

## Supplemental information

### Toll-like receptor 7 contributes to inflammation, organ injury, and mortality during polymicrobial sepsis in mice

Wenling Jian, MD, PhD<sup>2,3</sup>; Lili Gu, PhD<sup>1</sup>; Brittney Williams, MD<sup>1</sup>, Yan Feng, MD, PhD<sup>1,2</sup>; Wei Chao, MD, PhD<sup>1,2</sup>; Lin Zou, MD, PhD<sup>1,2</sup>

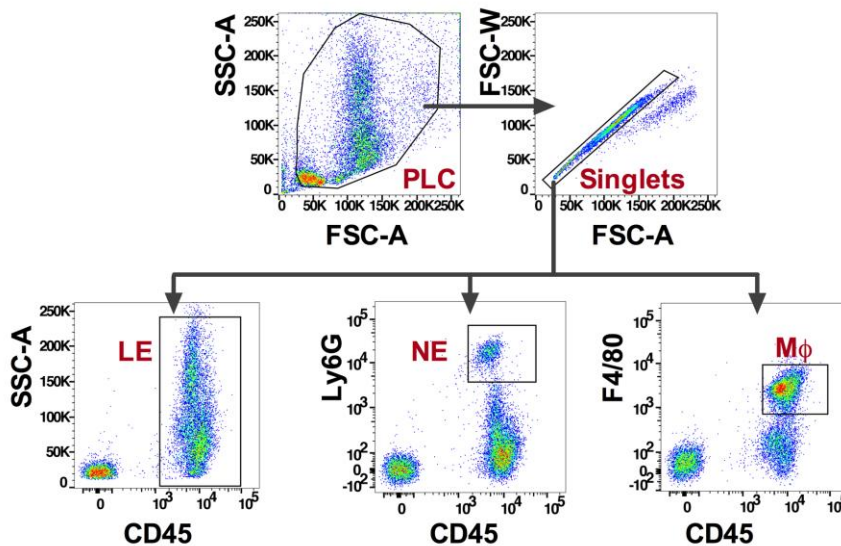
<sup>1</sup> Translational Research Program, Department of Anesthesiology and Center for Shock Trauma Anesthesiology Research, University of Maryland School of Medicine, Baltimore, MD

<sup>2</sup> Department of Anesthesia, Critical Care and Pain Medicine, Massachusetts General Hospital, Harvard Medical School, Boston, MA

<sup>3</sup> Department of Anesthesiology, the Second Xiangya Hospital, Central South University, Changsha, Hunan, China

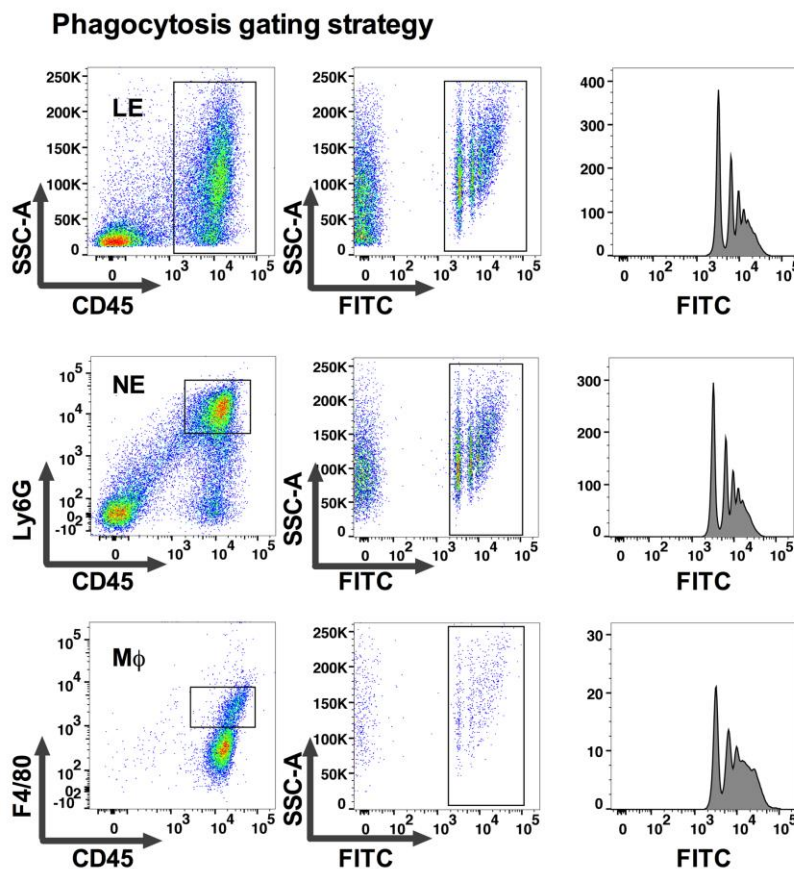
#### Supplement Figure 1

##### Gating strategy



**Supplement Figure 1: Gating strategy of a representative flow cytometry for identification of myeloid cell subsets in the peritoneal space.** Twenty-four hours after surgery, peritoneal cells (PLC) were harvested and doublets were excluded. Single-cell suspensions were analyzed for the expression of CD45<sup>+</sup> for leukocyte (LE), CD45<sup>+</sup>Ly6G<sup>+</sup> for neutrophil (NE) and CD45<sup>+</sup> F4/80<sup>low</sup> for small peritoneal macrophage (MΦ). FSC-A = forward scatter area, FSC-W = forward scatter width, SSC-A = side scatter area, LE= leukocyte , MΦ=macrophage, NE=Neutrophil.

**Supplement Figure 2**



**Supplement Fig. 2 - Gating strategy of phagocytic cells.** The 10<sup>8</sup> fluorescein isothiocyanate (FITC)-labeled carboxylate-modified microspheres (beads) in 200 μl were injected intraperitoneally at 23 hours following sham or CLP surgery. One hour later, peritoneal cells were collected and analyzed. Percentage of cells phagocytosing beads was calculated by the ratio of FITC-positive cells to total cells of each population (leukocyte: CD45<sup>+</sup>, neutrophils: CD45<sup>+</sup>Ly6G<sup>+</sup>, macrophages: CD45<sup>+</sup>F4/80<sup>low</sup>). The single cells phagocytic function was expressed as the mean fluorescence intensity of FITC-positive cells within each cell population. FSC-A = forward scatter area, FSC-W =

forward scatter width, SSC-A = side scatter area, LE= leukocyte , MΦ=macrophage,  
NE=Neutrophil.