Supplementary Material

Supporting Cells of the Human Olfactory Epithelium Co-Express the Lipid Scramblase TMEM16F and ACE2 and May Cause Smell Loss by SARS-CoV-2 Spike-Induced Syncytia

Andres Hernandez-Clavijo^a Kevin Y. Gonzalez-Velandia^a Uday Rangaswamy^a Giorgia Guarneri^a Paolo Boscolo-Rizzo^b Margherita Tofanelli^b Nicoletta Gardenal^b Remo Sanges^a Michele Dibattista^c Giancarlo Tirelli^b Anna Menini^a

^aNeuroscience Area, SISSA, Scuola Internazionale Superiore di Studi Avanzati, Trieste, Italy, ^bDepartment of Medical, Surgical and Health Sciences, Section of Otolaryngology, University of Trieste, Trieste, Italy, ^cDepartment of Basic Medical Sciences, Neuroscience and Sensory Organs, University of Bari A. Moro, Bari, Italy

Cluster_number	Cell_count	Cell_type	
0	3761	Olfactory HBCs/Respiratory HBCs	
1	3668	Fibroblasts/Stromal cells	
2	2178	CD8+ T Cells	
3	2026	Supporting cells	
4	1990	Pericytes	
5	1697	Vascular smooth muscle cells	
6	1269	Pericytes	
7	1242	CD4+ T Cells	
8	1210	Respiratory gland progenitor cells	
9	1199	Respiratory secretory cells	
10	1189	Supporting cells	
11	1181	Macrophages/Dendritic cells	
12	1176	Bowman's glands	
13	1101	Plasma cells	
14	856	Respiratory ciliated cells	
15	805	Immature neurons/Mature neurons	
16	581	Bowman's glands	
17	471	Bowman's glands	
18	365	Monocytes	
19	315	Plasma cells	
20	269	Natural killer cells	
21	263	B cells	
22	251	GBCs	
23	246	Olfactory ensheathing glia	
24	159	Mast cells	
25	93	Olfactory microvillar cells	
26	67	Olfactory HBCs/Respiratory HBCs	

Supplementary Table 1. Clusters, cell numbers and cell type.

Cell_type	Cell_count
Olfactory HBCs/Respiratory HBCs	3828
Fibroblasts/Stromal cells	3668
CD8+ T Cells	2178
Supporting cells	3215
Pericytes	3259
Vascular smooth muscle cells	1697
CD4+ T Cells	1242
Respiratory gland progenitor cells	1210
Respiratory secretory cells	1199
Macrophages/Dendritic cells	1181
Bowman's glands	2228
Plasma cells	1416
Respiratory ciliated cells	856
Immature neurons/Mature neurons	805
Monocytes	365
Natural killer cells	269
B cells	263
GBCs	251
Olfactory ensheathing glia	246
Mast cells	159
Olfactory microvillar cells	93

Supplementary Table 2. Overall cell count for each group of cell type.



Supplementary Fig. S1. *TMEM16F*, *ACE2* and *TMPRSS2* co-expression in cells from human nasal epithelium. (A-B) Co-expression of *ACE2* and *TMPRSS2* (A) or *TMEM16F* and *TMPRSS2*. (B) in the different cell clusters. Mean normalized expression levels are plotted. (C) Bar plot displaying the number of cells co-expressing *ACE2*, *TMEM16F* and *TMPRSS2* by cluster. (D) 3D-bar plot displaying the co-expression of *ACE2*, *TMPRSS2* and *TMEM16F* by cell cluster. Mean normalized expression levels are plotted. The three genes are co-expressed in respiratory secretory cells, respiratory ciliated cells, olfactory supporting cells, olfactory microvillar cells, Bowman's gland cells, horizontal basal cells and globose basal cells clusters.



Supplementary Fig. S2. Control experiments for immunohistochemistry in the absence of primary antibodies. (A-B) Section of human respiratory epithelium immunostained for (A) TMEM16F (green) and BT4 (red) (same slice as in Figure 3C); and (B) negative control avoiding primary antibodies. (C-D) Section of human olfactory epithelium immunostained for (C) TMEM16F (green) and TUJ1 (red) (same slice as in Fig.3D-F); and (D) negative control avoiding primary antibodies. (E-F) Section of human respiratory epithelium immunostained for (E) BT4 (green) and ACE2 (red) (same slice as in Figure 2C); and (B) negative control avoiding primary antibodies. Pseudocolor was modified in Figure 2C for easier visualization. Same brightness and contrast modification was applied to images from positive immunohistochemistry and its respective negative control. Nuclei were stained with DAPI (blue).