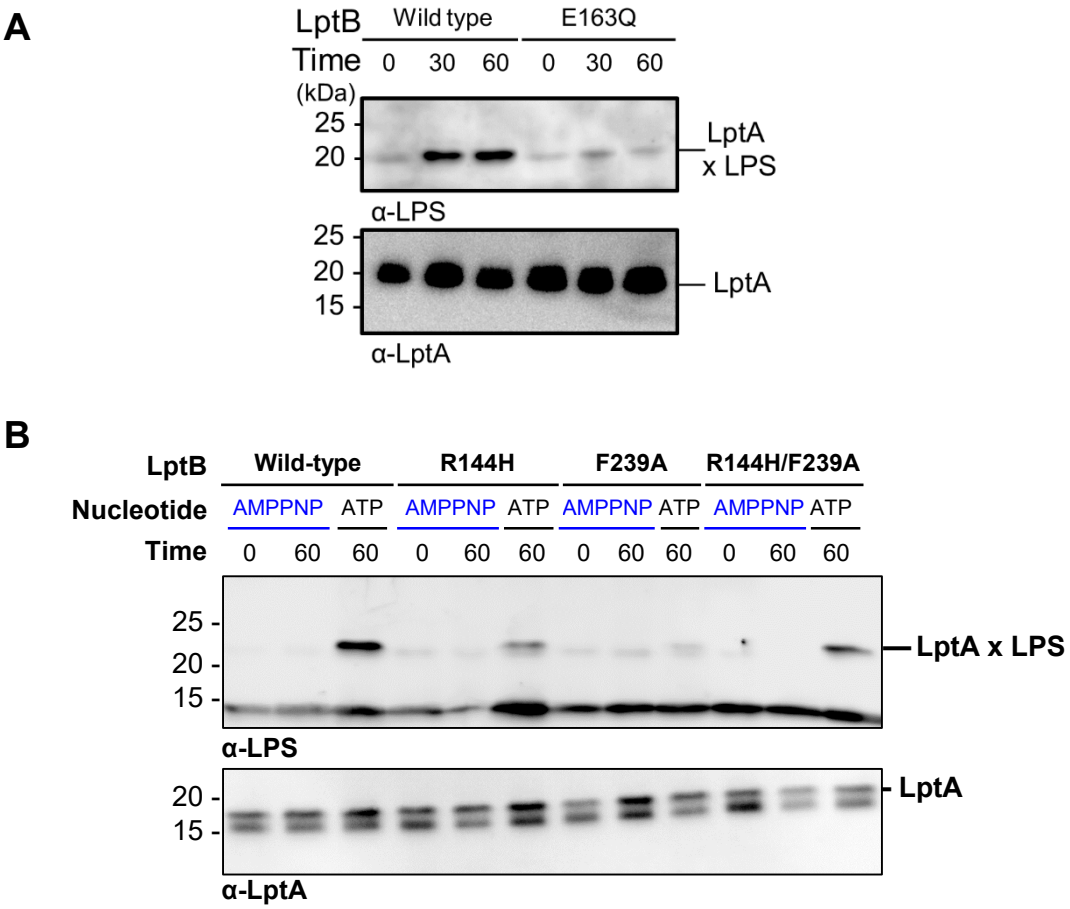

Figure S7: ATP hydrolysis is required for LPS release from proteoliposomes. **(A)** Complexes containing the catalytically dead LptB^{E163Q} variant are defective in extracting LPS from liposomes. Assay monitoring LPS release from liposomes containing LptB$_2$FGC complexes with LptB^{WT} and catalytically dead LptB^{E163Q} variants. Released LPS was UV-crosslinked to LptA^{I36pBpa} and the resulting LptA^{I36pBpa} x LPS adducts were detected by LPS immunoblot (top panel). Assays were initiated by the addition of ATP and incubated at 30°C for the indicated amount of time prior to crosslinking. LptA immunoblots are shown to confirm total LptA levels (bottom panel). Data are representative of three independent biological replicates. **(B)** *In vitro* assays for release of LPS from proteoliposomes containing LptB$_2$CFG complexes with different LptB variants were performed in the presence of either ATP or non-hydrolysable AMPPNP. After 60 min, LPS was cross-linked to LptA^{I36pBpa} and the resulting LPS-LptA^{I36pBpa} adducts (marked LptA x LPS) were detected by LPS immunoblot (top panel). Total levels of LptA^{I36pBpa} in the assay were confirmed by LptA immunoblotting (bottom panel). We also did not detect LPS transport by complexes containing LptB^{WT} or LptB$^{R144H/F239A}$ when using the non-hydrolysable ATP analogs α,β-methylene ATP (ApCpp), 2-chloroadenosine β, γ-methylene ATP (AppCp), ATP-γS, and AppNH$_2$.