Appendix Table 4. Annotation rationale
Metallothionein, FolR2 - Folate Receptor
macs - macrophages

Dataset	Cluster Annotation
Morse, Oxford BAL	**FABP4^{hi} AM**
Morse, Oxford BAL	**IGF1^{+} AM**
Morse, Oxford BAL	**Cycling Macs**
Morse	**IntmAM**
Morse, Oxford BAL	**CD206^{hi} FN1^{hi} AM**
Morse	**MT mac**
Morse	**CXCL10^{+} AM**
Morse	**IM**
Morse	**SPP1^{+} mac**
Morse	**HSP mac**
Morse	**monoMac**
Morse	**cMono**
Morse	**ncMono**
Morse	**DC**

Morse	**FOLR2^{+} IM**
Morse	**Intm AM1**
Morse	**MonoMac1 and MonoMac2**
Morse	**Mac UD**
Oxford BAL	**Mono-SPP1^{+} mac**
Oxford BAL	**cDC**
Oxford BAL	**pDC**

References:

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3. Morse, C. *et al*. Proliferating SPP1/
4. Mould, K.J. *et al*. Airspace Macroph
5. Li, X. *et al*. ScRNA-seq expression
6. Mulder, K. *et al*. Cross-tissue singl
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8. Chakarov, S. *et al*. Two distinct inte
9. Gibbings, S.L. *et al*. Three Unique I

and basis. AM - Alveolar macrophages; IM - interstitial macrophages, Intm - intermediate, MT - 2; SPP1 - Secreted Phosphoprotein 1 (Osteopontin); HSP - heat shock protein, Mono - monocytes,

Cluster defining transcripts and rationale for annotation including associated literature references

Cells express AM transcripts (as previously defined [1,2,3,4,5], including FABP4, CD52, C1QA, C1QB, SERPING1, SERPINE1, APOE, APOC1, CES1, INHBA, MRC1 RBP4). Display highest expression of

Cells express AM transcripts with a profile similar to FABP4^{hi} AM, but with additional and distinctly higher expression of certain AM associated genes including IGF1, IFIT 27, INHBA. On pathway enrichment noted to have increased cytokine signalling but reduced lipid and cholesterol homeostasis

Subset defined by expression of cell cycling transcripts including STMN1, TUBB, HIST1H4C, MKI67, TOP2A, CENP4 and expressing macrophage markers.

Intermediate AM - cluster expressing AM transcripts, but lower expression of mature AM marker compared to FABP4^{hi} AM (FABP4, SERPINEG1, SERPINE1, APOE, APOC1, CD52, C1QA, C1QB), and higher expression VSIG1, MARCO, ALDH2 and AGRP observed in monocyte and interstitial (tissue) macrophage populations. Likely represent AMs that have recently differentiated from monocyte and

AM population with lower expression of mature AM markers compared to FABP4^{hi} AM and IGF1^{+}1 AMs. Similar expression profile to Intm AMs with higher expression of VSIG, MARCO, ALDH2 and AGRP but distinct from Intm AM due to high expression of MRC1 and FN1 compared to other

Alveolar macrophage population defined by expression of Metallothionine (MT) transcripts as observed by others[1,4,5,6,7]. MT expression associated with exposure to scavenging and metabolism of heavy

Macrophage population with expression of AM transcripts and high expression of ISGs notably IFIT1,2,3, IFITM3 and Interferon related chemokines including CXCL10. Also express IDO1. A comparable population was described[6] and observed in other BAL scRNA seq studies[1,4,5].

Interstitial macrophages - macrophage population lacking expression of AM markers, however expressing markers typically associated with Interstitial macrophages including LYVE1, FOLR2 [2,6,7,8,9]. Notably lower expression of extracellular matrix associated proteins including osteopontin (SPP1) and

Macrophage population lacking expression of AM markers, expressing interstitial macrophage markers (at lower levels compared to IMs) and extracellular matrix associated markers notably SPP1, MMPs,

Heat Shock Protein-expressing macrophage population. Lacks expression of mature AM markers, expressing markers associated with interstitial macrophages but with high expression of transcripts

Macrophage population expressing markers associated with both monocytes and interstitial macrophages including low expression of C1Q transcripts typically absent in monocytes. Likely Population expressing established circulating classical monocyte-associated transcripts including S100 proteins S100A8/9/10/12, VCAN, FCN1, CORO1A. Low or minimal expression of transcripts Population with lower expression of classical monocyte associated transcripts compared to classical monocytes but increased expression of established non classical monocyte markers including FCGR3A Myeloid population expressing transcripts associated with DCs, notably cDC2 (typically larger lung population compared to cDC1). A separate cDC1 population was not identified therefore we have

Macrophage population with IM-associated markers as observed in IM population in the 5-study dataset. Additionally show high expression of markers associated with IM population subtype 1[2] including FOLR2 and HES1[6]. Lower expression of transcripts associated with extracellular matrix
An additional AM population to intm AM1 with a similar transcriptional profile to Intm AM; but less mature, expressing more monocyte than AM markers.
As identified in 5-study dataset, 2 populations of macrophages expressing transcripts associated with both monocytes and tissue macrophage populations.
A population of macrophages, with low expression of AM markers (distinguishing them from AM_UD present in 5-study dataset). However gene transcripts not otherwise easily attributable to any defined
Population present in BAL sample only but lacks expression of AM-associated transcripts. Express transcripts associated with tissue macrophage populations and classical monocytes. Potentially represent monocytes and tissue macrophage populations collected during harvesting or very recently migrated into the alveolar compartment. Comparable populations described in single cell RNA seq
Population expressing established DC markers but also high usage of iGEP monocyte programme. Likely
Population expressing established pDC markers but also high usage of iGEP monocyte programme. Like

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