Zeptomole per mL detection and quantification of edema factor in plasma by LC-MS/MS yields insights into toxemia and the progression of inhalation anthrax

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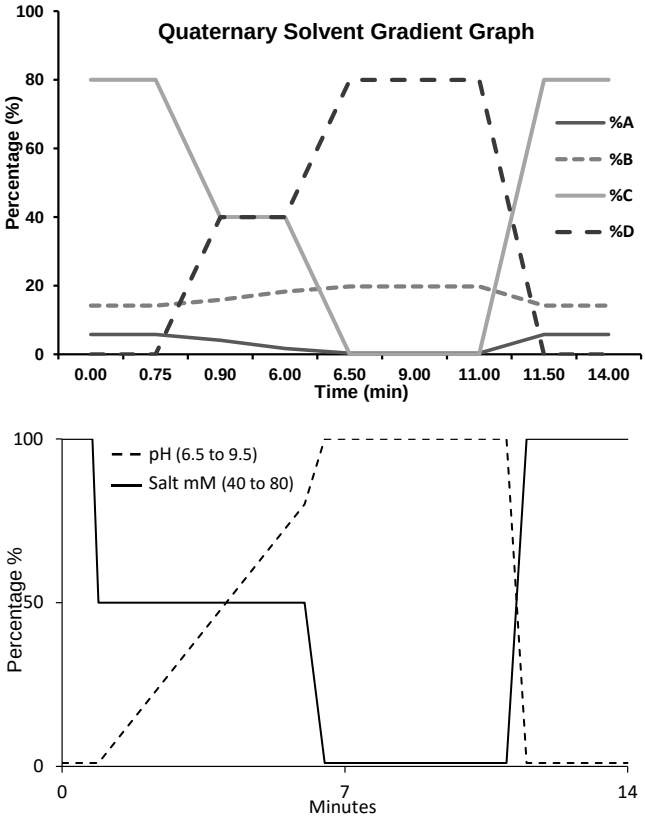
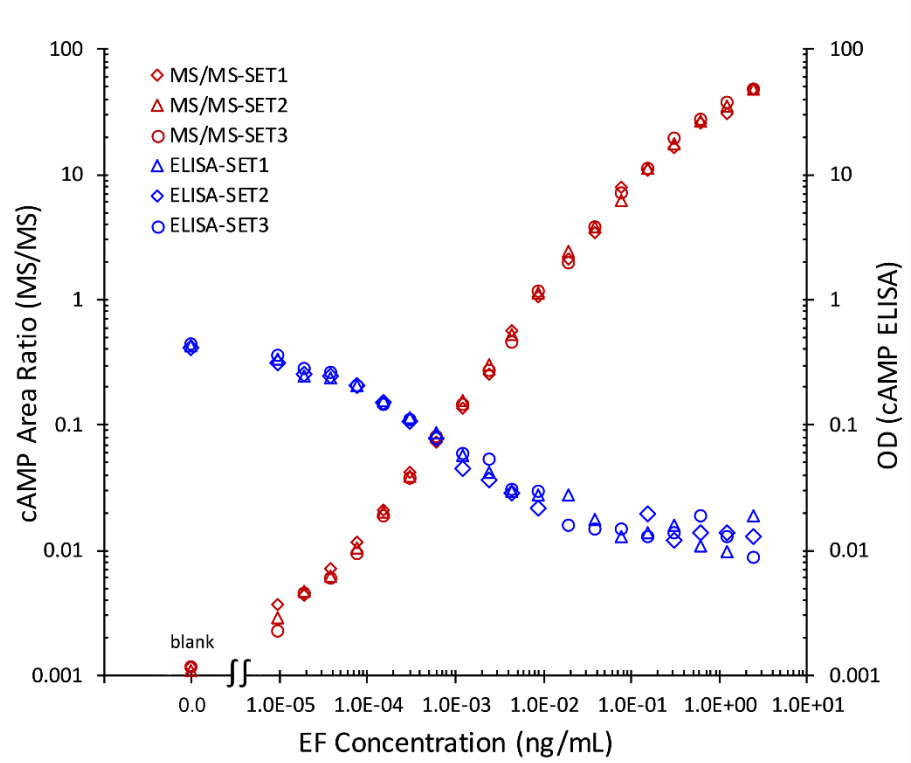
Supplementary Information

Online Resource 1 Solvent gradient program



Upper graph shows the time table of the quaternary solvent gradient program with corresponding percentage plot for solvents (A) 100 mM Acetic Acid, (B) 100 mM Ammonium Hydroxide, (C) 90% Acetonitrile with 10 mM Ammonium Acetate and (D) dH2O. Lower graph shows the pH and the salt concentration gradients as a result of mixing four solvents in the solvent program.

Online Resource 2 Comparison of competitive ELISA and LC-MS/MS for cAMP detection following EF capture/reaction



Three sets of EF were purified from plasma spiked at 2.5 ng/mL and titrated 2-fold serially down to 0.0000095 ng/mL, for 19-concentrations and blank, reacted with ATP in optimized buffer then reactions were split and analyzed by LC-MS/MS (cAMP area ratio) and competitive ELISA (OD at 405 nm), Direct cAMP kit.