DECIPHERING THE NEUROPROTECTIVE ROLE OF SIGMA1 RECEPTOR, AN IMPORTANT FUNCTION TO OVERCOME THE SYMPTOMS OF NEURODEGENERATIVE DISORDERS

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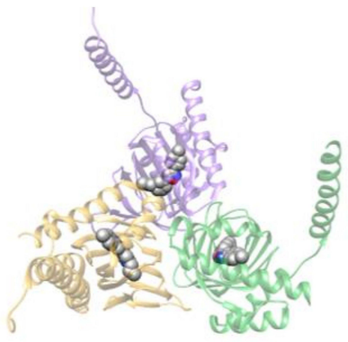
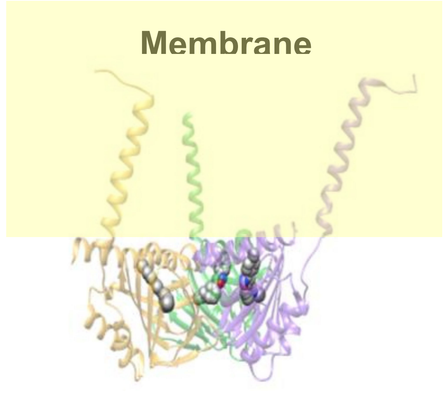
Sigma-1 receptor (σ1R)

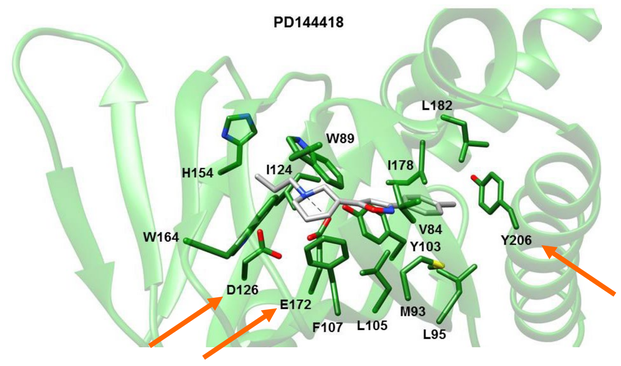
- expressed in the central nervous system
- **agonists** are **neuroprotective**
- anchored to cell and ER **membranes**
- experimentally determined **3D-structure**
  - X-ray crystallography
  - Resolution: 2.51-3.20 Å
  - Complexes: 1 agonist, 4 antagonists
  - No variations in the **ligand-binding region**



Top-view



Side-view



Ligand-binding site

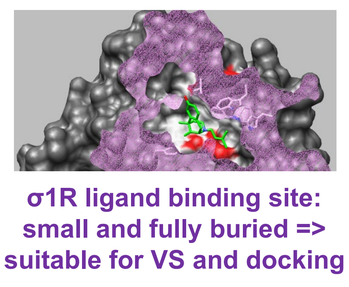
Identification of σ1R-binding neuroprotective drugs for HD therapy by Drug "repositioning" or "repurposing"

1) *In silico* analysis: Prediction of drug-σ1R interaction

a. Virtual Screening (VS)
- σ1R 3D structure: PDB ID: 5HK1 (best resolution)
- Ligands: ZINC **FDA-approved drugs** library
- Software: Autodock VINA
  - ➢ **Result: ranking by predicted affinity**

b. Computational docking
- Ligands: **20 drugs** with **highest predicted affinity**
- Software: Autodock Tools
  - ➢ **Result: predicted σ1R-drug complex structure and affinity**

c. Visual inspection
- **20 drug-σ1R predicted structures**
- Software: PyMol, InsightII
  - ➢ **Result: chosen 6 drugs based on potential interactions and clinical activity**
    - **Iloperidone, Paliperidone: Schizophrenia**
    - **Vilazadone: Depression**
    - **Flibanserin: Sexual desire hypoactivity**
    - **Nilotinib: Chronic myeloid leukemia**
    - **Linagliptin: Diabetes mellitus type 2**



σ1R ligand binding site: small and fully buried => suitable for VS and docking

2) *In vitro* analysis by SPR(*): Assessment of direct drug-σ1R interaction

FDA Name	ZINC ID	KD (μM)	Energy (kcal/mol) VINA	ATD1	ATD2
Flibanserin	52716421	4.9 ± 1.1	-11.6	-9.4	-10.0
Iloperidone	01548097	**5.1 ± 0.6**	-11.7	-10.2	-10.4
Linagliptin	03820029	9.6 ± 1.0	-11.7	-12.4	-12.4
Pridopidine	22063703	14.8 ± 1.0	-8.7		
Nilotinib	06716957	22.0 ± 3.0	-12.3	-7.8	-9.5
Paliperidone	04214700	46.0 ± 21	-12.2	-11.5	-11.9
Vilazodone	01542113	52.0 ± 9.0	-11.6	-9.2	-9.4

➢ **Result: All 6 selected drugs bind purified σ1R with affinity ~ pridopidine**

*: Surface Plasmon Resonance
**: Phase III clinical trials
ATD1: lowest energy pose of largest Autodock cluster
ATD2: lowest energy pose of lowest energy Autodock cluster

3) HD patients' skin fibroblasts: Assessment of drug agonist effect on cells

[1 μM]	HD 1	HD 2	CTRL 1	CTRL 2	KD (μM)	Energy (kcal/mol)		
Iloperidone	+	+			**5.1 ± 0.6**	-11.7		
Paliperidone	+	+	-	+	46.0 ± 21	-12.2		
Nilotinib	+	+		-	22.0 ± 3.0	-12.3	**Best 20**	
Flibanserin	+	-		+	4.9 ± 1.1	-11.6		
Linagliptin	+			+	9.6 ± 1.0	-11.7		
Vilazodone	+		+	+	52.0 ± 9.0	-11.6		
Pridopidine	+	+	+	+	**14.8 ± 1.0**	**-8.7**	**458th**	**1.7**
Other known ligands								
Haloperidol							**51st**	
N,N-dimethyltriptamine							**1002nd**	**3.8**

➢ **Result: HD fibroblasts growth and growth rate are increased (both or one patient) by all 6 selected drugs after 72 hours (=> all 6 are agonists) and cell death is decreased by 3 drugs (in *italic*)**

*: Both have **43 CAG repeats** in ***Htt*** and are at the **same initial HD stage**
: **VINA/ATD Energy differences **≤ 3 kcal/mol** are **not significant**

Conclusions

- ➢ The **Drug Repositioning** procedure identified **6 FDA-approved drugs** able to **improve HD fibroblasts phenotype**
- ➢ The 6 drugs are directly amenable to **clinical use** and can be used as **leads** for implemented therapeutics

Battista *et al.* (2021)
Int J Mol Sci

Current work

- **Ranking improvement** by *in silico* methods (e.g., by Artificial Intelligence methods)
- **Medium-scale** (i.e., **tens of compounds**) implementation of *in vitro* methods
- **Additional** HD cell models: **fibroblasts**; **iPSC-derived** neurospheres and **neurons**
- Investigation of **drug activity mechanism** (e.g., σ1R **antagonists**; involved **pathways**)
- Identification of the **endogenous σ1R ligand(s)** by Virtual Screening of large (tens of thousands) compound library
- Investigation of **σ1R ligand entrance mechanism** by Molecular Dynamics simulations