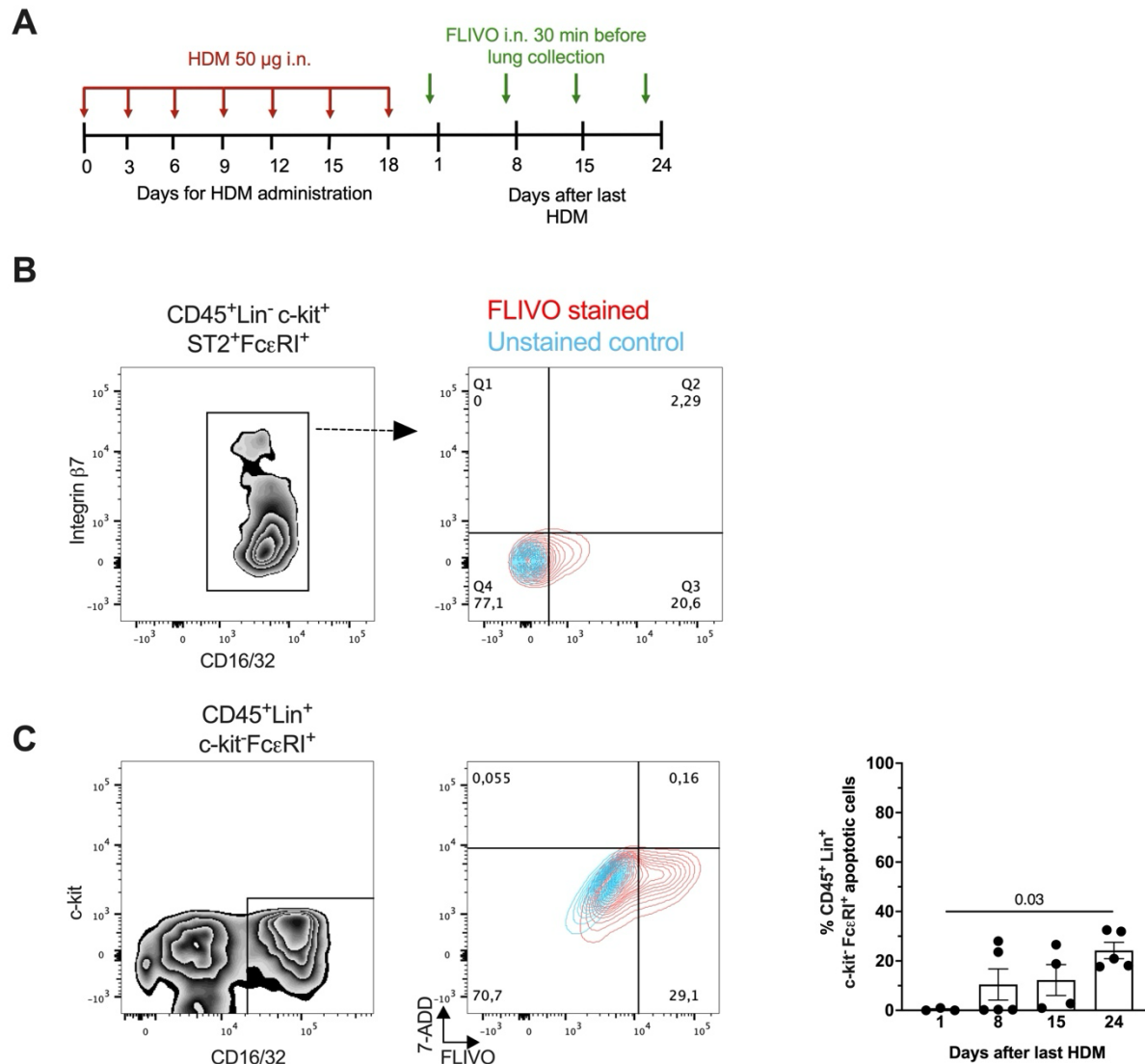
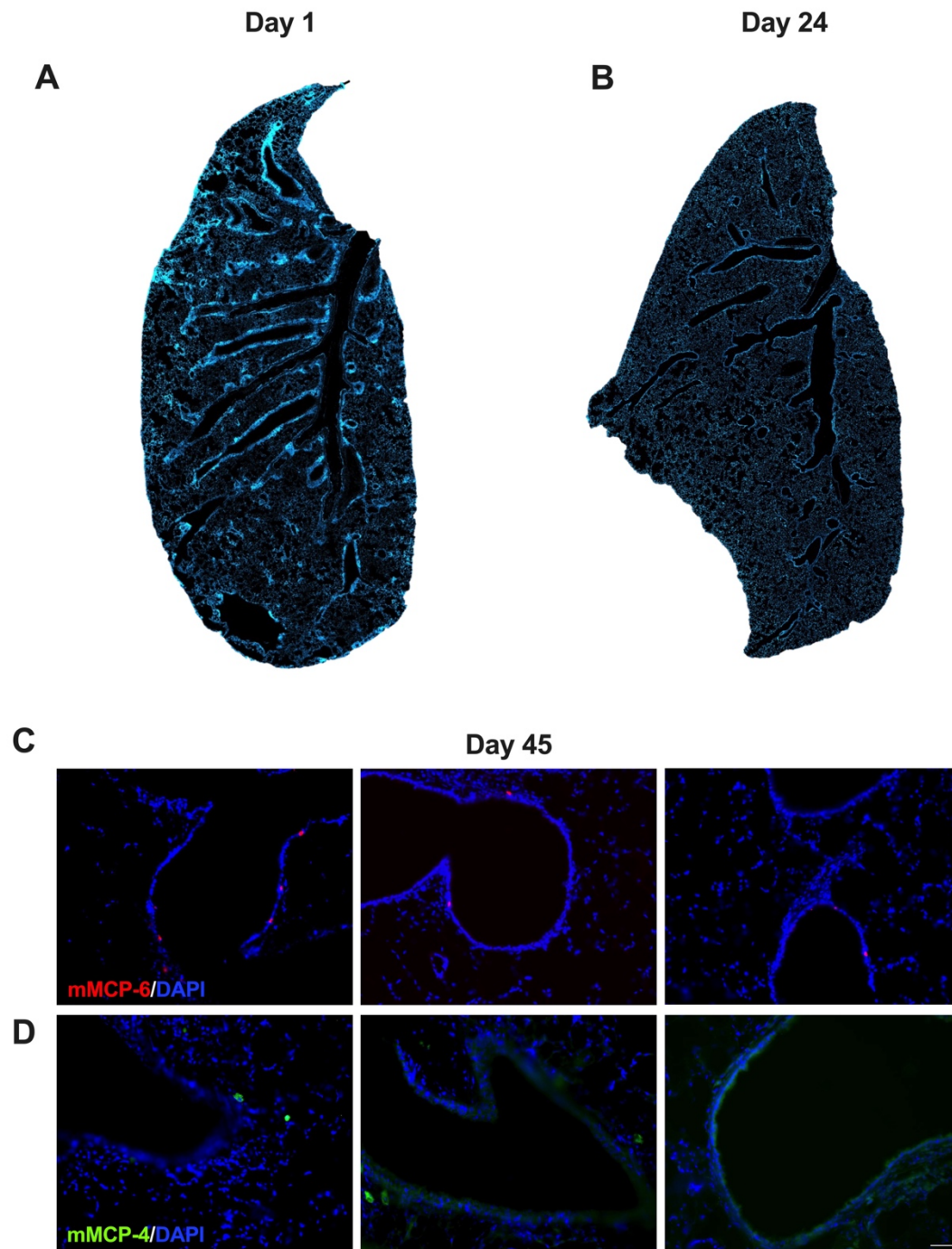


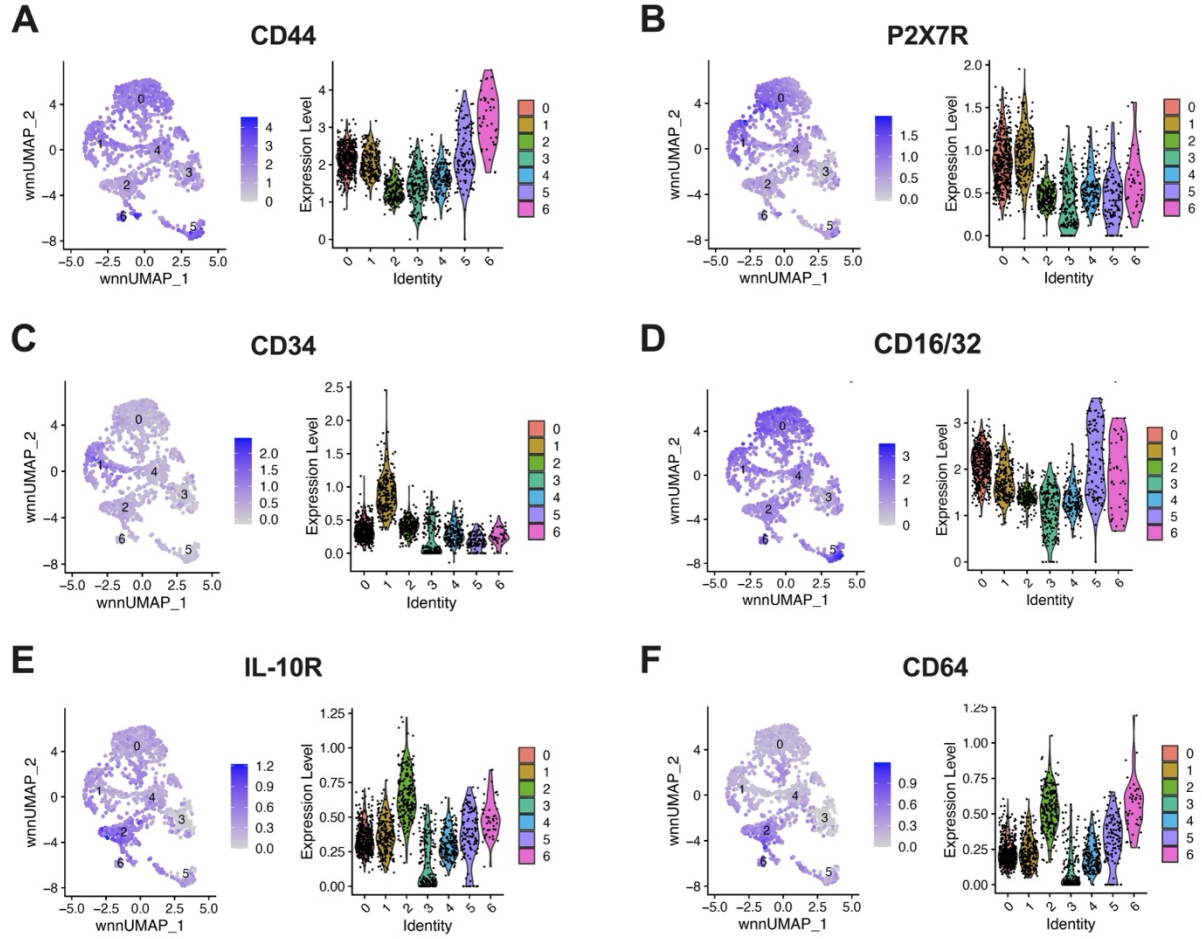
## Supplementary data



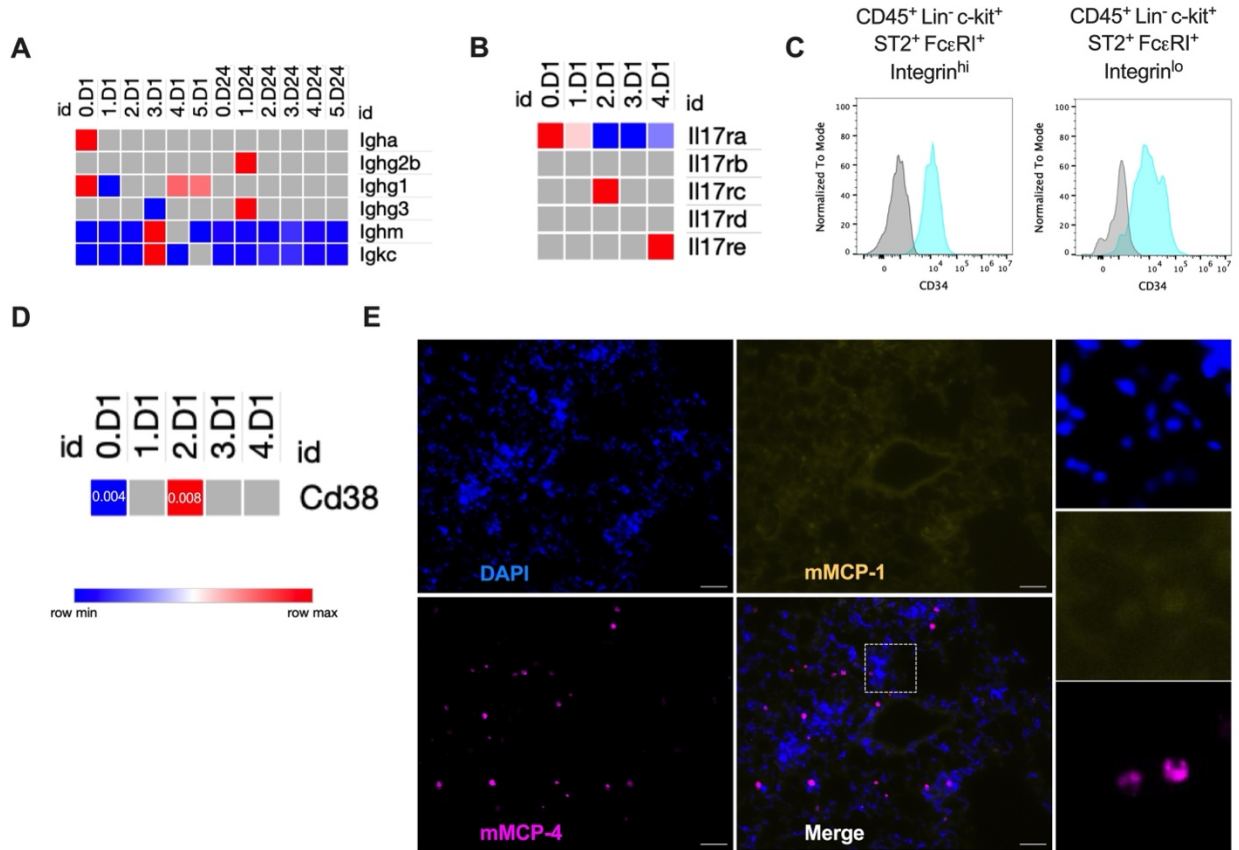
**Fig. S1. FLIVO Administration and apoptotic cell analysis in lung MCs populations.** **A** The fluorescently labeled poly-caspase inhibitor FLIVO was administrated 30 min before tissue collection at the indicated time points after the last HDM administration. **B-C** The proportion of apoptotic cells among the CD45<sup>+</sup> Lin<sup>-</sup> c-kit<sup>+</sup> ST2<sup>+</sup> FcεRI<sup>+</sup> MC populations (**B**), and among the whole CD45<sup>+</sup> Lin<sup>+</sup> c-kit<sup>+</sup> FcεRI<sup>+</sup> population (**C**) was investigated by flow cytometry. The lineage markers included: CD3, CD4, CD8b, CD19, B220, TER119, and Ly-6G. Bars represent the mean ± SEM of two independent experiments. Statistical differences were tested by one-way ANOVA with Tukey post-hoc test.



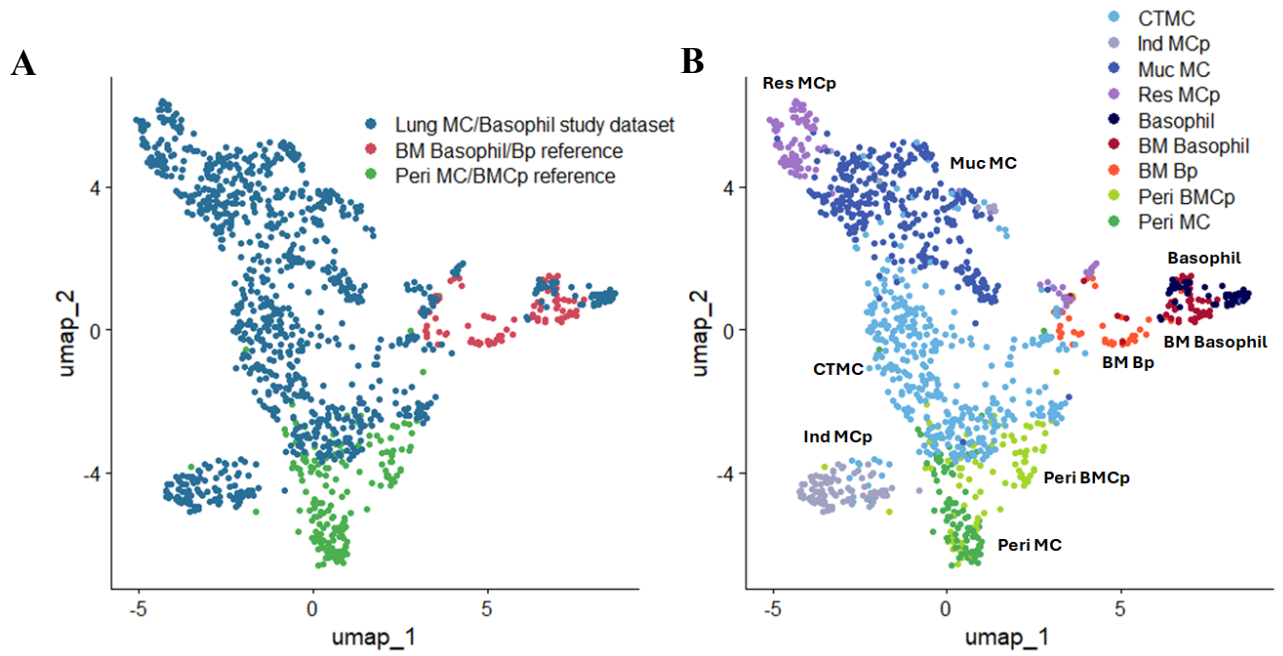
**Fig. S2. Lung inflammation is absent 24 days after the last HDM, and mMCP-6<sup>+</sup> and mMCP-4<sup>+</sup> cells can be observed even 45 days after last the HDM administration.** A-B Representative images of the left lobe lung were collected at day 1 or 24 after the last HDM administration and the general presence of inflammatory cells was visualized by DAPI. C-D In a single experiment, three mice subjected to the HDM protocol were left for 45 days post-HDM, and the lungs immunostained for mMCP-6<sup>+</sup> cells (C) and mMCP-4<sup>+</sup> cells (D) in the lung sections collected on day 45 after the last HDM administration. The scale bar represents 50  $\mu$ m.



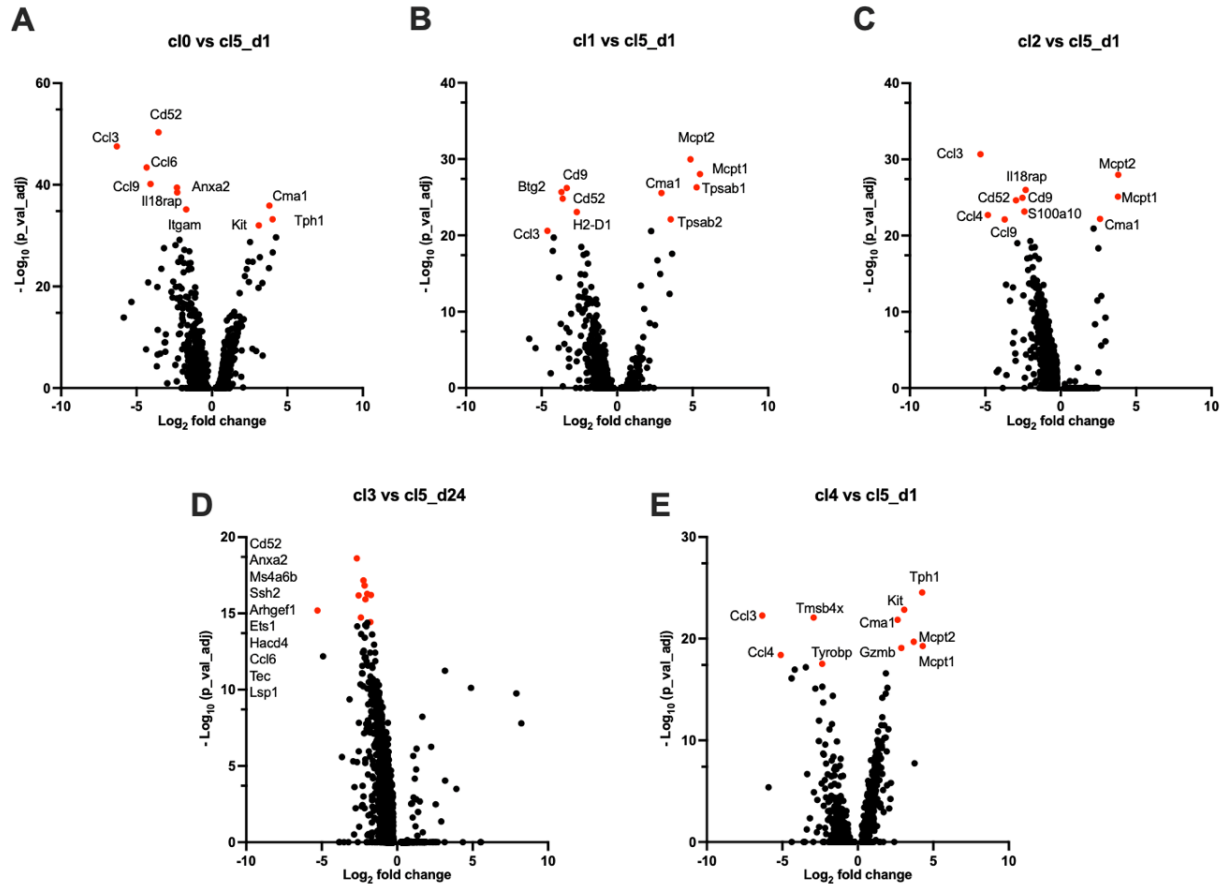
**Fig. S3. UMAPs and Violin plots illustrating additional oligo-tagged surface markers for the distinct clusters in the dataset. A-F** The UMAP highlights differentially expressed surface proteins across the CITE-seq identified cell populations, and the matching violin plots display the distribution and relative surface expression levels of CD44 (A), P2X7R (B), CD34 (C), CD16/32 (D), IL-10R (E), CD64 (F) among the seven clusters identified by CITE-seq.



**Fig. S4. Expression profiles of immunoglobulin heavy chain genes, IL-17 receptor subunits, and transcription factors in diverse clusters, and representative visualization of co-immunostaining for mMCP-4 and mMCP-1 in MCs at day 24.** **A-B** Heat maps showing the average expression of different subclasses of immunoglobulin heavy chain genes (**A**) and different IL-17 receptor subunits (**B**). Grey values represent the absence of expression. **C** The expression of CD34 by flow cytometry in lung MC populations. The Lin markers included: CD3, CD4, CD8b, CD19, B220, TER119, and Ly-6G. **D** The expression of the CD38 gene in the MC clusters at day 1. The numbers within the heat map indicate the average transcript levels in each group, while grey values denote the absence of expression. **E** Representative immunostaining of lung samples collected on day 24 post-final HDM showing the absence of mMCP-1 staining within the mMCP-4<sup>+</sup> cells in the submucosa and parenchyma. The scale bar represents 50 μm.



**Fig. S5: Projection of two reference datasets onto the cell type clusters identified by the scRNA-seq analysis in our study.** **A** UMAP visualization showing the integration of the dataset used in the current study (Lung MCs/basophil), and two reference datasets comprising of mouse bone marrow basophils and basophil progenitors (BM Basophil/Bp) and mouse peritoneal MCs and bipotent basophil MC progenitors (Peri MC/BMCp). **B** UMAP visualization of the clustering of the identified cell types in this study compared to those present in the reference datasets. BM, bone marrow; Peri, peritoneal; Bp, basophil progenitor; BMCp, basophil MC progenitor; MCp, MC progenitor; Ind, induced; Muc, mucosal; Res, resident.



**Fig. S6. Single-cell RNA-sequencing (scRNA-seq) analysis reveals that MC proteases are upregulated in clusters 0–4 compared with basophils (Cluster 5).** A–E Volcano plots illustrating the top 10 most significantly differentially expressed genes in each cluster (cl) 0–4, relative to basophils (cl5). Differential expression was evaluated at the indicated time points, with clusters 0, 1, 2, and 4 assessed on day 1 and Cluster 3 assessed on day 24. The top 10 DEGs in each cluster are labeled in red for clarity.



**Table S1.** Please see the separate excel file.

**Table S2.** Antibodies used in flow cytometry.

Target	Fluorochrome	Clone	Vendor
CD45	Alexa flour 700	30-F11	BD Biosciences
Lin: CD3	BV510	17A2	BD Biosciences
Lin: CD4	BV510	GK1.5	eBioscience
Lin: CD8b	BV510	eBioH35-17.2	eBioscience
Lin: CD19	BV510	ebio1D3	eBioscience
Lin: B220	BV510	RA3-6B2	eBioscience
Lin: TER-119	BV510	TER-119	eBioscience
Lin: Gr1	BV510	RB6-8C5	eBioscience
Lin: CD11b	BV510	M1/70	eBioscience
C-kit	BV421	2B8	eBioscience
CD16/32	BV605	2.4G2	BD Biosciences
Integrin $\beta$ 7	PE-Cy7	FIB504	eBioscience
Fc $\epsilon$ RI	PE	MAR-1	eBioscience
ST2	BV786	DIH9	BioLegend
CD34	BV421	SA376A4	BioLegend

**Table S3.** Oligo-tagged antibodies used in the CITE-seq experiment.

Target	Clone	Vendor
CD11b	M1/70	BioLegend
C-kit	2B8	BioLegend
Integrin $\beta$ 7	FIB504	BioLegend
Integrin $\alpha$ E	2E7	BioLegend
CD34	SA376A4	BioLegend
IL-10R	1B1.3a	BioLegend
CD64	X54-5/7.1	BioLegend
CD44	IM7	BioLegend
P2X7R	1F11	BioLegend
CD16/32	93	BioLegend

**Table S4.** Overrepresentation analysis of upregulated genes during the resolution of lung inflammation in the persistent mucosal MCs subset (cluster 1).

Gene Set	Description	Size	Overlap	Expect	Enrichment ratio	P-value	FDR	User input
P00006	Apoptosis signaling	103	5	0.58	8.60	1.46e-4	0.016	Hspa8; Jun; Nfkb1a; Birc3; Hspa1b
P06664	Gonadotropin-releasing hormone receptor	215	6	1.21	4.94	5.47e-4	0.03	Junb; Egr1; Nr4a1; Jund; Jun; Fosb
P00052	TGF- $\beta$ signaling	83	4	0.46	8.53	8.42e-4	0.031	Junb; Jund; Jun; Skil
P00054	Toll receptor signaling	36	2	0.20	9.84	0.016	0.46	Jun; Nfkb1a
P00031	Inflammation by cytokine signaling	210	4	1.18	3.37	0.02	0.54	Junb; Jund; Jun; Nfkb1a
P00010	B cell activation	54	2	0.30	6.56	0.03	0.66	Jun; Nfkb1a
P00053	T cell activation	64	2	0.36	5.53	0.04	0.77	Jun; Nfkb1a
P00049	Parkinson disease	85	2	0.47	4.16	0.08	1	Hspa8; Hspa1b
P00020	FAS signaling	26	1	0.14	6.81	0.13	1	Jun
P00017	DNA replication	31	1	0.17	5.71	0.16	1	H3f3b



**Table S5.** Antibodies used for immunofluorescence microscopy of mouse lung tissues.

<b>Primary Antibodies</b>					
<b>Target</b>	<b>Host</b>	<b>Dilution</b>	<b>Vendor</b>	<b>Catalog number</b>	<b>Conjugation</b>
mMCP-1	Rat	1:200	Thermo Fisher	14-5503-82	Unconjugated
mMCP-4	Rabbit	1:500	Gift (29)	Whole rabbit serum	Unconjugated
mMCP-6	Rabbit	1:500	R&D systems	MAB3736	Unconjugated
mMCP-8	Rat	1:200	BioLegend	647402	Unconjugated
Integrin $\beta 7$	Rat	1:50	BD	551082	APC
IL-17RA	Rabbit	1:200	Thermo Fisher	PA5-101326	Unconjugated
VEGFA	Rabbit	1:400	Thermo Fisher	MA5-32038	Unconjugated
Ki-67	Rat	1:1000	Thermo Fisher	14-5698-82	Unconjugated
AHR	Rat	1:200	Thermo Fisher	53-5925-80	Alexa 488
Avidin	-	1:2000	Vector laboratories	A-2001	Fluorescein
<b>Secondary Antibodies</b>					
<b>Target</b>	<b>Host</b>	<b>Dilution</b>	<b>Vendor</b>	<b>Catalog number</b>	<b>Conjugation</b>
Rat	Donkey	1:500	Thermo Fisher	A48269	Alexa 488
Rat	Donkey	1:500	Thermo Fisher	A48272	Alexa 647
Rabbit	Donkey	1:500	Thermo Fisher	A10042	Alexa 586