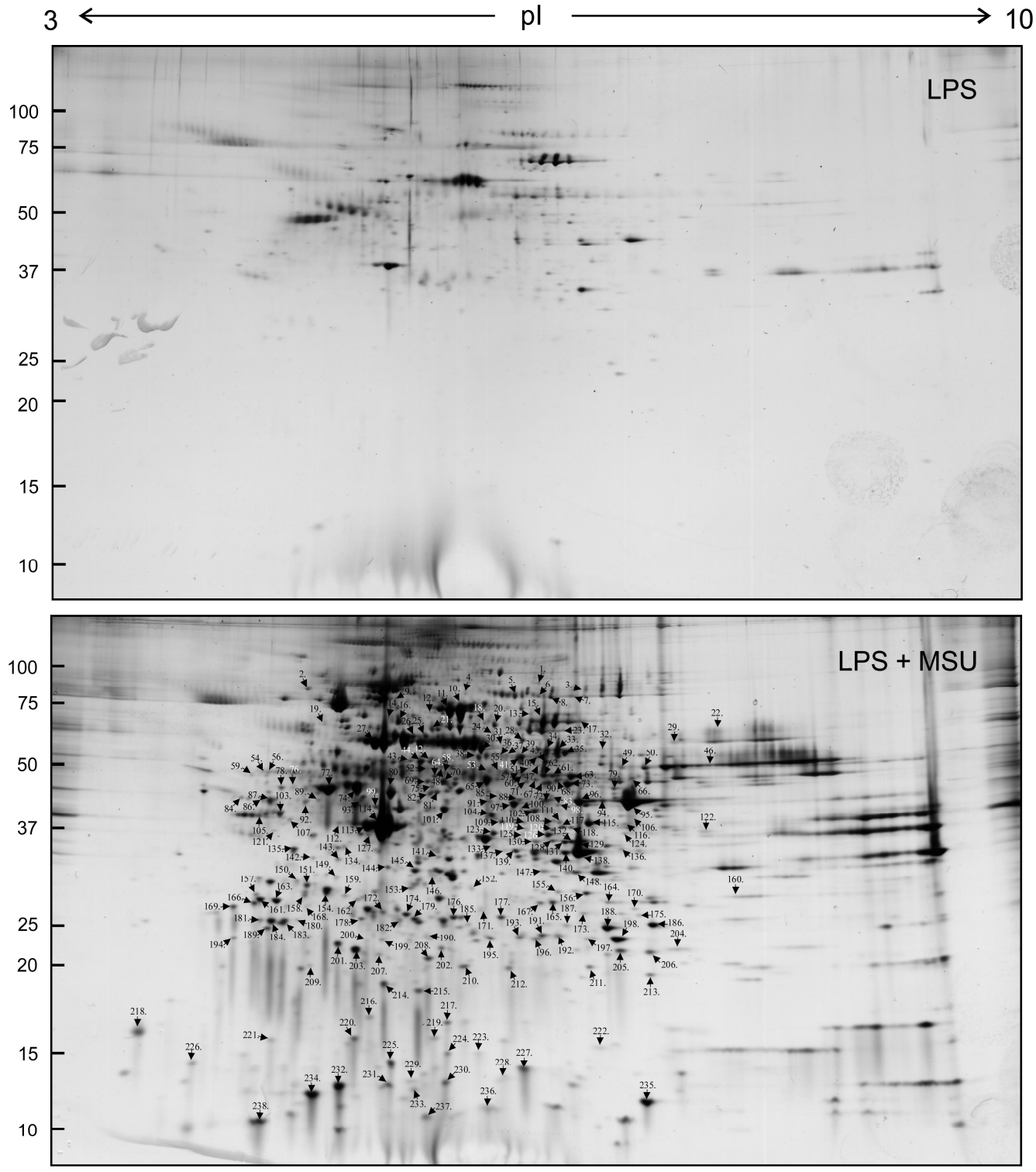

Figure S1. 2-DE based secretome analysis of LPS-primed and MSU-stimulated human primary macrophages.

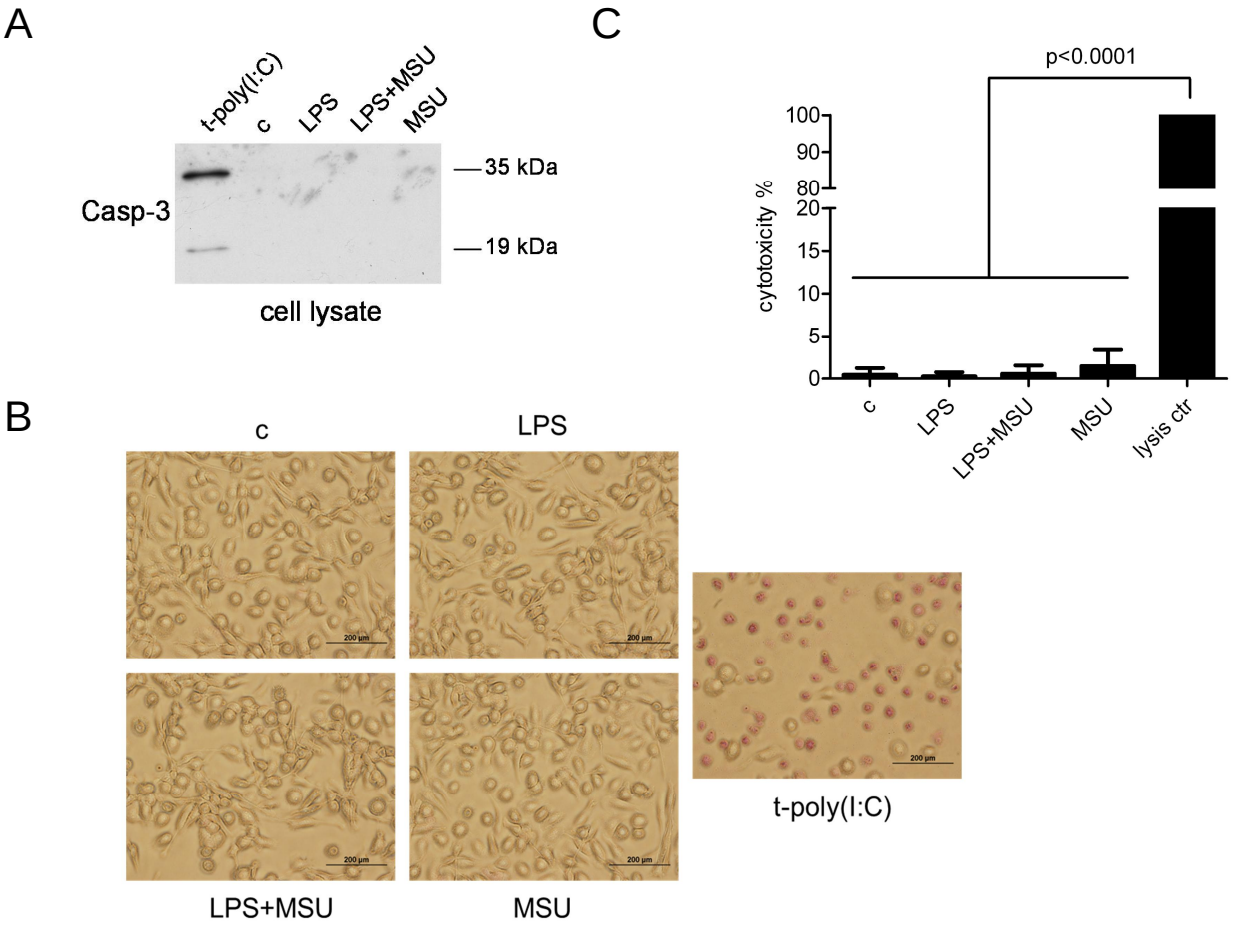

Figure S2. Lack of apoptosis and necrosis in MSU-stimulated macrophages (A) Macrophages were left untreated or primed with LPS (100 ng/ml) for 21 h after which they were left unstimulated or activated with MSU (100 µg/ml) for 3 h. After this total cellular lysates were prepared and Caspase-3 expression was analyzed by western blotting. In addition, macrophages were transfected with poly(I:C) (10 µg/ml; Sigma) for 18 h and cell lysate prepared from these cells was used as a positive control. Transfection was done using Lipofectamine (Invitrogen) according to manufacturer's instructions. Caspase-3 antibody was purchased from Cell signaling technology. (B) Cells were treated as in (A), and the number of apoptotic cells was assessed by Apopersentage assay (Biocolor Ltd), where a dye is selectively imported in apoptotic cells. Length of the scalebar in the pictures is 200µm. (C) Macrophages were left untreated or primed with LPS after which they were left unstimulated or activated with MSU. Cell culture supernatants were collected and lactate dehydrogenase (LDH) activity was measured. Supernatant from cells treated with 1% Triton-X was used as a positive control. Quantification of cytotoxicity was done by Cytotoxicity Detection Kit PLUS (LDH) kit (Roche Applied Science) according to manufacturer's instructions. Mean LDH activity (±SD) from five blood donors is shown. T-test was used to compare differences between the samples.

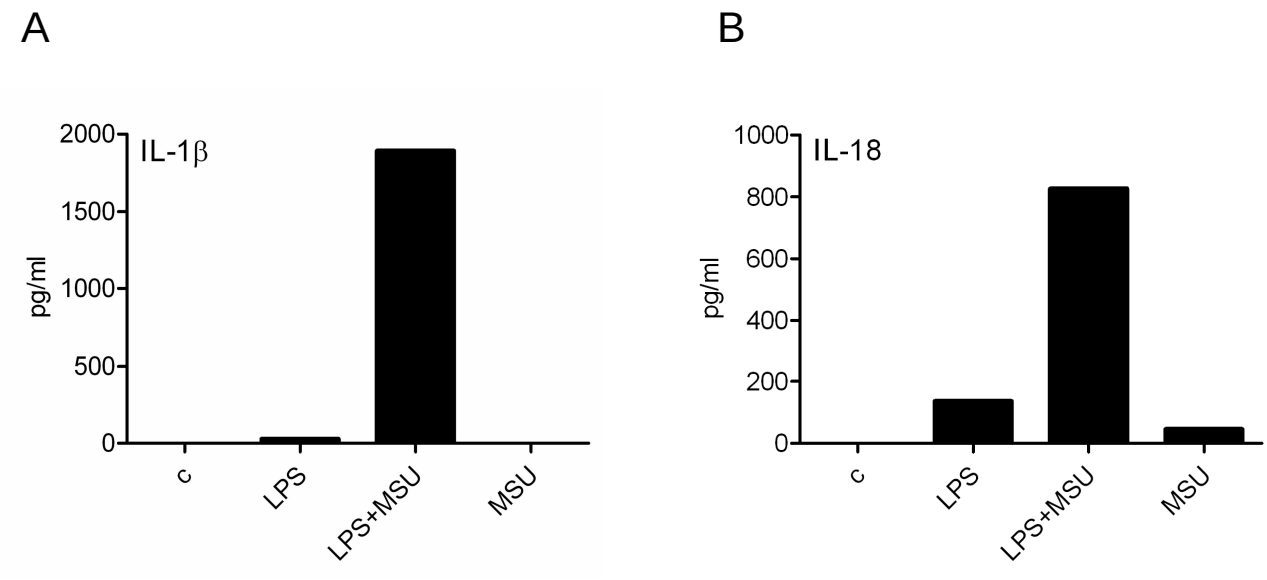

Figure S3. Inflammasome activation upon MSU stimulation was verified by measuring IL-1β and IL-18 secretion from cell culture supernatants. Macrophages were left untreated or primed with LPS (100 ng/ml) for 21 h after which they were left unstimulated or activated with MSU (100 μg/ml) for 3 h. After that cell culture supernatants were collected and cytokine levels measured with ELISA. IL-1β was analyzed with human IL-1β Eli-pair ELISA (Diaclone, Tepnel Research Product & Services) and IL-18 was analysed with human IL-18 module set (Bender MedSystems).