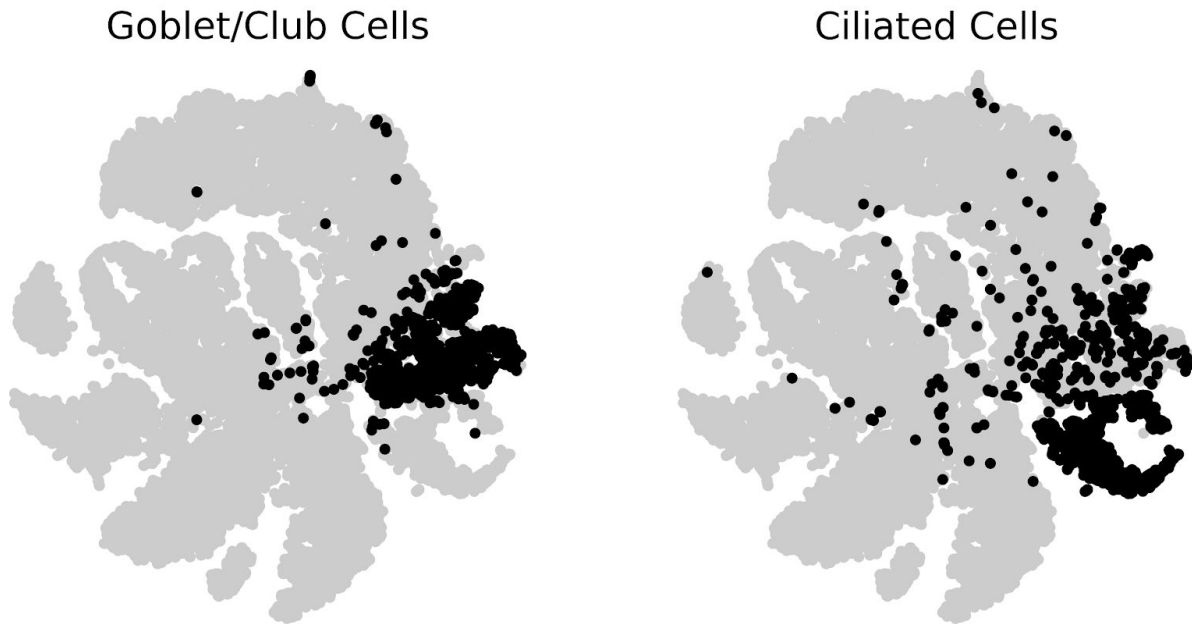


Supplementary Figures

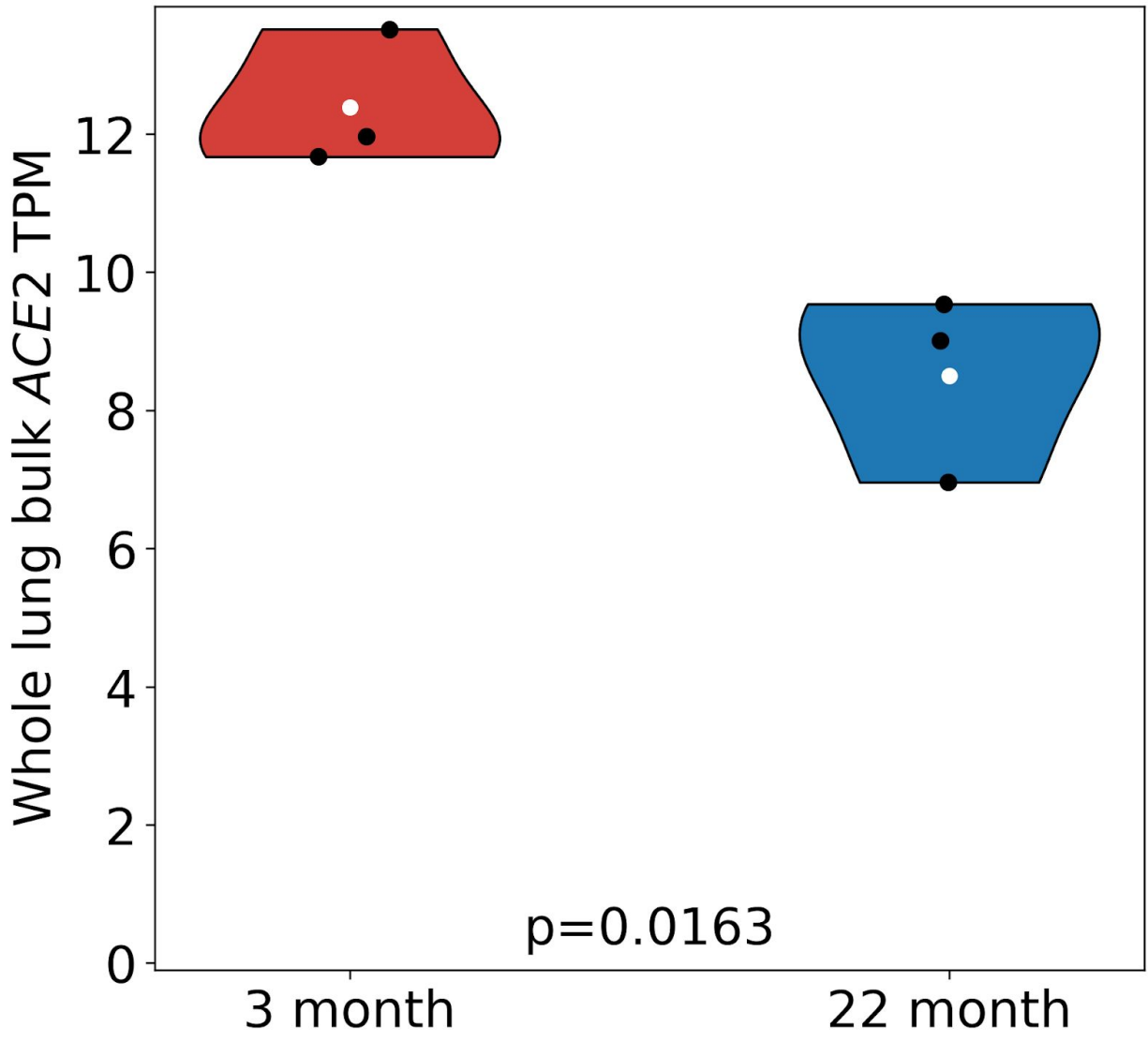
Decrease in *ACE2* mRNA expression in aged mouse lung

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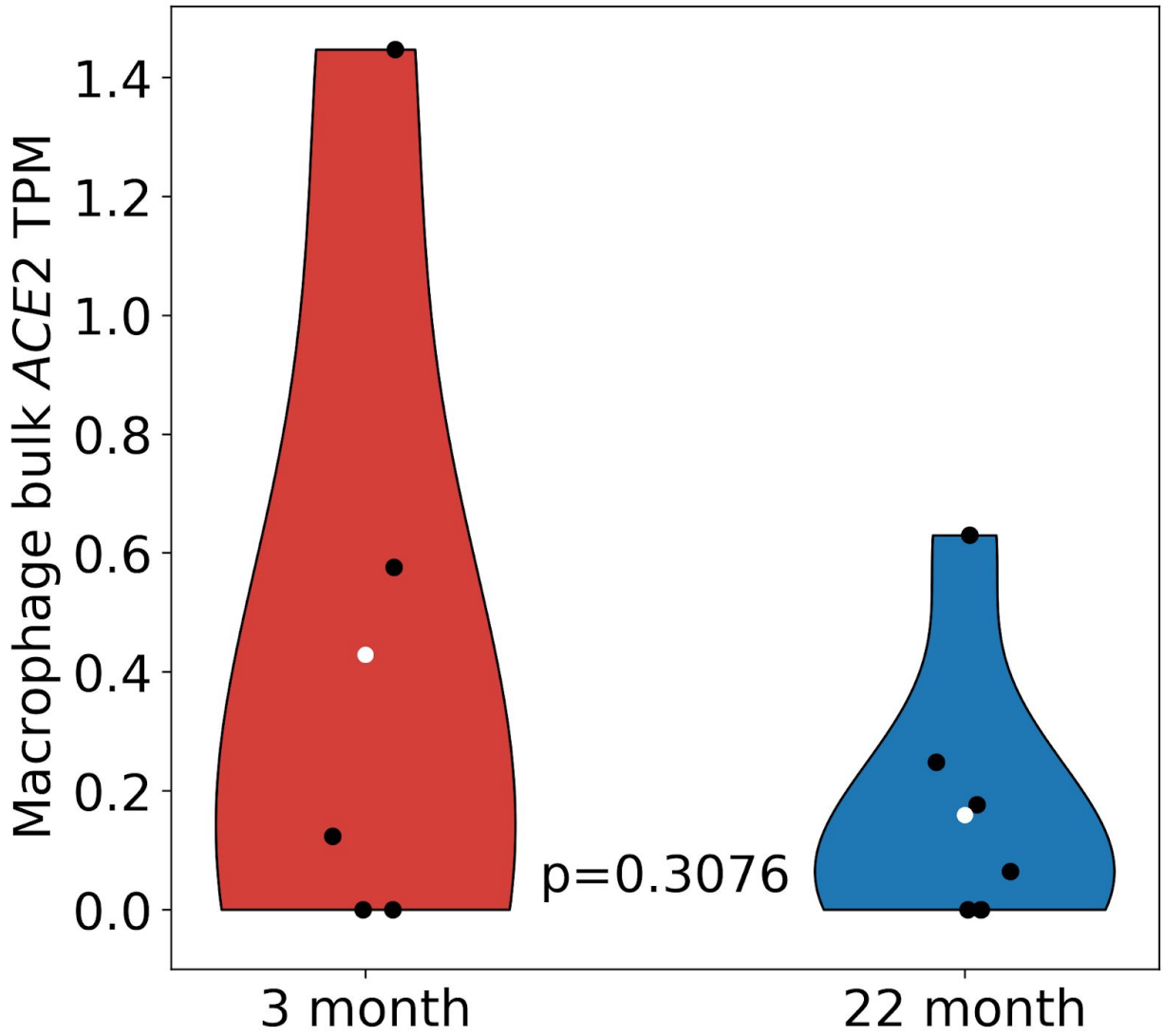
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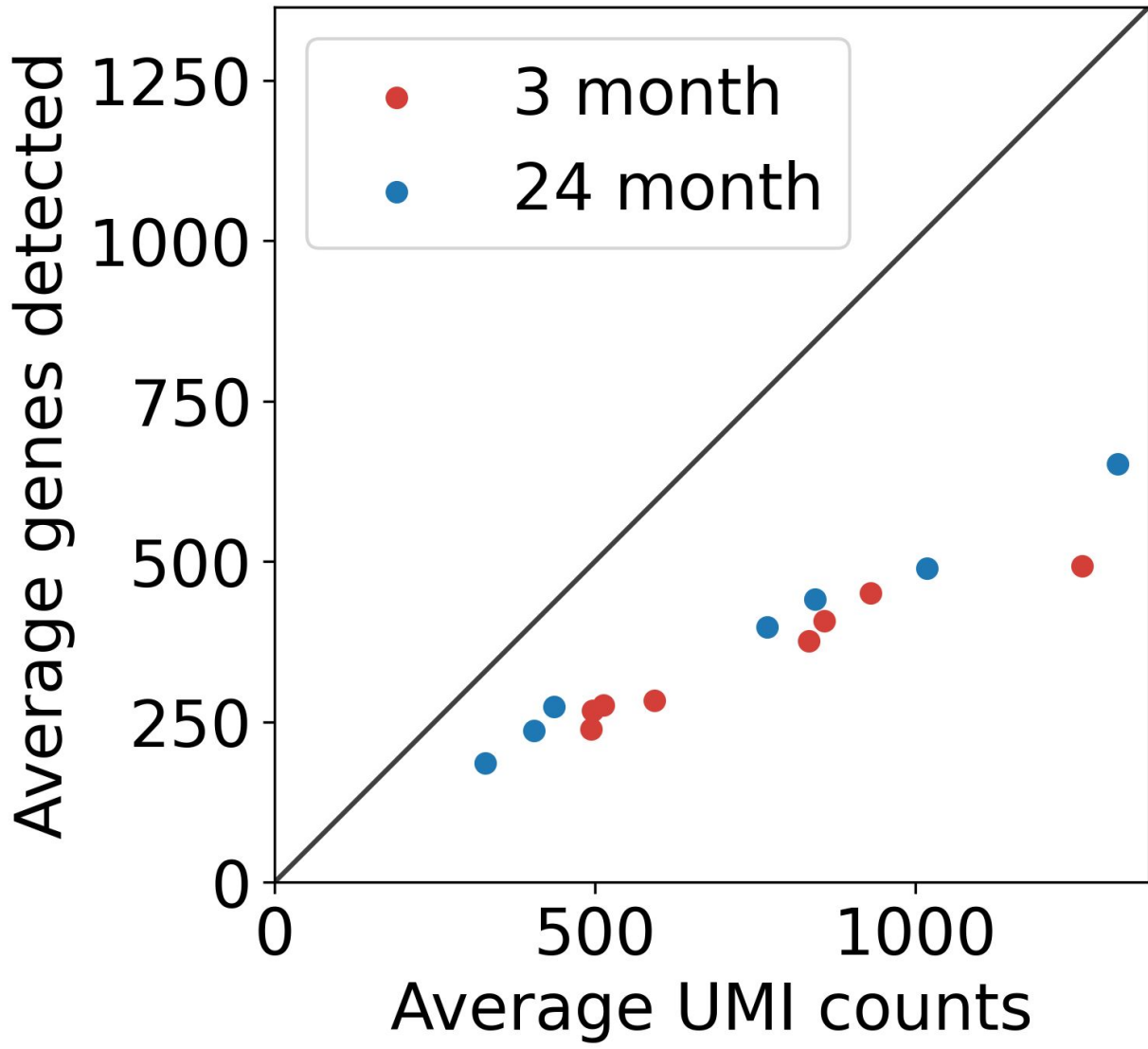
Supplementary Figure 1: t-distributed Stochastic Neighbor Embedding of 10 scVI components showing cell type in black.



Supplementary Figure 2: Comparison of *ACE2* mRNA expression in 3-month old and 22-month old whole lung bulk RNA-seq.

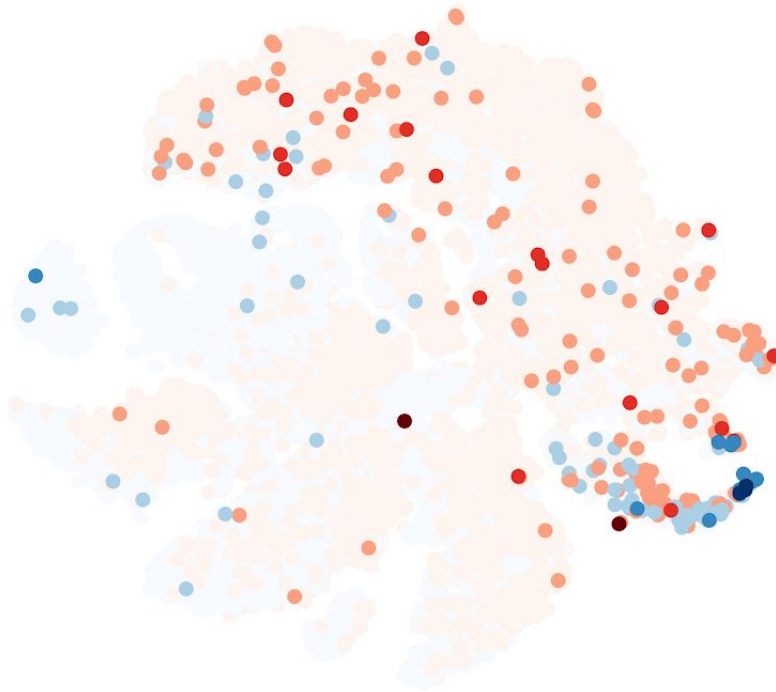


Supplementary Figure 3: Comparison of *ACE2* mRNA expression in 5 3-month old and 6 22-month old alveolar macrophage bulk RNA-seq.

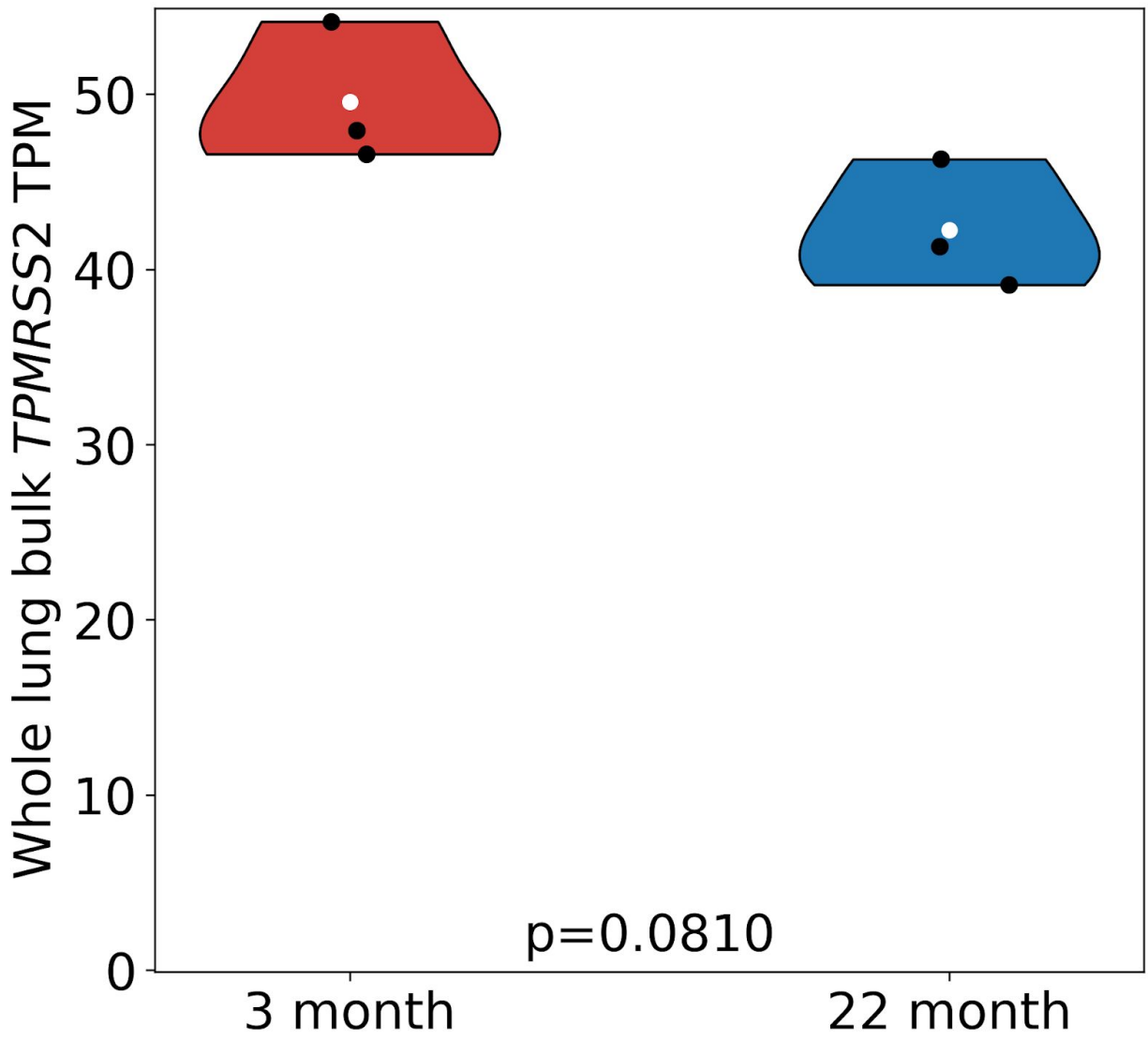


Supplementary Figure 4: Average number of UMIs and genes detected per cell colored by sample. There is no significant difference between 3-month old and 24-month old samples .

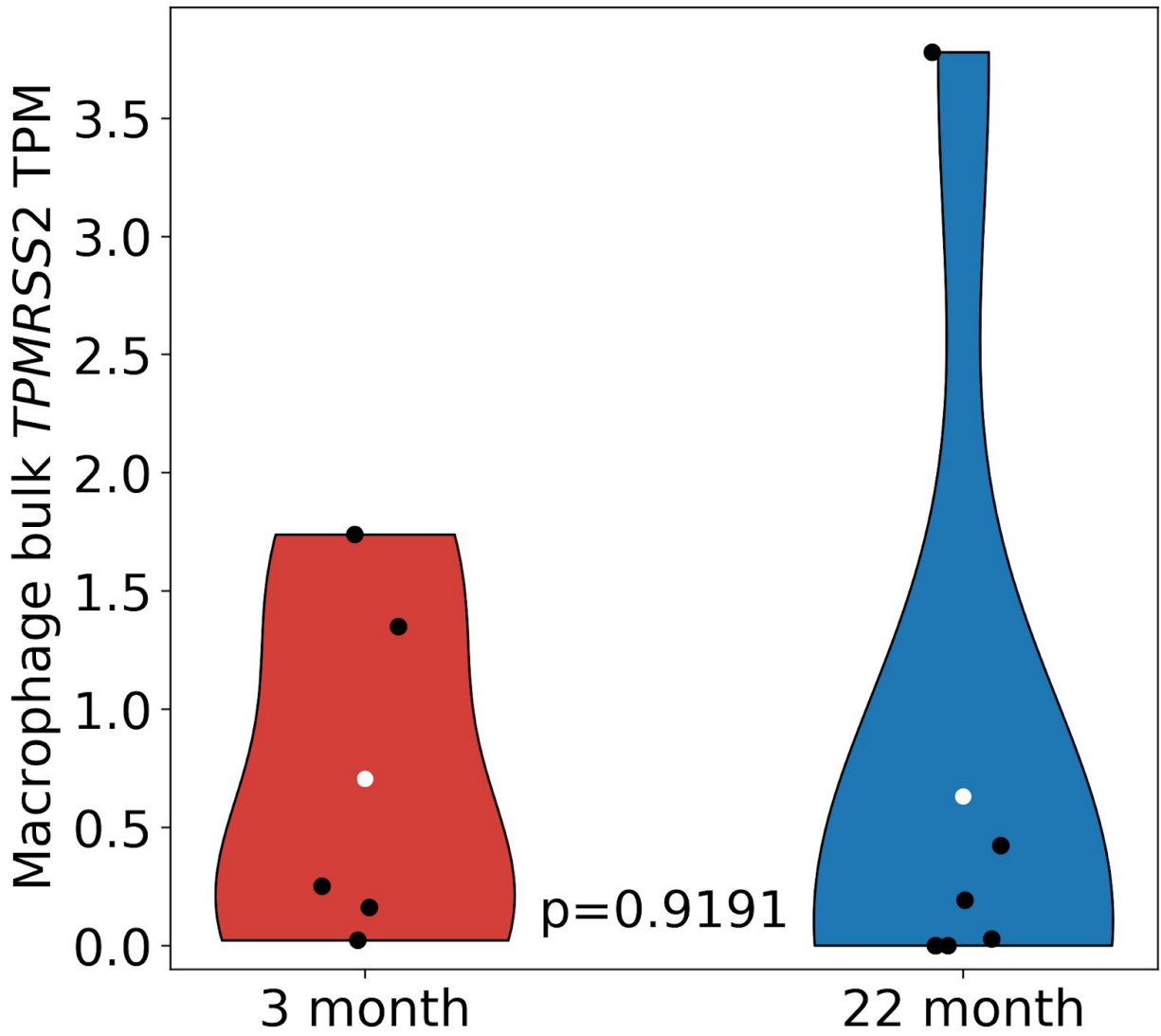
TMPRSS2 expression



Supplementary Figure 5: t-distributed Stochastic Neighbor Embedding showing cells that express *TMPRSS2* (3-month old cells in red, 24-month old cells in blue).



Supplementary Figure 6: Comparison of *TPMRSS2* mRNA expression in 3-month old and 22-month old whole lung bulk RNA-seq.



Supplementary Figure 7: Comparison of *TPMRSS2* mRNA expression in 5 3-month old and 6 22-month old alveolar macrophage bulk RNA-seq.

Supplementary Table 1: Claims that *ACE2* mRNA expression does not decrease with age.

Dataset	Claim	Refutation
GTEX ¹	<p>Chen <i>et al</i>.²: “<i>ACE2</i> expression generally decreases with age significantly or insignificantly.”</p> <p>Smith and Sheltzer³: “<i>ACE2</i> expression [in GTEX] was equivalent between...young individuals (<29 years) and elderly individuals (>70 years).”</p>	Multiple confounders that are difficult to correct for; RNA from cadavers may be of degraded quality; despite these issues evidence of slight decrease in <i>ACE2</i> expression with age.
Rat BodyMap ⁴	Smith and Sheltzer ³ : “...young rats (6 weeks) and elderly rats (104 weeks) [displayed equivalent levels of <i>ACE2</i> expression in the lung]”	Single-end 50bp bulk total RNA was quantified with Cufflinks v2.0.2 ⁵ (2012) which did not perform multiple rounds of EM for reads ambiguous between genes. <i>ACE2</i> is a member of a gene family. Furthermore, the Ribo-zero requires careful normalization.
Bulk RNA-seq from mice alveolar macrophages ⁶	We find that 22-month samples have increased expression of <i>ACE2</i> on average (0.47 fold change young/old p=0.1198). (Methods)	Increased <i>ACE2</i> expression in 24-month lung in FACS sorted bulk Epithelium (Club/Ciliated) is partly explained by different proportions of club/goblet cells to ciliated cells in comparison with proportions from single-cell RNA-seq (Figure 1c).

References

1. GTEx Consortium. The Genotype-Tissue Expression (GTEx) project. *Nat. Genet.* **45**, 580–585 (2013).
2. Chen, J. *et al.* Individual Variation of the SARS-CoV2 Receptor ACE2 Gene Expression and Regulation. (2020).
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5. Trapnell, C. *et al.* Transcript assembly and quantification by RNA-Seq reveals unannotated transcripts and isoform switching during cell differentiation. *Nat. Biotechnol.* **28**, 511–515 (2010).
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