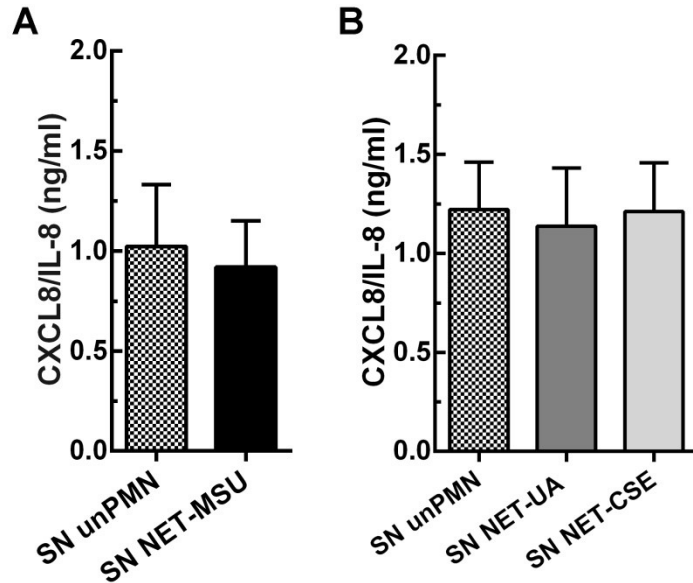


Figure S7. Concentrations of CXCL8/IL-8 in neutrophil in supernatants of neutrophils stimulated to undergo NETosis.



Neutrophils (5×10^6 /ml) were cultured with either MSU crystals (300 μ g/ml), UA (8 mg/dl), CSE (a tenfold dilution of a solution with a concentration corresponding to cigarettes containing 1.36 mg/ml tar) or left untreated for 1 h at 37°C. Then, after gentle pipetting, stimuli were removed by a quick spin in a microcentrifuge (12 sec) and pellets were suspended in fresh medium and incubated for three additional hours at 37°C under a humidified atmosphere with 5% CO₂. Afterwards, NET were detached from other cells and from the debris by a gentle 60 sec-long ultrasonic pulse and centrifugation. NET-containing supernatants (SN NET) as well as the supernatant of unstimulated neutrophils (SN unPMN) were collected and their concentrations of CXCL8/IL-8 were determined by ELISA. Data are depicted as the mean \pm SEM of 3 (A) and (5) independent experiments performed in triplicate.