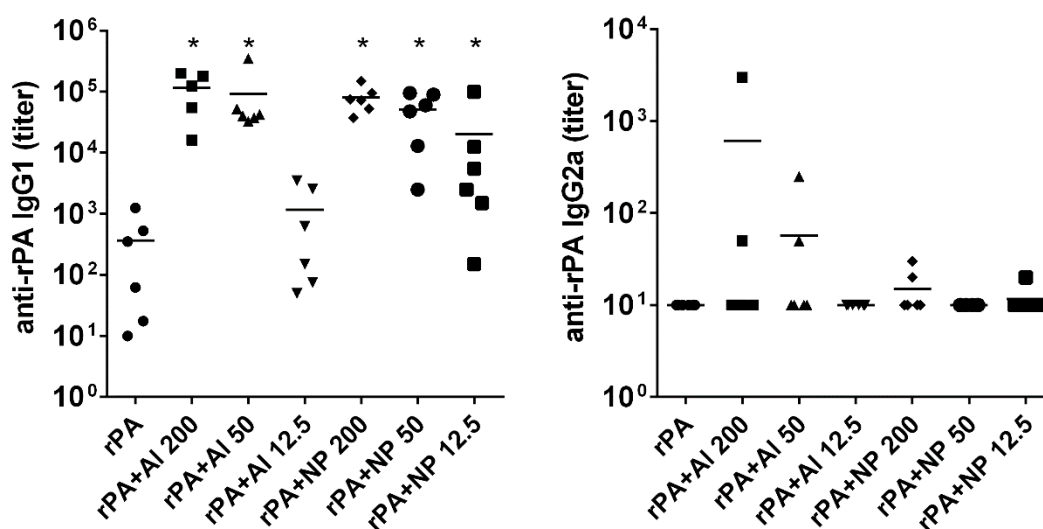


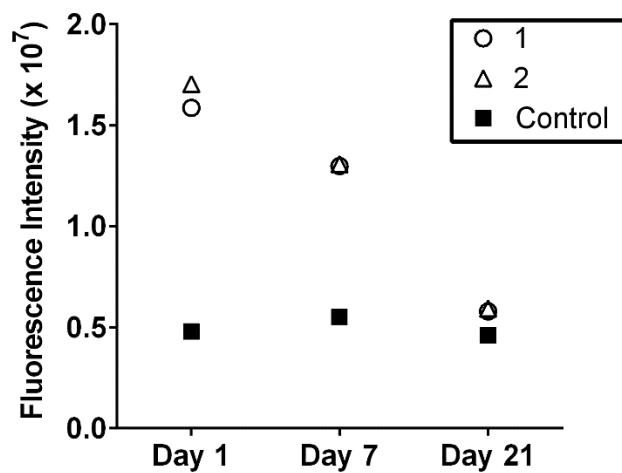
## Supporting Information

**Alpha-D-glucan Nanoparticulate Adjuvant Induces a Transient Inflammatory Response at the Injection Site and Targets Antigen to Migratory Dendritic Cells.**

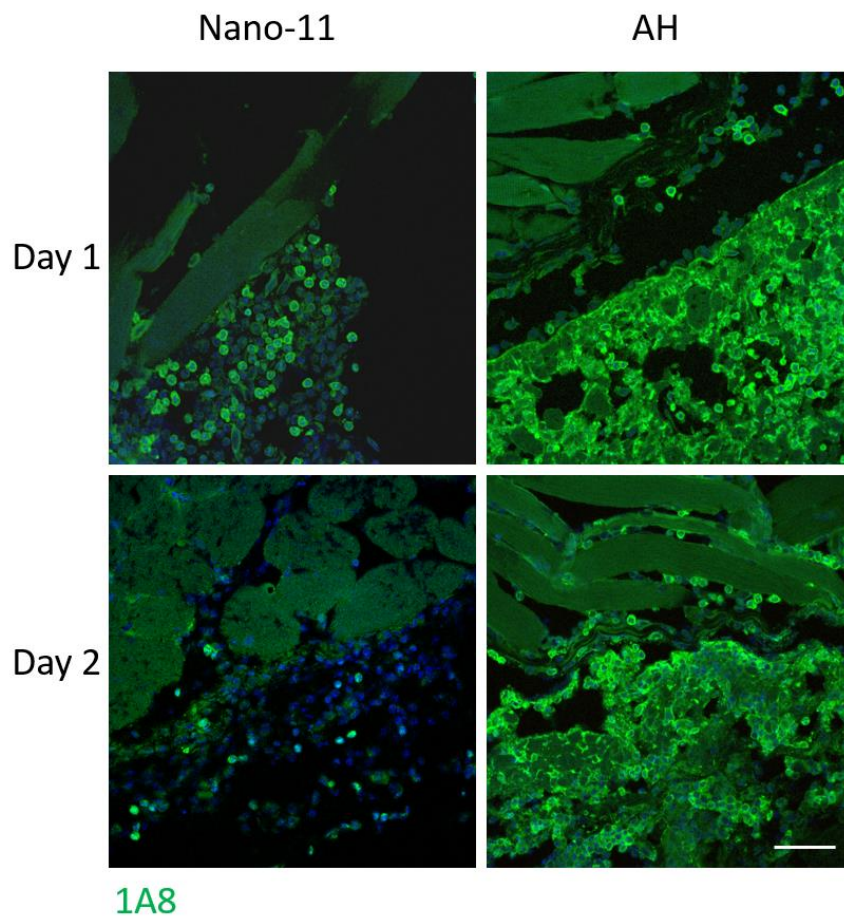
Fangjia Lu, Yung-Yi Mosley, Randol J Rodriguez Rosales, Brooke Carmichael, Srikanth Elesela, Yuan Yao, Harm HogenEsch\*



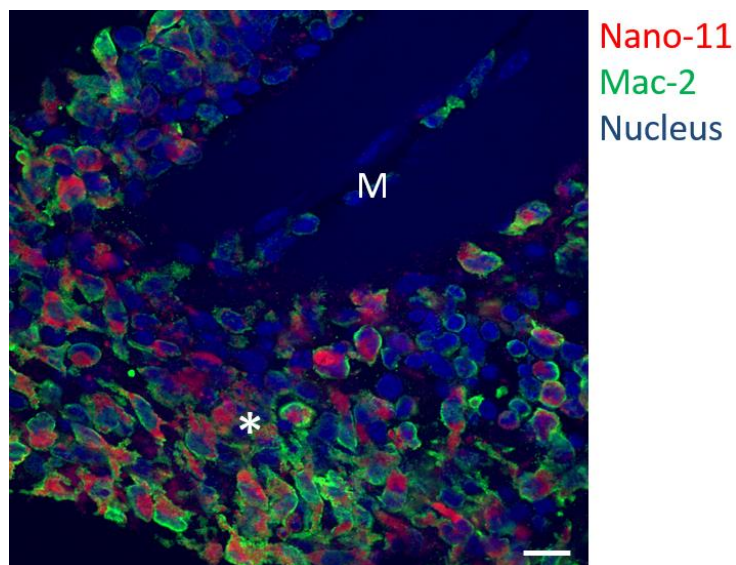
**Supplemental Figure 1.** Titers for rPA-specific IgG1 and IgG2a antibodies in serum after two injections of 2 μg recombinant protective antigen (rPA) from *Bacillus anthracis* alone or combined with either AH or Nano-11 at three different dosages (200 μg, 50 μg, and 12.5 μg). Titers were measured by ELISA. Each symbol represents a mouse and the bar indicates the geometric mean. \* p<0.05 vs. rPA alone.



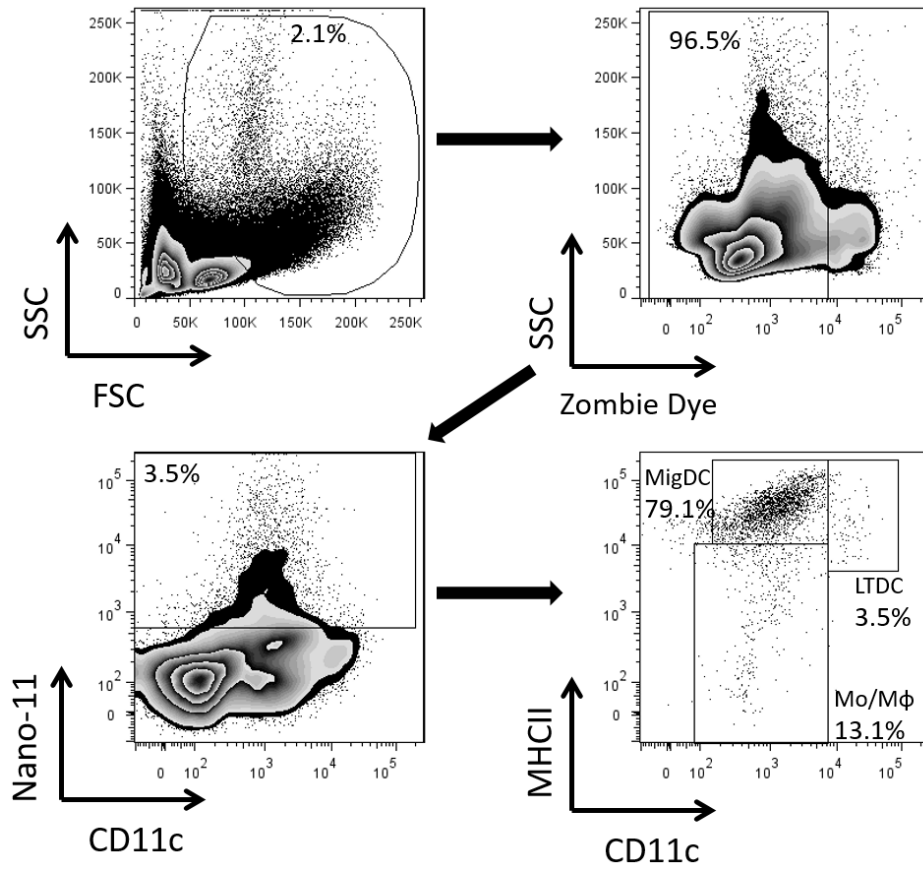
**Supplemental Figure 2.** Low levels of fluorescence in the draining (iliac) lymph node on Days 1, 7 and 21 after intramuscular injection of 200  $\mu\text{g}$  Alexa Fluor 647-labeled Nano-11 (mouse 1 and 2) or nonlabeled Nano-11 (Control) with 10  $\mu\text{g}$  OVA. Mice were euthanized at Day 1, 7, and 21 after injection, and the iliac lymph nodes were imaged.



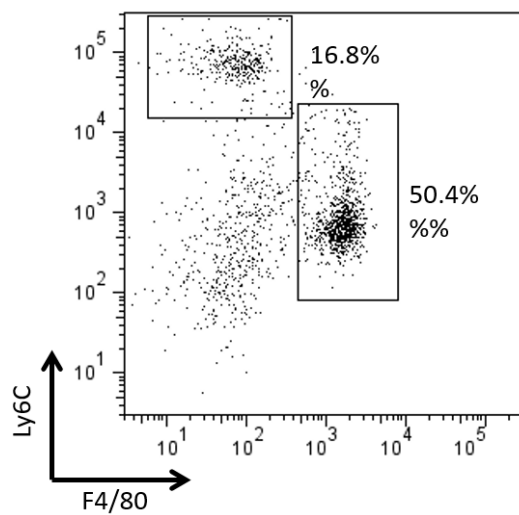
**Supplemental Figure 3.** Neutrophils at the injection sites on Day 1 and Day 2 for either Nano-11 or AH injected mice. Confocal images were taken under a 40X objective lens on a confocal microscope and are representative of four independent experiments. Bar=50 $\mu$ m



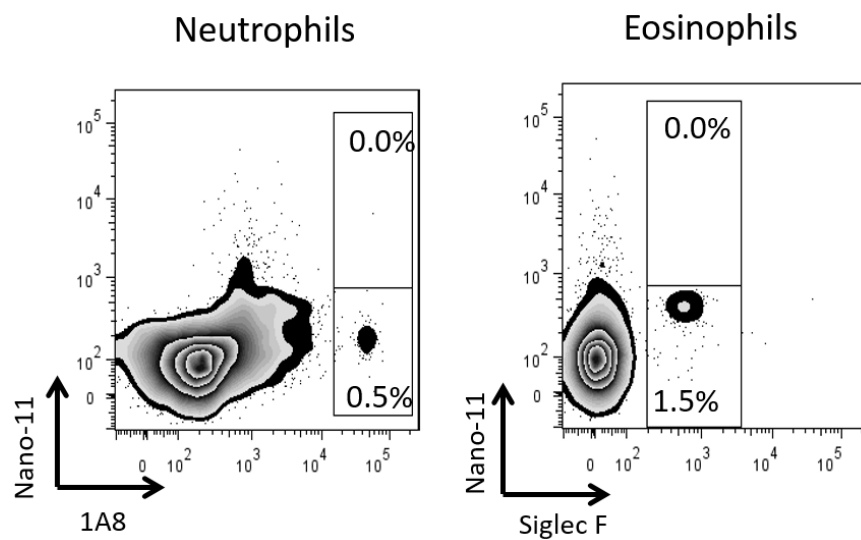
**Supplemental Figure 4.** Most Nano-11 at the injection site is inside Mac-2<sup>+</sup> cells by Day 2. Confocal images were taken under a 60X objective lens and are representative of two independent experiments. M: muscle, \*: injection site. Bar=20 $\mu$ m.



**Supplemental Figure 5.** Gating strategy for detecting and phenotyping Nano-11-containing myeloid cells in the draining lymph nodes demonstrated on a lymph node sample harvested on Day 2. Representative of three independent experiments is shown here.



**Supplemental Figure 6.** Phenotyping for the MHCII<sup>low-medium</sup>CD11c<sup>medium</sup> population in the draining lymph node. Representative of two independent experiments is shown here.



**Supplemental Figure 7.** Absence of Nano-11 from neutrophils and eosinophils in the draining lymph node. Data were obtained one day after immunization and are representative of two independent experiments.