Direct x-ray measurement of airway surface health in animal models: an update on the state-of-the-art

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Background

• Maintenance of adequate airway surface liquid (ASL) and proper functioning of the mucociliary transit (MCT) system in the pathophysiology of CF airway disease is well understood, but until recently these parameters have been challenging or impossible to measure in live animals.
• Our goal is to non-invasively assess ASL and MCT as outcome measures for developing genetic and pharmaceutical CF airway treatments, so we have developed synchrotron phase contrast x-ray imaging (PCXI) methods to enable monitoring of these measures of airway surface health.

Methods

• All methods were developed in rodents at the SPring-8 Synchrotron in Japan.
• The tracheae of a range of in vivo models were imaged, including normal and β-ENaC mice.
• After anaesthesia and intubation, MCT tracking particles (10–30 μm diameter) were insufflated into the airways (via an endotracheal tube) prior to imaging.
• Rodents were attached to an animal holder and placed supine in the X-ray beam. Image acquisition was triggered from a small animal ventilator, and vital signs were monitored from outside the imaging hutch.
• The effects of aerosolised drug delivery on airway hydration (e.g. hypertonic saline compared to isotonic saline) have been assessed using an Aeroneb vibrating mesh nebuliser in the ventilator inspiratory line.

Imaging setup

• A phase-grid was placed immediately before the trachea, and arranged to cover half the field-of-view (Fig 3), in order to simultaneously perform propagation-based (PB) and single-grid (SG) PCXI.

The SG-PCXI setup allows an airway surface image to be reconstructed from the sample-induced distortions of the grid observed 75 cm downstream (Fig 2), enabling the ASL depth to be visualised (Fig 3).

Results

• Increases in MCT activity (Fig. 6, 7) and ASL depth (qualitative impressions, analysis underway) were detected in mice following administration of aerosolised hypertonic or isotonic saline, but hypertonic saline produced greater rate increases and lasted longer. This agrees with cell culture studies assessed by optical microscopy.

Discussion

• High spatial resolution synchrotron phase-contrast x-ray imaging can reveal the depth of the ASL layer and be used for tracking MCT in live mouse nasal and tracheal airways at SPring-8.
• We are now translating MCT analysis methods to application in larger animal models at the Imaging and Medical Beamline (IMBL) at the Australian Synchrotron, but the coherence and flux density at the IMBL does not allow ASL depth assessment to be performed. In pilot IMBL studies MCT was visible in live sheep and pig tracheal segments, and live pig MCT studies are underway.

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