

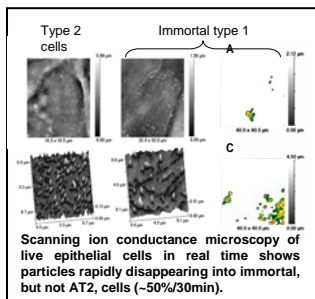
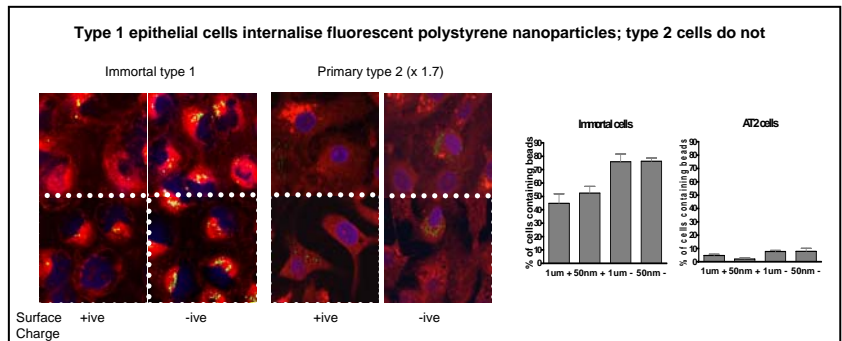
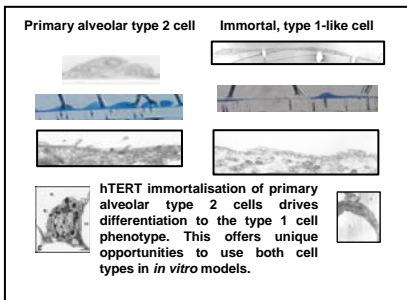
Background

- Inhalation of particulate air pollution has been associated with increased cardiopulmonary morbidity and mortality.
- The ultrafine component, enriched with nano-sized particles, has been implicated.
- Various mechanisms have been suggested.
- Particles may
 - aggravate pulmonary cells to produce mediators that impact on the cardiovascular
 - breach the alveolar-endothelial barrier to access the systemic circulation

DO ENGINEERED NANOPARTICLES POSE THE SAME PROBLEM? IF SO, BY WHICH MECHANISMS, AND WHICH PHYSICOCHEMICAL FEATURES OF ENGINEERED NANOPARTICLES ARE RESPONSIBLE?

In vitro model of human alveolus

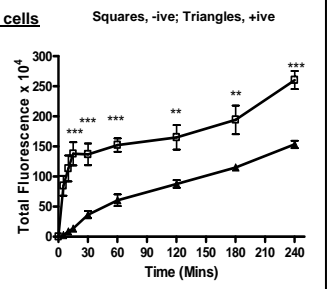
We have developed techniques to isolate primary human alveolar type 2 epithelial cells. It is not possible to isolate human alveolar type 1 epithelial cells, although these form >95% of the alveolar surface area and are the main target of inhaled toxicants. We have immortalised type 2 cells using hTERT to generate type 1 cells. These cells can be used to investigate nanoparticle uptake and toxicity *in vitro*.



Uptake of 50nm of fluorescent polystyrene particles by immortal, type 1-like cells

Our work shows that:

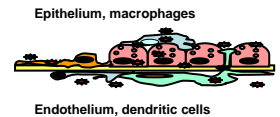
- More negatively charged particles are internalised than positively charged particles
- Uptake of negatively charged particles is initially extremely fast
- Approximately 71% and 57% respectively of negative and positively charged particles were taken up by passive mechanisms.
- Neither caveolae nor clathrin coated vesicles appear to be important in active uptake.



Future work

In vitro: Using simple and complex *in vitro* models of the human alveolar unit to determine nanoparticle-cell interactions at the alveolar epithelial-endothelial barrier (eg metal nanoparticles, nanofibres, nanotubes).

In vivo: To examine effects/translocation of nanoparticles in i) experimental models of cardiopulmonary disease eg emphysema - smoking mice/rats, and ii) genetic mouse models eg SPD deficiency, caveolin KO.



Questions:

- Which physicochemical features (if any) of nanoparticles are likely to initiate cellular reactivity and by which molecular mechanisms. Are these mechanisms toxic and/or pro-inflammatory? Are they likely to impact on the cardiovascular?
- Does modification of the nanoparticles (either deliberately to modify functionality, or incidentally due to adsorption of components in body fluids such as lung lining liquid) affect their structure-function and particle-cell interactions. Are these modifications protective or detrimental to cells?
- Do nanoparticles affect the functional integrity of the alveolar epithelial and endothelial monolayer?
- Do nanoparticles breach the alveolar epithelial-endothelial gas-blood barrier? Do they translocate through cells or between cells? If so, which compartments/organs are targeted?

Using designer particles and commercial particles, state of the art live cell imaging at the level of the electron microscope, labelled particles (eg stable isotopes, radiolabelled, fluorescent core labelled), inhalation facilities in partnership with National-Nanoparticle Inhalation Research Centre.