HIGH FRAME RATE IMAGING OF NASAL FLUID DOSING DYNAMICS

Martin Donnelley[1,3,4], Karen Siu[6,8,7], Kaye Morgan[5], David Parsons[1,2,3,4]

1. Respiratory and Sleep Medicine, Women's and Children's Hospital, Adelaide, South Australia
2. Women's and Children's Health Research Institute, Adelaide, South Australia
3. School of Paediatrics and Reproductive Health, and 4. Centre for Stem Cell Research, University of Adelaide, South Australia,
5. School of Physics, and 6. Monash Biomedical Imaging, Monash University, Melbourne, Victoria
7. Australian Synchrotron, Melbourne, Victoria

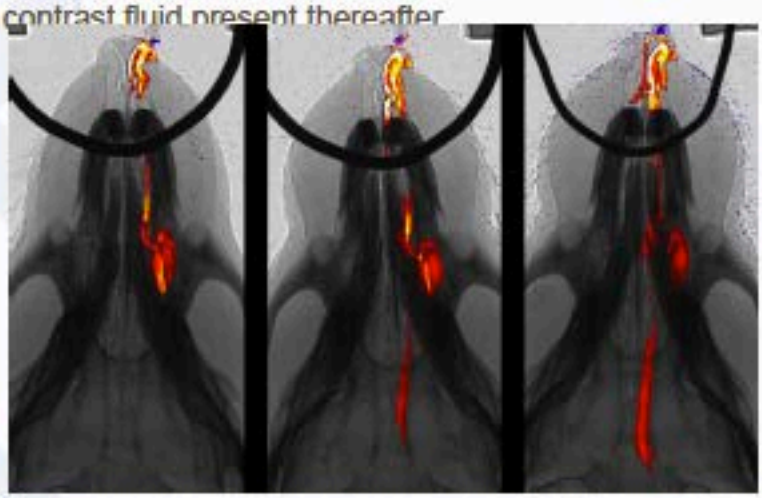
BACKGROUND: The mouse nose is commonly used as an *in vivo* model site for developing gene therapy treatments for CF airway disease. We have used synchrotron X-ray imaging to determine the fate of bolus doses of fluids delivered into live mouse nasal airways to help understand the variability we observe in the success of both reporter-gene expression and electrophysiological measurements of CFTR gene function after vector fluid delivery into mouse nasal airways.
Here, we improved image capture techniques to permit real-time high-resolution monitoring and document the detailed dose movement in live mouse nasal airways.

(F1)

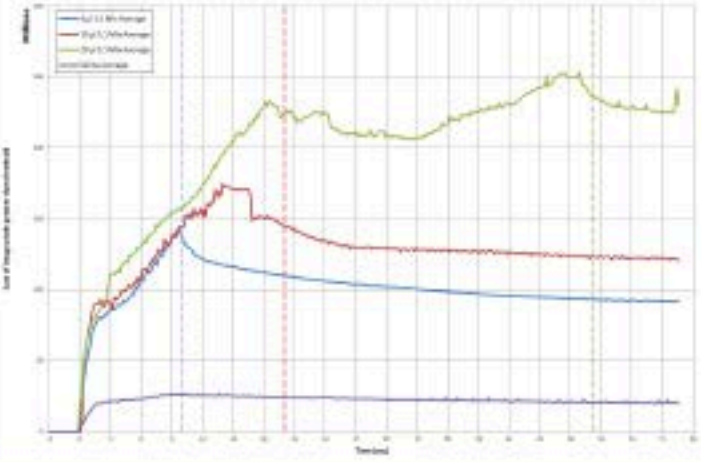
METHODS: Eighteen anaesthetised C57Bl/6 mice were imaged at ~6.67 Hz on the BL20B2 beamline at the SPring-8 synchrotron **(F1)**. After 15 seconds of baseline a 4, 10 or 20 µl sample of iodine-based contrast fluid was delivered over 10 sec **(F2)** to mimic standard doses used in gene-transfer studies. Imaging continued for a further 5 min. Background subtraction and pseudo-colouring revealed fluid position over time. The volume of contrast fluid (i.e. the amount of artificial colour) present in the nose in each image frame was also determined semi-quantitatively for every mouse.

(F2)

RESULTS: The extent of fluid distribution was dose-dependent. Again, contrast fluid was well-tolerated. The 4 µl dose usually remained in the anterior and olfactory region, while the 10 µl and 20 µl doses increasingly moved fluid into the nasaopharyngeal airway towards the lung but with variability in the coverage and persistence on airway surfaces. On average (F5) there was consistent airway filling in the first 10 sec. For the 4 µl dose there was a gradual small reduction after instillation ceased. At the highest dose (20 µl) the amount of fluid continued to increase until approximately 19 sec (at which point approximately 10 µl of fluid had been delivered). There was substantial variability in the amount of contrast fluid present thereafter.



(F4)



(F5)

CONCLUSION: High frame-rate X-ray imaging can now reveal the real-time dynamics of this commonly used dosing technique in mice. Clear in vivo visualisation of nasal airways reveals that outcome variability may be influenced by the heterogeneous dose distribution that occurs in these anatomically complex nasal airways.

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