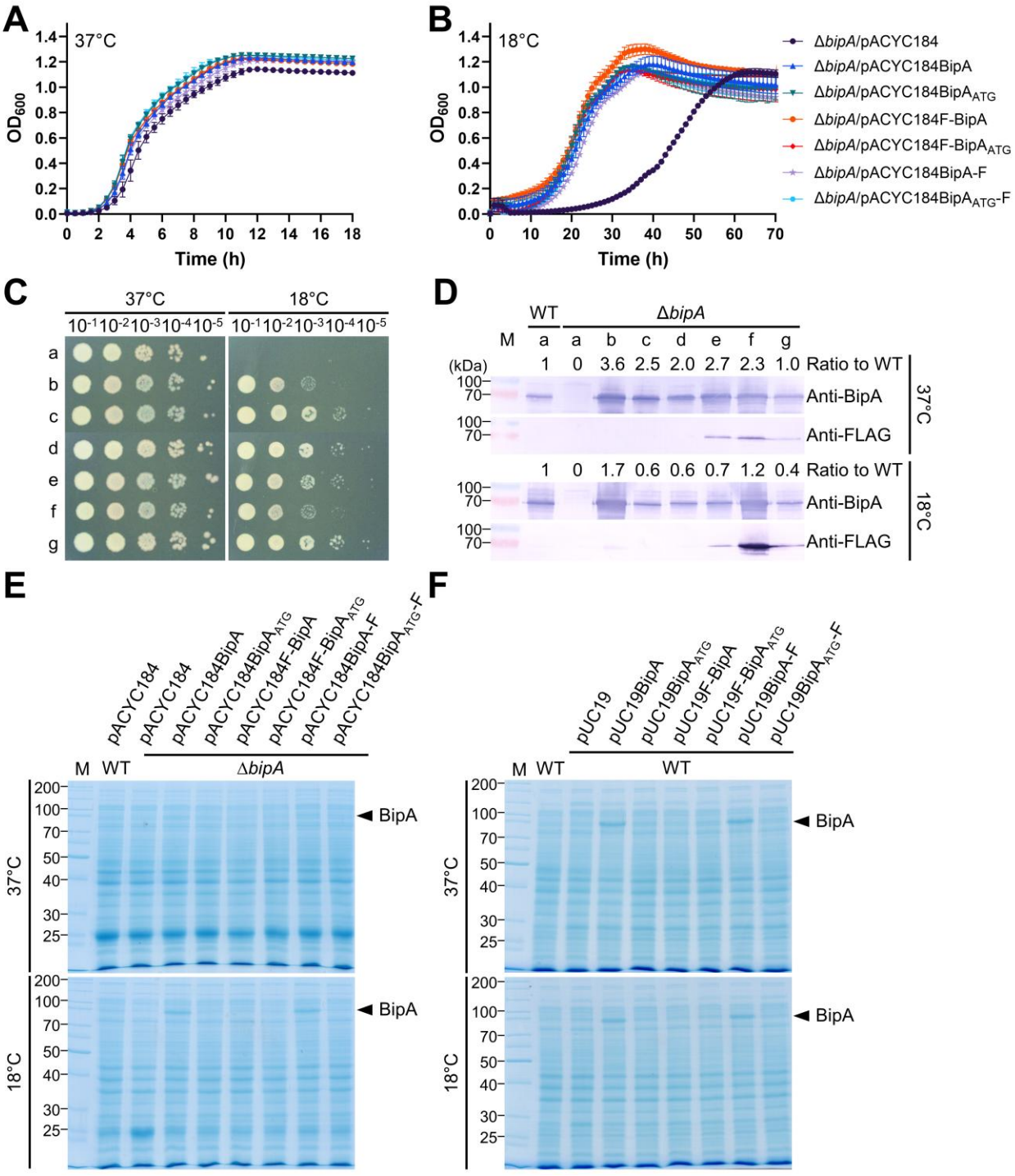


Fig. S3. Effects of BipA complementation and overexpression on cell growth. (A and B) Δ*bipA* strains carrying pACYC184-based BipA variants were diluted 1:200 into fresh LB medium supplemented with chloramphenicol and grown at 37°C (A) and 18°C (B). The data presented are the mean ± SD of three independent experiments. The same symbols are used in both panels. (C) Overnight cultures were serially

diluted (10^{-1}–10^{-5}), spotted onto LB agar plates supplemented with chloramphenicol, and incubated at the indicated temperatures. (a) pACYC184 (empty vector); (b–g) Δ*bipA* strains carrying the pACYC-based BipA constructs shown in Fig. 3A. (D) Cells grown at the indicated temperatures were harvested, resuspended in 1× SDS sample buffer, and analyzed using western blotting with anti-BipA or anti-FLAG antibodies. Labels a–g are as defined in Panel C. Relative BipA protein expression was quantified from the anti-BipA immunoblots using ImageJ after background subtraction and normalization to the wild-type sample on the same blot, which was assigned a value of 1.0. (E and F) Boiled samples prepared for western blotting analysis were subjected to SDS–PAGE followed by Coomassie blue staining. Samples were prepared from Δ*bipA* complementation strains carrying pACYC184-based BipA variants (E) and MG1655 overexpression strains carrying pUC19-based BipA variants (F). Cells were grown at 37°C or 18°C before sample preparation. Black arrowheads indicate BipA. M, molecular weight marker.