



Figure S1. AdeB and Ethidium interactions. (A) Fluorescence polarization (FP) assay shows Et binds to AdeB with an affinity of $2.5 \pm 0.1 \mu M$. (B) FP shows R6G binds AdeB with an affinity of $3.1 \pm 0.1 \mu M$. (C) Kinetics of intracellular accumulation of Et in the *A. baumannii* *Ab $\Delta 3$* cells lacking the AdeABC, AdeIJK and AdeFGH efflux pumps. Et accumulates inside of cells with all three pump systems are knocked out (green, 0 μM Et; blue, 0.5 μM Et; yellow, 1 μM Et; gray, 2 μM Et; red, 4 μM Et; and cyan, 8 μM Et). (D) Complementing triple knockout cells with AdeABC restores extrusion and halts accumulation of Et inside of cells (green, 0 μM Et; blue, 0.5 μM Et; yellow, 1 μM Et; gray, 2 μM Et; red, 4 μM Et; and cyan, 8 μM Et).