

Supplementary material

A Boron-containing Analogue Of Acetaminophen Induces Analgesic Effect in Hot-Plate Test And Limited Hepatotoxicity

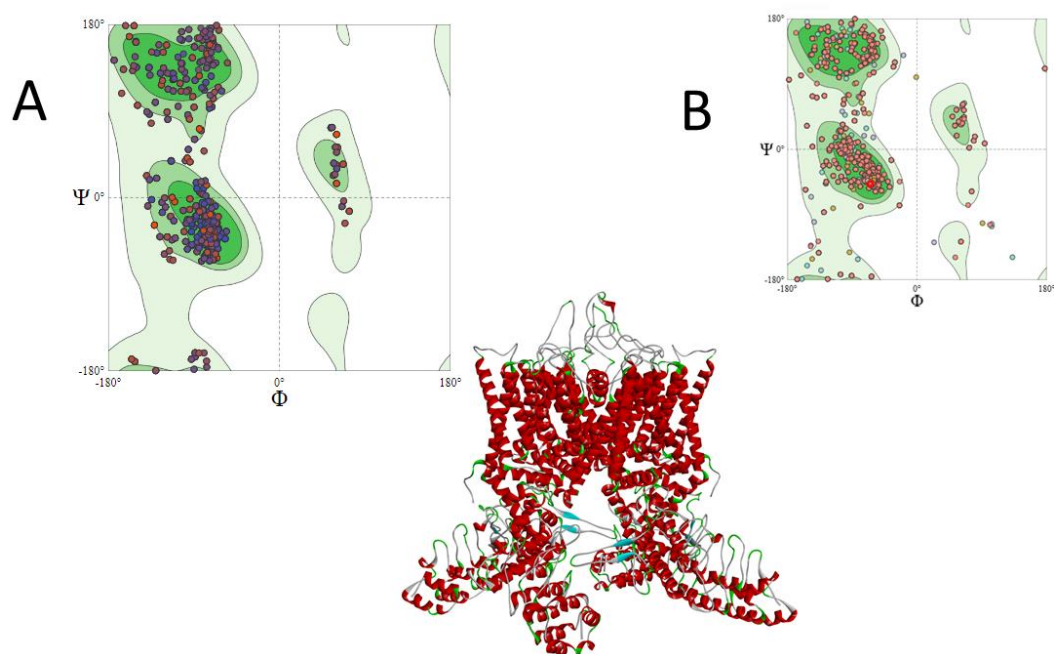


FIGURE S1. Ramachandran plots of designed protein from Swiss model. A. For rat TRPV1 (PDB code: 5IRX) B. For human TRPV1 (built by homology) and C. The complete 3D-structure of modeled human TRPV1 protein.

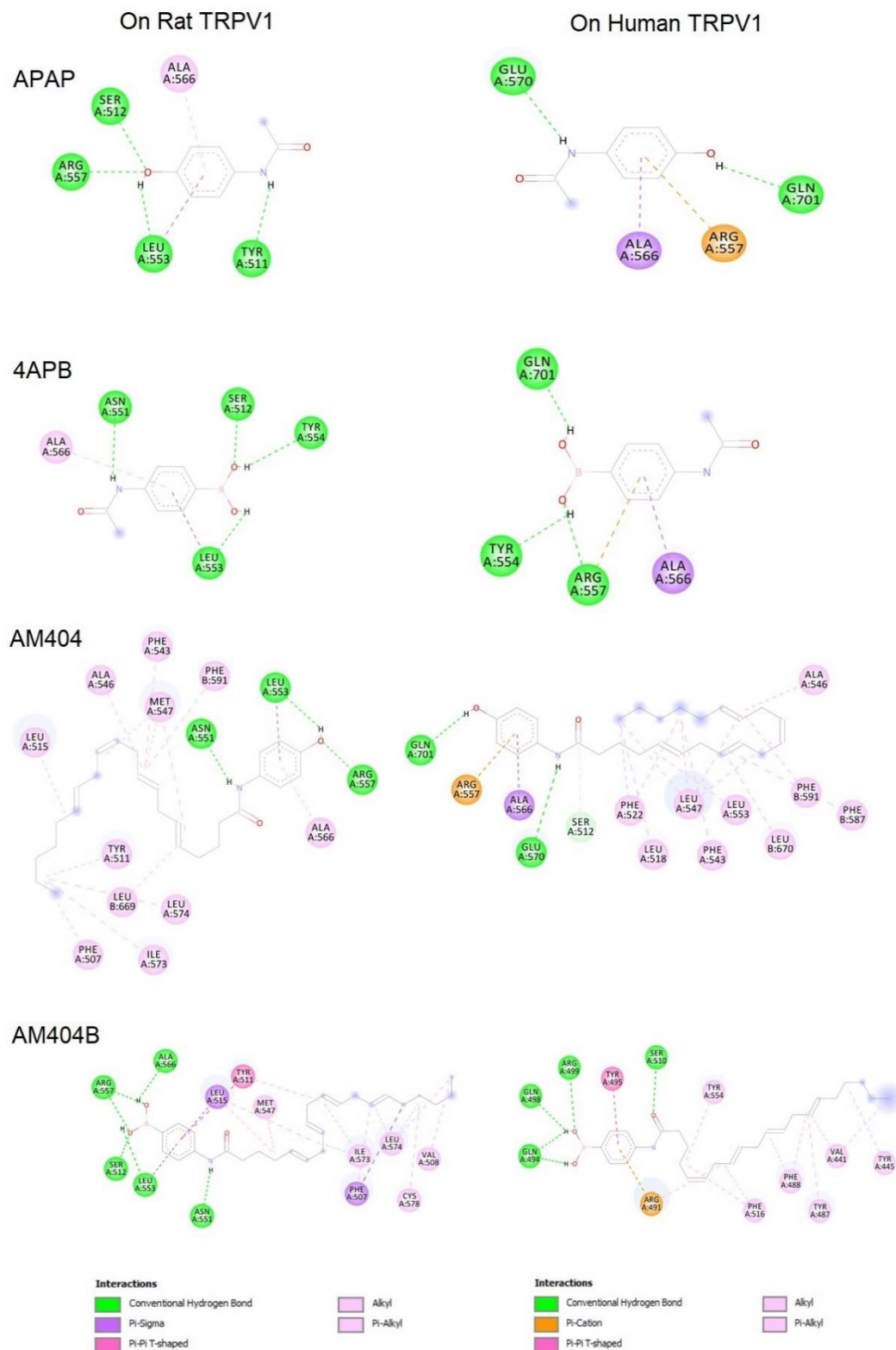


FIGURE S2. The 2D-schemes of interactions of APAP, 2APB and its metabolites (AM404 and AM404B, respectively) on the rat-TRPV1(at left) or human-TRPV1 (at right).

Table S1. Results from Swiss-model server structural assessment for the TRPV1 used proteins.

PDB ID	GMQE	QSQE	IDENTITY (From template)	METHOD	RAMACHANDRAN PLOT
5IRX	0.73	0.85	N.A.	EM	89.21 Favoured 0.12% E64Pro F64Pro
Modeled Human TRPV1 (Based on 3J5P)	0.69	0.86	92.31%	EM	91% Favoured angles (SWISS MODEL)

Table S2. Evaluated compounds with their labeled codes.

	CODE	NAME
1	3CysA	3-(S-Cysteiny)l acetaminophen
2	3CysB	3-(S-Cysteiny)l-4-acetamidophenylboronic acid
3	3GluA	3-(S-Glutathionyl) acetaminophen
4	3GluB	3-(S-Glutathionyl)-4-acetamidophenylboronic acid
5	3HA	3-Hydroxyacetaminophen
6	3HAGl	3-Hydroxyacetaminophen Glucuronide
7	3HASu	3-Hydroxyacetaminophen Sulphate
8	3HB	3-Hydroxy-4-acetamidophenylboronic acid
9		3-Hydroxy-4-acetamidophenylboronic acid
	3HBGL	Glucuronide
10	3HBSu	3-Hydroxy-4-acetamidophenylboronic acid Sulphate
11	3MA	3-Methoxyacetaminophen
12	3MB	3-Methoxy-4-acetamidophenylboronic acid
13	3MeA	3-(S-Mercapto)acetaminophen
14	3MeB	3-(S-Mercapto)-4-acetamidophenylboronic acid
15	3MeTA	3-(S-ThioMethyl)acetaminophen
16	3MeTB	3-(S-Thiomethyl)-4-acetamidophenylboronic acid
17	4AB	4-aminophenylboronic acid
18	4AP	4-aminophenol
19	4-APB	4-Acetamidophenylboronic acid
20	AcGl	acetaminophen Glucuronide
21	AcSu	Acetaminophen Sulphate
22	AM404	N-arachidonoylaminophenol
23	AM404B	N-arachidonoyl-4-aminophenylboronic acid
24	ANA	Anandamide
25	APAP	Acetaminophen
26	BGl	4-Acetamidophenylboronic acid Glucuronide
27	Bsu	4-Acetamidophenylboronic acid Sulphate
28	Cap	Capsaicin
29	Capz	Capsazepine
30	NAP	N-acetyl-p-benzoquinone imine
31	NAPB	N-acetyl-p-benzoboronic acid imine
32	PbenB	1-benzoquinone-4-boric acid
33	Pbenz	1, 4-benzoquinone

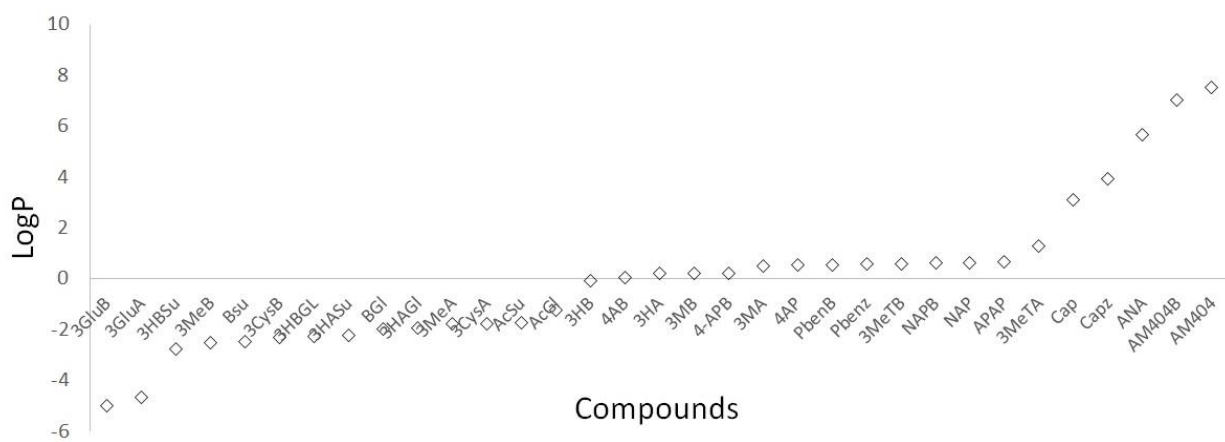


FIGURE S3. Compounds ordered considering the LogP (octanol-water partition coefficient) calculated for each tested compound from molinspiration.com. Abbreviations are as in Table S2.

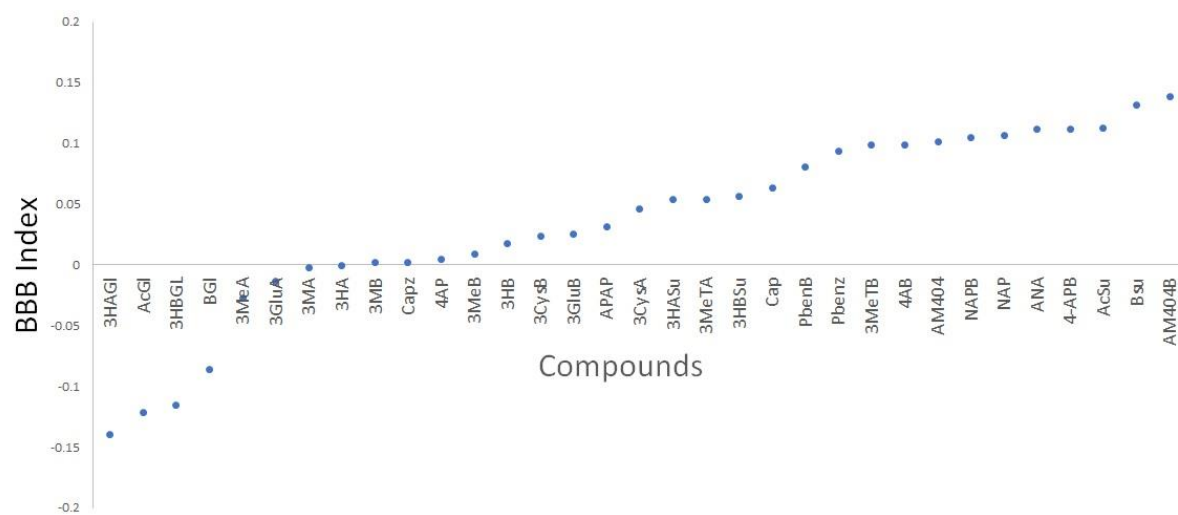


FIGURE S4. Blood brain barrier index calculated for each compound from prediction server (cbligand.org).

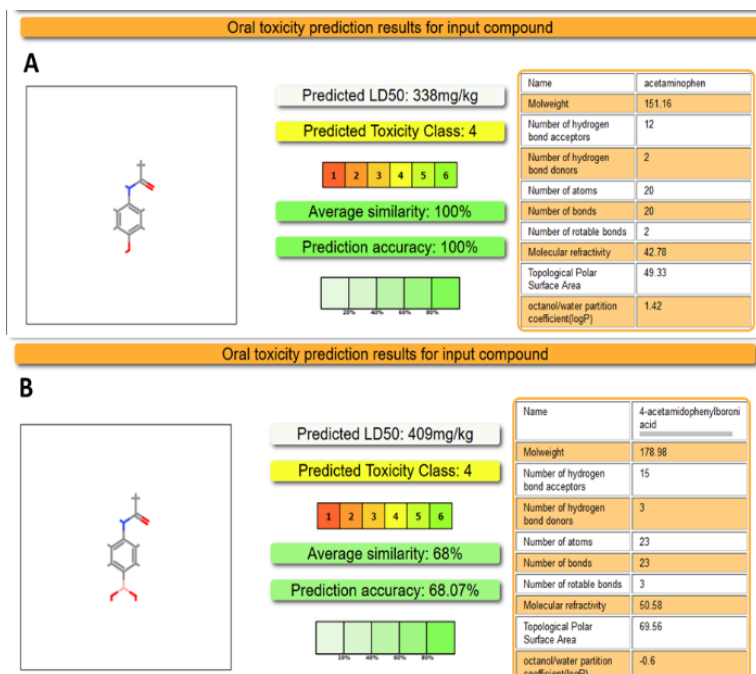


Figure S5. PROTOX II prediction of LD₅₀ values for APAP(A) and 4-APB(B).

A

Toxicity Model Report				
Classification	Target	Shorthand	Prediction	Probability
Organ toxicity	Hepatotoxicity	dtl	Active	0.74
Toxicity end points	Carcinogenicity	carcino	Inactive	0.51
Toxicity end points	Immunotoxicity	immuno	Inactive	0.99
Toxicity end points	Mutagenicity	mutagen	Inactive	0.90
Toxicity end points	Cytotoxicity	cyto	Inactive	0.62
Tox21-Nuclear receptor signaling pathways	Aryl hydrocarbon Receptor (AHR)	nr_ahr	Inactive	0.90
Tox21-Nuclear receptor signaling pathways	Androgen Receptor (AR)	nr_ar	Inactive	1.0
Tox21-Nuclear receptor signaling pathways	Androgen Receptor Ligand Binding Domain (AR-LBD)	nr_ar_lbd	Inactive	1.0
Tox21-Nuclear receptor signaling pathways	Aromatase	nr_aromatase	Inactive	1.0
Tox21-Nuclear receptor signaling pathways	Estrogen Receptor Alpha (ER)	nr_er	Inactive	0.94
Tox21-Nuclear receptor signaling pathways	Estrogen Receptor Ligand Binding Domain (ER-LBD)	nr_er_lbd	Inactive	0.99
Tox21-Nuclear receptor signaling pathways	Peroxisome Proliferator Activated Receptor Gamma (PPAR-Gamma)	nr_ppar_gamma	Inactive	1.0
Tox21-Stress response pathways	Nuclear factor (erythroid-derived 2)-like 2/antioxidant response element (nrf2/ARE)	nr_nfe2l3	Inactive	0.98
Tox21-Stress response pathways	Heat shock factor response element (HSE)	nr_hse	Inactive	0.98
Tox21-Stress response pathways	Mitochondrial Membrane Potential (MMP)	nr_mmp	Inactive	0.96
Tox21-Stress response pathways	Phosphoprotein (Tumor Suppressor) p53	nr_p53	Inactive	0.95
Tox21-Stress response pathways	ATPase family AAA domain-containing protein 5 (ATAD5)	nr_atad5	Inactive	0.97

B

Toxicity Model Report				
Classification	Target	Shorthand	Prediction	Probability
Organ toxicity	Hepatotoxicity	dtl	Inactive	0.52
Toxicity end points	Carcinogenicity	carcino	Inactive	0.72
Toxicity end points	Immunotoxicity	immuno	Inactive	0.99
Toxicity end points	Mutagenicity	mutagen	Inactive	0.64
Toxicity end points	Cytotoxicity	cyto	Inactive	0.62
Tox21-Nuclear receptor signaling pathways	Aryl hydrocarbon Receptor (AHR)	nr_ahr	Inactive	0.86
Tox21-Nuclear receptor signaling pathways	Androgen Receptor (AR)	nr_ar	Inactive	0.99
Tox21-Nuclear receptor signaling pathways	Androgen Receptor Ligand Binding Domain (AR-LBD)	nr_ar_lbd	Inactive	0.93
Tox21-Nuclear receptor signaling pathways	Aromatase	nr_aromatase	Inactive	0.96
Tox21-Nuclear receptor signaling pathways	Estrogen Receptor Alpha (ER)	nr_er	Inactive	0.90
Tox21-Nuclear receptor signaling pathways	Estrogen Receptor Ligand Binding Domain (ER-LBD)	nr_er_lbd	Inactive	0.91
Tox21-Nuclear receptor signaling pathways	Peroxisome Proliferator Activated Receptor Gamma (PPAR-Gamma)	nr_ppar_gamma	Inactive	0.96
Tox21-Stress response pathways	Nuclear factor (erythroid-derived 2)-like 2/antioxidant response element (nrf2/ARE)	nr_nfe2l3	Inactive	0.88
Tox21-Stress response pathways	Heat shock factor response element (HSE)	nr_hse	Inactive	0.88
Tox21-Stress response pathways	Mitochondrial Membrane Potential (MMP)	nr_mmp	Inactive	0.96
Tox21-Stress response pathways	Phosphoprotein (Tumor Suppressor) p53	nr_p53	Inactive	0.83
Tox21-Stress response pathways	ATPase family AAA domain-containing protein 5 (ATAD5)	nr_atad5	Inactive	0.94

Fig S6. In Silico Toxicity Model Report for APAP(A) and 4-APB(B).

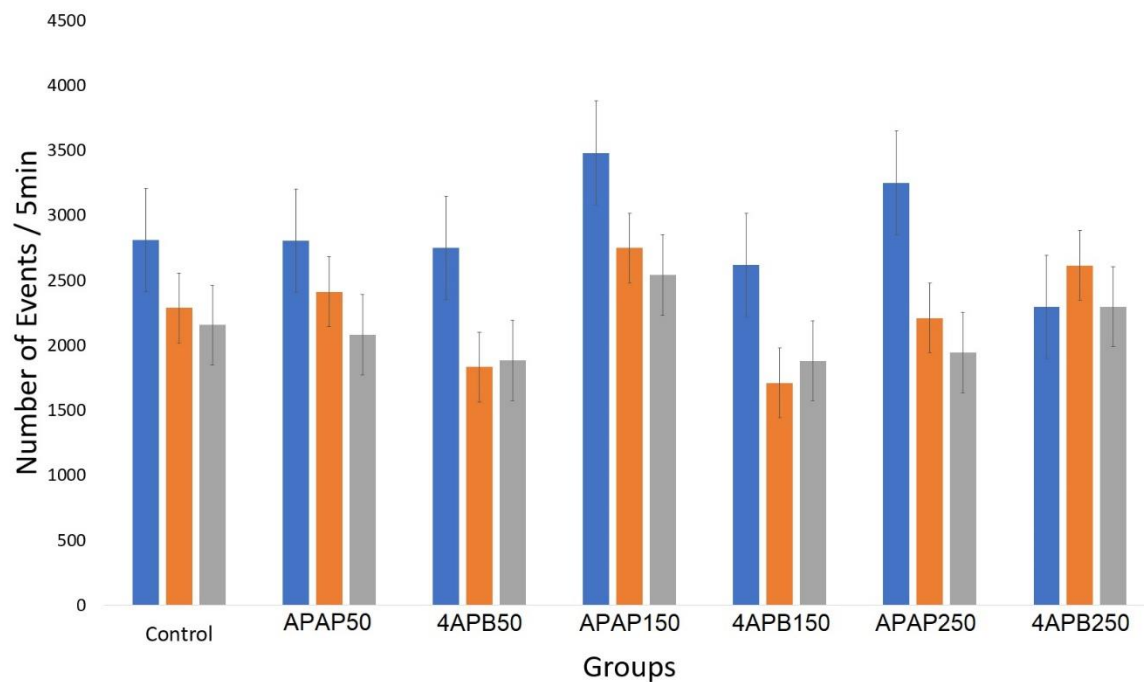


FIGURE S7: Number of events recorded for mice during 5 minutes of activity in an open field test. Prior to any procedure (blue), but also 2 h (orange) and 24 h (grey) after treatment administration.