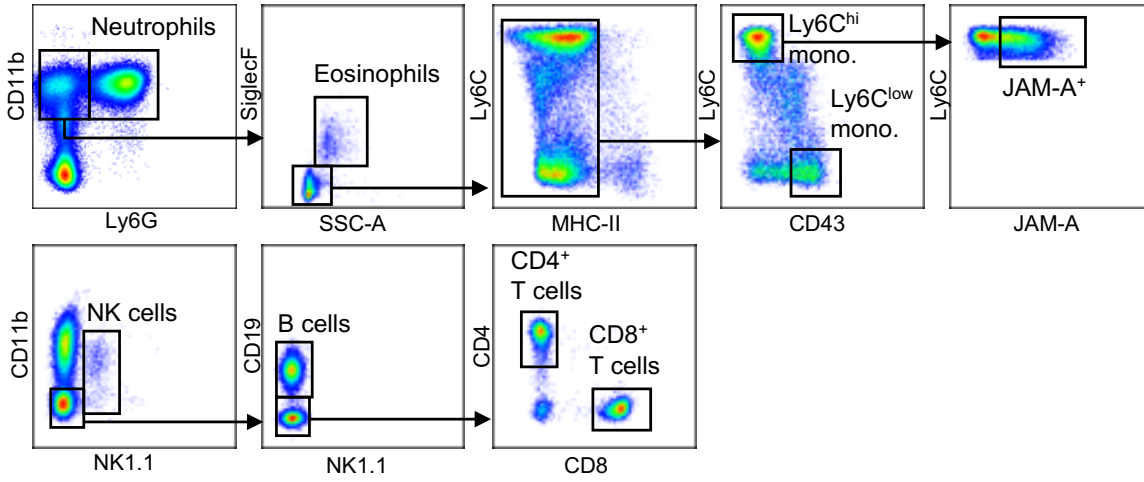
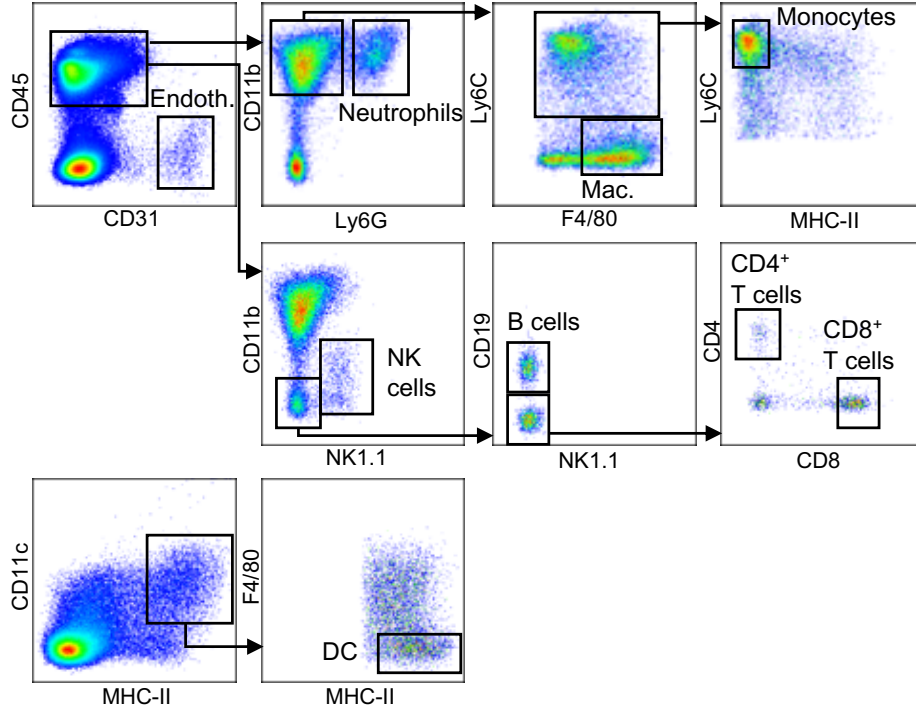


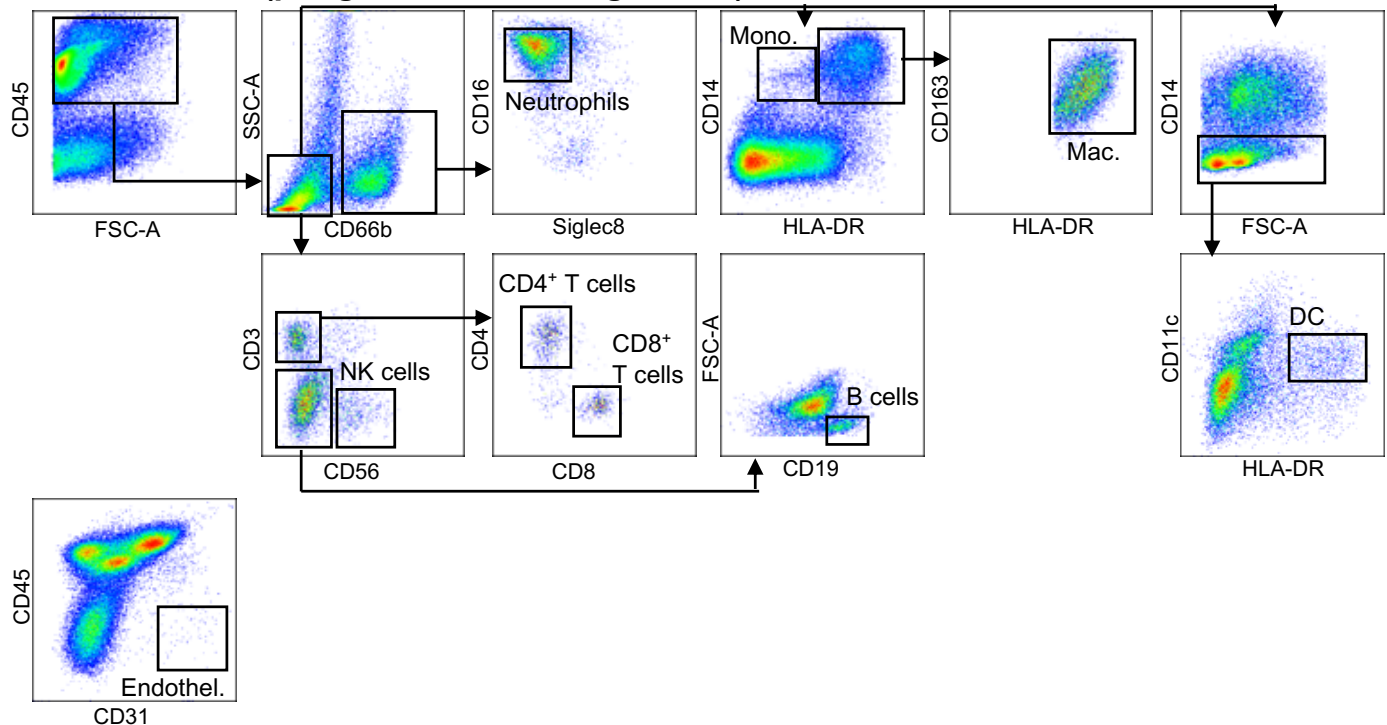
### Mouse blood (pre-gated on live, single cells)



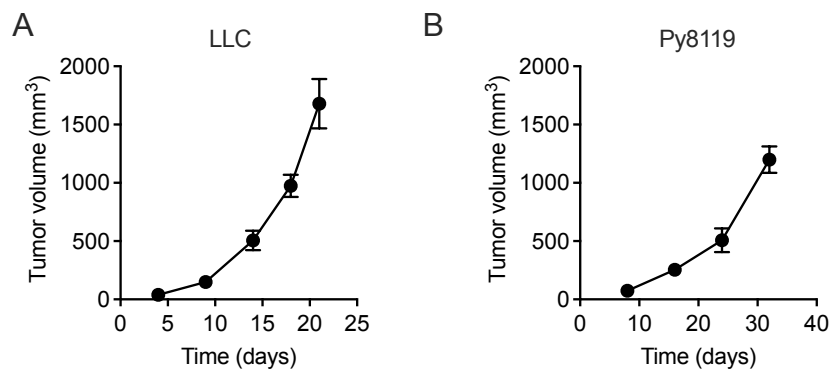
### Mouse tumor (pre-gated on live, single cells)



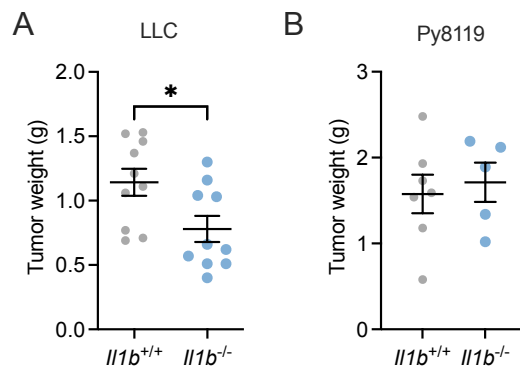
### Human tumor (pre-gated on live, single cells)



Supplementary Figure 1. Flow cytometry gating strategies used to identify cell populations



**Supplementary Figure 2. Tumor volumes at the time points shown in Figure 1E**  
(A) Tumor volume of sc. LLC tumors (n=6) from experiment shown in Figure 1E  
(B) Tumor volume of orthotopic Py8119 tumors (n=6) from experiment shown in Figure 1E  
Graphs show mean and SEM.

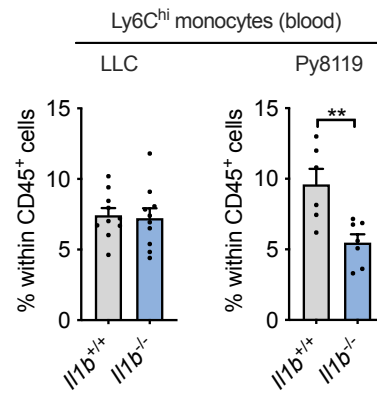


**Supplementary Figure 3. Tumor burden in *Il1b*<sup>-/-</sup> mice**

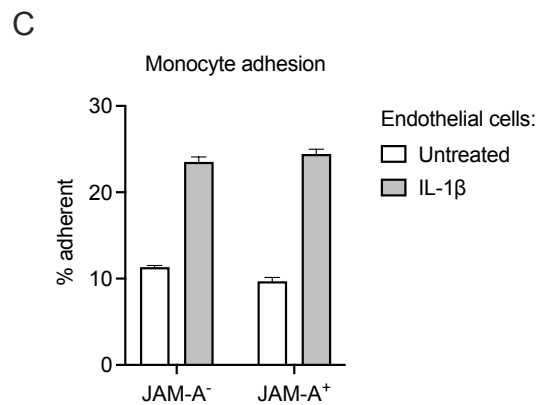
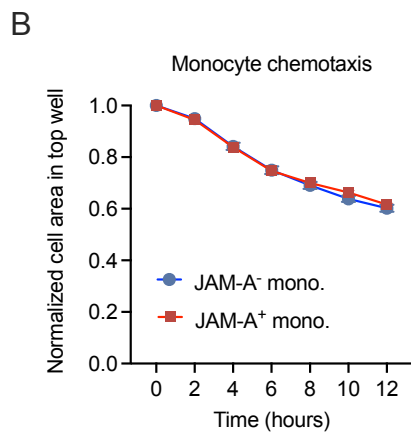
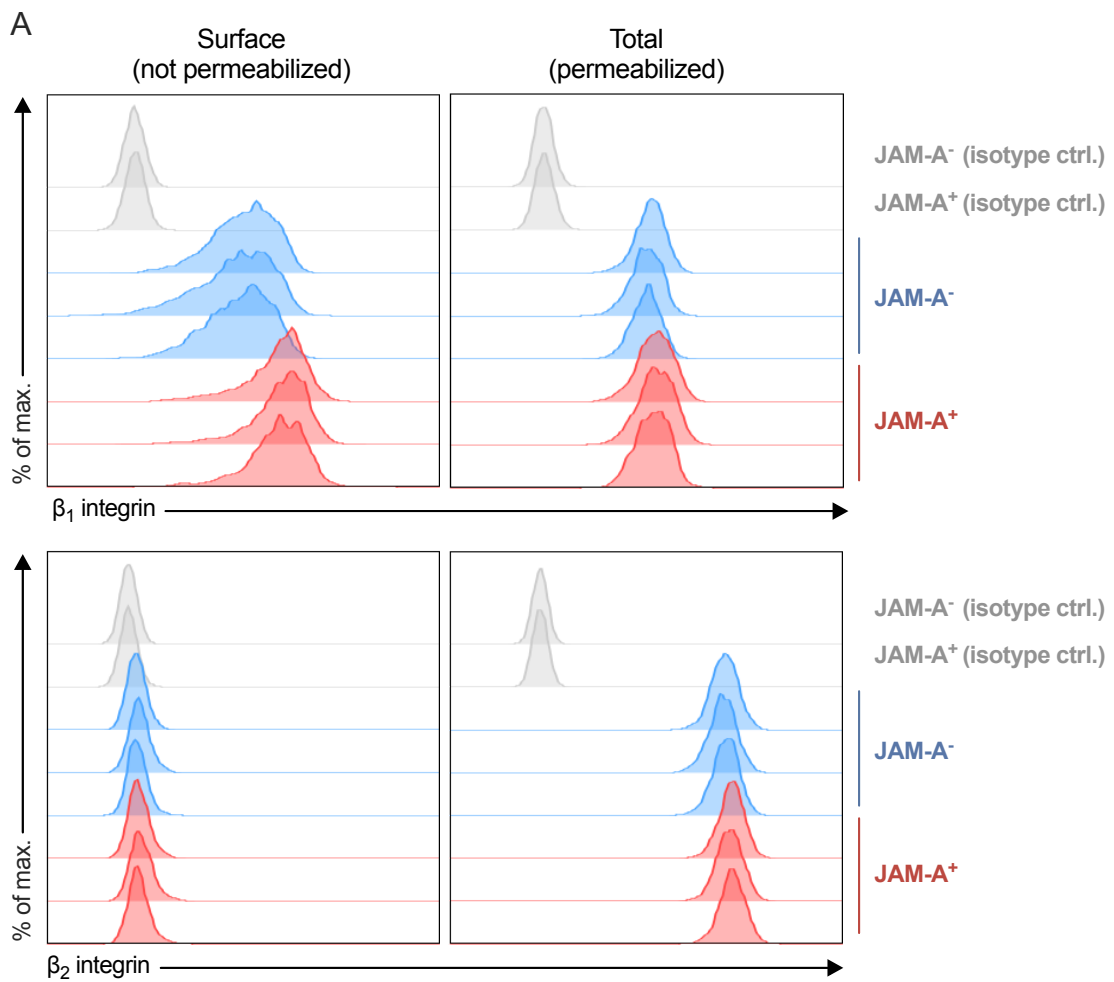
(A) LLC tumor weight in *Il1b*<sup>+/+</sup> and *Il1b*<sup>-/-</sup> mice (day 19 post-engraftment).

(B) Py8119 tumor weight in *Il1b*<sup>+/+</sup> and *Il1b*<sup>-/-</sup> mice (day 35 post-engraftment).

Graphs show mean and SEM.



**Supplementary Figure 4. Ly6C<sup>hi</sup> monocyte frequency in the peripheral blood of *I11b*<sup>-/-</sup> mice**  
Frequency of Ly6C<sup>hi</sup> monocytes in the peripheral blood of LLC (n=10) and Py8119 (n=6-7) tumor bearing *I11b*<sup>-/-</sup> and *I11b*<sup>+/+</sup> mice, assessed by flow cytometry. Graphs show mean and SEM.



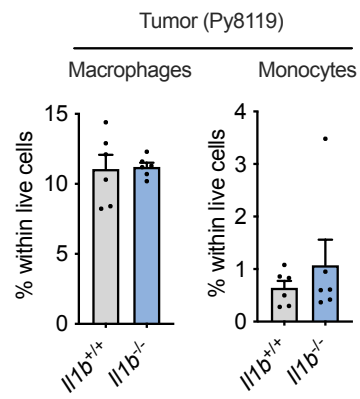
### Supplementary Figure 5. Phenotype comparison of JAM-A<sup>+</sup> and JAM-A<sup>-</sup> monocytes

(A) Expression of  $\beta_1$  and  $\beta_2$  integrins in peripheral blood JAM-A<sup>+</sup> and JAM-A<sup>-</sup> Ly6C<sup>hi</sup> monocytes from Py8119 tumor-bearing mice stained with or without membrane permeabilization (for examining total or surface levels, respectively).

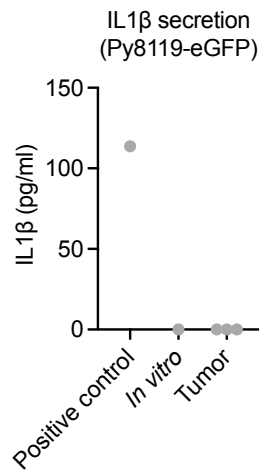
(B) Transwell migration of JAM-A<sup>+</sup> and JAM-A<sup>-</sup> Ly6C<sup>hi</sup> monocytes isolated from the blood of Py8119 tumor-bearing mice (n=8). CCL2 was used as a chemotactic stimulus in the bottom well.

(C) Adhesion of JAM-A<sup>+</sup> and JAM-A<sup>-</sup> Ly6C<sup>hi</sup> monocytes isolated from the blood Py8119 tumor-bearing mice (n=3). Proportion of adherent monocytes on untreated or IL1 $\beta$ -pretreated endothelial monolayers.

Integrin levels were assessed by flow cytometry. Graphs in panels B and C show mean and SEM.

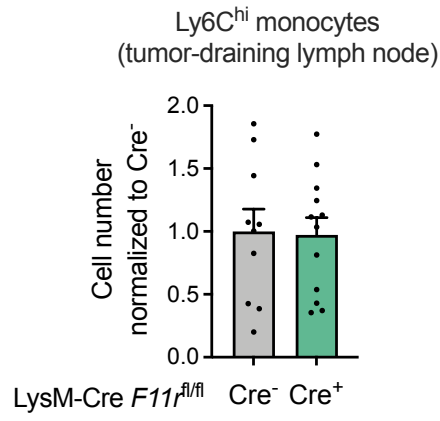


**Supplementary Figure 6. Macrophage and monocyte abundance in tumors of *Il1b<sup>-/-</sup>* mice**  
Frequency of macrophages and monocytes in Py8119 tumors of *Il1b<sup>-/-</sup>* and *Il1b<sup>+/+</sup>* mice, assessed by flow cytometry (n=6). Graphs shows mean and SEM.



**Supplementary Figure 7. Lack of IL1β secretion by Py8119 cancer cells**

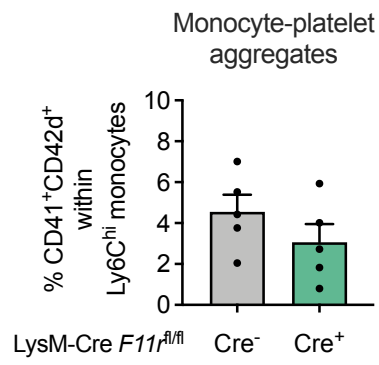
IL1β production by Py8119-eGFP cancer cells measured using ELISA. Positive control: Recombinant mouse IL1β; *In vitro*: Py8119-eGFP cell culture supernatant; Tumor: Supernatant of Py8119-eGFP cancer cells which were sorted from established orthotopic tumors and cultured for 24 hours.



**Supplementary Figure 8. Ly6C<sup>hi</sup> monocyte abundance in the tumor-draining lymph node**

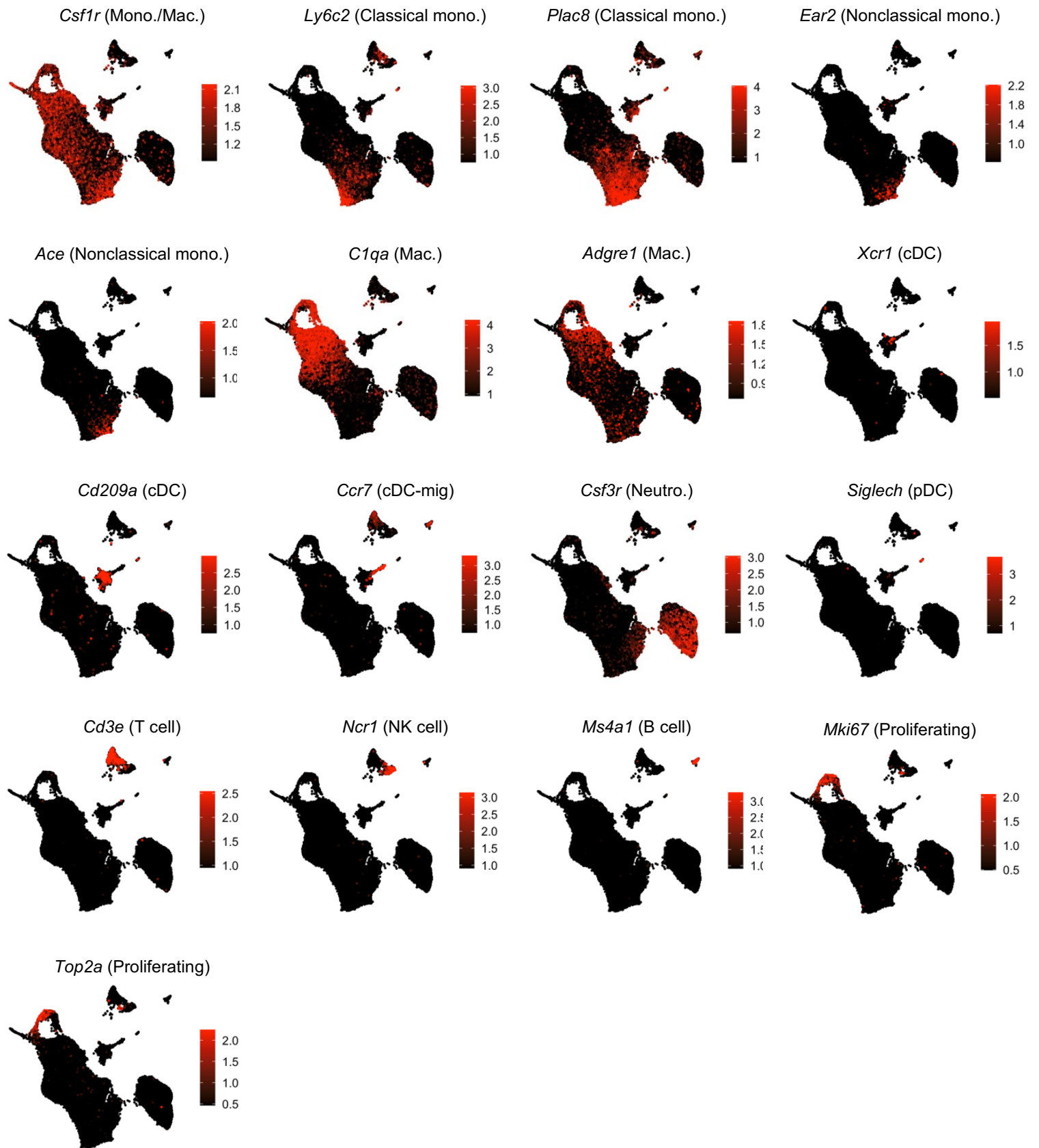
Number of Ly6C<sup>hi</sup> monocytes per tumor-draining lymph node determined by flow cytometry in LLC-eGFP tumor-bearing LysM-Cre *F11r<sup>fl/fl</sup>* mice, normalized to Cre<sup>-</sup> controls (n=10/12, data pooled from two experiments). Graph shows mean and SEM.



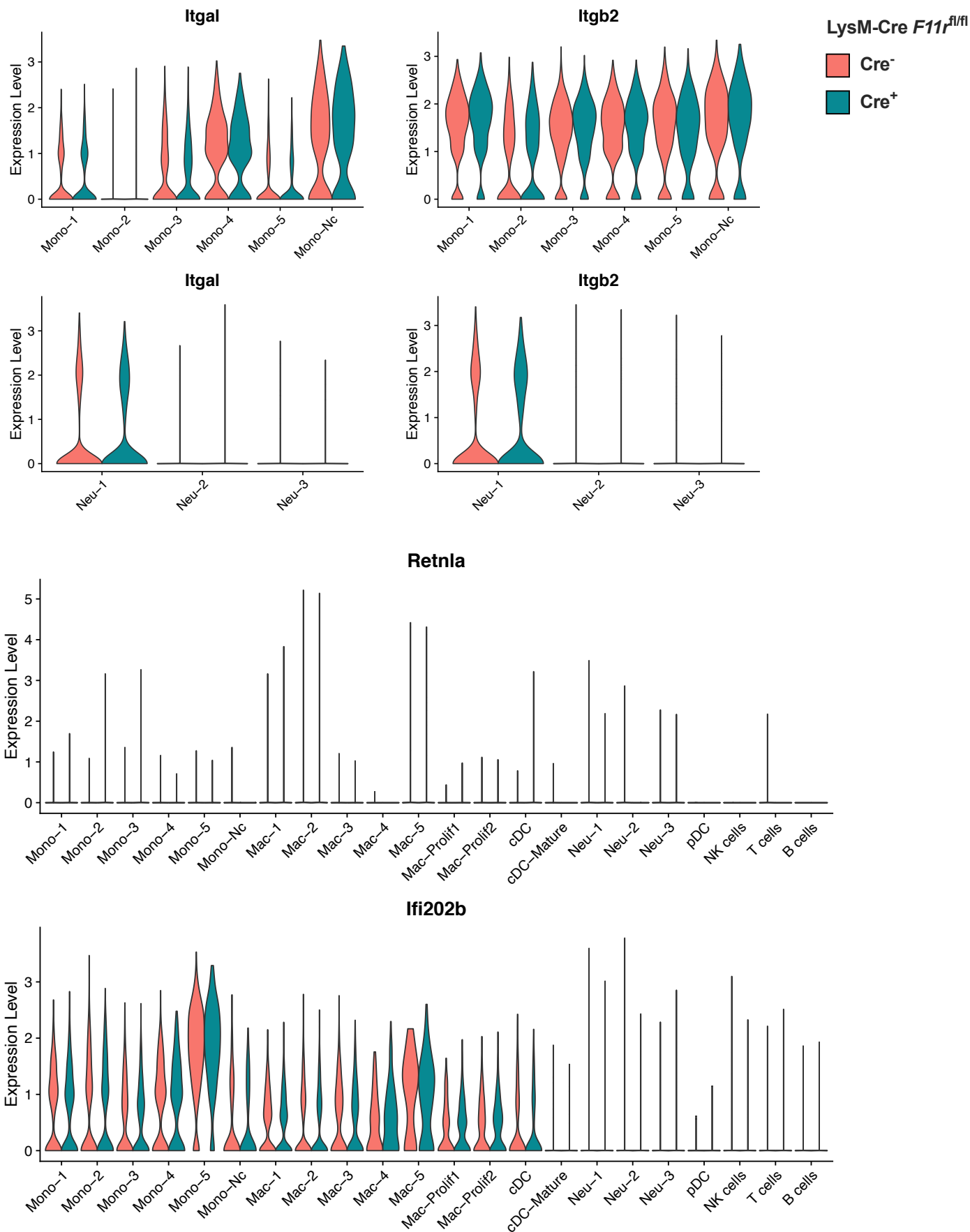


**Supplementary Figure 9. Monocyte-platelet aggregates in myeloid-specific JAM-A-deficient mice**

Frequency of monocytes in LLC-eGFP tumor-bearing LysM-Cre *F11<sup>fl/fl</sup>* Cre<sup>-</sup> and Cre<sup>+</sup> mice associated with platelets assessed by flow cytometry staining of platelet markers (CD41 and CD42d) on the cell surface. Graph shows mean and SEM.



**Supplementary Figure 10. UMAP plots colored based on expression of marker genes used to identify major cell populations in the single cell RNA-seq data**



**Supplementary Figure 11. Expression of *Itgal*, *Itgb2*, *Retnla* and *Ifi202b* in scRNAseq of LLC tumors**

Expression of  $\beta 2$  integrin subunits (*Itgal*, *Itgb2*) and previously reported JAM-A-regulated genes (*Retnla*, *Ifi202b*) across different immune cell states in LLC tumors of LysM-Cre  $F11r^{fl/fl}$  Cre<sup>+</sup> and Cre<sup>-</sup> mice.