Role of Gly117 in the cation/melibiose symport in MelB of *Salmonella typhimurium*

Lan Guan*, S. Vivek Jakkula, Alexey A. Hodkoff, and Yue Su

Supplemental Figures

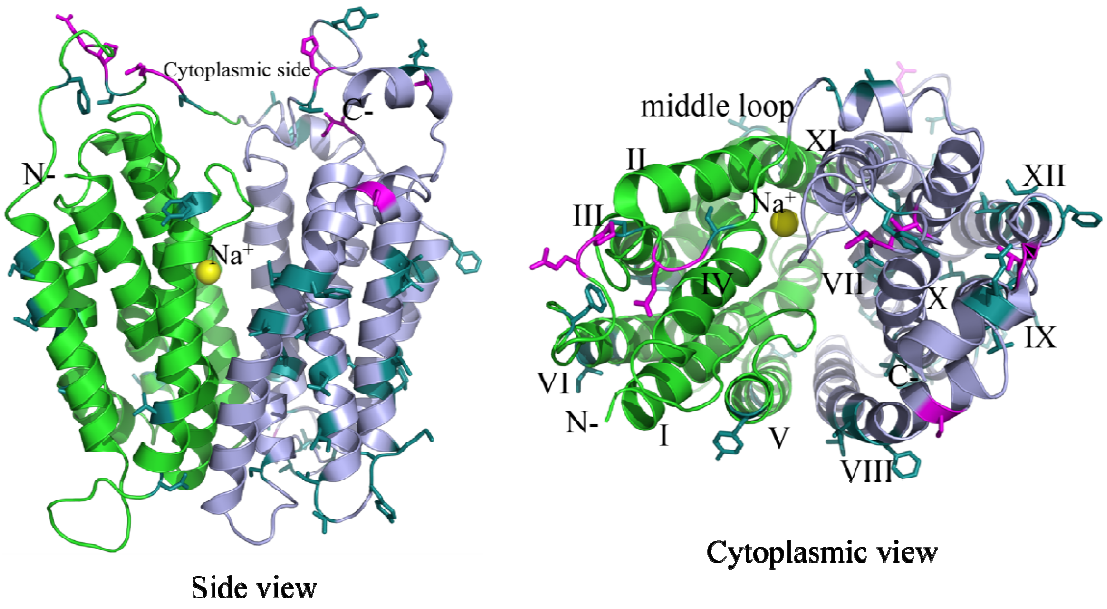

Fig. S1. Conservation between $MelB_{St}$ and $MelB_{Ec}$. Primary sequence alignment was performed using ClustalW 2.1 at the EBI (1,2). The sequence variations are projected on the threading model of $MelB_{Ec}$ (positions T6 to L448) (3) as shown in cyan (conserved changes) and magenta (non-conserved changes) sticks. The N- and C-terminal helix bundles are shown in green and blue colors, respectively. A Na^+ atom (yellow sphere) is docked between helices II and IV.

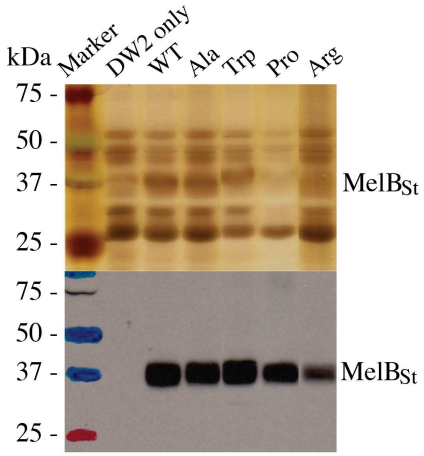

Fig. S2. Protein expression. Protein expression and membrane preparation were carried out as described in *Materials and Methods*. Crude membranes (15 μg) were loaded onto each well of SDS-12%PAGE. Total membrane proteins were detected by the silver staining (*top panel*), and $MelB_{St}$ was immunodetected using anti-His antibody after Western blotting (*bottom panel*).

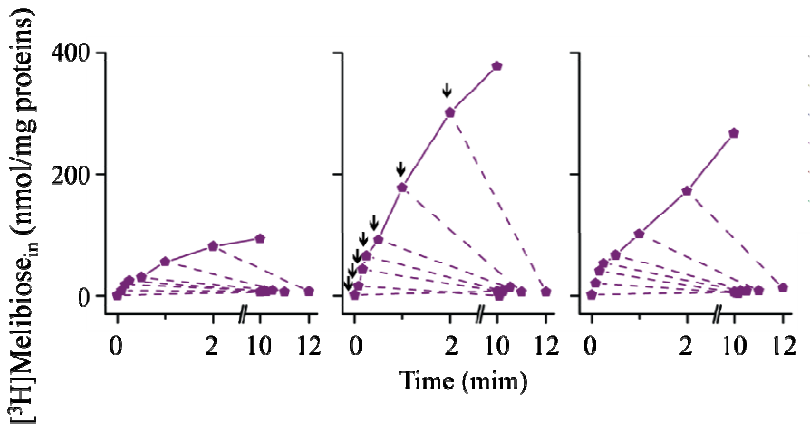

Fig. S3. Melibiose exchange. *E. coli* DW2 cells with the WT $MelB_{St}$ were washed and resuspended with 100 mM KP_i, pH 7.5, 10 mM $MgSO_4$, and adjusted to 0.7 mg/ml of

protein. Transport was initiated by adding melibiose (0.4 mM, 10 mCi/mmol) in the absence or presence of 20 mM NaCl or LiCl. The unlabeled melibiose at 100 mM was added at each time point during transport as shown by arrows, and incubated for 10 min prior to the filtration (dashed lines). Intracellular melibiose is plotted as a function of time.

REFERENCES

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