Docking of Dobutamine on Beta1 Adrenergic Receptor

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Abstract

Many disease treatments have used synthetic neurotransmitter drugs. This kind of drugs attaches and submits electrochemical messages to receptors to trigger downstream cascade for treatment propose. Docking experiment of drugs to their receptors in patients is not facile; spend both time and money; moreover it is also dangerous.

The advantages of computational simulation are inexpensive and obviously safety. This work utilizes engineering methodologies and analyses with AutoDock4 software and mathematical model to study docking of Dobutamine on the β_1-adrenergic receptor. As a result of the injection of Dobutamine and the uptake of Dobutamine by the patients, the patients' heart rate increased. Dobutamine attached to the sinoatrial (SA) node cells at particular sites on the membrane surface. These sites are known as β_1-adrenergic receptors. As there are no experimental results available for this docking procedure this work describes the calculations involved in estimating the energies and possible positions of docking. The minimum total energy from prediction of energies is 10.49 kcal/mol and all binding lengths of possible positions of docking are less than 2.5 Å. Understanding docking of drugs on their receptors not only raising inquiring questions about problems, but is also leading to the improvement of strategies to treat the patients.

Keywords: AutoDock, Dobutamine, Sinoatial Node, β_1-Adrenergic Receptor

1. Introduction

Dobutamine is a sympathetic nervous system drug used for treating people who have problems with heart failure and cardiogenic shock. Dobutamine ($C_{18}H_{23}NO_3$), has a 2 minute half-life in the human body, and is a drug that provides direct stimulation to the β_1-adrenergic receptor. However Dobutamine also has a small effect in the stimulation of β_2 and α_1 receptors. The average molecular weight and monoisotopic molecular weight of Dobutamine is 301.3801 and 301.1678. The docking of Dobutamine to these receptors occurs at the SA node cell wall that is a small mass of specialized tissue located in the right atrium of the heart.

Beta receptor is a class of G-protein-coupled receptors. There are three known types of beta receptor, β_1, β_2, and β_3. β_1-adrenergic receptors

(ADRB1) are located mainly in the heart and in the kidneys.

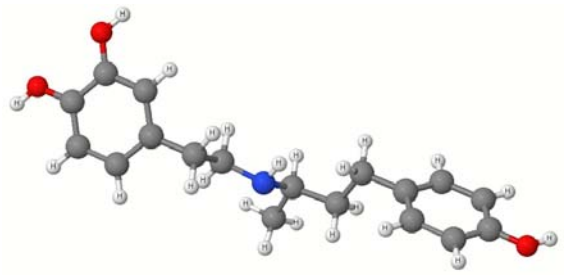

Fig. 1 The chemical structure of Dobutamine

2. Using AutoDock4

The AutoDock4 software, free available software under the GNU General Public License, is designed to predict the interaction of the ligand to a set of grids describing the target protein. AutoDock4 has been widely used in many academic, governmental and non-profit institutions around the world because AutoDock4 not only save times and cost in managing the experiments but also provides high quality predictions. To utilized AutoDock4 software calculated, there are several performed in four major steps: preparation of coordinate, precalculation of atomic affinities, docking of ligands, and analysis of results.

In coordinate preparation step, the Protein Data Bank (PDB) of the ligand and the target protein are used. The extended PDB format provides standard representation for macromolecular structure data that includes polar hydrogen atoms, but not hydrogen atoms bonded to carbon atoms. AutoDock4 software converts PDB format to PDBQT coordinate files including atomic partial charges, atom types and information on the torsional degrees of freedom.

The pre-calculation of atomic affinities step, involves using the AutoGrid procedure whereby the protein is embedded in a three-dimensional grid and a probe atom is placed at each grid point. The energy of interaction of this single atom with the protein is assigned to the grid point.

The docking of ligands step is carried out using one of several search methods. The most efficient method is a Lamarckian Genetic Algorithm (LGA), For typical systems, AutoDock is run several times to obtain several docking conformations Analysis of the predicted energy and the consistency of results are combined to identify the best solution.

The last step involves the analysis of results. AutoDockTools includes a number of methods for analyzing the results of docking simulations. These include tools for clustering results by conformational similarity, visualizing conformations, visualizing interactions between ligands and proteins, and visualizing the affinity potentials created by AutoGrid.

Each docking by AutoDock4 requires at least four input files. (1) a PDBQT file for the ligand, dobutamine data file from PDB file, (2) a PDBQT file for the receptor, β_1-adrenergic receptor data file from PDB file, (3) a grid parameter file (GPF) for the AutoGrid calculation, and (4) a docking parameter file (DPF) for AutoDock4 calculation.

3. Docking Positions of Dobutamine Atoms

The results for docking of Dobutamine into the β_1-adrenergic receptor by the AutoDock4 program are given in terms of ten interactions. The possible docking positions of the dobutamine atoms specified in cartesian coordinates. Docking is assumed to be assured when the length between bonded atoms is less than 2.5×10^{-10} m. That is if the Dobutamine atom is length within

2.5 x 10^{-10} m of the atoms of β_1-adrenergic receptor, then bonding is assumed to have taken place. Therefore, the nearest atom between the β_1 receptor and each atom of Dobutamine can be calculated from

$$d_{\min} = \sqrt{(x_b - x_d)^2 + (y_b - y_d)^2 + (z_b - z_d)^2} \quad (1)$$

Where as d_{min} is the least distance between Dobutamine and β_1 receptor atom, (x_b, y_b, z_b) is the position of β_1 atom, and (x_d, y_d, z_d) is the position of Dobutamine atom.

In all cases the Dobutamine had at least one O-bond connected to the β_1-adrenergic receptor. The result of the calculated docking positions for Dobutamine is shown in figure 3. The most likely configurations of all ten possible docking positions are the first and the fourth docked positions, figure 3, involving 3 O-bonds on the β_1-adrenergic receptor.

4.Docking of Dobutamine onto the Sinoatrial Node Model

There are five possible sites on the Dobutamine molecule that docking can occur. The motion of the Dobutamine from the blood to the cell membrane was analyzed in three scales. The flow in the region most distant from the surface was a continuum region, the interaction at the blood cell size level was a Monte Carlo process and the interactions with the receptors was undertaken using a direct simulation method known as molecular dynamics.

4.1. Continuum Scale

The two dimensional Navier Stokes equation is

$$\rho\left(\frac{\partial \hat{u}}{\partial t} + \hat{u} \bullet \nabla \hat{u}\right) + \nabla p = \mu \nabla^2 \hat{u} + \hat{F} \quad (2)$$

The blood flow velocity closest to the membrane was used as the bulk flow input to the blood cell scale calculation.

4.2. Blood Cell Scale

The Monte Carlo method was used. The blood is considered to be composed of water, erythrocyte, albumin, angiotensin II and Dobutamine. The solution starts with the Landau equation which in the test particle form below has been described as a generalized diffusion equation in velocity space, Chandrasekhar, (1942). Expressed in a non-dimensional form it becomes

$$\partial \varphi_\tau = \partial_{v_r}(-F_r + 0.5\partial_s T_{rs})\varphi \quad (3)$$

where φ is the velocity distribution, the v_r differentiation is with respect to non-dimensional velocity $v/2kT$, subscript τ is differentiation with respect to the non-dimensional time defined below.

$$F_r = -8v^{-1}G(v)v_r \quad (4)$$

$$T_{rs} = 2v^{-1}H(v)\delta_{rs} + 2v^{-3}E(v)v_r v_s \quad (5)$$

and H, G and E are tabulated Chandrasekhar, 1942 [1]. The non-dimensional [2] time is Balescu 1975

$$t = \frac{\beta^{3/2} Bn}{m^{1/2}} \tau \qquad (6)$$

where m is the mass, n the number density, $\beta = 1/kT$ and B is defined as

$$B = 8\pi^5 \int_0^{l_m} l^3 V_l dl \qquad (7)$$

The movement of the blood components assumes they are sufficiently far apart so that collisions between the components will not occur. This is the usual assumption made for the application of the Landau equation. Under these circumstances the force on an ion will consist of a drag due to $G(v)$ and a random force due to $H(v)$. The time scale is as defined in equation (6).

4.3. Molecular Dynamics Scale

The interaction time scale is as defined in equation (7). The convective step is then implemented. This is achieved by choosing a short length of time ΔT. The particles then move with the velocity v attained at the end of time ΔT for a distance $v\Delta T$. New cells are then formed and the process repeated. The boundary conditions as described above are applied at the end of each time step ΔT. The value of ΔT was determined as follows [3]. Within a cell containing N particles the particle with the largest total interaction cross section σ_i is chosen for collision where

$$\sigma_i = \sum_{j=1}^{N} \frac{|c_i - c_j|}{c_i} \sigma_{oij} \qquad (8)$$

The cross section is very difficult to calculate in the present case as the particles are so large. Thus two possible interactions were considered. In one case the particles were considered to carry a charge and the collision cross section σ_{oij} is given in terms of the deflection angle χ_m. In the other case the particles were considered to be hard spheres. The two cases were compared to judge the importance of the cross sectional approximation. The procedure then continues by choosing two colliding particles and time t calculated by

$$t_i = -\xi \times \frac{n}{\sigma_i N} \qquad (9)$$

Where ξ is a random number between 0 and 1, n is the number of molecules in the cell, N is the number density. This process is repeated for all cells. The geometry for the calculation of the diffusion of the Dobutamine in the sinoatrial node is complex as shown in figure 2 from [4].

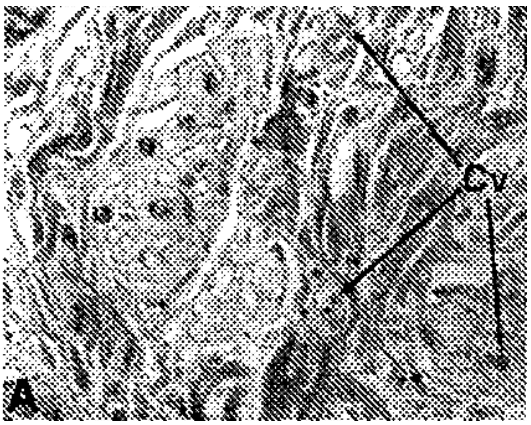

*Fig.*2 Sinoatrial node histologic section [4]

The arrows point to capillaries. The length of the centre arrow is approximately 50 μm long. The distance between the capillaries is then 48μm and 81μm. An accurate calculation of a docking

Table.1 Minimum distance docking of dobutamine atom on β_1 receptor

dobutamine atom number	dobutamine atom name	beta I atom	docking 1	2	3	4	5	6	7	8	9	10
1	N	d_{min} (A)	2.030879	2.227681	1.898663	1.576158	2.454867	2.200735	2.238256	2.108333	1.845344	2.227313
		number	3513	3513	3447	1379	3513	3447	3513	3513	1379	3513
		name	H	H	H	H	H	H	H	H	H	H
2	C	d_{min} (A)	2.677225	2.583053	2.013816	2.338299	2.402119	1.862699	2.573506	2.671232	2.867135	2.570462
		number	4013	4068	1377	745	4068	699	4068	4068	1377	4068
		name	O	O	H	O	O	H	O	O	H	O
3	C	d_{min} (A)	1.944167	2.134034	1.594833	2.231108	2.085795	1.609373	2.117294	2.007401	2.00139	2.11445
		number	3518	4017	1441	696	4017	1441	3518	3518	699	3518
		name	H	H	H	H	H	H	H	H	H	H
4	C	d_{min} (A)	1.539398	1.590693	1.932249	2.207014	1.669642	1.916372	1.596855	1.55737	1.763982	1.617808
		number	4058	4058	1379	1379	4058	1379	4058	4058	1379	4058
		name	H	H	H	H	H	H	H	H	H	H
5	C	d_{min} (A)	1.621223	1.602072	1.535142	1.566899	1.554165	1.63029	1.677494	1.647287	2.333864	1.638116
		number	4060	4060	1377	733	4060	1377	4060	4060	696	4060
		name	H	H	H	H	H	H	H	H	H	H
6	C	d_{min} (A)	2.165752	2.196894	1.794599	2.802463	2.250478	1.957262	2.24155	2.191733	2.062712	2.23033
		number	4058	4058	4058	1379	4058	4058	4058	4058	1379	4058
		name	H	H	H	H	H	H	H	H	H	H
7	NH	d_{min} (A)	2.288844	2.567856	2.234437	2.24444	2.869532	2.498398	2.621016	2.418747	1.795337	2.581187
		number	3513	3513	4057	745	3513	4057	3513	3513	3451	3513
		name	H	H	H	O	H	H	H	H	H	H
8	C	d_{min} (A)	2.170715	2.246317	2.582906	2.495677	2.345294	2.383681	2.247627	2.188144	2.629409	2.257475
		number	4057	4057	3513	795	4057	1379	4057	4057	3513	4057
		name	H	H	H	H	H	H	H	H	H	H
9	C	d_{min} (A)	2.958106	2.87004	2.008759	2.311648	2.885334	2.190704	2.812965	2.904862	1.938495	2.829062
		number	696	696	1379	3518	696	3513	696	696	1344	696
		name	H	H	H	H	H	H	H	H	H	H
10	C	d_{min} (A)	1.860613	1.818934	2.696378	1.748065	1.887303	2.273451	1.822039	1.857809	2.286053	1.790515
		number	696	696	1344	4027	696	3518	696	696	1344	696
		name	H	H	H	H	H	H	H	H	H	H
11	C	d_{min} (A)	1.821568	1.756665	2.28425	2.59943	1.744243	2.172315	1.72938	1.794543	2.246716	1.759987
		number	4109	4109	795	4031	4109	795	4109	4109	795	4109
		name	H	H	H	H	H	H	H	H	H	H
12	C	d_{min} (A)	2.54318	2.512944	2.451141	2.275417	2.488342	2.402601	2.537733	2.528501	2.558469	2.497912
		number	4057	4057	3518	1326	1441	795	4057	4057	4017	4057
		name	H	H	H	H	H	H	H	H	H	H
13	C	d_{min} (A)	1.512946	1.567118	2.679387	2.440869	1.555964	2.177795	1.568651	1.519822	2.730006	1.580801
		number	3447	3447	3513	795	3447	1379	3447	3447	3513	3447
		name	H	H	H	H	H	H	H	H	H	H
14	C	d_{min} (A)	1.475312	1.433906	2.190991	1.763739	1.428864	2.059255	1.448871	1.474129	1.948739	1.455699
		number	4109	4109	795	852	4109	795	4109	4109	4027	4109
		name	H	H	H	H	H	H	H	H	H	H
15	H	d_{min} (A)	1.428794	1.399358	2.643142	1.481419	1.446255	2.044915	1.431993	1.438138	1.691545	1.475687
		number	4109	4109	795	4031	4109	1326	4109	4109	4027	4109
		name	H	H	H	H	H	H	H	H	H	H
16	O	d_{min} (A)	2.134419	2.103551	1.965254	1.465858	1.994417	1.661771	2.072198	2.08517	1.821955	2.111291
		number	3444	3444	4017	1326	1441	1327	3444	3444	4017	3444
		name	H	H	H	H	H	H	H	H	H	H
17	OH	d_{min} (A)	2.120306	2.169942	1.705083	1.85392	1.962373	1.658157	2.106909	2.103151	1.99951	2.193864
		number	3382	3382	4027	1275	3444	796	3382	3382	4066	3382
		name	O	O	H	H	H	H	O	O	H	O
18	C	d_{min} (A)	1.970847	2.321681	1.978692	2.489798	2.149166	1.793044	2.404331	2.151246	1.590392	2.345011
		number	3518	3518	4109	746	795	704	3518	3518	1441	3518
		name	H	H	H	O	H	H	H	H	H	H
19	C	d_{min} (A)	2.348851	2.042652	2.011218	2.240003	1.88545	1.960554	2.046179	2.169074	2.247458	2.057044
		number	795	795	4109	4109	795	4109	795	795	3444	795
		name	H	H	H	H	H	H	H	H	H	H
20	C	d_{min} (A)	1.63237	1.591354	2.2235	2.010432	2.734474	2.106401	1.630084	1.579186	1.682649	1.585354
		number	795	795	4109	704	1327	4109	795	795	4057	795
		name	H	H	H	H	H	H	H	H	H	H
21	C	d_{min} (A)	2.492761	2.227842	2.273213	1.750646	2.027591	2.065505	2.170675	2.311158	2.344815	2.224597
		number	795	738	4057	1441	1379	4057	738	738	4057	738
		name	H	H	H	H	H	H	H	H	H	H
22	C	d_{min} (A)	2.191487	2.384114	2.353264	1.911312	2.214161	2.456784	2.377623	2.404786	2.613967	2.396655
		number	1379	1379	4057	1441	1379	4057	1379	1379	3382	1379
		name	H	H	H	H	H	H	H	H	O	H
23	C	d_{min} (A)	1.951145	1.999663	2.610412	2.317097	2.292541	2.577731	1.901815	2.000821	1.486098	1.974797
		number	1379	1379	4057	3444	738	4107	1379	1379	3390	1379
		name	H	H	H	H	H	O	H	H	H	H
24	C	d_{min} (A)	2.367853	2.664385	2.465423	1.905485	1.70999	1.951352	2.653151	2.544194	1.716079	2.624299
		number	1332	1332	3390	4057	795	3390	1332	1332	3390	1332
		name	H	H	H	H	H	H	H	H	H	H
25	O	d_{min} (A)	1.992535	2.021996	1.880573	1.868062	1.99621	2.05256	1.918517	1.979128	2.43563	1.943889
		number	732	733	4088	3391	732	4088	733	732	3380	733
		name	H	H	H	H	H	H	H	H	H	H
26	OH	d_{min} (A)	1.925967	1.93669	1.856748	1.720646	2.024015	1.917644	1.845033	1.866101	1.954102	1.88807
		number	1378	733	4050	3444	1378	4088	733	733	4162	733
		name	O	H	O	H	O	H	H	H	H	H

process would require detailed knowledge concerning the cell structure, the location of the interstitial fluid, the capillary lengths, the number of capillaries normally active etc. Within the limits discussed below the diffusion equation (9) can be approximately solved.

$$\frac{\partial C_N}{\partial t} = D \frac{\partial^2 C_N}{\partial^2 x} \qquad (10)$$

Although the diffusion process is three dimensional [5], due to the uncertainties in the present case only a one dimensional solution will be considered. The molecular dynamics region was the region above the surface and below the Monte Carlo region. The β_1-adrenergic receptor molecule raises approximately 50Å above the cell surface. Thus the lower surface of the Monte Carlo region was placed at 57Å above the cell surface. If a Dobutamine molecule entered the molecular dynamics region it was allowed to proceed at its current velocity to the cell surface. The density of β_1-adrenergic receptors of β_1-adrenergic receptors was obtained from [6] as 7.7 pmol/mL. Assuming that 30% of the receptors would be activated at a given time a random number was generated and if it was greater than the probability of hitting a receptor a collision was considered to occur. An arbitrary impact parameter was chosen for the Dobutamine molecule as well as an arbitrary rotational angle. The molecule was then allowed to proceed through the molecular dynamics region until it intercepted the receptor.

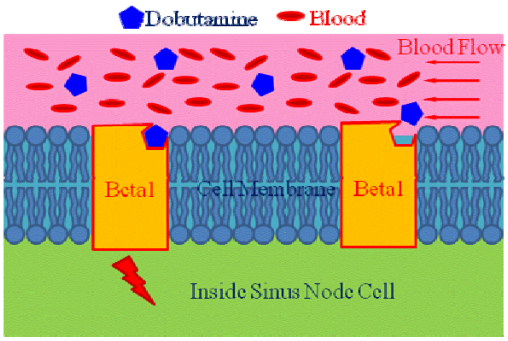

*Fig.*3 Docking of Dobutamine onto the SA Node Model

If the appropriate atoms on the Dobutamine were within 3Å of a docking site, as shown in table 1, then a docking was considered to occur. At this time the receptor was removed from the cell as thus the density of receptors in the cell was reduced. New Dobutamine molecule was introduced at the midpoint of the region of interest.

5. Docking of Dobutamine Model Result

The docking of Dobutamine model was undertaken for four different dosages of Dobutamine 10, 20, 30, and 40 mics. Because this model required extensive CPU time, each simulation dosage was run until 200 molecules of Dobutamine docked into the β_1-adrenergic receptor. This was considered sufficient time as based on the dosage time probably the Dobutamine would be released by the receptor in this time. All dosages results look like the linear curve are linear except 10 mics. For 10 mics dosage, the curve is beginning to look exponential. If simulated this model long enough all curve should be exponential.

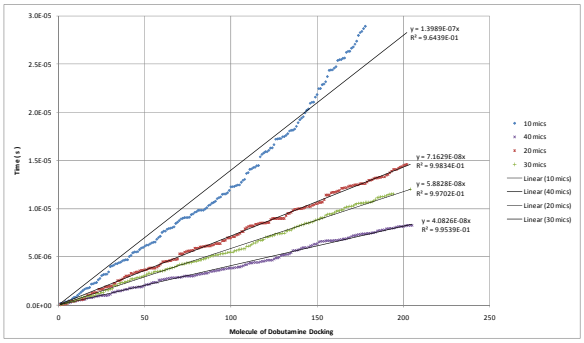

*Fig.*4 Number of Dobutamine molecules with time

6. Conclusion

The locations of dobutamine atoms that docked into the β_1-adrenergic receptor were found by the AutoDock4 software. The best position for docking is the lowest free energy level. The data from AutoDock4 software results corresponding to the pairs of bonding atoms were used as the parameters for simulation docking of dobutamine model. The quantity of dobutamine molecules docking for each dosage as a function of time period was determined using the procedures described above.

7. References

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