

Supplemental Figure 3. Arsenicals inhibit caspase-1 through indirect mechanisms that do not involve sequestration into a large molecular weight complex or phosphorylation events. (A) Sucrose lysates from Balb/cJ BMDMs pre-treated with LPS (1  $\mu$ g/mL, 2 h) were mixed with active recombinant caspase-1 (1 U/50  $\mu$ L) in the presence or absence of NaAsO<sub>2</sub> or positive control inhibitor Boc-D-CMK. Lysates were incubated at 37°C for 3 h and IL-1 $\beta$  cleavage was assessed by Western blot. (B) RAW264.7 cells were incubated with 50  $\mu$ M As<sub>2</sub>O<sub>3</sub> or heat shocked at 42°C for 60 min followed by lysis in sucrose lysis buffer. Supernatants and pellet were analyzed for caspase-1 by Western blot after centrifugation (10,000 x g for 10 min). (C) Sucrose lysates from RAW264.7 cells pre-treated with LPS (1  $\mu$ g/mL, 2 h) followed by As<sub>2</sub>O<sub>3</sub> (50  $\mu$ M, 1 h) were spiked with varying concentrations of phosphatase or kinase inhibitors prior to treatment with active recombinant caspase-1 (1 U/50  $\mu$ L, 37°C, 3 h). IL-1 $\beta$  cleavage was assessed by Western blot.