MAPLE TREE PUBLISHING HOUSE

INTERNATIONAL RESEARCH JOURNAL OF MULTIDISCIPLINARY TECHNOVATION (IRJMT)

http://www.mapletreejournals.com/index.php/IRJMT
Received 03 September 2019 ISSN 2582-1040
Accepted 30 October 2019 2019; 1(6); 253-257
Published online 02 November 2019

Role of AMPK signaling in Repigmentation- An Insilico study

M.Thenmozhi[1*], A.Murugesan[2] S.T.Kumaravel[3]

[1]*Assistant Professor, Department of Biotechnology, Selvam College of Technology, Tamil Nadu, India.*
[2] *Professor, Department of Mechanical Engineering, KSRangasamy College of Technology, Tamil Nadu, India.*
[3]*Assistant Professor, Department of Mechanical Engineering, Paavai College of Engineering, TamilNadu, India.*

**Corresponding author E-Mail ID: thenmozhimsel@gmail.com*

Doi: https://doi.org/10.34256/irjmtcon34

ABSTRACT

Vitiligo is an epidermal disorder causes depigmented patches resulted from the loss of melanocytes, Autoimmunity hypotheses strongly supports that the immune system compartments responsible in the development of vitiligo. Adenosine MonoPhosphate kinase (AMPK) signaling plays a role in regimentation in vitiligo. In this present study, set of ligands selected to dock against AMPK protein in the AMP binding site using FlexX software. Based on the scores and protein-ligand interactions selected ligands were analyzed for its binding affinity and protein ligand stability for its further drug development process.

Keywords: Vitiligo, autoimmunity, AMP, AMPK,

1. INTRODUCTION

Vitiligo is a rare and chronic autoimmune disease in which the body attacks its own melanocytes in the epidermis, causing depigmentation in irregular patches of skin and hair. Vitiligo affects 0.5-2% of the world population [Ray *etal*., 1994]. Currently there is no cure for Vitiligo. These with the disease often use topical creams and concealers to diminish the appearance of depigmented patches. Those with severe cases have sometimes resorted to skingrafts and photo therapies. Corticosteroids have been used in the treatment of vitiligo since 1970[Doghim *etal.*,2011].

ransrepression of NF-kB leads to suppression of immune system. Physical interaction between GR and p-65 subunit of NF-kB leads to inactivation of NF-kB. Because N-terminal of p-65 subunit has the strong capacity for transactivating potential of NF-kB. Interaction between N-terminal subunit p-65 and two critical aminoacids in the C-terminal zinc finger leads to the inactivation of NF-kB results to down regulate the immune system [Liberman *et al.*,2007].

AdenosineMonophosphateprotien kinase (AMPK) signaling mediates many cellular processes. From all AMPK signaling activation leads to Nf-kB deactivation, it helps in autoimmunity suppression in vitiligo patients. As well as imbalance of glucose uptakes found in vitiligo patients, high glucose levels inactivate AMPK so using AMPK activators helps to improve glucose metabolism it helps in repigmentation (Viollet.B *et al.*,2009, 2011).

In this present study set of 6 ligands which are as follows Ascorbic acid, Berberine, Chrysin, Kaempferol, Pinocembrine, Piperine allowed to binding in the AMP binding site and also AMP allowed to binding in the AMP binding site then compare the results (scores) of all the 6 compounds with AMP result (score). There were no docking studies carried out to study the protein-ligand stability based on the scores for AMPKinase with these set of ligands.

MATERIALS AND METHODS

Protein Preparation

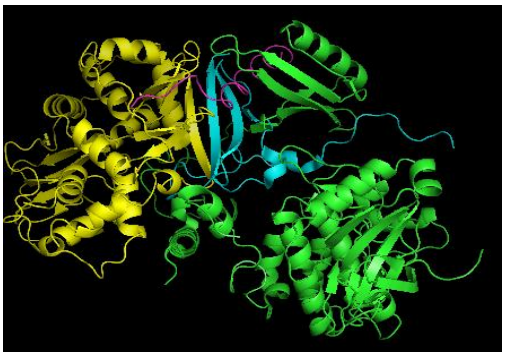


Fig1: Protein AMPK (2Y94)

Protein structure was downloaded from web based server www.rcsb.org. Selected protein structure was prepared using following steps using FlexX software.

Chain selection, Cofactor selection, if the structure is ligand bound in the next step the reference ligand must be selected to set active site pocket where the input ligands allowed binding into it properly. In non-ligand bounded structures active site pockets are predicting by specifying active site surrounding residues in this step. In this step we have to specify the residues included for docking analysis. Also if the protein contains water molecules we have to eliminate from the protein. Then the preprocessing of receptor done for further docking calculations.

Ligands Preparation

Ligands are imported in SDformat files then the docking process will be carried out in FlexX software.

Docking calculations

Based on the mentioned algorithm, FlexX docking program performed for the set of ligands docked against set of protein structures.

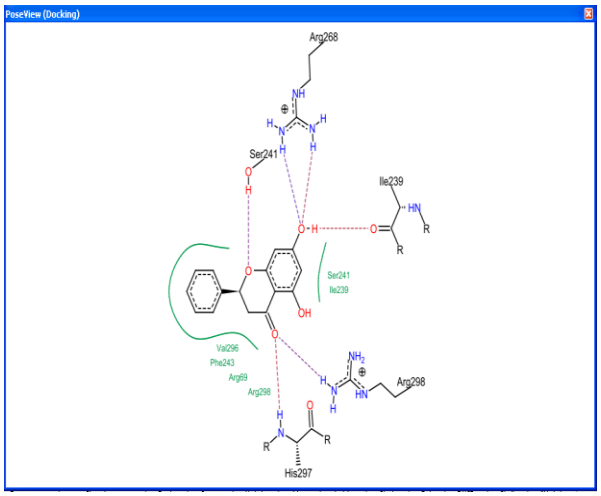
Table1 Interaction energies and molecular hydrogen bond interactions of selected ligands against AMPK protein.

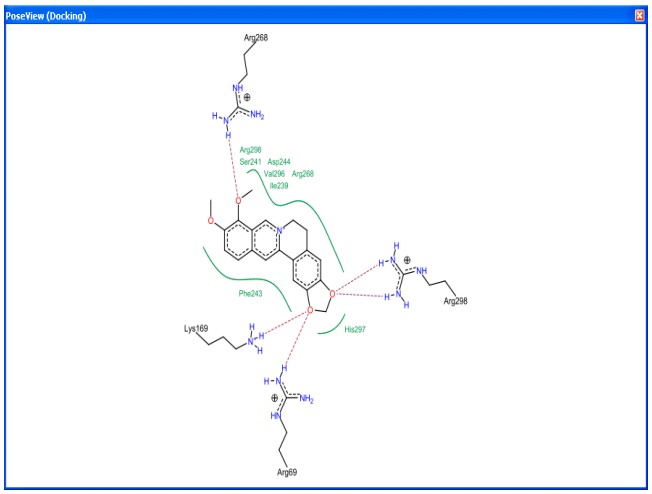
S.No	Ligand name	Ligand atom: AMPK	Interacting Residues	H-bond Length (Å)	FlexX scores
1	Berberine	O2…HH21 O2…HZ2 O3…HH12 HH…O1 HH21…O1	ARG69 LYS169 ARG268 ARG298 ARG298	1.69 2.09 2.01 1.97 1.82	-24.4
2	Piperine	O2…HH21 O2…HZ2 O1…HH12 O3…H O3…HH11	ARG69 LYS169 ARG268 HIS297 ARG298	1.95 1.91 2.08 1.82 1.70	-25.1961
3	Pinocembrine	H31…O O1…HG O4…HH11 O4…HH21 O2…H O2…HH11	ILE239 SER241 ARG268 ARG268 HIS297 ARG298	1.93 2.03 2.01 2.09 1.60 1.94	-20.693

4	Chrysin	H29…OD2 O4…HH11 O3…H O3…HH11	ASP244 ARG268 HIS297 ARG298	1.67 1.68 1.65 2.07	-21.7023
5	Ascorbic acid	O5…HZ2 H17…OD2 H19…OD2 H19…OD2 H…O6 HH…O6	LYS169 ASP244 ASP244 ASP244 HIS297 ARG298	1.84 2.23 2.23 1.91 2.01 1.53	-17.2892

(A) Pinocembrine

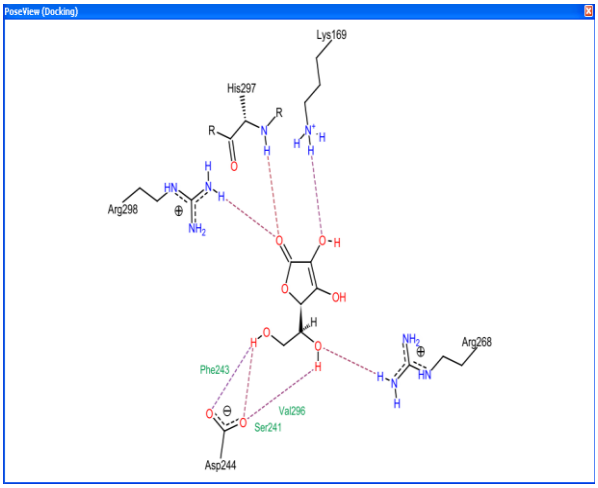
(B) Berberine





(C) Piperine

(D)Chrysin



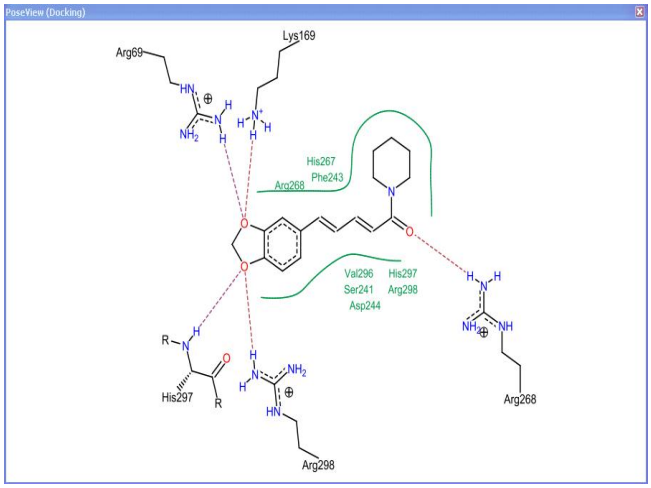


Figure 3 (i) Docked confirmation of AMPK with top ranked docked poses of selected ligands showing Interactions with non-ligand residues involved in the hydrogen (Black colour) & Hydrophobic interactions (Green color).

The results are in the form of spread sheets it consists of FlexX score, Total score- total docking score, RMS score, etc.

(E) Ascorbic acid

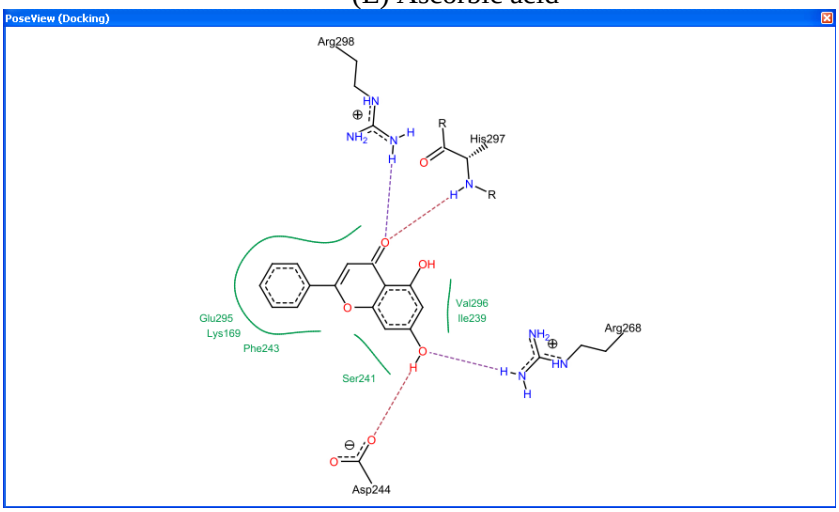


Figure 3 (ii): Docked confirmation of AMPK with top ranked docked poses ofselected ligands showing Interactions with non-ligand residuesinvolved in the hydrogen (Black colour) & Hydrophobicinteractions (Green color).

AMPK activation with the above said ligands (Ascorbic acid, Berberine, Chrysin, Pinocembrine, Piperine) studied computationally since it is traditionally using in a blind manner to cure this disorder. FlexX, CLC drug software employed to study the ligand binding affinity as well as protein-ligand stability. AMP binding site residues in the crystal structure of AMPK protein were compared with the docking results binding site residues of the selected ligands in the AMPK protein. It helps us to ensure the preliminary *insilco* analysis proved the activation efficacy. According to FlexX scores the ligand efficacy descended as follows (Table1) Piperine> Berberine> Chrysin > Pinocembrine > Ascorbic acid.

Analysis of hydrogen bond

Based on the results from FlexX, when analyzing hydrogen bond forming amino acids here AMP binding site residues are compared with the resulting interacting residues. LYS169A, SER241A, ASP244A, ARG268A, ARG298A, HIS297A these are the amino acid residues similar to AMP interacting amino acid residues (Figure 3). So the testing ligand components may also have the property to activate AMPK protein like AMP molecule. Some additional interacting residues ARG69A, ILE239A were found in the binding site. All the interacting residues are nearer to the ligand which shows the protein-ligand stability.

Analysis of Hydrophobic interactions

ILE239, SER241, PHE243, ASP244, HIS267, ARG268, VAL296, HIS297, ARG298, are the hydrophobic amino acid residues found in top scoring docked poses of ligands (Berberine and Piperine) against AMPK protein

From the results, Piperine, and Berberine could be the lead components based on the binding affinity (Docking scores) and protein-ligand stability (Molecular interactions). For AMPKII protein.

REFERENCES

1. Bosscher.K.D, Schmitz.L.M, Berghe.V.M, Plaisance.S, Fiers.W, Hageman.G, "Glucocorticoid-mediated repression of nuclearfactor- xB- dependent transcriptioninvolves direct interference with transactivation", *Proceedings of the national academy of sciences USA*,**94:**13504-13509;(1997).
2. Doghim.NN, Hassan.A.M,El-Ashmawy.AA,Gheida.S.F, "Topical combination of calcipotriol plus betamethasone dipropionate and narrowband in the treatment of vitiligo", *Life science journal*,**8(4):**186-197;(2011)
3. Kemp.E.H, Emhemad.S, Gowkrodger.D.J, Weetman.A.P, "Autoimmunity in vitiligo", In *Autoimune disorders-Pathogenic aspects,,* Cilo mavragani(ed);271-294, Intech Publishers, (2011).
4. Lee.Y.S, Kim.W.S, Kim.K.H, Yoon.M.J, Cho.H.J, Shen.Y. Ye.J.M, Lee.C.H, Oh.W.K, Kim.C.T, Behrens.C.H, Gosby.A, Kraegen.E.W, David.E.J, Kim.J.B, "Berberine, a natural plant product, activates AMP- activated protein kinase with beneficial metabolic effects in diabetic and insulin-resistant states" *Diabetes*, **55**:2256-2264;(2006).
5. Liberman.A.C, Refojo.D,Druker.J,Toscano.M, Rein.T,Arzt.E, " The activated glucocorticoid receptor inhibits the transcription factor T-bet by direct protein-protein interaction"*The FASEB journal*, **21**:1177-1188;(2007).
6. Liden.J,Delaunay.F,rafter.I,Gustafsson.J.A,Okret.S, "A new function for the C—terminal zinc finger of the glucocorticoid receptor –Repression of RelA transactivation" *The journal of biological chemistry*,**272**:21467-21472;(1997).
7. Nissen.R.M, Yamamoto.K.R, " The glucocorticoid receptor inhibits NF-xB by interfering with serine-2 phosphorylation of the RNA polymerase II corboxy terminaldomain", *Genes &development*,**14**:2314-2329;(2000).
8. Ray.A, Prefontaine.K.E, "Physical association and functional antagonism between the p-65subunit of transcription factor NF-xB and the glucocorticoid receptor" *Proceedings of the national academy of sciences,. USA.*,**91:**752-756;(1994).
9. Violett.B, Horman.S, Leclerc.J, Lantier.L, Marc.F, Billaud.M, Giri.S, Fabrizio.A, "AMPK inhibition in health and disease" *Critical revies in biochemistry and molecular biology*, **45(4)**:276-295; (2010).
10. Violett.B, Horman.S, Leclerc.J, Lantier.L, Marc.F, Billaud.M, Giri.S, Fabrizio.A, "Targetting AMPK pathway for the treatment of Type 2 Diabetes" Frontiers *in bioscience*, **14**:3380-3400;(2009).

Conflict of Interest

None of the authors have any conflicts of interest to declare.