

Supplemental Material

Table S1: Interactions between enzyme residues and carbohydrate

Substrate	Enzyme	Interactions		
		Substrate Contacts	H-Bonds (Donor Sugar)	H-Bonds (Donor Enzyme)
Sucrose	<i>B. subtilis</i>	D86, S164, L109, Y429, R343, D247, Y411, R360, R246, W163	Y429, D86	S164
Sucrose	<i>B. amyloliquefaciens</i>	R213, E307, R327, R400, E309	E307, E309	R327
Sucrose	<i>B. indica</i>	D98, F421, T361, R445 P409 H359 Y410, D439, D371, H101	D439, D371, D98	H101, R445
Sucrose	<i>G. oxydans</i>	H80, T365, D313, W79, R82, W346, D364, H301	D364, D313	R82, W79
Sucrose	<i>P. graminis</i>	R101, R434, H102, P398, P350, D99, Y399, H348	D99	R101
Sucrose	<i>V. natriegens</i>	R234, H345, Y230, E327, N232, Q325	E327, Q325, N232	H345, Q325
Fructose	<i>B. subtilis</i>	D247, Y411, S412, A342, D86, R246, E262, R343	D86, E262, D247	S412, R246
Fructose	<i>B. amyloliquefaciens</i>	R213, E307, R327, R400, E309	E309, D53	Y52
Fructose	<i>B. indica</i>	F244, S246, R248, D338, Q339	ASP338, GLN339	ARG248
Fructose	<i>G. oxydans</i>	S120, W41, N83, R194, W119, D195, E283	D195, E283	R194, N83, S120
Fructose	<i>P. graminis</i>	H102, D99, R434, F98, D428, R101	D99	H102, R101
Fructose	<i>V. natriegens</i>	R175, Y230, G179, N180, D181	D181	N180, Y230, D181, R175
Glucose	<i>B. subtilis</i>	D247, S164 D86, R246, L109, W163	D247, D86, S164	-
Glucose	<i>B. amyloliquefaciens</i>	W130, E309, R213, D214, L76, W52, S131	-	W52, S131
Glucose	<i>B. indica</i>	D98, R100, H101, P409, Y410, D439, R445	D98, R100, R445	H101
Glucose	<i>G. oxydans</i>	S120, R194, L65, W41, D195, W119	D195	S120, R194
Glucose	<i>P. graminis</i>	R434, H102, R101, F98, D428, D99	D99	H102, R101
Glucose	<i>V. natriegens</i>	R435, H100, D425, D97, P395, R99	D425, D97	R99

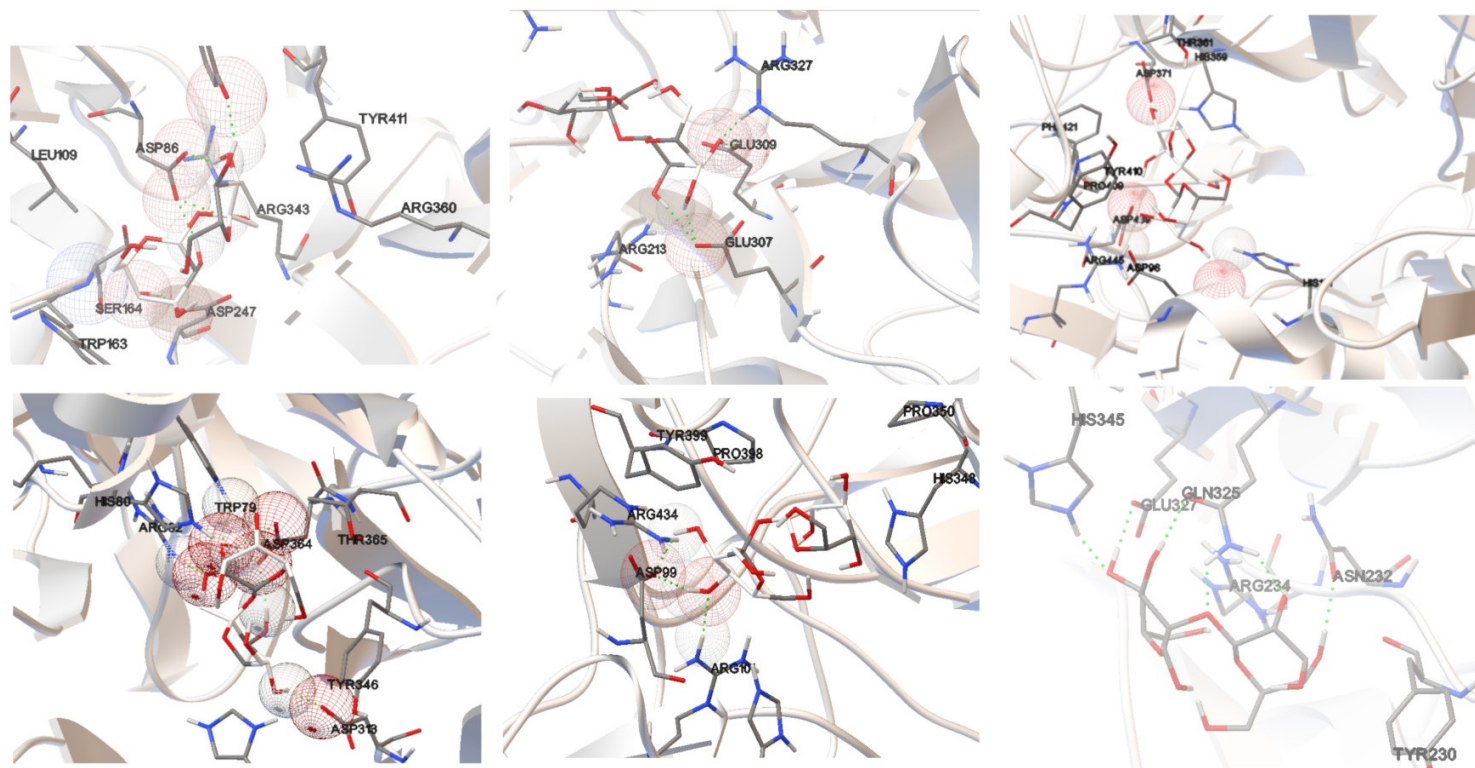


Figure S1: Models of LS from a) *B. subtilis*, (b) *B. amyloliquefaciens*, (c) *B. indica subsp. indica*, (d) *G. oxydans*, (e) *P. graminis* and (f) *V. natriegens* with sucrose docked within the active site. Sucrose is docked in a position representing the least binding energy state. Colouring represents electrostatic surface potential. The surface charge was calculated using AutoDock v 4.2 and the PDBqt file was uploaded to OPAL webserver for PQR map and visualization was done using PyMol APBS plugin.

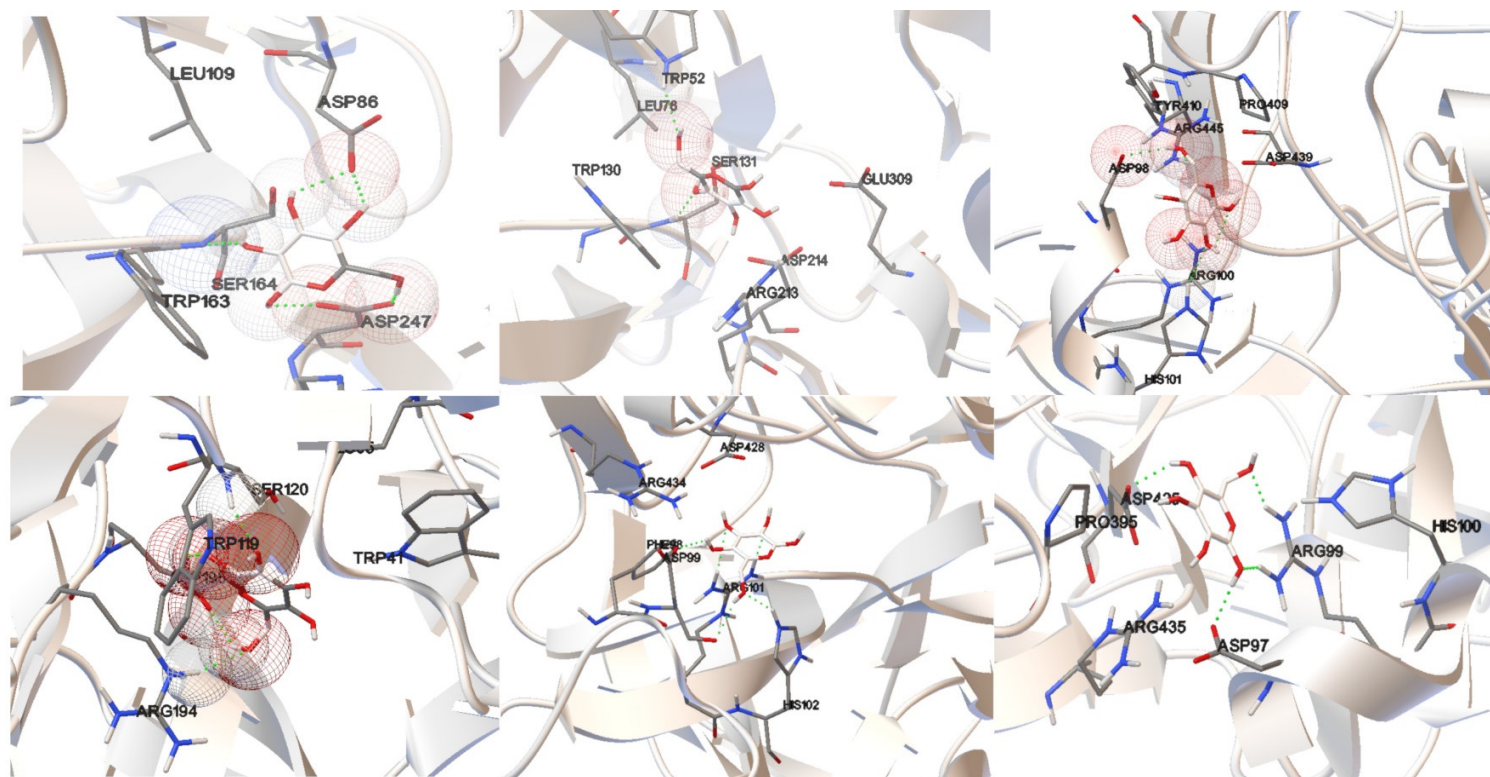


Figure S2: Models of LS from a) *B. subtilis*, (b) *B. amyloliquefaciens*, (c) *B. indica subsp. indica*, (d) *G. oxydans*, (e) *P. graminis* and (f) *V. natriegens* with glucose docked within the active site. Glucose is docked in a position representing the least binding energy state. Colouring represents electrostatic surface potential. The surface charge was calculated using AutoDock v 4.2 and the PDBqt file was uploaded to OPAL webserver for PQR map and visualization was done using PyMol APBS plugin.

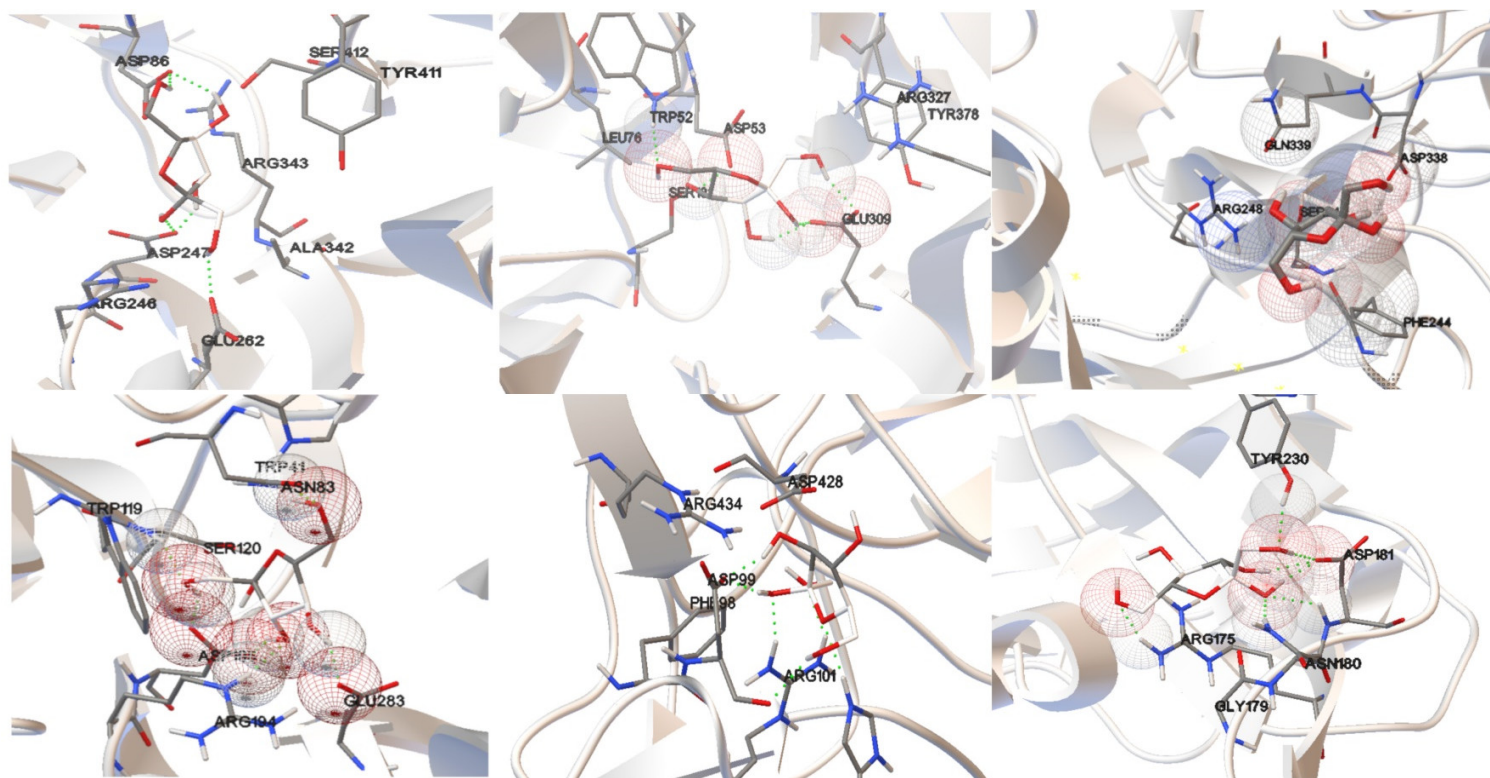


Figure S3: Models of LS from a) *B. subtilis*, (b) *B. amyloliquefaciens*, (c) *B. indica subsp. indica*, (d) *G. oxydans*, (e) *P. graminis* and (f) *V. natriegens* with fructose docked within the active site. Fructose is docked in a position representing the least binding energy state. Colouring represents electrostatic surface potential. The surface charge was calculated using AutoDock v 4.2 and the PDBqt file was uploaded to OPAL webserver for PQR map and visualization was done using PyMol APBS plugin.

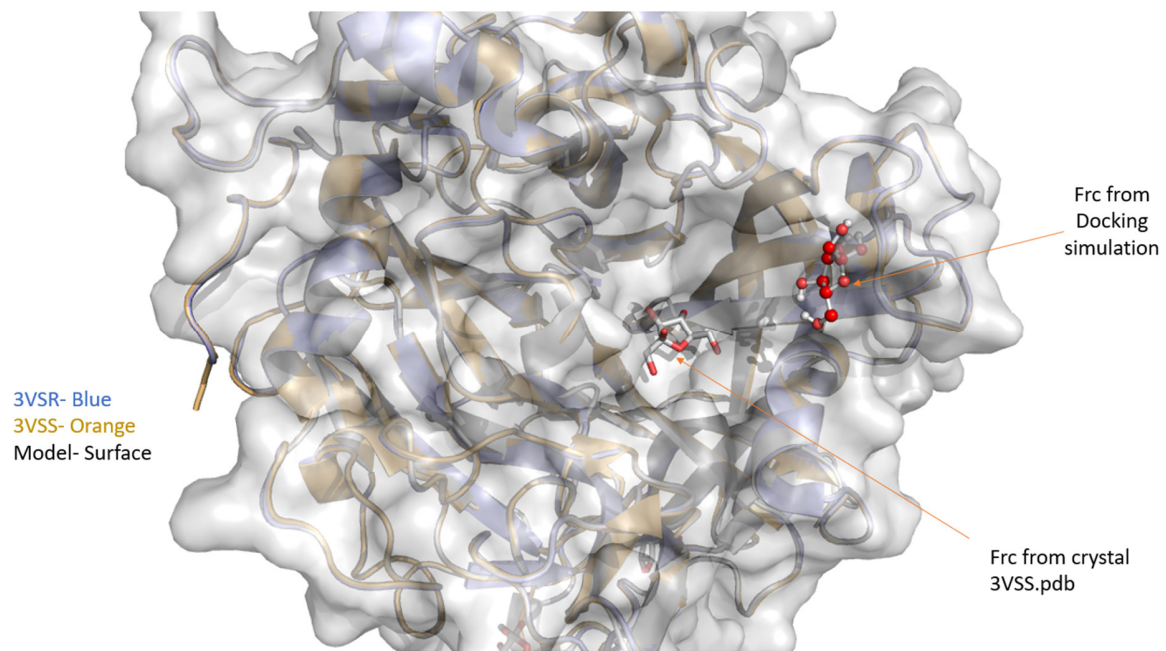
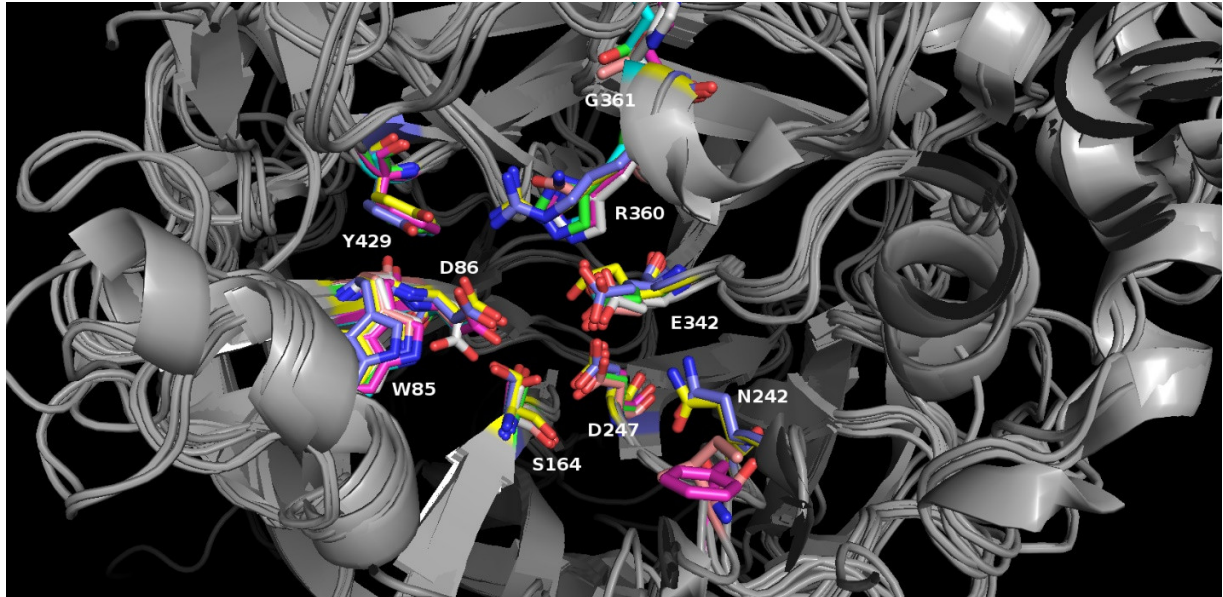


Figure S4: Comparison of the fructose docked in a position representing the least binding energy state generated by docking simulations using the unliganded (3VSR) LS homology model for *V. natriegens*. It was developed using *Microbacterium saccharophilum* K-1 beta-fructofuranosidase. The image shows 3VSR (unliganded), 3VSS (bound Frc).



*Figure S5: Overlay of the homology models developed for the LSs in this study.*

Table S2: Percent bioconversion of various monosaccharides, disaccharides, and trisaccharides as acceptor/donor molecules by various LSs as a function of time

		<i>Vibrio natriegens</i>	<i>Gluconobacter oxydans</i>	<i>Novosphingobium aromaticivorans</i>	<i>Burkholderia graminis</i>	<i>Berjinkia indica subsp. indica</i>
	Reaction time (h)	Average conversion (%)	Average conversion (%)	Average conversion (%)	Average conversion (%)	Average conversion (%)
<i>0.9 M Sucrose</i>	2	34.1 ± 11.9	33.9 ± 3.4	20.5 ± 7.3	16.9 ± 0.2	23.4 ± 0.7
	24	54.7 ± 6.4	61.5 ± 3.3	23.0 ± 6.0	61.0 ± 1.4	51.0 ± 1.0
	50	59.7 ± 6.1	77.9 ± 0.5	52.9 ± 3.8	62.5 ± 6.7	72.2 ± 2.1
<i>0.9 M Raffinose</i>	2	37.1 ± 2.9	92.7 ± 0.8	32.4 ±	54.9 ± 20.1	80.9 ±
	24	61.1 ± 2.1	48.4 ±	64.6 ± 6.0	30.4 ±	-
	50	73.0 ± 2.7	58.3 ± 5.4	64.0 ± 2.9	68.2 ± 5.5	66.5 ± 7.8
<i>0.45 M Raffinose / 0.9 M Sucrose</i>	2	52.2 ± 14.1	27.2 ±	26.4 ± 3.	11.7 ± 4.9	7.7 ±
	24	52.2 ± 3.2	20.5 ± 5.0	27.6 ± 2.6	36.7 ±	15.9 ±
	50	69.7 ± 12.4	71.0 ± 6.0	56.9 ± 2.8	69.7 ± 3.4	66.3 ± 7.9
<i>0.45 M Galactose / 0.9 M Sucrose</i>	2	73.3 ± 4.9	52.3 ± 5.7	12.9 ±	48.0 ± 3.0	58.8 ± 1.8
	24	76.2 ± 1.2	50.4 ± 3.1	41.7 ± 19.7	48.9 ± 7.3	66.6 ± 0.3
	50	34.2 ± 2.9	45.9 ± 2.4	46.9 ± 0.7	58.2 ± 2.5	46.2 ± 4.3
<i>0.45 M Maltose / 0.9 M Sucrose</i>	2	38.3 ± 15.3	34.4 ±	55.2 ±	59.8 ±	62.2 ± 0.2
	24	38.0 ±	41.7 ± 11.9	68.8 ± 3.2	58.6 ±	73.8 ± 0.1
	50	72.0 ± 5.4	52.5 ± 6.4	38.6 ± 0.2	56.1 ± 11.3	51.3 ± 1.1
<i>0.45 M Lactose / 0.9 M Sucrose</i>	2	68.7 ± 4.2	46.2 ± 8.6	67.9 ± 12.0	53.07 ± .02	54.9 ± 3.2
	24	67.1 ± 28.4	65.8 ± 8.2	18.9 ± 13.2	53.5 ± 7.5	78.1 ± 1.5
	50	60.4 ± 17.0	44.1 ± 1.8	67.2 ± 4.0	78.7 ± 2.6	71.1 ± 1.0
<i>0.45 M Xylose / 0.9 M Sucrose</i>	2	84.3 ± 1.8	60.6 ± 4.0	84.0 ± 4.0	62.1 ± 11.4	60.2 ± 13.4
	24	79.3 ±	66.8 ± 7.1	73.6 ± 2.0	62.3 ± 10.2	73.3 ± 2.4
	50	70.0 ± 8.5	62.8 ± 6.1	73.1 ± 4.3	67.2 ± 5.0	73.1 ± 0.3
<i>0.45 M Sorbitol / 0.9 M Sucrose</i>	2	59.3 ± 2.8	39.0 ± 25.0	29.8 ± 17.2	25.9 ± 4.3	49.6 ± 3.9
	24	59.4 ± 1.3	45.0 ± 0.9	43.4 ±	-	55.7 ± 1.2
	50	50.1 ± 2.0	45.4 ± 5.1	43.8 ± 2.4	42.0 ± 6.1	36.1 ± 2.8
<i>0.45 M Catechol / 0.9 M Sucrose</i>	2	-	6.8 ±	29.8 ± 17.2	-	50.3 ± 4.1
	24	20.1 ±	-	17.3 ±	-	-
	50	45.2 ± 16.5	-	38.8 ± 0.6	36.4 ± 18.3	61.1 ± 5.1