Appendix 1

Results from the docking of standard ligands with a structure derived from the crystal structure of *Torpedo californica* AChE in complex with aricept (2.50 Á) (PDB ID: 1eve) (arranged according to lowest binding energy).

Ligand	Cluster rank	Lowest binding energy (kcal/mol)	No. in Cluster	Inhibition constant, K_i (μM)
(-)- Huprine x	1	-7.54	56	2.98
Δ^9-THC	1	-7.31	5	4.35
Donepezil	1	-7.06	8	6.72
(-)-Galanthamine	1	-7.03	24	7.09
Δ^9-THC	5	-6.87	23	9.16
Huperzine B	1	-6.85	72	9.52
BW284C51	1	-6.63	6	13.90
Huperzine A	1	-6.58	35	14.91
D-Tubocurarine	1	-6.52	25	16.70
Tacrine	1	-6.51	28	16.90
Tacrine	3	-6.27	55	25.32
MPTP	1	-6.07	88	35.60
Propidium	1	-6.02	27	38.58
MF268	1	-5.73	20	63.36
Decidium	1	-5.49	1	94.15
Edrophonium	1	-4.52	28	489.08
Decamethonium	1	-4.05	64	1070.00
Acetylthiocholine	1	-3.54	23	2530.00
Acetylthiocholine	2	-3.46	50	2910.00
Acetylcholine	1	-3.39	55	3280.00
Ambenonium	1	-0.78	1	266860.00
Gallamine	1	-0.67	2	323570.00

Appendix 2

Results from the docking of standard ligands with a structure derived from the crystal structure of *Torpedo californica* AChE in complex with aricept (2.50 Å) (PDB ID: 1eve) (arranged according to highest number in cluster).

Ligand	Cluster rank	Lowest binding energy (kcal/mol)	No. in cluster	Inhibition constant, K_i (μM)
MPTP	1	-6.07	88	35.60
Huperzine B	1	-6.85	72	9.52
Decamethonium	1	-4.05	64	1070.00
(-)- Huprine x	1	-7.54	56	2.98
Acetylcholine	1	-3.39	55	3280.00
Tacrine	3	-6.27	55	25.32
Acetylthiocholine	2	-3.46	50	2910.00
Huperzine A	1	-6.58	35	14.91
Edrophonium	1	-4.52	28	489.08
Tacrine	1	-6.51	28	16.90
Propidium	1	-6.02	27	38.58
D-Tubocurarine	1	-6.52	25	16.70
(-)-Galanthamine	1	-7.03	24	7.09
Acetylthiocholine	1	-3.54	23	2530.00
Δ^9-THC	5	-6.87	23	9.16
MF268	1	-5.73	20	63.36
Donepezil	1	-7.06	8	6.72
BW284C51	1	-6.63	6	13.90
Δ^9-THC	1	-7.31	5	4.35
Gallamine	1	-0.67	2	323570.00
Ambenonium	1	-0.78	1	266860.00
Decidium	1	-5.49	1	94.15

Appendix 3

Results from the docking of standard ligands with a structure derived from the crystal structure of *Torpedo californica* AChE in complex with aricept (2.50 Å) (PDB ID: 1eve).

CR	LIGAND																			
	ACL (CAS)		ACLTHIO (CAS)		AMB (PAS)		BW284C51 (BTH)		DECA (BTH)		DECI (PAS)		DONE (BTH)		EDR (CAS)		GALLMN (PAS)		(-)-GLNTM (CAS)	
	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS
1	-3.39 (55)	PAS	-3.54 (23)	MDL	-0.78 (1)	O	-6.63 (6)	BTH	-4.05 (64)	MDL	-5.49 (1)	PAS	-7.06 (8)	BTH	-4.52 (28)	PAS	-0.67 (2)	PAS	-7.03 (24)	CAS
2	-3.28 (6)	MDL	-3.46 (50)	PAS	-0.71 (1)	PAS	-6.32 (7)	BTH	-3.96 (6)	BTH	-5.00 (2)	PAS	-6.88 (8)	BTH	-4.35 (12)	PAS	-0.43 (3)	PAS	-6.88 (3)	CAS
3	-3.20 (3)	MDL	-3.26 (4)	PAS	-0.62 (1)	O	-6.19 (2)	BTH	-3.70 (11)	PAS	4.62 (1)	PAS	-6.70 (8)	PAS	-4.21 (29)	PAS	-0.43 (1)	PAS	-6.68 (2)	MDL
4	-3.10 (6)	PAS	-3.19 (4)	PAS	-0.60 (1)	O	-6.01 (8)	BTH	-3.50 (6)	BTH	-4.22 (1)	PAS	-6.61 (8)	BTH	-3.91 (1)	PAS	-0.43 (2)	PAS	-6.58 (14)	MDL
5	-3.07 (1)	CAS	-3.11 (8)	O	-0.37 (2)	O	-5.65 (2)	BTH	-3.17 (1)	BTH	-4.09 (3)	PAS	-6.48 (2)	BTH	-3.86 (3)	PAS	-0.37 (2)	PAS	-6.56 (18)	PAS
6	-3.04 (6)	MDL	-3.11 (6)	PAS	-0.28 (1)	O	-5.17 (5)	BTH	-3.03 (2)	MDL	-3.96 (1)	PAS	-6.34 (1)	PAS	-3.77 (2)	CAS	-0.03 (1)	PAS	-6.51 (2)	PAS
7	-2.86 (5)	O	-3.10 (1)	CAS	-0.22 (1)	PAS	-4.96 (5)	BTH	-3.02 (4)	O	-3.81 (1)	PAS	-6.30 (3)	BTH	-3.76 (3)	O	-0.01 (2)	PAS	-6.49 (5)	PAS
8	-2.80 (9)	O	-2.74 (1)	O	-0.22 (1)	O	-4.95 (1)	BTH	-1.49 (1)	O	-3.75 (2)	PAS	-6.15 (3)	BTH	-3.74 (7)	CAS	+0.08 (1)	O	-6.23 (4)	O
9	-2.71 (1)	CAS	-2.51 (1)	O	-0.18 (1)	O	-4.93 (1)	BTH	-1.46 (2)	O	-3.48 (1)	PAS	-6.11 (2)	BTH	-3.72 (10)	O	+0.11 (1)	PAS	-6.12 (3)	PAS
10	-2.55 (1)	MDL	-2.49 (1)	O	-0.17 (1)	O	-4.60 (1)	PAS	-1.42 (1)	O	-3.47 (1)	PAS	-5.98 (2)	BTH	-3.68 (3)	CAS	+0.26 (1)	PAS	-5.97 (1)	O
11–100		O		O		PAS/ O		PAS/ O /BTH		O		PAS/ O		PAS/ O BTH/		O		PAS/ O		O
√/ X (*/100)		(2) X		(1) X		(4) X		(44) √		(13) X		(20) √		(46) √		(12) X		(36) √		(27) √

CR = Cluster rank, PAS = Peripheral anionic site, CAS = Catalytic site, LBE = Lowest binding energy (kcal/mol), BS = Binding site, O = Bind to other site compared to PAS and CAS, MDL = Bind in between PAS and CAS, BTH = Bind at PAS and CAS, X = The binding location is incorrect according to literature, √ = The binding location is correct according to literature, (*/100) = Total conformation (the binding location is correct according to the literature) over 100 runs .

CR	LIGAND																	
	HUPA (CAS)		HUPB (CAS)		(-)-HUP X (CAS)		MF268 (PAS)		MPTP (CAS)		PROP (PAS)		TAC (CAS)		Δ^9-THC (PAS)		TUBO (PAS)	
	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS
1	-6.58 (35)	CAS	-6.85 (72)	CAS	-7.54 (56)	CAS	-5.73 (20)	MDL	-6.07 (88)	CAS	-6.02 (27)	PAS	-6.51 (28)	PAS	-7.31 (5)	MDL	-6.52 (25)	PAS
2	-6.21 (19)	PAS	-6.48 (3)	CAS	-7.32 (17)	MDL	-5.01 (1)	MDL	-5.73 (3)	PAS	-5.34 (1)	PAS	-6.30 (4)	PAS	-7.22 (5)	MDL	-5.89 (7)	PAS
3	-5.92 (6)	PAS	-6.41 (11)	PAS	-7.17 (3)	MDL	-5.00 (6)	MDL	-5.39 (4)	PAS	-4.81 (3)	PAS	-6.27 (55)	MDL	-7.16 (8)	MDL	-5.34 (4)	PAS
4	-5.88 (6)	MDL	-6.34 (4)	MDL	-6.07 (2)	O	-4.98 (12)	MDL	-5.38 (3)	PAS	-4.66 (1)	PAS	-5.59 (1)	O	-7.00 (8)	MDL	-5.02 (1)	PAS
5	-5.86 (13)	O	-6.32 (3)	PAS	-5.92 (2)	PAS	-4.59 (1)	MDL	-4.54 (1)	O	-4.63 (1)	PAS	-5.49 (3)	O	-6.87 (23)	MDL	-4.87 (7)	O
6	-5.79 (2)	PAS	-6.11 (1)	O	-5.82 (11)	O	-4.54 (5)	MDL	-3.54 (1)	O	-4.39 (3)	O	-5.43 (1)	O	-6.76 (4)	MDL	-4.85 (4)	O
7	-5.57 (1)	O	-5.99 (1)	PAS	-5.69 (2)	O	-4.44 (3)	MDL	-	-	-4.25 (2)	PAS	-5.39 (1)	O	-6.69 (2)	MDL	-4.68 (1)	O
8	-5.32 (6)	O	-5.72 (2)	O	-5.34 (2)	O	-4.42 (1)	MDL	-	-	-4.24 (1)	PAS	-5.29 (2)	O	-6.13 (1)	PAS	-4.65 (3)	O
9	-5.26 (1)	O	-5.57 (1)	O	-4.92 (3)	O	-4.33 (2)	MDL	-	-	-4.17 (1)	PAS	-5.26 (2)	O	-5.05 (6)	O	-4.55 (1)	O
10	-5.23 (1)	O	-5.13 (1)	O	-4.90 (2)	O	-4.31 (1)	MDL	-	-	-4.12 (1)	PAS	-4.87 (1)	O	-4.89 (1)	O	-4.49 (2)	O
11-100		O		O		-		PAS/ MDL /O	-	-		PAS/ O		O		O		O
(*/100)		(35)		(75)		(56)		(4)		(88)		(40)		(-)		(1)		(37)
√/ X		√		√		√		X		√		√		X		X		√

CR = Cluster rank, PAS = Peripheral anionic site, CAS = Catalytic site, LBE = Lowest binding energy (kcal/mol), BS = Binding site, O = Bind to other site compared to PAS and CAS, MDL = Bind in between PAS and CAS, BTH = Bind at PAS and CAS, X = The binding location is incorrect according to literature, √ = The binding location is correct according to literature, (*/100) = Total conformation (the binding location is correct according to the literature) over 100 runs (continued).

Appendix 4

Results from the docking of standard ligands with a structure derived from the crystal structure of *Torpedo californica* AChE in complex with aricept (2.50 Å) (PDB ID: 1eve) (with residue Phe330 replaced by Tyr) (arranged according to lowest binding energy).

Ligand	Cluster rank	Lowest binding energy (kcal/mol)	No. in cluster	Inhibition constant, K_i (μM)
Donepezil	1	-7.01	1	7.26
(-)-Galanthamine	1	-6.95	8	8.04
(-)-Galanthamine	2	-6.78	21	10.77
(-)- Huprine x	1	-6.70	22	12.36
D-Tubocurarine	1	-6.58	22	15.05
Huperzine B	1	-6.54	16	15.97
Δ^9-THC	1	-6.45	3	18.77
Tacrine	1	-6.26	16	25.62
Δ^9-THC	3	-6.09	10	34.15
Huperzine A	1	-6.07	4	35.78
Huperzine B	5	-6.04	21	37.24
Propidium	1	-5.96	20	43.13
Huperzine A	7	-5.87	20	49.82
Decidium	1	-5.86	1	50.62
Donepezil	2	-5.73	6	63.12
BW284C51	1	-5.60	2	78.22
MPTP	1	-5.48	10	96.54
MPTP	2	-5.48	53	96.75
MF268	1	-5.14	9	171.01
Edrophonium	1	-4.49	69	511.22
Decamethonium	1	-3.70	41	1960.00
Acetylthiocholine	1	-3.52	32	2650.00
Acetylcholine	1	-3.42	45	3090.00
Gallamine	1	-1.10	2	157350.00
Ambenonium	1	-0.88	1	225440.00

Appendix 5

Results from the docking of standard ligands with a structure derived from the crystal structure of *Torpedo californica* AChE in complex with aricept (2.50 Å) (PDB ID: 1eve) (with residue Phe330 replaced by Tyr) (arranged according to highest number in cluster).

Ligand	Cluster rank	Lowest binding energy (kcal/mol)	No. in cluster	Inhibition constant, K_i (µM)
Edrophonium	1	-4.49	69	511.22
MPTP	2	-5.48	53	96.75
Acetylcholine	1	-3.42	45	3090.00
Decamethonium	1	-3.70	41	1960.00
Acetylthiocholine	1	-3.52	32	2650.00
(-)- Huprine x	1	-6.70	22	12.36
D-Tubocurarine	1	-6.58	22	15.05
(-)-Galanthamine	2	-6.78	21	10.77
Huperzine B	5	-6.04	21	37.24
Huperzine A	7	-5.87	20	49.82
Propidium	1	-5.96	20	43.13
Huperzine B	1	-6.54	16	15.97
Tacrine	1	-6.26	16	25.62
MPTP	1	-5.48	10	96.54
Δ^9-THC	3	-6.09	10	34.15
MF268	1	-5.14	9	171.01
(-)-Galanthamine	1	-6.95	8	8.04
Donepezil	2	-5.73	6	63.12
Huperzine A	1	-6.07	4	35.78
Δ^9-THC	1	-6.45	3	18.77
BW284C51	1	-5.60	2	78.22
Gallamine	1	-1.10	2	157350.00
Ambenonium	1	-0.88	1	225440.00
Decidium	1	-5.86	1	50.62
Donepezil	1	-7.01	1	7.26

Appendix 6

Results from the docking of standard ligands with a structure derived from the crystal structure of *Torpedo californica* AChE in complex with aricept (2.50 Å) (PDB ID: 1eve) (with residue Phe330 replaced by Tyr).

CR	LIGAND																			
	ACL (CAS)		ACLTHIO (CAS)		AMB (PAS)		BW284C51 (BTH)		DECA (BTH)		DECI (PAS)		DONE (BTH)		EDR (CAS)		GALLMN (PAS)		(-)-GLNTM (CAS)	
	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS
1	-3.42 (45)	PAS	-3.52 (32)	PAS	-0.88 (1)	O	-5.60 (2)	PAS	-3.70 (41)	PAS	-5.86 (1)	PAS	-7.01 (1)	BTH	-4.49 (69)	PAS	-1.10 (2)	PAS	-6.95 (8)	PAS
2	-3.25 (5)	CAS	-3.46 (22)	PAS	-0.65 (1)	O	-5.35 (1)	MDL	-3.68 (1)	BTH	-5.06 (1)	PAS	-5.73 (6)	PAS	-4.02 (3)	CAS	-0.65 (1)	PAS	-6.78 (21)	PAS
3	-3.18 (9)	PAS	-3.43 (4)	PAS	-0.41 (1)	PAS	-5.26 (2)	PAS	-3.57 (2)	BTH	-3.91 (1)	PAS	-5.63 (6)	PAS	-3.96 (1)	PAS	-0.24 (2)	PAS	-6.76 (16)	PAS
4	-2.90 (3)	O	-3.31 (2)	MDL	-0.30 (1)	O	-4.82 (2)	MDL	-3.35 (12)	PAS	-3.88 (1)	PAS	-5.12 (1)	O	-3.92 (1)	MDL	-0.07 (1)	PAS	-6.28 (3)	PAS
5	-2.89 (1)	MDL	-3.27 (2)	CAS	-0.28 (1)	O	-4.54 (1)	PAS	-3.21 (2)	BTH	-3.64 (1)	O	-4.98 (1)	PAS	-3.72 (3)	O	-0.05 (3)	PAS	-6.26 (1)	PAS
6	-2.81 (3)	O	-3.22 (23)	O	-0.21 (1)	O	-4.40 (4)	PAS	-2.94 (2)	PAS	-3.63 (2)	PAS	-4.98 (1)	PAS	-3.69 (14)	O	-0.01 (1)	PAS	-6.25 (10)	O
7	-2.77 (17)	O	-3.17 (1)	PAS	-0.18 (1)	O	-4.28 (1)	PAS	-2.80 (2)	BTH	-3.56 (1)	O	-4.90 (1)	PAS	-3.50 (1)	CAS	+0.14 (1)	PAS	-6.09 (3)	PAS
8	-2.71 (2)	O	-2.85 (4)	O	-0.12 (1)	O	-4.16 (2)	PAS	-2.72 (1)	PAS	-3.24 (1)	PAS	-4.84 (4)	PAS	-3.48 (1)	O	+0.17 (3)	PAS	-5.09 (7)	O
9	-2.58 (2)	O	-2.54 (1)	O	-0.12 (1)	O	-4.16 (1)	O	-2.70 (9)	O	-3.21 (1)	PAS	-4.77 (1)	O	-3.24 (1)	O	+0.19 (1)	PAS	-4.89 (2)	O
10	-2.56 (3)	O	-2.52 (1)	O	+0.09 (1)	O	-3.98 (3)	O	-2.62 (1)	PAS	-2.84 (1)	PAS	-4.72 (2)	PAS	-3.20 (1)	O	+0.35 (1)	PAS	-4.85 (4)	O
11-100		O		O		PAS/ O		PAS/ O		PAS/ O		PAS/ O		PAS/ O		O		PAS/ O		O
(*/100) √/ X		(5) X		(2) X		(3) X		(3) X		(19) X		(16) √		(1) √		(4) X		(27) √		(-) X

CR = Cluster rank, PAS = Peripheral anionic site, CAS = Catalytic site, LBE = Lowest binding energy (kcal/mol), BS = Binding site, O = Bind to other site compared to PAS and CAS, MDL = Bind in between PAS and CAS, BTH = Bind at PAS and CAS, X = The binding location is incorrect according to literature, √ = The binding location is correct according to literature, (*/100) = Total conformation (the binding location is correct according to the literature) over 100 runs.

CR	LIGAND																	
	HUPA (CAS)		HUPB (CAS)		(-)-HUP X (CAS)		MF268 (PAS)		MPTP (CAS)		PROP (PAS)		TAC (CAS)		Δ^9-THC (PAS)		TUBO (PAS)	
	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS
1	-6.07 (4)	CAS	-6.54 (16)	CAS	-6.70 (22)	PAS	-5.14 (9)	PAS	-5.48 (10)	PAS	-5.96 (20)	PAS	-6.26 (16)	CAS	-6.45 (3)	PAS	-6.58 (22)	PAS
2	-5.99 (14)	PAS	-6.24 (18)	PAS	-6.44 (10)	PAS	-4.30 (6)	O	-5.48 (53)	PAS	-4.62 (1)	PAS	-6.23 (13)	CAS	-6.19 (2)	PAS	-6.04 (2)	PAS
3	-5.97 (1)	CAS	-6.11 (17)	CAS	-6.41 (18)	PAS	-4.19 (8)	PAS	-5.25 (11)	CAS	-4.61 (1)	PAS	-5.70 (7)	PAS	-6.09 (10)	PAS	-5.54 (5)	PAS
4	-5.97 (1)	CAS	-6.11 (9)	O	-6.23 (1)	CAS	-4.11 (1)	MDL	-4.81 (13)	O	-4.59 (1)	PAS	-5.57 (19)	O	-5.78 (1)	MDL	-5.10 (1)	PAS
5	-5.94 (16)	PAS	-6.04 (21)	PAS	-6.12 (7)	O	-4.11 (5)	PAS	-4.73 (3)	O	-4.57 (3)	PAS	-5.57 (1)	PAS	-5.72 (3)	PAS	-5.05 (2)	PAS
6	-5.88 (1)	CAS	-6.04 (6)	PAS	-5.83 (14)	O	-4.10 (4)	PAS	-4.56 (1)	O	-4.44 (1)	PAS	-5.51 (1)	O	-5.72 (6)	PAS	-5.01 (1)	PAS
7	-5.87 (20)	O	-5.73 (3)	O	-5.62 (2)	O	-4.10 (1)	O	-4.01 (6)	O	-4.27 (1)	PAS	-5.36 (3)	O	-5.68 (3)	PAS	-4.94 (6)	O
8	-5.68 (1)	PAS	-5.67 (3)	PAS	-5.54 (2)	O	-4.05 (3)	PAS	-3.90 (3)	O	-4.22 (1)	PAS	-5.35 (4)	O	-5.67 (3)	PAS	-4.87 (5)	O
9	-5.62 (2)	O	-5.61 (1)	O	-5.34 (1)	O	-3.98 (1)	PAS	-	-	-3.74 (1)	O	-5.33 (5)	PAS	-5.52 (1)	PAS	-4.77 (3)	PAS
10	-5.55 (8)	O	-5.57 (1)	PAS	-5.34 (1)	O	-3.82 (1)	PAS	-	-	-3.73 (1)	PAS	-5.32 (1)	CAS	-5.44 (2)	MDL	-4.75 (2)	O
11-100		PAS/ O		O		O		PAS/ O/ MDL	-	-		PAS/ O		PAS/ O		O		O
(*/100)		(7)		(33)		(1)		(42)		(11)		(72)		(30)		(31)		(33)
√/ X		√		√		X		√		X		√		√		√		√

CR = Cluster rank, PAS = Peripheral anionic site, CAS = Catalytic site, LBE = Lowest binding energy (kcal/mol), BS = Binding site, O = Bind to other site compared to PAS and CAS, MDL = Bind in between PAS and CAS, BTH = Bind at PAS and CAS, X = The binding location is incorrect according to literature, √ = The binding location is correct according to literature, (*/100) = Total conformation (the binding location is correct according to the literature) over 100 runs (continued).

Appendix 7

Results from the docking of standard ligands with a structure derived from the crystal structure of *Torpedo californica* AChE in complex with aricept (2.50 Á) (PDB ID: 1eve) (with residue Phe330 replaced by Tyr and minimized) (arranged according to lowest binding energy).

Ligand	Cluster rank	Lowest binding energy (kcal/mol)	No. in cluster	Inhibition constant, K_i (µM)
Huperzine A	1	-6.96	15	7.86
(-)-Galanthamine	1	-6.74	37	11.48
(-)- Huprine x	1	-6.56	18	15.51
D-Tubocurarine	1	-6.51	33	16.79
Huperzine B	1	-6.48	10	17.78
Decidium	1	-6.44	2	19.09
Donepezil	1	-6.39	4	20.66
Δ^9-THC	1	-6.36	4	21.94
(-)- Huprine x	4	-6.32	29	23.41
Tacrine	1	-6.26	20	25.83
Huperzine B	5	-6.13	35	32.28
BW284C51	1	-6.07	6	35.25
Δ^9-THC	4	-5.69	7	67.72
Propidium	1	-5.68	13	68.15
Donepezil	4	-5.55	7	85.78
MPTP	1	-5.44	27	102.14
MPTP	2	-5.41	33	107.53
MF268	1	-4.60	10	421.66
Edrophonium	1	-4.28	59	725.03
Decamethonium	1	-3.67	8	2040.00
Decamethonium	2	-3.58	34	2370.00
Acetylthiocholine	1	-3.42	39	3100.00
Acetylcholine	1	-3.25	39	4120.00
Ambenonium	1	-2.19	1	24900.00
Gallamine	1	-0.38	1	524600.00

Appendix 8

Results from the docking of standard ligands with a structure derived from the crystal structure of *Torpedo californica* AChE in complex with aricept (2.50 Å) (PDB ID: 1eve) (with residue Phe330 replaced by Tyr and minimized) (arranged according to highest number in cluster).

Ligand	Cluster rank	Lowest binding energy (kcal/mol)	No. in cluster	Inhibition constant, K_i (μM)
Edrophonium	1	-4.28	59	725.03
Acetylcholine	1	-3.25	39	4120.00
Acetylthiocholine	1	-3.42	39	3100.00
(-)-Galanthamine	1	-6.74	37	11.48
Huperzine B	5	-6.13	35	32.28
Decamethonium	2	-3.58	34	2370.00
MPTP	2	-5.41	33	107.53
D-Tubocurarine	1	-6.51	33	16.79
(-)- Huprine x	4	-6.32	29	23.41
MPTP	1	-5.44	27	102.14
Tacrine	1	-6.26	20	25.83
(-)- Huprine x	1	-6.56	18	15.51
Huperzine A	1	-6.96	15	7.86
Propidium	1	-5.68	13	68.15
Huperzine B	1	-6.48	10	17.78
MF268	1	-4.60	10	421.66
Decamethonium	1	-3.67	8	2040.00
Donepezil	4	-5.55	7	85.78
Δ^9-THC	4	-5.69	7	67.72
BW284C51	1	-6.07	6	35.25
Donepezil	1	-6.39	4	20.66
Δ^9-THC	1	-6.36	4	21.94
Decidium	1	-6.44	2	19.09
Ambenonium	1	-2.19	1	24900.00
Gallamine	1	-0.38	1	524600.00

Appendix 9

Results from the docking of standard ligands with a structure derived from the crystal structure of *Torpedo californica* AChE in complex with aricept (2.50 Å) (PDB ID: 1eve) (with residue Phe330 replaced by Tyr and minimized).

CR	LIGAND																			
	ACL (CAS)		ACLTHIO (CAS)		AMB (PAS)		BW284C51 (BTH)		DECA (BTH)		DECI (PAS)		DONE (BTH)		EDR (CAS)		GALLMN (PAS)		(-)-GLNTM (CAS)	
	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS
1	-3.25 (39)	PAS	-3.42 (39)	PAS	-2.19 (1)	PAS	-6.07 (6)	PAS	-3.67 (8)	BTH	-6.44 (2)	PAS	-6.39 (4)	PAS	-4.28 (59)	PAS	-0.38 (1)	PAS	-6.74 (37)	PAS
2	-3.17 (19)	PAS	-3.30 (1)	CAS	-2.05 (1)	PAS	-5.11 (1)	BTH	-3.58 (34)	PAS	-5.43 (3)	PAS	-5.98 (5)	O	-4.19 (13)	PAS	-0.13 (1)	PAS	-6.61 (13)	PAS
3	-3.09 (5)	O	-3.30 (10)	PAS	-2.03 (1)	PAS	-4.55 (5)	PAS	-3.32 (2)	BTH	-5.24 (1)	PAS	-5.76 (1)	O	-3.96 (3)	O	-0.08 (2)	PAS	-6.39 (2)	PAS
4	-3.09 (2)	O	-3.26 (2)	CAS	-1.57 (1)	O	-3.98 (1)	PAS	-3.19 (2)	BTH	-5.18 (1)	PAS	-5.55 (7)	O	-3.95 (4)	CAS	-0.00 (2)	PAS	-6.25 (7)	PAS
5	-2.91 (1)	O	-3.24 (14)	PAS	-1.42 (1)	PAS	-3.80 (4)	PAS	-3.09 (9)	PAS	-4.63 (1)	PAS	-5.16 (6)	O	-3.88 (5)	CAS	+0.08 (1)	PAS	-6.24 (7)	O
6	-2.89 (14)	O	-3.17 (13)	O	-1.38 (1)	PAS	-3.77 (1)	O	-2.66 (2)	PAS	-4.60 (2)	PAS	-5.12 (1)	O	-3.54 (2)	CAS	+0.28 (2)	PAS	-5.91 (10)	O
7	-2.84 (1)	O	-3.10 (2)	CAS	-0.99 (1)	PAS	-3.71 (1)	PAS	-2.56 (2)	PAS	-4.17 (1)	PAS	-5.09 (3)	O	-3.51 (2)	CAS	+0.30 (1)	O	-5.07 (6)	O
8	-2.82 (3)	CAS	-3.02 (1)	CAS	-0.91 (1)	O	-3.68 (1)	PAS	-2.38 (5)	O	-4.13 (2)	PAS	-5.02 (2)	O	-3.43 (2)	O	+0.37 (1)	PAS	-4.95 (1)	O
9	-2.73 (1)	CAS	-2.92 (1)	PAS	-0.89 (1)	O	-3.55 (1)	PAS	-2.33 (1)	PAS	-3.97 (1)	PAS	-4.91 (2)	O	-3.42 (3)	O	+0.44 (2)	PAS	-4.89 (5)	O
10	-2.67 (2)	O	-2.89 (1)	CAS	-0.81 (1)	O	-3.53 (2)	PAS	-2.28 (3)	O	-3.96 (2)	PAS	-4.87 (2)	O	-3.34 (1)	O	+0.46 (1)	PAS	-4.70 (2)	O
11-100		CAS/ O		MDL /O		PAS/ O		PAS/ O		PAS/ O		PAS/ O		PAS/ O		CAS/ O		PAS/ O		O
(*/100)		(4)		(7)		(10)		(1)		(12)		(30)		(-)		(14)		(24)		(-)
√/ X		X		X		√		X		√		√		X		X		√		X

CR = Cluster rank, PAS = Peripheral anionic site, CAS = Catalytic site, LBE = Lowest binding energy (kcal/mol), BS = Binding site, O = Bind to other site compared to PAS and CAS, MDL = Bind in between PAS and CAS, BTH = Bind at PAS and CAS, X = The binding location is incorrect according to literature, √ = The binding location is correct according to literature, (*/100) = Total conformation (the binding location is correct according to the literature) over 100 runs.

CR	LIGAND																	
	HUPA (CAS)		**HUPB (CAS)**		**(-)-HUP X (CAS)**		**MF268 (PAS)**		**MPTP (CAS)**		**PROP (PAS)**		**TAC (CAS)**		**Δ^9-THC (PAS)**		**TUBO (PAS)**	
	LBE	**BS**	**LBE**	**BS**	**LBE**	**BS**	**LBE**	**BS**	**LBE**	**BS**	**LBE**	**BS**	**LBE**	**BS**	**LBE**	**BS**	**LBE**	**BS**
1	-6.96 (15)	CAS	-6.48 (10)	CAS	-6.56 (18)	PAS	-4.60 (10)	PAS	-5.44 (27)	PAS	-5.68 (13)	PAS	-6.26 (20)	CAS	-6.36 (4)	PAS	-6.51 (33)	PAS
2	-5.86 (1)	CAS	-6.44 (8)	CAS	-6.44 (1)	CAS	-4.58 (1)	PAS	-5.41 (33)	CAS	-5.07 (5)	PAS	-6.26 (17)	CAS	-5.88 (2)	PAS	-5.72 (3)	PAS
3	-5.76 (8)	O	-6.23 (12)	O	-6.38 (10)	PAS	-4.46 (6)	PAS	-5.26 (2)	CAS	-4.97 (3)	PAS	-5.72 (4)	O	-5.84 (3)	PAS	-5.44 (1)	PAS
4	-5.71 (8)	MDL	-6.20 (12)	CAS	-6.32 (29)	PAS	-4.40 (1)	PAS	-5.12 (15)	PAS	-4.69 (1)	PAS	-5.63 (3)	O	-5.69 (7)	PAS	-4.94 (5)	O
5	-5.55 (9)	MDL	-6.13 (35)	PAS	-6.19 (1)	CAS	-4.38 (10)	PAS	-4.70 (14)	O	-4.65 (1)	PAS	-5.34 (13)	O	-5.65 (3)	PAS	-4.94 (3)	O
6	-5.42 (4)	O	-6.10 (8)	PAS	-5.75 (16)	O	-4.07 (1)	PAS	-4.53 (2)	O	-4.58 (1)	PAS	-5.31 (11)	PAS	-5.63 (4)	PAS	-4.87 (3)	PAS
7	-5.40 (3)	PAS	-5.59 (3)	PAS	-5.61 (5)	O	-4.04 (3)	PAS	-4.48 (2)	O	-4.46 (1)	PAS	-5.21 (5)	O	-5.59 (3)	PAS	-4.80 (2)	O
8	-5.38 (6)	PAS	-5.57 (1)	PAS	-5.54 (3)	O	-4.01 (2)	PAS	-4.32 (2)	O	-4.44 (3)	PAS	-5.17 (2)	O	-5.34 (2)	PAS	-4.71 (5)	O
9	-5.37 (18)	O	-5.53 (1)	O	-5.33 (1)	O	-4.00 (1)	O	-3.97 (1)	O	-3.74 (1)	O	-5.17 (8)	PAS	-5.30 (1)	PAS	-4.48 (2)	O
10	-5.29 (7)	O	-5.47 (1)		-5.26 (2)	O	-4.00 (1)	PAS	-3.89 (1)	O	-3.69 (1)	O	-5.16 (2)	PAS	-4.99 (7)	O	-4.42 (1)	O
11-100		PAS/ O		PAS/ O		O		PAS/ CAS/ O /MDL		O		PAS/ O		PAS/ O		PAS/ O		O
(*/100) √/ X		(16) √		(30) √		(2) X		(49) √		(35) X		(30) √		(37) √		(31) √		(40) √

CR = Cluster rank, PAS = Peripheral anionic site, CAS = Catalytic site, LBE = Lowest binding energy (kcal/mol), BS = Binding site, O = Bind to other site compared to PAS and CAS, MDL = Bind in between PAS and CAS, BTH = Bind at PAS and CAS, X = The binding location is incorrect according to literature, √ = The binding location is correct according to literature, (*/100) = Total conformation (the binding location is correct according to the literature) over 100 runs (continued).

Appendix 10

Results from the docking of standard ligands with a structure derived from the crystal structure of *Torpedo californica* AChE in complex with aricept (2.50 Å) (PDB ID: 1eve) (with residue Phe330 replaced by Tyr, Ser81 replaced by Thr, Glu73 replaced by Thr and Gln74 replaced by Leu) (arranged according to lowest binding energy).

Ligand	Cluster rank	Lowest binding energy (kcal/mol)	No. in cluster	Inhibition constant, K_i (μM)
(-)-Galanthamine	1	-7.01	32	7.23
(-)- Huprine x	1	-6.72	21	11.95
D-Tubocurarine	1	-6.70	19	12.24
Huperzine A	1	-6.59	14	14.72
Δ^9-THC	1	-6.59	9	14.77
Huperzine B	1	-6.53	4	16.33
(-)- Huprine x	3	-6.43	27	19.21
Δ^9-THC	2	-6.35	16	22.29
Decidium	1	-6.29	2	24.43
Tacrine	1	-6.27	22	25.41
D-Tubocurarine	2	-6.27	22	25.45
Donepezil	1	-6.26	6	25.65
Huperzine A	2	-6.22	36	27.65
Huperzine B	4	-6.18	35	29.39
Propidium	1	-6.14	16	31.64
BW284C51	1	-5.80	8	56.07
MPTP	1	-5.73	72	63.13
MF268	1	-5.49	10	94.10
Edrophonium	1	-4.49	72	512.67
Decamethonium	1	-3.69	6	1980.00
Decamethonium	2	-3.68	36	2020.00
Acetylthiocholine	1	-3.52	50	2610.00
Acetylcholine	1	-3.41	57	3180.00
Ambenonium	1	-1.35	1	102700.00
Gallamine	1	-1.14	2	147020.00

Appendix 11

Results from the docking of standard ligands with a structure derived from the crystal structure of *Torpedo californica* AChE in complex with aricept (2.50 Å) (PDB ID: 1eve) (with residue Phe330 replaced by Tyr, Ser81 replaced by Thr, Glu73 replaced by Thr and Gln74 replaced by Leu) (arranged according to highest number in cluster).

Ligand	Cluster rank	Lowest binding energy (kcal/mol)	No. in cluster	Inhibition constant, K_i (μM)
Edrophonium	1	-4.49	72	512.67
MPTP	1	-5.73	72	63.13
Acetylcholine	1	-3.41	57	3180.00
Acetylthiocholine	1	-3.52	50	2610.00
Decamethonium	2	-3.68	36	2020.00
Huperzine A	2	-6.22	36	27.65
Huperzine B	4	-6.18	35	29.39
(-)-Galanthamine	1	-7.01	32	7.23
(-)- Huprine x	3	-6.43	27	19.21
Tacrine	1	-6.27	22	25.41
D-Tubocurarine	2	-6.27	22	25.45
(-)- Huprine x	1	-6.72	21	11.95
D-Tubocurarine	1	-6.70	19	12.24
Propidium	1	-6.14	16	31.64
Δ^9-THC	2	-6.35	16	22.29
Huperzine A	1	-6.59	14	14.72
MF268	1	-5.49	10	94.10
Δ^9-THC	1	-6.59	9	14.77
BW284C51	1	-5.80	8	56.07
Decamethonium	1	-3.69	6	1980.00
Donepezil	1	-6.26	6	25.65
Huperzine B	1	-6.53	4	16.33
Decidium	1	-6.29	2	24.43
Gallamine	1	-1.14	2	147020.00
Ambenonium	1	-1.35	1	102700.00

Appendix 12

Results from the docking of standard ligands with a structure derived from the crystal structure of *Torpedo californica* AChE in complex with aricept (2.50 Á) (PDB ID: 1eve) (with residue Phe330 replaced by Tyr, Ser81 replaced by Thr, Glu73 replaced by Thr and Gln74 replaced by Leu).

CR	LIGAND																			
	ACL (CAS)		ACLTHIO (CAS)		AMB (PAS)		BW284C51 (BTH)		DECA (BTH)		DECI (PAS)		DONE (BTH)		EDR (CAS)		GALLMN (PAS)		(-)-GLNTM (CAS)	
	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS
1	-3.41 (57)	PAS	-3.52 (50)	PAS	-1.35 (1)	PAS	-5.80 (8)	PAS	-3.69 (6)	BTH	-6.29 (2)	PAS	-6.26 (6)	PAS	-4.49 (72)	PAS	-1.14 (2)	PAS	-7.01 (32)	PAS
2	-3.38 (1)	MDL	-3.52 (6)	CAS	-1.30 (1)	PAS	-4.81 (2)	PAS	-3.68 (36)	PAS	-5.78 (1)	PAS	-5.96 (4)	PAS	-4.20 (6)	PAS	-0.93 (1)	PAS	-6.95 (14)	PAS
3	-3.30 (2)	CAS	-3.46 (20)	PAS	-0.49 (1)	O	-4.27 (3)	PAS	-3.55 (2)	BTH	-5.53 (2)	PAS	-5.82 (1)	MDL	-3.88 (1)	PAS	-0.89 (1)	PAS	-6.88 (1)	PAS
4	-3.11 (4)	PAS	-3.34 (2)	PAS	-0.44 (1)	PAS	-3.95 (2)	PAS	-3.50 (5)	BTH	-5.13 (1)	PAS	-5.59 (1)	PAS	-3.78 (1)	CAS	-0.45 (1)	PAS	-6.73 (6)	PAS
5	-2.81 (1)	O	-3.25 (3)	PAS	+0.88 (1)	O	-3.94 (1)	PAS	-3.48 (13)	PAS	-4.90 (1)	PAS	-5.48 (5)	PAS	-3.65 (1)	O	+0.14 (1)	PAS	-6.46 (3)	PAS
6	-2.75 (19)	O	-3.06 (9)	O	+0.11 (1)	O	-3.94 (1)	PAS	-3.00 (1)	BTH	-4.84 (1)	PAS	-5.36 (6)	PAS	-3.64 (10)	O	+0.35 (1)	PAS	-6.18 (14)	O
7	-2.71 (1)	MDL	-2.69 (2)	O	+0.12 (1)	O	-3.84 (1)	PAS	-2.94 (2)	BTH	-4.49 (1)	PAS	-5.23 (1)	PAS	-3.47 (1)	O	+0.40 (1)	O	-5.89 (1)	CAS
8	-2.56 (1)	PAS	-2.49 (1)	O	+0.16 (1)	PAS	-3.83 (2)	PAS	-2.91 (1)	MDL	-4.49 (1)	PAS	-4.97 (2)	PAS	-3.45 (1)	CAS	+0.42 (1)	PAS	-4.90 (4)	O
9	-2.49 (3)	O	-2.40 (2)	O	+0.20 (1)	O	-3.78 (1)	PAS	-2.78 (6)	O	-4.38 (1)	PAS	-4.89 (1)	PAS	-3.43 (1)	CAS	+0.42 (1)	O	-4.87 (3)	O
10	-2.38 (3)	O	-2.28 (1)	O	+0.30 (1)	O	-3.67 (3)	PAS	-2.72 (1)	PAS	-4.07 (1)	PAS	-4.86 (1)	PAS	-3.07 (1)	O	+0.57 (1)	O	-4.81 (1)	O
11-100		O		O		PAS/ O		PAS/ O		PAS/ O		PAS/ O		PAS/ O		O		PAS/ O		O
(*/100) √/ X		(2) X		(6) X		(7) √		(-) X		(16) √		(31) √		(-) √		(3) X		(28) √		(1) X

CR = Cluster rank, PAS = Peripheral anionic site, CAS = Catalytic site, LBE = Lowest binding energy (kcal/mol), BS = Binding site, O = Bind to other site compared to PAS and CAS, MDL = Bind in between PAS and CAS, BTH = Bind at PAS and CAS, X = The binding location is incorrect according to literature, √ = The binding location is correct according to literature, (*/100) = Total conformation (the binding location is correct according to the literature) over 100 runs.

CR	LIGAND																	
	HUPA (CAS)		HUPB (CAS)		(-)-HUP X (CAS)		MF268 (PAS)		MPTP (CAS)		PROP (PAS)		TAC (CAS)		Δ^9-THC (PAS)		TUBO (PAS)	
	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS
1	-6.59 (14)	CAS	-6.53 (4)	CAS	-6.72 (21)	PAS	-5.49 (10)	PAS	-5.73 (72)	PAS	-6.14 (16)	PAS	-6.27 (22)	CAS	-6.59 (9)	PAS	-6.70 (19)	PAS
2	-6.22 (36)	PAS	-6.49 (34)	CAS	-6.55 (22)	PAS	-5.11 (1)	MDL	-5.26 (3)	PAS	-5.53 (4)	PAS	-6.25 (15)	CAS	-6.35 (16)	PAS	-6.27 (22)	PAS
3	-6.09 (4)	PAS	-6.18 (9)	PAS	-6.43 (27)	PAS	-4.92 (6)	PAS	-5.24 (12)	CAS	-5.52 (6)	PAS	-5.55 (23)	PAS	-5.90 (2)	PAS	-6.05 (1)	PAS
4	5.97 (3)	PAS	-6.18 (35)	PAS	-6.42 (2)	CAS	-4.68 (5)	O	-5.04 (1)	MDL	-5.15 (1)	PAS	-5.54 (12)	O	-5.84 (4)	MDL	-5.56 (1)	PAS
5	-5.85 (10)	O	-5.98 (8)	O	-5.95 (1)	PAS	-4.62 (2)	PAS	-4.77 (2)	O	-4.83 (2)	PAS	-5.45 (3)	O	-5.79 (1)	PAS	-5.51 (3)	PAS
6	-5.71 (2)	PAS	-5.81 (3)	PAS	-5.81 (16)	O	-4.50 (4)	PAS	-4.72 (7)	O	-4.56 (3)	PAS	-5.43 (9)	PAS	-5.72 (3)	PAS	-5.30 (1)	PAS
7	-5.35 (7)	O	-5.77 (4)	O	-5.64 (3)	O	-4.45 (6)	PAS	-4.29 (2)	O	-4.55 (1)	PAS	-5.35 (3)	O	-5.53 (1)	PAS	-5.28 (2)	O
8	-5.23 (7)	O	-5.70 (1)	PAS	-5.59 (2)	O	-4.37 (2)	PAS	-3.79 (1)	O	-4.48 (1)	O	-5.32 (1)	PAS	-5.50 (2)	PAS	-5.17 (1)	PAS
9	-5.11 (1)	O	-5.07 (2)	O	-5.19 (1)	O	-4.27 (3)	PAS	-	-	-4.43 (1)	PAS	-5.20 (3)	O	-5.39 (2)	PAS	-5.16 (7)	O
10	-4.94 (3)	O	-	-	-4.92 (2)	O	-4.24 (2)	PAS	-	-	-3.93 (1)	PAS	-5.07 (1)	O	-5.33 (1)	PAS	-4.78 (1)	O
11-100		O	-	-		O		PAS/ O/ MDL	-	-		PAS/ O		O		PAS/ O		O
(*/100) √/ X		(14) √		(38) √		(2) X		(55) √		(12) X		(46) √		(37) √		(39) √		(48) √

CR = Cluster rank, PAS = Peripheral anionic site, CAS = Catalytic site, LBE = Lowest binding energy (kcal/mol), BS = Binding site, O = Bind to other site compared to PAS and CAS, MDL = Bind in between PAS and CAS, BTH = Bind at PAS and CAS, X = The binding location is incorrect according to literature, √ = The binding location is correct according to literature, (*/100) = Total conformation (the binding location is correct according to the literature) over 100 runs (continued).

Appendix 13

Results from the docking of standard ligands with a structure derived from the crystal structure of *Torpedo californica* AChE in complex with aricept (2.50 Å) (PDB ID: 1eve) (with residue Phe330 replaced by Tyr, Ser81 replaced by Thr, Glu73 replaced by Thr and Gln74 replaced by Leu followed by energy minimization) (arranged according to lowest binding energy).

Ligand	Cluster rank	Lowest binding energy (kcal/mol)	No. in cluster	Inhibition constant, K_i (μM)
(-)-Galanthamine	1	-6.95	31	8.09
Huperzine A	1	-6.89	25	8.87
Δ^9-THC	1	-6.62	4	14.02
D-Tubocurarine	1	-6.58	18	14.96
Huperzine B	1	-6.56	16	15.47
(-)- Huprine x	1	-6.54	16	16.02
Donepezil	1	-6.45	1	18.58
(-)- Huprine x	3	-6.32	26	23.19
Tacrine	1	-6.28	21	25.07
D-Tubocurarine	2	-6.27	32	25.30
Huperzine B	4	-6.13	50	32.07
Δ^9-THC	3	-6.05	10	36.75
BW284C51	1	-5.86	7	50.29
Decidium	1	-5.81	1	55.06
Propidium	1	-5.65	19	72.00
MPTP	1	-5.48	27	95.76
Tacrine	3	-5.43	38	104.53
MPTP	2	-5.38	43	114.15
MF268	1	-4.66	13	385.30
Edrophonium	1	-4.36	65	635.49
Decamethonium	1	-3.65	5	2120.00
Decamethonium	2	-3.62	36	2220.00
Acetylthiocholine	1	-3.47	60	2860.00
Gallamine	1	-1.42	3	90420.00
Ambenonium	1	-0.78	1	266740.00

Appendix 14

Results from the docking of standard ligands with a structure derived from the crystal structure of *Torpedo californica* AChE in complex with aricept (2.50 Å) (PDB ID: 1eve) (with residue Phe330 replaced by Tyr, Ser81 replaced by Thr, Glu73 replaced by Thr and Gln74 replaced by Leu followed by energy minimization) (arranged according to highest number in cluster).

Ligand	Cluster rank	Lowest binding energy (kcal/mol)	No. in cluster	Inhibition constant, K_i (μM)
Edrophonium	1	-4.36	65	635.49
Acetylthiocholine	1	-3.47	60	2860.00
Huperzine B	4	-6.13	50	32.07
MPTP	2	-5.38	43	114.15
Tacrine	3	-5.43	38	104.53
Decamethonium	2	-3.62	36	2220.00
D-Tubocurarine	2	-6.27	32	25.30
(-)-Galanthamine	1	-6.95	31	8.09
MPTP	1	-5.48	27	95.76
(-)- Huprine x	3	-6.32	26	23.19
Huperzine A	1	-6.89	25	8.87
Tacrine	1	-6.28	21	25.07
Propidium	1	-5.65	19	72.00
D-Tubocurarine	1	-6.58	18	14.96
Huperzine B	1	-6.56	16	15.47
(-)- Huprine x	1	-6.54	16	16.02
MF268	1	-4.66	13	385.30
Δ^9-THC	3	-6.05	10	36.75
BW284C51	1	-5.86	7	50.29
Decamethonium	1	-3.65	5	2120
Δ^9-THC	1	-6.62	4	14.02
Gallamine	1	-1.42	3	90420.00
Donepezil	1	-6.45	1	18.58
Decidium	1	-5.81	1	55.06
Ambenonium	1	-0.78	1	266740.00

Appendix 15

Results from the docking of standard ligands with a structure derived from the crystal structure of *Torpedo californica* AChE in complex with aricept (2.50 Å) (PDB ID: 1eve) (with residue Phe330 replaced by Tyr, Ser81 replaced by Thr, Glu73 replaced by Thr and Gln74 replaced by Leu followed by energy minimization).

CR	LIGAND																			
	ACL (CAS)		ACLTHIO (CAS)		AMB (PAS)		BW284C51 (BTH)		DECA (BTH)		DECI (PAS)		DONE (BTH)		EDR (CAS)		GALLMN (PAS)		(-)-GLNTM (CAS)	
	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS
1	-3.25 (45)	PAS	-3.47 (60)	PAS	-0.78 (1)	O	-5.86 (7)	PAS	-3.65 (5)	BTH	-5.81 (1)	PAS	-6.45 (1)	BTH	-4.36 (65)	PAS	-1.42 (3)	PAS	-6.95 (31)	PAS
2	-3.11 (12)	PAS	-3.39 (3)	CAS	-0.28 (1)	PAS	-5.35 (2)	PAS	-3.62 (36)	PAS	-5.53 (2)	PAS	-6.07 (2)	PAS	-4.31 (9)	PAS	-1.30 (2)	PAS	-6.82 (19)	PAS
3	-2.96 (1)	CAS	-3.28 (3)	MDL	-0.11 (1)	O	-4.94 (3)	PAS	-3.50 (12)	PAS	-4.52 (1)	PAS	-6.07 (2)	BTH	-3.97 (2)	CAS	-0.77 (1)	PAS	-6.61 (13)	PAS
4	-2.93 (11)	O	-3.24 (6)	PAS	+0.00 (1)	O	-4.40 (1)	PAS	-3.12 (6)	PAS	-4.48 (1)	PAS	-5.21 (2)	PAS	-3.96 (3)	MDL	-0.56 (2)	PAS	-6.43 (8)	PAS
5	-2.91 (1)	CAS	-3.24 (6)	PAS	+0.25 (1)	O	-4.24 (2)	PAS	-3.12 (7)	PAS	-4.39 (1)	PAS	-5.14 (3)	PAS	-3.95 (2)	CAS	-0.49 (1)	PAS	-6.32 (1)	CAS
6	-2.88 (3)	MDL	-3.23 (2)	MDL	+0.32 (1)	PAS	-4.21 (3)	PAS	-2.63 (5)	O	-3.95 (1)	PAS	-5.09 (4)	PAS	3.59 (5)	O	-0.45 (1)	PAS	-5.80 (5)	O
7	-2.85 (5)	PAS	-3.11 (2)	MDL	+0.37 (1)	O	-4.03 (2)	PAS	-2.39 (1)	PAS	-3.92 (2)	PAS	-4.91 (2)	PAS	-3.44 (1)	O	-0.37 (1)	PAS	-4.89 (2)	O
8	-2.75 (2)	CAS	-3.10 (11)	O	+0.52 (1)	O	-3.76 (2)	PAS	-2.32 (1)	PAS	-3.85 (1)	PAS	-4.81 (1)	PAS	-3.44 (1)	CAS	-0.30 (2)	O	-4.88 (1)	O
9	-2.68 (1)	CAS	-3.07 (1)	PAS	+0.53 (1)	O	-3.73 (1)	PAS	-2.14 (1)	PAS	-3.75 (1)	PAS	-4.71 (2)	O	-3.42 (7)	O	-0.20 (1)	PAS	-4.78 (1)	O
10	-2.58 (2)	O	-2.95 (1)	CAS	+0.69 (1)	O	-3.71 (2)	PAS	-2.12 (1)	O	-3.64 (2)	O	-4.71 (2)	PAS	-3.37 (1)	CAS	-0.16 (3)	PAS	-4.68 (1)	O
11-100		O		O		O		PAS/ O		PAS/ O		PAS/ O		PAS/ O /BTH		O		PAS/ O		O
(*/100) √/ X		(5) X		(4) X		(2) X		(-) X		(5) √		(24) √		(4) √		(6) X		(38) √		(1) X

CR = Cluster rank, PAS = Peripheral anionic site, CAS = Catalytic site, LBE = Lowest binding energy (kcal/mol), BS = Binding site, O = Bind to other site compared to PAS and CAS, MDL = Bind in between PAS and CAS, BTH = Bind at PAS and CAS, X = The binding location is incorrect according to literature, √ = The binding location is correct according to literature, (*/100) = Total conformation (the binding location is correct according to the literature) over 100 runs.

CR	LIGAND																	
	HUPA (CAS)		HUPB (CAS)		(-)-HUP X (CAS)		MF268 (PAS)		MPTP (CAS)		PROP (PAS)		TAC (CAS)		Δ9-THC (PAS)		TUBO (PAS)	
	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS
1	-6.89 (25)	CAS	-6.56 (16)	CAS	-6.54 (16)	PAS	-4.66 (13)	PAS	-5.48 (27)	CAS	-5.65 (19)	PAS	-6.28 (21)	CAS	-6.62 (4)	PAS	-6.58 (18)	PAS
2	-5.90 (22)	PAS	-6.55 (15)	CAS	-6.53 (22)	PAS	-4.51 (2)	MDL	-5.38 (43)	PAS	-5.16 (3)	PAS	-6.27 (14)	CAS	-6.22 (1)	PAS	-6.27 (32)	PAS
3	-5.86 (1)	CAS	-6.15 (8)	PAS	-6.32 (26)	PAS	-4.42 (9)	PAS	-5.19 (10)	PAS	-4.71 (1)	PAS	-5.43 (38)	PAS	-6.05 (10)	PAS	-5.78 (1)	PAS
4	-5.80 (1)	CAS	-6.13 (50)	PAS	-6.01 (4)	PAS	-4.39 (7)	PAS	-5.07 (4)	PAS	-4.65 (2)	PAS	-5.38 (8)	O	-5.85 (9)	PAS	-5.50 (2)	PAS
5	-5.73 (3)	PAS	-6.12 (2)	CAS	-5.72 (14)	O	-4.34 (2)	PAS	-4.71 (9)	O	-4.55 (5)	PAS	-5.26 (3)	O	-5.84 (8)	PAS	-5.31 (6)	PAS
6	-5.71 (2)	O	-5.77 (2)	PAS	-5.64 (1)	O	-4.34 (1)	MDL	-4.45 (1)	O	-4.38 (1)	PAS	-5.25 (1)	PAS	-5.81 (6)	PAS	-5.27 (1)	PAS
7	-5.70 (20)	PAS	-5.49 (2)	O	-5.57 (4)	O	-4.18 (1)	PAS	-4.30 (3)	O	-4.32 (1)	PAS	-5.21 (2)	PAS	-5.75 (2)	PAS	-5.21 (3)	O
8	-5.51 (1)	PAS	-5.25 (1)	O	-5.51 (1)	O	-4.14 (7)	PAS	-4.13 (1)	O	-4.30 (1)	PAS	-5.17 (1)	PAS	-5.17 (5)	PAS	-4.94 (1)	PAS
9	-5.34 (12)	O	-5.02 (1)	O	-5.45 (1)	O	-3.97 (2)	PAS	-4.04 (1)	O	-4.06 (1)	PAS	-5.17 (2)	O	-5.05 (1)	O	-4.52 (2)	O
10	-5.30 (1)	O	-4.94 (1)	O	-5.02 (1)	O	-3.97 (4)	PAS	-3.64 (1)	O	-4.04 (1)	O	-5.14 (1)	O	-4.98 (1)	MDL	-4.52 (2)	O
11-100		O		O		O		PAS/ O /MDL		-		PAS/O		PAS/ O		O		O
(*/100) √/ X		(27) √		(33) √		(-) X		(57) √		(27) √		(37) √		(35) √		(45) √		(61) √

CR = Cluster rank, PAS = Peripheral anionic site, CAS = Catalytic site, LBE = Lowest binding energy (kcal/mol), BS = Binding site, O = Bind to other site compared to PAS and CAS, MDL = Bind in between PAS and CAS, BTH = Bind at PAS and CAS, X = The binding location is incorrect according to literature, √ = The binding location is correct according to literature, (*/100) = Total conformation (the binding location is correct according to the literature) over 100 runs (continued).

Appendix 16

Results from the docking of standard ligands with a structure derived from the crystal structure of mouse acetylcholinesterase - decidium complex (2.35 A°) (PDB ID: 1j07) (arranged according to lowest binding energy).

Ligand	Cluster rank	Lowest binding energy (kcal/mol)	No. in cluster	Inhibition constant, K_i (μM)
Δ^9-THC	1	-7.45	4	3.49
(-)- Huprine x	1	-7.29	11	4.55
Huperzine B	1	-7.12	57	6.03
Huperzine A	1	-7.11	20	6.16
(-)- Huprine x	2	-7.04	27	6.87
(-)-Galanthamine	1	-7.03	4	7.09
Δ^9-THC	3	-6.84	8	9.63
BW284C51	1	-6.54	1	16.21
D-Tubocurarine	1	-6.50	4	17.12
Tacrine	1	-6.43	24	19.47
Propidium	1	-5.94	3	44.55
Donepezil	1	-5.84	2	52.77
(-)-Galanthamine	8	-5.84	14	52.76
MPTP	1	-5.72	28	63.66
Donepezil	2	-5.67	4	69.83
MF268	1	-5.44	5	103.74
MPTP	3	-5.36	54	118.58
Edrophonium	1	-4.69	16	366.58
Decidium	1	-4.63	2	401.84
D-Tubocurarine	14	-4.36	24	632.31
MF268	3	-4.31	9	692.63
Edrophonium	4	-4.27	28	744.12
Acetylthiocholine	1	-4.17	40	880.28
Acetylcholine	1	-3.94	41	1280.00
Decamethonium	1	-3.50	7	2710.00
Decamethonium	3	-3.31	16	3740.00
Gallamine	1	-0.56	1	387660.00
Ambenonium	1	-0.41	1	504260.00

Appendix 17

Results from the docking of standard ligands with a structure derived from the crystal structure of mouse acetylcholinesterase - decidium complex (2.35 A°) (PDB ID: 1j07) (arranged according to highest number in cluster).

Ligand	Cluster rank	Lowest binding energy (kcal/mol)	No. in cluster	Inhibition constant, K_i (μM)
Huperzine B	1	-7.12	57	6.03
MPTP	3	-5.36	54	118.58
Acetylcholine	1	-3.94	41	1280.00
Acetylthiocholine	1	-4.17	40	880.28
Edrophonium	4	-4.27	28	744.12
MPTP	1	-5.72	28	63.66
(-)- Huprine x	2	-7.04	27	6.87
Tacrine	1	-6.43	24	19.47
D-Tubocurarine	14	-4.36	24	632.31
Huperzine A	1	-7.11	20	6.16
Decamethonium	3	-3.31	16	3740.00
Edrophonium	1	-4.69	16	366.58
(-)-Galanthamine	8	-5.84	14	52.76
(-)- Huprine x	1	-7.29	11	4.55
MF268	3	-4.31	9	692.63
Δ^9-THC	3	-6.84	8	9.63
Decamethonium	1	-3.50	7	2710.00
MF268	1	-5.44	5	103.74
Donepezil	2	-5.67	4	69.83
(-)-Galanthamine	1	-7.03	4	7.09
Δ^9-THC	1	-7.45	4	3.49
D-Tubocurarine	1	-6.50	4	17.12
Propidium	1	-5.94	3	44.55
Decidium	1	-4.63	2	401.84
Donepezil	1	-5.84	2	52.77
Ambenonium	1	-0.41	1	504260.00
BW284C51	1	-6.54	1	16.21
Gallamine	1	-0.56	1	387660.00

Appendix 18

Results from the docking of standard ligands with a structure derived from the crystal structure of mouse acetylcholinesterase – decidium complex (2.35 A°) (PDB ID: 1j07).

CR	LIGAND																			
	ACL (CAS)		ACLTHIO (CAS)		AMB (PAS)		BW284C51 (BTH)		DECA (BTH)		DECI (PAS)		DONE (BTH)		EDR (CAS)		GALLMN (PAS)		(-)-GLNTM (CAS)	
	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS
1	-3.94 (41)	O	-4.17 (40)	O	-0.41 (1)	O	-6.54 (1)	PAS	-3.50 (7)	O	-4.63 (2)	PAS	-5.84 (2)	O	-4.69 (16)	PAS	-0.56 (1)	O	-7.03 (4)	PAS
2	-3.67 (25)	PAS	-3.68 (10)	PAS	+0.11 (1)	PAS	-5.71 (1)	BTH	-3.35 (10)	O	-4.57 (1)	PAS	-5.67 (4)	PAS	-4.58 (19)	PAS	-0.44 (1)	O	-6.74 (8)	PAS
3	-3.46 (6)	PAS	-3.58 (19)	PAS	+0.43 (1)	PAS	-4.94 (1)	O	-3.31 (16)	PAS	-4.53 (1)	PAS	-5.48 (1)	BTH	-4.30 (2)	O	-0.24 (2)	O	-6.66 (3)	CAS
4	-3.17 (2)	O	-3.36 (3)	PAS	+0.48 (1)	O	-4.91 (2)	O	-3.30 (2)	BTH	-4.46 (1)	PAS	-5.43 (1)	PAS	-4.27 (28)	O	+0.07 (1)	O	-6.64 (7)	PAS
5	-3.03 (2)	O	-3.29 (1)	O	+0.53 (1)	PAS	-4.43 (1)	PAS	-3.23 (5)	O	-4.31 (1)	PAS	-5.42 (1)	PAS	-4.11 (7)	O	+0.26 (1)	O	-6.41 (6)	PAS
6	-2.92 (1)	CAS	-3.16 (3)	O	+0.54 (1)	O	-4.41 (1)	O	-3.18 (6)	O	-3.78 (1)	PAS	-5.14 (4)	PAS	-4.04 (1)	O	+0.27 (1)	O	-6.01 (2)	CAS
7	-2.92 (2)	PAS	-3.10 (1)	PAS	+0.59 (1)	O	-4.27 (1)	O	-3.16 (4)	PAS	-3.53 (1)	O	-5.06 (1)	PAS	-4.04 (10)	PAS	+0.28 (2)	O	-5.95 (1)	PAS
8	-2.78 (5)	O	-3.06 (3)	O	+0.67 (1)	O	-4.06 (1)	O	-2.89 (3)	O	-3.44 (1)	PAS	-5.02 (2)	O	-3.96 (3)	O	+0.30 (1)	PAS	-5.84 (14)	O
9	-2.62 (1)	O	-2.98 (7)	O	+0.70 (1)	PAS	-3.89 (2)	O	-2.85 (2)	PAS	-3.40 (1)	PAS	-4.87 (4)	O	-3.90 (1)	O	+0.30 (1)	O	-5.82 (1)	O
10	-2.57 (2)	O	-2.96 (1)	MDL	+0.70 (1)	PAS	-3.77 (1)	O	-2.84 (6)	O	-3.33 (1)	PAS	-4.81 (1)	PAS	-3.85 (1)	CAS	+0.37 (2)	O	-5.72 (1)	O
11-100		O		CAS/ O		PAS/ O		PAS/ O\		PAS/ O		PAS/ O		PAS/ O		PAS/ O/ CAS		PAS/ O		O
(*/100)		(1)		(5)		(9)		(1)		(2)		(20)		(1)		(3)		(14)		(5)
√/ X		X		X		X		X		X		√		X		X		X		X

CR = Cluster rank, PAS = Peripheral anionic site, CAS = Catalytic site, LBE = Lowest binding energy (kcal/mol), BS = Binding site, O = Bind to other site compared to PAS and CAS, MDL = Bind in between PAS and CAS, BTH = Bind at PAS and CAS, X = The binding location is incorrect according to literature, √ = The binding location is correct according to literature, (*/100) = Total conformation (the binding location is correct according to the literature) over 100 runs.

CR	LIGAND																	
	HUPA (CAS)		HUPB (CAS)		(-)-HUP X (CAS)		MF268 (PAS)		MPTP (CAS)		PROP (PAS)		TAC (CAS)		Δ^9-THC (PAS)		TUBO (PAS)	
	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS
1	-7.11 (20)	O	-7.12 (57)	O	-7.29 (11)	PAS	-5.44 (5)	PAS	-5.72 (28)	PAS	-5.94 (3)	PAS	-6.43 (24)	O	-7.45 (4)	PAS	-6.50 (4)	PAS
2	-6.66 (18)	O	-6.57 (10)	PAS	-7.04 (27)	PAS	-4.78 (3)	PAS	-5.53 (1)	PAS	-5.71 (1)	PAS	-6.37 (21)	PAS	-7.34 (4)	MDL	-6.00 (1)	PAS
3	-6.59 (20)	PAS	-6.37 (31)	PAS	-6.83 (9)	PAS	-4.31 (9)	O	-5.36 (54)	O	-5.16 (2)	PAS	-6.35 (21)	PAS	-6.84 (8)	PAS	-5.68 (1)	PAS
4	-6.44 (3)	O	-5.77 (1)	O	-6.70 (2)	PAS	-4.31 (1)	PAS	-5.21 (1)	CAS	-4.99 (3)	PAS	-6.31 (15)	O	-6.76 (2)	PAS	-5.31 (2)	PAS
5	-6.22 (2)	O	-5.68 (1)	CAS	-6.69 (6)	O	-4.30 (2)	PAS	-5.21 (1)	PAS	-4.76 (1)	O	-6.29 (3)	PAS	-6.61 (6)	PAS	-5.17 (2)	PAS
6	-6.04 (1)	O	-	-	-6.57 (20)	O	-4.13 (1)	PAS	-5.19 (7)	PAS	-4.72 (1)	PAS	-6.19 (3)	PAS	-6.48 (7)	PAS	-5.14 (2)	O
7	-5.96 (7)	PAS	-	-	-6.34 (9)	PAS	-4.11 (1)	PAS	-5.19 (3)	O	-4.64 (2)	O	-5.83 (2)	O	-6.27 (1)	PAS	-5.13 (6)	O
8	-5.79 (3)	CAS	-	-	-6.07 (1)	O	-4.11 (1)	PAS	-5.06 (1)	MDL	-4.40 (1)	PAS	-5.77 (3)	O	-6.07 (7)	O	-4.94 (1)	O
9	-5.58 (9)	PAS	-	-	-6.05 (1)	O	-4.05 (2)	PAS	-4.33 (1)	O	-4.33 (1)	PAS	-5.24 (4)	O	-5.69 (1)	PAS	-4.87 (2)	PAS
10	-5.28 (1)	O	-	-	-5.69 (1)	O	-4.03 (3)	PAS	-4.21 (1)	O	-4.30 (1)	PAS	-5.10 (1)	O	-5.63 (1)	PAS	-4.64 (3)	O
11-100		PAS/ O	-	-		O		PAS/ O/ MDL		O		PAS/ O		O		PAS/ O		PAS/O
(*/100)		(3)		(1)		(-)		(33)		(1)		(17)		(0)		(32)		(18)
√/ X		X		X		X		√		√		√		X		√		√

CR = Cluster rank, PAS = Peripheral anionic site, CAS = Catalytic site, LBE = Lowest binding energy (kcal/mol), BS = Binding site, O = Bind to other site compared to PAS and CAS, MDL = Bind in between PAS and CAS, BTH = Bind at PAS and CAS, X = The binding location is incorrect according to literature, √ = The binding location is correct according to literature, (*/100) = Total conformation (the binding location is correct according to the literature) over 100 runs (continued).

Appendix 19

Results from the docking of standard ligands with a structure derived from the crystal structure of mouse acetylcholinesterase in complex with propidium (2.25 A°) (PDB ID: 1n5r) (arranged according to lowest binding energy).

Ligand	Cluster rank	Lowest binding energy (kcal/mol)	No. in cluster	Inhibition constant, K_i (μM)
Δ^9-THC	1	-7.69	8	2.31
Donepezil	1	-7.46	2	3.38
(-)- Huprine x	1	-7.22	8	5.08
(-)-Galanthamine	1	-6.87	4	9.27
Huperzine A	1	-6.81	25	10.24
Huperzine B	1	-6.59	50	14.89
Tacrine	1	-6.43	40	19.39
D-Tubocurarine	1	-6.43	5	19.42
(-)- Huprine x	6	-6.41	33	20.03
Δ^9-THC	7	-6.17	10	30.03
Propidium	1	-5.90	5	47.51
(-)-Galanthamine	11	-5.79	14	57.25
Propidium	2	-5.70	6	66.54
MPTP	1	-5.65	30	72.26
Decidium	1	-5.54	1	86.81
MPTP	3	-5.44	47	102.34
Donepezil	8	-5.19	5	156.84
BW284C51	1	-5.02	2	208.70
MF268	1	-4.75	2	328.00
MF268	2	-4.70	7	359.77
Edrophonium	1	-4.59	14	430.13
D-Tubocurarine	14	-4.29	19	722.24
Edrophonium	3	-4.20	26	828.56
Acetylthiocholine	1	-4.01	33	1150.00
Acetylcholine	1	-3.89	31	1400.00
Decamethonium	1	-3.79	4	1670.00
Acetylcholine	2	-3.65	33	2130.00
Decamethonium	2	-3.58	18	2370.00
Ambenonium	1	-0.82	1	249110.00
Gallamine	1	-0.80	1	261310.00

Appendix 20

Results from the docking of standard ligands with a structure derived from the crystal structure of mouse acetylcholinesterase in complex with propidium (2.25 A°) (PDB ID: 1n5r) (arranged according to highest number in cluster).

Ligand	Cluster Rank	Lowest Binding Energy (kcal/mol)	No in Cluster	Inhibition Constant, K_i (µM)
Huperzine B	1	-6.59	50	14.89
MPTP	3	-5.44	47	102.34
Tacrine	1	-6.43	40	19.39
Acetylcholine	2	-3.65	33	2130.00
Acetylthiocholine	1	-4.01	33	1150.00
(-)- Huprine x	6	-6.41	33	20.03
Acetylcholine	1	-3.89	31	1400.00
MPTP	1	-5.65	30	72.26
Edrophonium	3	-4.20	26	828.56
Huperzine A	1	-6.81	25	10.24
D-Tubocuraine	14	-4.29	19	722.24
Decamethonium	2	-3.58	18	2370.00
Edrophonium	1	-4.59	14	430.13
(-)-Galanthamine	11	-5.79	14	57.25
Δ^9-THC	7	-6.17	10	30.03
(-)- Huprine x	1	-7.22	8	5.08
Δ^9-THC	1	-7.69	8	2.31
MF268	2	-4.70	7	359.77
Propidium	2	-5.7	6	66.54
Donepezil	8	-5.19	5	156.84
Propidium	1	-5.90	5	47.51
D-Tubocuraine	1	-6.43	5	19.42
Decamethonium	1	-3.79	4	1670.00
(-)-Galanthamine	1	-6.87	4	9.27
BW284C51	1	-5.02	2	208.70
Donepezil	1	-7.46	2	3.38
MF268	1	-4.75	2	328.00
Ambenonium	1	-0.82	1	249110.00
Decidium	1	-5.54	1	86.81
Gallamine	1	-0.80	1	261310.00

Appendix 21

Results from the docking of standard ligands with a structure derived from the crystal structure of mouse acetylcholinesterase in complex with propidium (2.25 A°) (PDB ID: 1n5r).

CR	LIGAND																			
	ACL (CAS)		ACLTHIO (CAS)		AMB (PAS)		BW284C51 (BTH)		DECA (BTH)		DECI (PAS)		DONE (BTH)		EDR (CAS)		GALLMN (PAS)		(-)-GLNTM (CAS)	
	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS
1	-3.89 (31)	O	-4.01 (33)	O	-0.82 (1)	PAS	-5.02 (2)	PAS	-3.79 (4)	BTH	-5.54 (1)	MDL	-7.46 (2)	BTH	-4.59 (14)	PAS	-0.80 (1)	O	-6.87 (4)	PAS
2	-3.65 (33)	PAS	-3.68 (11)	PAS	+0.19 (1)	O	-5.01 (1)	BTH	-3.58 (18)	O	-4.49 (2)	PAS	-6.20 (1)	PAS	-4.53 (22)	PAS	-0.54 (1)	O	-6.74 (11)	PAS
3	-3.38 (5)	PAS	-3.47 (21)	PAS	+0.19 (1)	O	-4.62 (2)	O	-3.54 (3)	MDL	-4.40 (1)	PAS	-6.00 (1)	BTH	-4.20 (26)	O	-0.45 (2)	PAS	-6.58 (6)	PAS
4	-3.26 (4)	O	-3.31 (2)	PAS	+0.26 (1)	O	-4.56 (1)	PAS	-3.31 (6)	PAS	-4.29 (1)	PAS	-5.76 (2)	PAS	-4.11 (8)	O	-0.21 (1)	O	-6.55 (2)	CAS
5	-3.12 (2)	O	-3.26 (7)	O	+0.49 (1)	O	-4.47 (1)	MDL	-3.29 (3)	PAS	-4.12 (1)	PAS	-5.68 (2)	O	-4.03 (6)	O	-0.17 (2)	PAS	-6.16 (4)	PAS
6	-3.05 (5)	O	-3.22 (4)	O	+0.52 (1)	O	-4.30 (1)	BTH	-3.28 (4)	O	-3.73 (1)	PAS	-5.37 (4)	PAS	-3.97 (4)	PAS	-0.15 (1)	O	-6.16 (1)	PAS
7	-2.99 (1)	O	-3.11 (6)	O	+0.56 (1)	O	-4.28 (1)	PAS	-3.24 (1)	BTH	-3.64 (1)	PAS	-5.28 (1)	PAS	-3.93 (1)	O	-0.04 (2)	O	-6.07 (2)	PAS
8	-2.82 (2)	O	-2.97 (8)	O	+0.66 (1)	O	-4.19 (1)	BTH	-3.16 (11)	O	-3.19 (1)	O	-5.19 (5)	PAS	-3.91 (3)	O	+0.03 (1)	O	-6.07 (2)	CAS
9	-2.78 (2)	PAS	-2.81 (1)	O	+0.66 (1)	O	-4.19 (1)	PAS	-3.13 (4)	O	-3.18 (1)	PAS	-5.09 (1)	PAS	-3.79 (1)	PAS	+0.04 (1)	PAS	-6.04 (2)	PAS
10	-2.77 (1)	CAS	-2.72 (2)	O	+0.68 (1)	O	-4.18 (1)	O	-3.07 (3)	BTH	-3.10 (1)	O	-4.98 (1)	O	-3.78 (4)	CAS	+0.07 (2)	O	-5.94 (3)	O
11-100		O		O		PAS/ O		PAS/ O/ BTH		PAS/ O BTH		PAS/ O		PAS/ O		PAS/ O/ CAS		PAS/ O		O
(*/100)		(1)		(-)		(4)		(4)		(9)		(17)		(3)		(6)		(19)		(4)
√/ X		X		X		√		X		√		√		√		X		X		X

CR = Cluster rank, PAS = Peripheral anionic site, CAS = Catalytic site, LBE = Lowest binding energy (kcal/mol), BS = Binding site, O = Bind to other site compared to PAS and CAS, MDL = Bind in between PAS and CAS, BTH = Bind at PAS and CAS, X = The binding location is incorrect according to literature, √ = The binding location is correct according to literature, (*/100) = Total conformation (the binding location is correct according to the literature) over 100 runs.

CR	LIGAND																	
	HUPA (CAS)		HUPB (CAS)		(-)-HUP X (CAS)		MF268 (PAS)		MPTP (CAS)		PROP (PAS)		TAC (CAS)		Δ^9-THC (PAS)		TUBO (PAS)	
	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS
1	-6.81 (25)	O	-6.59 (50)	O	-7.22 (8)	PAS	-4.75 (2)	PAS	-5.65 (30)	PAS	-5.90 (5)	PAS	-6.43 (40)	PAS	-7.69 (8)	PAS	-6.43 (5)	PAS
2	-6.70 (25)	PAS	-6.56 (8)	PAS	-6.92 (4)	PAS	-4.70 (7)	O	-5.62 (5)	PAS	-5.70 (6)	PAS	-6.37 (16)	PAS	-7.41 (3)	MDL	-6.40 (5)	PAS
3	-6.59 (10)	O	-6.31 (36)	PAS	-6.91 (13)	PAS	-4.41 (1)	MDL	-5.44 (47)	O	-5.70 (3)	PAS	-6.23 (7)	PAS	-6.93 (8)	MDL	-5.88 (3)	PAS
4	-6.33 (1)	O	-5.82 (3)	O	-6.85 (5)	PAS	-4.33 (5)	PAS	-5.25 (1)	O	-5.66 (1)	PAS	-6.17 (6)	O	-6.51 (6)	PAS	-5.43 (11)	PAS
5	-5.84 (1)	O	-5.76 (1)	O	-6.66 (4)	O	-4.30 (1)	PAS	-5.18 (7)	PAS	-4.98 (2)	PAS	-6.14 (6)	O	-6.51 (1)	PAS	-5.26 (3)	PAS
6	-5.81 (2)	CAS	-5.19 (1)	O	-6.41 (33)	O	-4.19 (3)	PAS	-5.12 (1)	MDL	-4.90 (2)	PAS	-5.99 (10)	O	-6.41 (4)	PAS	-5.22 (1)	PAS
7	-5.77 (7)	PAS	-4.78 (1)	O	-6.25 (4)	PAS	-4.18 (1)	PAS	-5.11 (1)	PAS	-4.80 (1)	PAS	-5.94 (4)	O	-6.17 (10)	O	-5.03 (5)	O
8	-5.72 (1)	PAS	-	-	-6.22 (2)	O	-4.05 (1)	PAS	-4.40 (2)	O	-4.59 (2)	O	-5.93 (1)	O	-6.15 (3)	PAS	-5.03 (6)	O
9	-5.41 (2)	O	-	-	-6.17 (1)	O	-3.92 (2)	PAS	-4.27 (1)	O	-4.34 (1)	PAS	-5.88 (1)	O	-6.13 (2)	PAS	-5.02 (5)	O
10	-5.39 (4)	PAS	-	-	-5.91 (1)	MDL	-3.91 (1)	PAS	-4.06 (5)	O	-4.33 (1)	PAS	-5.82 (1)	O	-5.49 (1)	PAS	-4.53 (1)	O
11-100		PAS/ O	-	-		PAS/ O		PAS/ O	-	-		PAS/ O		O		O		PAS/ O
(*/100) √/ X		(2) X		(-) X		(-) X		(29) √		(-) X		(28) √		**(-)** X		(25) √		(29) √

CR = Cluster rank, PAS = Peripheral anionic site, CAS = Catalytic site, LBE = Lowest binding energy (kcal/mol), BS = Binding site, O = Bind to other site compared to PAS and CAS, MDL = Bind in between PAS and CAS, BTH = Bind at PAS and CAS, X = The binding location is incorrect according to literature, √ = The binding location is correct according to literature, (*/100) = Total conformation (the binding location is correct according to the literature) over 100 runs (Continued).

Appendix 22

Results from the docking of standard ligands with a structure derived from the crystal structure of mouse acetylcholinesterase in complex with succinylcholine (2.05 A°) (PDB ID: 2ha2) (arranged according to lowest binding energy).

Ligand	Cluster rank	Lowest binding energy (kcal/mol)	No. in cluster	Inhibition constant, K_i (μM)
(-)- Huprine x	1	-7.62	16	2.59
(-)- Huprine x	3	-7.50	26	3.20
Δ^9-THC	1	-7.19	18	5.40
Huperzine A	1	-7.00	37	7.44
(-)-Galanthamine	1	-6.98	33	7.62
Huperzine B	1	-6.98	32	7.67
Donepezil	1	-6.88	5	9.04
BW284C51	1	-6.48	6	17.78
Tacrine	1	-6.40	11	20.21
Tacrine	4	-6.33	27	23.02
D-Tubocuraine	1	-6.21	7	28.22
Decidium	1	-6.13	2	32.03
Propidium	1	-5.95	5	43.20
MPTP	1	-5.64	48	73.44
MF268	1	-5.27	4	136.70
D-Tubocuraine	11	-4.71	16	354.12
Edrophonium	1	-4.61	32	419.22
MF268	8	-4.51	9	491.22
Decamethonium	1	-3.88	19	1420.00
Acetylthiocholine	1	-3.76	33	1760.00
Acetylthiocholine	2	-3.59	46	2320.00
Acetylcholine	2	-3.53	30	2580.00
Gallamine	1	-0.92	1	210000.00
Ambenonium	1	-0.17	1	754450.00

Appendix 23

Results from the docking of standard ligands with a structure derived from the crystal structure of mouse acetylcholinesterase in complex with succinylcholine (2.05 A°) (PDB ID: 2ha2) (arranged according to highest number in cluster).

Ligand	Cluster Rank	Lowest Binding Energy (kcal/mol)	No in Cluster	Inhibition Constant, K_i (μM)
MPTP	1	-5.64	48	73.44
Acetylthiocholine	2	-3.59	46	2320.00
Huperzine A	1	-7.00	37	7.44
Acetylthiocholine	1	-3.76	33	1760.00
(-)-Galanthamine	1	-6.98	33	7.62
Edrophonium	1	-4.61	32	419.22
Huperzine B	1	-6.98	32	7.67
Acetylcholine	2	-3.53	30	2580.00
Tacrine	4	-6.33	27	23.02
(-)- Huprine x	3	-7.50	26	3.20
Decamethonium	1	-3.88	19	1420.00
Δ^9-THC	1	-7.19	18	5.40
(-)- Huprine x	1	-7.62	16	2.59
D-Tubocurarine	11	-4.71	16	354.12
Tacrine	1	-6.40	11	20.21
MF268	8	-4.51	9	491.22
D-Tubocurarine	1	-6.21	7	28.22
BW284C51	1	-6.48	6	17.78
Donepezil	1	-6.88	5	9.04
Propidium	1	-5.95	5	43.20
MF268	1	-5.27	4	136.70
Decidium	1	-6.13	2	32.03
Ambenonium	1	-0.17	1	754450.00
Gallamine	1	-0.92	1	210000.00

Appendix 24

Results from the docking of standard ligands with a structure derived from the crystal structure of mouse acetylcholinesterase in complex with succinylcholine (2.05 A°) (PDB ID: 2ha2).

CR	LIGAND																			
	ACL (CAS)		ACLTHIO (CAS)		AMB (PAS)		BW284C51 (BTH)		DECA (BTH)		DECI (PAS)		DONE (BTH)		EDR (CAS)		GALLMN (PAS)		(-)-GLNTM (CAS)	
	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS
1	-3.65 (14)	O	-3.76 (33)	O	-0.17 (1)	O	-6.48 (6)	BTH	-3.88 (19)	BTH	-6.13 (2)	PAS	-6.88 (5)	BTH	-4.61 (32)	PAS	-0.92 (1)	O	-6.98 (33)	PAS
2	-3.53 (30)	PAS	-3.59 (46)	PAS	+0.20 (1)	O	-5.73 (3)	BTH	-3.83 (12)	O	-4.82 (1)	PAS	-6.80 (2)	BTH	-4.48 (3)	PAS	-0.57 (1)	O	-6.67 (3)	PAS
3	-3.44 (14)	O	-3.45 (5)	PAS	+0.21 (1)	O	-5.71 (2)	BTH	-3.74 (9)	BTH	-4.33 (1)	PAS	-6.69 (3)	BTH	-4.39 (8)	O	+0.00 (1)	PAS	-6.53 (1)	PAS
4	-3.37 (11)	PAS	-3.41 (2)	PAS	+0.25 (1)	O	-5.49 (5)	BTH	-3.67 (3)	O	-3.80 (1)	PAS	-6.45 (4)	BTH	-4.30 (4)	O	+0.19 (1)	O	-6.47 (3)	CAS
5	-3.22 (7)	O	-3.38 (5)	O	+0.31 (1)	O	-5.18 (4)	BTH	-3.45 (2)	BTH	-3.78 (1)	PAS	-6.36 (1)	O	-4.19 (21)	O	+0.28 (1)	PAS	-6.46 (1)	CAS
6	-3.22 (3)	O	-3.11 (2)	O	+0.52 (1)	O	-4.93 (1)	PAS	-3.44 (4)	BTH	-3.74 (1)	PAS	-6.23 (4)	BTH	-4.04 (17)	O	+0.38 (2)	O	-6.37 (1)	PAS
7	-3.04 (4)	PAS	-3.03 (1)	O	+0.55 (1)	O	-4.67 (1)	O	-3.36 (7)	O	-3.64 (1)	PAS	-6.20 (4)	PAS	-3.85 (3)	O	+0.53 (3)	O	-6.27 (6)	CAS
8	-2.94 (2)	O	-2.70 (1)	O	+0.56 (1)	O	-4.61 (1)	BTH	-3.24 (4)	PAS	-3.58 (1)	O	-6.18 (4)	PAS	-3.84 (1)	O	+0.53 (1)	O	-6.21 (2)	CAS
9	-2.83 (1)	CAS	-2.17 (1)	O	+0.59 (1)	O	-4.48 (1)	PAS	-3.13 (1)	BTH	-3.42 (1)	PAS	-6.08 (2)	BTH	-3.77 (3)	CAS	+0.54 (2)	O	-5.80 (1)	CAS
10	-2.66 (2)	O	-2.16 (1)	O	+0.65 (1)	O	-4.47 (1)	O	-2.86 (3)	O	-3.38 (1)	O	-5.94 (2)	PAS	-3.45 (3)	O	+0.76 (1)	O	-5.79 (6)	O
11-100		O		O		O		PAS/ O		PAS/ O		PAS/ O		PAS/ O/ BTH		O		PAS/ O		O
(*/100)		(1)		(-)		(O)		(21)		(35)		(16)		(23)		(3)		(13)		(13)
√/ X		X		X		X		√		√		√		√		X		X		X

CR = Cluster rank, PAS = Peripheral anionic site, CAS = Catalytic site, LBE = Lowest binding energy (kcal/mol), O = Bind to other site compared to PAS and CAS, MDL = Bind in between PAS and CAS, BTH = Bind at PAS and CAS, X = The binding location is incorrect according to literature, √ = The binding location is correct according to literature, (*/100) = Total conformation (the binding location is correct according to the literature) over 100 runs.

CR	LIGAND																	
	HUPA (CAS)		HUPB (CAS)		(-)-HUP X (CAS)		MF268 (PAS)		MPTP (CAS)		PROP (PAS)		TAC (CAS)		Δ^9-THC (PAS)		TUBO (PAS)	
	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS
1	-7.00 (37)	PAS	-6.98 (32)	PAS	-7.62 (16)	CAS	-5.27 (4)	PAS	-5.64 (48)	O	-5.95 (5)	PAS	-6.40 (11)	MDL	-7.19 (18)	PAS	-6.21 (7)	O
2	-6.41 (18)	O	-6.59 (11)	O	-7.53 (2)	PAS	-4.77 (6)	PAS	-5.48 (2)	PAS	-5.71 (4)	PAS	-6.37 (22)	MDL	-7.18 (6)	MDL	-6.14 (11)	PAS
3	-6.35 (14)	O	-6.52 (9)	CAS	-7.50 (26)	PAS	-4.74 (2)	MDL	-5.33 (9)	CAS	-5.38 (2)	PAS	-6.34 (19)	O	-6.87 (7)	PAS	-5.67 (3)	PAS
4	-6.01 (1)	PAS	-6.50 (29)	O	-7.08 (2)	PAS	-4.74 (3)	MDL	-5.30 (3)	CAS	-5.31 (2)	PAS	-6.33 (27)	PAS	-6.71 (4)	MDL	-5.52 (1)	O
5	-5.87 (3)	O	-6.03 (2)	O	-6.90 (1)	PAS	-4.70 (2)	PAS	-5.13 (1)	PAS	-4.81 (1)	PAS	-6.20 (2)	PAS	-6.21 (1)	PAS	-5.27 (2)	O
6	-5.82 (1)	CAS	-5.74 (2)	O	-6.54 (1)	CAS	-4.68 (4)	PAS	-5.09 (21)	PAS	-4.75 (1)	PAS	-6.13 (2)	O	-5.89 (1)	PAS	-5.03 (1)	PAS
7	-5.80 (4)	O	-5.18 (8)	O	-6.38 (17)	O	-4.54 (1)	PAS	-4.40 (2)	O	-4.64 (1)	PAS	-5.87 (3)	O	-5.67 (3)	O	-4.93 (1)	PAS
8	-5.63 (1)	O	-4.89 (1)	O	-5.69 (1)	O	-4.51 (9)	MDL	-4.20 (2)	O	-4.37 (1)	PAS	-5.51 (2)	CAS	-5.03 (4)	O	-4.85 (3)	O
9	-5.22 (1)	CAS	-4.85 (1)	O	-5.67 (1)	O	-4.37 (1)	MDL	-4.15 (10)	O	-4.23 (1)	PAS	-5.24 (4)	O	-4.96 (2)	O	-4.84 (1)	O
10	-4.96 (3)	O	-4.81 (3)	O	-5.48 (1)	O	-4.32 (4)	PAS	-3.86 (1)	O	-4.22 (1)	O	-5.15 (1)	O	-4.88 (2)	O	-4.75 (3)	O
11-100		O		O		O		PAS/ O/ MDL		O		PAS/ O		O		O		O
(*/100) √/ X		(2) X		(9) X		(17) √		(29) √		(12) X		(19) √		(2) X		(27) √		(16) X

CR = Cluster rank, PAS = Peripheral anionic site, CAS = Catalytic site, LBE = Lowest binding energy (kcal/mol), O = Bind to other site compared to PAS and CAS, MDL = Bind in between PAS and CAS, BTH = Bind at PAS and CAS, X = The binding location is incorrect according to literature, √ = The binding location is correct according to literature, (*/100) = Total conformation (the binding location is correct according to the literature) over 100 runs (continued).

Appendix 25

Results from the docking of standard ligands with a structure derived from the crystal structure of mouse acetylcholinesterase in complex with gallamine (2.20 A°) (PDB ID: 1n5m) (arranged according to lowest binding energy).

Ligand	Cluster rank	Lowest binding energy (kcal/mol)	No. in cluster	Inhibition constant, K_i (μM)
(-)- Huprine x	1	-7.63	13	2.55
Δ^9-THC	1	-7.39	11	3.81
Huperzine A	1	-7.00	37	7.36
Huperzine B	1	-6.99	72	7.54
Donepezil	1	-6.81	1	10.26
(-)-Galanthamine	1	-6.80	5	10.39
(-)-Galanthamine	2	-6.68	15	12.76
Tacrine	1	-6.56	21	15.55
Tacrine	4	-6.31	30	23.83
(-)- Huprine x	8	-6.20	19	28.34
Propidium	1	-5.83	7	53.16
BW284C51	1	-5.71	1	65.52
MPTP	1	-5.64	21	73.48
D-Tubocurarine	1	-5.52	1	90.42
Donepezil	5	-5.39	3	111.03
MPTP	4	-5.20	62	154.43
Decidium	1	-5.00	1	215.58
MF268	1	-4.84	3	284.58
Edrophonium	1	-4.65	23	392.12
Edrophonium	3	-4.38	33	617.34
D-Tubocurarine	21	-4.25	25	761.06
BW284C51	6	-4.05	3	1080.00
Acetylthiocholine	1	-3.91	29	1370.00
Acetylcholine	2	-3.67	40	2040.00
Decamethonium	1	-3.63	7	2170.00
Acetylthiocholine	4	-3.56	35	2440.00
Decidium	7	-3.55	2	2480.00
Decamethonium	4	-3.30	16	3820.00
Gallamine	1	-1.17	1	138640.00
Ambenonium	1	-0.73	1	293990.00

Appendix 26

Results from the docking of standard ligands with a structure derived from the crystal structure of mouse acetylcholinesterase in complex with gallamine (2.20 A°) (PDB ID: 1n5m) (arranged according to highest number in cluster).

Ligand	Cluster rank	Lowest binding energy (kcal/mol)	No. in cluster	Inhibition constant, K_i (μM)
Huperzine B	1	-6.99	72	7.54
MPTP	4	-5.2	62	154.43
Acetylcholine	2	-3.67	40	2040.00
Huperzine A	1	-7.00	37	7.36
Acetylthiocholine	4	-3.56	35	2440.00
Edrophonium	3	-4.38	33	617.34
Tacrine	4	-6.31	30	23.83
Acetylthiocholine	1	-3.91	29	1370.00
D-Tubocurarine	21	-4.25	25	761.06
Edrophonium	1	-4.65	23	392.12
MPTP	1	-5.64	21	73.48
Tacrine	1	-6.56	21	15.55
(-)- Huprine x	8	-6.2	19	28.34
Decamethonium	4	-3.30	16	3820.00
(-)-Galanthamine	2	-6.68	15	12.76
(-)- Huprine x	1	-7.63	13	2.55
Δ^9-THC	1	-7.39	11	3.81
Decamethonium	1	-3.63	7	2170.00
Propidium	1	-5.83	7	53.16
(-)-Galanthamine	1	-6.8	5	10.39
BW284C51	6	-4.05	3	1080.00
Donepezil	5	-5.39	3	111.03
MF268	1	-4.84	3	284.58
Decidium	7	-3.55	2	2480.00
Ambenonium	1	-0.73	1	293990.00
BW284C51	1	-5.71	1	65.52
Decidium	1	-5.00	1	215.58
Donepezil	1	-6.81	1	10.26
Gallamine	1	-1.17	1	138640.00
D-Tubocurarine	1	-5.52	1	90.42

Appendix 27

Results from the docking of standard ligands with a structure derived from the crystal structure of mouse acetylcholinesterase in complex with gallamine (2.20 A°) (PDB ID: 1n5m).

CR	LIGAND																			
	ACL (CAS)		ACLTHIO (CAS)		AMB (PAS)		BW284C51 (BTH)		DECA (BTH)		DECI (PAS)		DONE (BTH)		EDR (CAS)		GALLMN (PAS)		(-)-GLNTM (CAS)	
	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS
1	-3.68 (29)	O	-3.91 (29)	O	-0.73 (1)	PAS	-5.71 (1)	BTH	-3.63 (7)	PAS	-5.00 (1)	PAS	-6.81 (1)	PAS	-4.65 (23)	PAS	-1.17 (1)	O	-6.80 (5)	PAS
2	-3.67 (40)	PAS	-3.65 (13)	PAS	-0.39 (1)	O	-5.44 (1)	PAS	-3.52 (1)	BTH	-4.56 (1)	PAS	-6.79 (2)	BTH	-4.64 (24)	PAS	-1.04 (1)	O	-6.68 (15)	PAS
3	-3.40 (15)	PAS	-3.60 (4)	PAS	+0.05 (1)	O	-4.54 (1)	PAS	-3.38 (4)	PAS	-4.54 (1)	PAS	-6.21 (1)	PAS	-4.38 (33)	O	-0.60 (1)	PAS	-6.54 (10)	PAS
4	-2.93 (1)	PAS	-3.56 (35)	PAS	+0.11 (1)	O	-4.41 (1)	PAS	-3.30 (16)	O	-4.50 (1)	PAS	-5.65 (1)	PAS	-4.11 (5)	PAS	-0.54 (1)	PAS	-6.53 (7)	CAS
5	-2.90 (3)	PAS	-3.47 (4)	PAS	+0.22 (1)	O	-4.35 (1)	PAS	-3.28 (6)	O	-4.36 (1)	PAS	-5.39 (3)	PAS	-3.82 (2)	O	-0.42 (1)	O	-6.14 (1)	CAS
6	-2.81 (1)	O	-3.37 (1)	O	+0.29 (1)	O	-4.05 (3)	O	-3.23 (16)	O	-3.85 (1)	PAS	-5.23 (2)	PAS	-3.81 (1)	PAS	-0.33 (1)	PAS	-6.09 (1)	PAS
7	-2.66 (1)	O	-2.98 (5)	O	+0.37 (1)	PAS	-3.91 (1)	PAS	-3.16 (4)	O	-3.55 (2)	PAS	-4.95 (1)	PAS	-3.66 (1)	MDL	-0.23 (1)	O	-5.55 (9)	O
8	-2.66 (1)	O	-2.48 (1)	O	+0.42 (1)	O	-3.87 (1)	O	-3.11 (1)	BTH	-3.43 (1)	O	-4.81 (1)	PAS	-3.45 (1)	CAS	-0.04 (1)	O	-5.48 (7)	O
9	-2.65 (2)	O	-2.45 (1)	O	+0.54 (1)	O	-3.81 (2)	O	-3.03 (4)	PAS	-3.03 (1)	PAS	-4.79 (1)	PAS	-3.25 (2)	O	+0.13 (1)	O	-5.39 (2)	O
10	-2.59 (1)	O	-2.43 (1)	O	+0.57 (1)	O	-3.63 (1)	O	-2.84 (7)	O	-3.03 (1)	O	-4.70 (1)	PAS	-3.24 (2)	O	+0.18 (1)	O	-5.28 (1)	O
11-100		O		O		PAS/ O		PAS/ O		PAS/ O		PAS/ O		PAS/ O		O		PAS/ O		O
(*/100) √/ X		(0) X		(-) X		(4) √		(1) √		(2) X		(17) √		(2) X		(1) X		(11) X		(8) X

CR = Cluster rank, PAS = Peripheral anionic site, CAS = Catalytic site, LBE = Lowest binding energy (kcal/mol), BS = Binding site, O = Bind to other site compared to PAS and CAS, MDL = Bind in between PAS and CAS, BTH = Bind at PAS and CAS, X = The binding location is incorrect according to literature, √ = The binding location is correct according to literature, (*/100) = Total conformation (the binding location is correct according to the literature) over 100 runs.

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CR	LIGAND																	
	HUPA (CAS)		HUPB (CAS)		(-)-HUP X (CAS)		MF268 (PAS)		MPTP (CAS)		PROP (PAS)		TAC (CAS)		Δ^9-THC (PAS)		TUBO (PAS)	
	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS	LBE	BS
1	-7.00 (37)	O	-6.99 (72)	O	-7.63 (13)	PAS	-4.84 (3)	PAS	-5.64 (21)	PAS	-5.83 (7)	PAS	-6.56 (21)	O	-7.39 (11)	PAS	-5.52 (1)	O
2	-6.83 (14)	PAS	-6.88 (21)	PAS	-7.08 (7)	PAS	-4.80 (3)	PAS	-5.37 (8)	PAS	-5.36 (5)	PAS	-6.46 (10)	O	-7.02 (4)	MDL	-5.15 (2)	PAS
3	-6.57 (5)	O	-6.02 (5)	PAS	-7.07 (4)	CAS	-4.77 (2)	PAS	-5.30 (2)	CAS	-5.14 (2)	PAS	-6.33 (23)	PAS	-6.91 (4)	MDL	-5.12 (4)	O
4	-6.28 (10)	PAS	-5.72 (1)	CAS	-6.99 (10)	PAS	-4.68 (1)	PAS	-5.20 (62)	O	-4.96 (1)	PAS	-6.31 (30)	PAS	-6.47 (3)	PAS	-5.01 (3)	PAS
5	-6.09 (5)	PAS	-5.71 (1)	O	-6.70 (6)	PAS	-4.32 (3)	PAS	-4.76 (1)	CAS	-4.85 (1)	PAS	-6.26 (9)	PAS	-6.45 (1)	PAS	-4.83 (2)	O
6	-5.79 (8)	CAS	-	-	-6.37 (9)	O	-4.22 (2)	PAS	-4.14 (6)	O	-4.73 (1)	O	-6.12 (1)	PAS	-6.44 (5)	PAS	-4.76 (1)	O
7	-5.53 (4)	O	-	-	-6.24 (13)	O	-3.97 (2)	O	-	-	-4.65 (2)	PAS	-5.35 (2)	O	-6.19 (3)	PAS	-4.69 (1)	PAS
8	-5.38 (1)	CAS	-	-	-6.20 (19)	O	-3.92 (1)	PAS	-	-	-4.30 (2)	O	-5.09 (2)	O	-5.74 (1)	PAS	-4.67 (2)	PAS
9	-4.87 (3)	O	-	-	-6.16 (2)	PAS	-3.91 (2)	PAS	-	-	-4.28 (1)	PAS	-4.85 (1)	O	-5.59 (8)	O	-4.65 (3)	PAS
10	-4.80 (1)	O	-	-	-5.96 (2)	O	-3.87 (2)	MDL	-	-	-4.13 (1)	PAS	-4.55 (1)	O	-5.36 (2)	PAS	-4.63 (1)	O
11-100		O	-	-		O		PAS/ O/ MDL	-	-		PAS/ O	-	-		PAS/ O		O
(*/100) √/ X		(9) X		(1) X		(4) X		(31) √		(3) X		(25) √		(-) X		(29) √		(11) X

CR = Cluster rank, PAS = Peripheral anionic site, CAS = Catalytic site, LBE = Lowest binding energy (kcal/mol), BS = Binding site, O = Bind to other site compared to PAS and CAS, MDL = Bind in between PAS and CAS, BTH = Bind at PAS and CAS, X = The binding location is incorrect according to literature, √ = The binding location is correct according to literature, (*/100) = Total conformation (the binding location is correct according to the literature) over 100 runs (continued).

Appendix 28

Results from the docking of PAS binding ligands (Ligplot analysis).

CS	No. in cluster	BS	LBE	Residues involved in hydrogen bonding	Length (Å)	No. of residues involved in hydrophobic interaction	CS	No. in cluster	BS	LBE	Residues involved in hydrogen bonding	Length (Å)	No. of residues involved in hydrophobic interaction
				AMBENONIUM							**DECIDIUM**		
a	1	PAS	-2.70	O3-TYR 72:OH	2.76	5	a	1	PAS	-5.39	N34-SER293:OG	2.72	7
b	1	O	-0.78	N7-GLU73:OE2 N8-GLU73:OE2	2.74 2.70	4	b	1	PAS	-5.49	N35-PHE284:O	2.65	8
c	1	O	-0.88	-	-	8	c	1	PAS	-5.86	N34-SER286:O N34-ARG289:O N35-ASP72:OD2	2.94 2.85 2.76	7
d	1	PAS	-2.19	N7-ASP285:OD1 N8-ASP285:OD2	2.61 2.81	5	d	2	PAS	-6.44	N35-TYR330:OH N35-ASP72:OD2 N34-SER286:O	2.81 2.62 2.93	6
e	1	PAS	-1.35	N7-ASP285:OD1	2.71	4	e	2	PAS	-6.29	N34-SER286:O N34-ARG289:O N35-ASP72:OD2 N35-TYR330:OH	2.93 2.85 2.67 2.76	8
f	1	O	-0.78	N7-ASP285:OD2 N8-ASP285:OD2	3.06	3	f	1	PAS	-5.81	N34-ASP285:OD1 N34-SER286:O	3.13 2.83	11
g	1	O	-0.41	-	-	7	g	2	PAS	-4.63	N34-SER293:O	2.98	6
h	1	PAS	-0.82	N8-GLY342:O	2.72	3	h	1	MDL	-5.54	-	-	10
i	1	PAS	-0.73	N7-GLU292:OE2 N8-GLU292:OE2	3.07 2.79	4	i	1	PAS	-5.00	N35-TYR72:OH N34-SER293:OG	2.69 2.58	8
j	1	O	-0.17	N7-GLY305:O N8-GLY 305:O	3.11 2.85	5	j	2	PAS	-6.13	N34-SER293:O N34-SER293:OG N35-TYR72:OH	2.95 2.92 3.01	11

CS = Crystal structure, BS = Binding site, LBE = Lowest binding energy (kcal/mol), PAS = Peripheral anionic site, CAS = Catalytic site, O = Bind to other site compared to PAS and CAS, MDL = Bind in between PAS and CAS.

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CS	No. in cluster	BS	LBE	Residues involved in hydrogen bonding	Length (Å)	No. of residues involved in hydrophobic interaction	CS	No. in cluster	BS	LBE	Residues involved in hydrogen bonding	Length (Å)	No. of residues involved in hydrophobic interaction
				GALLAMIN							**MF 268**		
a	1	PAS	+0.38	-	-	7	a	6	PAS	-4.98	N48-TYR 124:OH O16-PHE295:N O16-ARG296:N	2.68 2.98 3.18	8
b	2	PAS	-0.67	-	-	7	b	20	MDL	-5.73	O16-PHE288:N O16-ARG289:N N14-SER286:O N48-ASP72:OD2	2.95 2.82 2.94 2.83	6
c	2	PAS	-1.10	-	-	9	c	9	PAS	-5.14	O1-PHE288:N	2.81	8
d	1	PAS	-0.38	-	-	8	d	10	PAS	-4.60	N14-TYR70:OH	3.01	7
e	2	PAS	-1.14	-	-	9	e	10	PAS	-5.49	O16-PHE288:N O16-ARG289:N N14-SER286:O N48-TYR70:OH	2.84 3.15 2.97 3.09	6
f	3	PAS	-1.42	-	-	7	f	13	PAS	-4.66	O1-PHE288:N N14-TYR70:OH	2.81 2.94	6
g	1	O	-0.56	-	-	8	g	5	PAS	-5.44	N48-TYR124:OH N14-SER293:O O16-PHE295:N O16-ARG296:N	2.56 2.86 2.91 3.05	6
h	1	O	-0.80	-	-	10	h	2	PAS	-4.75	-	-	8
i	1	O	-1.17	-	-	9	i	3	PAS	-4.84	N48-TYR124:OH	2.90	7
j	1	O	-0.92	-	-	9	j	4	PAS	-5.27	N48-TYR124:OH N14-ARG296:O O16-PHE295:N	2.75 2.83 2.91	7

CS = Crystal structure, BS = Binding site, LBE = Lowest binding energy (kcal/mol), PAS = Peripheral anionic site, CAS = Catalytic site, O = Bind to other site compared to PAS and CAS, MDL = Bind in between PAS and CAS (Continued).

CS	No. in cluster	BS	LBE	Residues involved in hydrogen bonding	Length (Å)	No. of residues involved in hydrophobic interaction	CS	No. in cluster	BS	LBE	Residues involved in hydrogen bonding	Length (Å)	No. of residues involved in hydrophobic interaction
				PROPIDIUM							**Δ^9-THC**		
a	3	PAS	-5.48	N21-TYR124:OH	2.79	7	a	13	PAS	-6.88	O1-PHE295:N	3.06	9
b	27	PAS	-6.02	N20-TYR70:OH N21-PHE284:O	2.52 2.72	8	b	5	MDL	-7.31	O2-PHE330:OC	2.63	10
c	20	PAS	-5.96	N20-TYR 70:OH N21-PHE284:O	2.60 2.78	11	c	3	PAS	-6.45	-	-	9
d	13	PAS	-5.68	N21-PHE284:O N20-TYR70:OH	2.71 2.78	8	d	4	PAS	-6.36	-	-	11
e	16	PAS	-6.14	N20-TYR70:OH N21-PHE284:O	2.52 2.83	10	e	9	PAS	-6.59	-	-	8
f	19	PAS	-5.65	N20-TYR70:OH N21-PHE284:O	2.77 2.71	7	f	4	PAS	-6.62	-	-	9
g	3	PAS	-5.94	N20-TYR72:OH N21-GLN291:O	2.93 2.66	5	g	4	PAS	-7.45	O2-PHE295:N	3.07	9
h	5	PAS	-5.90	N21-GLN291:O	2.58	8	h	8	PAS	-7.69	O2-PHE295:N	3.04	12
i	7	PAS	-5.83	N20-TYR72:OH N21-GLN291:O	2.79 2.65	9	i	11	PAS	-7.39	O2-PHE295:N	3.11	10
j	5	PAS	-5.95	N21-SER293:O N20-ASP74:OD2	2.84 3.15	4	j	18	PAS	-7.19	O2-PHE295:N	3.14	12

CS = Crystal structure, BS = Binding site, LBE = Lowest binding energy (kcal/mol), PAS = Peripheral anionic site, CAS = Catalytic site, MDL = Bind in between PAS and CAS (Continued).

CS	No. in cluster	BS	LBE	Residues involved in hydrogen bonding	Length (Å)	No. of residues involved in hydrophobic interaction
				D-TUBOCURARINE		
a	3	PAS	-6.01	O2-SER293:OG O2-ARG296:O O4-GLU292:N N85-HIS 287:O	2.97 3.22 3.24 3.02	6
b	25	PAS	-6.52	O4-ASP285:OD1 O2-SER286:O	2.66 2.75	9
c	22	PAS	-6.58	O4-ASP285:OD1 O2-SER286:0	2.52 2.75	9
d	33	PAS	-6.51	O4-ASP285:OD1 N85-TRP279:O O2-SER286:O O2-PHE288:N	2.62 3.25 2.86 3.16	7
e	19	PAS	-6.70	N85-TRP279:O O2-SER286:O O2-PHE288:N O4-ASP285:OD1	3.31 2.83 3.24 2.69	7
f	18	PAS	-6.58	O4-ASP285:OD1 O2-SER286:O	2.61 2.75	9
g	4	PAS	-6.50	N85-LEU289:O O2-SER293:O	3.31 2.85	6
h	5	PAS	-6.43	O2-ASP74:OD1 O2-TYR72:OH	2.70 3.11	5
i	1	O	-5.52	O4-GLN181:OE1 O2-PRO50:O O1-ARG45:NH2	2.96 3.13 2.73	6
j	7	O	-6.21	O4-GLN181:OE1 O2-PRO50:O	2.90 2.58	5

CS = Crystal structure, BS = Binding site, LBE = Lowest binding energy (kcal/mol), PAS = Peripheral anionic site, CAS = Catalytic site, O = Bind to other site compared to PAS and CAS (Continued).

Appendix 29

Results from the docking of CAS binding ligands (Ligplot analysis).

CS	No. in cluster	BS	LBE	Residues involved in hydrogen bonding	Length (Å)	No. of residues involved in hydrophobic interaction	CS	No. in cluster	BS	LBE	Residues involved in hydrogen bonding	Length (Å)	No. of residues involved in hydrophobic interaction
				ACETYLCHOLINE							**ACETYLTHIOCHOLINE**		
a	36	PAS	-3.76	O3-PHE295:N O5-ARG296:N	2.90 2.92	6	a	33	PAS	-3.77	O1-ARG 296:N O1-PHE295:N	2.81 2.82	6
b	55	PAS	-3.39	O5-ARG289:N	2.95	5	b	23	MDL	-3.54	O1-SER200:OG O1-HIS440:NE2	2.70 2.85	7
c	45	PAS	-3.42	O5-ARG289:N	2.95	5	c	32	PAS	-3.52	O1-PHE288:N	2.83	6
d	39	PAS	-3.25	O5-ARG289:N	3.01	5	d	39	PAS	-3.42	O1-PHE288:N	2.84	7
e	57	PAS	-3.41	O5-ARG289:N	2.98	5	e	50	PAS	-3.52	O1-PHE288:N	2.93	7
f	45	PAS	-3.25	O5-ARG289:N	2.97	5	f	60	PAS	-3.47	O1-PHE288:N	2.83	9
g	41	O	-3.94	O5-THR438:N	2.96	7	g	40	O	-4.17	O1-THR438:N	2.84	8
h	31	O	-3.89	O5-THR438:N	2.99	7	h	33	O	-4.01	O1-THR438:N	2.81	8
i	29	O	-3.68	O5-THR438:N	2.95	7	i	29	O	-3.91	O1-THR438:N	3.09	6
j	14	O	-3.65	O5-THR438:N	3.04	6	j	33	O	-3.76	O1-THR438:N	2.80	7

CS = Crystal structure, BS = Binding site, LBE = Lowest binding energy (kcal/mol), PAS = Peripheral anionic site, CAS = Catalytic site, O = Bind to other site compared to PAS and CAS, MDL = Bind in between PAS and CAS.

CS	No. in cluster	BS	LBE	Residues involved in hydrogen bonding	Length (Å)	No. of residues involved in hydrophobic interaction	CS	No. in cluster	BS	LBE	Residues involved in hydrogen bonding	Length (Å)	No. of residues involved in hydrophobic interaction
EDROPHONIUM							**(-)-GALANTAMINE**						
a	19	PAS	-4.68	O1-ARG296:N O1-PHE295:N	2.78 2.86	6	a	33	PAS	-7.09	O19-TYR337:OH	2.70	9
b	28	PAS	-4.52	O1-ARG289:N O1-ARG289:O	2.81 2.69	6	b	24	CAS	-7.03	N11-ASP72:OD2	2.62	6
c	69	PAS	-4.49	O1-ARG289:N O1-ARG289:O	2.91 2.73	7	c	8	PAS	-6.95	O19-TYR121:OH O18-TYR121:OH N11-TYR70:OH	3.14 2.73 2.92	5
d	59	PAS	-4.28	O1-ARG289:N O1-ARG289:O	2.90 2.66	7	d	37	PAS	-6.74	N11-TYR121:OH	2.71	7
e	72	PAS	-4.49	O1-ARG289:N O1-ARG289:O	2.86 2.69	7	e	32	PAS	-7.01	N11-TYR70:OH	3.03	6
f	65	PAS	-4.36	O1-ARG289:N	3.08	6	f	31	PAS	-6.95	O18-ARG289:N N11-TYR121:OH	3.10 2.76	7
g	16	PAS	-4.69	O1-ARG296:N	2.81	5	g	4	PAS	-7.03	O18-ARG296:N N11-TYR124:OH O19-TYR341:O	3.13 2.69 3.02	6
h	14	PAS	-4.59	O1-ARG296:N	2.75	4	h	4	PAS	-6.87	O18-ARG296:N O18-PHE295:N O19-TYR341:O N11-TYR124:OH	3.20 2.99 2.99 2.67	5
i	23	PAS	-4.65	O1-ARG296:O O1-ARG296:N	2.69 2.65	6	i	5	PAS	-6.80	O18-ARG296:N N11-TYR124:OH	3.02 2.85	7
j	32	PAS	-4.61	O1-ARG296:N O1-ARG296:O	2.97 2.78		j	33	PAS	-6.98	O19-ASP74:OD2 O19-TYR124:OH N1-SER293:O	3.04 3.07 2.88	7

CS = Crystal structure, BS = Binding site, LBE = Lowest binding energy (kcal/mol), PAS = Peripheral anionic site, CAS = Catalytic site (continued).

CS	No. in cluster	BS	LBE	Residues involved in hydrogen bonding	Length (Å)	No. of residues involved in hydrophobic interaction	CS	No. in cluster	BS	LBE	Residues involved in hydrogen bonding	Length (Å)	No. of residues involved in hydrophobic interaction
HUPERZINE A							**HUPERZINE B**						
a	16	CAS	-7.13	N13-SER203:OE1	2.72	9	a	18	CAS	-7.16	N2-GLU202:OE1	2.70	9
b	35	CAS	-6.58	N5-TYR121:OH N13-TYR334:OH N13-ASP72:OD2	2.65 3.20 2.54	3	b	72	CAS	-6.85	O1-HIS440:NE2 O1-SER200:OG N2-TYR121:OH	2.90 2.87 2.70	3
c	4	CAS	-6.07	N13-GLU199:OE1	2.51	7	c	16	CAS	-6.54	N2-GLU199:OE1	2.63	7
d	15	CAS	-6.96	N13-TYR330:OH N5-TYR130:OH O15-TYR130:OH	3.08 2.94 2.83	5	d	10	CAS	-6.48			
e	14	CAS	-6.59	N5-TYR130:OH N13-TYR330:OH	3.32 2.62	4	e	4	CAS	-6.53	N2-GLU199:OE1	2.65	6
f	25	CAS	-6.89	N13-TYR330:OH O15-TYR130:OH N5-TYR130:OH	2.91 2.91 3.10	5	f	16	CAS	-6.56	N2-GLU199:OE1	2.63	7
g	20	O	-7.11	O15-THR438:N N13-GLU81:OE2 N13-MET85:SD	3.02 2.58 3.30	6	g	57	O	-7.12	N38-GLU452:OE2 N2-GLU81:OE2	2.81 2.65	7
h	25	O	-6.81	O15-THR438:N N13-THR436:OG1 N13-GLU452:OE1 N13-TYR465:OH	2.98 2.68 2.62 2.57	4	h	50	O	-6.59	N2-GLU81:OE2	2.71	8
i	37	O	-7.00	N13-GLU452:OE2 N13-GLU452:OE1	2.77 2.64	7	i	72	O	-6.99	N2-THR436:OG1 N2-GLU452:OE1	2.76 2.94	6
j	37	PAS	-7.00	N13-SER293:O N13-TYR341:O N5-ARG296:O	2.86 2.48 3.03	5	j	3	PAS	-6.98	N38-TYR124:OH N2-TYR341:O	2.73 2.71	4

CS = Crystal structure, BS = Binding site, LBE = Lowest binding energy (kcal/mol), PAS = Peripheral anionic site, CAS = Catalytic site , O = Bind to other site compared to PAS and CAS (continued).

CS	No. in cluster	BS	LBE	Residues involved in hydrogen bonding	Length (Å)	No. of residues involved in hydrophobic interaction	CS	No. in cluster	BS	LBE	Residues involved in hydrogen bonding	Length (Å)	No. of residues involved in hydrophobic interaction
				(-)-HUPRINE X							**MPTP**		
a	7	PAS	-7.27	N15-TYR341:O N42-ARG296:O	2.52 2.70	6	a	33	PAS	-5.58	N13-TYR72:OH	2.93	5
b	56	CAS	-7.54	N15-ASP72:OD2	2.41	5	b	88	CAS	-6.07	N13-ASP72:OD2	2.70	5
c	22	PAS	-6.70	N15-ARG289:O	2.63	7	c	10	PAS	-5.48	N13-SER286:O	2.79	9
d	18	PAS	-6.56	N15-ARG289:O	2.51	7	d	27	PAS	-5.44	N13-SER286:O	2.78	9
e	21	PAS	-6.72	N15-ARG289:O	2.56	8	e	72	PAS	-5.73	N13-TYR70:OH	2.85	6
f	16	PAS	-6.54	N15-ARG289:O	2.68	7	f	27	CAS	-5.48	N13-ASN85:OD1	3.06	10
g	11	PAS	-7.29	N15-SER293:O N15-TYR341:O	3.06 2.54	6	g	28	PAS	-5.72	N13-SER293:O	2.91	6
h	8	PAS	-7.22	N42-ARG296:O N15-TYR341:O	3.12 2.36	6	h	30	PAS	-5.65	N13-SER 293:O	2.99	5
i	13	PAS	-7.63	N42-ARG296:O N15-TYR341:O	3.16 2.38	6	i	21	PAS	-5.64	N13-SER293:O	2.90	5
j	16	CAS	-7.62	N15-ASP74:OD2	2.42	6	j	48	O	-5.64	N13-ASP131:OD2	3.10	4

CS = Crystal structure, BS = Binding site, LBE = Lowest binding energy (kcal/mol), PAS = Peripheral anionic site, CAS = Catalytic site , O = Bind to other site compared to PAS and CAS (continued).

CS	No. in cluster	BS	BE	Residues involved in hydrogen bonding	Length (Å)	No. of residues involved in hydrophobic interaction
				TACRINE		
a	20	CAS	-6.40	N1-ASP74:OD2 N1-THR83:OG1	2.84 2.93	7
b	28	PAS	-6.51	N1-TYR121:OH	2.57	6
c	16	CAS	-6.26	N1-GLN69:OE1 N1-TRP84:O	2.58 2.60	7
d	20	CAS	-6.26	N1-GLN69:OE1 N1-TRP84:O	2.60 2.60	7
e	22	CAS	-6.27	N1-TRP84:O NI-GLN69:OE1	2.60 2.50	5
f	21	CAS	-6.28	N1-TRP84:O N1-GLN69:OE1	2.63 2.53	9
g	24	O	-6.43	N1-GLU452:OE1 N1-THR436:OG1	2.64 2.56	7
h	40	PAS	-6.43	N1-ASP74:OD1	2.40	5
i	21	O	-6.56	N1-THR436:OG1 N1-GLU452:OE1	2.52 2.59	8
j	11	MDL	-6.40	N1-ASP74:OD2	2.48	3

CS = Crystal structure, BS = Binding site, LBE = Lowest binding energy (kcal/mol), PAS = Peripheral anionic site, CAS = Catalytic site , O = Bind to other site compared to PAS and CAS (continued).

Appendix 30

Results from the docking of both sites (PAS & CAS) binding ligands (Ligplot analysis).

CS	No. in cluster	BS	LBE	Residues involved in hydrogen bonding	Length (Å)	No. of residues involved in hydrophobic interaction	CS	No. in cluster	BS	LBE	Residues involved in hydrogen bonding	Length (Å)	No. of residues involved in hydrophobic interaction
				BW284C5							**DECAMETHONIUM**		
a	2	PAS	-5.33	-	-	8	a	2	BTH	-3.54	-	-	11
b	6	BTH	-6.63	O30-PHE288:N	2.75	9	b	64	MDL	-4.05	-	-	6
c	2	PAS	-5.60	O30-PHE288:N	2.88	6	c	41	PAS	-3.70	-	-	10
d	6	PAS	-6.07	-	-	6	d	8	BTH	-3.67	-	-	9
e	8	PAS	-5.80	-	-	9	e	6	BTH	-3.69	-	-	8
f	7	PAS	-5.86	-	-	9	f	5	BTH	-3.65	-	-	10
g	1	PAS	-6.54	-	-	9	g	7	O	-3.50	-	-	11
h	2	PAS	-5.02	-	-	9	h	4	BTH	-3.79	-	-	9
i	1	BTH	-5.71	O30-PHE295:N	2.98	10	i	7	PAS	-3.63	-	-	9
j	6	BTH	-6.48	O30-PHE295:N	2.89	8	j	19	BTH	-3.88	-	-	10

CS = Crystal structure, BS = Binding site, LBE = Lowest binding energy (kcal/mol), PAS = Peripheral anionic site, CAS = Catalytic site, O = Bind to other site compared to PAS and CAS, MDL = Bind in between PAS and CAS.

CS	No. of cluster	Observed Binding Site	Lowest Binding Energy (kcal/mol)	Residues involved in hydrogen bonding	Length (Å)	Hydrophobic Interactions (No. of residues)
				DONEPEZIL		
A	5	PAS	-5.89	-	-	8
B	8	BTH	-7.06	O58-ARG289:N	2.77	7
c	1	BTH	-7.01	O58-ARG289:N N3-TYR121:OH	2.78 2.75	8
d	4	PAS	-6.39	N3-ASP285:OD1 O58-ARG289:N O2-TYR121:OH	2.81 3.21 3.00	9
e	6	PAS	-6.26	O58-PHE288:N O58-ARG289:N N3-ASP285:OD1	3.07 3.20 2.84	9
f	1	BTH	-6.45	N3-TYR121:OH O58-PHE288:N	2.84 2.90	7
g	2	O	-5.84	N3-ASP131:OD2	2.99	11
h	2	BTH	-7.46	N3-TYR124:OH O58-ARG296:N	2.78 2.69	9
I	1	PAS	-6.81	O58-TYR72:OH N3-TYR124:OH	2.93 2.81	9
J	5	BTH	-6.88	N3-TYR124:OH	2.91	8

CS = Crystal structure, BS = Binding site, LBE = Lowest binding energy (kcal/mol), PAS = Peripheral anionic site, CAS = Catalytic site, BTH = Bind at PAS and CAS, O = Bind to other site compared to PAS and CAS (continued).

Appendix 31: Abstract and certificate for oral presentation at the 1st Pharmaceutical Sciences Conference & Exhibition (PSCE), Penang, Malaysia, 27th-28th September 2010:

Following is the abstract for oral presentation at the 1st Pharmaceutical Sciences Conference & Exhibition (PSCE), Penang, Malaysia, 27th-28th September 2010:

MODEL BUILDING FOR VIRTUAL SCREENING OF COMPOUND LIBRARIES FOR ANTICHOLINESTERASE ACTIVITY

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Recent studies have identified that acetylcholinesterase (AChE) accelerates the formation of amyloid fibrils and stable complexes with amyloid beta (Aβ) that produce the senile plaques characteristic of Alzheimer's disease (AD). Our aim was to identify a structure to be used for the virtual screening on commercially available compound libraries to predict their ability to interact with the Aβ binding site thus preventing or delaying the degeneration of cholinergic neurons. The AChE crystal structures (PDB code: 1B41, 1EVE, 1JO7, 1N5M, 1N5R and 2HA2) were extracted from the protein data bank and the ligands removed. 19 standard acetylcholinesterase inhibitors were docked into the resulting structures using Auto Dock Software and the results analysed in order to find the most suitable model for virtual screening to identify peripheral anionic site (PAS) binding ligands. The docking results using a crystal structure of human acetylcholinesterase in complex with fasciculin II (PDB CODE: 1B41) have shown that most of the PAS ligands bound near the aromatic residue Trp 286. This is in agreement with site directed mutagenesis studies. Therefore, the structure obtained from this crystal structure was found to be the most suitable for identifying PAS ligands.

Appendix 32

Abstract and certificates for poster presentation at the 25th Scientific Meeting of The Malaysian Society of Pharmacology and Physiology (MSPP), Universiti Putra Malaysia, Malaysia, 25th-26th May 2011 (Awarded a Best Poster Prize in Poster Presentation Contest):

Following is the abstract for poster presentation at the The 25th Scientific Meeting of The Malaysian Society of Pharmacology and Physiology (MSPP), Universiti Putra Malaysia, Malaysia, 25th-26th May 2011:

ANTICHOLINESTERASE ACTIVITY OF CHALCONES AND THEIR MOLECULAR INTERACTIONS

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Acetylcholinesterase (AChE) plays a crucial role in the metabolism of the neurotransmitter, acetylcholine. Hence, AChE inhibitors can be used to increase the level of acetylcholine present in the synapses between the cholinergic neurons to improve cholinergic function in Alzheimer's disease (AD). Recent studies have shown AChE enhances the aggregation of amyloid beta (Aβ) peptide fragments into amyloid fibrils to form senile plaques characteristic of AD, and the presence of AChE inhibitors is known to suppress this AChE-induced amyloid fibril formation. In this study, we investigated anticholinesterase activity of chalcones using Ellman colorimetric assay and studied their molecular interactions by molecular docking experiments. For computational study, the AChE crystal structure, human AChE in complex with Fasciculin II (PDB code: 1B41) was extracted from the protein data bank and the ligand Fasciculin II removed. Chalcones with anticholinesterase activity were docked to this crystal structure using AutoDock 4.0 software to study protein-ligand binding interactions. Fifty-one chalcones were investigated for their anticholinesterase activity. The cholinesterase assay predicted that ten compounds exhibited more than 20% inhibition and one gave more than 50% inhibition at 10 µM, whereas the positive controls tacrine and propidium gave 60% and 40% inhibition at the same concentration, respectively. The docking experiments further demonstrated that these ligands either bind to the catalytic site near to the aromatic residue Trp 86 or at peripheral anionic site near the aromatic residue Trp 286, or bind to both Trp 86 and Trp 286. This study successfully showed that some chalcones have the potential to inhibit acetylcholinesterase activity and may improve cholinergic function in AD.

Keywords: Alzheimer's disease, Acetylcholinesterase, Ellman assay, Chalcones, Molecular docking

APPENDIX 33

Published articles