

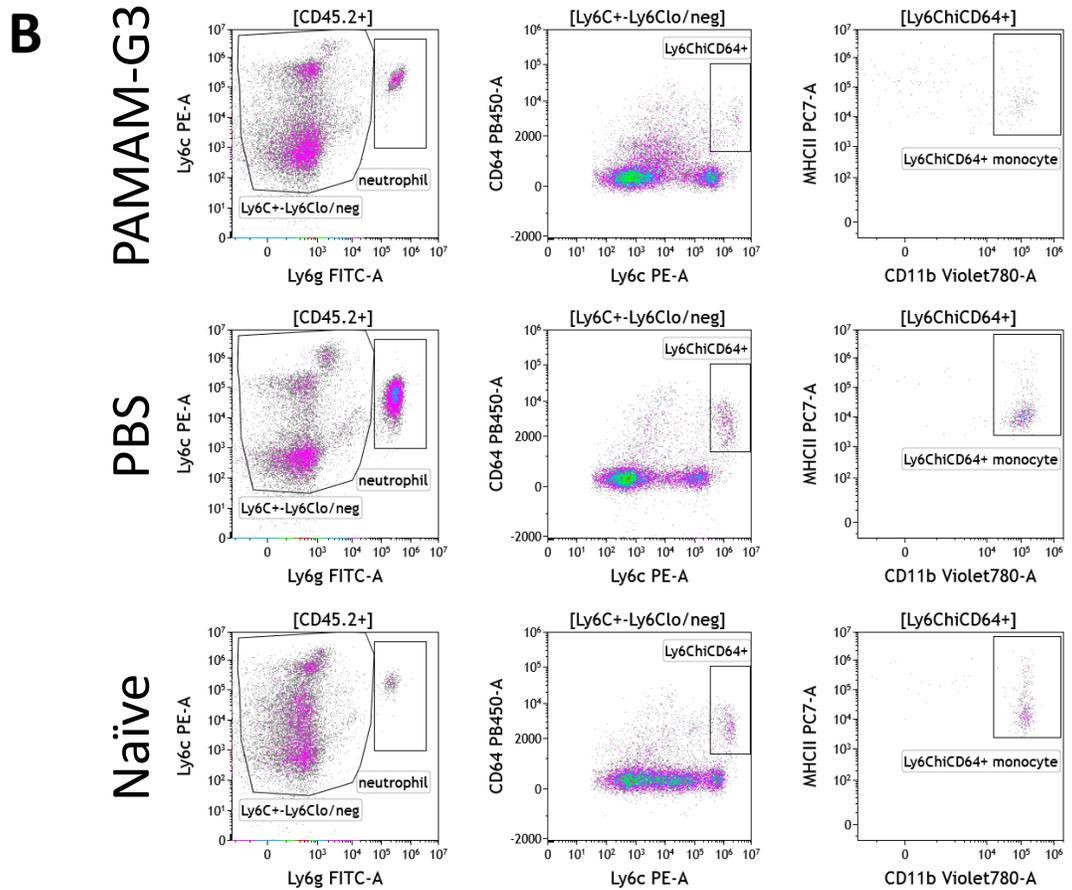
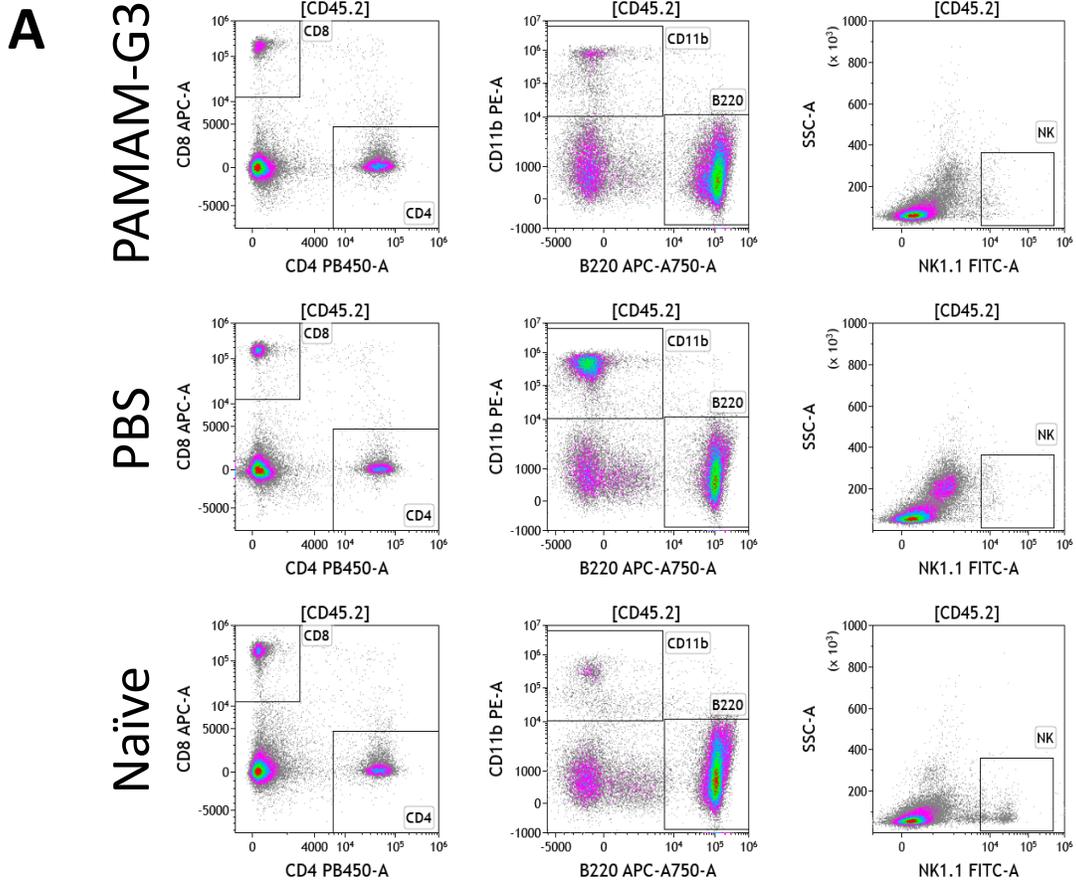
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Supplemental Information

Controlling cancer-induced inflammation with a nucleic acid scavenger prevents lung metastasis in murine models of breast cancer

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Supplementary Figure S1



Supplementary Figure S1. Gating strategy to examine immune cell landscape in spleen of tumor-bearing mice treated with PAMAM-G3 or PBS.

The intravenous experimental lung metastasis 4T1 mouse model of breast cancer described in Figure 1 was used for this analysis. Spleens were collected from PAMAM-G3 or PBS treated tumor-bearing mice on day 15 and naïve non-tumor-bearing mice. Splenocytes were analyzed for immune cell populations as described in Figure 2 and in Methods. One representative mouse from each group is shown.

A. Immune cell populations:

CD4 T cells: CD45.2+CD4+

CD8 T cells: CD45.2+CD8+

Innate/myeloid cells: CD45.2+CD11b+

B cells: CD45.2+B220+

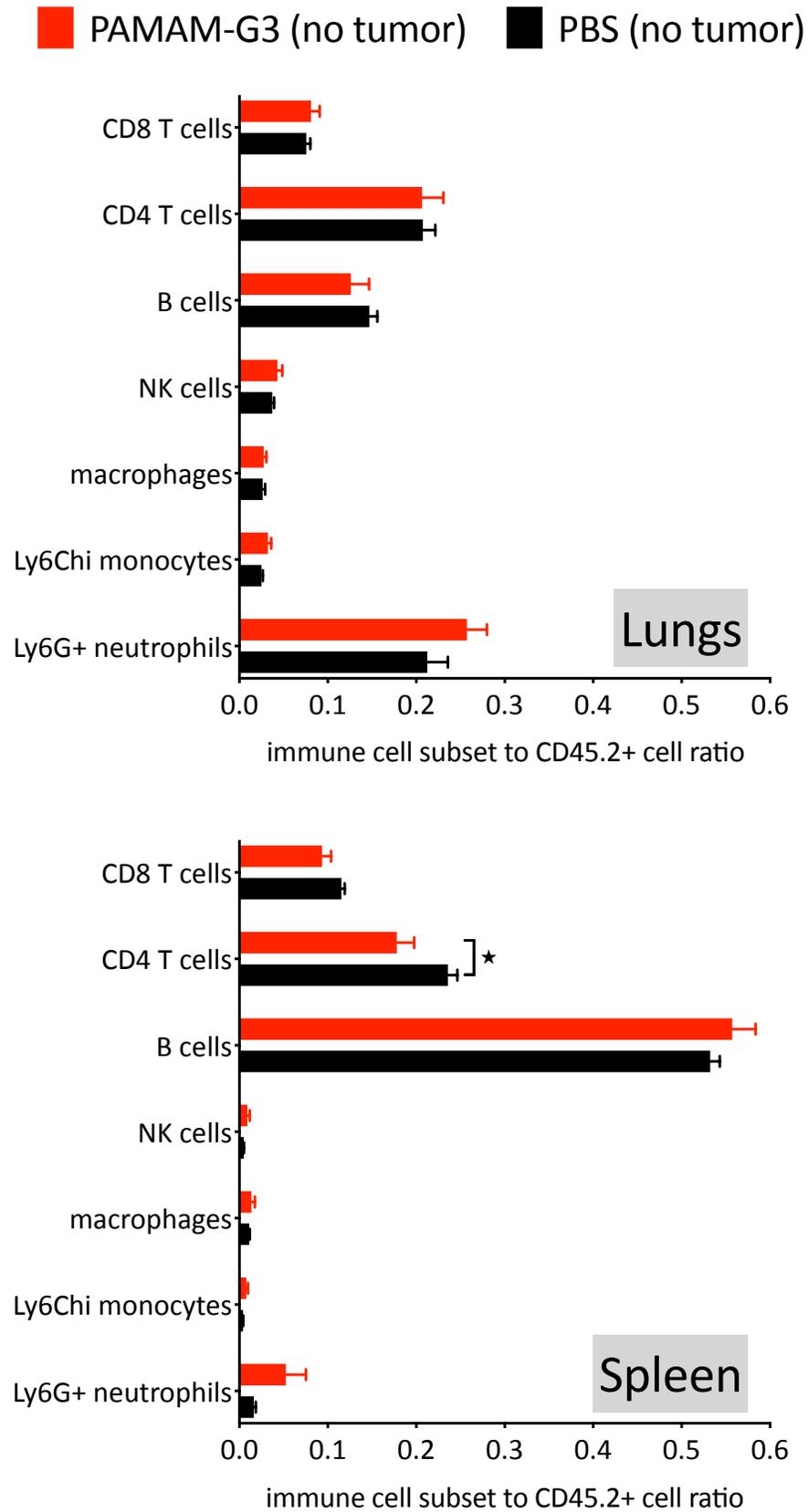
NK cells: CD45.2+NK1.1+

B. Immune cell populations:

Neutrophils: CD45.2+Ly6C+Ly6G+

Inflammatory monocytes: CD45.2+Ly6G-Ly6C^{hi}CD64+MHCII+CD11b+
(MHCII, class II MHC)

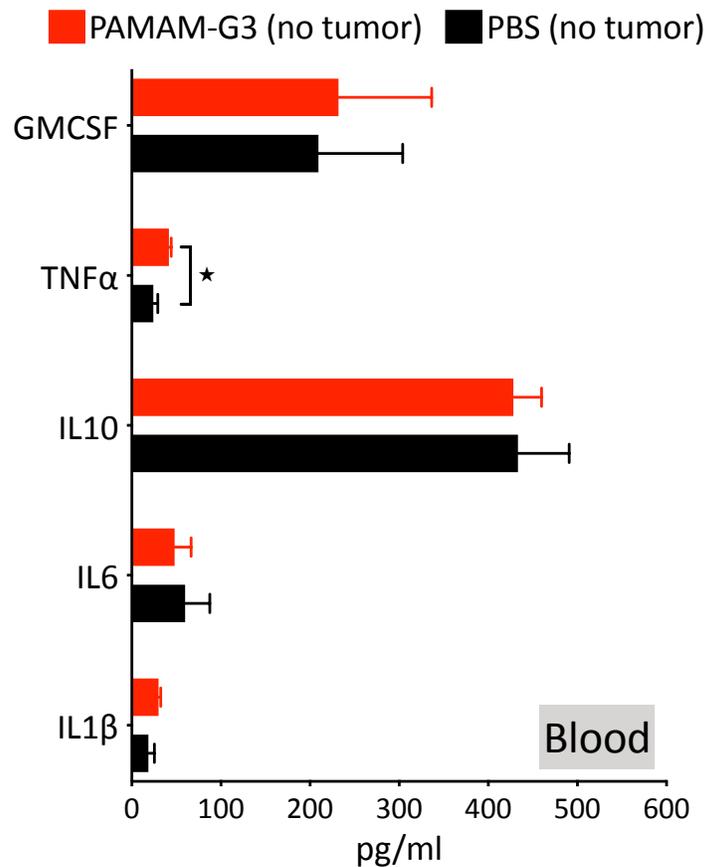
Supplementary Figure S2



Supplementary Figure S2. Immune cell landscape in lungs and spleen of naïve mice with no tumors treated with PAMAM-G3 or PBS.

Naïve female Balb/c mice with no tumors were injected intraperitoneally with PAMAM-G3 at 20 mg/kg or PBS, 6 times at 3-day interval. One day after the last injection, mice were euthanized and lungs and spleens were harvested and processed for immune cell analysis via flow cytometry. Immune cell subsets in lungs and spleen were identified using the gating strategy described in Figure 2, Supplementary Figure S1 and as described in Methods. Data represent an average of 5 mice per group \pm standard error of mean (SEM). The ratio of immune cell subsets to total CD45.2+ immune cells in lungs (top) and spleen (bottom) is presented in the figure. * $p < 0.05$, unpaired t-test.

Supplementary Figure S3



Supplementary Figure S3. Circulating pro-inflammatory cytokine levels in blood of naïve mice with no tumors treated with PAMAM-G3 or PBS.

Naïve female Balb/c mice with no tumors were injected intraperitoneally with PAMAM-G3 at 20 mg/kg or PBS, 6 times at 3-day interval. One day after the last injection, mice were euthanized and blood was harvested. Plasma was used for analysis. Analysis was conducted using the BioLegend LEGENDplex™ bead-based multiplex assay. Data represent an average of 5 mice per group \pm SEM. * $p < 0.05$, unpaired t-test.