

SUPPORTING INFORMATION

Membrane trafficking of the bacterial adhesin GspB and the accessory Sec transport machinery

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Figure S1

Figure S2

Figure S3

References

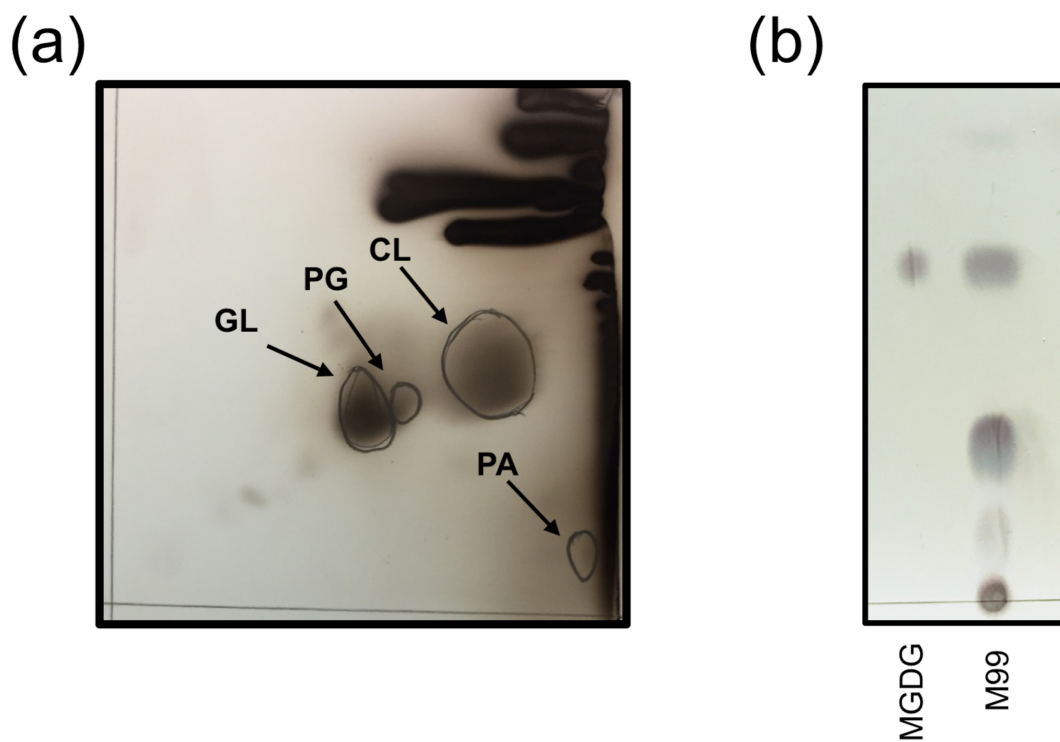


Figure S1. Lipid analysis of *S. gordonii* strain M99. Cells were extracted and analyzed by (a) 2D TLC and stained with cupric sulfate for total lipids or (b) 1D TLC and stained with orcinol for glycolipids. CL, cardiolipin; PG, phosphatidylglycerol; PA, phosphatidic acid; GL, glycolipids; MGDG, monoglucosyldiacylglycerol; M99, lipid extract of *S. gordonii* M99 cells. Lipid extraction and 2D TLC analysis of *S. gordonii* M99 cells was performed using a previously published protocol (1). For 1D TLC, *S. gordonii* M99 lipid were extracted using 2:1 MeOH/CHCl₃. The organic layer was dried using vacuum centrifugation. Lipids were resuspended in 9:1 CHCl₃/MeOH. TLC was run using an HPTLC plate (Sigma) with a 13:5:1 CHCl₃/MeOH/H₂O solvent system. Glycolipids were stained using a 5% orcinol stain.



Figure S2. Lipid binding of wild type SP-AST with PC liposomes. Representative coomassie stained SDS-PAGE gel is shown. Protein-lipid co-floitation assay was performed as described in the methods.

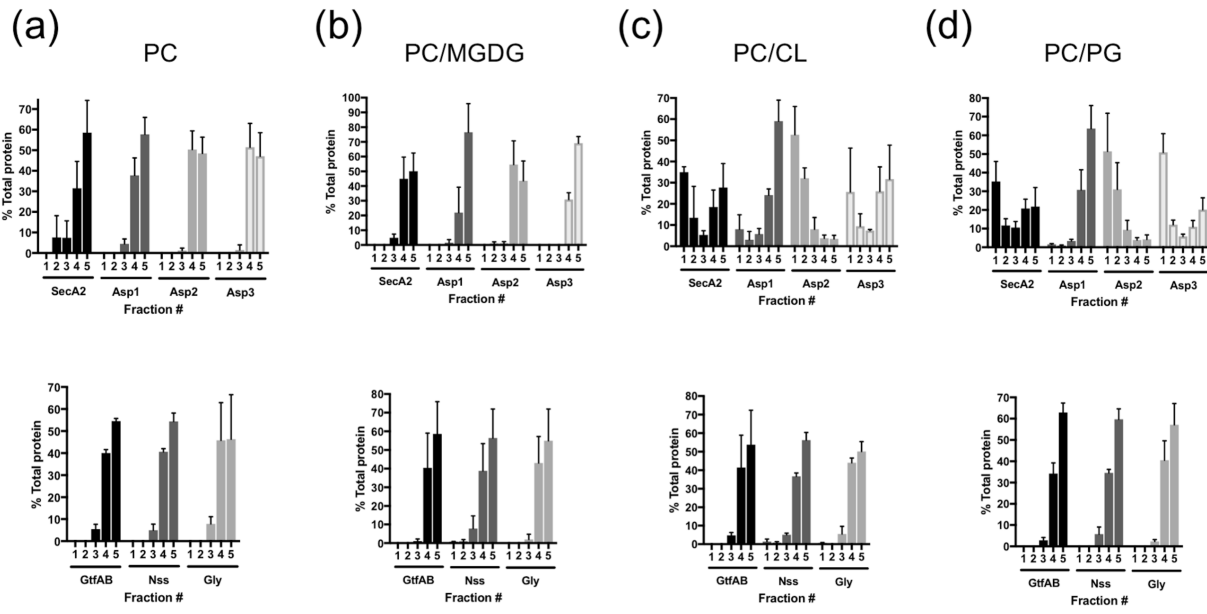


Figure S3. Densitometry quantitation of aSec protein binding to PC/CL in gradient fractions from Figure 5. (A) Lipid binding of aSec transport and glycosylation proteins with aa) PC liposomes (b) PC/MGDG liposomes (c) PC/CL liposomes, and (d) PC/PB liposomes. Independent experiments were performed in triplicate, except with MGDG, which was performed in duplicate. Plotted are mean values \pm SD.

References

1. Mishra, N. N., Tran, T. T., Seepersaud, R., Garcia-de-la-Maria, C., Faull, K., Yoon, A., Proctor, R., Miro, J. M., Rybak, M. J., Bayer, A. S., Arias, C. A., and Sullam, P. M. (2017) Perturbations of Phosphatidate Cytidylyltransferase (CdsA) Mediate Daptomycin Resistance in *Streptococcus mitis/oralis* by a Novel Mechanism. *Antimicrob Agents Chemother* **61**