SUPPORTING INFORMATION

Structure-based protein engineering enables prenyl donor switching of a fungal aromatic prenyltransferase

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Table S1 Primers used in this study and protein yields

Mutant	Primer	Sequence (5`→3`)	Yield [mg/L]*
fgaPT2			3.3
M328A	328_GNW_f	TGCCGCTT**GCA**GCCAATTTCACCCTGCACCAGAATGACC	2.1
	328_GNW_r	AATTGGC**TGC**AAGCGGCAGCCTCTCGTCTGGGATAACC	
M328V	328_NNN_f	TGCCGCTT**GTG**GCCAATTTCACCCTGCACCAGAATGACC	1.5
	328_NNN_r	AATTGGC**CAC**AAGCGGCAGCCTCTCGTCTGGGATAACC	
M328L	328_CTA_f	TGCCGCTT**CTA**GCCAATTTCACCCTGCACCAGAATGACC	1.6
	328_CTA_r	AATTGGC**TAG**AAGCGGCAGCCTCTCGTCTGGGATAACC	
M328I	328_AWW_f	TGCCGCTT**ATT**GCCAATTTCACCCTGCACCAGAATGACC	1.6
	328_AWW_r	AATTGGC**AAT**AAGCGGCAGCCTCTCGTCTGGGATAACC	
M328P	328_NNN_f	TGCCGCTT**CCT**GCCAATTTCACCCTGCACCAGAATGACC	1.8
	328_NNN_r	AATTGGC**AGG**AAGCGGCAGCCTCTCGTCTGGGATAACC	
M328W	328_NNN_f	TGCCGCTT**TGG**GCCAATTTCACCCTGCACCAGAATGACC	2.7
	328_NNN_r	AATTGGC**CCA**AAGCGGCAGCCTCTCGTCTGGGATAACC	
M328F	328_TTT_f	TGCCGCTT**TTT**GCCAATTTCACCCTGCACCAGAATGACC'	1.5
	328_TTT_r	AATTGGC**AAA**AAGCGGCAGCCTCTCGTCTGGGATAACC	
M328Y	328_TNT_f	TGCCGCTT**TAT**GCCAATTTCACCCTGCACCAGAATGACC	2.5
	328_TNT_r	AATTGGC**ATA**AAGCGGCAGCCTCTCGTCTGGGATAACC	
M328T	328_NNN_f	TGCCGCTT**ACG**GCCAATTTCACCCTGCACCAGAATGACC	2.0
	328_NNN_r	AATTGGC**CGT**AAGCGGCAGCCTCTCGTCTGGGATAACC	
M328Q	328_NNN_f	TGCCGCTT**CAA**GCCAATTTCACCCTGCACCAGAATGACC	1.6
	328_NNN_r	AATTGGC**TTG**AAGCGGCAGCCTCTCGTCTGGGATAACC	
M328G	328_NNN_f	TGCCGCTT**GGG**GCCAATTTCACCCTGCACCAGAATGACC	2.2
	328_NNN_r	AATTGGC**CCC**AAGCGGCAGCCTCTCGTCTGGGATAACC	
M328S	328_NNN_f	TGCCGCTT**TCT**GCCAATTTCACCCTGCACCAGAATGACC	1.6
	328_NNN_r	AATTGGC**AGA**AAGCGGCAGCCTCTCGTCTGGGATAACC	
M328C	328_TNT_f	TGCCGCTT**TGT**GCCAATTTCACCCTGCACCAGAATGACC	2.6
	328_TNT_r	AATTGGC**ACA**AAGCGGCAGCCTCTCGTCTGGGATAACC	
M328N	328_AWW_f	TGCCGCTT**AAT**GCCAATTTCACCCTGCACCAGAATGACC	3.2
	328_AWW_r	AATTGGC**ATT**AAGCGGCAGCCTCTCGTCTGGGATAACC	
M328K	328_AWW_f	TGCCGCTT**AAA**GCCAATTTCACCCTGCACCAGAATGACC	2.7
	328_AWW_r	AATTGGC**TTT**AAGCGGCAGCCTCTCGTCTGGGATAACC	
M328R	328_NNN_f	TGCCGCTT**CGG**GCCAATTTCACCCTGCACCAGAATGACC	1.7
	328_NNN_r	AATTGGC**CCG**AAGCGGCAGCCTCTCGTCTGGGATAACC	
M328H	328_CAT_f	TGCCGCTT**CAT**GCCAATTTCACCCTGCACCAGAATGACC	3.1
	328_CAT_r	AATTGGC**ATG**AAGCGGCAGCCTCTCGTCTGGGATAACC	
M328E	328_GNW_f	TGCCGCTT**GAA**GCCAATTTCACCCTGCACCAGAATGACC	2.0
	328_GNW_r	AATTGGC**TTC**AAGCGGCAGCCTCTCGTCTGGGATAACC	
M328D	328_GAC_f	TGCCGCTT**GAC**GCCAATTTCACCCTGCACCAGAATGACC	1.5
	328_GAC_r	AATTGGC**GTC**AAGCGGCAGCCTCTCGTCTGGGATAACC	
L263A_M328A	L263A_f	CTACCTG**GCA**GAGCAGATGGTTTCACTAGAAGCCATGGAGG	1.3
	L263A_r	TCTGCTC**TGC**CAGGTAGATCTTGATTCTCGACTTGGCAGG	
L263A_M328A_Y398F	Y398F_f	CCACGCC**TTT**ATATCCTTCTCCTACAGGGACCGTACC	1.3
	Y398F_r	AAGGATAT**AAA**GGCGTGGAGGTAGTTAAGTTTGTCATGATCCG	

* The purity of the recombinant proteins was determined at 75% − 80%. The protein yields were calculated by taking the impurities into consideration.

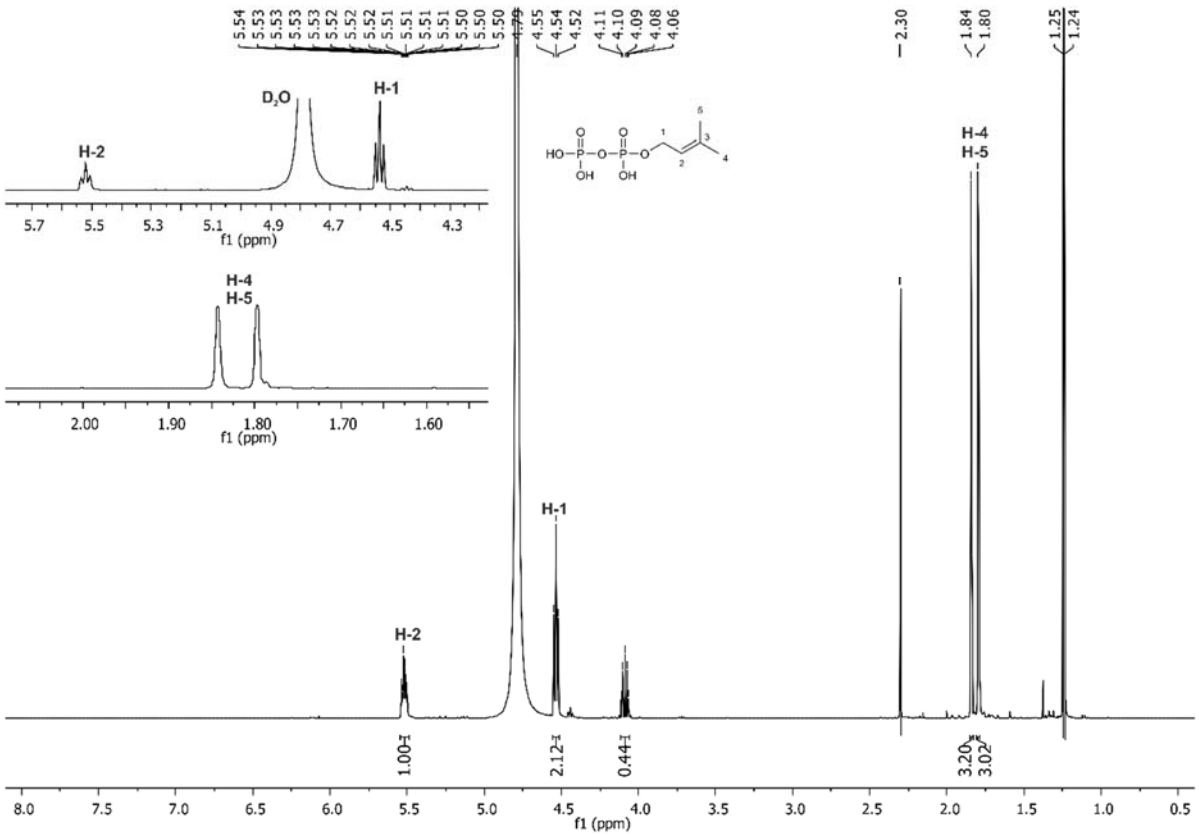

Figure S1 ^{1}H NMR spectrum of dimethylallyl diphosphate (DMAPP) (500 MHz, D_2O)

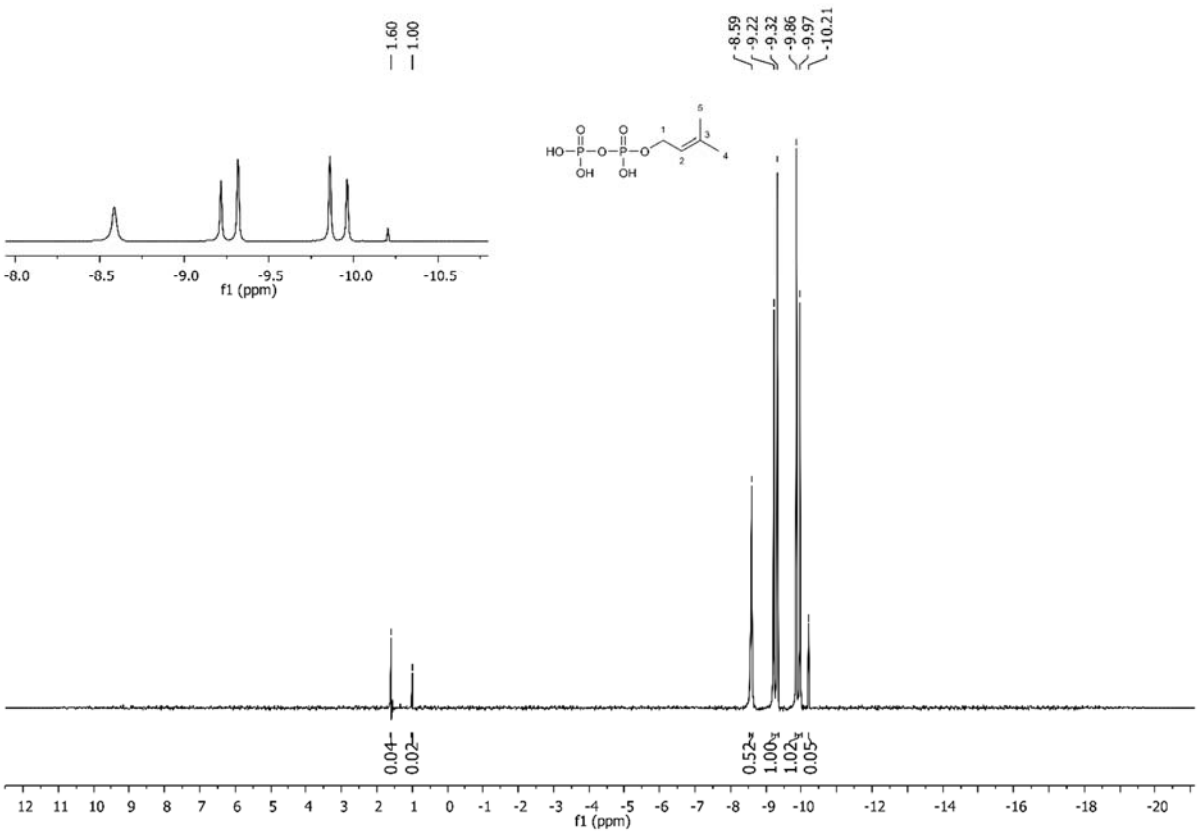

Figure S2 ^{31}P NMR spectrum of dimethylallyl diphosphate (DMAPP) (200 MHz, D_2O)

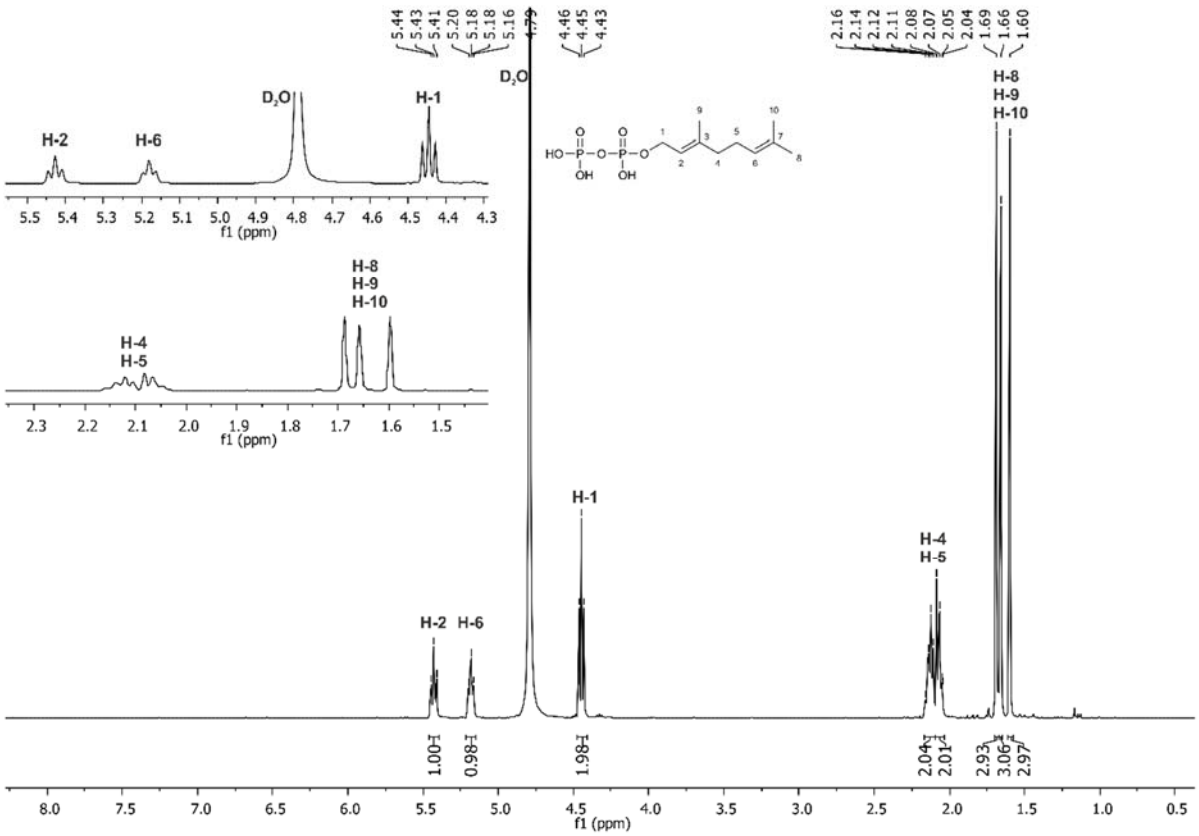

Figure S3 ^{1}H NMR spectrum of geranyl diphosphate (GPP) (500 MHz, D_2O)

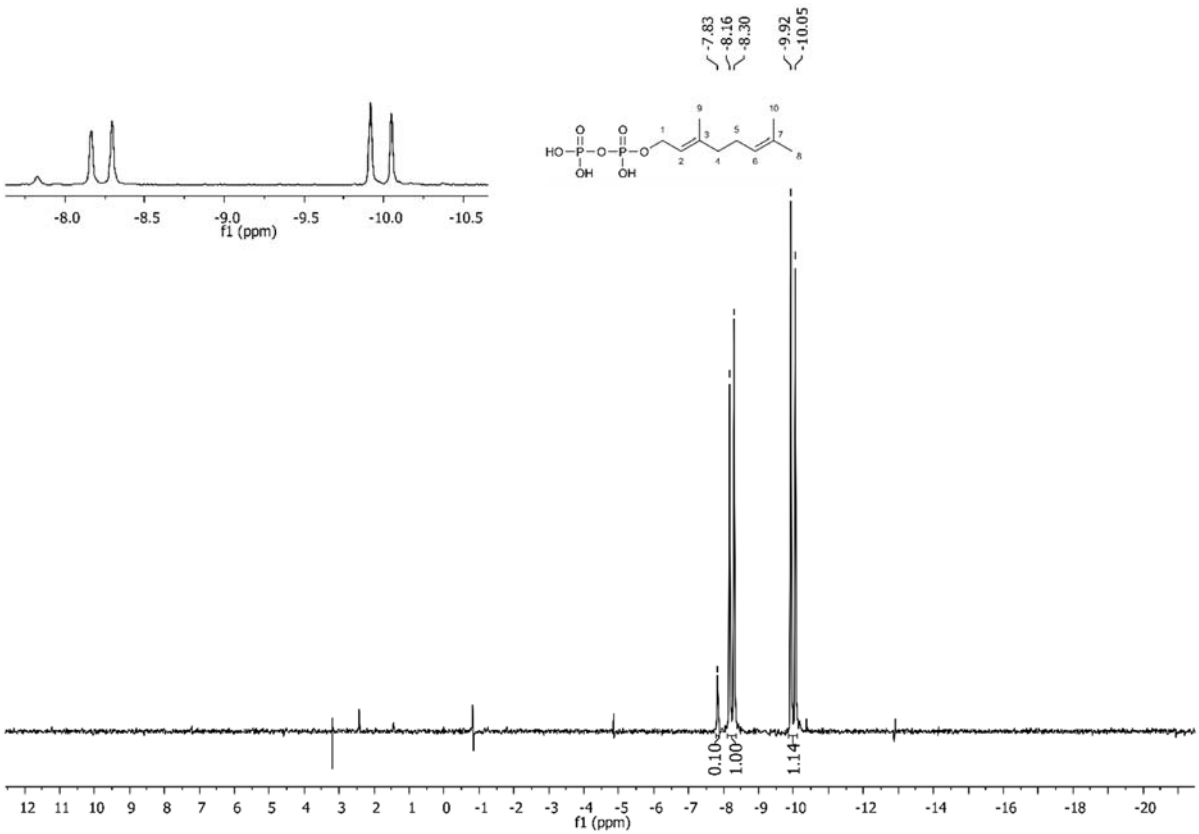

Figure S4 ^{31}P NMR spectrum of geranyl diphosphate (GPP) (200 MHz, D_2O)

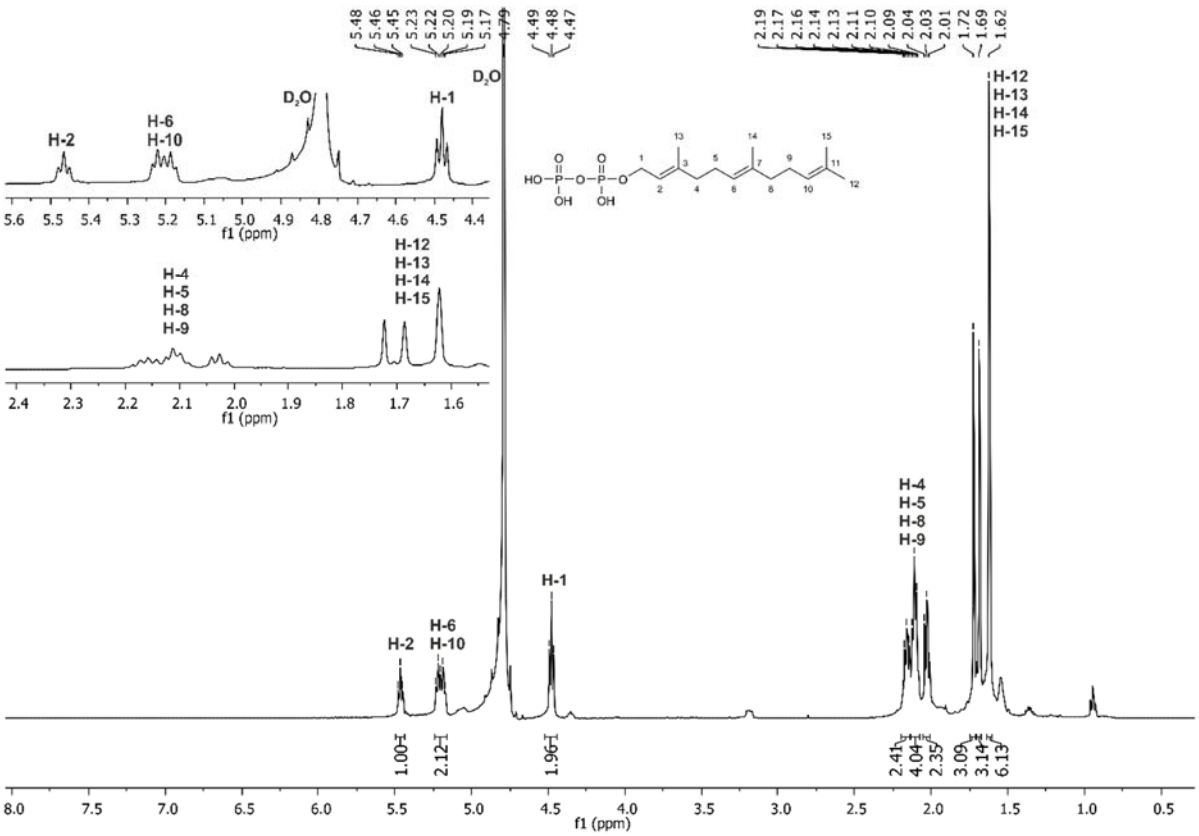

Figure S5 ^{1}H NMR spectrum of farnesyl diphosphate (FPP) (500 MHz, D_2O)

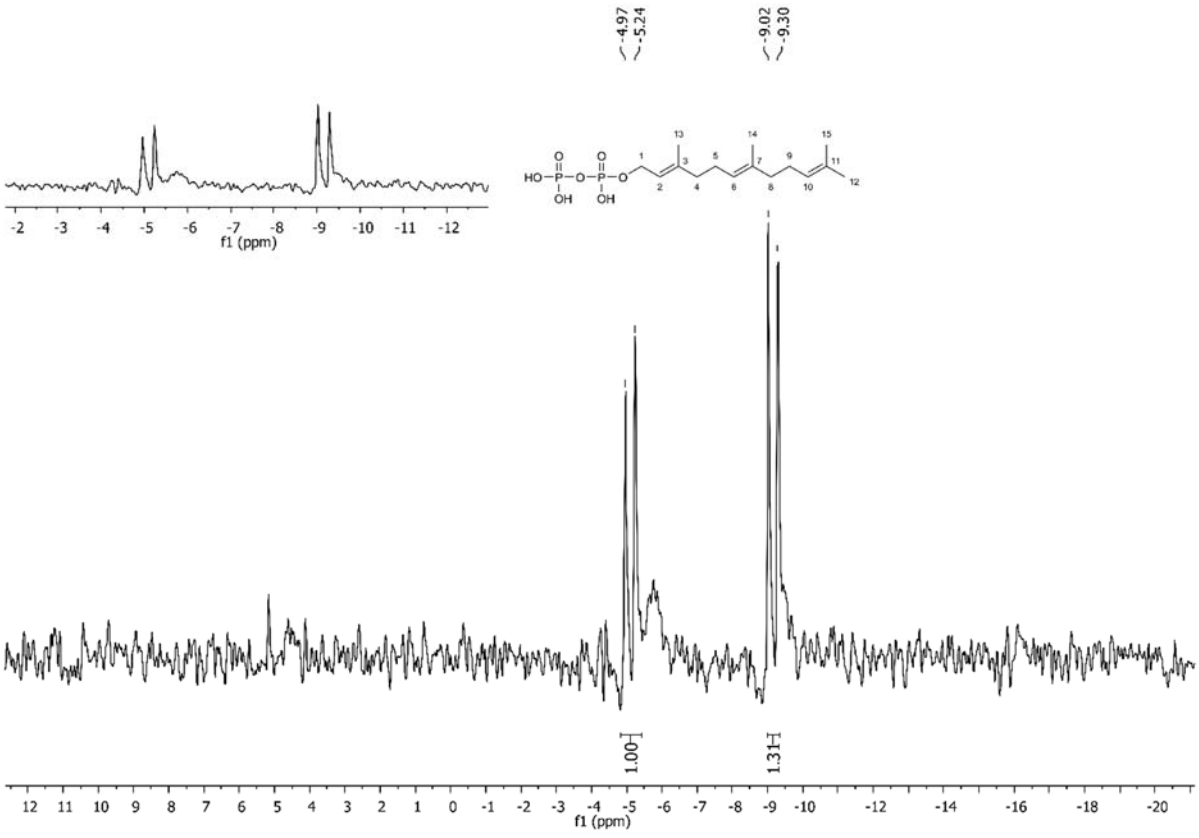

Figure S6 ^{31}P NMR spectrum of farnesyl diphosphate (FPP) (200 MHz, D_2O)

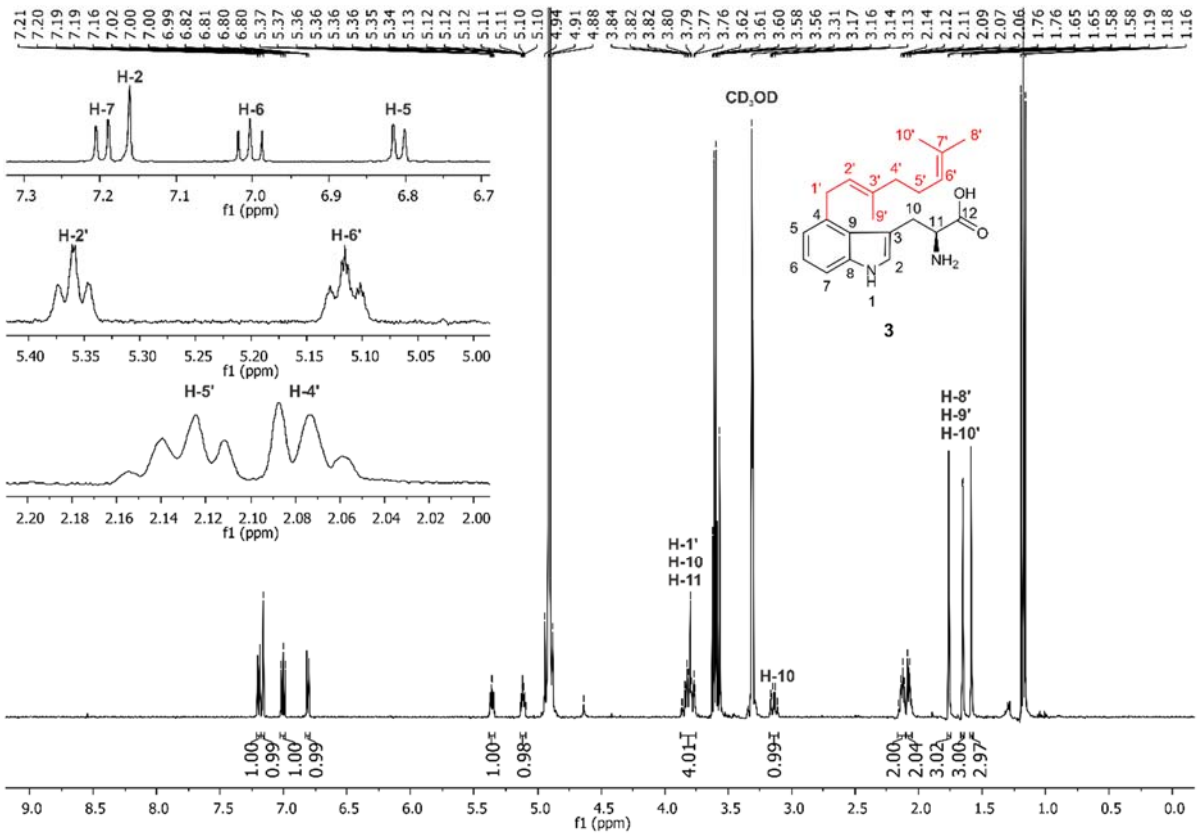

Figure S7 ^{1}H NMR spectrum of 4-geranyltryptophan (**3**) (500 MHz, CD_3OD)

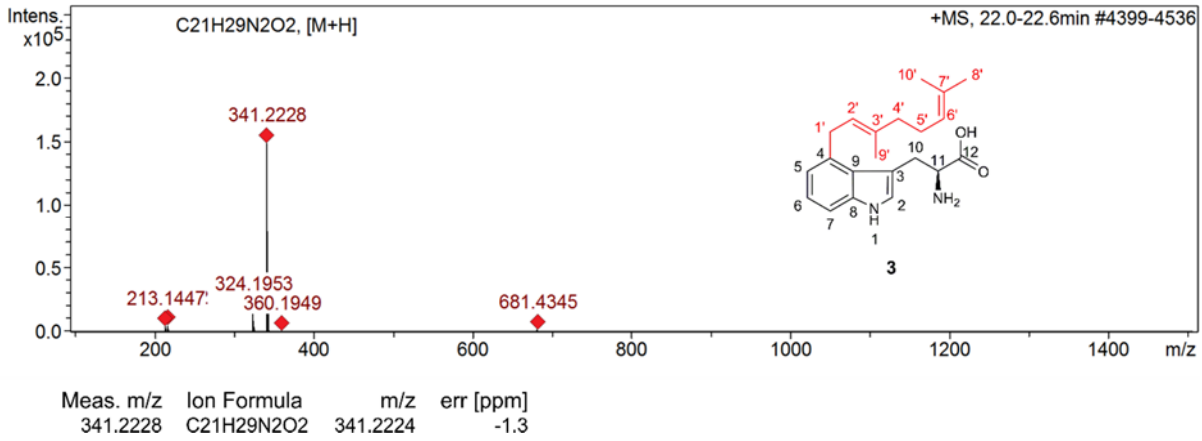

Figure S8 Positive HR-ESI-MS spectrum of 4-geranyltryptophan (**3**)

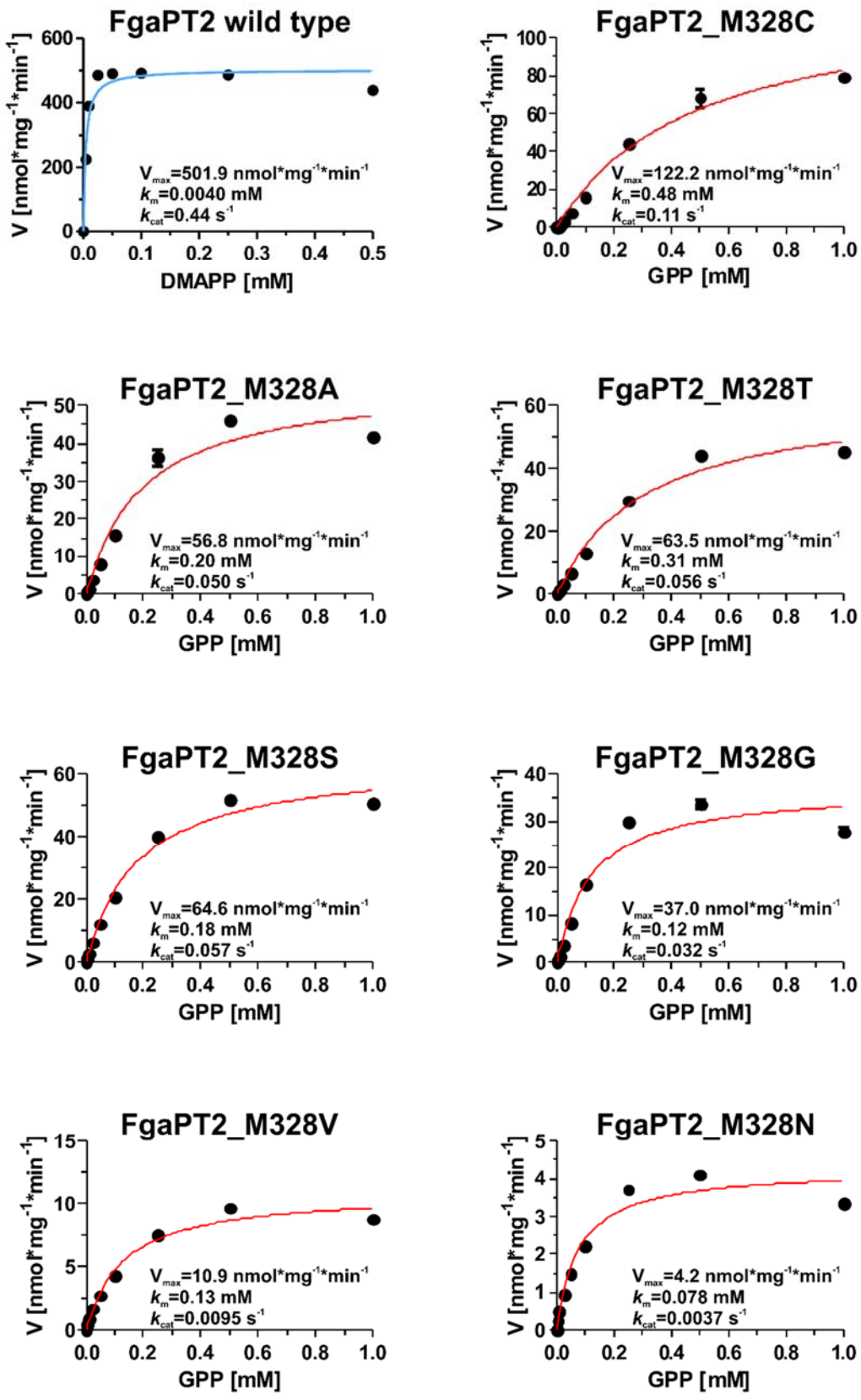

Figure S9 Kinetic parameters of FgaPT2 wild type and FgaPT2_M328 mutants toward DMAPP and GPP with **1**