ELECTRONIC SUPPLEMENTARY INFORMATION

ONIOM Calculations on Serotonin Degradation by Monoamine Oxidase B: Insight into Oxidation Mechanism and Covalent Reversible Inhibition

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Table S1. Important geometrical parameters calculated at ONIOM (M062x/631+G(d,p):PM6) level.

(A) For small model with covalent FAD (M_S-cFAD)

	RC	TS	PC_{inv}	$TS_{N5\text{-}PC}$	$N5_{yl}$	TS_{H2O}	PC
θ(C9-C8-C7-C6)	10.06	8.36	6.06	9.09	7.75	9.53	6.32
θ(C9-C9a-C5a-C6)	8.59	7.70	6.90	8.85	6.46	12.54	8.22
θ(N10-C9a-C5a-N5)	6.37	4.95	3.57	2.30	1.05	12.39	5.61
θ(N10-C10a-C4a-N5)	2.24	6.94	9.86	18.76	19.03	4.37	5.98
θ(N1-C10a-C4a-C4)	2.29	4.91	-0.96	9.03	8.46	3.56	-9.90
θ(N1-C2-N3-C4)	7.54	4.96	4.03	12.25	11.22	2.48	6.73
θ(N1-C10a-C4a-N5)	-176.90	-173.00	-167.45	-161.49	-161.51	-172.87	-175.01
θ(N10-C10a-C4a-C4)	-178.56	-175.15	176.35	-170.72	-171.01	-179.20	171.09
θ(C9a-N10-C10a-C4a)	-2.06	-10.70	-19.87	-28.61	- 32.25	5.78	-20.64
θ(C10a-C4a-N5-C5a)	2.03	3.41	7.20	3.22	6.55	-5.87	14.39
θ(C6-C5a-N5-C4a)	178.09	-175.32	-169.36	-167.13	-161.70	167.20	-167.69
θ(C5a-N5-C4a-C4)	178.75	171.36	173.69	160.98	157.78	-170.62	174.33
θ(C14-H15-N5)		152.13					
R(N11-C4a)	2.73	2.59	3.00	2.90	2.86	3.51	3.09

(B) For small model with noncovalent FAD (M_S-nFAD)

	RC	TS	PC_{inv}	$TS_{N5\text{-}PC}$	$N5_{yl}$
θ(C9-C8-C7-C6)	3.75	2.91	2.31	2.40	2.26
θ(C9-C9a-C5a-C6)	3.68	3.67	3.31	2.84	2.17
θ(N10-C9a-C5a-N5)	2.09	1.03	-2.42	-4.74	-5.16
θ(N10-C10a-C4a-N5)	2.45	5.69	9.12	17.63	19.32
θ(N1-C10a-C4a-C4)	-0.68	1.07	2.95	8.38	11.32
θ(N1-C2-N3-C4)	10.52	4.70	5.93	11.56	13.55
θ(N1-C10a-C4a-N5)	-174.90	-174.91	-169.69	-162.49	-161.58
θ(N10-C10a-C4a-C4)	-178.34	-178.34	-178.24	-171.51	-167.78
θ(C9a-N10-C10a-C4a)	-2.33	-2.33	-7.66	-26.76	-29.69
θ(C10a-C4a-N5-C5a)	-5.42	-5.42	-6.98	-0.41	0.39
θ(C6-C5a-N5-C4a)	176.38	-176.69	-179.10	-167.21	-164.42
θ(C5a-N5-C4a-C4)	-178.29	-178.30	-179.79	162.48	157.51
θ(C14-H15-N5)		149.68			
R(N11-C4a)	2.79	2.61	3.04	2.91	2.92

(C) For big model with covalent FAD (M_B-cFAD) and noncovalent FAD (M_B-nFAD)

	M_B-cFAD				M_B-nFAD			
	RC	**TS**	**PC_{inv}**	**$N5_{yl}$**	**RC**	**TS**	**PC_{inv}**	**$N5_{yl}$**
θ(C9-C8-C7-C6)	9.49	6.67	5.39	5.28	6.44	4.13	0.80	2.56
θ(C9-C9a-C5a-C6)	8.56	6.67	4.78	4.81	6.30	4.64	1.68	2.00
θ(N10-C9a-C5a-N5)	7.15	4.88	0.48	-1.50	4.53	3.02	-0.30	-1.59
θ(N10-C10a-C4a-N5)	4.80	7.60	20.29	19.73	5.38	5.99	13.33	18.24
θ(N1-C10a-C4a-C4)	5.07	8.25	11.53	8.55	1.94	3.67	7.61	11.12
θ(N1-C2-N3-C4)	7.13	7.51	14.79	11.73	10.93	11.59	18.68	17.77
θ(N1-C10a-C4a-N5)	-175.34	-173.16	-161.70	-161.28	-175.60	-176.20	-165.77	-164.00
θ(N10-C10a-C4a-C4)	-174.80	-170.99	-166.47	-170.44	-177.08	-174.14	-173.30	-166.64
θ(C9a-N10-C10a-C4a)	-1.74	-11.77	-37.52	-39.91	0.71	-4.81	-22.12	-34.54
θ(C10a-C4a-N5-C5a)	-1.67	3.54	9.75	12.03	-6.38	-2.24	2.66	8.85
θ(C6-C5a-N5-C4a)	176.71	-174.19	-155.17	-153.97	173.39	179.89	-166.97	-156.44
θ(C5a-N5-C4a-C4)	177.85	166.84	149.28	150.25	-176.83	175.32	164.43	150.33
θ(C14-H15-N5)		152.10				151.08		
R(N11-C4a)	2.78	2.63	3.24	2.84	2.81	2.72	3.02	2.90

Table S2. Calculated energies (E), Gibbs free energies (G) in au and imaginary frequencies for small and big models.

(A) From PM6 and CPCM//M06-2X/6-31+G(d,p)//PM6 calculations for M_S-cFAD

	M_S-cFAD				
	E	**G**	**Single point E**	**G correction**	**Imaginary frequency**
RC	-1.17382502	-0.231974	-4363.804717	0.941852	
TS	-1.12639747	-0.186326	-4363.767506	0.940072	-1691.7
PC_{-+}	-1.19593130	-0.248288	-4363.805305	0.947449	
TS_{inv}	-1.19588461	-0.247872	-4363.806857	0.948013	-147.07
PC_{inv}	-1.20160732	-0.252339	-4363.814550	0.949268	
$TS_{N5\text{-}PC}$	-1.18754511	-0.242536	-4363.816629	0.945009	-141.02
$N5_{yl}$	-1.19665223	-0.249548	-4363.805566	0.947104	
TS_{H2O}	-1.18365757	-0.243322	-4363.797434	0.940336	-880.35
PC	-1.19306579	-0.248614	-4363.805498	0.944452	

(B) From ONIOM (M062X/6-31+G(d,p):PM6 calculations for M_S-cFAD and M_S-nFAD

	M_S-cFAD			M_S-nFAD		
	E	**G**	**Imaginary frequency**	**E**	**G**	**Imaginary frequency**
RC	-2112.141806	-2111.123297		-2113.348535	-2112.31121	
TS	-2112.102057	-2111.086940	-1284.51	-2113.304300	-2112.27284	-1370.88
PC_{inv}	-2112.148398	-2111.129189		-2113.351678	-2112.31566	
$TS_{N5\text{-}PC}$	-2112.139992	-2111.116063	-94.79	-2113.348480	-2112.30804	-110.06
$N5_{yl}$	-2112.141004	-2111.117167		-2113.349169	-2112.30883	
TS_{H2O}	-2112.128868	-2111.114748	-927.68	-	-	
PC	-2112.137444	-2111.120295		-	-	

(C) From PM6 calculations for M_B-cFAD and M_B-nFAD

	M_B-cFAD			M_B-nFAD		
	E	**G**	**Imaginary frequency**	**E**	**G**	**Imaginary frequency**
RC	-1.80851232	0.108749		-1.83687050	0.093597	
TS	-1.76685682	0.148773	-1634.51	-1.79406471	0.135126	-1636.45
PC_{inv}	-1.84018863	0.080058		-1.86586533	0.070013	
$TS_{N5\text{-}PC}$	-1.83585971	0.081824	-151.22	-1.86194113	0.071056	-116.54
$N5_{yl}$	-1.84432014	0.075080		-1.87400333	0.066156	

(D) From ONIOM (M062X/6-31+G(d,p):PM6 calculations for M_B-cFAD and M_B-nFAD

	M_B-cFAD			M_B-nFAD		
	E	**G**	**Imaginary frequency**	**E**	**G**	**Imaginary frequency**
RC	-2112.771263	-2110.785479		-2113.976982	-2111.97318	
TS	-2112.729699	-2110.749556	-1224.9	-2113.934608	-2111.93429	-1328.35
PC_{inv}	-2112.775427	-2110.790833		-2113.969874	-2111.97087	
$TS_{N5\text{-}PC}$	-	-		-	-	
$N5_{yl}$	-2112.779482	-2110.785078		-2113.980879	-2111.96999	

Table S3. NBO partial charges[a] of important atoms for M_S-cFAD calculated at the CPCM/M062X/6-31+G(d,p)//PM6 level.

	RC	**TS**	**PC** $_{-+}$	**TS_{inv}**	**PC_{inv}**	**$TS_{N5\text{-}PC}$**	**$N5_{yl}$**	**TS_{H2O}**	**PC**
O1	-0.67	-0.71	-0.83	-0.83	-0.83	-0.80	-0.81	-0.79	-0.76
C4	0.74	0.70	0.64	0.64	0.64	0.66	0.67	0.66	0.67
C4a	0.12	-0.01	-0.11	-0.11	-0.12	-0.12	-0.15	-0.10	-0.08
N5	-0.37	-0.51	-0.67	-0.67	-0.71	-0.60	-0.54	-0.70	-0.69
N11	-0.98	-0.82	-0.71	-0.71	-0.71	-0.79	-0.92	-0.74	-0.75
H12	0.43	0.49	0.51	0.51	0.51	0.48	0.44	0.53	0.55
H13	0.41	0.44	0.47	0.47	0.47	0.46	0.43	0.43	0.40
C14	-0.29	-0.18	0.27	0.27	0.28	0.21	0.12	0.22	0.15
H15	0.23	0.32	0.47	0.47	0.47	0.48	0.53	0.47	0.47
H16	0.23	0.27	0.26	0.27	0.27	0.28	0.28	0.24	0.23
N1	-0.66	-0.68	-0.70	-0.69	-0.70	-0.69	-0.68	-0.75	-0.69
C10	0.48	0.50	0.46	0.46	0.46	0.48	0.49	0.52	0.50
FAD	-0.27	-0.66	-0.98	-0.98	-0.99	-0.77	-0.49	-0.79	-0.12
SER	0.02	0.22	0.82	0.83	0.85	0.67	0.39	0.13	-0.02

[a]FAD and SER represent the NBO charge sum of all atoms in flavin and serotonin, repectively.

Table S4. Benchmarking PM6 geometries: Critical distances (R in Å), the error with respect to M06-2X/6-31+G** (ΔR), average absolute error (AAE) calculated for the benzylamin + FAD model structure [a] and alignment of 3-D structures from PM6 (blue) and M06-2X/6-31+G** (green) optimizations.

		C4a–N5		C4a-N11		N5-αH		C14-αH		N5-C14		N11-C14		AAE
		R	ΔR	R	ΔR	R	ΔR	R	ΔR	R	ΔR	R	ΔR	
PM6	RC	1.31	0.02	3.07	0.18	2.72	0.18	1.12	0.03	3.56	0.24	1.49	0.02	0.11
	TS	1.37	0.02	2.48	-0.17	1.24	0.08	1.53	0.04	2.54	0.01	1.40	0.04	0.06
	$N5_{yl}$	1.45	0.01	2.76	-0.06	1.05	0.02	2.19	0.04	1.62	-0.03	1.45	0.05	0.03
HF/6-31G*	RC	1.26	-0.03	3.25	0.36	3.25	0.71	1.08	-0.01	3.82	0.50	1.46	-0.01	0.27
	TS	1.35	0.00	2.86	0.21	1.17	0.01	1.45	-0.04	2.57	0.04	1.34	-0.02	0.05
	$N5_{yl}$	1.44	0.00	2.83	0.01	1.01	-0.02	2.10	-0.05	1.59	-0.06	1.41	0.01	0.03
HF/6-31+G**	RC	1.26	-0.03	3.28	0.39	3.87	1.33	1.08	-0.01	3.82	0.50	1.46	-0.01	0.38
	TS	1.35	0.00	2.84	0.19	1.18	0.02	1.44	-0.05	2.56	0.03	1.34	-0.02	0.05
	$N5_{yl}$	1.45	0.01	2.83	0.01	1.01	-0.02	2.11	-0.04	1.59	-0.06	1.41	0.01	0.03
B3LYP/6-31G*	RC	1.30	0.01	3.15	0.26	2.81	0.27	1.09	0.00	3.58	0.26	1.48	0.01	0.14
	TS	1.36	0.01	2.75	0.10	1.16	0.00	1.52	0.03	2.56	0.03	1.38	0.02	0.03
	$N5_{yl}$	1.41	-0.03	2.87	0.05	1.02	-0.01	2.47	0.32	2.12	0.47	1.35	-0.05	0.16
B3LYP/6-31+G**	RC	1.29	0.00	3.58	0.69	2.81	0.27	1.09	0.00	3.15	-0.17	1.48	0.01	0.19
	TS	1.36	0.01	3.03	0.38	1.18	0.02	1.47	-0.02	2.60	0.07	1.38	0.02	0.09
	$N5_{yl}$	1.41	-0.03	2.86	0.04	1.02	-0.01	2.47	0.32	2.12	0.47	1.35	-0.05	0.15
CAM-B3LYP/6-31+G**	RC	1.29	0.00	3.12	0.23	2.71	0.17	1.09	0.00	3.45	0.13	1.47	0.00	0.09
	TS	1.36	0.01	2.69	0.04	1.15	-0.01	1.51	0.02	2.56	0.03	1.36	0.00	0.02
	$N5_{yl}$	1.44	0.00	2.81	-0.01	1.03	0.00	2.16	0.01	1.66	0.01	1.40	0.00	0.01
WB97XD/6-31+G**	RC	1.29	0.00	2.95	0.06	2.52	-0.02	1.09	0.00	3.52	0.20	1.47	0.00	0.05
	TS	1.36	0.01	2.69	0.04	1.14	-0.02	1.49	0.00	2.53	0.00	1.36	0.00	0.01
	$N5_{yl}$	1.44	0.00	2.82	0.00	1.03	0.00	2.15	0.00	1.64	-0.01	1.40	0.00	0.00
M06-2X/6-31+G*	RC	1.29	0.00	2.87	-0.02	4.05	1.51	1.09	0.00	3.72	0.40	1.47	0.00	0.16
	TS	1.36	0.01	2.63	-0.02	1.16	0.00	1.51	0.02	2.55	0.02	1.36	0.00	0.01
	$N5_{yl}$	1.44	0.00	2.83	0.01	1.03	0.00	2.15	0.00	1.64	-0.01	1.40	0.00	0.00
M06-2X/6-31+G**	RC	1.29	0.00	2.89	0.00	2.54	0.00	1.09	0.00	3.32	0.00	1.47	0.00	0.00
	TS	1.35	0.00	2.65	0.00	1.16	0.00	1.49	0.00	2.53	0.00	1.36	0.00	0.00
	$N5_{yl}$	1.44	0.00	2.82	0.00	1.03	0.00	2.15	0.00	1.65	0.00	1.40	0.00	0.00

[a]See ref. 36 for the details of the calculation. Except M06-2X/6-31+G**, the distances were taken from ref.36.

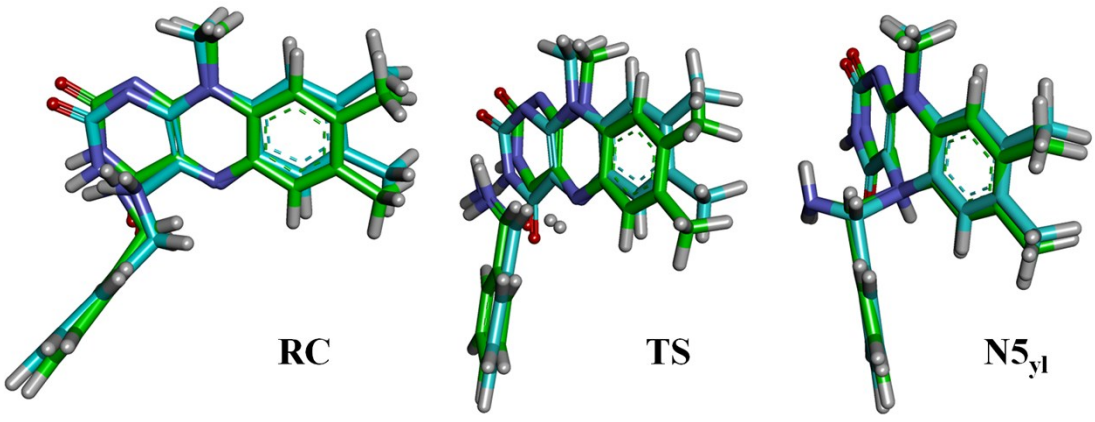

Table S5. Benchmarking PM6: Gibbs free energies (au) and activation energies (kcal/mol) calculated by different methods for the benzylamin + FAD model structure.[a]

	RC	**TS**	**$N5_{yl}$**	**ΔG (kcal/mol)**
PM6	0.28477	0.33973	0.26976	34.5
HF/6-31G*	-1191.28661	-1191.15263	-	84.1
HF/6-31+G**	-1191.32881	-1191.22859	-1191.28389	62.8
DFT/B3LYP/6-31G*	-1198.69942	-1198.64745	-1198.75103	32.6
DFT/B3LYP/6-31+G**	-1198.78332	-1198.73415	-1198.75104	30.9
CAMB3LYP/6-31+G**	-1198.16079	-1198.10515	-1198.13412	34.9
WB97XD/6-31+G**	-1198.38122	-1198.33161	-1198.36454	31.1
M06-2X/6-31+G*	-1198.23440	-1198.17995	-1198.21673	34.2
M06-2X/6-31+G**	-1198.26379	-1198.215558	-1198.24791	30.3

[a]See ref. 36 for the details of the calculation. Except M06-2X/6-31+G**, the values were taken from ref.36.

Table S6. ONIOM energy decomposition (au) and relative energies (kcal/mol) of stationary points along the reaction coordinate of small model (M_S-cFAD) and ΔS values. (FS:Full System, HL:High Layer, LL:Low Layer)

	$E_{FS,PM6}$	$E_{HL,PM6}$	$E_{HL,M062X}$	[a]E_{ONIOM}	[b]$E_{LL,PM6}$	[c]$\Delta S_{LL,PM6}$
RC	-1.152113090 (0.0)	-0.303913663 (0.0)	-2111.293607 (0.0)	-2112.141806 (0.0)	-0.848199428 (0.0)	
TS	-1.106513589 (28.6)	-0.253532381 (31.6)	-2111.249076 (27.9)	-2112.102057 (24.9)	-0.852981208 (-3.0)	-0.055119422
PC_{inv}	-1.172206040 (-12.6)	-0.313433584 (-6.0)	-2111.282652 (6.8)	-2112.141425 (0.2)	-0.858772456 (-6.6)	
$TS_{N5\text{-}PC}$	-1.180587719 (-17.9)	-0.324919273 (-13.2)	-2111.284324 (5.8)	-2112.139992 (1.1)	-0.855668446 (-4.7)	0.061523241
$N5_{yl}$	-1.188561977 (-22.9)	-0.333958732 (-18.8)	-2111.286401 (4.5)	-2112.141004 (0.5)	-0.854603246 (-4.0)	
TS_{H2O}	-1.154231060 (-1.3)	-0.304823633 (-0.6)	-2111.279461 (8.9)	-2112.128868 (8.1)	-0.849407427 (-0.8)	0.052479283
PC	-1.164833666 (-8.0)	-0.319274378 (-9.6)	-2111.291885 (1.1)	-2112.137444 (2.7)	-0.845559289 (1.6)	

[a]$E_{ONIOM} = E_{FS,PM6} + E_{HL,M062x} - E_{HL,PM6}$

[b]$E_{LL,PM6}$ is the contribution of low layer PM6 energy: $E_{LL,PM6} = E_{FS,PM6} - E_{HL,PM6}$

[c]$\Delta S_{LL,PM6} = \{E_{FS,PM6}(\text{reactant}) - E_{HL,PM6}(\text{reactant})\} - \{E_{FS,PM6}(\text{product}) - E_{HL,PM6}(\text{product})\}$

See ref. 32 for detailed discussion on ΔS.

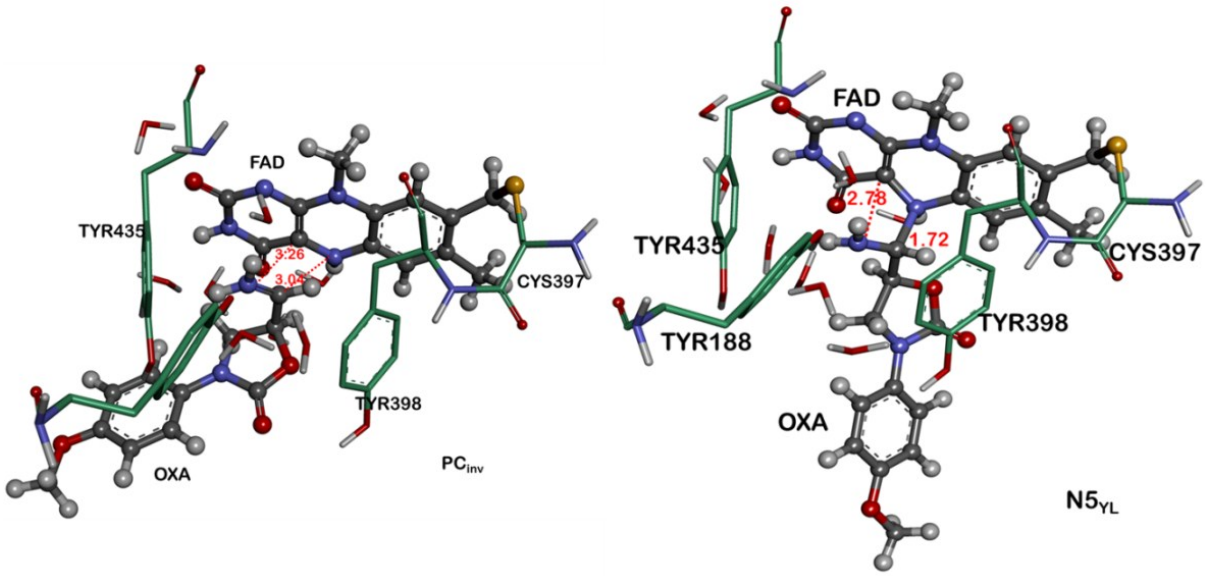

Figure S1. Optimized structures of PC_{inv} and $N5_{yl}$ for 5-(aminomethyl)-3-(4-methoxyphenyl)-2-oxazolidinone at M062x/6-31+G(d,p):PM6 level.

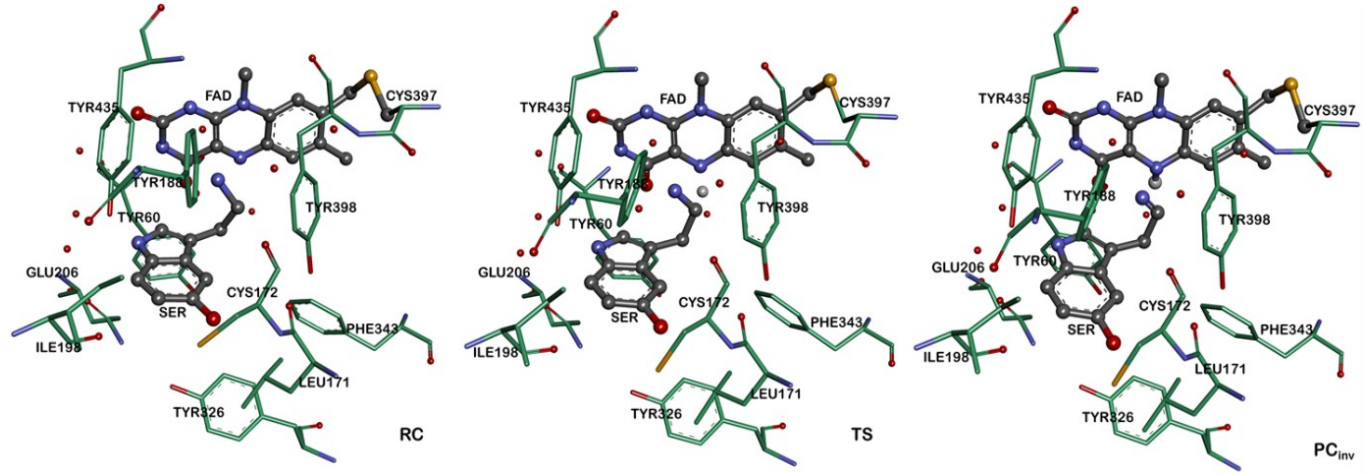

Figure S2. Optimized structures of the rate determining first step of serotonin oxidation by MAO for big model MB-cFAD obtained from M06-2X/6-31+G(d,p):PM6 calculations.

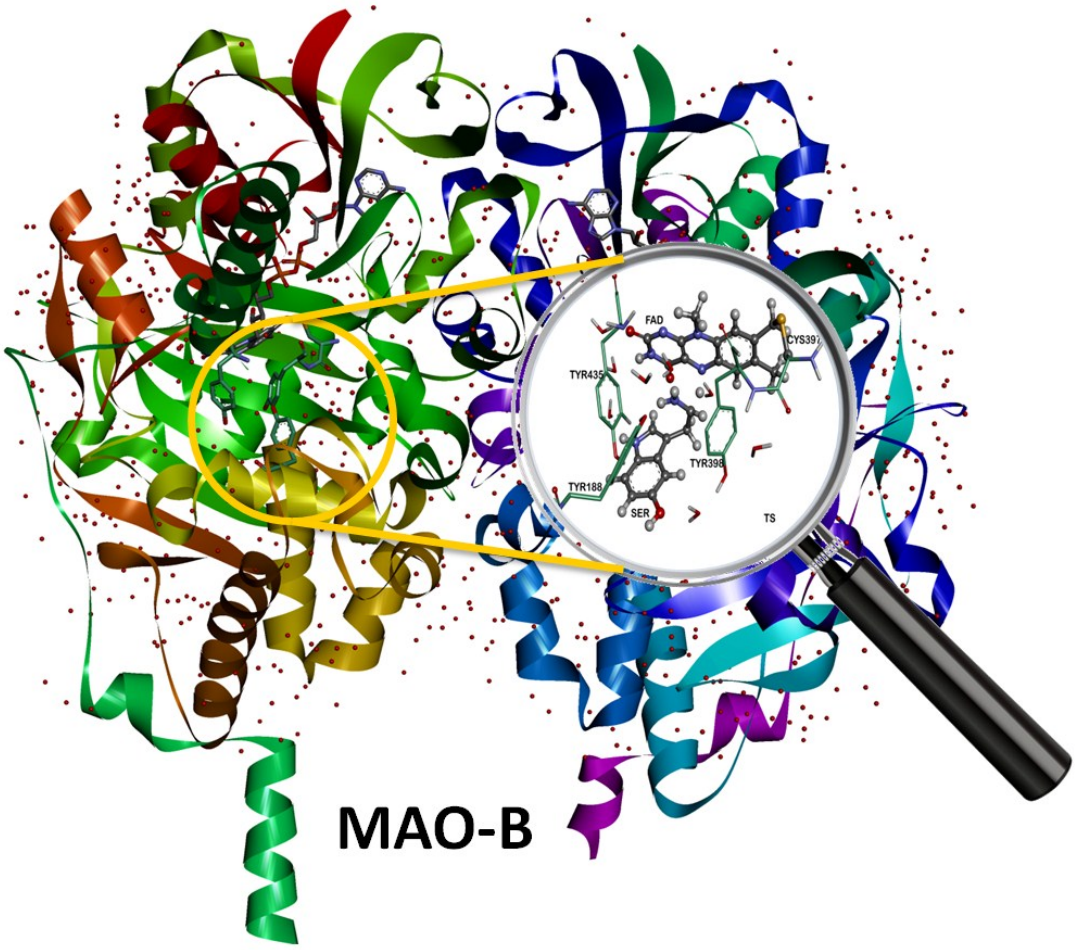

Figure S3. Active site model inside the real enzyme.