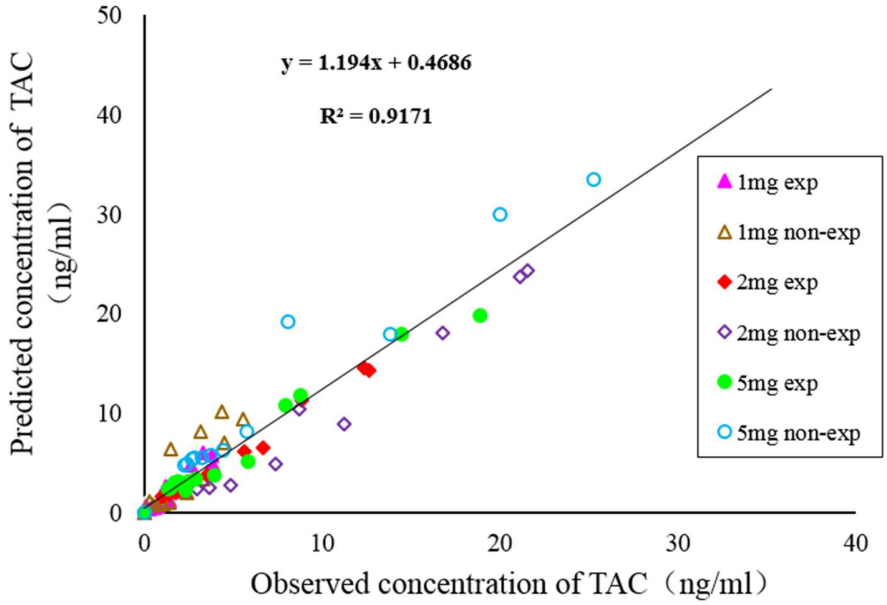

Figure S1. Linear analysis of the predicted concentration and observed concentration of tacrolimus

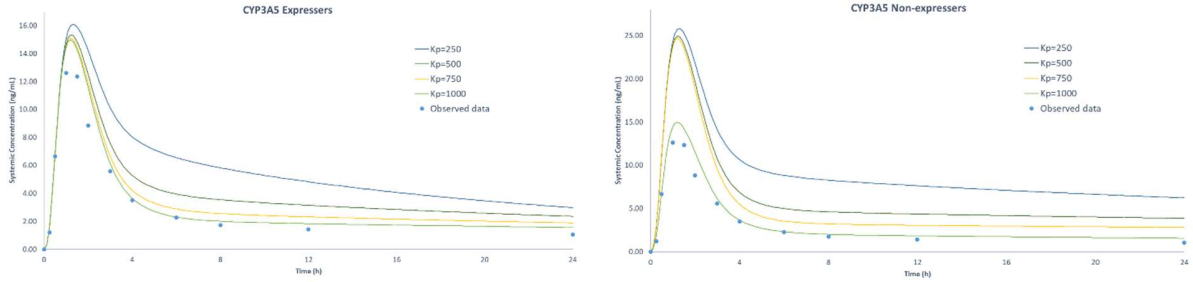

Figure S2. Sensitivity analysis of adipose distribution on tacrolimus pharmacokinetics in CYP3A5 expressers and non-expressers. Blood concentration of tacrolimus following a single oral dose of 2 mg was simulated with K_P value of adipose tissue varying from 250 to 1000 by 250.

Table S1 Data points for RI and TDI assays with mean and standard variation values

Figure 2A								
	0.2 μM tacrolimus		**0.4 μM tacrolimus**		**0.8 μM tacrolimus**		**1.6 μM tacrolimus**	
STA	**Mean**	**SD**	**Mean**	**SD**	**Mean**	**SD**	**Mean**	**SD**
0 μM	8.5682	0.1101	7.3548	1.2723	4.5004	0.1763	2.3328	0.0625
0.125 μM	17.0000	1.0000	9.0441	1.0494	7.1353	0.6206	2.6233	0.6790
0.25 μM	21.0000	1.0000	14.8861	0.4893	8.3071	0.6418	3.9072	0.6564
0.5 μM	35.0000	3.0000	23.9051	1.2189	13.9244	1.6474	5.0160	0.7723
Figure 2B								
	0.2 μM tacrolimus		**0.4 μM tacrolimus**		**0.8 μM tacrolimus**		**1.6 μM tacrolimus**	
STA	**Mean**	**SD**	**Mean**	**SD**	**Mean**	**SD**	**Mean**	**SD**
0 μM	8.0000	2.7759	7.5682	0.1101	6.3548	1.2723	5.1363	1.1036
0.125 μM	15.0000	0.3663	14.0109	1.4711	7.5773	1.6034	5.6992	0.6790
0.25 μM	23.2012	1.0138	15.6487	0.8529	12.0521	2.6740	5.8700	0.6564
0.5 μM	30.8928	3.5033	26.2841	1.6422	14.9975	2.4334	8.0120	0.7723
Figure 2C								
	0.2 μM tacrolimus		**0.4 μM tacrolimus**		**0.8 μM tacrolimus**		**1.6 μM tacrolimus**	
SIA	**Mean**	**SD**	**Mean**	**SD**	**Mean**	**SD**	**Mean**	**SD**
0 μM	19.7089	0.5288	9.0000	1.4052	5.2438	1.4052	1.5764	1.1963
2.4 μM	20.0000	1.1414	10.0000	0.8362	3.3452	0.8362	1.4662	0.0139
7.2 μM	18.8974	1.8284	10.0565	1.1621	3.6950	1.1621	1.7598	0.1414
12 μM	20.3445	1.2828	9.8175	1.2527	4.5304	1.2327	1.6431	0.0012
Figure 2D								
	0.2 μM tacrolimus		**0.4 μM tacrolimus**		**0.8 μM tacrolimus**		**1.6 μM tacrolimus**	
SIA	**Mean**	**SD**	**Mean**	**SD**	**Mean**	**SD**	**Mean**	**SD**
0 μM	28.0000	2.0288	14.9302	3.4052	8.4000	1.4052	4.4803	1.1963
2.4 μM	32.0000	1.1414	17.6167	0.8362	9.2000	0.8362	6.2790	0.0139
7.2 μM	43.0000	3.8284	21.6176	1.1621	12.0300	1.1621	6.9243	0.1414
12 μM	60.0000	2.8284	30.1990	3.2527	15.0500	1.2327	7.2381	0.0012
Figure 3A										
	2 μM STA		1 μM STA		0.5 μM STA		0.25 μM STA		0 μM STA	
Time	**Mean**	**SD**	**Mean**	**SD**	**Mean**	**SD**	**Mean**	**SD**	**Mean**	**SD**
30 min	3.0051	0.3321	3.5554	0.3885	3.9183	0.0745	4.2407	0.0685	4.6052	0.0000
20 min	3.1694	0.3080	3.8785	0.2297	3.9359	0.0519	4.2986	0.0755	4.6052	0.0000
10 min	3.5615	0.2198	4.0515	0.1355	4.0778	0.1705	4.3386	0.0879	4.6052	0.0000
5 min	4.0322	0.1220	4.1658	0.1597	4.2393	0.1871	4.4164	0.0627	4.6052	0.0000
0 min	4.6052	0.0000	4.5902	0.0263	4.6010	0.0073	4.6010	0.0087	4.6052	0.0000
Figure 4A										
	16 μM SIA		8 μM SIA		4 μM SIA		2 μM SIA		0 μM SIA	
Time	**Mean**	**SD**	**Mean**	**SD**	**Mean**	**SD**	**Mean**	**SD**	**Mean**	**SD**
30 min	4.0738	0.0015	4.1876	0.0077	4.2490	0.0142	4.3520	0.0200	4.6052	0.0000

20 min	4.2397	0.0074	4.2781	0.0201	4.3925	0.0448	4.4602	0.0351	4.6052	0.0000
10 min	4.3776	0.0019	4.3979	0.0093	4.4420	0.0329	4.5165	0.0465	4.6052	0.0000
5 min	4.4518	0.0063	4.5164	0.0047	4.5505	0.0164	4.5754	0.0186	4.6052	0.0000
0 min	4.6052	0.0000	4.6029	0.0023	4.6052	0.0000	4.6052	0.0000	4.6052	0.0000
Figure 4C										
	16 μM SIA		8 μM SIA		4 μM SIA		2 μM SIA		0 μM SIA	
Time	**Mean**	**SD**	**Mean**	**SD**	**Mean**	**SD**	**Mean**	**SD**	**Mean**	**SD**
30 min	4.1607	0.0015	4.2282	0.0077	4.2859	0.0142	4.3791	0.0200	4.6052	0.0000
20 min	4.2585	0.0074	4.4028	0.0201	4.4869	0.0448	4.4427	0.0351	4.6052	0.0000
10 min	4.3701	0.0019	4.4150	0.0093	4.5031	0.0329	4.5218	0.0465	4.6052	0.0000
5 min	4.4523	0.0063	4.4632	0.0047	4.4998	0.0164	4.5539	0.0186	4.6052	0.0000
0 min	4.5539	0.0000	4.6052	0.0023	4.6052	0.0000	4.6052	0.0000	4.6052	0.0000

Table S2. Optimized detection parameters and LC–MS/MS conditions

Analytes	**MRM transition(m/z)**	**DP (V)**	**CE (V)**	**LLOQ (ng/ml)**	**Retention time (min)**
Tacrolimus	821.5→768.6	25	30	1	3.75
Ascomycin	810.7→757.6	100	30	—	3.75
6β-hydroxyl testosterone	305.3→269.2	78	22	10	2.79
Prednisolone	361.3→343.2	22	15	—	2.79

Table S3 Parameters used for PBPK modeling of schisantherin A and schisandrin A

Parameters	**Schisantherin A**		**Schisandrin A**	
	Value	**Ref.**	**Value**	**Ref.**
Molecular weight (g/mol)	536.58	-	416.52	-
Compound type	neutral	-	neutral	-
Log $P_{o:w}$	1.7	In-house data	5.265	[1]
$f_{u\ plasma}$	0.083	[2]	0.0334	[2]
$f_{u\ gut}$	1	default value	1	default value
B/P	0.63	Predicted by Gastroplus®	0.69	Predicted by Gastroplus®
Intrinsic solubility (mg/mL)	0.01	Predicted by Gastroplus®	0.000668	Predicted by Gastroplus®
$P_{app,caco-2}$ ($\times 10^{-6}$cm/s)	34.41	[3]		

PSA (Å^2)	-		55.38	https://pubchem.ncbi.nlm.nih.gov/
HBD	-		0	https://pubchem.ncbi.nlm.nih.gov/
K_p				
Adipose			36.4	
Brain			7.4	
Gut			210.3	
Heart	-	Predicted by Simcyp®	39.5	In-house data
Kidney			5.9	
Liver			3.9	
Lung			19	
Spleen			7.6	
$CL_{int,HLM}$(μL/min/mg protein)	24	In-house data	130	In-house data

Note: Log $P_{o:w}$, octanol/water partition coefficient; fu, fraction of unbound drug in plasma; B/P, blood to plasma partition ratio; $P_{app,caco-2}$,apparent permeability of Caco-2 cell line; PSA, polar surface area; HBD, hydrogen bond donors; K_p, tissue to plasma partition coefficients; $CL_{int,HLM}$, intrinsic clearance in human liver microsomes.

Table S4 Parameters used for PBPK modeling of tacrolimus

Parameter	Tacrolimus	
	Value	**Ref.**
Molecular weight (g/mol)	804.02	Drug label
Compound type	neutral	Drug label
Log$P_{o:w}$	3.26	[4]
$f_{u\ plasma}$	0.013	[4]
$f_{u\ gut}$	1	[5]
B/P	35	[4]
Intrinsic solubility (mg/mL)	0.012	[6]
$P_{app,caco-2,(pH\ 7.4:7.4),tacrolimus}$ (×10^{-6}cm/s)	13.1	[7]
$P_{app,caco-2,(pH\ 7.4:7.4)\ midazolam}$ (×10^{-6}cm/s)	32.4	[7]
CYP3A4 13-DMT *V*max	8 pmol/min/pmol rCYP3A4	[8]
CYP3A5 13-DMT *V*max	17 pmol/min/pmol rCYP3A5	[8]
CYP3A4 13-DMT *K*m, u	0.21 μM	[8]
CYP3A5 13-DMT *K*m, u	0.21 μM	[8]
CYP3A4 12-HT *V*max	0.6 pmol/min/pmol rCYP3A4	[8]
CYP3A5 12-HT *V*max	1.4 pmol/min/pmol rCYP3A5	[8]
CYP3A4 12-HT *K*m, u	0.29 μM	[8]
CYP3A5 12-HT *K*m, u	0.35 μM	[8]
CYP3A4/5 ISEF	0.24 (BD SUP)	Simcyp®
$CL_{R,\ human}$ (L/h^{-1})	0	[9]

Note: Log $P_{o:w}$, octanol/water partition coefficient; *f*u, fraction of unbound drug in plasma; B/P, blood to plasma partition ratio; $P_{app,caco-2}$,apparent permeability of Caco-2 cell line; 13-DMT, 13-O-desmethyl tacrolimus; 12-HT, 12-hydroxy tacrolimus; CL_R, renal clearance.

Table S5 C_{max} and AUC of tacrolimus under different K_p values

	CYP3A5 expresser		CYP3A5 non-expresser	
K_p	C_{max}[a] (ng/mL)	AUC[b] (ng/mL·h)	C_{max} (ng/mL)	AUC (ng/mL·h)
250	16.10 (1.14[c])	137.27 (2.25)	25.84 (1.06)	216.28 (1.82)
500	15.34 (1.09)	100.06 (1.64)	24.98 (1.03)	145.93 (1.23)
750	15.11 (1.07)	82.37 (1.35)	24.71 (1.02)	117. 92 (1.00)
1000	14..99 (1.06)	72.04 (1.18)	24.58 (1.01)	102.80 (1.16)
Observed	14.09	60.83	24.28	119.02

a Cmax: Maximal blood concentration; b AUC: Area under the curve; c fold error

Table S6 Demographic information used for PBPK modeling of tacrolimus

Characteristics	**1 mg tacrolimus**		**2 mg tacrolimus**		**5 mg tacrolimus**	
	CYP3A5 expressers	CYP3A5 non-expressers	CYP3A5 expressers	CYP3A5 non-expressers	CYP3A5 expressers	CYP3A5 non-expressers
Number (n)	12	26	31	40	12	12
Age (years)	23.0±1.0	23.0±3.3	23.0±1.7	23.0±1.8	24.0±1.4	21.0±0.5
Sex (male/female)	Male 100%		Male 100%		Male 100%	
Body weight (kg)	82.0±18.6	85.0±16.5	68.0±8.3	68.0±8.3	82.0±18.6	82.0±18.6
Height (cm)	179.0±8.1	178.0±7.8	174.0±6.4	174.0±6.7	179.0±8.1	179.0±8.1
Hematocrit (%)	44.0±2.7	43.0±3.0	46.0±4.0	46.0±3.8	44.0±2.7	44.0±2.7

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