

Online supplementary data

Title: Bacteria-driven peribronchial lymphoid neogenesis in bronchiectasis and cystic fibrosis

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Supplementary Table S1. Clinical characteristics of patients

	Control (n=17)	Bronchiectasis (n=8)	Cystic fibrosis (n=17)
Female, n (%)	11 (65%)	6 (75%)	8 (47%)
Age, years (mean \pm SD)	67 \pm 10	55 \pm 16	34 \pm 9
FEV₁, % predicted	100 \pm 18	78 \pm 35	22 \pm 6
CFTR genotype, n			
Phe508del / Phe508del	Unknown	Unknown	10
Phe508del / Other	Unknown	Unknown	5
Other	Unknown	Unknown	2
Bronchial colonization*, n			
<i>P. aeruginosa</i>	0	4	16
<i>S. aureus</i>	0	2	16
<i>H. influenzae</i>	0	0	0
<i>S. maltophilia</i>	0	2	0
<i>A. xylosoxydans</i>	0	1	1

*, Some patients displayed colonization with two or more pathogens; FEV₁: Forced expiratory volume in one second, CFTR: Cystic fibrosis transmembrane conductance regulator

Supplementary Table S2. Primary antibodies used for immunohistochemical staining in human and in mouse lung sections

Antigen	Source	Manufacturer	Reference	Dilution	Unmasking
Human sections					
Human CD20	Mouse monoclonal	Dako	M0755	1:200	Citrate microwave
Human CD3	Rabbit polyclonal	Dako	A0452	1:200	Citrate microwave
Human FDC	Mouse monoclonal	Dako	M7157	1:50	Citrate microwave
Human PNAd	Rat monoclonal	Santa Cruz	sc19602	1:50	None
Human PCNA	Rabbit polyclonal	Calbiochem	PC474	1:200	None
Human CXCL12	Rabbit polyclonal	Santa Cruz	Sc28876	1:50	Citrate microwave
Human CXCL13	Goat polyclonal	R&D	AF801	1:50	Citrate microwave
Mouse IL-17A	Rabbit polyclonal	Santa Cruz	sc7927	1:50	None
Mouse sections					
Mouse CD20	Goat polyclonal	Santa Cruz	sc7735	1:250	None
Mouse CD3	Rat monoclonal	Santa Cruz	sc101442	1:200	Protease
Mouse FDC	Rat monoclonal	BD Pharmingen	Antigen M1	1:50	Protease
Mouse PNAd	Rat monoclonal	BD Pharmingen	553863	1:50	None
Mouse PCNA	Rabbit polyclonal	Calbiochem	PC474	1:200	Protease
Mouse CXCL12	Rabbit polyclonal	Santa Cruz	sc28876	1:50	Citrate microwave
Mouse CXCL13	Goat polyclonal	R&D	AF470	1:20	Citrate microwave
Mouse IL-17A	Rabbit polyclonal	Santa Cruz	sc7927	1:50	None

Supplementary Table S3. Identification of lymphoid and myeloid cell populations by flow cytometry in the bronchoalveolar lavage.

Cell Subset	Phenotype
Granulocytes	CD45 ⁺ CD11b ⁺ CD11c ⁺ NK1.1 ⁻ Ly-6G ⁺
Monocytes/Macrophages	CD45 ⁺ CD11b ⁺ CD11c ⁻ NK1.1 ⁻ Ly-6G ⁻ Ly-6C ⁻
CD4 ⁺ Tcells	CD45 ⁺ CD4 ⁺ CD8 ⁻ TCRβ ⁺
CD8 ⁺ Tcells	CD45 ⁺ CD4 ⁻ CD8 ⁺ TCRβ ⁺
γδ T cells	CD45 ⁺ CD4 ⁻ TCRγδ ⁺
Myeloid dendritic cells	CD45 ⁺ CD11b ⁺ CD11c ⁺ Ly-6C ⁻
B cells	CD45 ⁺ CD19 ⁺

Supplementary Table S4. Antibodies used for detection of myeloid and lymphoid cells by flow cytometry in the bronchoalveolar lavage

Antibodies used for detection of myeloid cells			
Antigen	Manufacturer	Reference	Dilution
Ly6C biotinylated	BD Pharmingen	557359	1:100
SAV PB	Invitrogen	S11222	1:600
CD11b APC	eBioscience	170112832	1:200
CD11c PE Cy7	BD Pharmingen	558079	1:100
NK1.1 PE	BD Pharmingen	557391	1:100
Ly6G FITC	BD Pharmingen	551460	1:100
CD45 Pcp Cy5.5	BD Pharmingen	550994	1:100
Antibodies used for detection of lymphoid cells			
Antigen	Manufacturer	Reference	Dilution
TCRβ biotinylated	BD Pharmingen	553169	1:100
SAV PE Cy7	BD Pharmingen	557598	1:100
CD4 PB	BD Pharmingen	558107	1:100
CD8 APC H7	MACS	130096602	1:50
CD19 FITC	BD Pharmingen	553785	1:100
NK1.1 Pcp Cy5.5	BD Pharmingen	551114	1:100
TCRgd PE	BD Pharmingen	553178	1:100
CD45 APC	BD Pharmingen	559864	1:100