

Supplementary Figure S1. Overview of all antibodies used for immunohistochemistry

Target	Class	Host	Isotype	Conjugate	Conc	Vendor	Cat #
Acetyl- α -Tubulin (Lys40) (D20G3)	Monoclonal	Rabbit	IgG	AF 647	1:50	Cell Signaling Technology	81502
Mucin 5AC	Polyclonal	Rabbit	IgG		5 μ g/ml	Abcam	ab78660
CDH26	Polyclonal	Rabbit	IgG		1:100	Invitrogen	PA5-103396
VANGL1	Polyclonal	Rabbit	IgG		1:100	Invitrogen	PA5-55231
ZO-1 (1A12)	Monoclonal	Mouse	IgG	AF 488	1:50	Invitrogen	339188
Anti-Rabbit (Secondary)	Polyclonal	Goat	IgG	AF488	1:1000	Invitrogen	A-11008
Anti-Mouse (Secondary)	Polyclonal	Goat	IgG	AF488	1:1000	Invitrogen	A-11001

Supplementary Figure S2. Validation and introduction of immune component to airway chip. **A)** Phase images of naïve and cytokine-activated macrophages showing the ability of the macrophages to respond to cytokine activity. **B)** To demonstrate that the CD14-positive cells were effectively turned into immune macrophages, we show that only the differentiated macrophages produce a robust secretion of TNF alpha in response to LPS (1µg/ml) as compared to undifferentiated CD14-positive cells and macrophages not exposed to LPS (N=5, p=.05). **C)** Introduction of macrophages into the vascular side of the airway chip shows that these circulating macrophages do produce significant elevation in both IL-6 and TNF alpha (N=4, p=.01).

