Research Article

In silico Docking Analysis of Anticancer Drugs against Casein Kinase II

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ABSTRACT

Casein Kinase II plays a casual role in tumorigenesis. It phosphorylates and positively modulates the activity of several proteins that plays a critical role in cell survival. In present study, molecular docking is performed to study the interactions between drugs from chemical origin like Doxorubicin, Methotrexate, Mitomycin C, Ofatumumab and Berbamine and a natural product based drug Teniposide and the protein. The chemical structure of the drugs was retrieved from PubChem and docked using Patchdock. The present study suggested that Teniposide has higher affinity to suppress casein kinase II.

Keywords: Docking, Anticancer drugs, Cancer, Casein Kinase II.

INTRODUCTION

Casein Kinase II (CK2) is a constitutively active serine/threonine protein kinase composed of two 44 kDa catalytic α-subunits and two 26 kDa regulatory β-subunits in a $\alpha_2\beta_2$ configuration to form stable heterotetramers. CK2 holoenzyme undergoes autophosphorylation at two serine residues (S2/S3) of its β-subunit. Casein Kinase 2 (CK2), a highly conserved, multifunctional serine/threonine protein kinase, is critically important for regulation of a plethora of processes in eukaryotes, such as cell proliferation, differentiation and death. It is expressed in all tissues; in particular, its amount and activity are elevated in tumor cells. Due to key regulatory role CK2 plays in cell fate determination in cancer cells, there is a tremendous interest in development of CK2-specific therapies and drugs.[1] CK2 is known to phosphorylate tumor suppressors such as AKT, PTEN and p53 causing inhibition of apoptosis in cancer cells. Apart from its key involvement in regulating normal cell growth and development, enhanced CK2 activity was observed in variety of cancers, including breast, prostate, lungs, leukaemia and brain.[2,3] Over expression of protein kinase CK2 is an unfavourable prognostic marker in several cancers. CK2 has emerged as a relevant therapeutic target. Elevated CK2 activity has been uniformly documented in different human cancer types and has been correlated with aggressive tumor behaviour.[4]

Cancer is a major cause of death and number of new cases as well as number of individuals having cancer is increasing continuously. Cancer is associated with multiple genetic and regulatory aberrations in the cell. It is a highly heterogeneous disease, both morphologically and genetically. Analysis of cancer pathways shows a number of interrelated markers responsible for oncogenesis.[5] Selection of a therapeutic target is a fuzzy task. In the present study, the focus is on CK2 as a target and its inhibition by an active compound is studied by *in silico* docking analysis. Casein kinase II is a serine/threonine protein kinase that phosphorylates acidic proteins such as casein. It is involved in various cellular processes, including cell cycle control, apoptosis, and circadian rhythm. The kinase exists as a tetramer and is composed of an alpha, an alpha-prime, and two beta subunits. The alpha subunits contain the catalytic activity while the beta subunits undergo autophosphorylation. The protein encoded by this gene represents the alpha subunit. While this gene is found on chromosome 20, a related transcribed pseudogene is found on chromosome 11.

Three transcript variants encoding two different proteins have been found for this gene. A number of structurally unrelated CK2 inhibitors, tested on a variety of cells derived from tumours, including lymphomas, leukaemia, multiple myeloma and prostate carcinoma, display a pro-apoptotic effect which is roughly proportional to their in vitro inhibitory potency.[6]

MATERIALS AND METHODS

Protein Preparation

Casein Kinase II alpha subunit contain catalytic activity was chosen as a receptor molecule because of its role in cancer. The protein sequence for CK2 subunit alpha has NCBI accession number NP_001886.1.

The protein structure is available at Protein Databank PDB (http://www.pdb.org/) as 4MD8_E having 99% identity and 100% query coverage as per PSI BLAST (http://blast.ncbi.nlm.nih.gov/).

Hydrogen atoms were added to the protein using CHARMM force field on Accelyrs Discovery Studio client software.

Binding Site Analysis

Active Sites of the receptor were analysed by MetaPocket (http://metpocket.eml.org).[7] Binding sites are the

distribution of surrounding residues in the active site (Table 1). The centre of active site is selected as grid map value for preparation of the grids for docking.

Ligand Preparation

Through literature study the list of CK2 inhibitors including available inhibitors and natural inhibitors was made. The structure of the ligands was downloaded from PubChem database (http://pubchem.ncbi.nlm.nih.gov/). The drug files are available in sdf format and were converted to pdb format using Open Babel software. The ligand preparation included 2D-3D conversion, verifying and optimizing the structures. The energy was minimized using Accelyrs Discovery studio client software.

Docking

Patchdock was used to dock receptor and ligand molecules. The algorithm of PatchDock is inspired by object recognition and image segmentation techniques used in computer vision. It is based on shape complimentarily.[8,9] Docking was compared to assembling a jigsaw puzzle. When solving the puzzle the algorithm try to match two pieces by picking one piece and searching for the complementary one.

RESULTS AND DISCUSSION

Three top active sites predicted by Metapocket are shown in Figure 1 in forms of spheres. Active site 1 has the largest pocket and can be the potential drug binding site.

Table 1 has listed all important amino acids forming the active sites.

Few drugs as listed in Table 2 having anticancer properties were chosen with SMILES and 3D structure and were docked against CK2 to study their interaction using PATCHDOCK at default settings. The software was assessed using Score, Area and atomic contact energy of the protein-drug interaction.

Score provided by Patchdock is the geometric shape complementarily scores, Area is the approximate interface area of the complex and ACE is atomic contact energy, it is used to calculate the free energy of transferring side chains from protein interior into water. ACE provide a reasonable accurate and rapidly evaluated salvation component of free energy.[10]

Drug- receptor interactions were evaluated to know their activity using 3D structure of receptor from protein data bank. The binding mode of drug with receptor was studied and analysis suggested that binding pattern was varied with ligand's chemical nature as all the ligands docked at the same active site still their interacting residues are different (Figure 2).

The binding free energy calculated by ACE parameter of PATCHDOCK (Table 3) showed that among 6 drug molecules, Teniposide drug (plant derived) showed maximum binding energy i.e - 305.24 Kcal/mol which explains that it binds to the protein's active site more strongly. This could be exemplified based on the interaction studies among the drug and receptor (Figure 3).
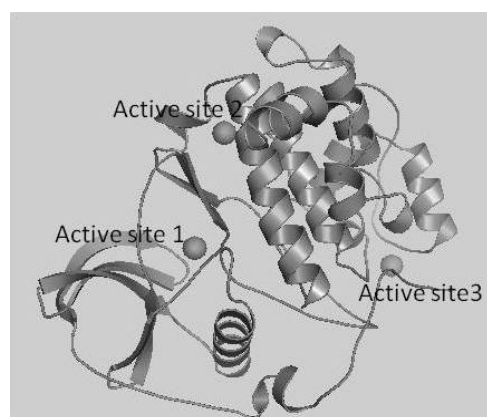

Figure 1: Protein Structure showing active sites

Table 1: Showing Binding Site Residues

S. No	Active site	Residue in active site	Centre of Active site
1	MPT1	R43,L45,E55,K64,H115,N117,V116,N118,V66,K44,D120,G46,T119,V53,M163,R45,E121,K68,F113,S51,I95,E114,G48,I174,H160,D175,K49,V162,E81,Y50,N161,P159,L124,I164,Y125,L85,W176,L70,K158,K77,E177,D156,L178,S194,V73,K74,A193,R80,K75,K76,R195,Y26,R191,V192,K198,E180,R155,N189,N238,W24,D25,E27,V190,F181,H183,Y188,Q186,D187,I78,L111,I82,K71,P72,H154,A179	70.421 9.267 26.578
2	MPT2	D120,E121,K122,H160,N118,M163,T119,E230,P159,F197,S224,PRO231,Y196,K158,V162,Y125,I164,L124,M221,M225,C220,S194,L128,I133,Q126,T127,L292,R228,K229,F232,F233,H234,N241	74.460 25.176 25.817
3	MPT3	V204,E205,I263,E264,L265,D266,E187,Y206,Q207,M208,P4,Q186,V5,N262,G3,S2	98.362 6.516 15.024

Alphabets are denoting single letter amino acids, numbers denote the position of amino acid and Centre of active sites are the X, Y and Z co-ordinates of the pocket.
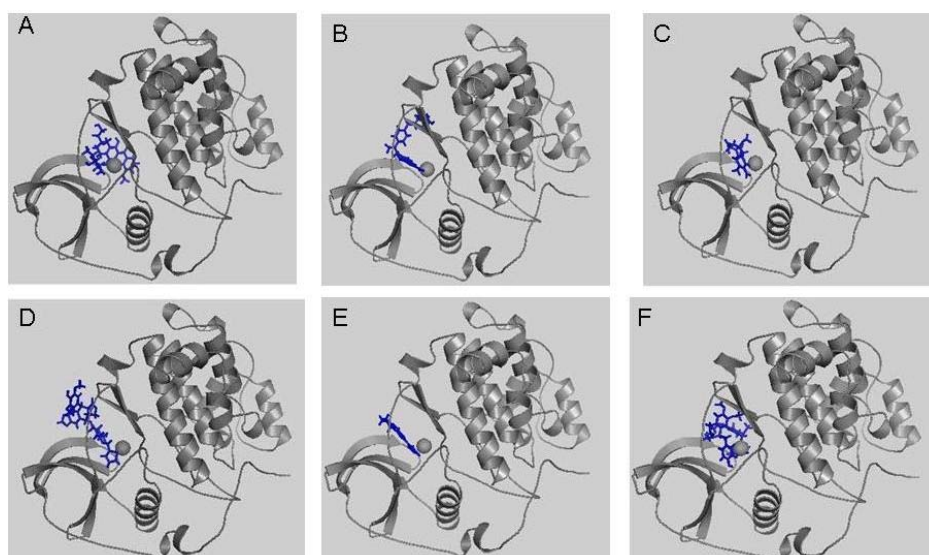
Figure 2: Drug molecule a) Doxorubicin b) Methotrexate c) Mitomycin d) Teniposide e) Ofatumumab and f) Berbamine binding with active site of CKII.

Table 2: List of Drug molecules with their structures

S. No	CID	Name	Smiles	3D Structure
1	31703	Doxorubicin	COC1=CC=CC2=C(O)C3=C(C(=C4C(CC(O)(CC4=C3O)C(=O)CO)OC5CC(N)C(O)C(C)O5)O)C(=C12)O	
2	126941	Methotrexate	CN(CC1=CN=2N=C(N)N=C(N)C2=N1)C3=CC=C(C=C3)C(=O)NC(CCC(O)=O)C(O)=O	
3	5746	Mitomycin C	COC12C3NC3CN1C4=C(O)C(=C(N)C(=C4C2COC(N)=O)O)C	
4	452548	Teniposide	COC1=C(O)C(=CC(=C1)C2C3C(COC3=O)C(OC4OC5COC(OC5C(O)C4O)C6=CC=CS6)C7=CC8=C(OCO8)C=C27)OC	
5	6918251	Ofatumumab	C1COCC2=C1N=CC3=NC(=C4C=CON4)N=C23	
6	275182	Berbamine	CN1CCC2=CC(=C3C=C2C1CC4=CC=C(C=C4)OC5=C(C=CC(=C5)CC6C7=C(O3)C(=C(C=C7CCN6C)OC)OC)O)OC	

Table 3: Docking statistics of drugs with CK2

S. No	Complex	Score	Area	Ace
1	Doxorubicin	5860	687.20	-161.42
2	Methotrexate	5746	687.70	-298.77
3	Mitomycin C	4600	525.10	-217.61
4	Teniposide	6258	800.70	-305.24
5	Ofatumumab	3700	430.90	-192.92
6	Berbamine	6268	843.00	-289.22

Table 4: Interactions among Residues of Drugs and CKII

| S. No	Complex	H-Bond Between Amino Acid and Drug	Pi Interactions
1	Doxorubicin	i) TYR50:H1 LIG:O ii) LYS158:H11 LIG:O	NIL
2	Methotrexate	i) HIS160:H5 LIG:O ii) VAL162:O LIG:H	NIL
3	Mitomycin C	i) ARG47:H1 LIG:O ii) ARG47:O LIG:H	ILE174
4	Teniposide	i) ARG43:H10 LIG:O ii) ARG43:H1 LIG:O iii) ASN118:H6 LIG:O iv) LEU45:O LIG:H	ARG43,LYS68
5	Ofatumumab	i) ASN118:H1 LIG:O	NIL
6	Berbamine	i) ASN118:H6 LIG:O	LYS68
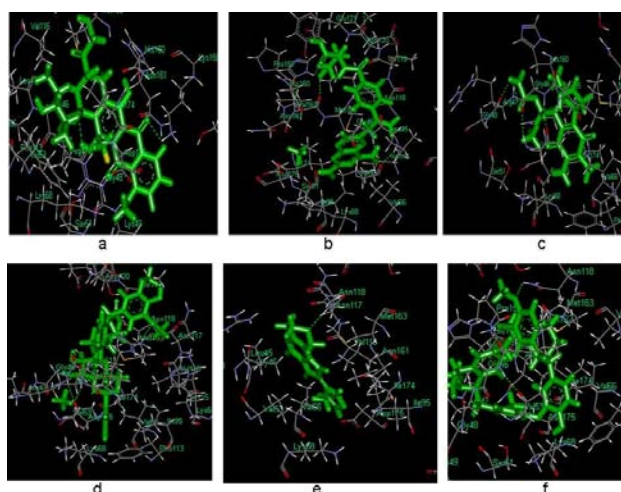
Figure 3: Drug molecule a) Doxorubicin b) Methotrexate c) Mitomycin C d) Teniposide e) Ofatumumab and f) Berbamine binding with active site of CKII. H-Bonds are shown with green dotted lines.

As in Figure 3 (dotted green lines) and Table 4 interaction between protein's amino acid and drug's atom is shown. Figure 3d is Teniposide drug which has strong binding i.e four hydrogen bonds with receptor molecule i.e. i) ARG[43]:H10 LIG:O ii) ARG[43]:H1 LIG:O iii) ASN[118]:H6 LIG:O and iv) LEU[45]:O LIG:H respectively.

CONCLUSION

The result of present study shows that Teniposide has good interactions in form of four hydrogen bonds and contact energy-305.24 Kcal/Mol when compared with other drug compounds taken in this study. Further experimental studies should be performed to validate the experiment. Therefore, these results may offer advantages in treatment of cancer.

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Source of Support: Nil, **Conflict of Interest:** None.