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# The biological effects of repeated exposure of Normal Human Bronchial Epithelial (NHBE) cells to the PM<sub>10</sub> fraction of indoor dust



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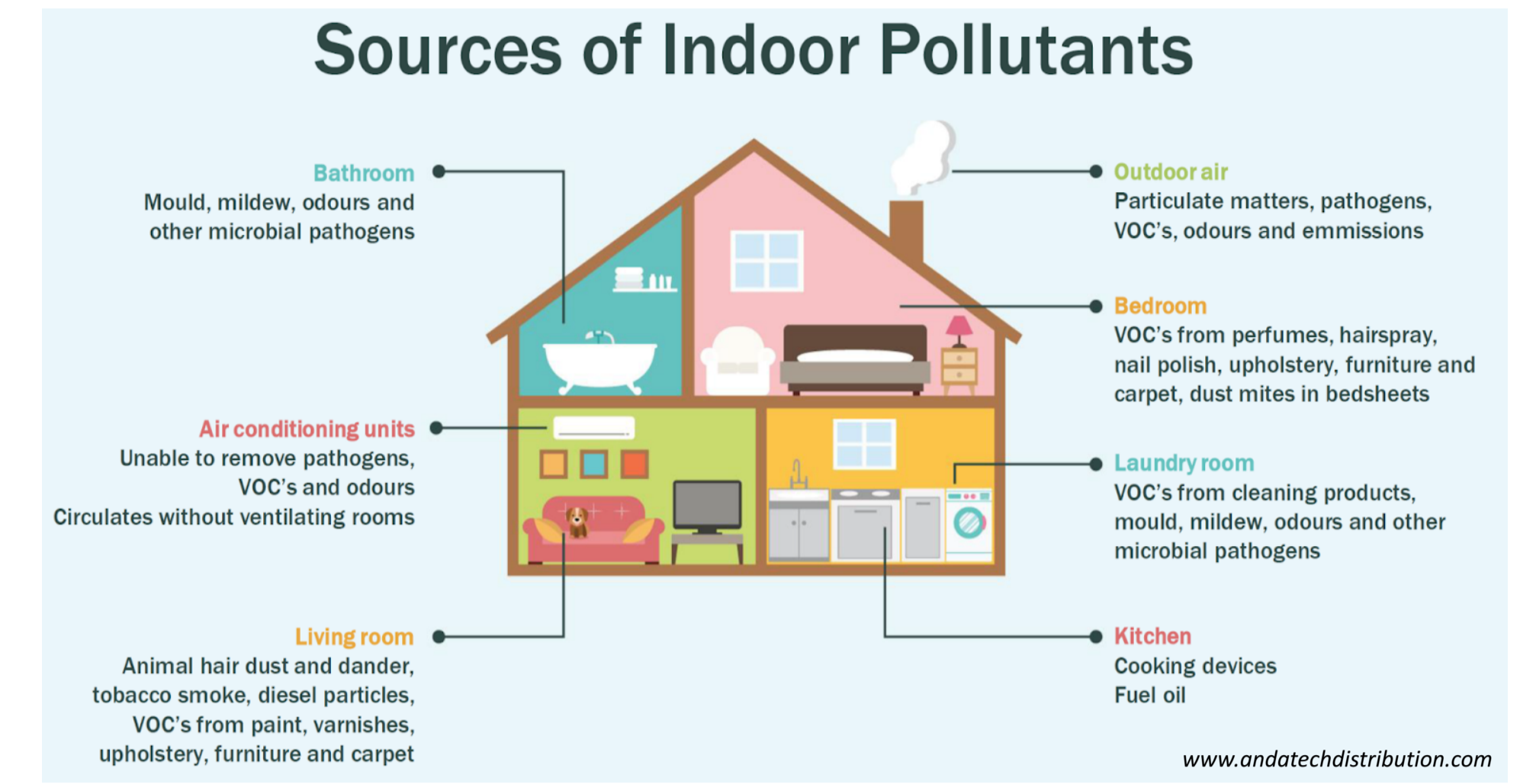
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## INTRODUCTION

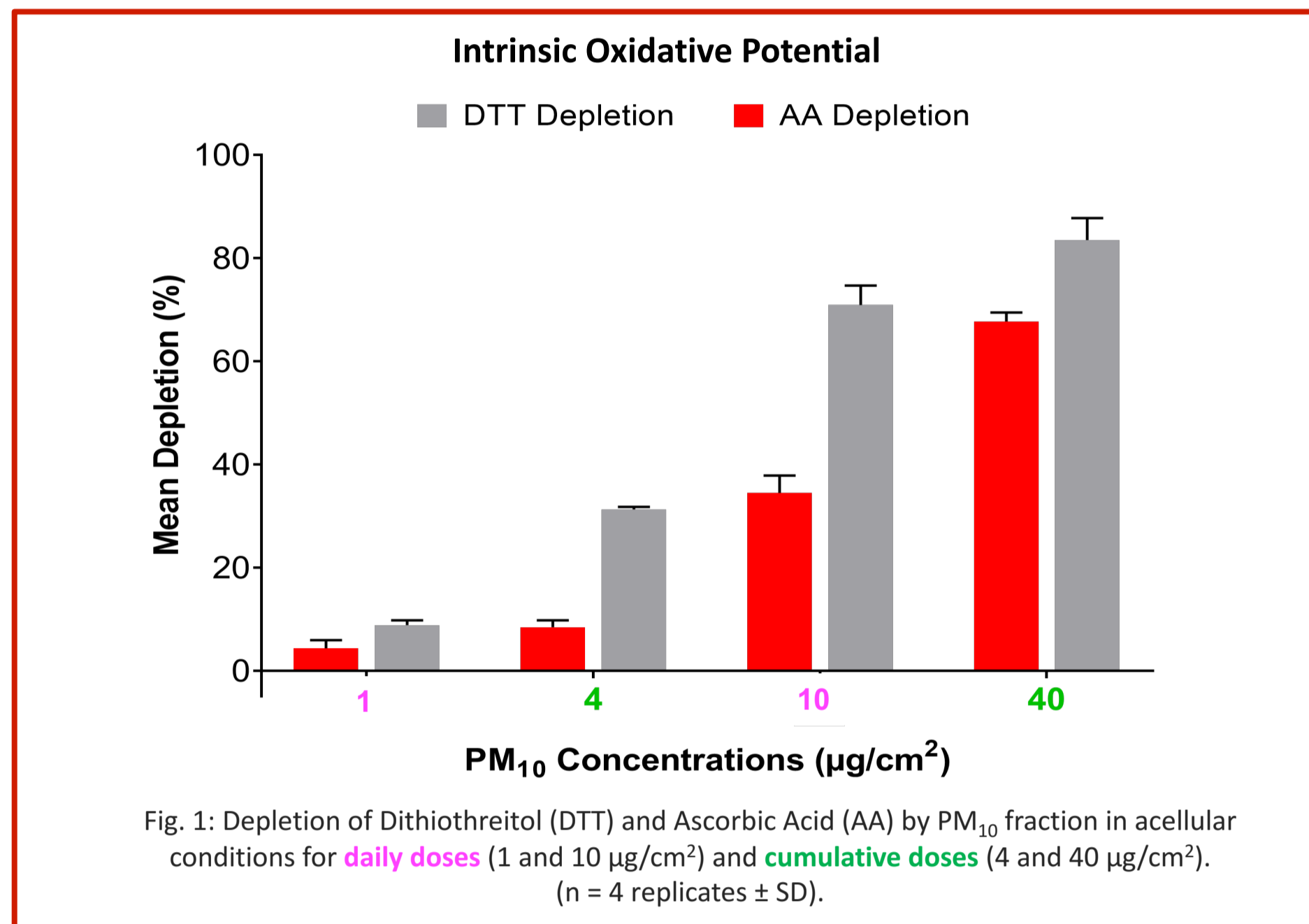
- World Health Organization reported 8 million deaths every year globally related to air pollution. Among these, 4.3 million were attributed to air pollution from house-hold sources and 3.7 million die because of ambient air pollution (1)
- Related health effects have been studied mainly from outdoor pollution but as we spend 90% of our time indoor, the awareness about the impact of indoor air quality has emerged more recently (2)
- The most severe human health effects of air pollution have been attributed to particulate matter (PM) (3)
- The chemical composition of indoor particles is very complex because it is a mixture of different sources and components (4)
- Exposure to indoor PM is associated with adverse health effects related essentially to respiratory and cardiovascular diseases (5)



## AIMS OF THE STUDY

Study the biological markers of cellular and molecular alterations of NHBE differentiated human cells cultured at the Air-Liquid Interface (ALI) after repeated exposure to aerosolized indoor PM<sub>10</sub> issued from standard reference material "Organic Contaminants in Dust House" (SRM® 2585, NIST®) in order to mimic realistic exposure conditions *in vitro*

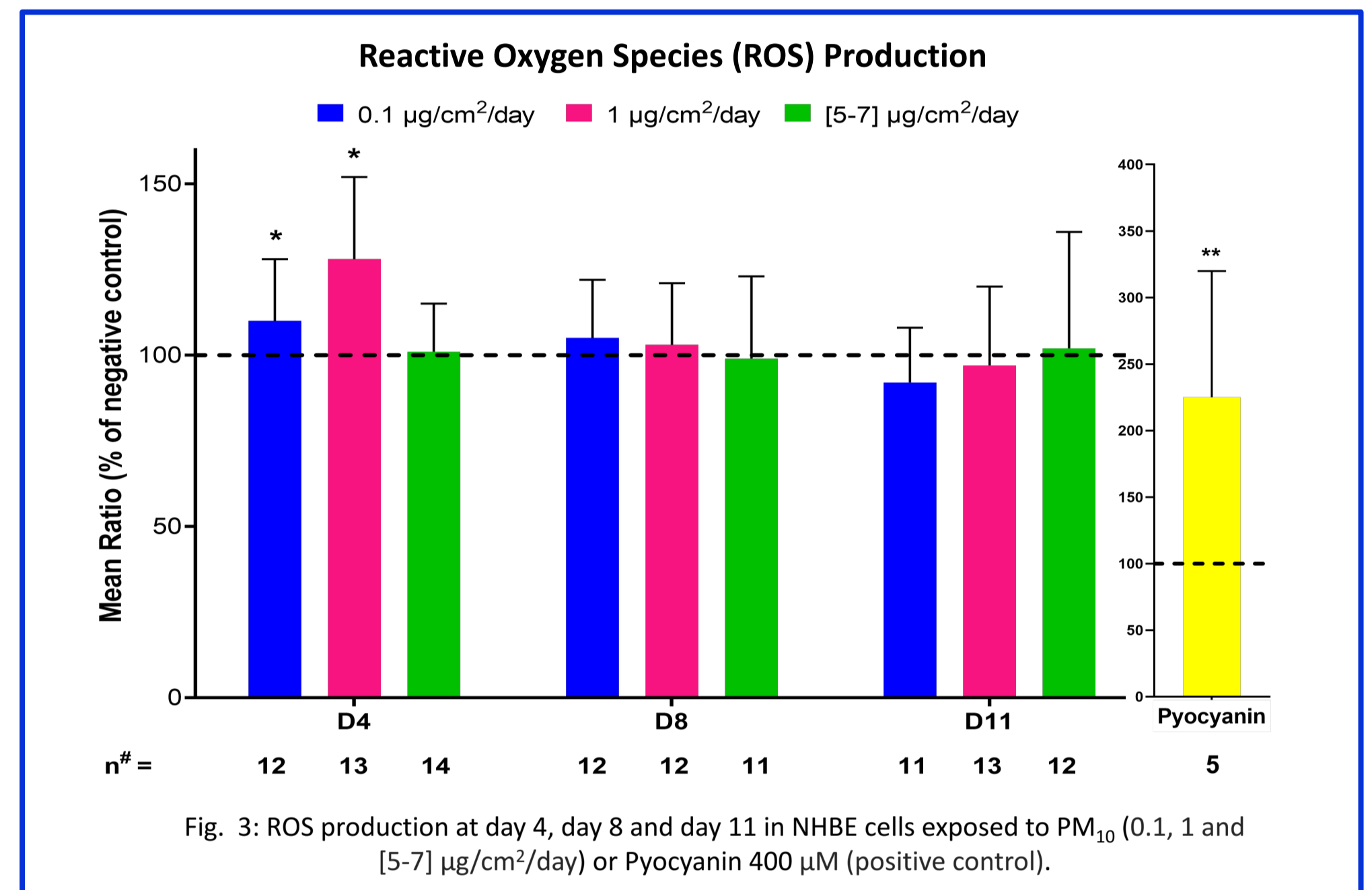
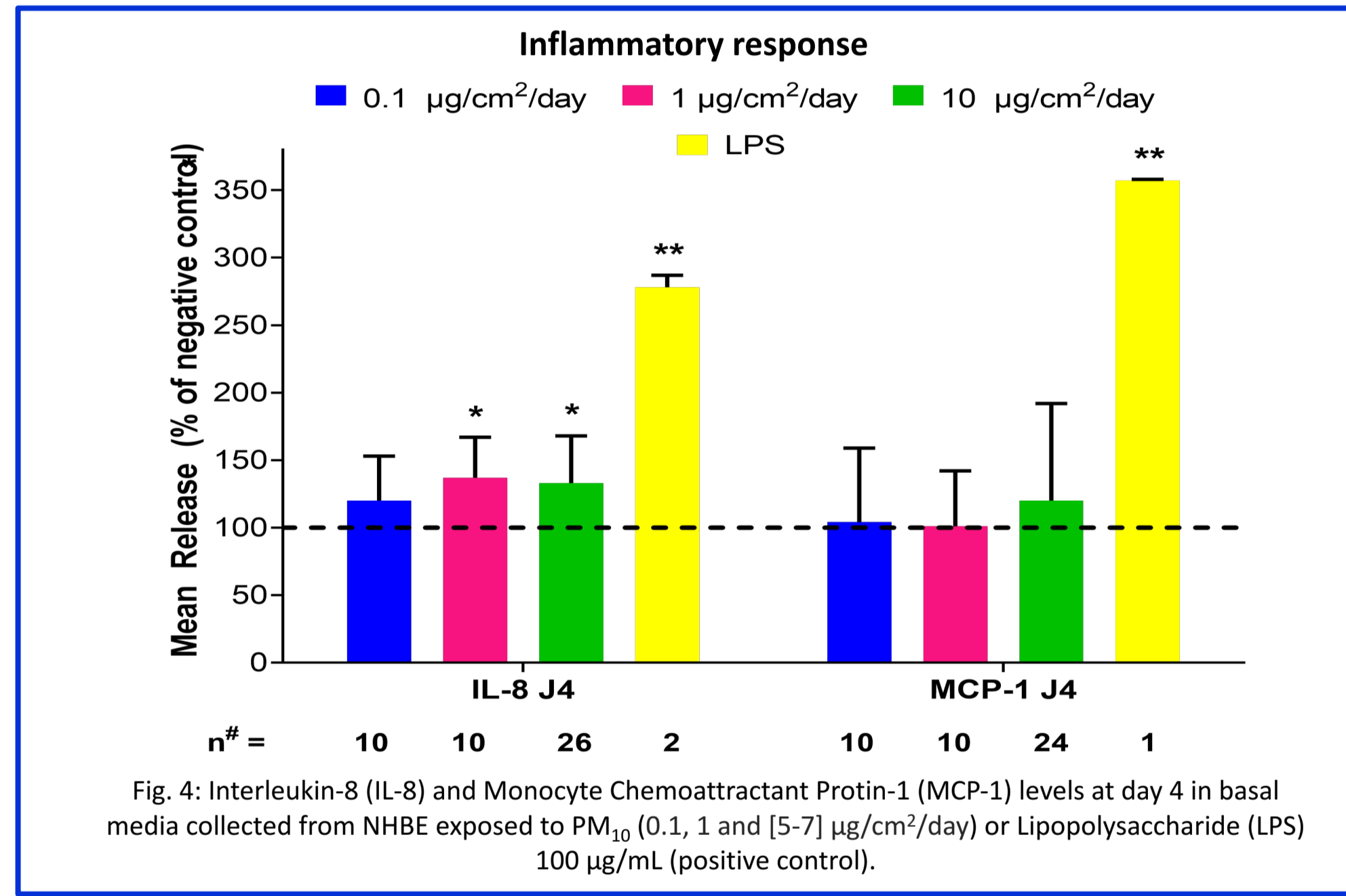
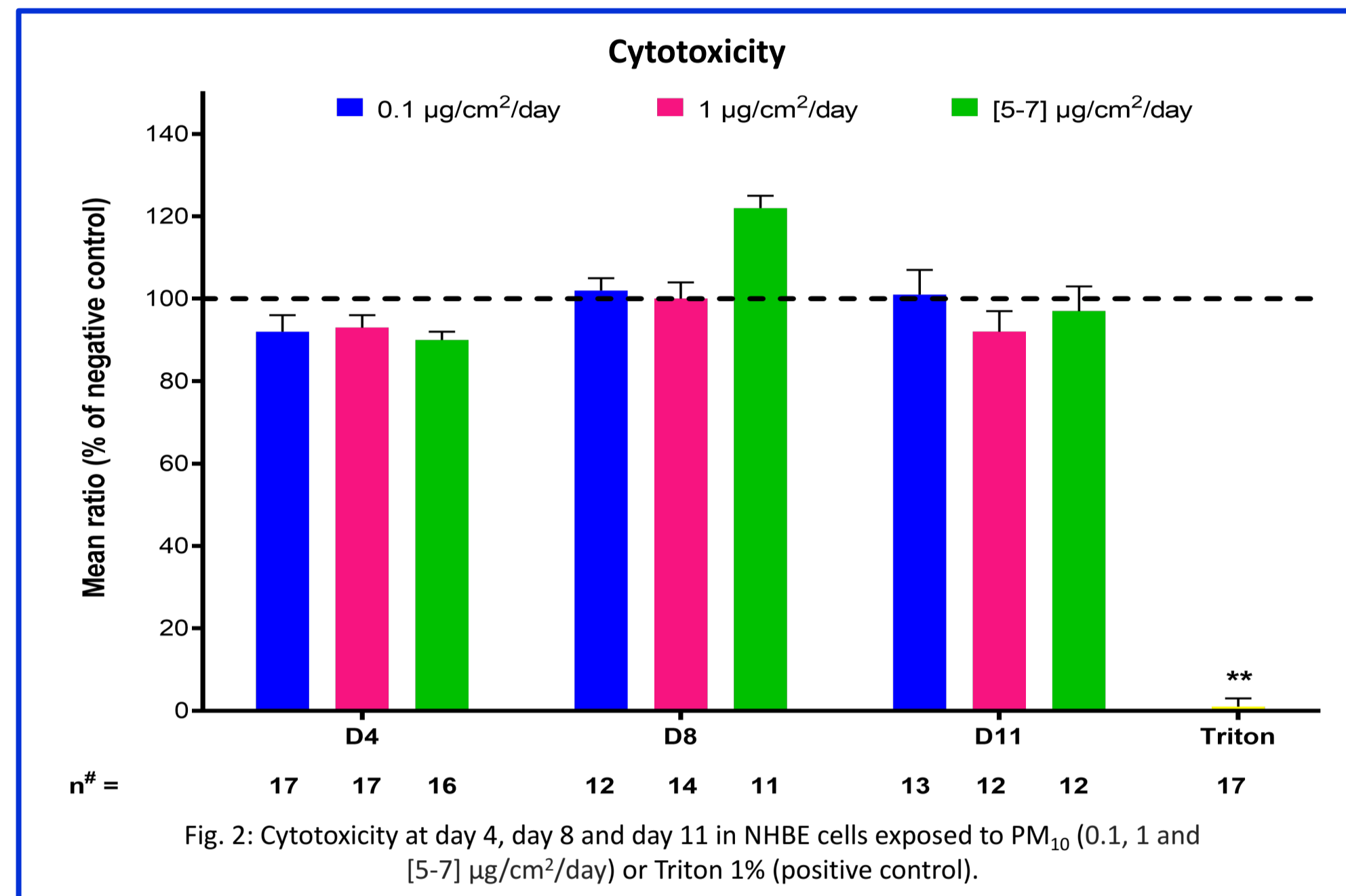
## RESULTS



- Fig 1: Depletion of antioxidant substrates increased in a dose-dependent manner for both daily and cumulative doses, suggesting an intrinsic oxidative potential

**Intrinsic Inflammatory Potential**  
 PM<sub>10</sub> endotoxin level : 2660 ng/g of PM<sub>10</sub>

- Endotoxins are issued from gram – bacteria and could contribute to pro-inflammatory process. Endotoxins are present in SRM® 2585 PM<sub>10</sub> whereas no viable fungal nor bacterial flora was detected



Statistical significance vs. results for negative control condition was performed using Wilcoxon non-parametric test: \*p < 0.05, \*\*p < 0.01.  
 #: number of replicates

- Fig. 2: No significant cytotoxicity was observed whatever the conditions tested in accordance with homogenous TEER measurements (data not shown)

- Fig. 3: A transient and significant increase of ROS production was observed only at day 4 for the two lowest doses

- Fig. 4: A weak but significant increase in IL-8 production was observed at day 4 only for the two highest doses

## CONCLUSIONS & PERSPECTIVES

- Limited biological responses were detected despite the chemical content, the intrinsic capacity of these PM<sub>10</sub> to generate ROS and the presence of endotoxin. The differentiated NHBE cell model appears thus resistant to the toxic effects of PM<sub>10</sub> fraction isolated from SRM® 2585 in our realistic exposure conditions
  - To complete and explain this behaviour, expression of a panel of gene involved in oxidative stress and genotoxicity is currently being analyzed
  - Moreover, since the mucus produced by bronchial cells has antioxidant properties that could potentially limit the ROS effects, bronchial mucins will be further quantified

### Perspective

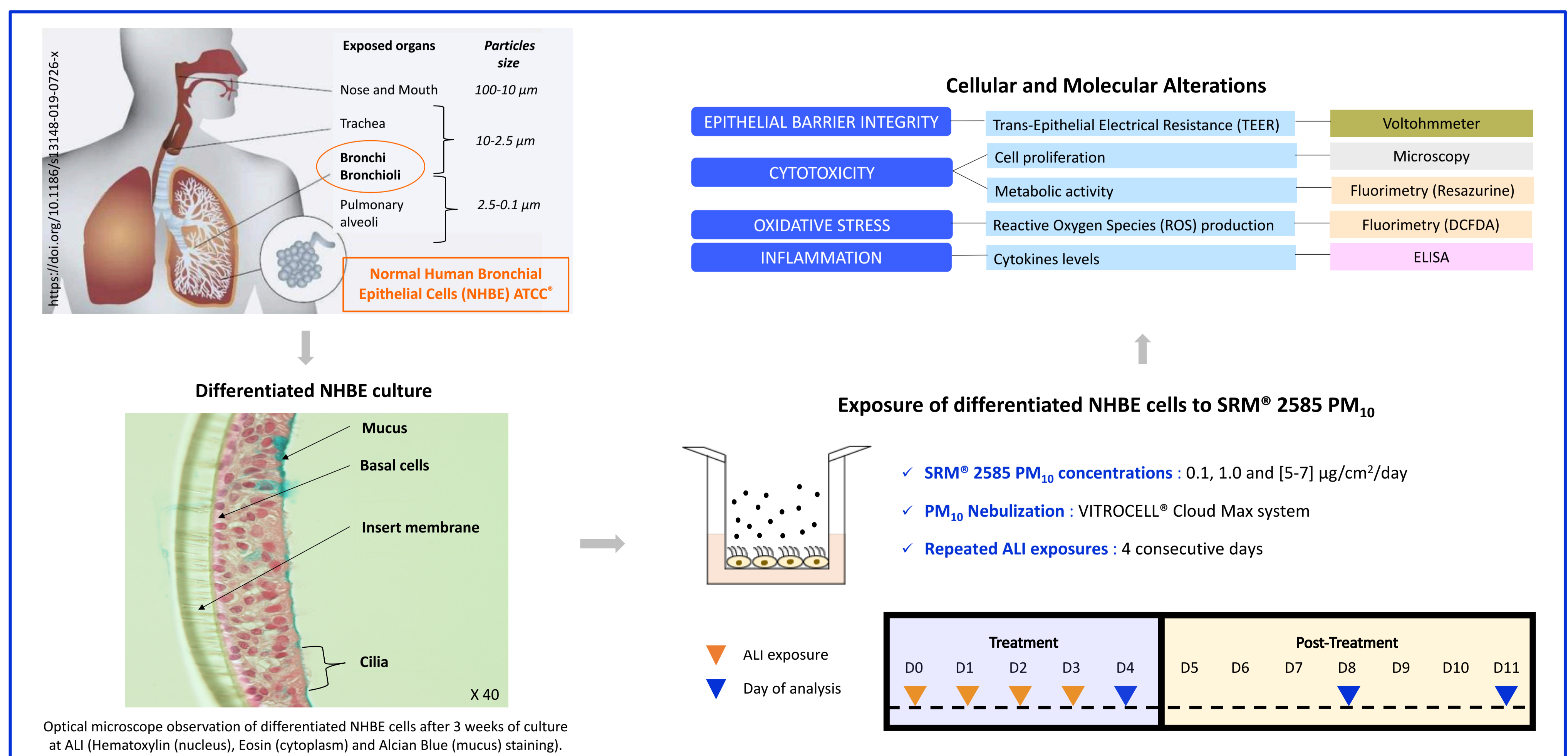
- The NHBE cells will be co-exposed to SRM® 2585 PM<sub>10</sub> and supernatants collected from human macrophages exposed to SRM® 2585 PM<sub>2.5</sub> (Poster n° 1185, "Impact of repeated exposure to indoor dust on human macrophage polarization profile"). The possible effect on inflammatory response will be thus studied in NHBE cells

## MATERIALS & METHODS

| Chemical Family                                | Total number of substances |
|--|----------------------------|
| Polycyclic Aromatic Hydrocarbons (PAHs)        | 66                         |
| Polychlorinated biphenyl (PCB) congeners       | 42                         |
| Chlorinated pesticides                         | 14                         |
| Polybrominated diphenyl ether (PBDE) congeners | 18                         |
| Phosphorous flame retardants                   | 5                          |
| Perfluorinated alkyl acids (PFAAs)             | 8                          |
| Synthetic musks                                | 5                          |

"Organic Contaminants in Dust House" (SRM® 2585) → Size separation → PM<sub>10</sub> fraction

| SRM® 2585 PROPERTIES                     | Assay                          |
|--|--------------------------------|
| Endotoxin levels                         | Limulus Amebocyte Lysate Assay |
| Pro-oxidative intrinsic potential        | Antioxidant Depletion          |
| Microbial content (fungal and bacterial) | Culture on MEA medium          |



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