



S2 Fig. Stability of the flagellar switch in enriched *flg* ON and OFF populations during growth in liquid and solid media. (A) Asymmetric PCR-digestion assay with template from enriched *C. difficile* R20291 *flg* ON and OFF isolates grown on an agar surface for 24 hours and 48 hours. T₀ represents the cultures used to inoculate the agar plates. (B) Asymmetric PCR-digestion assay of *C. difficile* R20291 *flg* ON and OFF isolates grown in BHIS medium collected at T₀, two exponential phase time points (EXP, OD₆₀₀ 0.5 and 1.0), and two stationary phase time points (STAT, OD₆₀₀ 1.8 and overnight, O/N). Images are representative from two independent experiments with four replicates of each *flg* phase. (C) Growth curve of *C. difficile* R20291 *flg* ON and OFF isolates in BHIS medium. Data are combined from two independent experiments each with two replicates of each *flg* phase, and means and standard deviations are shown.