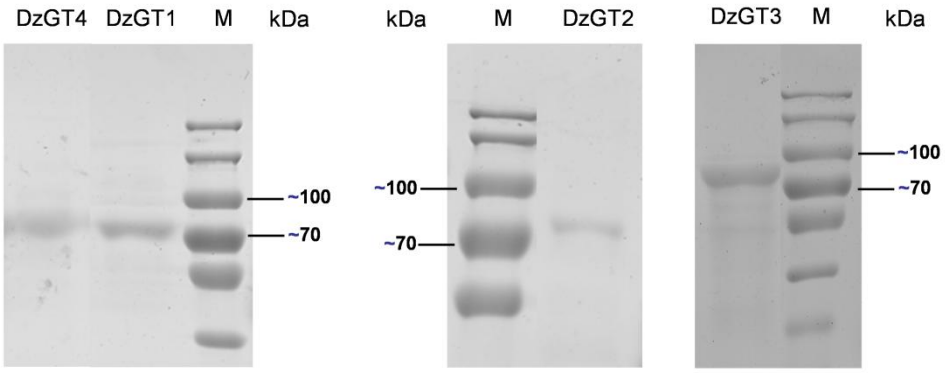
Supplementary Material

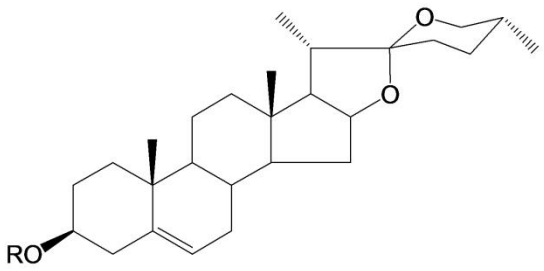
1 Supplementary Figures and Tables

1.1 Supplementary Figures

Supplementary Figure 1. The amino acid sequence alignment of DzGT1-DzGT4. The putative plant secondary product glycosyltransferase (PSPG) box is highlighted by red colour.



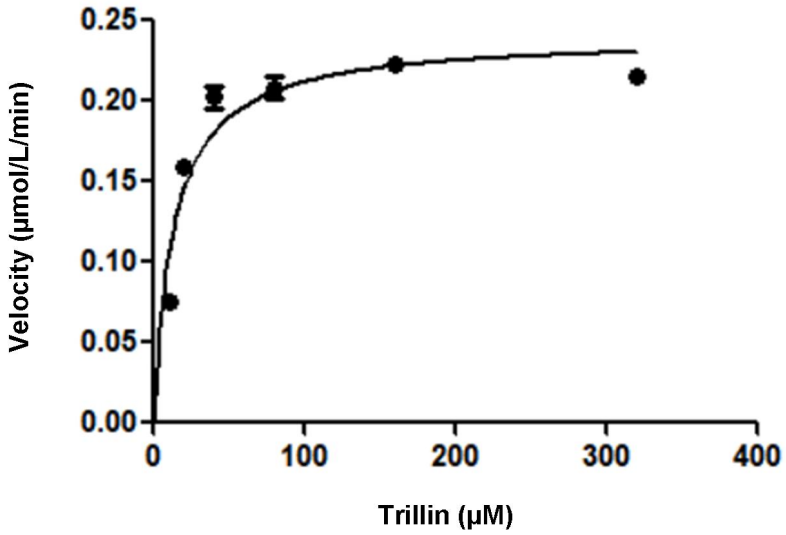
Supplementary Figure 2. SDS-PAGE electrophoresis analysis of the purified recombinant DzGTs.



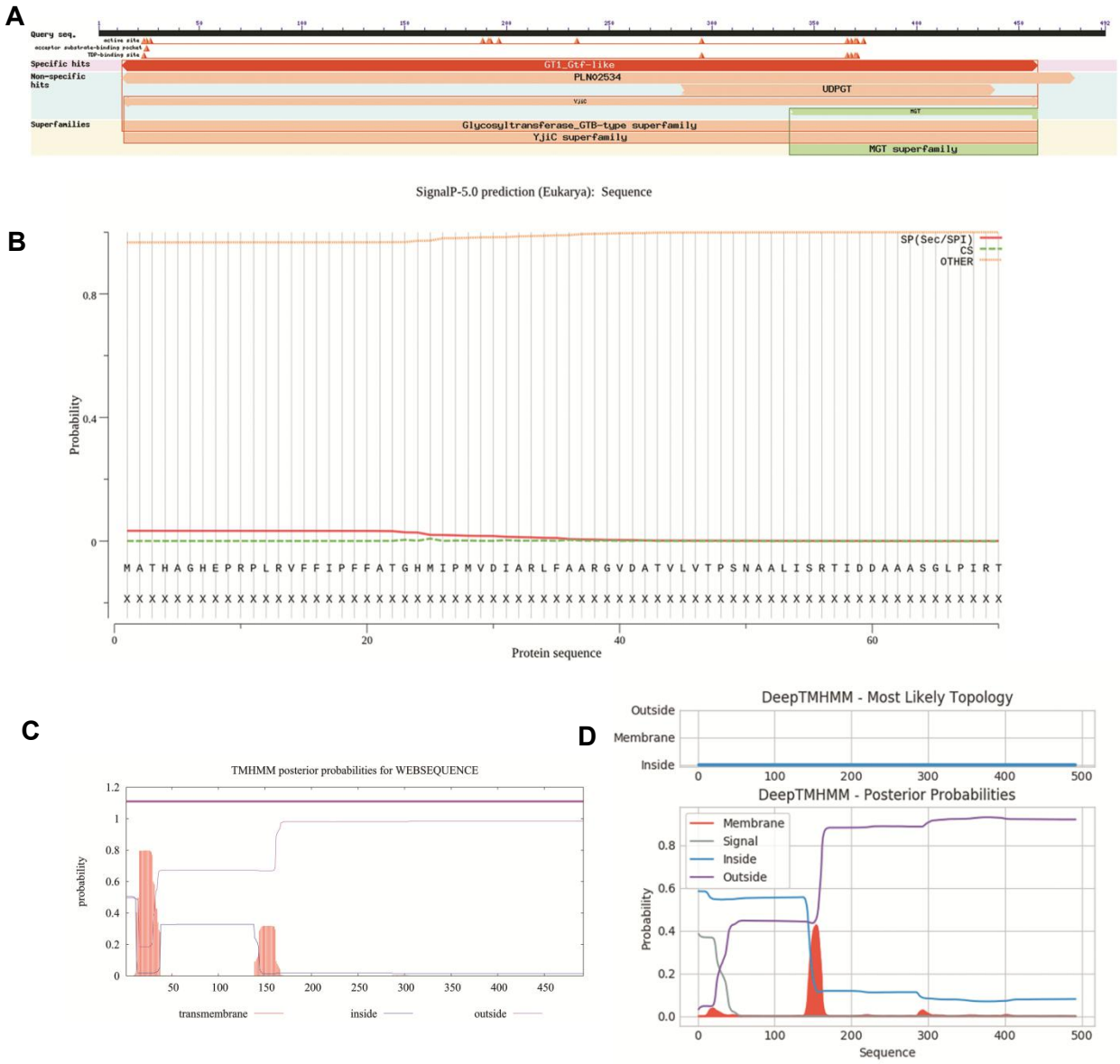
R

Trillin:	Glc
PSA:	Rha(1→2)-Glc
Dioscin:	Rha(1→4)-[Rha(1→2)]-Glc
Deltonin:	Glc(1→4)-[Rha(1→2)]-Glc
Gracillin:	Glc(1→3)-[Rha(1→2)]-Glc
Zingiberensis newsaponin:	Glc(1→3)-Glc(1→4)-[Rha(1→2)]-Glc

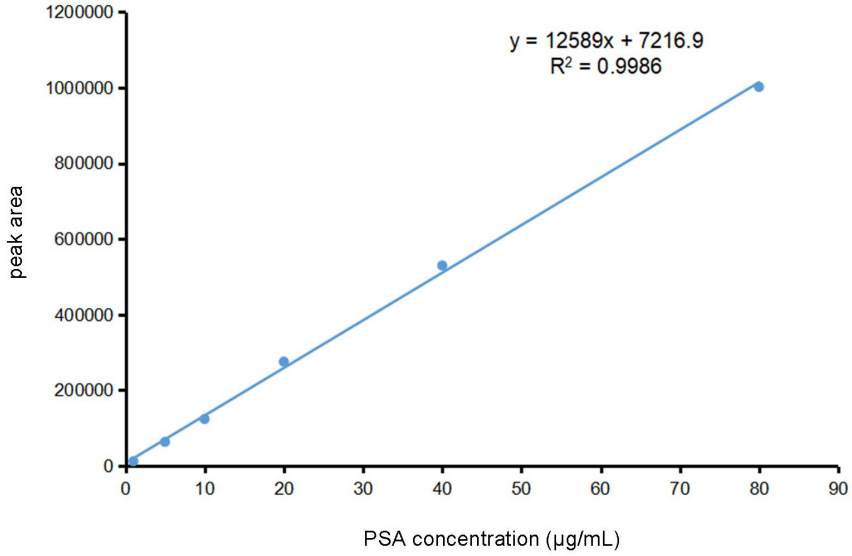
Supplementary Figure 3. Structure of the substrates used for the enzyme assays of DzGT1.



Supplementary Figure 4. Nonlinear regression analysis of DzGT1 with trillin using the GraphPad Prism 5.01 software.The best-fit regression equations is V = 0.24[S]/(13.05 + [S]). V, enzyme velocity (µmol/L/min); [S], substrate (trillin) concentration.



Supplementary Figure 5. Bioinformatic analysis of DzGT1. **(A)** Putative conserved domains of DzGT1 detected by the NCBI conserved domains database. **(B)** Signal peptide prediction of DzGT1 with SignalP 5.0. **(C, D)** Transmembrane topology prediction of DzGT1 with TMHMM2 and DeepTMHMM.



Supplementary Figure 6. The calibration curve of PSA. The linear fit equation is y = 12589x+ 7216.9 and R^2 = 0.9986.

1.2 Supplementary Tables

Supplementary Table 1. Information of the glycosyltransferases used in the phylogenetic analysis

Protein	GenBank accession Number	Plant	Function
UGT80A1	CAB06081	*Avena sativa*	Sterol 3-*O*-glucosyltransferase
UGT80B1	OAP13743	*Arabidopsis thaliana*	Sterol 3-*O*-glucosyltransferase
GhSGT1	AHX00584	*Gossypium hirsutum*	Sterol 3-*O*-glucosyltransferase
SGTL1	ABC96116	*Withania somnifera*	Sterol 3-*O*-glucosyltransferase
Dz3GT1	AVI57699	*Dioscorea zingiberensis*	Sterol 3-*O*-glucosyltransferase
Dz3GT2	AVI57700	*D. zingiberensis*	Sterol 3-*O*-glucosyltransferase
GmSGT3	NP_001240857	*Glycine max*	soyasaponin III rhamnosyltransferase
StSGT3	NP_001274799	*Solanum tuberosum*	β-solanine/β-chaconine rhamnosyltransferase

Supplementary Table 2. Primers used for the gene amplifications in this study

Name	Primers 5' to 3' sequence
For pMD18-T /pGEX-2T	
DzGT1-F	GATCTGGTTCCGCGTGGATCCATGGCCACCCATGCC
DzGT1-R	TCAGTCAGTCACGATGAATTCTCAAGCGGTAGCAGCATC
DzGT2-F	GATCTGGTTCCGCGTGGATCCATGACTTCAAAAGCCAAAGA
DzGT2-R	TCAGTCAGTCACGATGAATTCTTATTGCTTGGACAGATTCA
DzGT3-F	GATCTGGTTCCGCGTGGATCCATGGGGACTGAAACAAAAAC
DzGT3-R	TCAGTCAGTCACGATGAATTCCTAGTTTGAAGAGATTTCTACTTT
DzGT4-F	GATCTGGTTCCGCGTGGATCCATGGGATCCAACAGTACTACT
DzGT4-R	TCAGTCAGTCACGATGAATTCCTAGTATAAACTAGGTACTGGTCT
For real-time PCR	
Actin-F	TGCTGGATTCTGGTGATGGT
Actin-R	CCCGTTCTGCGGTAGTAGTG
DzGT1-F	TTCCTCCGCTCCGAAAACA
DzGT1-R	CGAACCACGTCCGCATATTG