Supporting Information

Neumann et al. 10.1073/pnas.1001681107

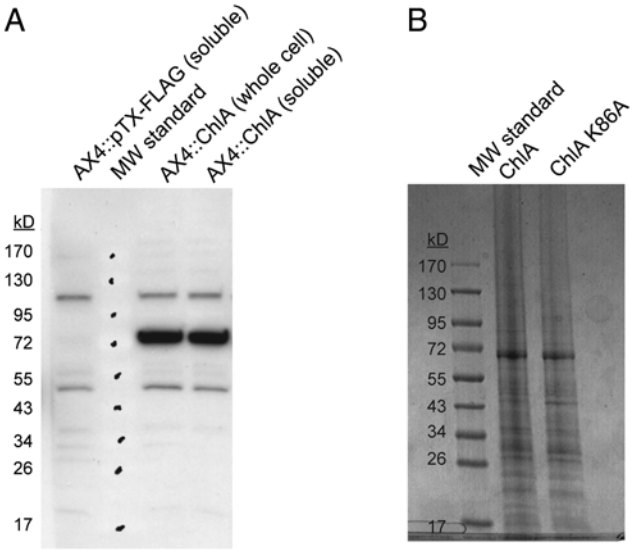

Fig. S1. Analysis of protein expression and purification. (*A*) Western blot analysis showing expression of ChlA protein and recovery in the soluble fraction of the cell lysate. Proteins were blotted with an α-c-myc anitbody with HRP conjugate and detected with ECL Plus reagent (Amersham). (*B*) SDS-PAGE analysis of ChlA and ChlA K86A proteins after affinity purification with an α-FLAG agarose resin. (Molecular weight standards are approximate due to lot-specific variation in the prestained standards.)

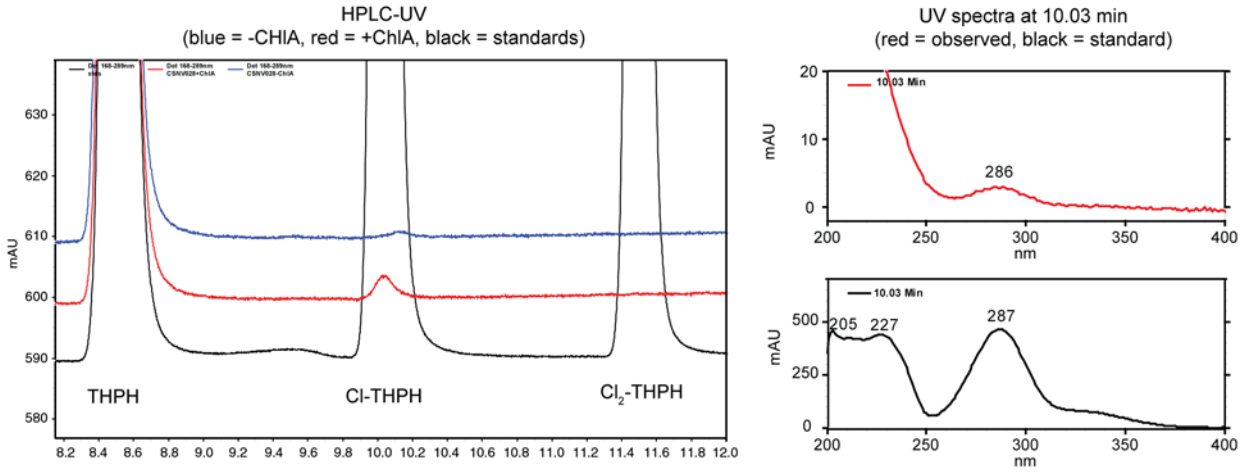

Fig. S2. HPLC-UV analysis of in vitro THPH halogenation reactions containing purified ChlA (red) or lacking ChlA (blue). A peak coeluting with authentic Cl-THPH showed $\lambda_{max} = 286$ nm (standard $\lambda_{max} = 287$ nm). Spectral differences below 250 nm are attributable to the organic phase of HPLC eluent.

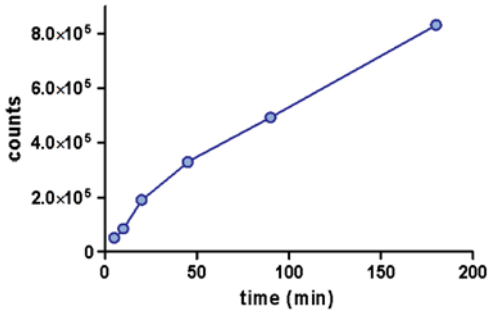

Fig. S3. ChlA-mediated formation of Cl-THPH from THPH follows a roughly linear time course. *y* axis displays counts of Cl-THPH from LCMS analysis.

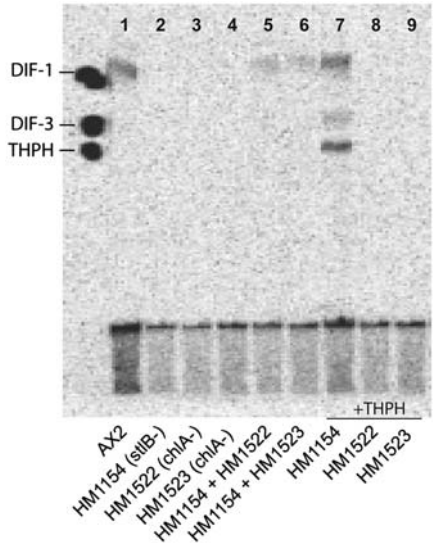

Fig. S4. 36Cl$^-$ radio-TLC analysis of DIF-1 production in vivo. DIF-1 is detectable in the parental strain Ax2 (lane 1), but not *stlB*$^-$ or *chlA*$^-$ strains (lanes 2–4). Codevelopment of *stlB*$^-$ and *chlA*$^-$ strains restores DIF-1 production (lanes 5–6). Addition of THPH to substratum restores DIF-1 production in the *stlB*$^-$ strain (lane 7), but not in the *chlA*$^-$ strains (lane 8–9) indicating that unintentional disruption of *stlB* or its promoter is not responsible for the lack of DIF-1 in *chlA*$^-$ strains.

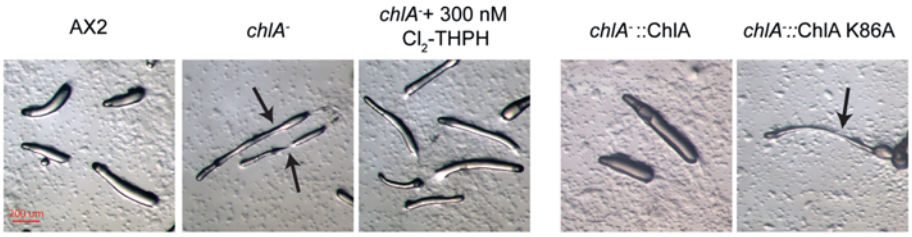

Fig. S5. Slug morphologies of wildtype (Ax2), mutant and complemented strains. The elongated, fragile slugs (arrows) characteristic of DIF-less development and observed in the *chlA*$^-$ mutant are complemented by development in the presence of Cl_2-THPH or by expression of ChlA, but not by expression of ChlA K86A.