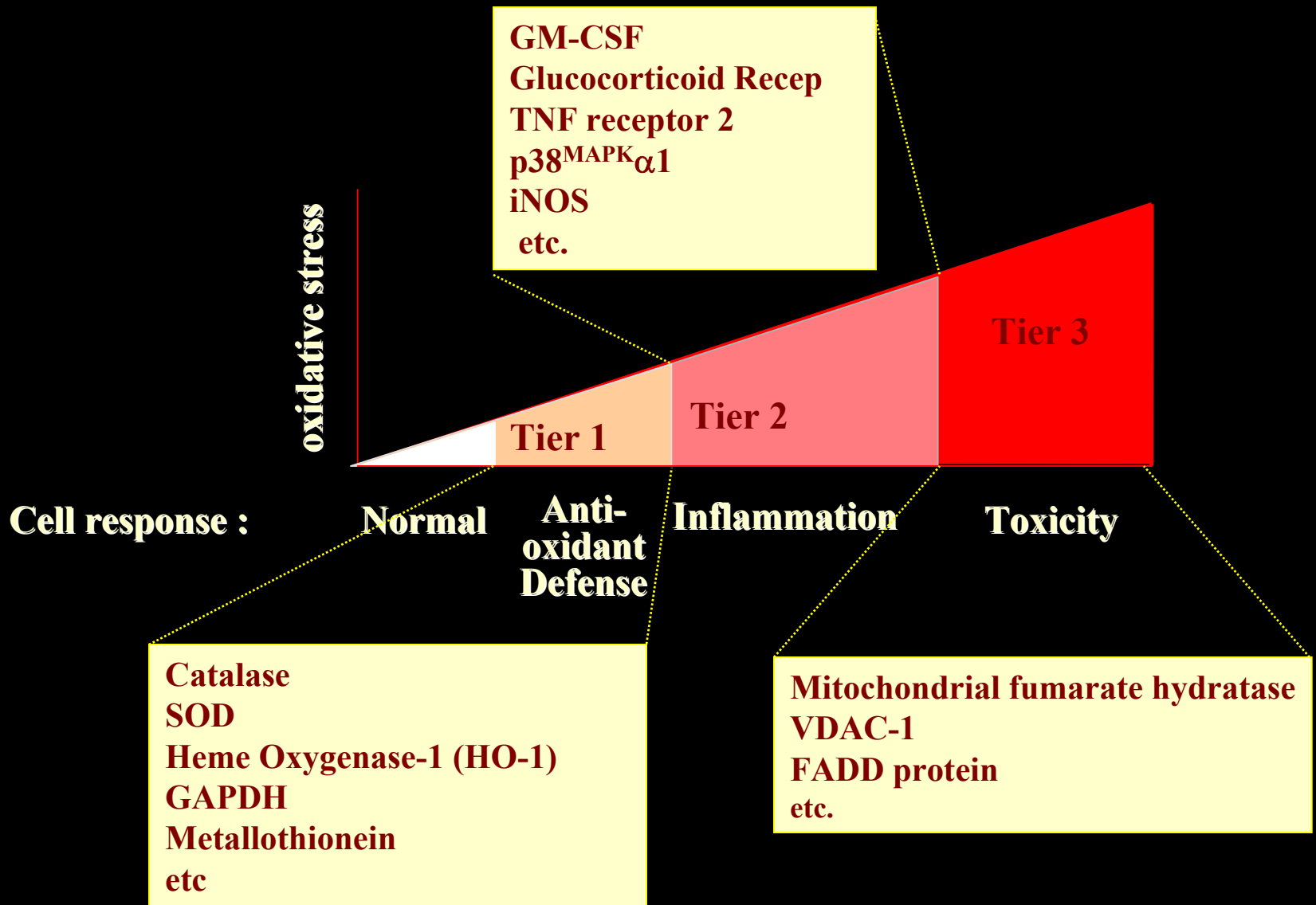


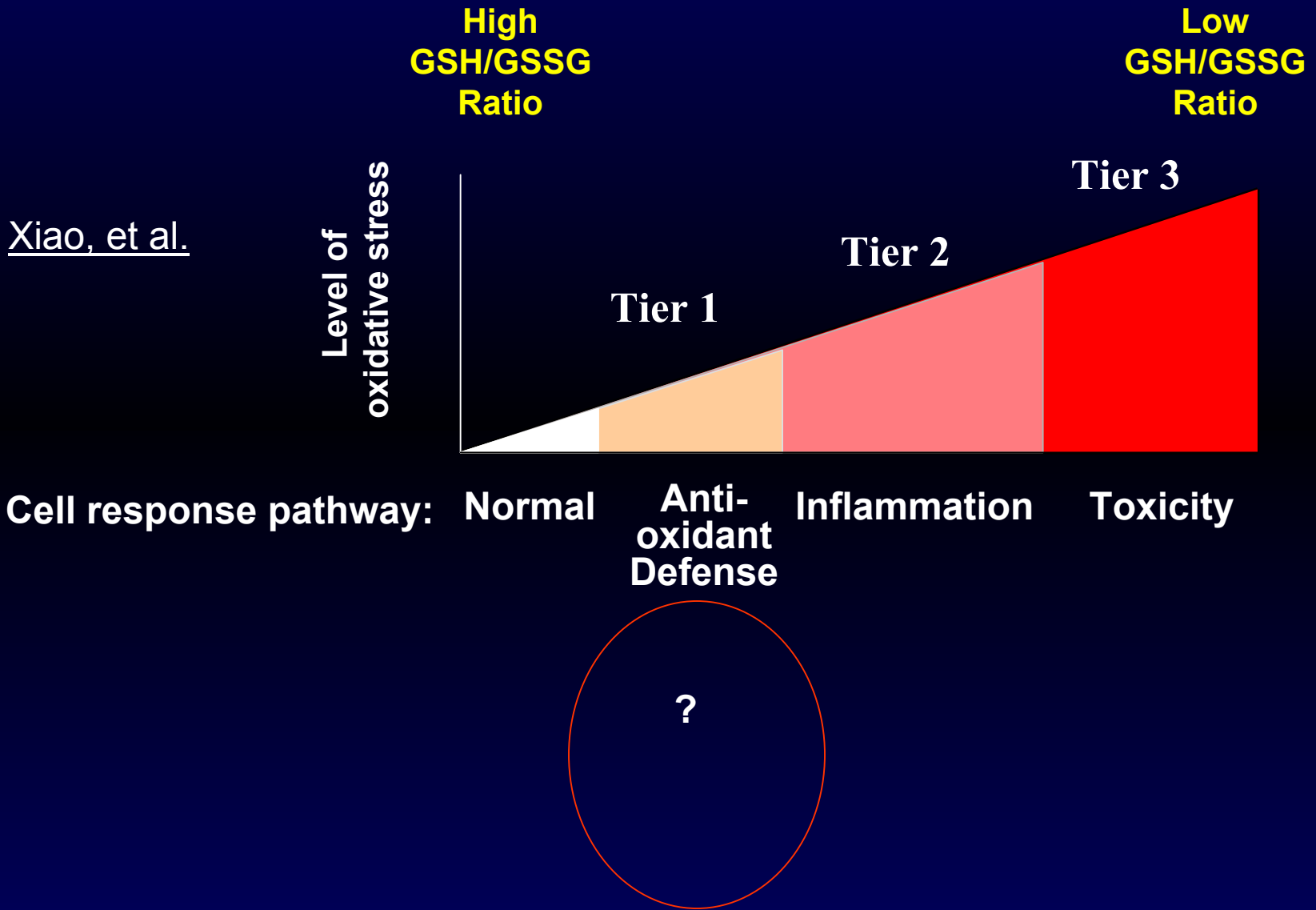
Oxidative stress response ?
Inflammatory markers ?

Dose-dependant induction of new Oxidative Stress Proteins by DEP



Macrophage/epithelial oxidative stress analysis (in vitro)





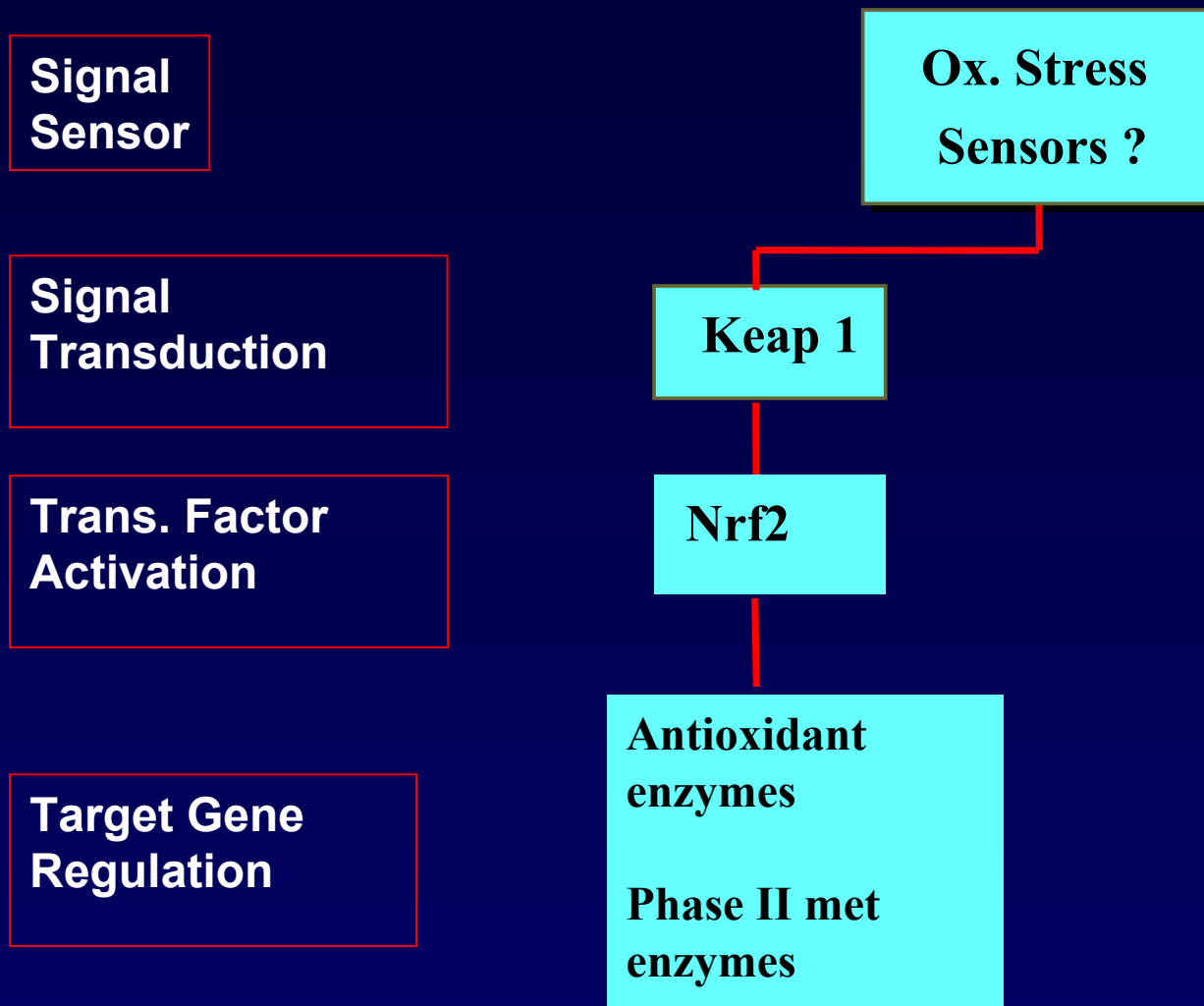
Extrapolation of the Stratified Oxidative Stress hypothesis to Asthma & Atherosclerosis

Prediction: people with defective anti-oxidant defense will more readily develop airway inflammation in response to exposure to particulate pollutants

.... and may help to answer

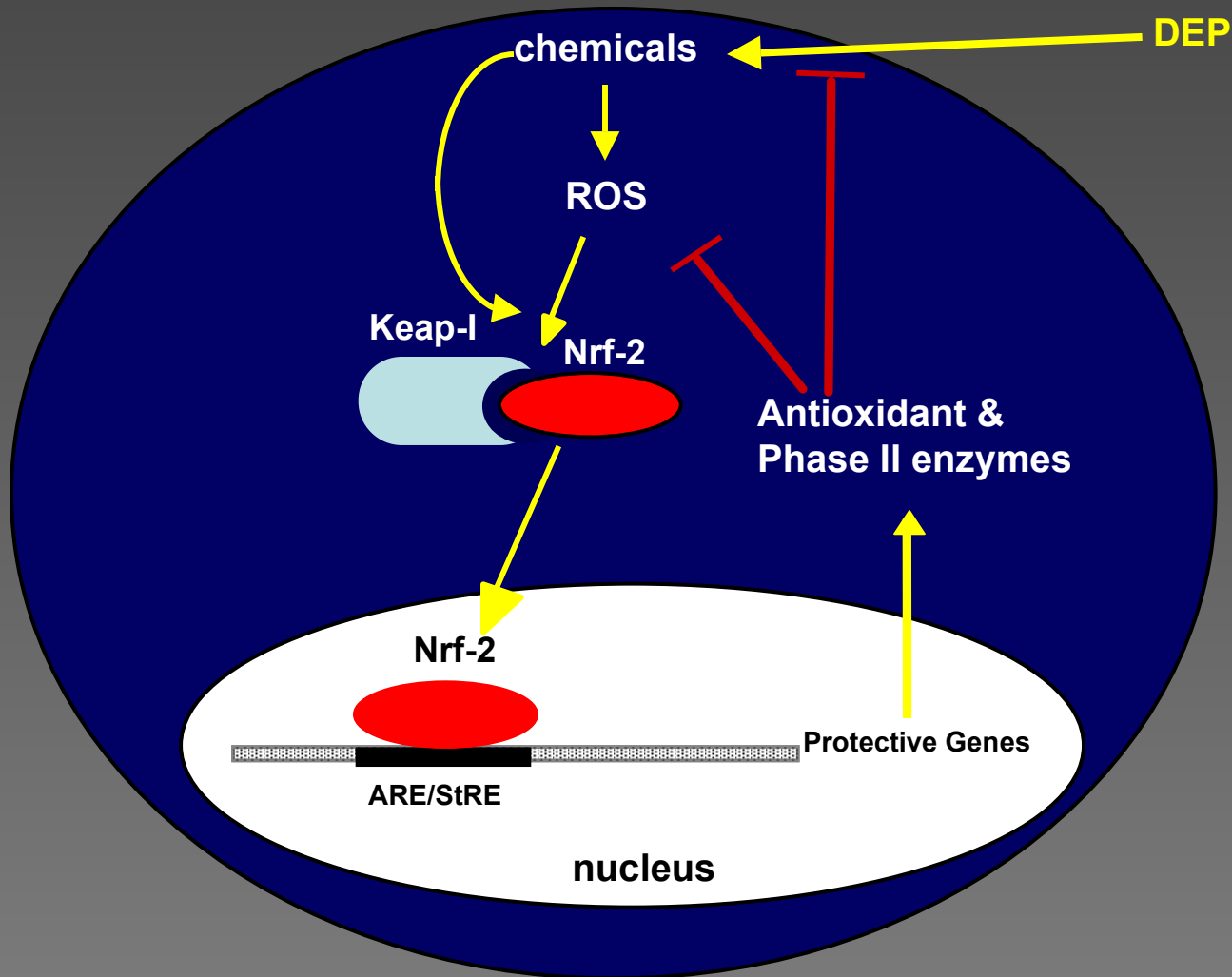
Who is susceptible to PM?

Adaptive Genetic Programming

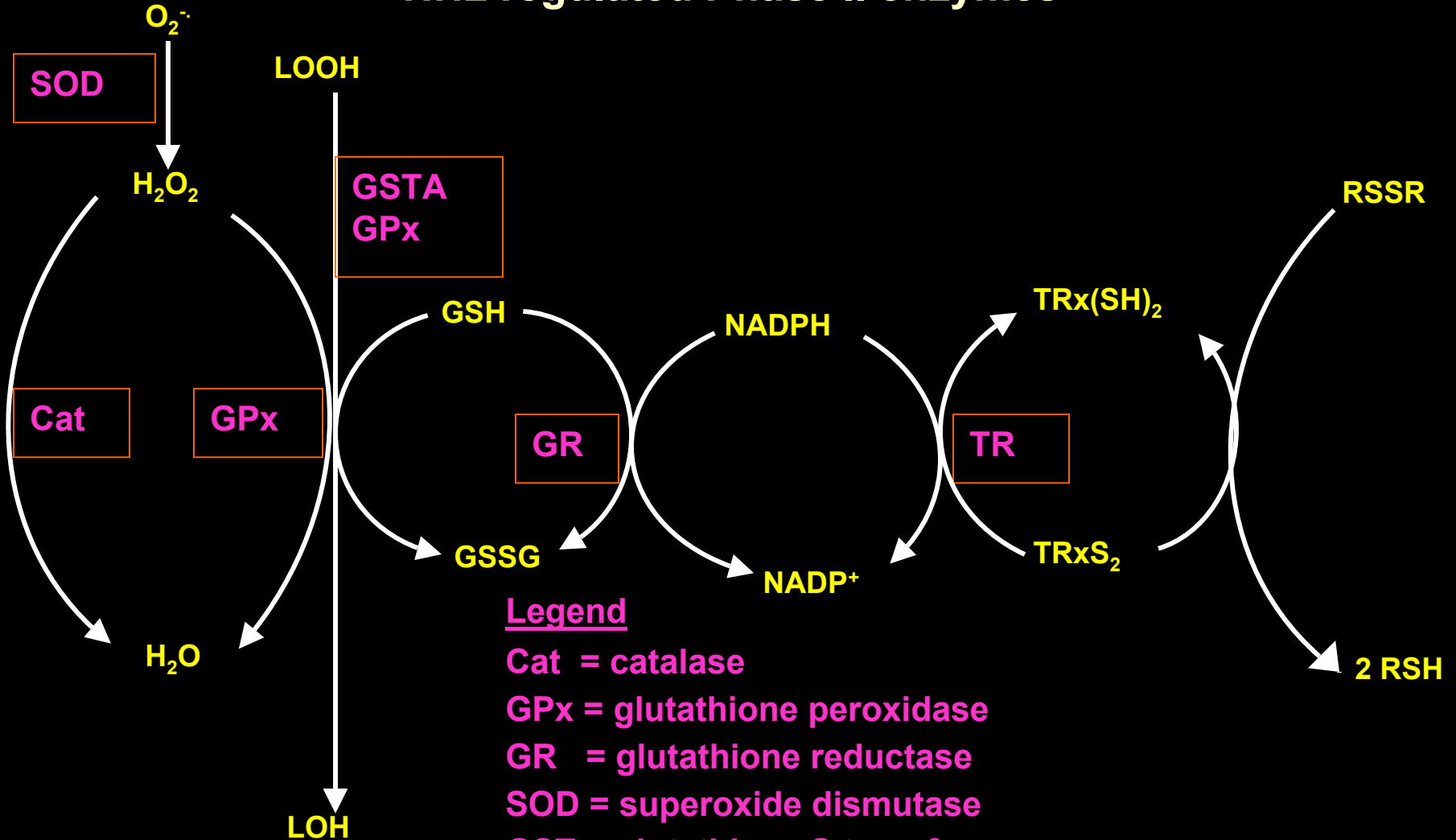


Role of the Antioxidant Response Element (ARE)

and the transcription factor, Nrf-2



Nrf2 regulated Phase II enzymes



Legend

Cat = catalase

GPx = glutathione peroxidase

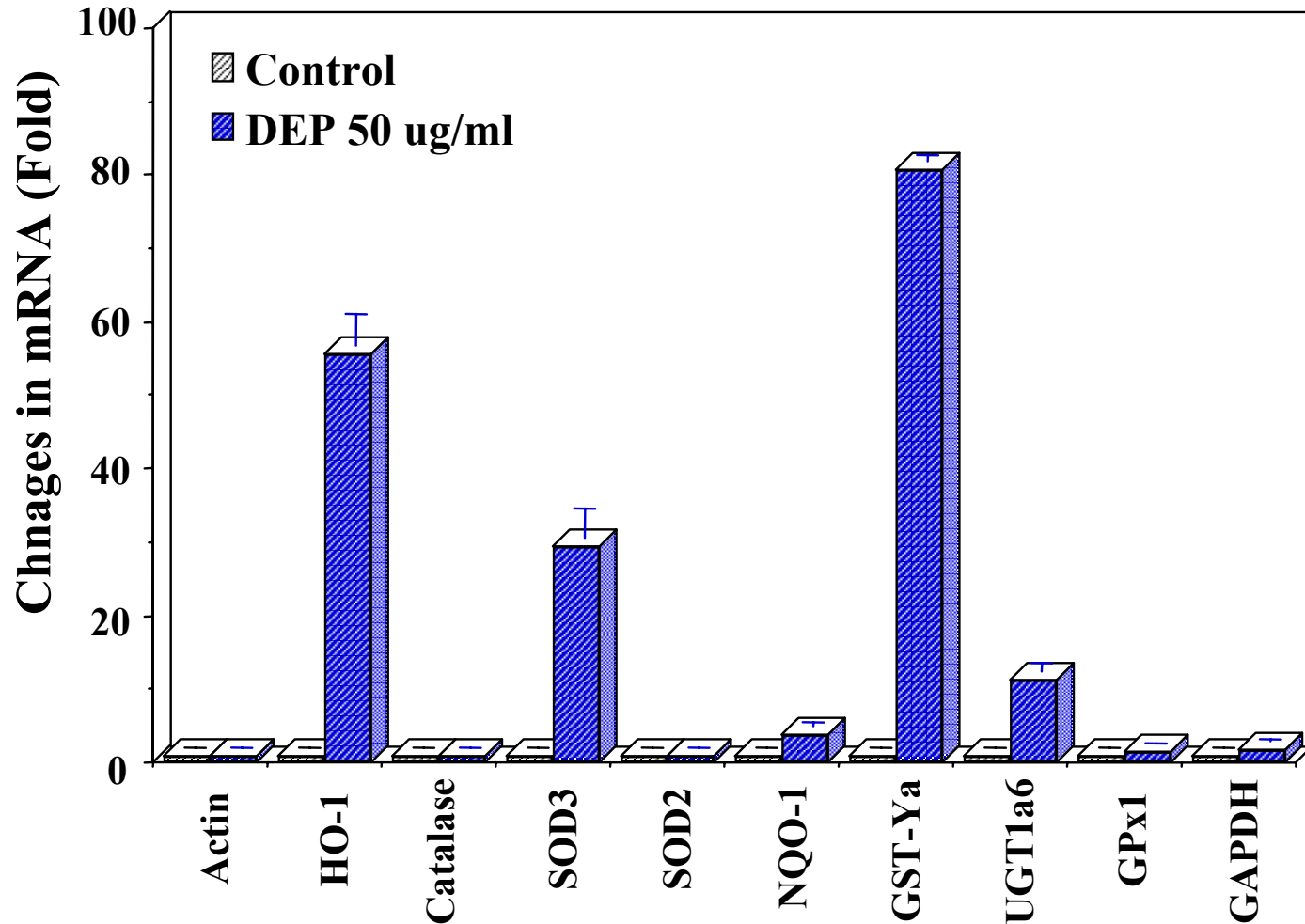
GR = glutathione reductase

SOD = superoxide dismutase

GST = glutathione S transferase

TR = thioredoxin reductase

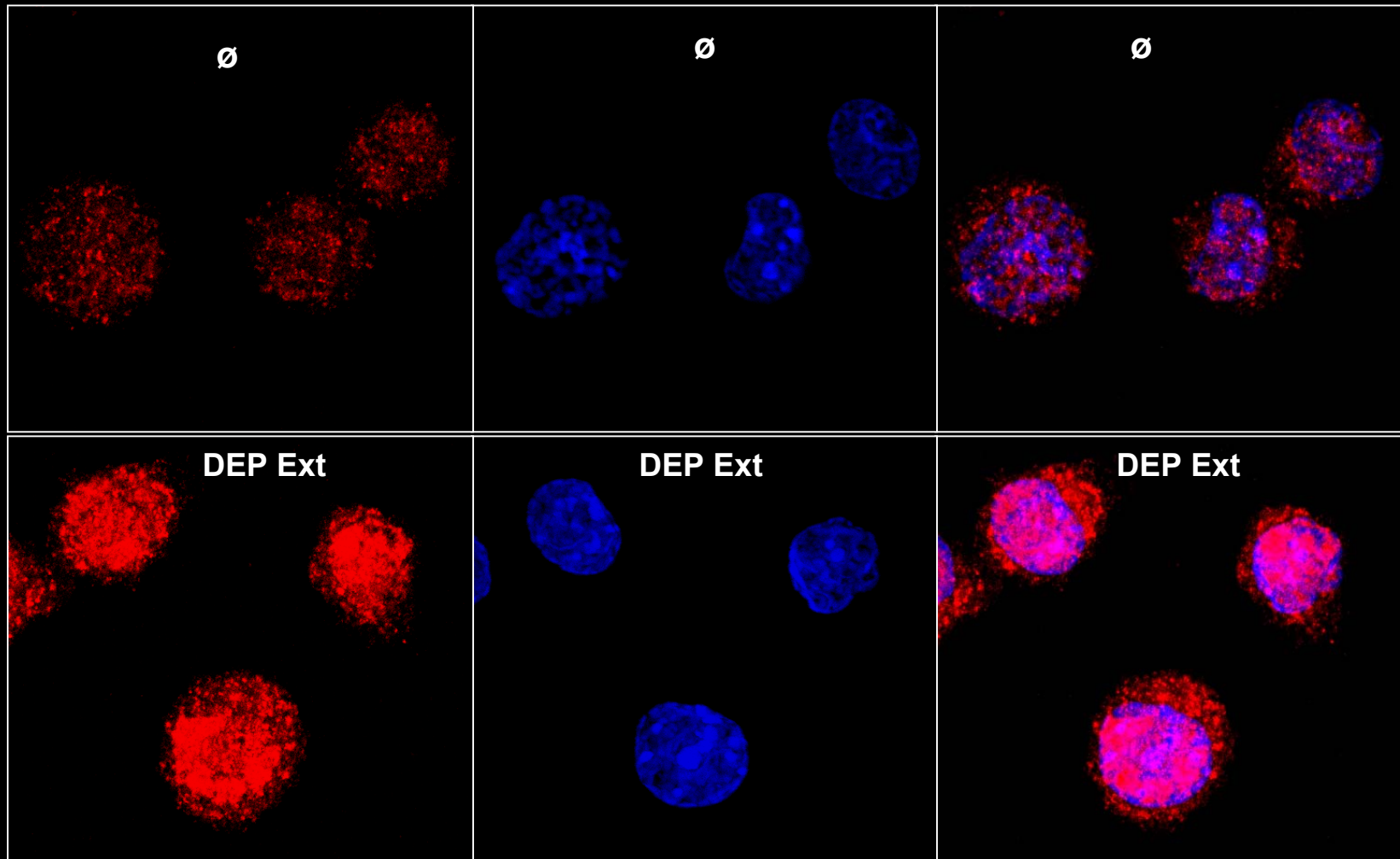
Phase II Enzyme expression by real-time PCR in macrophages



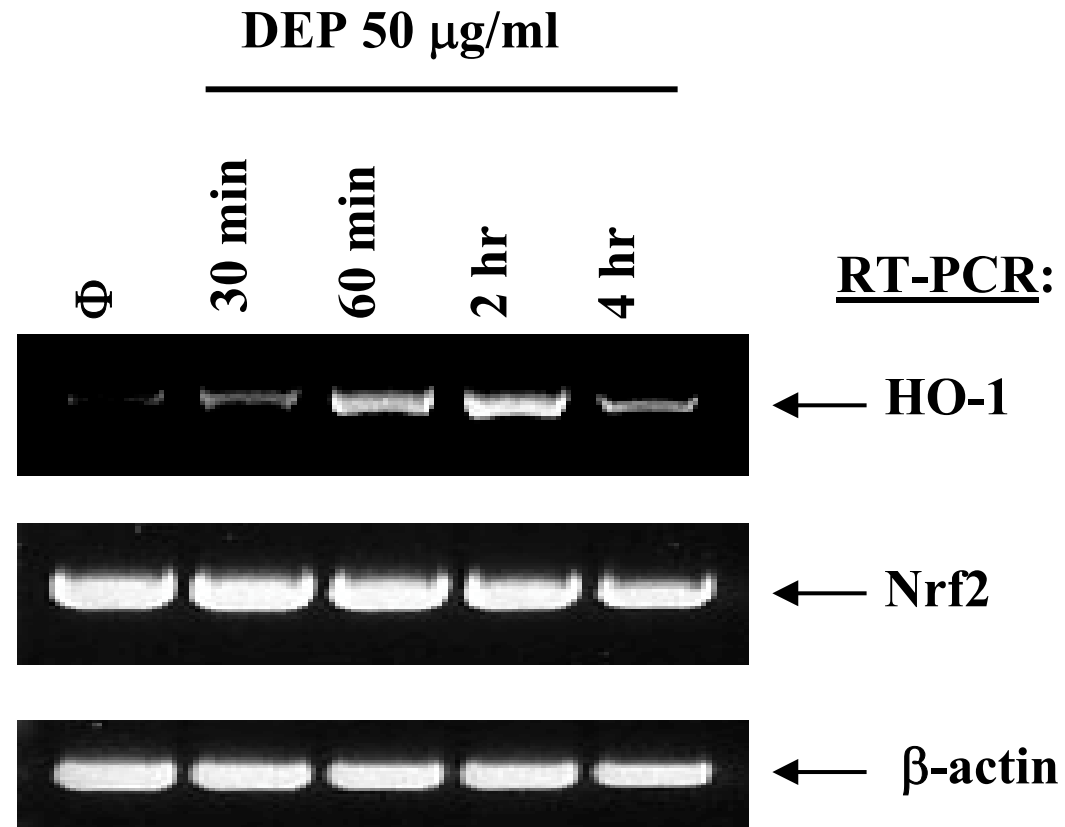
Anti-Nrf2

DAPI

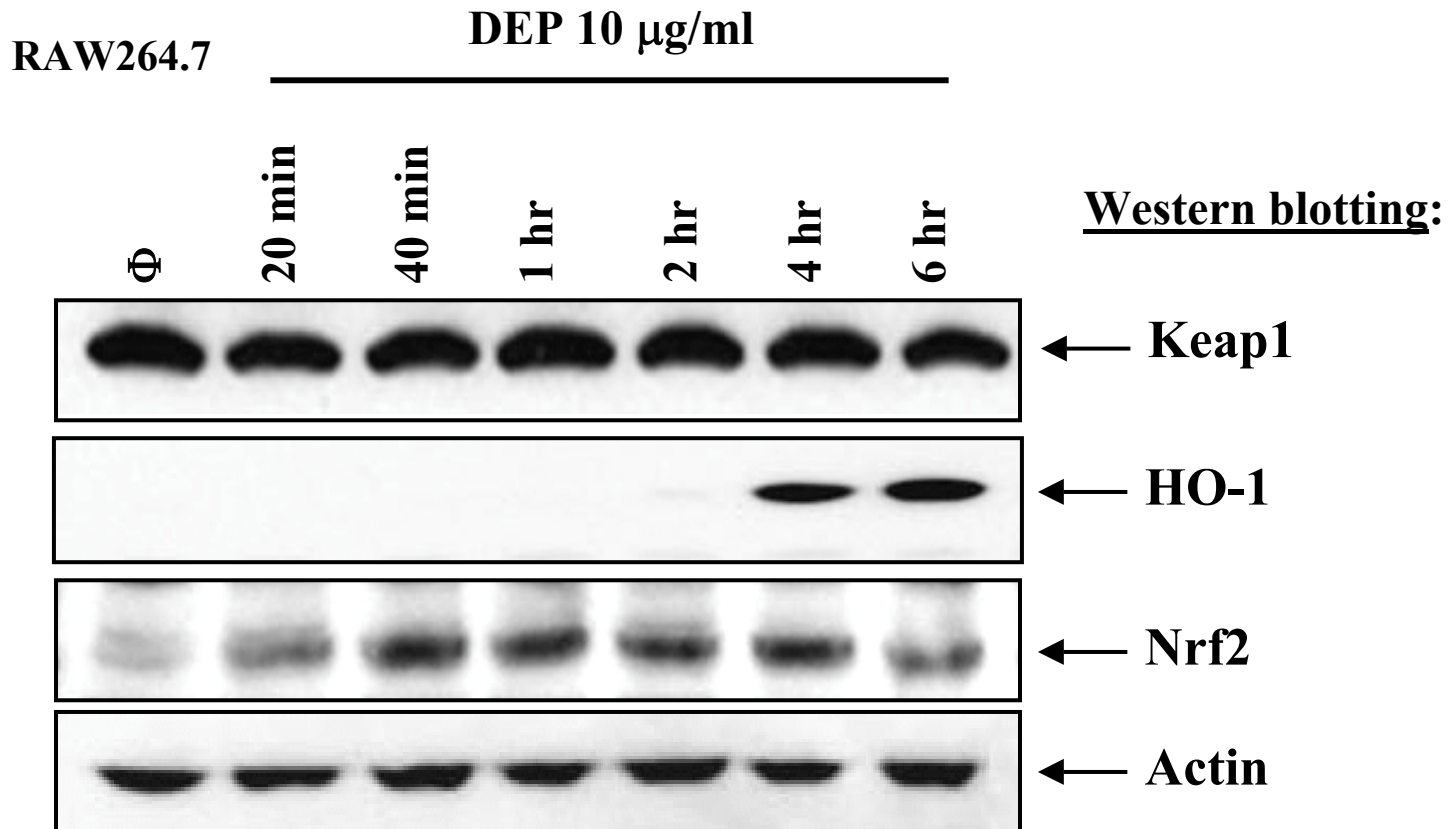
Composite



Nrf2 message is not increased during DEP treatment

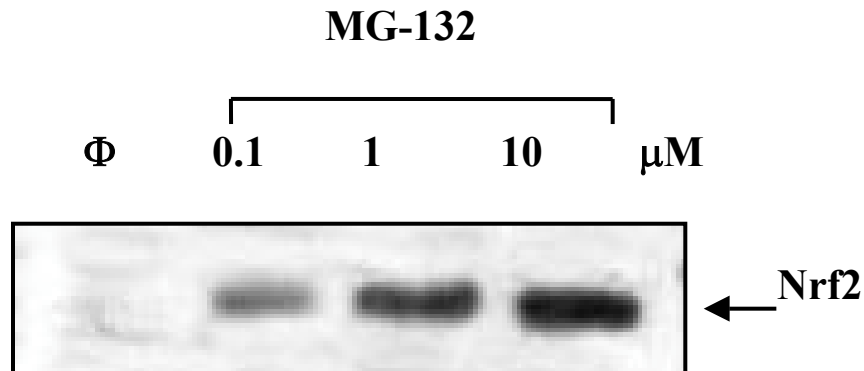


... but Nrf2 protein levels are increased by DEP treatment



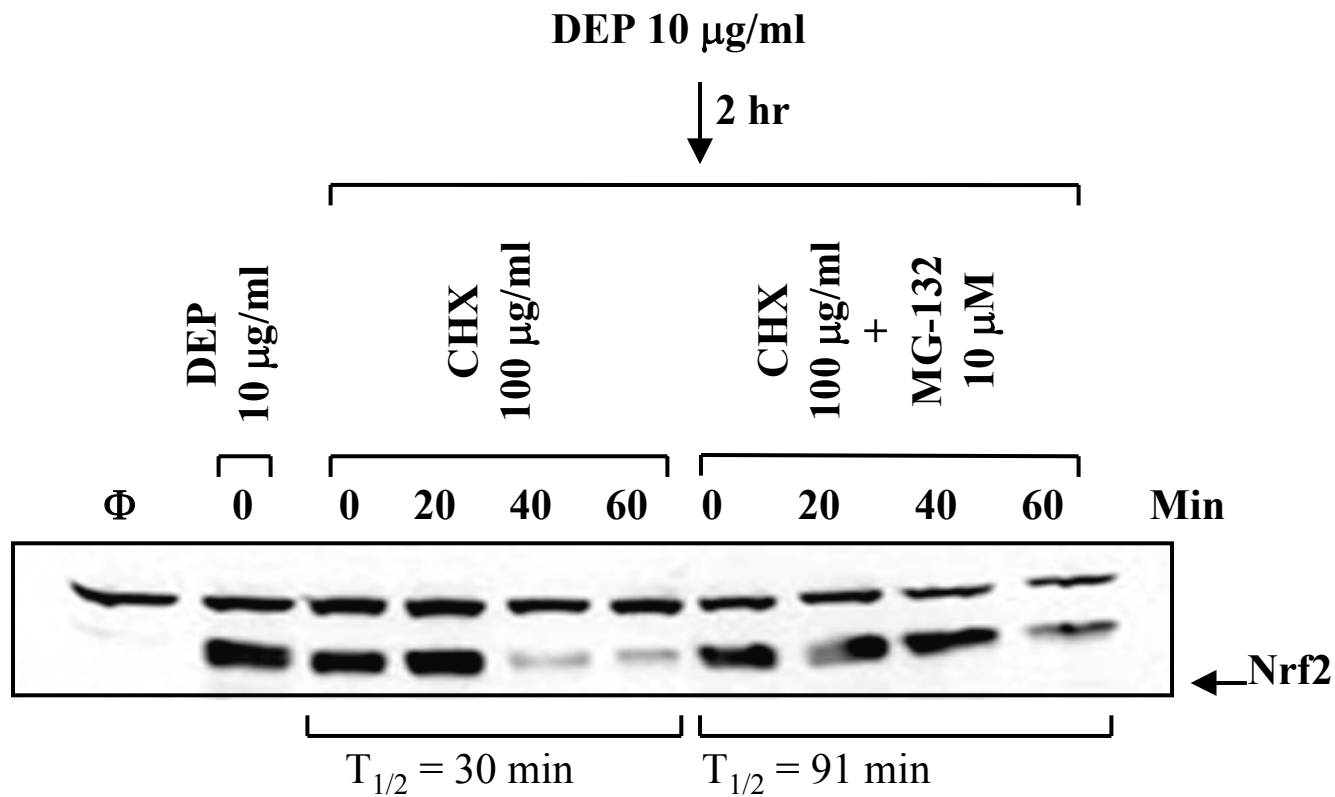
...suggesting post-transcriptional regulation

Nrf2 protein levels are increased by MG-132 - an inhibitor of the 26S proteasome

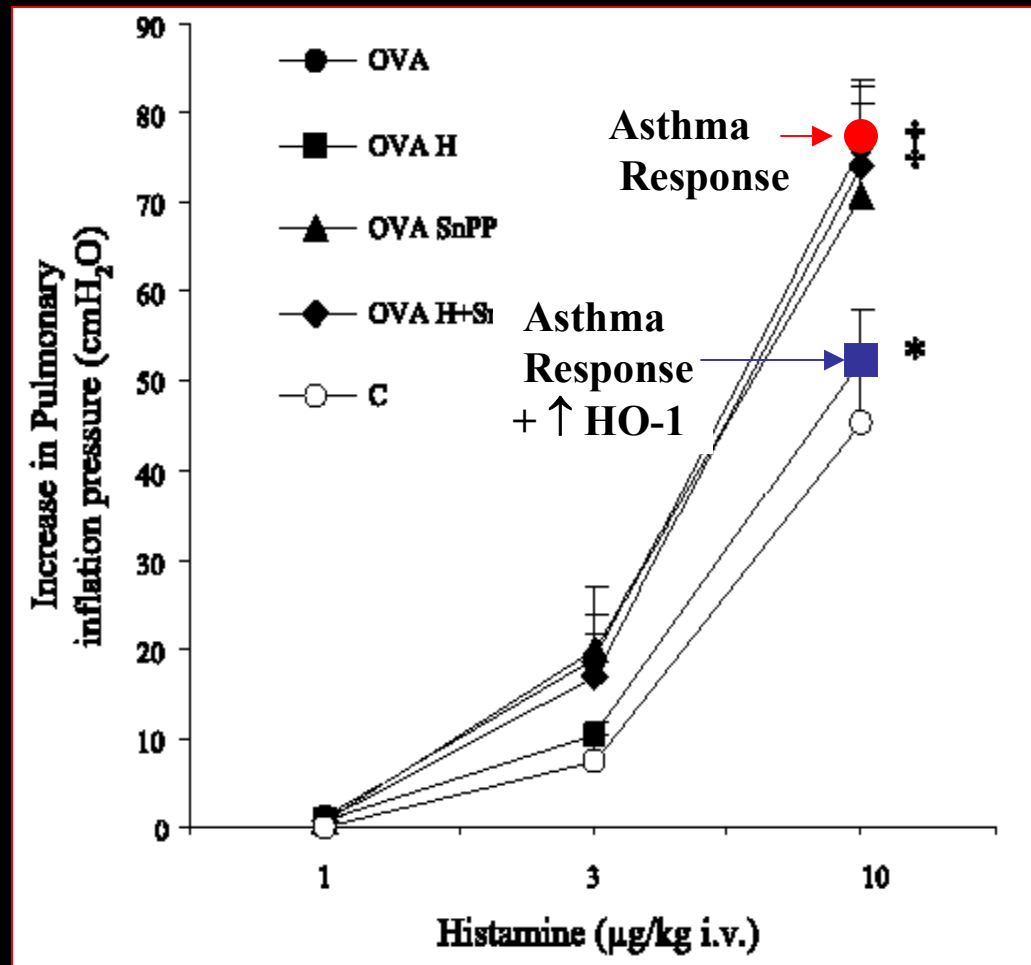


...demonstrating that Nrf2 levels are regulated by proteasomal degradation

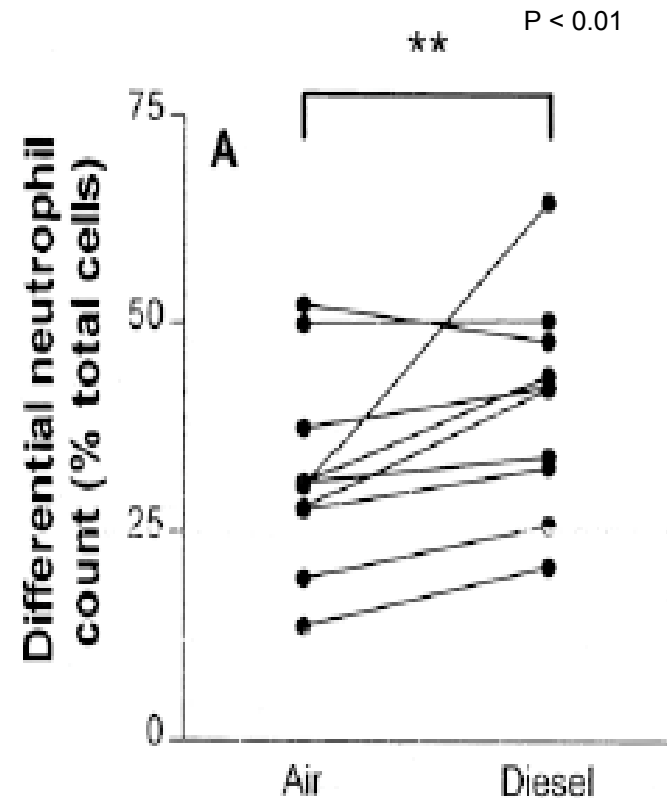
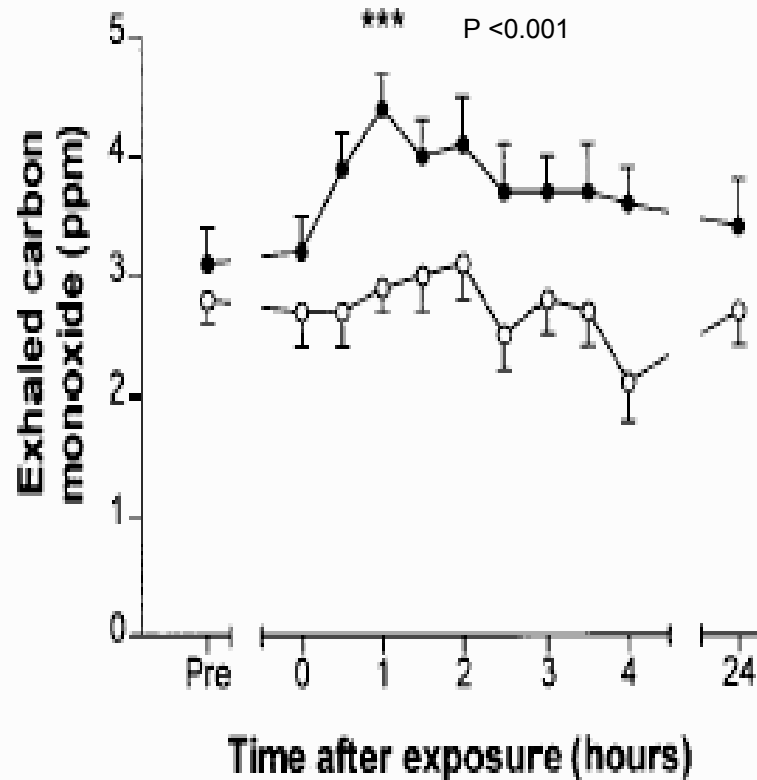
Nrf2 protein accumulation is regulated through an effect on protein stability (half life)



Heme Oxygenase attenuates Allergen-induced Airway Inflammation and Hyperreactivity in Guinea Pigs



CO exhalation is a sensitive clinical marker for the effects of DEP on the HO-1 system in vivo

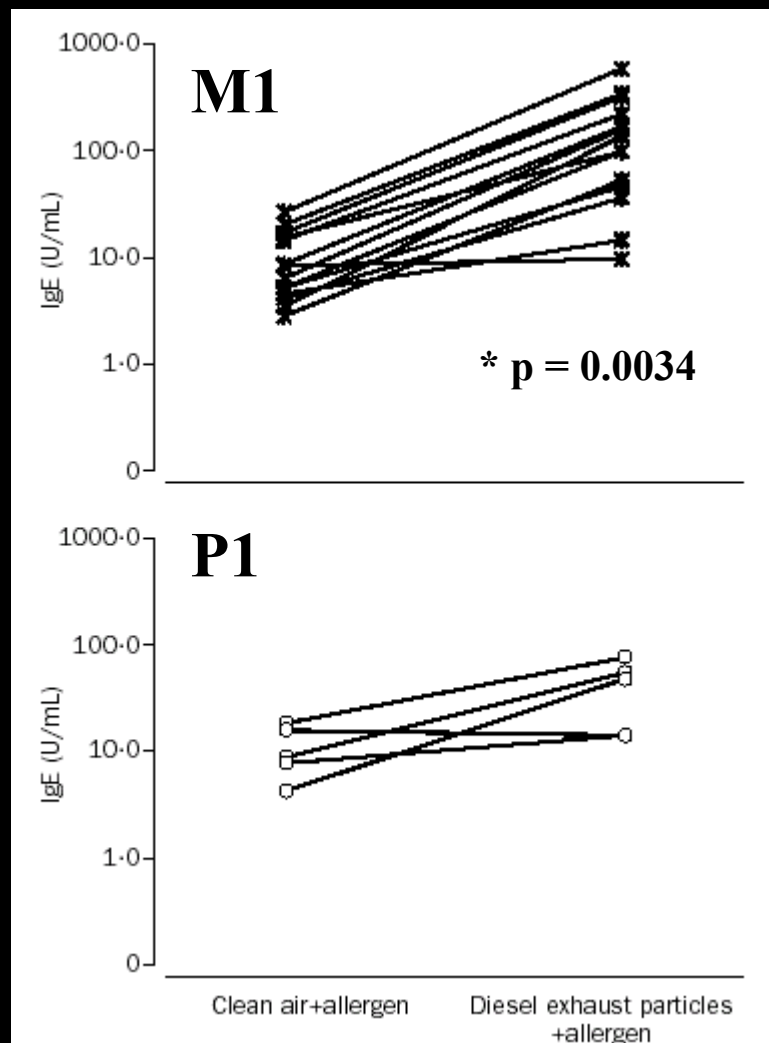


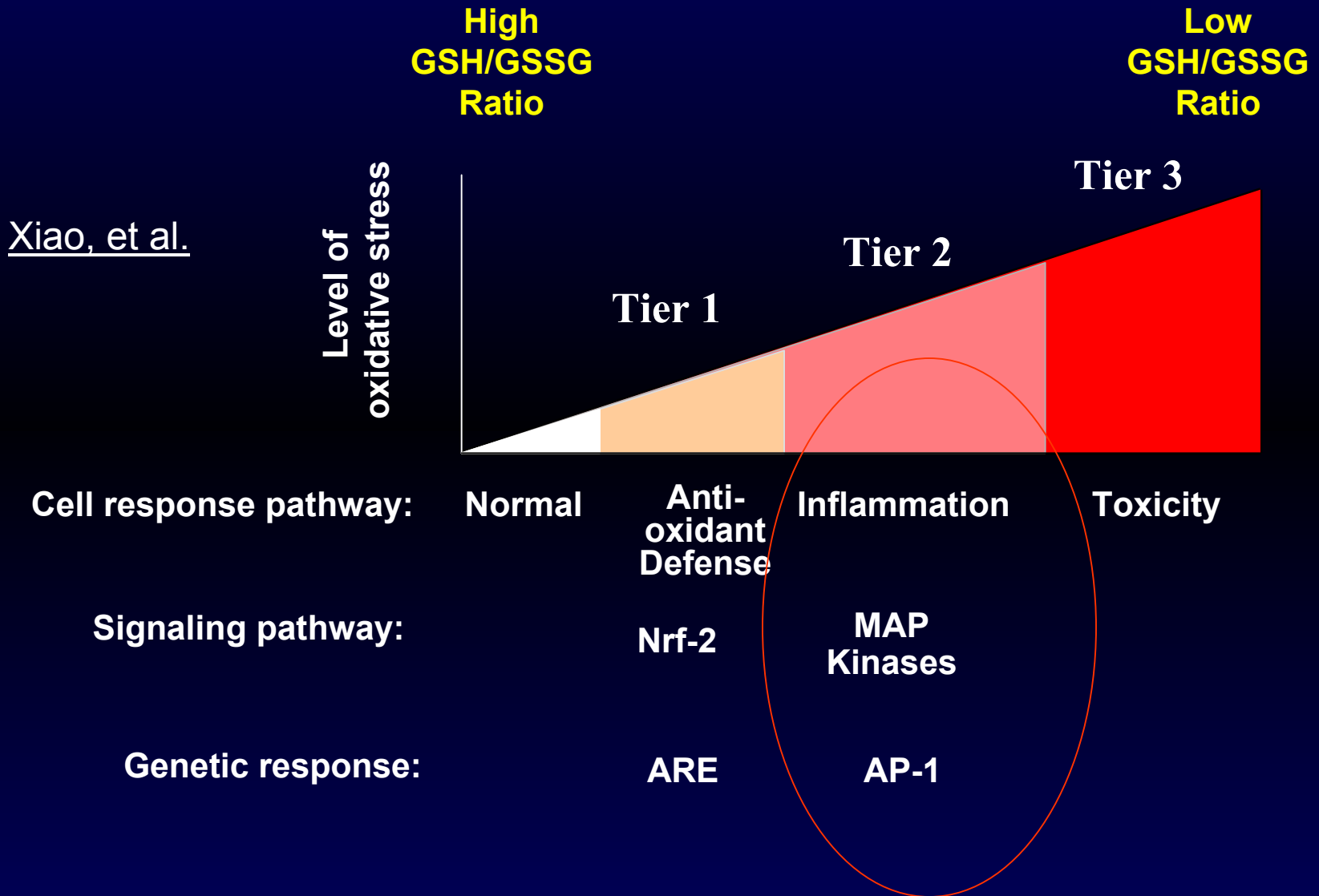
Altered Phase II Antioxidant Defense in Asthma

Association of GST genotypes/polymorphisms with asthma, including occupational asthma

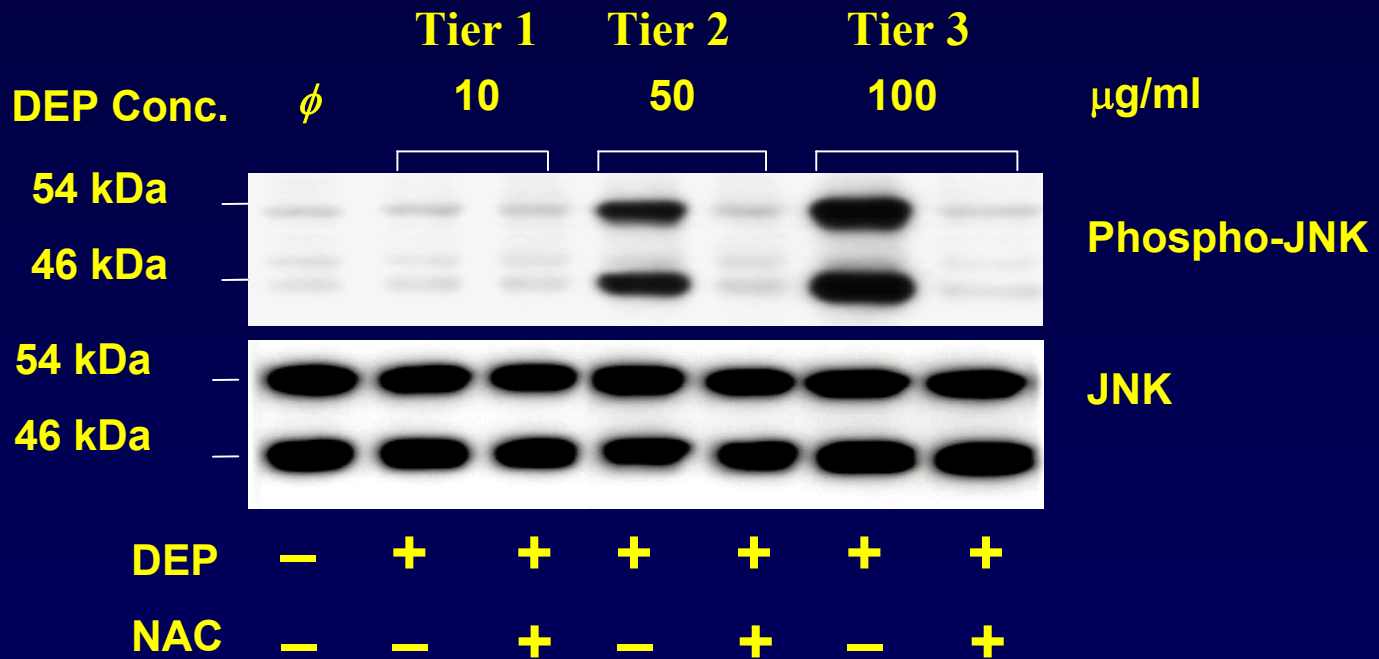
- a. **GST-M1 null genotype: Asthma risk ↑ 3.5 fold**
- b. **GST-P1 (Val¹⁰⁵/Val¹⁰⁵) 6-fold lower risk of asthma vs Ile¹⁰⁵/Ile¹⁰⁵ genotype**
- c. **GST-P1 (Val¹⁰⁵/Val¹⁰⁵) protects vs TDI-induced asthma**

Effect of GST M1 and P1 genotypes on xenobiotic enhancement of allergic responses



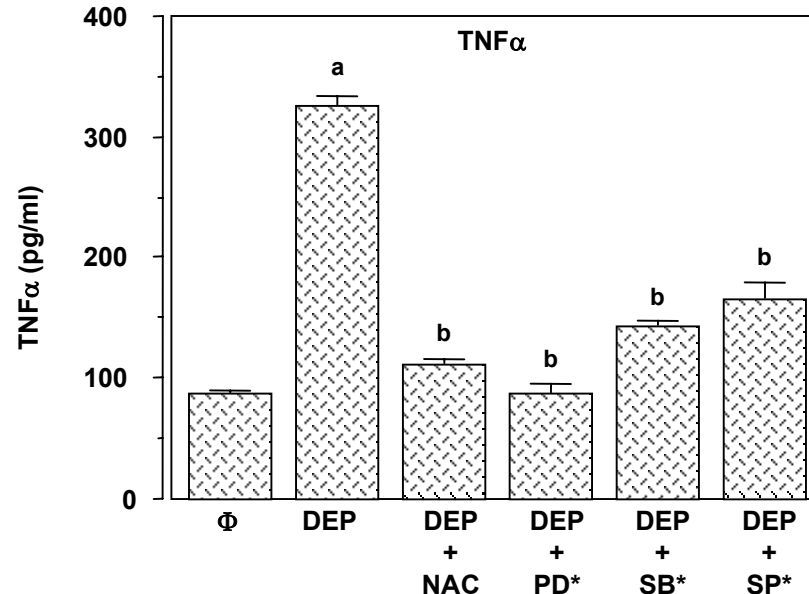
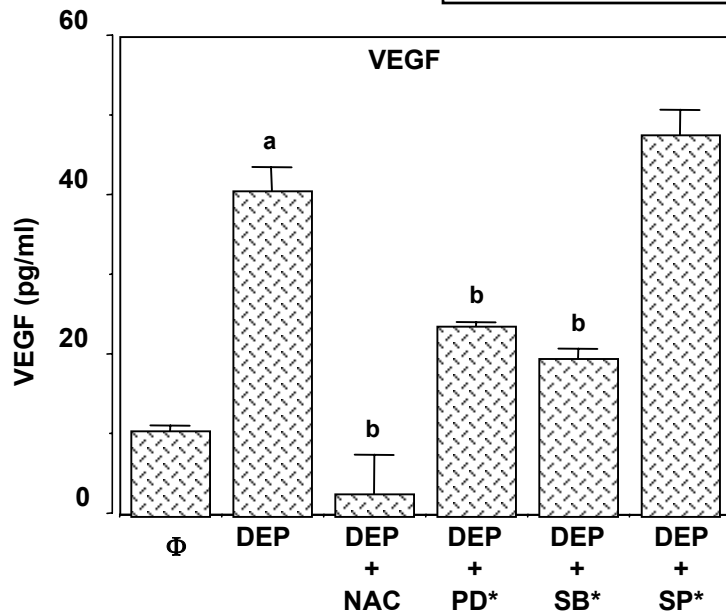
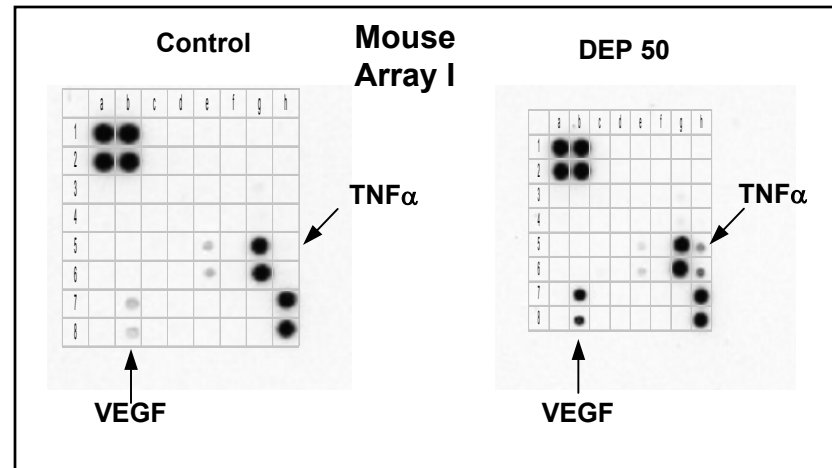


MAP kinase activation at intermediary dose range



Cytokine Array Analysis and ELISA to demonstrate Pro-inflammatory Effects of DEP

RAW 264.7

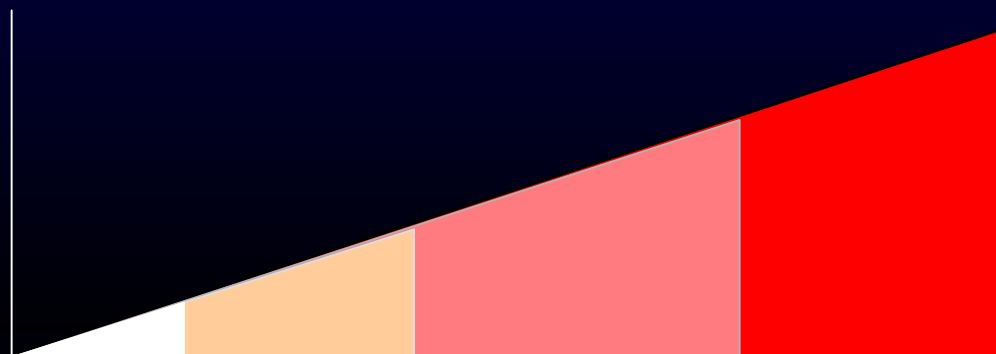


Xiao, et al.

High
GSH/GSSG
Ratio

Low
GSH/GSSG
Ratio

Level of
oxidative stress



Cell response pathway:

Normal

Anti-oxidant
Defense

Inflammation

Toxicity

Signaling pathway:

—

Nrf-2

MAPK

Mitochondrial
perturbation

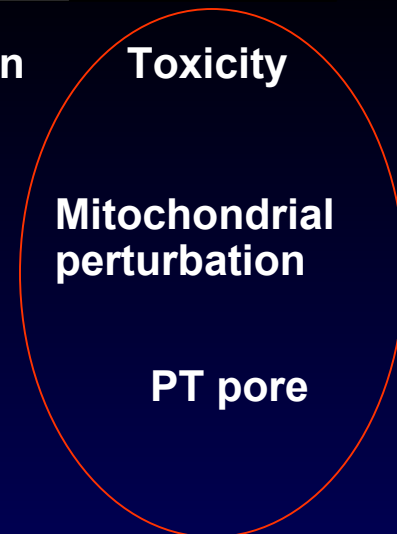
Genetic response:

—

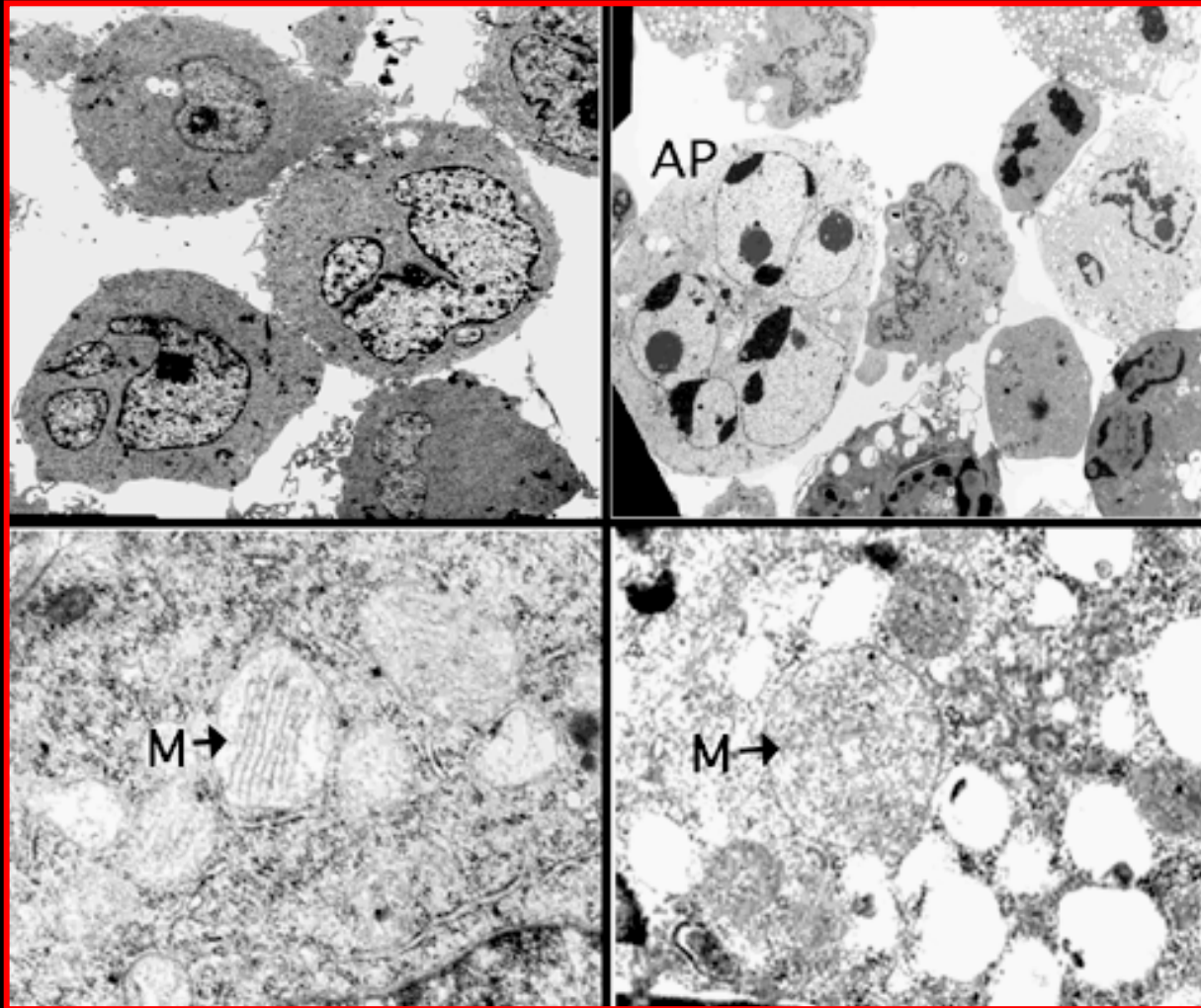
ARE

AP-1

PT pore



Electron micrograph showing that organic DEP chemicals induce apoptosis via mitochondrial effects



Untreated

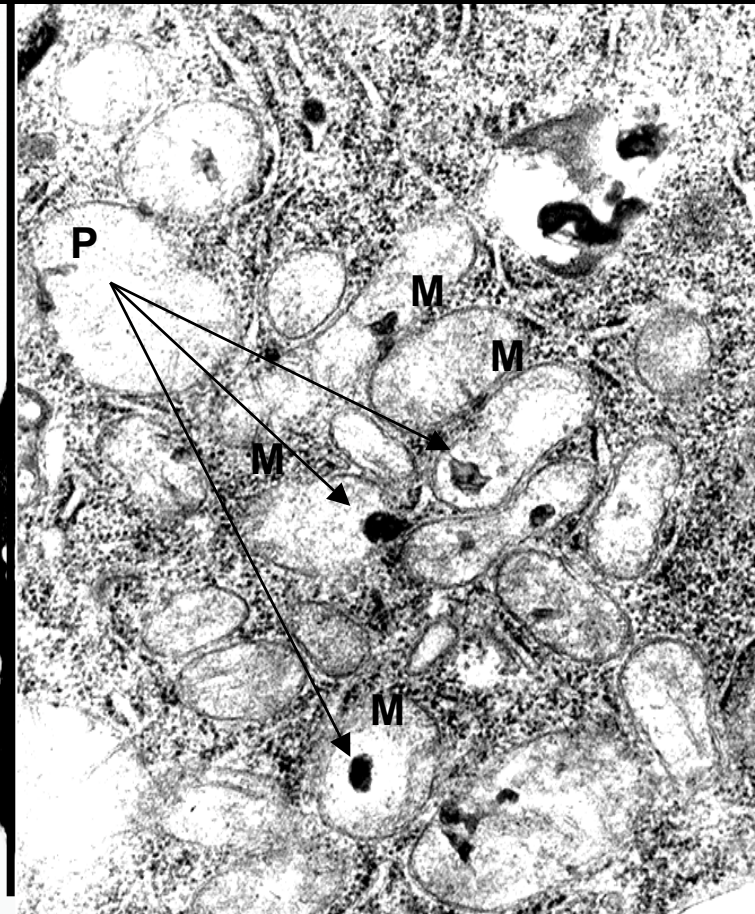
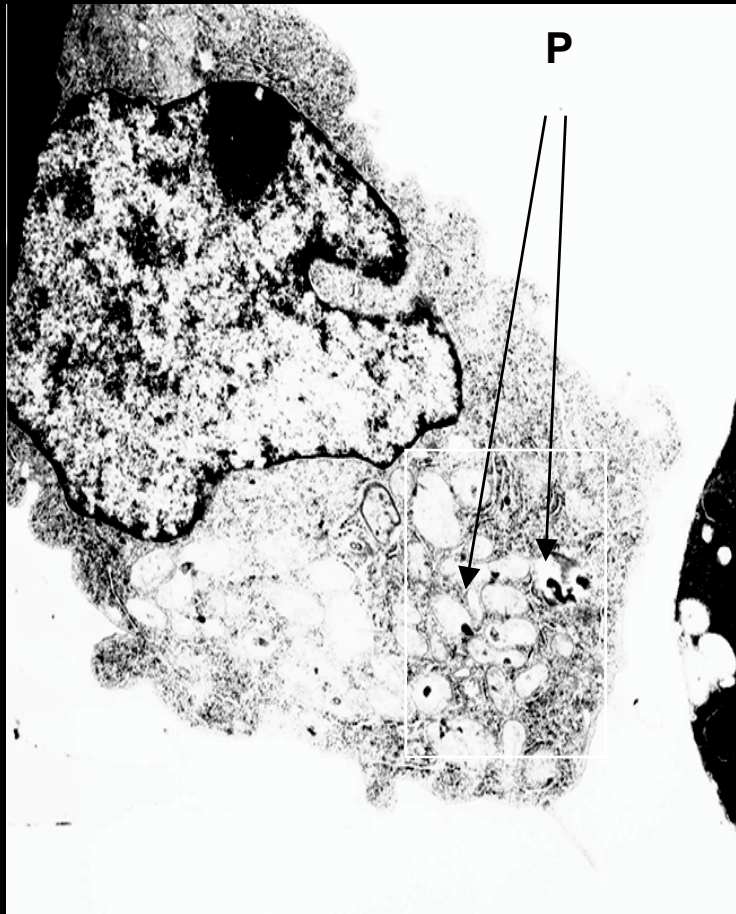
DEP extract

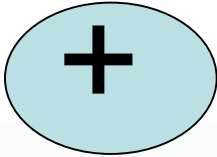
Ultrafines lodge in and destroy mitochondria

Mag. x 6000

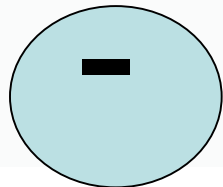
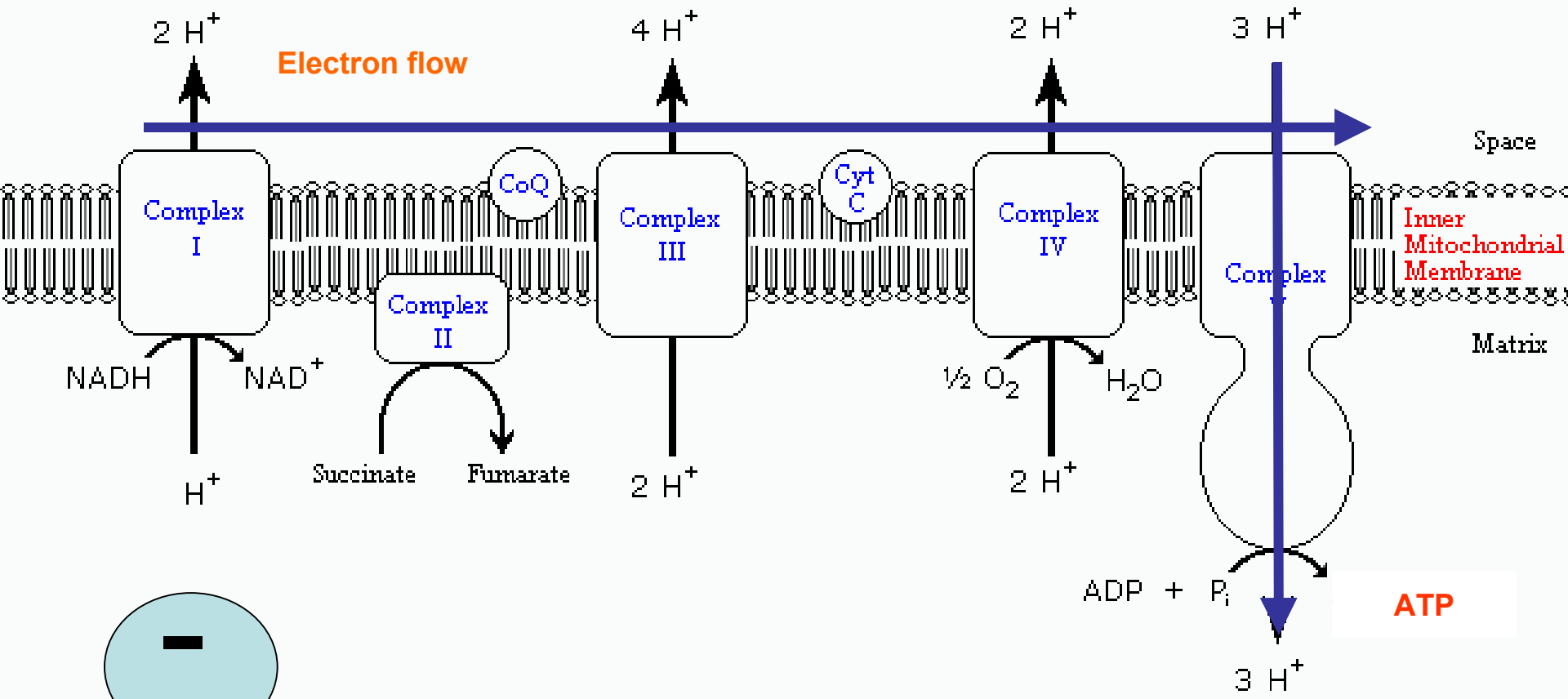
Mag. x 21000

UFP





Mitochondria membrane potential



Differential Toxic Effects by different DEP components

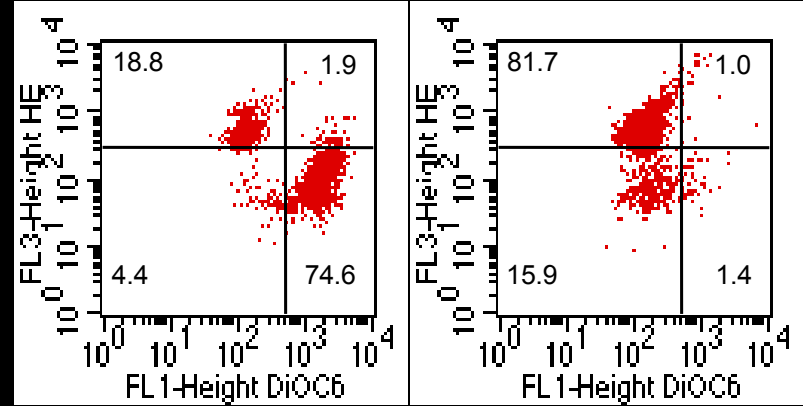
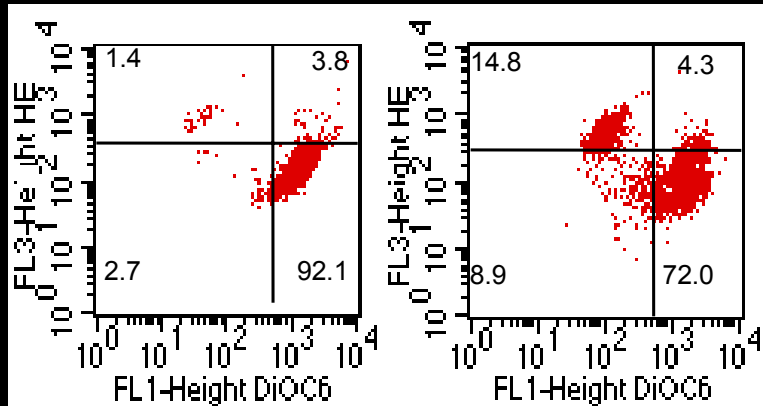
Superoxide radical

Control
(Aliphatic)

DEP

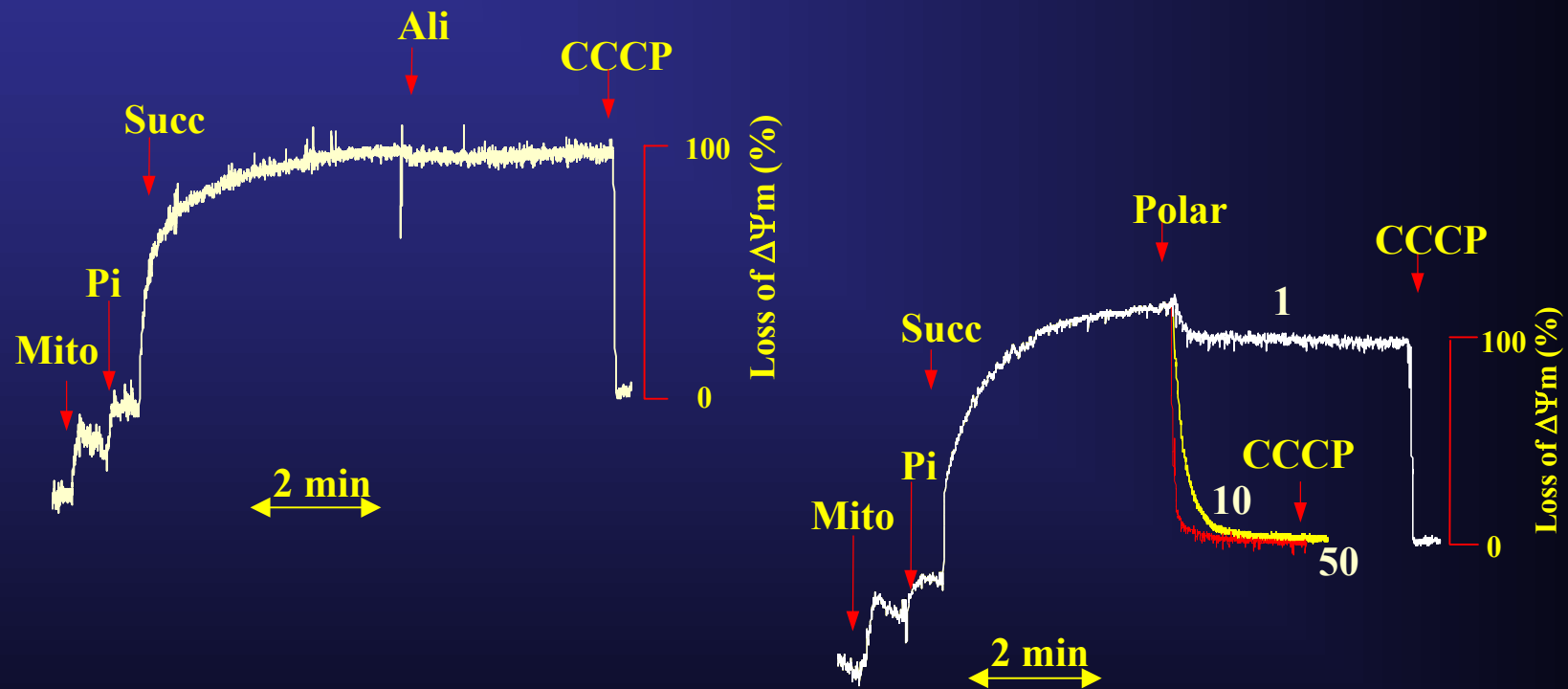
Aromatic
(PAHs)

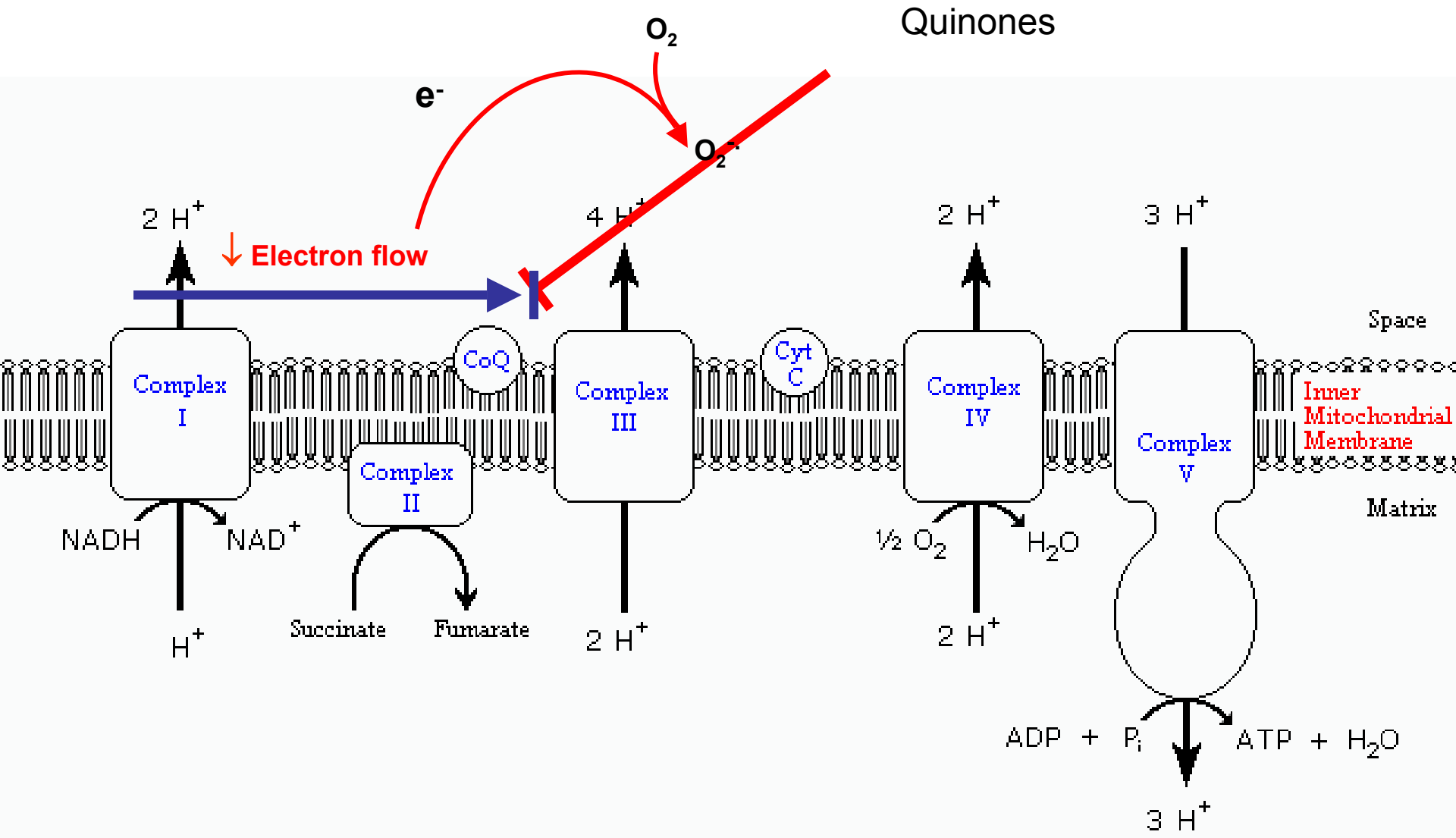
Polar
(Quinones)



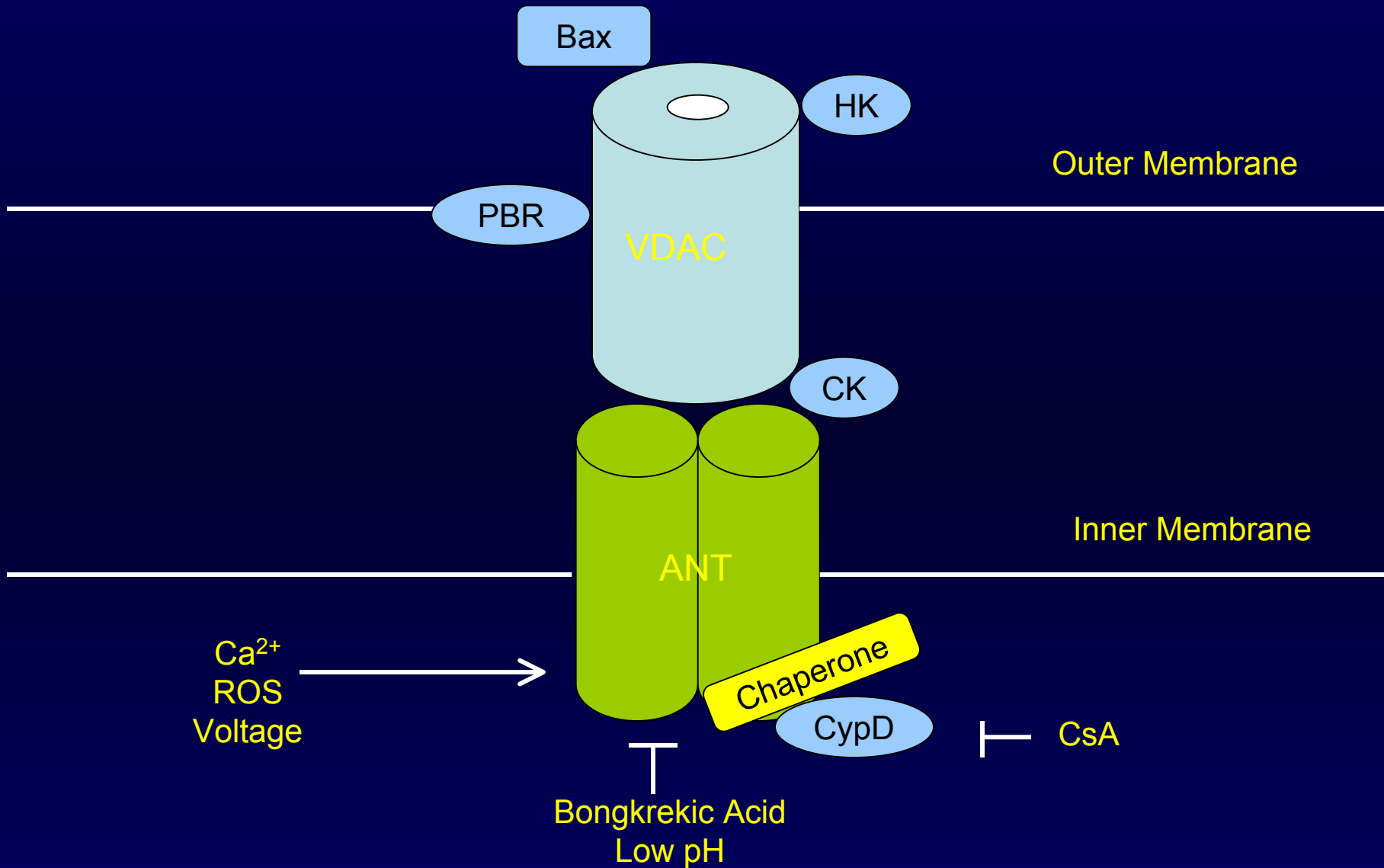
Mitochondrial membrane potential (DiOC6) 

Polar material induces rapid mitochondrial depolarization

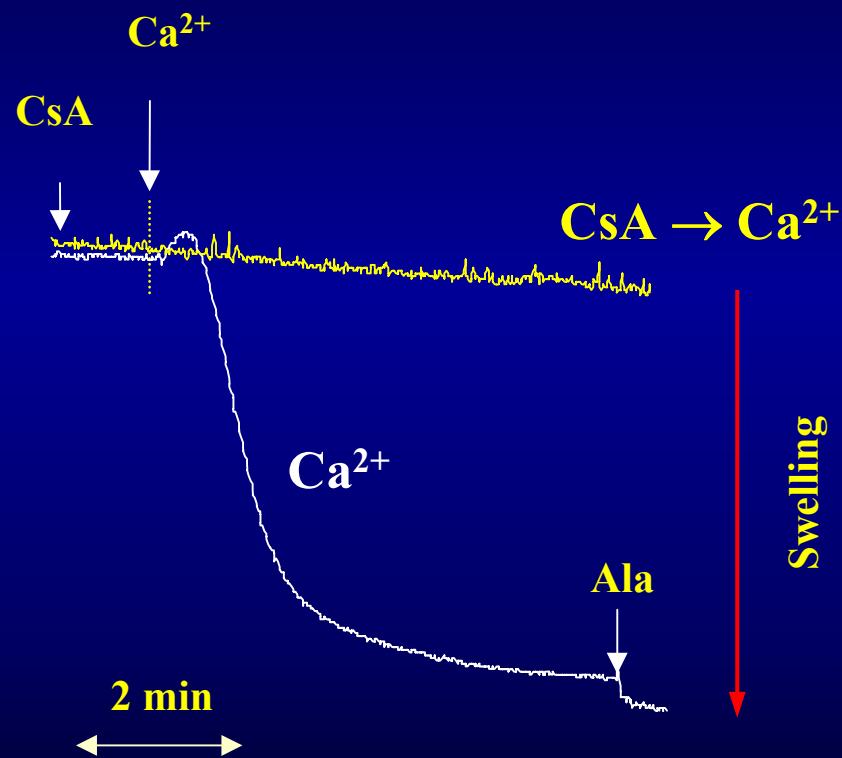




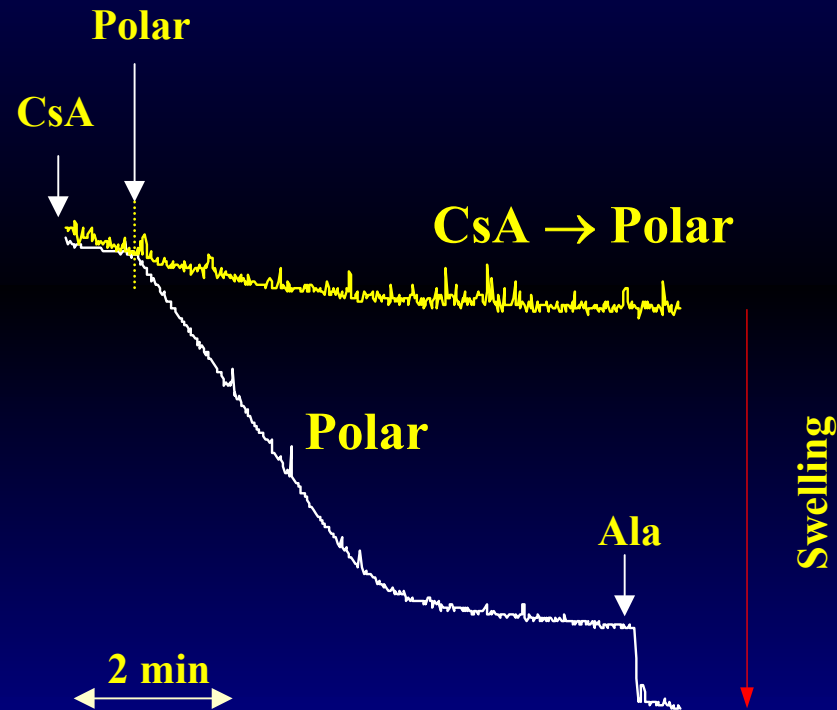
Tian Xia



Calcium dependant PT pore opening is sensitive to the effect of Cyclosporin A

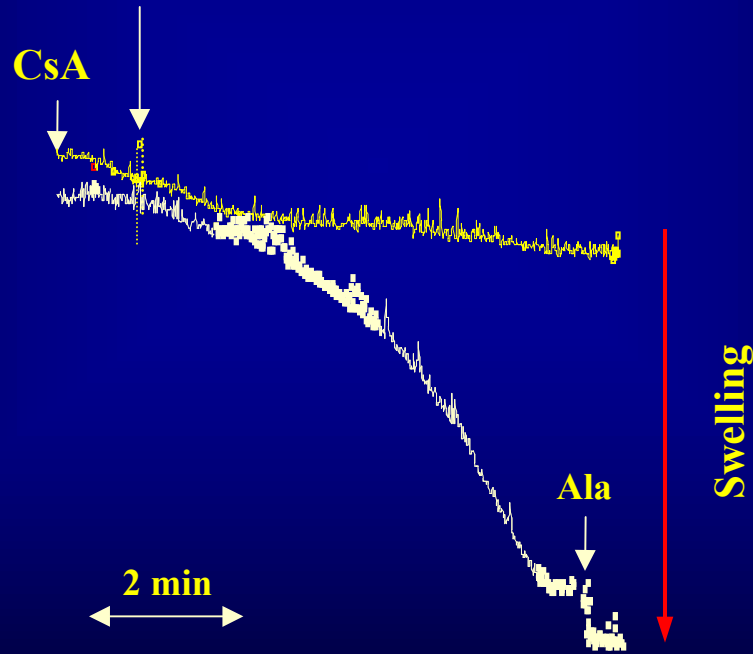


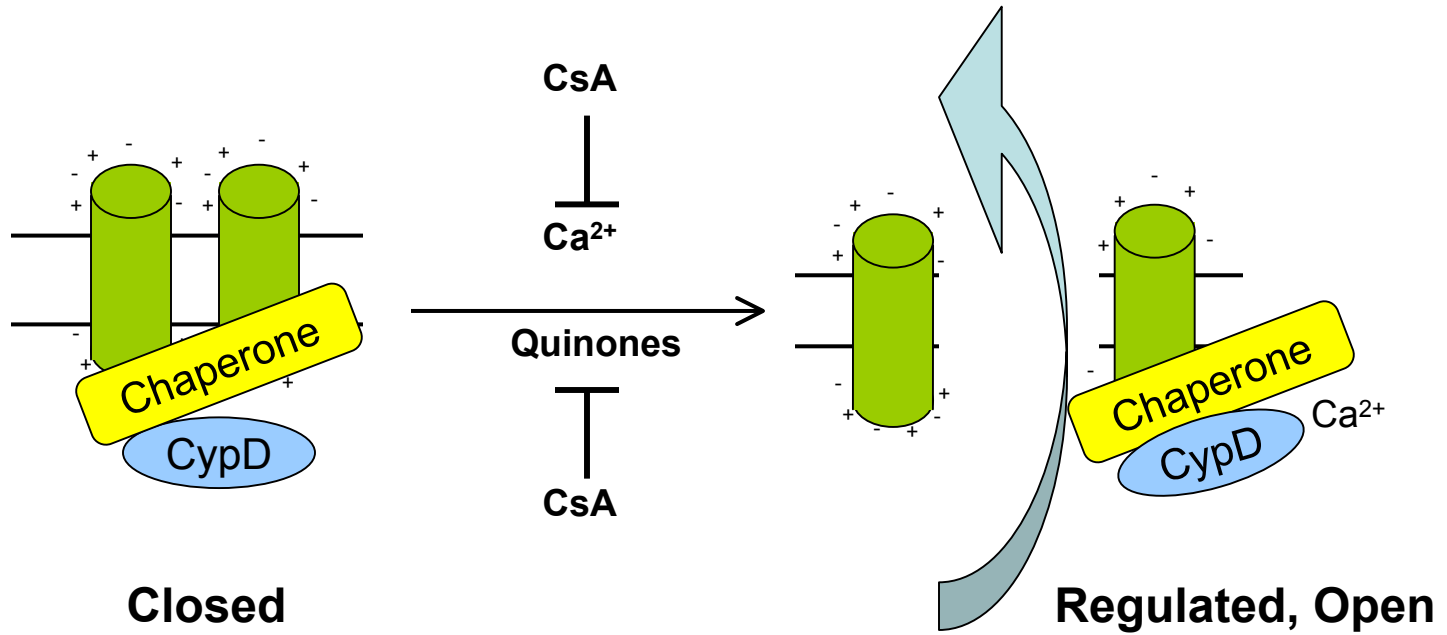
Polar-induced PT transition is Ca^{2+} dependant and CsA inhibitable



PT opening by redox cycling DEP quinones is Ca^{2+} dependant and CsA inhibitable

Phenanthraquinone



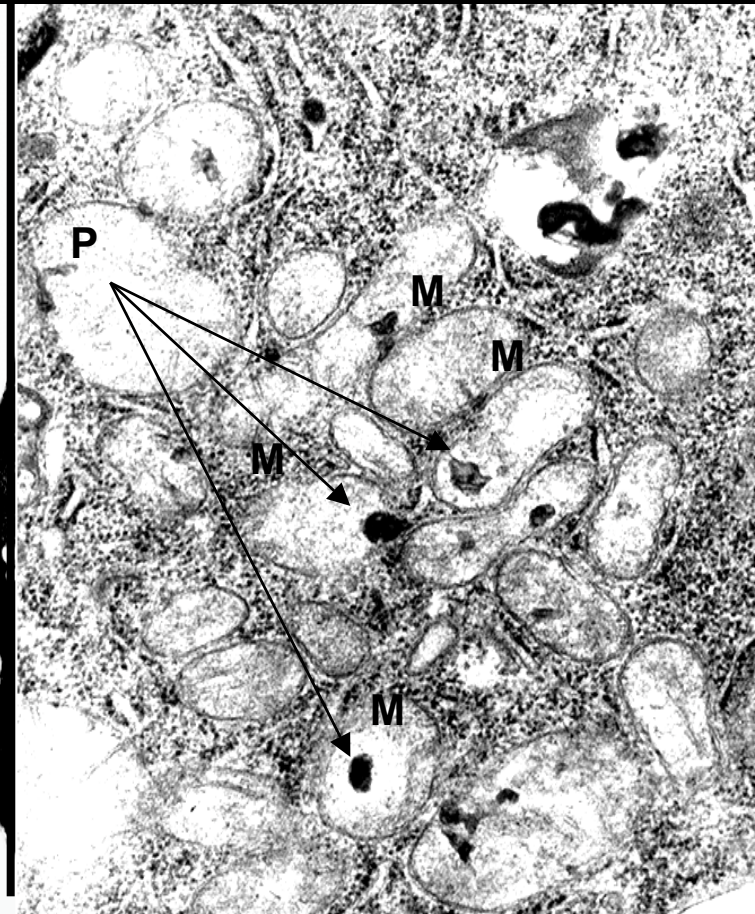
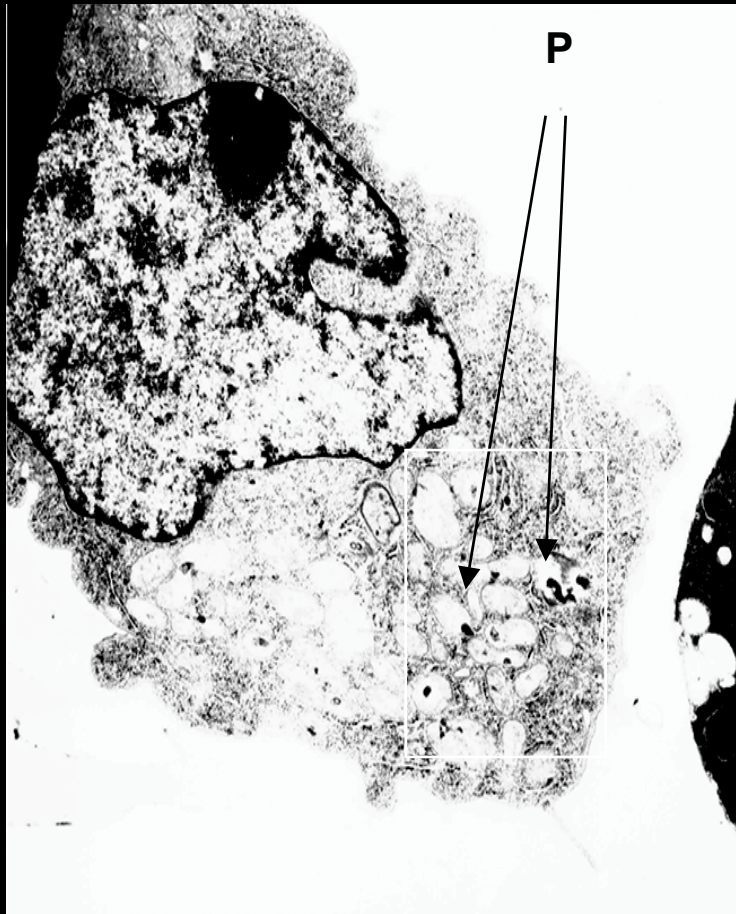


Ultrafines lodge in and destroy mitochondria

Mag. x 6000

Mag. x 21000

UFP



Ambient Ultrafine Particles induce PT pore opening

