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Computational Approach on The Drug Affinity Studies of Substituted (4-Aminophenyl) Benzothiazole Derivatives Against Colon Cancer.

Teenu James, Arya Rajendran, Shilpa Martin, Reshma Easo, Navneeth C Krishnan, Asha Asokan Manakadan, and Saranya TS*.

Department of Pharmaceutical Chemistry & Analysis, Amrita School of Pharmacy, Kochi – 682041, Amrita Vishwa Vidyapeetham, Amrita University, AIMS Health Science Campus, Ponekkara P. O., Kerala, India

ABSTRACT

Colon cancer is the growth arising from the inner wall of colon, which is a part of large intestine. Wnt-signalling pathway has been recognized as a crucial factor in carcinogenesis. Mutations in this pathway lead to the accumulation of beta –catenin which initiates oncogenesis. Thus beta catenin is selected as the target to resist against the uncontrolled growth. In this work, a total of 10 substituted (4-aminophenyl) benzothiazole derivatives were selected for docking studies. These molecules were selected based on their potent antitumor, antifungal, antimicrobial, anthelmintic.antidiabetic, and anticonvulsant, anti-inflammatory and antimalarial properties. The primary and secondary characterizations of protein(PDB ID:3SLA) were achieved from pdb structures using the aid of protparam and sopma tools.Insilico docking analysis were carried out, using Argus lab version 4.0 based on their scoring functions and Lipinski rule of 5 .It revealed that these derivatives have shown better docking score than the standard drug RALTITREXED; hence, can be used to reduce one of its side effect inflammation , and showed strong anticancer activity by the suppression of beta catenin, thus making them possible inhibitors against colon carcinogenesis. The main objective of our research work is to conclude that substituted (4-aminophenyl) benzothiazole derivatives are better drugs than RALTITREXED as it shows higher binding energy with the modeled protein. So these substituted derivatives can be used as suitable drug of choice against colon cancer.

Keywords: Benzothiazole, beta- catenin, Argus lab, RALTITREXED.

Corresponding author

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INTRODUCTION

Cancer is a condition of uncontrolled or abnormal growth or cell proliferation. Colon cancer is a condition where the cancer starts from the inner lining of the colon which is a part of large intestine. It is the third most common type of cancer and second most death causing one. Most of the colon cancers start as a polyp in the colon which will further develop to cancerous cells and cause death ; but all the polyps may not be cancerous. Differentiation into cancer cells depends upon the kind of the polyp formed. There are 2 types of polyps:

- Adenomatous polyps (adenomas)[1] : since these can change into cancer cells , adenomas are known as pre-cancerous condition.
- Hyperplastic polyps[1] and inflammatory polyps: these are common but not pre-cancerous.

Colon cancer starts from the lining of the colon;they further move to the lymph nodes and then to the distant areas of the body and become lethal. Colon cancer has been diagnosed in about 48,400 men .Some of these people were cancer-free, while others still had evidence of cancer and may have been undergoing treatment. There is an increased risk of colon cancer in low income countries and a significantly higher proportion of early- onset cancers. There is a rising incidence of colon cancer in young adults from diverse geographic and ethnic backgrounds which could be linked to environmental pollution or lifestyle factors such as obesity,physical inactivity and a diet rich in processed foods.[2]

Wnt signalling pathway has been recognized as playing an important role in the carcinogenesis. Activation of this pathway initiates the inhibitory activities of the destruction complex (APC, PP2A, GSK3, CK1alpha) that blocks beta-catenin [3] . Cytosolic beta-catenin then enters the nucleus to activate TCF/LEF transcriptional response elements [4] binding the DNA, which instigates the regulation of numerous genes, each contributing to over expression of beta catenin that results in mutations thereby causing tumors .

Signs of colon cancer includes:nausea, vomiting, loss of appetite, weight loss, constipation etc. Rectal bleeding and anaemic conditions can be seen in 50+aged persons.[5] . The main cause of this condition is the non- periodic life style of the people. Smoking, obesity, alcoholism[6] ,increased fat consumption, un-exercised life will lead to diseased state. People with high degree relatives (parents or siblings)will have a chance of genetical transmission of disease. Among the 4 stages of colon cancer, 3rd is the lethal condition stage.[7]

From the literatures it was found that benzothiazole derivatives play a major role in designing of new drugs since they have an interesting pharmacological profile including anti-allergic,anti- inflammatory, anti- tumor, analgesic, antimicrobial, anthelmintic, anti-leishimanial, anti-convulsant activities .It has various other chemical and biological activity.[8]

Molecular docking [9]is a simulation method that is broadly used in modern drug design, predicts the conformation of a receptor-ligand complex, in which the receptor can be either a protein or a nucleic acid, and the ligand is a molecule. This computer simulation can generate many possible positions for the ligand in the protein-binding pocket. Docking is frequently used to predict the binding orientation of small drug candidates to their protein targets which in turn predict the affinity and activity of the chemical molecule.

The main objective of our research work was to compare docking scores of substituted benzothiazole derivatives with the standard drug on the basis of Lipinski rule of five and their docking studies carried out by using Argus lab 4.0. and thus obtaining a primary idea on the affinity and activity of the respective chemical compound.

MATERIALS AND METHODS

DATA BASE :

PROTEIN DATA BANK (PDB)

Protein data bank is database designed specificaly to store information about the three dimensional

stuctural datas of complex and large molecules such as proteins .PDB format is used for the primary and secondary analysis of the protein and also for further docking studies. PDB has a large array of protien information and identification from where the specific drug target was selected.

DOCKING TOOLS AND SOFTWARES:

PROTPARAM:

Expasy Protparam [10] is a server used for primary characterization by calculating various physiochemical parameters of a protein using the protein sequence. The parameters gravy [11], aliphatic index [12]and instability index.[13] , extinction coefficient [14],half life were computed using the server.

SOPMA

Sopma is a software tool used for prediction of the secondary characterization of proteins and nucleic acid sequences which constitute the formation of protein structures such as alpha helices, beta strands, random coil and Pi helices. Based on all these parameters the appropriate protein was selected as the target site. Using castp software number of target sites were found out.

CASTp

Castp is known as Computer Atlas Of Surface Topography of Proteins which is a web server. It is a bioinformatic tool developed to identify the binding sites and interior voids which is a characteristic feature of every proteins,based on which whose binding activities can be screened. This pocket detection tool uses the theories like alpha shape and pocket algorithm of computational geometry to identify the target sites. Castp measures the area of binding site,volume of the void, accessibility to bind for different ligands.

EVALUATION OF DRUG-LIKENESS:

MOLINSPIRATION

Molinspiration is a software for determination of drug likeness based on factors like milogp, molecular weight ,nOHNH,nON,nviolations and bioavailability of the compoud by the application of Lipinski rule of five [15]. The selection of the ligand was done using
molinspiration.

The online softwares like corina,Swiss pdbv,chemsketch provided the three- dimensional structures for the further process involved in computational chemistry. ARGUSLAB , was an important machinery in developing a proper binding of the target protein with the ligand which is the aim of our study . Finally, docking was carried out and appropriate ligands were selected which gave an idea about the drug showing less side effect and maximal potency based on the docking scores obtained.

RESULT AND DISCUSSION

The protein beta catenin was selected from the destruction complex of the Wnt signaling pathway which is involved in the colon carcinogenesis.From pdb format sequence, the primary and secondary characterization of beta catenin were carried out by considering certain parametrers that has mentioned below. From table 1.1 and 1.2, the protien was selected with reference to their negative gravy value, high half life, high aliphatic index , high instability index and high random coil . The drug likeness of the benzothiazole derivatives which possess a high anti-tumour activity were calculated on the basis of Lipinski rule of 5 using molinspiration. Substituted 4-amino phenyl benzothiazole derivatives and substituted hydroxy benzothiazole derivatives were identified, showing better druglikeness.The 10 ligands were docked with the protein having PDB ID :3SLA and calculated the docking scores .

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TABLE 1.1 PROTPARAM OF BETA CATENIN

PDB ID	NO:OF AMINO ACID	MOLECULAR WEIGHT	THEORECTAL pI	HALF LIFE	INSTABILITY INDEX	ALIPHATIC INDEX	GRAVY	EXTINCTION COEFFICIENT
4DJS	518	56430.7	8.59	5.5hrs	45.31	110.54	0.096	29045
3SL9	55	6165.7	4.49	30hrs	51.11	65.64	-1.016	
3SLA	168	18315.3	8.96	30hrs	45.62	107.44	0.073	4595
3TX7	527	57449.8	8.43	1.9hrs	44.34	110.32	0.083	30535
3DIW	124	13722.6	8.05	30hrs	59.68	85.56	-0.448	8480
2Z6H	644	70337.8	6.03	1.4hrs	42.87	100.75	-0.084	48985
2GL7	550	60167.8	8.23	1.9hrs	45.66	108.36	0.021	30535
2G57	28	2964.6	6.92		49.46	52.50	-0.518	6990
1T08	519	56515.8	8.59	4.4hrs	44.87	110.52	0.099	29045
1TH1	532	58026.4	8.44	1.3hrs	44.38	110.56	-0.083	30535
1QZ7	533	58141.5	8.26	1.3hrs	44.68	110.36	0.076	30535
1P22	435	49629.7	8.44	1.9hrs	45.77	91.20	-0.279	86245
1LUJ	514	56002.2	8.81	7.2hrs	44.83	110.45	0.099	29045
1JDH	529	57632	8.44	4.4hrs	44.21	111.19	0.0103	30535
1JPW	540	58920.4	7.99	30hrs	44.64	108.93	0.058	32025
1G3J	532	58026.4	8.44	1.3hrs	44.38	110.56	0.083	30535

The protein 3SLA was selected from table 1.1 based on the highest half life, highest negative GRAVY value and lowest extinction coefficient on the comparison basis with other proteins.

TABLE 1.2 SOPMA OF BETA CATENIN

PDB ID	ALPHA HELIX	Pi HELIX	BETA TURN	RANDOM COIL
4DJS	0	0	0	10
3SL9	7	0	3	2
3SLA	20	0		19
3TX7	117	0	15	22
3DIW	305	0	57	94
2Z6H	341	0	69	150
2GL7	320	0	57	102
2G57	6	0	4	13
1T08	302	0	57	91
1THI	305	0	57	96
1QZ7	305	0	58	96
1P22	120	0	57	112
1LUJ	295	0	56	94
1JBH	303	0	56	96
1JPW	304	0	58	104
1G3J	305	0	57	96

The table 1.2 was useful to select the protein based on the value of random coil obtained with the help of sopma. The highest value of random coil was selected taking into consideration the other parameters which is already obtained from the table 1.1 of protparam. Using all these information we selected the protein 3SLA.

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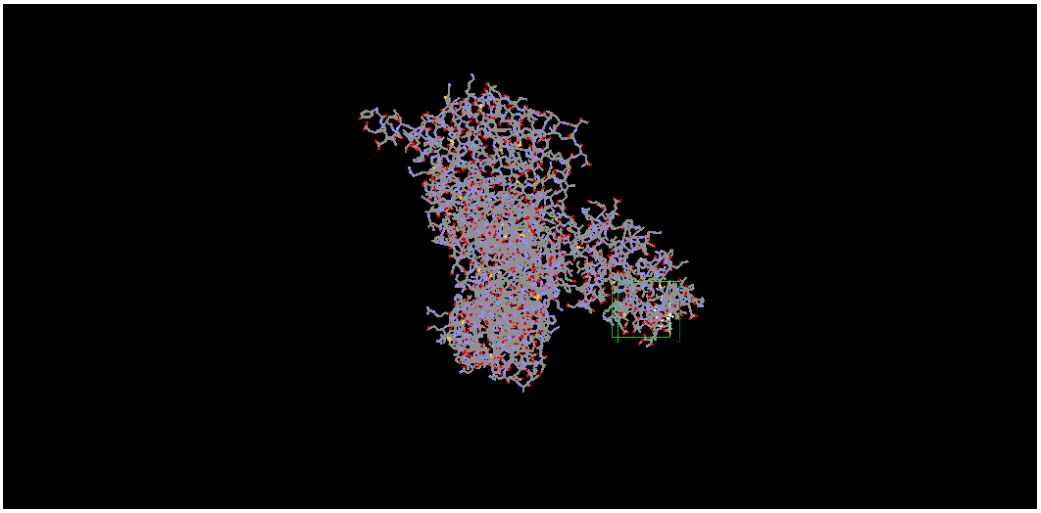

TABLE 1.3 DOCKING SCOREOF BETA CATENIN (PDB ID:3SLA)

SL NO:	LIGANDS	DOCKING SCORE(KCAL/MOLE)
1	2-(4-AMINOPHENYL)BENZOTHIAZOLE	-7.22035
2	4-(4-AMINOPHENYL)BENZOTHIAZOLE	-7.46144
3	5-(4-AMINOPHENYL)BENZOTHIAZOLE	-7.67075
4	6-(4-AMINOPHENYL)BENZOTHIAZOLE	-7.59074
5	7-(4-AMINOPHENYL)BENZOTHIAZOLE	-7.97738
6	2- HYDROXY BENZOTHIAZOLE	-6.72232
7	4- HYDROXY BENZOTHIAZOLE	-6.73926
8	5- HYDROXY BENZOTHIAZOLE	-6.58864
9	6- HYDROXY BENZOTHIAZOLE	-6.689
10	7- HYDROXY BENZOTHIAZOLE	-6.55215
11	RALTITREXED(STANDARD)	-7.0825

The compound 7-(4-aminophenyl) benzothiazole was identified to have the highest docking score when compared with the standard drug RALTITREXED.

CONCLUSION

The objective of the work was to emphasize the binding affinity of target and the chemical compound, thereby arrived at a primary conclusion that these benzothiazole derivatives(ligand) showed more anti- tumour activity with less inflammatory side effect than standard drug RALTITREXED.

Docking studies[16-17] were carried out using Argus lab 4.0 to study the mode of binding of the target protein beta catenin with ligand-benzothiazole derivatives. From table 1.0 , it is clear that the most promising target protein that can be selected is PDB ID:3SLA ,for it has the suitable parameters that is already mentioned above. The ligand benzothiazole was substituted with different functional groups like hydroxy, amino, and halo groups. Out of these,4-aminophenyl and hydroxy substituted benzothiazole derivaties had good drug likeness satisfying the Lipinskii rule of five which is aided by molinspiration technique. These substitued derivatives was then docked with the target protein betacatenin (PDB ID:3SLA) to obtain the binding energy. 2(4-aminophenyl) benzothiazole, 4 (4-aminophenyl)benzothiazole, 5(4-aminophenyl)benzothiazole,6(4-aminophenyl) benzothiazole, and 7(4-aminophenyl)benzothiazole possess a better docking scores which was concluded from table 1.3 ;out of it 7(4-aminophenyl)benzothiazole have shown higher docking score when compared to the standard drug RALTITREXED. Hence, the ligand 7(4-aminophenyl) benzothiazole can be considered as a good anticancer agent whose potency and the activity can be well confirmed on other invitro and in vivo studies which can lead to the development of drugs having more potent pharmacological activities with less side effects for treating colon carcinogenesis.

REFERENCES

[1] American Cancer Society, Cancer Facts and Figures 2010. Atlanta, American cance society; 2010.

[2] American Cancer Society, Colorectal Cancer Facts and Figures 2014-2016. Atlanta, American cancer society;2014.
[3] MacDonald, Tamai K. Wnt/Beta-Catenin Signaling: Components, Mechanisms and Diseases. 2009; 17:9–26.
[4] Markowitz SD, Bertagnolli MM. Molecular Origins of Cancer: Molecular Basis of Colorectal Cancer. 2009; 2449–60.
[5] Astin M, Griffin T. Neal, RD. Rose, P. Hamilton. The Diagnostic Value of Symptoms for Colorectal Cancer In Primary Care: A Systematic Review. The British Journal of General Practice. 2011; 61(586): 231–43.
[6] Fedirko V.Tramacere, I. Bagnardi, V. Alcohol Drinking and Colorectal Cancer Risk: An Overall and Dose-Response Meta-Analysis of Published Studies. Annals of Oncology 2011.;22 (9): 1958–72.
[7] American Cancer Society, Colorectal Cancer Facts and Figures 2014-2016. Atlanta American cancer society; 2014.
[8] Rajinder Kumar ,Uday Kalindh, Amendeep Kaur. Benzothiazole Derivatives and its Biological Activities; A Review .2012 ;3:167.
[9] Saoussen lakehal ,Fouad Ferkous,Khairedine Kraim ; Molecular Docking Study on Xanthone Derivatives Towards Alpha-Glucosidase . 2016; 3:11.
[10] Wilkins MR et al. Methods of Molecular Biology 1999 ;112: 531.
[11] Kyte J & Doolittle RF. Journal of Molecular Biology. 1982 ;157: 105.
[12] Ikai A. Journal of Biochemistry. 1980; 88: 1895.
[13] Guruprasad K et al. Protein Engineering. 1990 ; 4:125
[14] Gill SC & von Hippel PH. Anal ysis of Biochemistry. 1989 ;182: 319.
[15] Lipinski CA. Lead- And Drug-Like Compounds: The Rule Of Five Revolution. 2004; (4): 337–341.
[16] Research Journal Of Pharmacy And Technology Year:2015, Volume:8, Issue:12 First page : (1673) Last page : (1678) In silico Docking Approach of Coumarin Derivatives as an Aromatase Antagonist Thomas Reshma, Hari R., Joy Josna, Krishnan Saranya, Swathy A.N., Nair Sruthy S., Manakadan Asha Asokan, Sathianarayanan, Saranya T.S.
[17] In silico design, synthesis and in vitro studies of some novel 4-phenyl-4Hchromene derivatives as antioxidant and anti-inflammatory agents Subin Mary Zachariah*, Reshmi K. P., Vidya Viswanad, Nithya P. S., Fahina Beegum S. C. and Rajan V. K.,7(7) 2015 ,32-40.