

Title: The 3D structure prediction of TAS2R38 bitter receptors bound to agonists phenylthiocarbamide (PTC) and 6-n-Propylthiouracil (PROP)

Supplementary Information

Methods of Molecular Dynamics Simulation

We performed molecular dynamics (MD) simulations of the predicted structure of bitter taste receptor with and without ligand for 10 ns in explicit lipid bilayer and water. We carried out MD simulations using NAMD including explicit water and a periodically infinite lipid to determine the interactions of the protein with lipid and water. We used the CHARMM22 force field parameters for the protein, the TIP3 model for water, and the CHARMM27 force field parameters for the lipids. Quantum charges from DFT/6311G** method were used for these ligands. We started with the predicted protein structure, stripped away the lipid molecules, and inserted it in a periodic structure of 1-palmytoil-2-oleoylsn-glycero-3-phosphatidylcholine (POPC). In this process, we eliminated lipid molecules within 5 Å of the protein. Then, we inserted this in a box of water molecules and eliminated waters within 5 Å of the lipid and protein. Chloride ions were added to neutralize the charge of the system. The membrane and water molecules were minimized with the protein fixed, and then equilibrated for 500 ps in an NPT simulation. Finally, the entire system was minimized, and then 10 ns of NPT simulation was run. All NPT simulations were run using Langevin dynamics with a damping coefficient of 1 ps^{-1} and a bath temperature of 310 K. The pressure was kept constant by Nosé-Hoover Langevin piston pressure control, with a target pressure of 1 atm and barostats oscillation and damping times of 200 fs. The step size was 1 fs, with periodic boundary conditions applied. The full system (Figure S4) contains the predicted protein, 101 lipid molecules, 7528 water molecules, and 19 chlorine ions for a total of 41570 atoms per periodic cell for apo-

PAV protein. That of apo-AVI protein contains the predicted protein, 102 lipid molecules, 7498 water molecules, and 19 chlorine ions for a total of 41619 atoms per periodic cell. That of PAV protein bound to PTC contains the predicted protein, 104 lipid molecules, 5626 water molecules, and 19 chlorine ions for a total of 36284 atoms per periodic cell. That of AVI protein bound to PTC contains the predicted protein, 105 lipid molecules, 7387 water molecules, and 19 chlorine ions for a total of 41706 atoms per periodic cell. The box size is 75 Å by 75 Å by 85 Å. We then used the NAMD program to carry out 10 ns of NPT MD with a bath temperature of 310 K.

Table I. BiHelix and ComBiHelix results for the PAV variation of bitter taste receptors in the different receptor templates.

Receptor Templates	Rotational Angle							Total Energy (Kcal/mol)
	<i>H1</i>	<i>H2</i>	<i>H3</i>	<i>H4</i>	<i>H5</i>	<i>H6</i>	<i>H7</i>	
t β 1AR	30	330	60	90	180	270	30	598.1
h β 2AR	30	330	60	120	330	90	270	754.5
BovR	0	0	60	150	330	30	330	997.3
hAA _{2A} R	0	330	30	180	0	30	0	932.8

Figure S1 Sequence alignment against $\text{t}\beta\text{1AR}$, $\text{h}\beta\text{2AR}$, Rhodopsin and hAA2AR for TAS2R38 bitter taste receptor variants PAV.

PAV-TM1	14	VRSTFLFI	SVLEFAVGFLTNAFVFLVNFWD	43
$\text{t}\beta\text{1AR-TM1}$	40	WEAGMSLLMALVVLLI	VAGNVLVI AAI GST	69
PAV-TM2	55	CVLLCLSI	SRLFLHGLLFLSAI QLTHFQKLSE	86
$\text{t}\beta\text{1AR-TM2}$	74	TLTNLFI	TSACADLVVGLLVVPPGATLVVRG	105
PAV-TM3	94	AI I MLVMI	ANQANLWLAACLSLLYCSKLI RFSHT	127
$\text{t}\beta\text{1AR-TM3}$	110	GSFLCELWTSLDVL	CVTASI ETLCVI AI DRYLAI	143
PAV-TM4	140	I SQMLLGI	I LCSCI CTVLCVWCFFSR	165
$\text{t}\beta\text{1AR-TM4}$	154	TRARAKVI	I CTVVAI SALVSFLPI MM	179
PAV-TM5	190	NLFYSFLFCYLWSVPPFLLFLVSSGMLTVSLGR		222
$\text{t}\beta\text{1AR-TM5}$	204	NRAYAI ASSI	I SFYI PLLI MI FVALRVYREAKE	236
PAV-TM6	244	KALKSLVSFFCFFVI	SSCAAFI SVPLLI	271
$\text{t}\beta\text{1AR-TM6}$	284	REHKALKTLGI	I MGVTLCWLPFFLVNI	311
PAV-TM7	276	KI GVMVCVGI	MAACPSGHAAVLI S	299
$\text{t}\beta\text{1AR-TM7}$	321	PDWLFVAFNWLGYANSAMNPI	I YC	344
PAV-TM1	11	SYEVRSTFLFI	SVLEFAVGFLTNAFVFLVNFWD	42
$\text{h}\beta\text{2AR-TM1}$	29	DEVVVVGMI	VMSLI VLA I VFGNVLVI TAI AK	60
PAV-TM2	56	VLLCLSI	SRLFLHGLLFLSAI QLTHFQKLS	85
$\text{h}\beta\text{2AR-TM2}$	67	VTNYFI	TSACADLVMLAVVPPGAAHI LM	96
PAV-TM3	95	I I MLVMI	ANQANLWLAACLSLLYCSKLI RFSHT	128
$\text{h}\beta\text{2AR-TM3}$	103	NFWCEFWTSI	DVLCVTASI ETLCVI AVDRYFAI T	136
PAV-TM4	139	KI SQMLLGI	I LCSCI CTVLCVWC	161
$\text{h}\beta\text{2AR-TM4}$	149	KARVI	I LMVVI VSGLTSFLPI QM	171
PAV-TM5	190	NLFYSFLFCYLWSVPPFLLFLVSSGMLTVSLGR		222
$\text{h}\beta\text{2AR-TM5}$	197	QAYAI ASSI	VSFYVPLVI MVFVYSRVFQEAQRQ	229
PAV-TM6	244	KALKSLVSFFCFFVI	SSCAAFI SVPLLI	271
$\text{h}\beta\text{2AR-TM6}$	267	KEHKALKTLGI	I MGTFTLCWLPFFI VNI	294
PAV-TM7	277	I GVMVCVGI	MAACPSGHAAI LI S	299
$\text{h}\beta\text{2AR-TM7}$	306	EYVI LLNWI	GYVNSGFNPLI YCR	328
PAV-TM1	13	EVRSTFLFI	SVLEFAVGFLTNAFVFLVNFWD	42
rhodop-TM1	35	WQFSMLAAYMFLLI	MLGFPI NFLTLVYTVQ	64
PAV-TM2	57	LLCLSI	SRLFLHGLLFLSAI QLTHFQKLSE	86
rhodop-TM2	71	PLNYI LLNLAVADLF	MVFGGFTTTLYTSLH	100
PAV-TM3	95	I I MLVMI	ANQANLWLAACLSLLYCSKLI RFSHT	127
rhodop-TM3	107	PTGCNLEGFFATLGGEI	ALWSLVVLAI ERYVVV	139
PAV-TM4	142	QMLLGI	I LCSCI CTVLCVWCFFS	164
rhodop-TM4	151	NHAI MGVAFTVWMALACAAPPLV		173
PAV-TM5	189	LNLFYSLFCYLWSVPPFLLFLVSSG		214
rhodop-TM5	200	NESFVI YMFVVHFI	I PLI VI FFCYGO	225
PAV-TM6	244	KALKSLVSFFCFFVI	SSCAAFI SVPLLI	271
rhodop-TM6	247	EKEVTRMVI	I MVI AFLI CWLPYAGVAFY	274
PAV-TM7	277	I GVMVCVGI	MAACPSGHAAI L	297
rhodop-TM7	286	I FMTI PAFFAKTS	SAVYNPVI Y	306
PAV-TM1	14	VRSTFLFI	SVLEFAVGFLTNAFVFLVNF	41
hAA2AR-TM1	5	GSSVYI	TVELAI AVLAI LGNVLCWAVW	32
PAV-TM2	57	LLCLSI	SRLFLHGLLFLSAI QLTHFQK	83
hAA2AR-TM2	41	TNYFVVS	LAAADI AVGVLA I PFAI TI S	67
PAV-TM3	95	I I MLVMI	ANQANLWLAACLSLLYCSKLI RFSHT	127
hAA2AR-TM3	75	HGCLFI	ACFVLVLTQSSI FSL LAI AI DRYI AI R	107
PAV-TM4	139	KI SQMLLGI	I LCSCI CTVLCVW	160
hAA2AR-TM4	119	TRAKGI	I AI CWVLSFAI GLTPM	140
PAV-TM5	190	NLFYSFLFCYLWSVPPFLLFLVSSGMLTVS		219
hAA2AR-TM5	175	NYMVYFNFFACVLVPLLLMLGVYLR	I FLAA	204
PAV-TM6	242	HI KALKSLVSFFCFFVI	SSCAAFI SVPLLI	271
hAA2AR-TM6	222	RSTLQKEVHAAKSLAI	I VGLFALCWLP LHI	251
PAV-TM7	277	I GVMVCVGI	MAACPSGHAAVLI S	299
hAA2AR-TM7	269	LMYLAI	VL SHTNSVVNPFI YAYR	291

Figure S2. The prediction of 7 hydrophobic regions from PredicTM

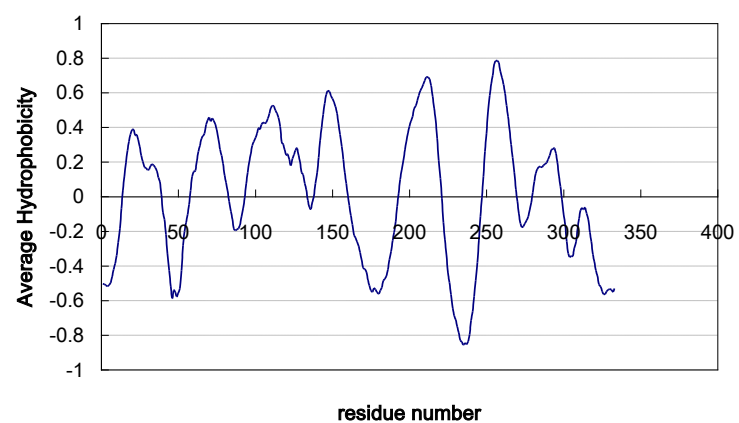


Figure S3 Predicted 3D structures of bitter taste receptors PAV based on the four templates (a) t β 1AR, (b) h β 2AR, (c) Rhodopsin and (d) hAA_{2A}R from BiHelix. (Residue A262 in the TM6 are highlighted here)

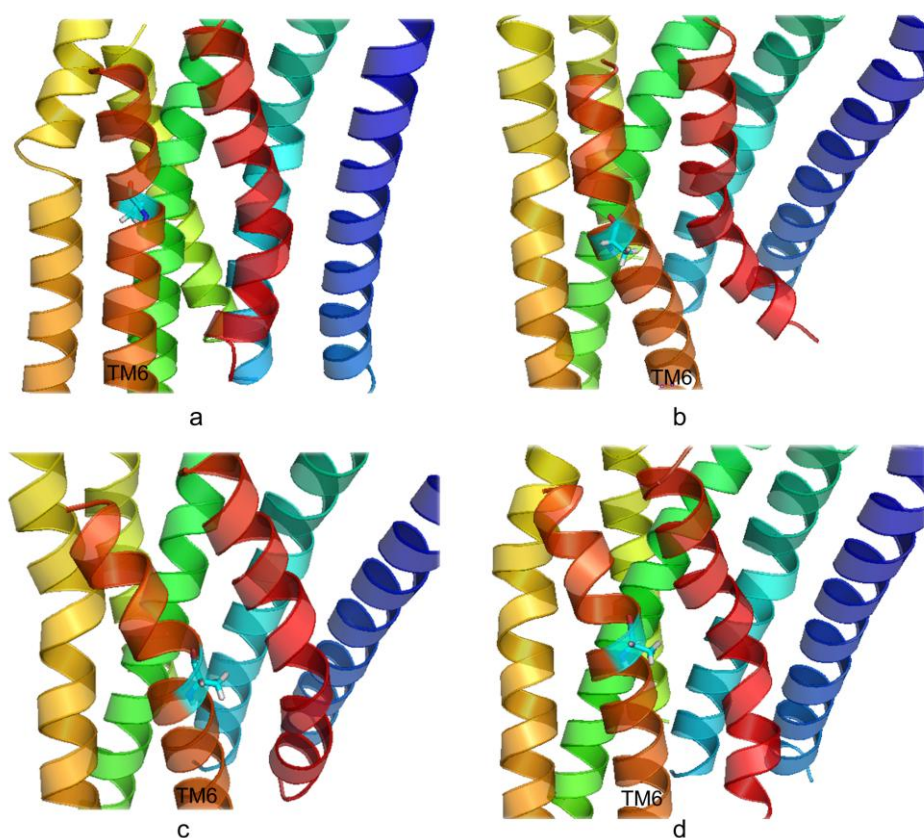


Figure S4. The molecular dynamics simulation box of TAR2S38 bitter receptor with lipid and water.

The EC region is at the top.

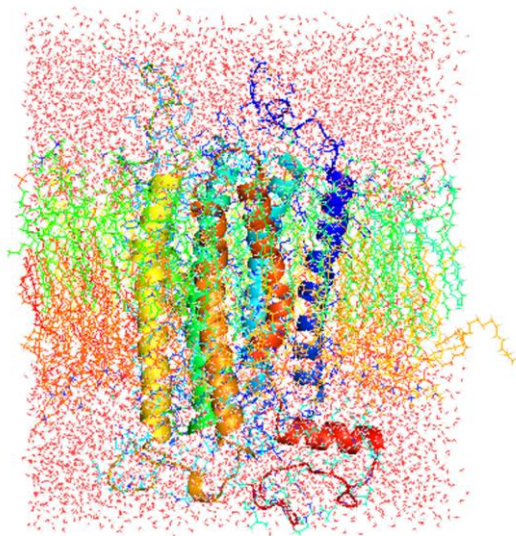
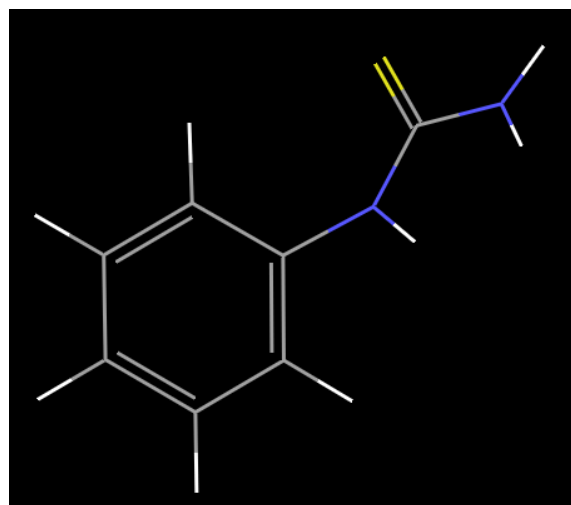
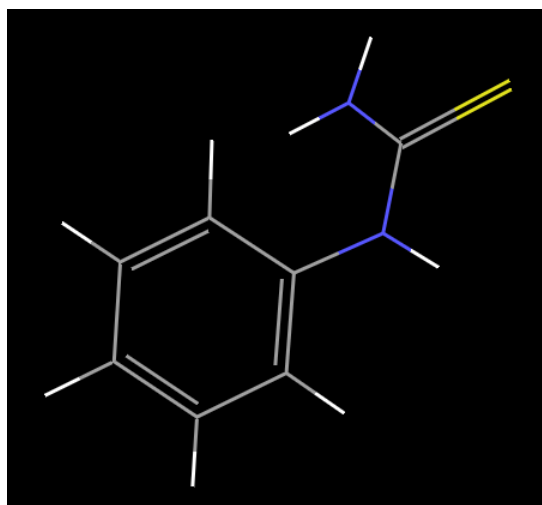
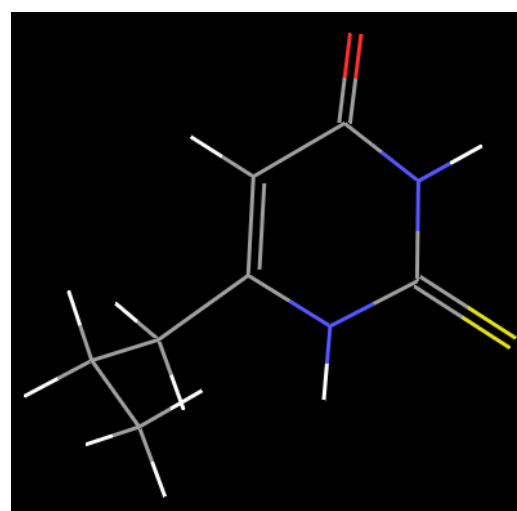
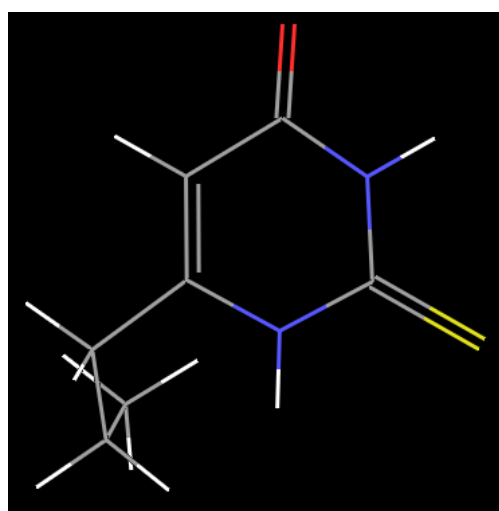
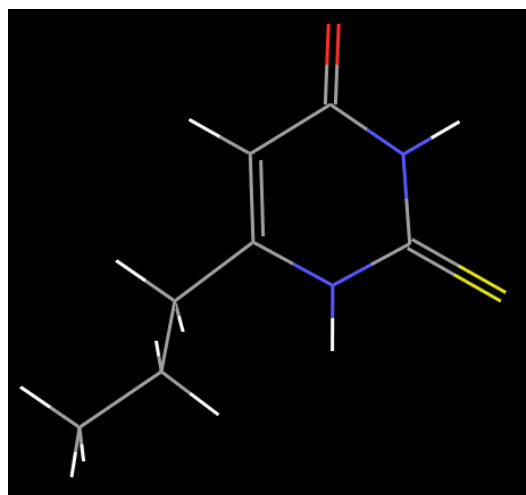
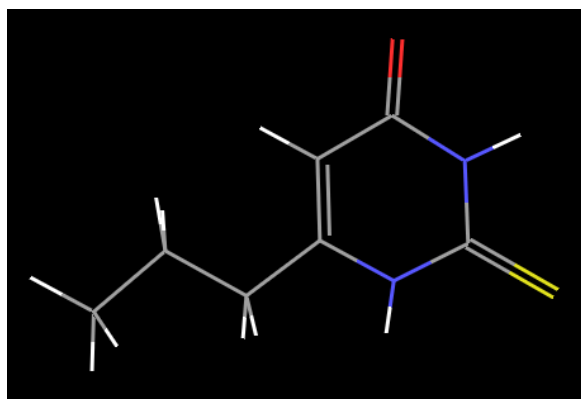


Figure S5 The 2 conformations of PTC and 4 of PROP docked to bitter taste receptor hTAS2R38.



a



b

Figure S6 Predicted binding sites of agonists in bitter taste receptors. (a) PTC in hTAS2R38_{PVV}, (b) PROP in hTAS2R38_{PVV}, (c) PTC in hTAS2R38_{AAI}, (d) PROP in hTAS2R38_{AAI}.

