5-19-2022

E-FLOAT: Extractable floating liquid gel-based organ-on-a-chip for airway tissue modeling under airflow

Siwan Park

Edmond W. K. Young

E-FLOAT: EXTRACTABLE FLOATING LIQUID GEL-BASED ORGAN-ON-A-CHIP FOR AIRWAY TISSUE MODELING UNDER AIRFLOW

Siwan Park, University of Toronto, Canada
siwan.park@mail.utoronto.ca
Edmond W. K. Young, University of Toronto, Canada

Key Words: Lung-on-a-Chip, Airway Epithelium, Matrix Interactions, Particulate Matter, Microfluidics

Chronic respiratory diseases (CRDs) such as asthma and cystic fibrosis represent major global health challenges and are responsible for >4 million deaths each year.[1] These respiratory diseases are exacerbated by exposure to allergens, pollutants, particulate matter (PM), and viruses. A major problem is that existing lung models for testing drug delivery do not represent real human tissues. Thus, there is a need for advanced *in vitro* models that can facilitate cellular and molecular research of the respiratory system and potentially serve as a drug screening platform for drug development. The development of emergent "organ-on-a-chip" technologies has enabled us to recreate key features of various tissues and organs within engineered microfluidic devices. However, current microfluidic lung-on-a-chip devices do not recapitulate the mechanobiological effect of airflow-induced shear stress on the epithelial cells of larger airways. Also, the existing devices are only limited to on-chip analysis that may hinder the compatibility with existing assessment tools. We have developed a lung-on-a-chip system referred to as E-FLOAT for airway tissue modelling that is arrayable, scalable, and contains a removable cell-ladened hydrogel construct capable of withstanding airflow. We also have developed an airflow system that allows culturing of airway cells under the shear flow to accurately represent the aerodynamic conditions in the native airways. The E-FLOAT device was fabricated by micro-milling the poly(methyl methacrylate) (PMMA) plastic layers and bonding the layers via liquid solvent bonding.[2] (Fig 1a) The device incorporates an anchored suspended hydrogel where we cultured the airway epithelial cells (AECs) under airflow. The airflow system controls the flow rate, humidity, and temperature of the air for the purpose of effectively inducing accurate mucociliary differentiation of AECs. In this study, AECs were cultured under three different conditions: *submerged in media, static air-liquid interface (ALI), or with airflow*. After experiments, the hydrogels were extracted for various downstream analyses such as immunostaining, scanning electron microscopy (SEM) and histology sectioning. Airflow culture showed the most accurate recapitulation of native airway epithelium. AECs showed high viability and more localized tight junction expression when cultured with airflow, suggesting that airflow promotes improved barrier function compared to the other two conditions. Furthermore, the number of mucin-producing goblet cell differentiation with airflow was shown to be more representative of the native airway epithelium (~10% of epithelial population) (Fig 1b). Also, the AECs cultured with airflow showed more physiological expression of long cilia (6~7μm), while cells in submerged and ALI conditions showed shorter microvilli (0.5-1μm) (Fig 1c). Finally, as a demonstration, physiological deposition of carbon black particulates via airflow was achieved and confirmed by SEM (Fig 1e).
E-FLOAT has unique features that allow for the extraction of the cell-laden hydrogel, providing access to various analysis tools that are not accessible with other lung-on-a-chip devices. In addition, since it uses a biocompatible hydrogel, the experiments can be extended to freely add the components that make up for the airway tissue construct. E-FLOAT has potential to serve as a disease model for CRDs and a drug screening platform, with potential applications toward understanding nanoparticle toxicity and particular matter exposures leading to the exacerbation of CRDs.

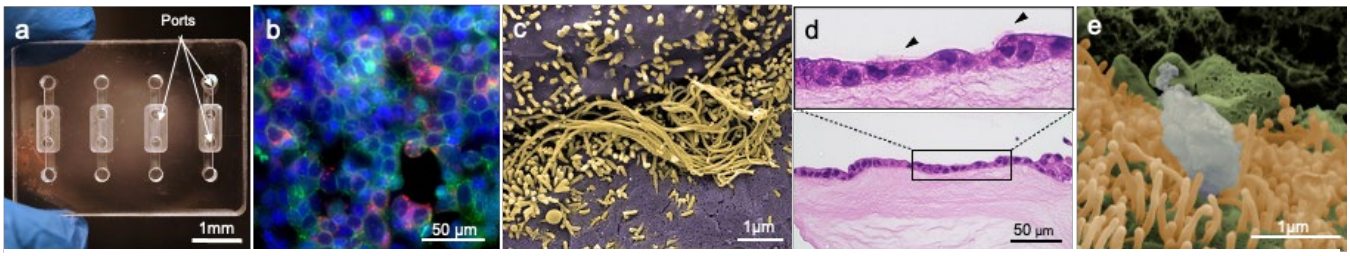

Figure 1. (a) Photograph of fabricated E-FLOAT device (b) Immunostained AECs cultured under flow condition. ZO-1 (green); MUC5AC (red); Hoechst (blue). (c) SEM image of AEC cilia expression after being cultured under airflow. (d) Histology sectioning and H&E staining of the extracted hydrogel. (e) SEM image of physiological particulate (carbon black) deposition experiment on the airway epithelium.

[1] T. Ferkol, *et al*, Annals of the American Thoracic Society, vol. 11, no. 3, 404-406, 2014.
[2] A.M.D. Wan, *et al*, Lab Chip, vol. 15, no. 18, 3785-3792, 2015