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Catechin Derivatives as Inhibitors of Programmed Cell Death 1 Receptor (Pd –1), a Predominant Factor of *Homo Sapiens* in the Development of Oral Squamous Cell Cancer

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Abstract

One of several mechanisms of tumour-mediated immune suppression is the expression of co-inhibitory molecules by tumour. Upon engagement to their ligands these molecules can suppress effector lymphocytes in the periphery and in the tumour microenvironment. The PD-1 is one of the central signalling molecules that may inhibit T cell immunity when bound to its ligands (PD-L1 or PD-L2) by inducing T cell apoptosis and anergy. PD- L1 expression reduces the number of T cells in human oral squamous cell carcinoma. Further it has been suggested that the development of a strategy to block the interactions of PD-L1 with PD-1 would be a useful tool for inhibiting tumour growth. The three-dimensional structure of GTF-SI was retrieved from RCSB database. The possible binding sites of PD-1 were searched using binding site prediction 3DLIGANDSITE, an online tool. A total of 500 ligands in 2D format were generated from the basic structure of catechin with the help of software ACD chemsketch. Rapid virtual screenings of these compounds were performed in the docking tool iGEMDOCK v2.0. The molecular docking of ligands was performed using AutoDock 4.0 software. In the present study, (2S, 3R)-2-(3,4-dihydroxyphenyl)-3,4-dihydro-2H-chromene-3,5,7-triol has found to have very good inhibitory property based on molecular docking study. Further the compound shows a good ADMET properties based on studies in OSIRIS. Hence it is concluded that (2S,3R)-2-(3,4-dihydroxyphenyl)-3,4-dihydro-2H-chromene-3,5,7-triol is an excellent drug candidate in the control of oral squamous cell carcinoma.

Keywords: *Programmed death-1, oral squamous cell carcinoma, catechin derivaties, molecular docking, AutoDock.*

1. Introduction

One of several mechanisms of tumour-mediated immune suppression is the expression of co-inhibitory molecules by tumour. Upon engagement to their ligands these molecules can suppress effector lymphocytes in the periphery and in the tumour microenvironment[1, 2].

The PD-1 is one of the central signalling molecules that may inhibit T cell immunity when bound to its ligands (PD-L1 or PD-L2) by inducing T cell apoptosis and anergy[3]. Programmed death-1 (PD-1) is an immunoglobulin superfamily member related to CD28 and CTLA-4. PD-1 is induced on T cells, B cells, and monocytes on activation. Several evidences indicate that PD-1 plays a crucial role in regulating peripheral tolerance and autoimmunity[4, 5].

PD-1 has two ligands: PD-1 ligand 1 (PD-L1; B7-H1) and PD-1 ligand 2 (PD-L2; B7-dendritic cell) [6, 7]. PD-L1 and PDL2 are involved in the negative regulation of cellular and humoral immune responses by engaging PD-1 receptor[8].Recent studies have also shown that the PD-L/PD-1 pathway might play critical roles in tumour immunity [9, 10]. PD-L1 on tumours or antigen-presenting cells in tumour environment has been proposed to promote tumour growth and induce apoptosis of tumour-reactive T cells expressing PD-1 [11].

Oral cancer ranks from the sixth to eighth most common cancer around the world, with a great variability in incidence among countries[12]. In the oral cavity, the squamous cell carcinoma is more prevalent than the other types of cancer[13]. It has been recently shown that PD-L1 expression reduces

the number of T cells in human oral squamous cell carcinoma. Further it has been suggested that the development of a strategy to block the interactions of PD-L1 with PD-1 would be a useful tool for inhibiting tumour growth[14].

It has been found that tea catechins and related polyphenols have anti-cancer activity[15]. Hence an attempt has been made in this study to evaluate catechin and its derivates as the inhibitors of PD- 1 by molecular docking method.

2. Materials And Methods

Protein preparation

The three-dimensional structure of GTF-SI was retrieved from RCSB database (http://www.rcsb.org/pdb/explore/explore.do?structureId=2m2d)[16]. Its RCSB code is 2M2D.

Active site prediction

The possible binding sites of PD-1 were searched using binding site prediction 3DLIGANDSITE, an online tool (http://www.ncbi.nlm.nih.gov/pubmed/20513649)[17]. binding site thus obtained was selected for this study.

Generation and optimization of Ligand

The structure of catechin (Figure 1) was obtained from pubchem database. Its compound ID is 9064. Its IUPAC name is (2R,3S)-2- (3,4-dihydroxyphenyl)-3,4-dihydro-2H-chromene-3,5,7-triol. The catechin has a molecular weight of 290.3 and its xlogP value is 0.4. A total of 500 ligands in 2D format were generated from the basic structure of catechin with the help of software ACD chemsketch[18]. The ligands were saved in mol 2 format. The OPEN BABEL software (www.vcclab.org/lab/babel/start.html) was used to convert mol format to pdb format. Rapid virtual screenings of these compounds were performed in the docking tool iGEMDOCK v2.0[19]. A population size of 150 is set with 70 generation and one solution for quick docking. The ligands with low binding energy were selected for the further study. The selected ligands were then analyzed for drug- relevant properties based on "Lipinski's rule of five" and other drug like properties using OSIRIS Property Explorer (http://www.organicchemistry.org/prog/peo/), Mol soft: Drug- Likeness and molecular property explorer (http://www.molsoft.com/mprop/). On the basis of binding affinity and drug like properties, all these ligands were taken for further molecular docking study.

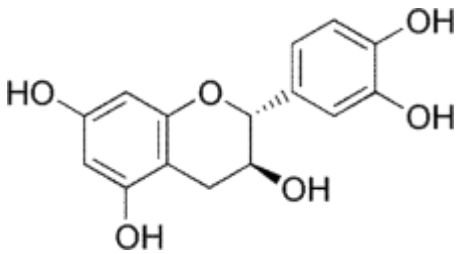

Figure 1: Structure of catechin

Protein-ligand docking

The docking of ligands was performed using AutoDock 4.0 software. Docking was performed to obtain a population of possible conformations and orientations for the ligands at the binding site and also its binding energy. Using the software, polar hydrogen atoms were added to the PD-1 and its non-polar hydrogen atoms were merged. All bonds of ligands were set to be rotatable. All calculations for protein-ligand flexible docking were done using the Lamarckian Genetic Algorithm (LGA) method. The grid box with a dimension of 126 x 126 x 126 points was used so as to cover the entire enzyme binding site and accommodate ligands to move freely. The best conformation was chosen with the lowest docked energy, after the docking search was completed.

3. Results and Discussion

The 3D structure of PD-1 is shown Figure 2. It is made up of 233 amino acids. Alpha helices are coloured magenta, beta sheets are coloured yellow, turns are coloured pale blue, and all other residues are coloured white.

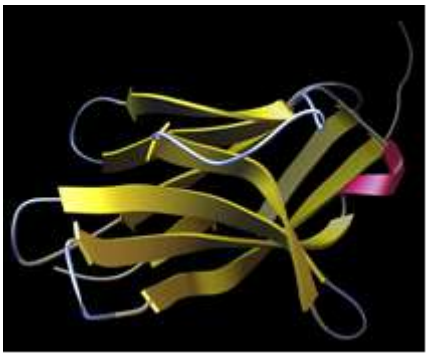

Figure 2: The 3D structure of PD-1 viewed with Rasmol structure colour scheme

Its binding site was predicted using 3DLIGANDSITE. The binding sites predicted are 62 SER, 63 PHE, 128 LEU, 129 ALA, and 130 PRO. The Figure 3 shows the 3D structure of PD-1 protein displaying its binding sites.

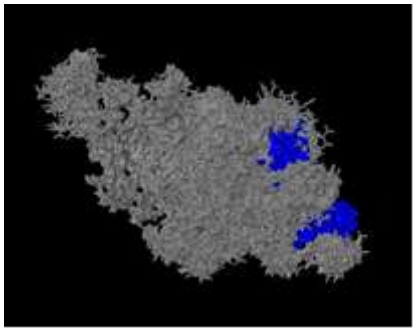

Figure 3: The 3D structure of PD-1 showing its binding site

A total of 500 ligands were derived from catechin using ACD chemsketch software. It was converted to pdb format using OPEN BABEL software. On virtual rapid screening with iGEMDOCK software, four compounds were found to have good fit with a low binding energy.

The Table 1 displays the results obtained in rapid virtual screening by iGEMDOCK of the four ligands. From the table it is clear that the four ligands have low total binding energies and thus were taken to further docking studies. Their docking pose is shown in Figure 4. The structure and the IUPAC name of the four ligands were shown in the Figure 5.

Table 1: The results of iGEMDOCK showing binding energies of four selected ligands

S. No.	Ligand	Total binding Energy (kcal/mol)	Vanderwaals force	H Bond	Electrostatic bond
1.	(2S,3S)-2-(3,4-dihydroxyphenyl) -3-methyl-3,4-dihydro-2H-chromene-5,7-diol	-80.2977	-68.7688	-11.529	0
2.	(2S)-2-(3,4-dihydroxyphenyl) -3,4-dihydro-2H-chromene-5,7-diol	-84.2311	-58.7018	-25.5293	0
3.	(2S,3R)-2-(3,4-dihydroxyphenyl) -3,4-dihydro-2H-chromene-3,5,7-triol	-79.8576	-55.5792	-24.2785	0
4.	(2R)-2-(4-hydroxy-3-methoxyphenyl) -6-methoxy-3,4-dihydro-2H- chromen-7-ol	-80.9772	-72.8105	-8.16669	0

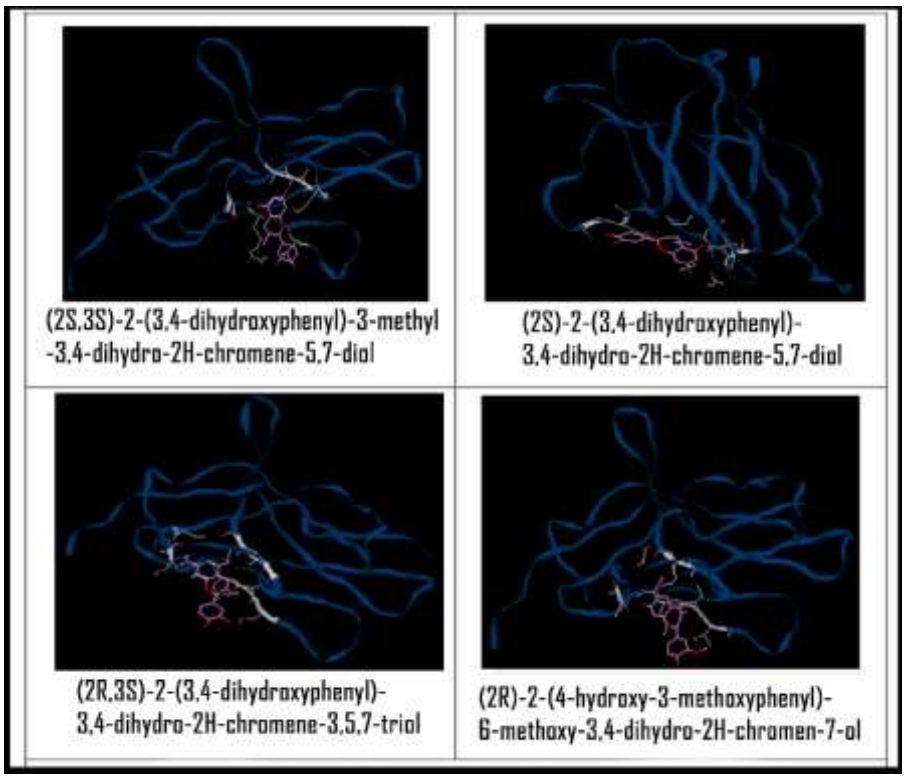

Figure 4: Docking pose of the four ligands with PD-1

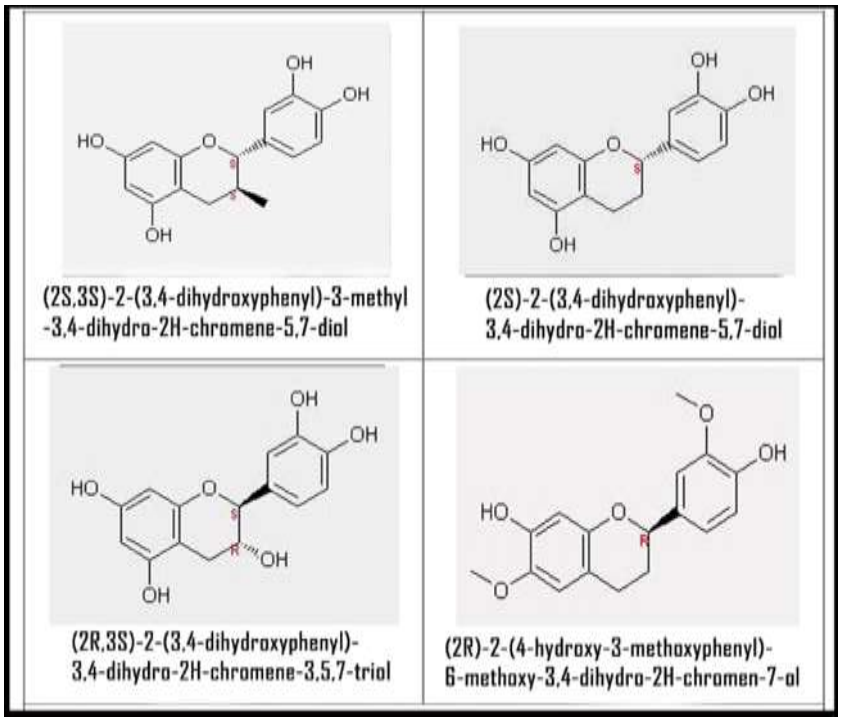

Figure 5: The structure and IUPAC name of catechin derivatives

The four ligands were subjected for its ADMET properties. The Table 2 depicts the values related to the Lipinski's rule of Five. From the table it is evident that all the four selected ligands obey the rule. The Table 3 shows the drug relevant properties of the four ligands. They all possess good drug score and drug likeness.

Table 2: The Lipinski's properties of the selected four ligands

S. No.	Ligand	Molecular weight	Xlog p	H bond donor	H bond acceptor
1.	(2S,3S)-2-(3,4-dihydroxyphenyl) -3-methyl-3,4-dihydro-2H-chromene-5,7- diol	288.299	2.76	4	5
2.	(2S)-2-(3,4-dihydroxyphenyl) -3,4-dihydro-2H-chromene-5,7-diol	274.272	2.531	4	5

3.	(2S,3R)-2-(3,4-dihydroxyphenyl) -3,4-dihydro-2H-chromene-3,5,7-triol	290.271	1.369	5	6
4.	(2R)-2-(4-hydroxy-3-methoxyphenyl) -6-methoxy-3,4-dihydro-2H-chromen-7-ol	302.326	2.741	2	5

Table 3: The drug relevant properties of selected four ligands

S. No.	Ligand	Drug likeness	Drug score	Mutagenic	Tumorigenic	Irritant
1.	(2S,3S)-2-(3,4-dihydroxyphenyl)-3-methyl-3,4-dihydro-2H-chromene- 5,7-diol	1.74	0.82	No	No	No
2.	(2S)-2-(3,4-dihydroxyphenyl)-3,4-dihydro-2H-chromene-5,7-diol	0.04	0.68	No	No	No
3.	(2S,3R)-2-(3,4-dihydroxyphenyl)-3,4-dihydro-2H-chromene-3,5,7-triol	1.92	0.87	No	No	No
4.	(2R)-2-(4-hydroxy-3-methoxyphenyl)-6-methoxy-3,4-dihydro-2H- chromen-7-ol	-1.32	0.61	No	No	No

The four ligands were subjected to molecular docking using AutoDock tools. The best confirmation of protein-ligand docking for the four ligands were selected based its total binding energy hydrogen bonding. The Table 4 depicts the results of the molecular docking. All the four ligands showed the low binding energy with the negative values. Its best docking pose is shown in Figure 6. The properties of hydrogen bonding of four ligands are given in Table 5. From the table is it is evident that all four ligands show good binding properties. Their hydrogen bonding property is shown in Figure 7.

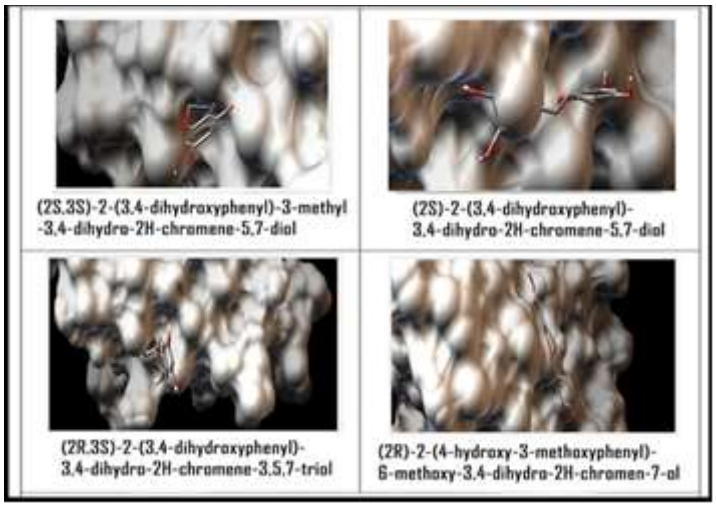

Figure 6: Docking pose of the four ligands with PD-1

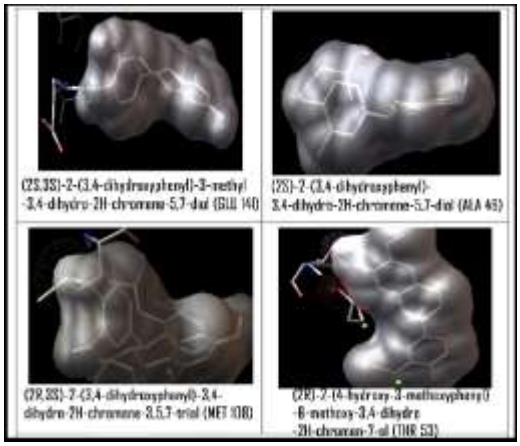

Figure 7: Hydrogen bond of the four ligands with PD-1

Among the four ligands, (2S, 3R)-2-(3,4-dihydroxyphenyl)-3,4-dihydro-2H-chromene-3,5,7-triol (Ligand 3) have excellent binding energy coupled with good ADMET properties.

Table 4: The results of AUTODOCK showing binding energies of four ligands

S. No.	Ligand	Total binding energy (kcal/mol)	Vanderwaals+ Hydrogen bond+ dissolvation energy	Electrostatic energy
1.	(2S,3S)-2-(3,4-dihydroxyphenyl)-3-methyl-3,4-dihydro-2H-chromene- 5,7-diol	-6.84 X 10^{12}	469000	-6.84 X 10^{12}
2.	(2S)-2-(3,4-dihydroxyphenyl)-3,4-dihydro-2H-chromene-5,7-diol	-6.65 X 10^{12}	230000	-6.65 X 10^{12}
3.	(2S,3R)-2-(3,4-dihydroxyphenyl)-3,4-dihydro-2H-chromene-3,5,7-triol	-6.65 X 10^{12}	487.17	-6.65 X 10^{12}
4.	(2R)-2-(4-hydroxy-3-methoxyphenyl)-6-methoxy-3,4-dihydro-2H- chromen-7-ol	-6.76 X 10^{12}	106000	-6.76 X 10^{12}

Table 5: The H bond properties of the four ligands

S. No.	Ligand	distance	energy	theta	Phi
1.	(2S,3S)-2-(3,4-dihydroxyphenyl)-3-methyl-3,4-dihydro-2H-chromene-5,7-diol	1.732	-7.632	174.298	132.552
2.	(2S)-2-(3,4-dihydroxyphenyl)-3,4-dihydro-2H-chromene-5,7-diol	1.439	1.782	130.712	100.341
3.	(2S,3R)-2-(3,4-dihydroxyphenyl)-3,4-dihydro-2H-chromene-3,5,7-triol	0.552	-4.682	161.013	127.725
4.	(2R)-2-(4-hydroxy-3-methoxyphenyl)-6-methoxy-3,4-dihydro-2H-chromen-7-ol	1.865	-3.155	141.186	135.0

4. Conclusion

PD-1 protein is one of the important factors involved in down regulation of T cell infiltration during oral squamous cell carcinoma when it binds with its ligand present on carcinoma cells. Hence blocking of PD-1 receptor will prevent the down regulation of T cell infiltration thereby can help the prevention of oral squamous cell carcinoma. In the present study, (2S, 3R)-2-(3,4-dihydroxyphenyl)- 3,4-dihydro-2H-chromene-3,5,7-triol has found to have very good inhibitory property based on molecular docking study. Further the compound shows a good ADMET properties based on studies in OSIRIS. Hence it is concluded that (2S,3R)-2-(3,4- dihydroxyphenyl)-3,4-dihydro-2H-chromene-3,5,7-triol is an excellent drug candidate in the control of oral squamous cell carcinoma.

References:

1. Hamanishi J, Mandai M, Iwasaki M, Okazaki T, Tanaka Y, Yamaguchi K, Higuchi T, Yagi H, Takakura K, Minato N, et al: Programmed cell death 1 ligand 1 and tumor-infiltrating CD8+ T lymphocytes are prognostic factors of human ovarian cancer. Proc Natl Acad Sci USA 2007, 104:3360– 3365.
2. Tsushima F, Tanaka K, Otsuki N, Youngnak P, Iwai H, Omura K, Azuma M: Predominant expression of B7-H1 and its immunoregulatory roles in oral squamous cell carcinoma. Oral Oncol 2006, 42:268–274.
3. Francisco LM, Sage PT, Sharpe AH: The PD-1 pathway in tolerance and autoimmunity. Immunol Rev 2010, 236:219– 242.
4. Greenwald RJ, Freeman GJ, SharpeAH.The B7 family revisited. Annu Rev Immunol 2005, 23:515-548.
5. Keir ME, Liang SC, Guleria I, et al. Tissue expression of PD-L1mediates peripheral T cell tolerance. J Exp Med 2006, 203:883-895.
6. LatchmanY,Wood CR, ChernovaT, et al. PD-L2 is a second
7. ligand for PD-1 and inhibits T cell activation.Nat Immunol 2001, 2:261-268.
8. Freeman GJ, Long AJ, Iwai Y, et al. Engagement of the PD-1 immunoinhibitory receptor by a novel B7 family member leads to negative regulation of lymphocyte activation. JExpMed 2000, 192:1027-1034.
9. Greenwald RJ, Freeman GJ, SharpeAH.The B7 family revisited. Annu Rev Immunol 2005, 23:515-548.
10. Curiel TJ,Wei S, Dong H, et al. Blockade of B7-1 improves myeloid dendritic cell-mediated antitumor immunity. NatMed 2003, 9:562-567.
11. Dong H, Strome SE, Salomao DR, et al.Tumor-associated B7-1 promotes T-cell apoptosis: a potential mechanism of immune evasion. Nat Med 2002, 8:793-800.
12. OhigashiY,ShoM,YamadaY, etal.Clinical significance of programmed death-1ligand-1and programmed death- 1ligand-2 expressioninhuman esophageal cancer. Clin CancerRes 2005, 11:2947-2953.
13. Scully C, Bedi R. Ethnicity and oral cancer.Lancet Oncol 200, 1:37-42.
14. Mignogna MD, Fedele S, Lo Russo L. The World Cancer Report and the burden of oral cancer. Eur J Cancer Prev 2004, 13(2):139–142
15. Cho YA, Yoon HJ, Lee JI, Hong SP, Hong SD. Relationship between the expressions of PD-L1 and tumor-infiltrating lymphocytes in oral squamous cell carcinoma. Oral Oncol. 2011, 47:1148-1153.
16. Semura M, Saeki K, Kimura T, Hayakawa S, Minami T, Sazuka M. Tea catechins and related polyphenols as anti- cancer agents. BioFactors, 2000, 13: 81–85.

17. Cheng, X, Veverka, V, Radhakrishnan, A. et al. Structure
18. and interactions of the human programmed cell death 1 receptor. J.Biol.Chem. 2013, 288: 11771-11785
19. Wass MN, Kelley LA and Sternberg MJ: 3DLigandSite: predicting ligand-binding sites using similar structures.*NAR* 2010, 38:469-473.
20. ACD/ChemSketch Freeware, version 10.00, Advanced Chemistry Development, Inc., Toronto, ON, Canada, 2012.
21. Yang JM and Chen CC: "GEMDOCK: A generic evolutionary method for molecular Docking." Proteins: Structure, Function, and Bioinformatics, 2004, 55:288-304.