ONLINE SUPPLEMENTAL MATERIAL

Supplemental Table 1: **Cytokine and chemokine levels in BALF samples from WT and MMP8$^{-/-}$ mice 4 h after IT LPS or IT PBS**

	Wild Type Mice				MMP-8$^{-/-}$ Mice			
	IT PBS		IT LPS		IT PBS		IT LPS	
	Mean (SEM) pg/BAL	N[J]	Mean (SEM) pg/BAL	N	Mean (SEM) pg/BAL	N	Mean (SEM) pg/BAL	N
KC[I]	812 (251)	7	10250 (4350)	18	676 (317)	11	7749 (1621)	20
MIP2-α	889 (344)	9	2542 (501)	16	385 (174)	10	4075 (1005)	20
LIX	410 (100)	4	511 (80)	17	250 (20)	5	750 (110)	16
LTB4	190 (50)	4	553 (140)	11	353 (208)	3	422 (95)	10
TNF-α	543 (314)	8	4202 (510)	20	293 (141)	7	4566 (794)	21
IL-1β	40 (18.8)	9	61 (24)	20	44 (19)	8	58 (32)	19
IL-6	6 (3)	4	678 (231)	11	3 (3)	4	694 (234)	10
GM-CSF	1 (1)	6	241 (85)	12	8 (6)	7	137 (45)	13
G-CSF	65 (12)	4	2411 (1050)	11	57 (15)	4	2878 (1204)	10
IL-10	N.D.[K]	5	7.0 (7.0)	14	N.D.	6	2.9 (2.9)	14

[I] Chemokines and cytokines were quantified in BALF samples harvested 4 h after IT LPS was instilled in WT and MMP-8$^{-/-}$ mice. Data are mean (SEM).

[J] N = number of mice per group.

[K] N.D. indicates not detected.

There were no statistically significant differences between PBS-treated WT vs. PBS-treated MMP8$^{-/-}$ mice or between LPS-treated WT vs. LPS-treated MMP8$^{-/-}$ mice for any of the mediators listed.

Supplemental Table 2: Cytokine and chemokine levels in BALF samples from WT and MMP8$^{-/-}$ mice 24 h after IT LPS or IT PBS

	Wild Type Mice				MMP-8$^{-/-}$ Mice			
	IT PBS		IT LPS		IT PBS		IT LPS	
	Mean (SEM) pg/BAL	N[M]	Mean (SEM) pg/BAL	N	Mean (SEM) pg/BAL	N	Mean (SEM) pg/BAL	N
KC[L]	38 (8)	6	503 (67)	12	41 (10)	5	357 (62)	12
MIP-2α	33 (10)	6	363 (53)	12	20 (3)	5	514 (63)	12
LIX	102 (21)	5	1066 (229)	13	160 (82)	6	1241 (172)	16
LTB4	398 (182)	7	525 (74)	13	816 (98)	7	674 (129)	15
TNF-α	79 (10)	4	368 (66)	10	77 (6)	4	534 (174)	10
IL-1β	3 (3)	4	9 (3)	10	1 (1)	4	5 (3)	10
IL-6	2 (1)	4	933 (216)	10	2 (2)	5	950 (220)	11
GM-CSF	53 (14)	4	26 (7)	10	47 (11)	4	29 (7)	10
G-CSF	76 (44)	5	6735 (2130)	10	76 (25)	5	6785 (1403)	10
IL-10	68 (4)	7	61 (8)	8	62 (2)	6	62 (5)	8

[L]Chemokines and cytokines were quantified in BALF samples harvested 24 h after PBS or LPS was instilled by the IT route in WT and MMP-8$^{-/-}$ mice.

[M]N = number of mice per group.

There were no statistically significant differences between PBS-treated WT vs. PBS-treated MMP8$^{-/-}$ mice or between LPS-treated WT vs. LPS-treated MMP8$^{-/-}$ for any of the mediators listed.

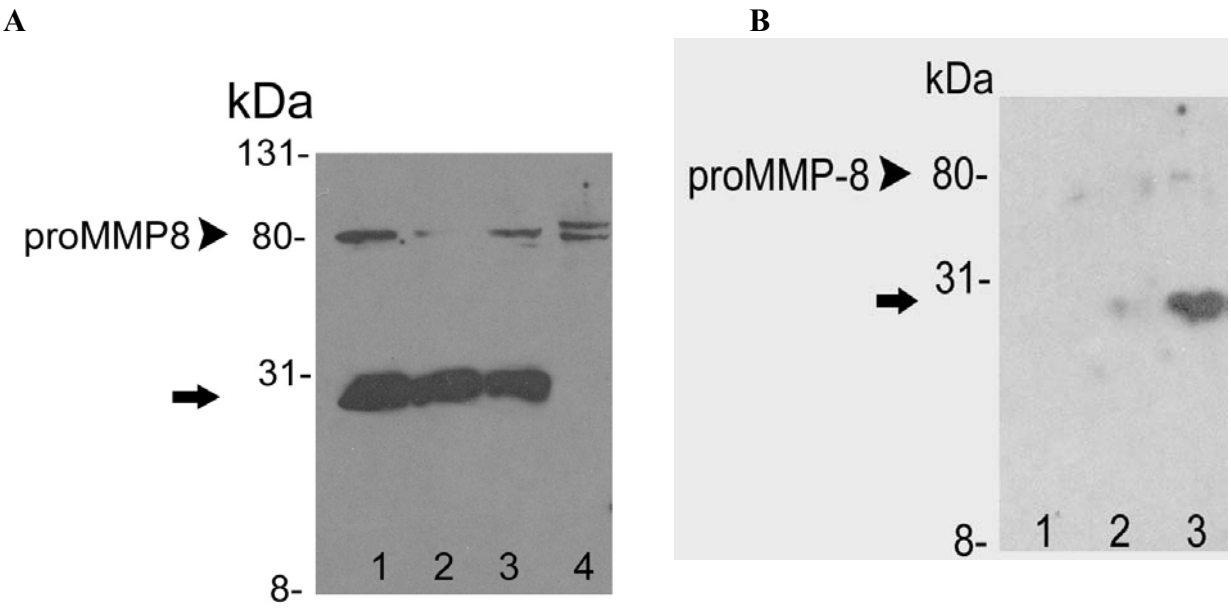
Supplemental Table 3: **MIP-1α increases PMN migration in vitro**

Stimulus[N]	**PMNs migrating per field**
Buffer	29.5 (5.4)[O]
10^{-7}M MIP-1α	67.0 (11.8)
10^{-7}M MIP-1α + goat anti-MIP-1α	24.6 (4.3)
10^{-7}M MIP-1α + goat IgG	60.5 (4.0)

[N] Human PMNs were tested for their capacity to migrate in Boyden microchemotaxis assay chambers in response to buffer, 10^{-7}M MIP-1α, or 10^{-7}M MIP-1α that was pre-incubated for 30 min at 37°C with or without a neutralizing antibody to MIP-1α or an isotype control antibody (both at 100 μg/ml).

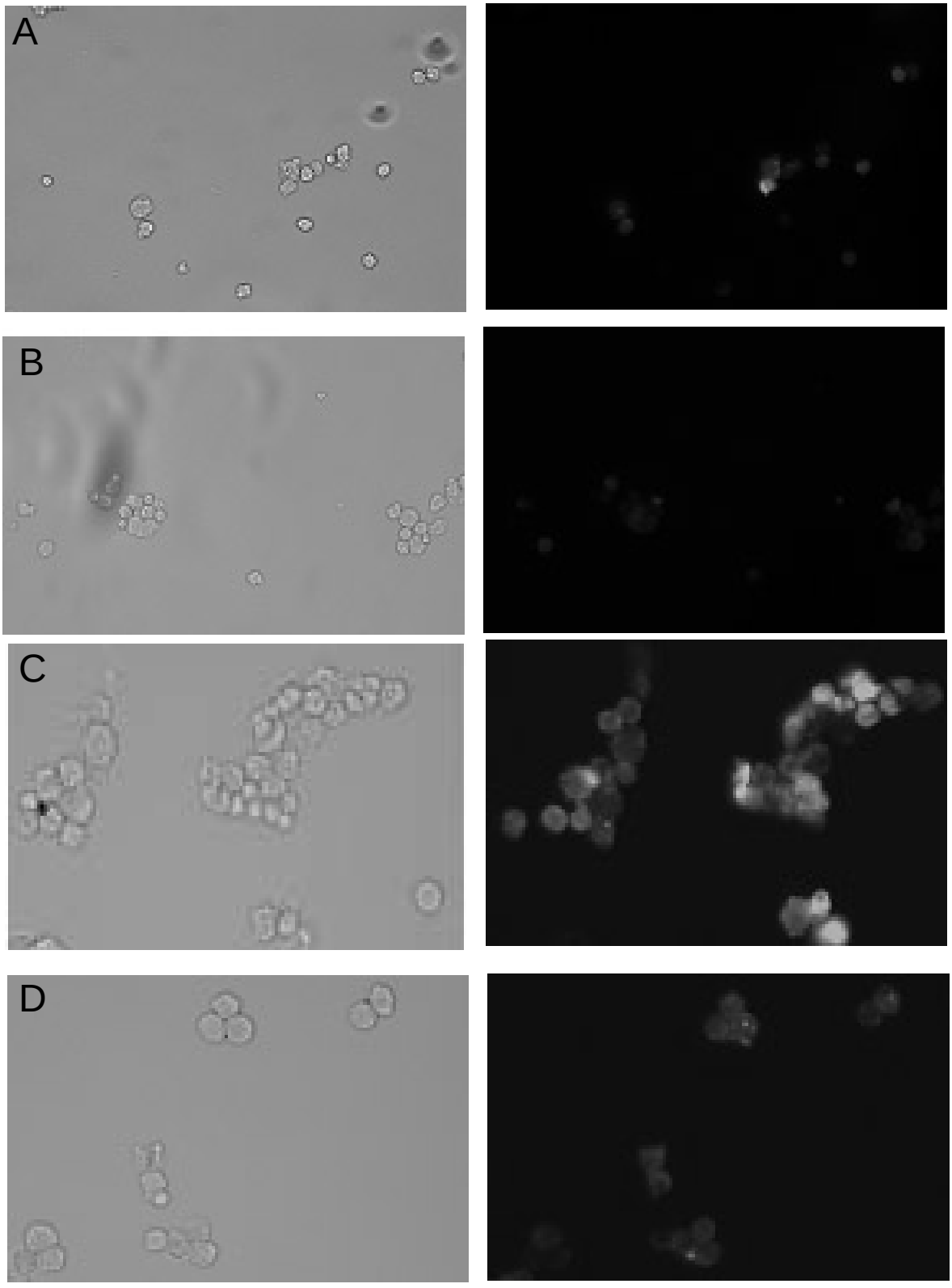
[O]Data are mean ± SD number of PMNs migrating per high magnification field counted in 3-6 wells per condition.

Supplemental Figure 1



Supplemental Fig. 1: Soluble, active MMP-8 is not detected in bronchoalveolar lavage cell-free supernatant samples from WT mice with LPS-mediated ALI. **A.** Four WT mice (lanes 1-4) were given IT LPS and after 24 h BAL was performed and BALF cell-free supernatant samples were immunoblotted for MMP-8 as described previously (2). Note that two forms of MMP-8 corresponding to latent pro-MMP-8 (~80 kDa; arrowhead), and a lower M_r fragment of MMP-8 (~27 Da; arrow) were detected in BALF samples. However, active soluble MMP-8 (M_r ~60 kDa) was not detected in BALF from any of the LPS-treated WT mice. **B.** As a control, BALF from two $MMP8^{-/-}$ mice (lanes 1 and 2) and one WT mouse (lane 3) were immunoblotted for MMP-8. Note the lack of all forms of MMP-8 in BALF from $MMP8^{-/-}$ mice as expected.

Supplemental Figure 2



Legend to Supplemental Figure 2: WT mice were given IT LPS (10 μg) and after 6 h (**A** and **B**) or 72 h (**C** and **D**), PMNs were harvested from the lung by BAL. Non-permeabilized cells were immunostained with rabbit anti-MMP-8 (A and C) or with non-immune rabbit IgG (B and D) followed by alexa-488 conjugated goat anti-rabbit $F(ab)_2$. Left panels show bright field images and right panels show cells examined under epi-fluorescence microscopy. Note that there is minimal surface staining for MMP-8 on BAL cells 6 h after IT LPS (in **A**) but intense staining for MMP-8 72 h after IT LPS (in **C**).