Supplementary Information

Molecular structure of a metal-independent bacterial glycosyltransferase that catalyzes the synthesis of histo-blood group A antigen

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Supplementary Figures

Figure S1. A multiple sequence alignment (using Clustal X2) of GT6 family members used in this study. Loop1, Loop2, Loop3 and C-term are secondary structural regions of BoGT6a that exhibit significant conformational changes upon ligand binding. A1-A7 are residues that contribute towards acceptor binding and are boxed in blue colour. D1-D14 are donor binding residues and are boxed in green colour. The residues in the box coloured in gold are the manganese ion binding D-*X*-D motif of GTA/B and/or α3GT. In BoGT6a this is replaced by N-*X*-N motif.

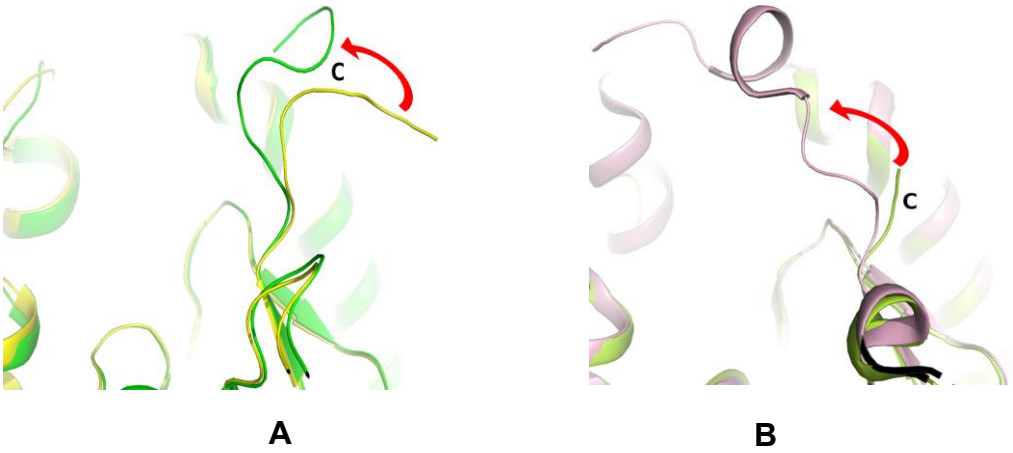

Figure S2. C-terminal conformational flexibility as observed in X-ray crystallographic structures of BoGT6a and α3GT. **(A)** BoGT6a (yellow) and BoGT6a-FAL (green); **(B)** α3GT (pea colour) and α3GT in complex with UDP and GalNAc (PDB id: 1GX4) (light purple).

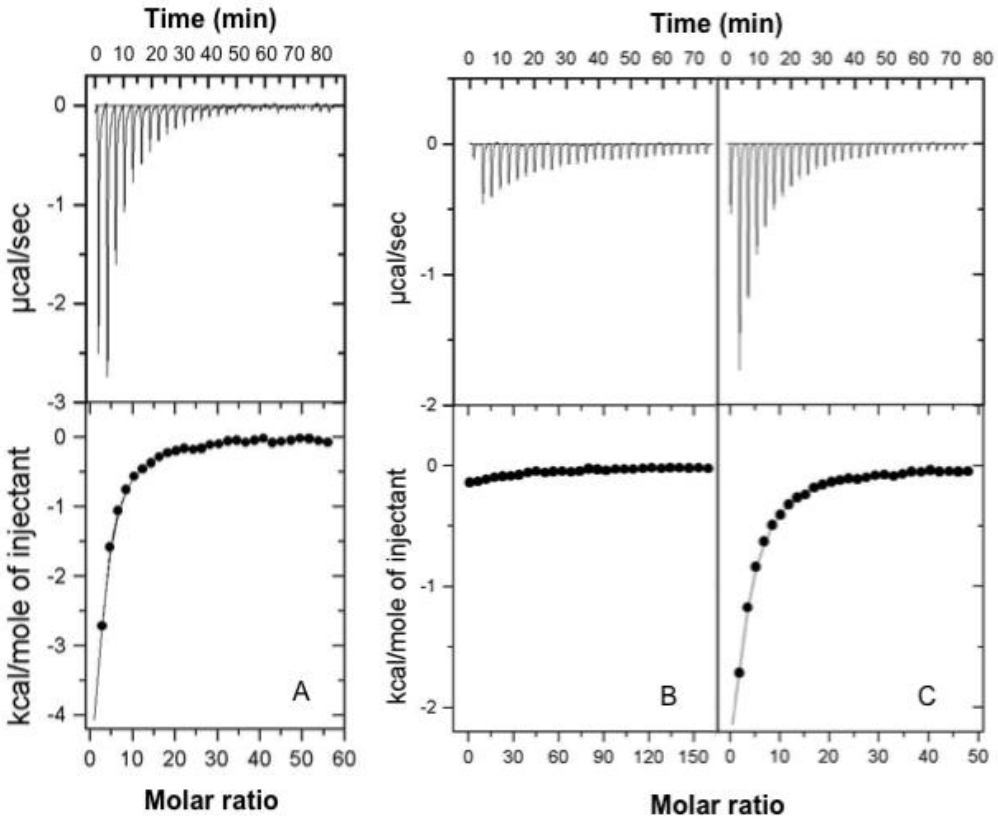

Figure S3. Isothermal titration of BoGT6a with ligands. Titrations were with (**A**) UDP-GalNAc (**B**) FAL and (**C**) FAL in the presence of 3 mM UDP (**D**). Both the ligand and sample were in 20mM HEPES buffer pH 7.5, containing 1 mM DTT and 0.2M NaCl and the reaction cell was maintained at 25 °C. The first injection in each titration was 5 µl of ligand and subsequent injections were 10

μl. The upper section of each panel shows the exotherms of binding for each injection of ligand and the lower, the integrated binding isotherm with the experimental points (filled circles) and fitted line that gave the values for thermodynamic parameters give in **Table 3.**