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Pharmacophore Based Screening of Epicatechin against Colon Cancer

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ABSTRACT

The present study deals with Pharmacophore based analog generation of Epicatechin derivatives against the PKC-alpha receptor for treating colon cancer. The chemical features of Epicatechin were analyzed by catalyst - Hiphop algorithm and its derivatives were generated based on Epicatechin Pharmacophore using Minimaybridge commercial database. Two different Epicatechin Pharmacophore analogs were acquired by virtual screening and docked against the PKC-alpha for screening the protein binding affinity. The Pharmacophore result gives HTS01501 possess -3.514 Kcal /mol highest dock score as like as Epicatechin.

Keywords: Colon cancer, Pharmacophore, Epicatechin, virtual screening, docking.

INTRODUCTION

Cancers that are confined within the wall of the colon are often curable with surgery while cancer that has spread widely around the body is usually not curable and management then focuses on extending the person's life via chemotherapy and improving quality of life. [1] Colorectal cancer (CRC) is the fourth most common cancer in men and the third in women worldwide. It is a frequent cancer, with more than 1 million new cases every year and a poor survival rate. Rapid increases in CRC incidence have been observed mainly in emerging economies. These increases are attributed to changes in diet, life style, and patterns of physical activity. In Western countries, only 55% of the patients are alive 5 years after diagnosis, with most patients dying from metastatic disease. [2]

The colon cancer is originating from the epithelial cells lining the colon or rectum of the gastrointestinal tract, most frequently because of mutations in the Wnt signaling pathway that artificially increase signaling activity. [3-4] Hence, it is necessary to control the cancer cell proliferation through a important signaling pathway. The Wnt signaling pathway is a network of proteins best known for their roles in embryogenesis and cancer. The Wnt pathway involves a large number of proteins that can regulate the production of Wnt signaling molecules, their interactions with receptors on target cells and the physiological responses of target cells that result from the exposure of cells to the extracellular Wnt ligands. Numerous studies suggest that activation of the Wnt/β-catenin signaling pathway plays an important role in human tumorigenesis. [5-7]

Wnts play important roles in cell fate specification, tissue patterning, and control of asymmetric cell division. The expression of Wnt genes is developmentally regulated in a coordinated temporal and spatial manner. [8] The following Fig. 1 shows the WNT signaling pathway.

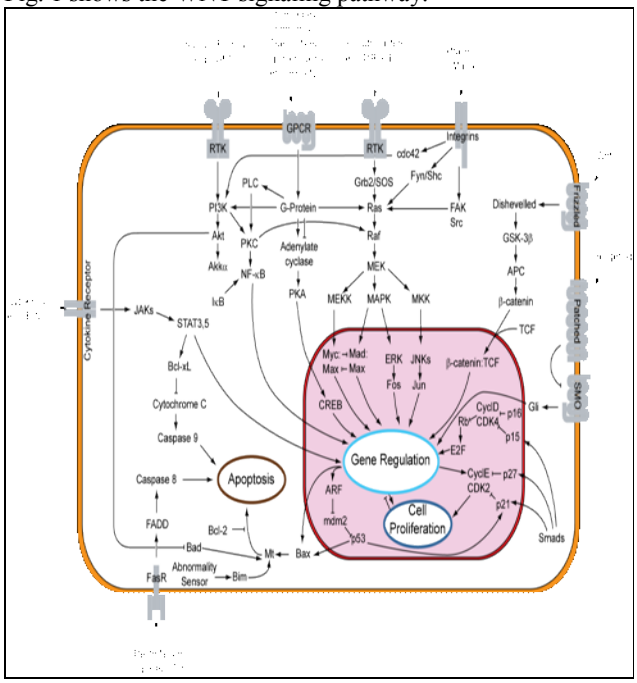

Fig. 1: WNT signaling pathway

Protein kinase C-alpha (PKC-alpha) is a calcium- and phospholipid dependent serine-threonine protein kinase of

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fundamental importance in signal transduction and growth regulation and act as a negative regulator of Wnt/beta-catenin signaling pathway. On other hand, PKC-alpha activation repressed the expression of cyclin D1 and c-myc, which are known beta-catenin target genes; it regulates colon cancer cell proliferation via beta-catenin phosphorylation/down-regulation and may facilitate the development of new strategies to treatment of colon cancer. [9] The targeting of Protein kinase C-alpha (PKC-alpha) is the one of the important drug target in colorectal cancer using Pharmacophore based drug designing .Many current study in natural products contain phytochemical constitute such as terpnoids, flavoniods. In particular, Flavanols are used for treating the various cancers on other way the chemotherapeutic drugs such as fluorouracil, capecitabine, UFT, leucovorin, irinotecan, or oxaliplatin. However, it these drugs cause more side effects to the human body. Hence, it is more important to look for better alternative compound for the treating the cancer using natural compounds.

MATERIALS AND METHODS

Retrieval of protein and ligand from database

The structure of the drug target protein PKC-alpha and its X-ray crystallography structure with 2.80Å was retrieved from protein data bank with its Identification number as 3IW4, commonly known as PDB ID is complexed with selective potent inhibitor 3-(1H-indol-3-yl)-4-[2-(4-methylpiperazin-1-yl)quinazolin-4-yl]-1H-pyrrole-2,5-dione and the structure of flavoniod Epicatechin was retrieved from Pubchem compound database.

Protein preparation

The raw protein from protein databank with PDB ID 3IW4 named Protein kinase C alpha (PKC-alpha) is further prepared for docking studies initially, all the Hetatms were removed and subsequently subjected for energy minimization to remove the bad steric clashes using tool smart minimizer for 1000 steps at RMS gradient of 0.1 and 0.03 respectively by applying the suitable force field CHARMm available through Accelrys life science software. [10]

Common feature Pharmacophore generation

Common feature pharmacophore are generated using the HipHop algorithm. Hip Hop identifies configurations or three-dimensional spatial arrangements of chemical features that are common to molecules in a training set. The configurations are identified by a pruned exhaustive search, starting with small sets of features and extending them until no larger common configuration is found. This option allows broader and more diverse pharmacophores to be generated. The resultant pharmacophores are ranked as they are built. The ranking is a measure of how well the molecules map onto the proposed pharmacophores, as well as the rarity of the pharmacophore model.

Pharmacophore feature based screening

The best Pharmacophore resultant from the Hiphop algorithm based on fit value was taken and its respective Pharmacophore based analogs are generated using search 3D minimaybridge database available in accelrys

Receptor- Ligand interaction

The molecular level interaction between protein and generated analogs were taken for docking studies using Ligand fit is Docking program available with DS2.5. [11] The largest cavity of protein named as Site 1 was selected consisting of 2116 points and partition level was set to and proceeded for docking with Number of Monte Carlo steps was set to "2 500 120, 4 1200 300, 6 1500 350, 10 2000 500,253000 750" with maximum 10 number of poses.

RESULTS AND DISCUSSION

Protein preparation

The protein with PDB id 3IW4 was initially prepared by applying the appropriate force field to avoid the steric clashes and non bonded interaction as shown in the (Fig. 2) followed by defining the appropriate binding site for the lead molecule to go and bind with the active site of the drug target protein, the site of the protein is predicted in automatically mode by using Flood filling algorithm as shown in the (Fig. 3).

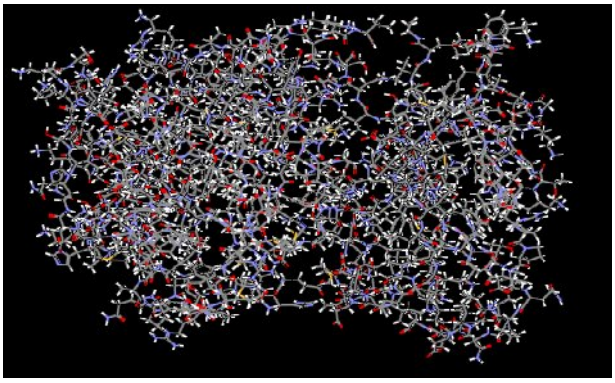

Fig. 2: 3IW4 Protein after applying force field

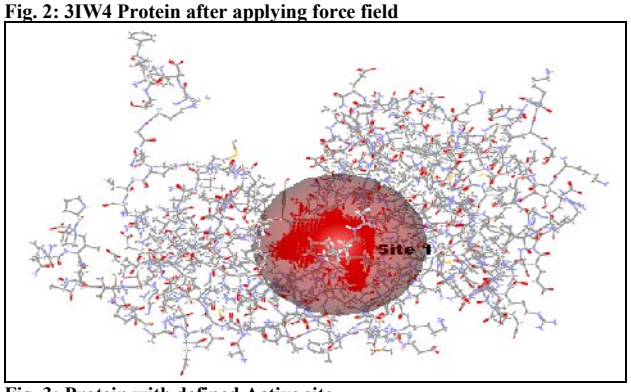

Fig. 3: Protein with defined Active site

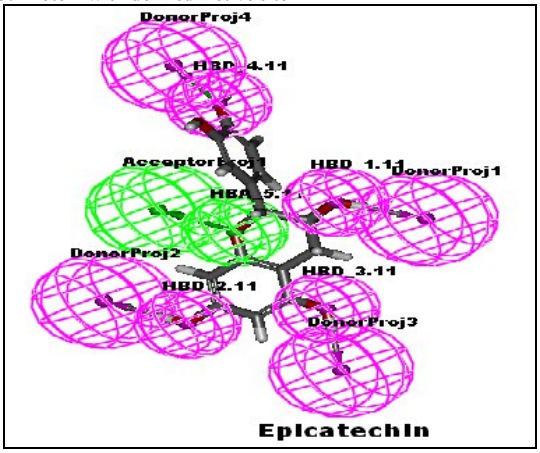

Fig. 4: The common Pharmacophore for the natural compound Epicatechin

Qualitative Pharmacophore generation

A pharmacophore is the ensemble of steric and electronic features necessary to ensure the optimal supramolecular interactions with a specific biological target structure, and to

trigger (or to block) its biological response. In this current study the chemical features natural compound epicathein were generated using the catalyst- Hip-Hop algorithm was shown in the (Fig. 4).

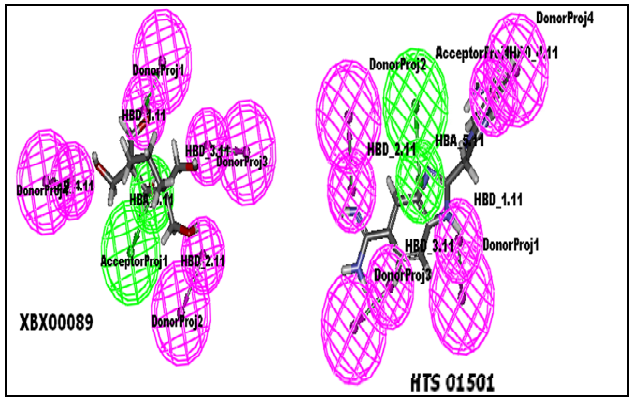

Fig. 5: Identical molecules of epicathechin shows Four Hydrogen bond donor and one acceptor with its respective projection

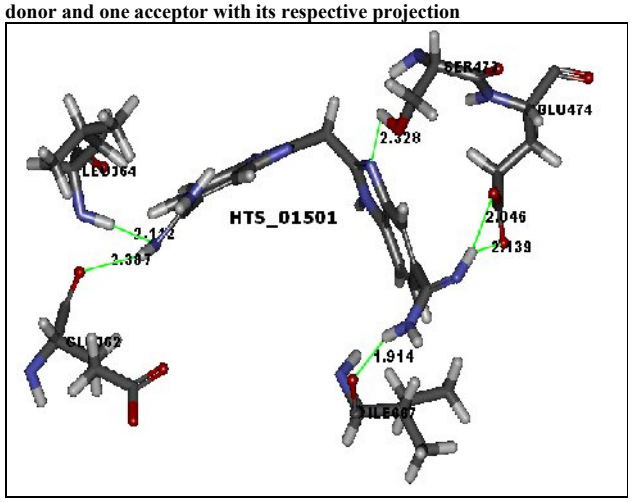

Fig. 6: Binding of HTS01501 with active site amino acid

Table 1: Dock score of the HTS01501 along with amino acid interaction and nature of bonding

Molecule	Amino acid	Distance in Å	Nature of Bonding	Dock score
HTS01501	LEU364	2.112	Hydrogen	53.911
	GLU362	2.362	Hydrogen	
	SER473	2.328	Hydrogen	
	GLU474	2.046 2.139	Hydrogen	
	ILE667	1.914	Hydrogen	

The first Pharmacophore of Epicatechin was taken to generate derivatives of Epicatechin using the search 3D minimaybridge commercially available database, which is integrated in the accelrys software. The (Fig. 5) shows the identical Pharmacophore based of molecules of Epicatechin with fit value of more than 80 % compared with original Pharmacophore.

Receptor –Ligand interaction

The pharmacophores, which is identical to Epicatechin such as XBX0089 AND HTS01501, were successfully docked to the active of the drug target Protein kinase C alpha. The docking results were tabulated and summarized in (Table 1)with the nature of molecular interaction between drug target receptor and leads .Among two leads HTS01501 shows more binding affinity with PKC-alpha receptor with binding energy value of -3.514 Kcal/mol, Whereas XBX00089 shows positive score binding energy value of about 14.575 Kcal/mol. Hence, molecule with lower binding energy is favorable for screening the molecules using Insilco method based on recptor-ligand interaction concept of virtual screening binding affinity between receptor .the interactions of HTS01501 with PKC-alpha was shown in the (Fig. 6)

Finally, this study reveals that natural compound Epicatechin found in the various medicinal plants serve as good candidate for treating the cancer ,on the similar way the Epicatechin derivatives which possess similar chemical features as like as Epicatechin can also be potential drug candidate against Protein kinase C alpha . Between the two compounds examined in this study, HTS01501 posses the highest dock score with good interaction potential, whereas compound XBX00089 has the highest internal energy with no bonding with active site due to positive internal energy. Hence, HTS01501 is chemically resembles as like as Epicatechin can be better drug candidate for treating the Colon cancers in future.

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REFERENCES

1. Bray F, Ren JS, Masuyer E, Ferlay J. Estimates of global cancer prevalence in 2008.
2. Brunetto de Farias C, Rosemberg DB, Heinen TE, Koehler-Santos P, Abujamra AL, Kapczinski F, Brunetto AL, Ashton-Prolla P, Meurer L, Bogo MR, Damin DC, Schwartsmann G, Roesler R. BDNF/TrkB Content and Interaction with Gastrin-Releasing Peptide Receptor Blockade in Colorectal Cancer. Oncology 2010; 79: 430
3. Ionov Y, Peinado MA, Malkhosyan S, Shibata D, Perucho M. Ubiquitous somatic mutations in simple repeated sequences reveals a new mechanism for colonic carcinogenesis. Nature 1993; 363: 558-561.
4. Chakravarthi S, Krishnan B, Madhavan M. Apoptosis, and expression of p53 in colorectal neoplasms". Indian J Med Res. 1999; 86: 95-102.
5. Polakis P. The oncogenic activation of beta-catenin. Curr. Opin. Genet. Dev. 1999; 9, 15-21.
6. Waltzer L, Bienz M. The control of beta-catenin and TCF during embryonic development and cancer. Cancer Metastasis Rev. 1999; 18: 231-246.
7. Behrens J. Control of beta-catenin signaling in tumor development. Ann N Y Acad Sci. 2000; 910:21-33.
8. Luu HH, Zhang R, Haydon R, Rayburn E, Kang Q, Si W, Park JK, Wang H, Peng Y, Jiang W, Tong-Chuan H. Wnt/ß-Catenin Signaling Pathway as Novel Cancer Drug Targets. Current Cancer Drug Targets 2004; 4:653-671.
9. Hanauske AR, Sundell K, Lahn M. The role of protein kinase C-alpha (PKC-alpha) in cancer and its modulation by the novel PKC-alpha-specific inhibitor aprinocarsen. Curr Pharm Des. 2004; 10:1923-36.
10. Momany FA, Rone RJ. Validation of the general purpose QUANTA 3.2/CHARMm force field. Comp. Chem. 1992; 13:888-900.
11. Venkatachalam CM, Jiang X, Oldfield T, Waldman M. Ligand Fit: a novel method for the shape-directed rapid docking of ligands to protein active sites. J. Mol. Graph. Model. 2003; 21:289-307.