

VERT– FOCUS EVENT

Empa Dübendorf, March 16, 2018

Effects of combustion and ambient aerosols on normal and diseased airway epithelia

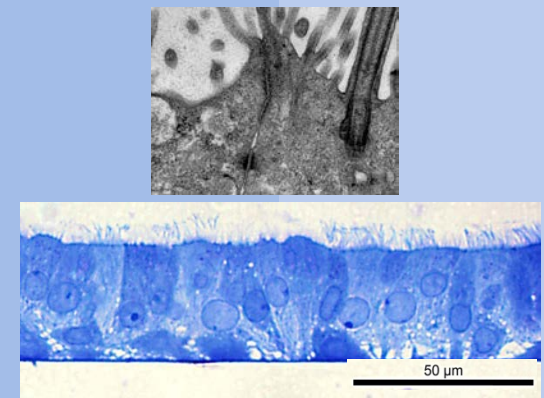
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Sources of anthropogenic
(nano)particles



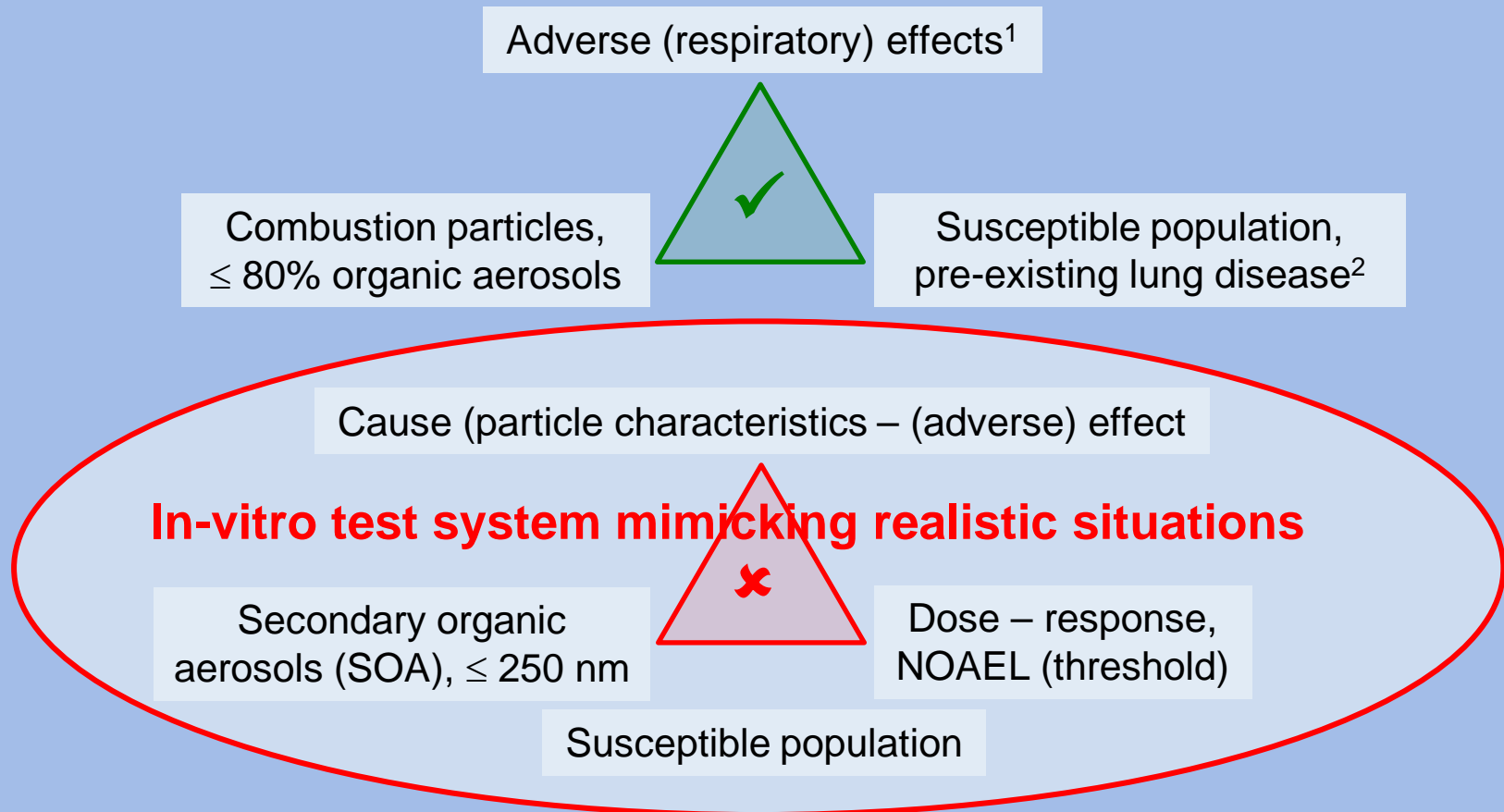
NACIVT – Nano Aerosol Chamber
for In-Vitro Toxicity, www.nacivt.ch



Re-differentiated human airway epithelia

PM_{2.5} – adverse health effects: 20 yrs. evidence – lacking knowledge today

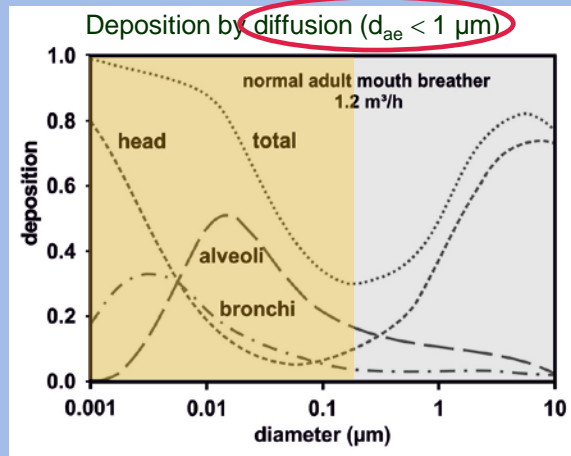
> Inhaled fine particulate matter (PM_{2.5})



¹Schwartz J et al. *JAWMA*, 1996; ²WHO, 2014

Aerosol inhalation – deposition – clearance

> Particle size, regional distribution in RT, deposition efficiency



> Lung compartments, primary target tissue, defense, disease

- Liquid lining layer

*surfactant film & aqueous phase

- Epithelium

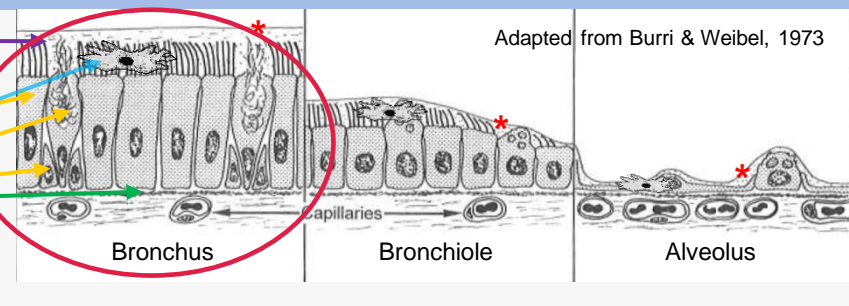
Ciliated cells

Secretory cells

Basal cells

- Basal lamina

- Macrophages



In-vitro model – representative for a part of an organ or tissue

> Requirements

- ➔ Well characterized aerosol
- ➔ Realistic deposition **Mechanical unit - NACIVT** on lung cell cultures
- ➔ Cell **Biological unit – Human Bronchial Epithelia** (surface)
- ➔ Meaningful parameters to characterize the biological response

Nano Aerosol Chamber for In-Vitro Toxicity (NACIVT), <http://www.nacivt.ch>

Aerosol (external production/source)

Aerosol conditioning (T; RH)

Aerosol distribution to 24 delivery tubes
Particle deposition on 24 cell cultures

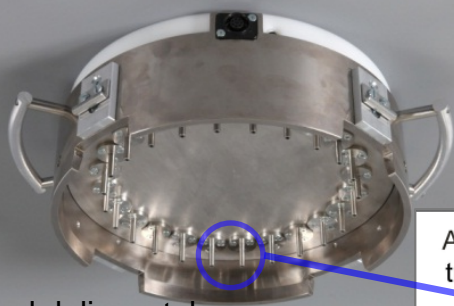
Custom electronics (aerosol
charger, pumps, flow control)

Online quantification of deposited particles
Chamber control & data logging

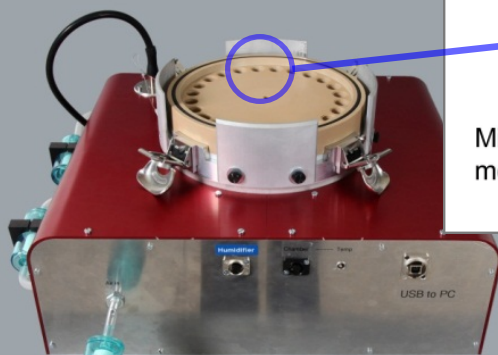
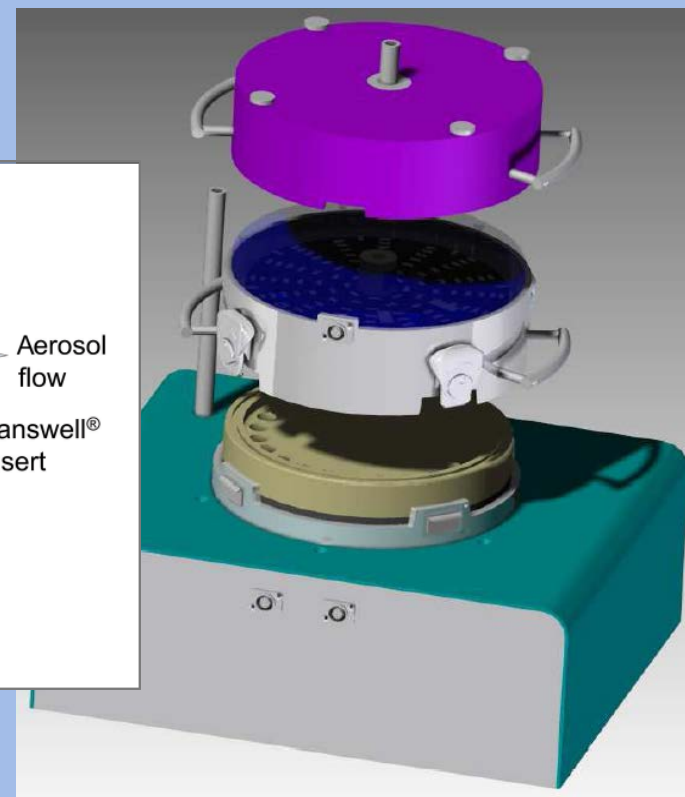
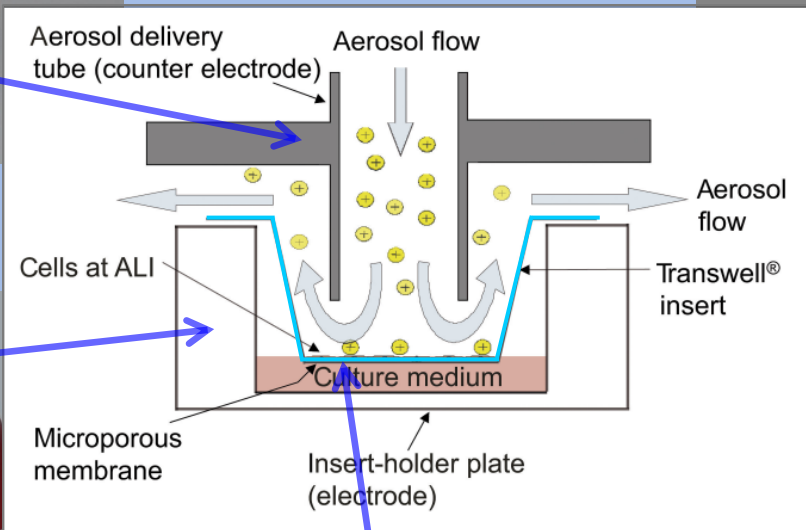
54×49×48 cm (wdh), 30.5 kg

- “All-in-one”, mobile system for direct use at any particle source
- Mimics particle deposition in lungs (T, RH, gas, air flow, N_P , N_{Dep})
- Simultaneous exposure of 24 cell cultures
- Controlled & stable conditions allowing long-term exposures

NACIVT – particle delivery and deposition



Aerosol delivery tubes
(d inner/outer = 0.9/1.4 mm)



Holder plate for ALI cell cultures on
microporous Transwell® inserts



Transwell® inserts

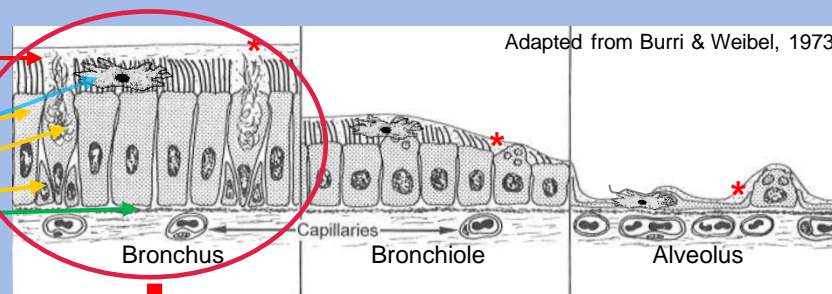
Re-differentiated Human Bronchial Epithelia (HBE)

> Liquid lining layer
*surfactant film & aqueous phase

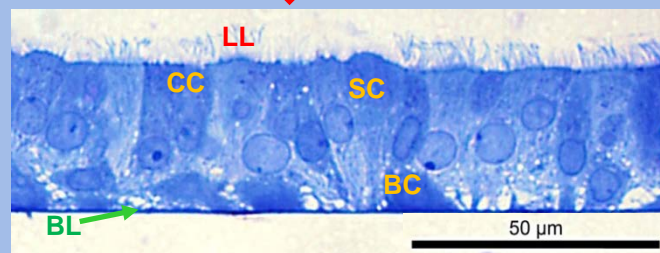
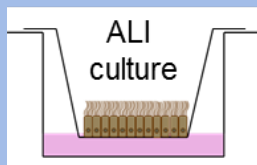
> Epithelium → Ciliated cells
Secretory cells

> Basal lamina → Basal cells

> Macrophages



> Air-liquid interface culture



Junctional complexes

Human bronchial epithelia (HBE)

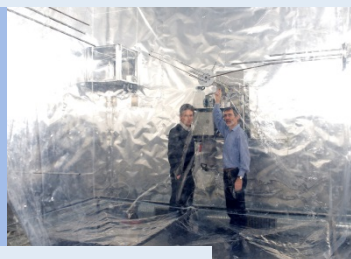
- > All differentiated cell types, basal lamina, junctional complexes
- > Permanent air-liquid interface (ALI)
- > Innate defense, repair, long life span (up to 1 year)
- > Normal and diseased (asthma, COPD/smokers, cystic fibrosis) HBE

Some examples of studies with combustion aerosols

- > Specific sources, POA & SOA – combustion of fossil fuels & wood

Setup for experiments with combustion aerosols – cars and stoves

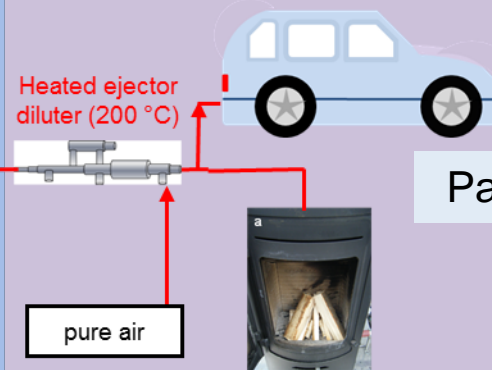
Atmospheric aging



Smog chamber



Emission injection system

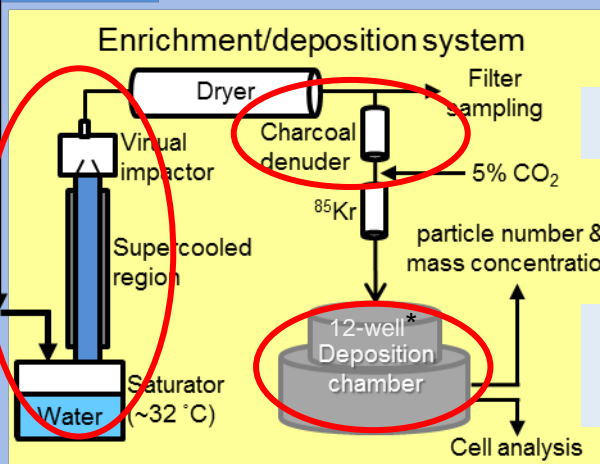


Particle source

Online measurements

- Number & mass conc.
- Mobility & aerodynamic diameter
- Non-refractory chemical composition
- Black carbon
- Nitrogen dioxide
- Total hydrocarbon
- Relative humidity
- Temperature

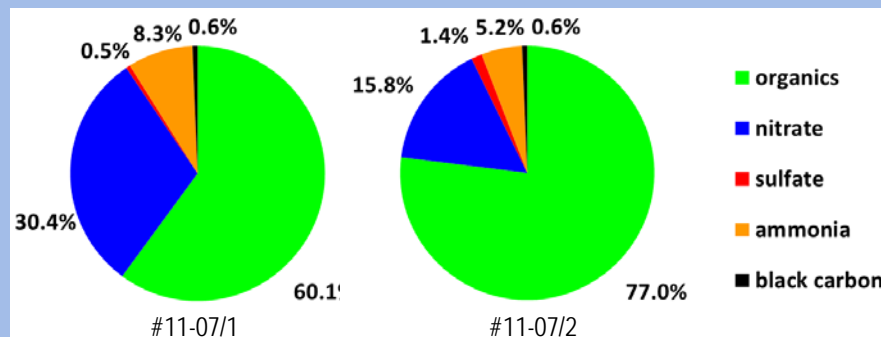
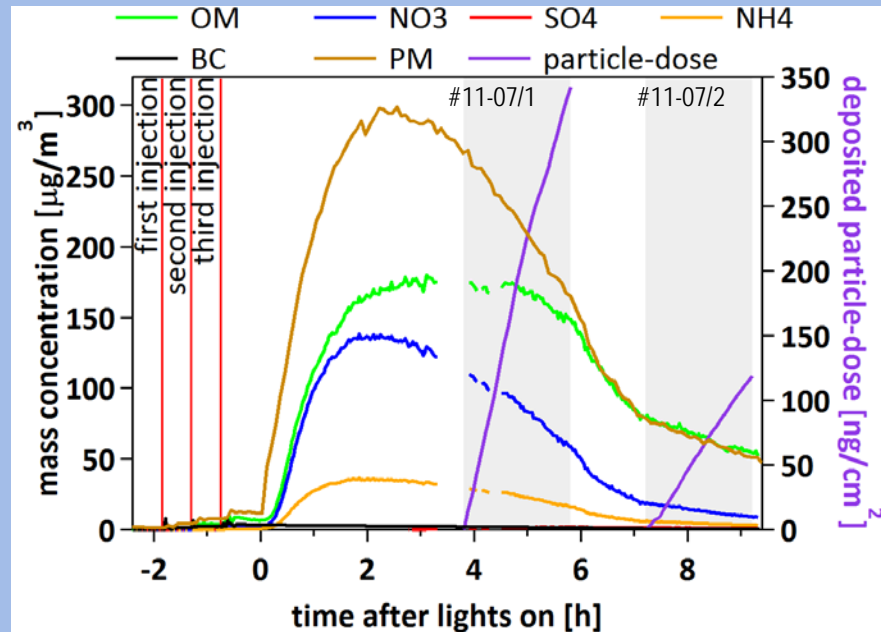
Aerosol conc. adjustment (VACES)



Removal of gases

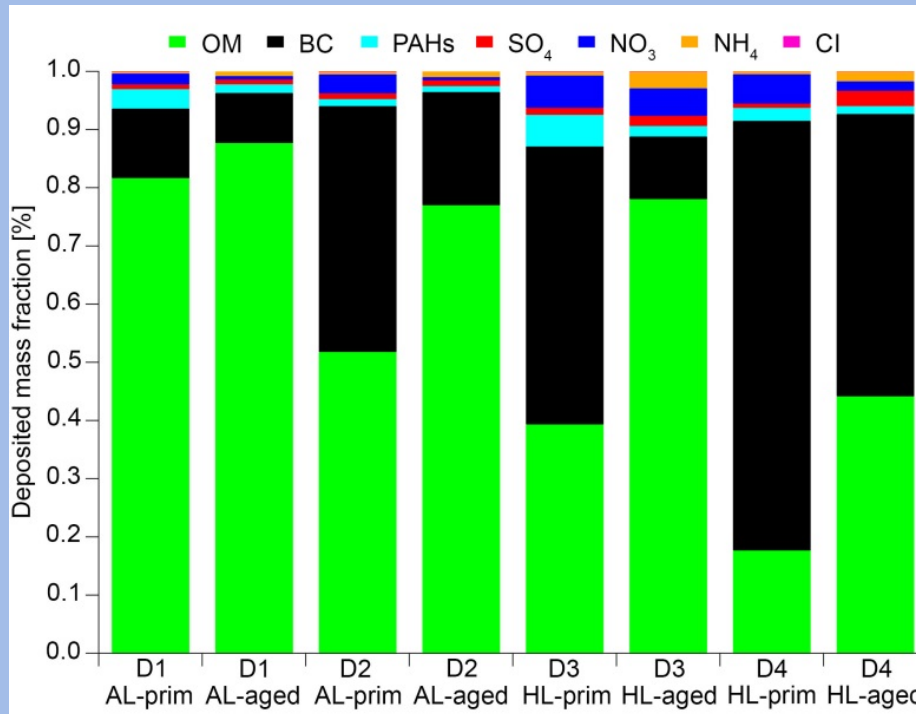
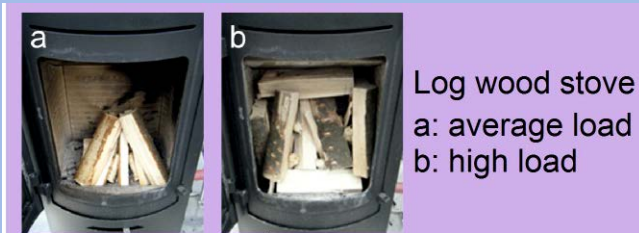
Particle deposition on cell cultures at ALI

Evolution of smog chamber experiment & chemical composition of aerosol (Euro 5, gasoline exhaust)



Chemical composition of wood combustion particles

Deposited dose: $199 \pm 58 \text{ ng/cm}^2$; equiv. to daily TB dose at ambient $400\text{--}1000 \text{ }\mu\text{g/m}^3 \text{ PM}$



- Primary OM: dominant fraction at AL
- eBC: most abundant at HL
- Higher OM:BC ratios in aged particles due to secondary OM formation

Gasoline, diesel & wood particles – acute (24h) responses to single, short-term (1-2h) exposure at realistic particle doses (TB 24h, PM <20-1000µg/m³)

Findings

- > Cytotoxicity ↑ with dose in normal & vulnerable HBE (G/SOA; W/POA,SOA)
- > Cytokine release ↓ (G/SOA;D/POA,SOA)
- > Oxidative stress ↑ (W/POA,SOA)
- > Adverse effects at lowest dose (G/SOA;D/POA,SOA)
- > Differences between normal & diseased HBE (G/SOA;W/POA,SOA)
- > Cause-effect: highest correlation with all particle fractions* (W/POA,SOA)
- > Effects of POA ~ SOA (D,W/POA,SOA)
- > Differences between simplistic, single-cell type cell lines and fully differentiated HBE (G/SOA; D, W/POA,SOA)

Consequences

- Impairment of epithelial key-defense mechanisms, rendering the epithelium more vulnerable to subsequent hazards
- No evidence for threshold (NOAEL)
- Confirms susceptibility of impaired epithelia
- Effects might be attributable to a combination of particle characteristics
- Higher SOA-toxicity not confirmed
- Use of HBE to be most appropriate in future in-vitro toxicity studies

Acknowledgements



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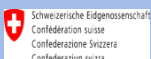
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