Bioscience, Biotechnology, and Biochemistry

Plasmid copy number mutation in *repA* gene encoding RepA replication initiatior of cryptic plasmid pHM1519 in *Corynebacterium glutamicum*

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Supplementary materials

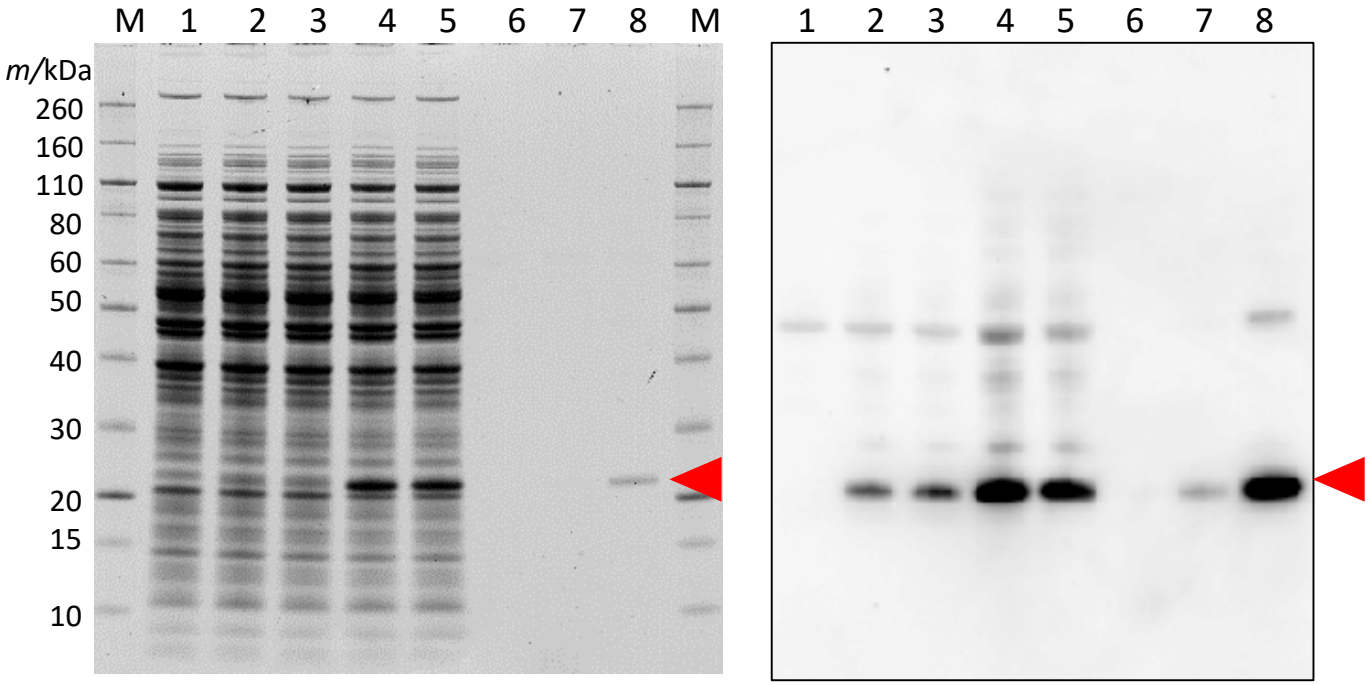

Fig. S1. SDS-PAGE analysis (A) and immunoblotting (B) of chloramphenicol acetyl transferase (CATase) produced from plasmids. For immunoblotting, rabbit anti-CATase antiserum was used. Cell-free extract of *Corynebacterium glutamicum* 2256L and purified CATase (16 mg/mL; Sigma-Aldrich) were employed as standards. Lanes: M, protein standard maker; 1, *C. glutamicum* 2256L (control); 2 and 3, strain 2256L harboring pPC4; 4 and 5, 2256L harboring pPC4H1; 6, 7 and 8, samples (4 μL/lane) of 10^{-3}, 10^{-2}, and 10^{-1} dilutions of CATase, respectively. The position of CATase is indicated with a red arrow.

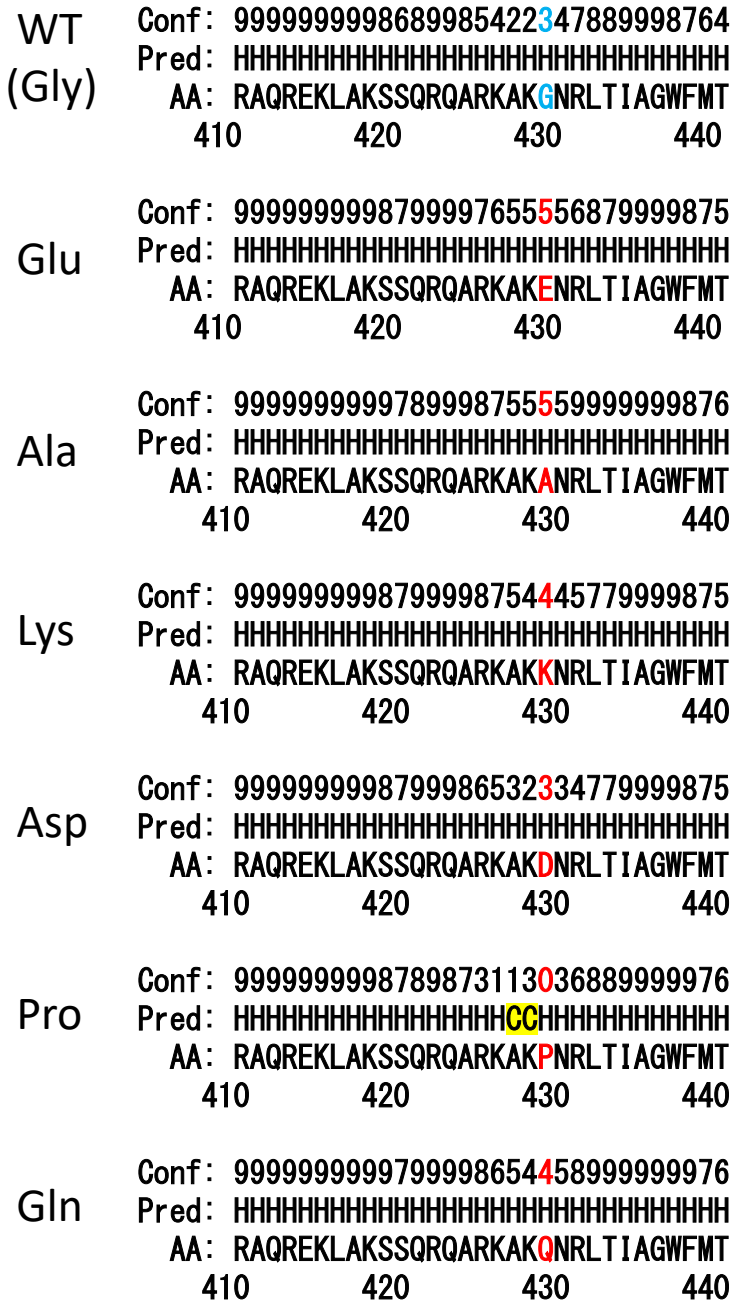

Fig. S2. Predicted secondary structure of the C-terminal region (from amino acids 410 to 440) of RepA and its mutants. The secondary structure was predicted using PSIPRED. The amino acid numbers of RepA are indicated below the sequence (AA). “H” stands for helix and “C” for coil. “Conf” represents the confidence of each prediction. The Gly (G) residue at position 429 of wild-type RepA and the confidence value for the prediction of its structure are indicated in blue. Each substituted residue and its confidence value is in red. A coil structure (highlighted in yellow) was only predicted on replacement of Gly429 to Pro.