INTERNATIONAL RESEARCH JOURNAL OF PHARMACY
www.irjponline.com
ISSN 2230 – 8407

Research Article

MOLECULAR DOCKING STUDIES OF NOVEL FURAN-AZETIDINONE HYBRID COMPOUNDS AS POTENTIAL ANTIFUNGAL AGENTS

Judy Jays [1]*, S Mohan [2], J Saravanan [2]
[1]Faculty of Pharmacy, M.S. Ramaiah University of applied sciences, M.S.R.I.T. Post, Bangalore, Karnataka, India
[2]PES college of pharmacy, Hanumanthnagar, Bangalore, Karnataka, India
*Corresponding Author Email: judyjays4@gmail.com

Article Received on: 25/11/18 Approved for publication: 11/01/19

DOI: 10.7897/2230-8407.100260

ABSTRACT

Antifungal resistance poses a significant clinical challenge for treating invasive fungal infections. *Candida albicans* is responsible for candidiasis including invasive fungal infections, where most patients are immunocompromised. Therefore the success of treatment depends significantly on the effectiveness of the antifungal agent. In this study, sixteen novel furan derivatives containing the azetidinone moiety were designed and synthesized to arrive at potentially effective antifungal agents. *In silico* antifungal activity was carried out to identify the specificity of the novel furan derivatives for the fungal proteins using 'Glide'. Molecular docking studies were conducted on two antifungal targets; Dihydrofolate reductase of *C.albicans* (PDB ID: 4HOE); N-myristoyl transferase of *C.albicans* (PDB ID: 1IYK). Molecular docking was carried out at the Standard Precision (SP) and Extra Precision (XP) mode. The docked poses were ranked according to their docking scores (GScore) and their binding energy with the enzyme. The results obtained for the docking of the title compounds with N-myristoyl transferase of *C.albicans* is quite promising. Molecular docking suggests that compounds 4d, 4e, and 4h are potential inhibitors of N-myristoyl transferase and are specific in binding at the active site of the enzyme. They form H-bond with THR 211 and pi-pi stacking interactions with PHE 117, TRY 354, and TRY 225 at the active site of the protein, similar to the standard drug. However the test compounds show low docking scores against Dihydrofolate reductase of *C.albicans* indicating that they may not be effective against it.

KEY WORDS: Furan, Azetidinone, Docking, *C.albicans*, Antifungal activity.

INTRODUCTION

Antifungal resistance poses a significant clinical challenge for treating invasive fungal infections due to the limited arsenal of systemically available antifungal agents.[1] Candida Species is the leading cause of healthcare associated bloodstream infections in US hospitals.[2] *Candida albicans* is responsible for candidiasis which comprises a host of fungal infections, including invasive fungal infections.[3] Invasive fungal infections have become one of the main causes of illness and mortality in immunocompromised patients.[4] Hence the success of treatment depends critically on the effectiveness of the antifungal agent. Since only limited classes of antifungals are available, the emergence of resistance significantly hinders the treatment.[5] Many synthetic compounds containing furan nucleus possess various pharmacological activities such as antibacterial, antifungal, antiviral, antidepressant, anti-inflammatory, anti-ulcer, diuretic and antihypertensive activities.[6,7] Azetidinones possess antimicrobial, antifungal, antibacterial, antiviral, anticonvulsant, antioxidant, antimycobacterial, and anthelmintic activities.[8,9] In this perspective, novel azetidinone-furan hybrids were synthesized with the objective of obtaining more potent antifungals.[10]

Molecular docking studies are used to explain the binding of the synthesized compounds with the target proteins to gain knowledge for structural optimization. The title compounds were docked onto the active site of the crystal structure of the enzymes. The mechanism of interaction of furan-azetidinone hybrids were studied on two target proteins of *C.albicans* - **N-myristoyl transferase (PDB ID: 1IYK) and Dihydrofolate reductase of (PDB ID: 4HOE).**N-myristoyl transferase (NMT) catalyzes the transfer of the rare fatty acid myristate from myristoyl-CoA to the N-terminal glycine of substrate proteins, and is found only in eukaryotic cells. NMT activity is essential for vegetative growth. Recent NMT inhibitors like benzofurans show *in vivo* antifungal activity and are promising selective fungal N-myristoyltransferase inhibitors.[11,12] Dihydrofolate reductase catalyses the reduction of dihydrofolate into tetrahydrofolate, and is critical for the biosynthesis of purines, thymidylate and some amino acids which are important for cell proliferation and cell growth.[13] Fungal dihydrofolate reductase is different from the human enzyme.[14] Hence inhibitors of fungal dihydrofolate reductase maybe useful as antibacterial agents.[15]

MATERIALS AND METHODS

Molecular Modeling and Scoring

Molecular modeling was carried out using GLIDE 2.0, running on a Intel® Core TM i3-2130 CPU@ 3.40GHz processor using Linux professional workstation.

Preparation of ligands[16]

The structures were drawn in 2D. The corresponding 3D structures were generated using the Chem draw 3D software. By using the standard bond lengths and bond angles, the geometry optimization was carried out with the help of standard OPLS_2005 force field. All the conformations were optimized for minimum energy.

Preparation of protein [17]

The crystal structures of ***C.albicans,*** dihydrofolate reductase complexed with NADPH and 5-[3-(2,5-dimethoxy-4-phenylphenyl)but-1-yn-1-yl]-6-methylpyrimidine-2,4-diamine (PDB code 4HOE) and ***C.albicans,*** **N-myristoyl transferase**, complexed with myristoyl-COA and peptidic inhibitor (PDB entry code 1IYK), was downloaded from the Protein Data Bank (PDB extracted from the Brookhaven Protein Database http://www.rcsb.org/pdb) and used for docking studies. The protein was prepared using the protein preparation wizard.[17] Co-crystalized ligand and water molecules were removed from the structure, H-atoms were added, disulphide bonds were created and side chains were fixed during protein preparation. The structure was then subjected to an energy refinement and energy minimization procedure.

Receptor Grid generation

The optimized protein with co-crystallized ligand was taken to generate a 3D (20x20x20 A°) grid at the active site of the target protein as per the standard protocol.The co-crystallized ligand molecule is removed and the prepared structure will is docked in its place. Receptor grid generation allows to define the position and size of active site for ligand docking.

Docking protocol [18-21]

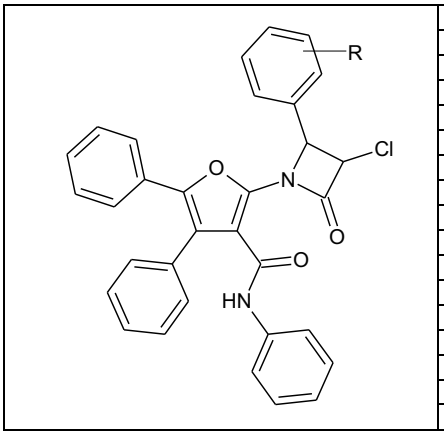
The binding of the furan derivatives was estimated using a variety of scoring functions that have been compiled into the single score (GScore). The G score integrates a number of popular scoring functions for ranking the affinity of ligand bound to the active site of a receptor.

GScore = 0.065*vdW+0.130*Coul +Lipo + Hbond + Metal + buryP + rotB +site

VdW=Van der Waals energy, Coul= Coulonb energy; Lipo=lipophilic term; Hbond=hydrogen bonding term; Metal=metal binding term; buryP=penalty for buried polar groups; rotB=penalty for freezing rotatable bonds; site=polar interactions in the active site

Glide XP (Extra precision) mode combines a powerful sampling protocol with a value of a custom scoring function designed to identify ligand poses. The chief purpose of XP method is to weed out false positives and provide a better correlation between good poses and good scores. The XP scoring function includes additional terms over SP (Standard precision) scoring function.

Table 1: Structures of the novel furan derivatives



Compound Code	R
4a	H
4b	2-ethyl
4c	4-ethyl
4d	4-chloro
4e	2,4-dichloro
4f	2-nitro
4g	4-nitro
4h	4-methoxy
4i	3-methoxy
4j	3-nitro
4k	3-chloro
4l	4-dimethyl amino
4m	3,4-dimethoxy
4n	3,4,5-trimethoxy
4o	2,6-dichloro
4p	Furfuryl

Table 2: Molecular docking results of furan derivatives N-myristoyl transferase

Compound Code	R	SP mode		XP mode		
		GScore	Emodel	GScore	Emodel	Interactions at active site
4e	2,4-dichloro	-6.968	-80.552	-10.896	-88.144	Phe 117,Tyr 354, Thr 211
Miconazole		-6.680	-57.104	-6.432	-65.489	Asp 412, Asp 112
Ketoconazole		-7.582	-67.082	-7.503	-88.869	Phe 240, Asp 412, Asp 110
Clotrimazole		-4.718	-28.043	-5.372	-35.957	Phe 117, Tyr 225, Phe 339

Table 3: Molecular docking results of furan derivatives with DHFR

Compound Code	R	SP mode		XP mode		
		GScore	Emodel	GScore	Emodel	Interactions at active site
4d	4-chloro	-6.453	-67.415	-6.494	-69.855	Phe 36, Arg 72
Miconazole		-6.680	-57.104	-6.432	-65.489	Ala 115, Glh 32,
Ketoconazole		-7.582	-67.082	-7.065	-68.387	Phe 36,
Clotrimazole		-4.718	-28.043	-3.889	-32.197	Phe 36, Ile 112,

Fig.1. N-myristoyl transferase of *C.albicans* (PDB ID: 1IYK)

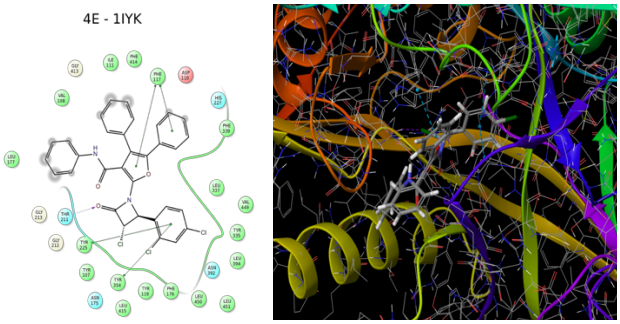


Fig.2. 2D and 3D Docked poses of Compound 4e with N-myristoyl transferase of *C.albicans* (PDB ID: 1IYK)

RESULTS AND DISCUSSION

To understand the molecular basis of interaction and affinity of binding of the furan-azetidinone hybrids analogues with the N-myristoyl transferase and Dihydrofolate reductase proteins, the ligands were docked into the active site of the respective proteins. Although docking was carried out in the SP and XP modes, the XP results are discussed as the SP mode may give some false positives. The docking results of the ligands with N-myristoyl transferase is given in Table 2, The docking score using GLIDE varied from -5.263 to -10.896. The GScore for docking of the standards Miconazole, Ketoconazole, and Clotrimazole with N-myristoyl transferase was -7.301, -6.357 & -7.301.

The results of docking with DHFR is given in Table 3. The Gscore of the ligands ranged from -3.983 to -6.494 while that of the standards Miconazole, Ketoconazole, and Clotrimazole was -6.432, -7.065 and -3.889 respectively.

This proves that synthesized novel compounds could be potential anti-fungal drugs. The GLIDE score can be used as a semi-quantitative descriptor for the ability of ligands to bind to a specific conformation of the protein receptor. Usually good ligand affinity for the receptor may be expected for low GLIDE score. Compound 4e (2, 4 –dichloro derivative) showed the best the inhibition for the N-myristoyl transferase enzyme with Gscore -10.896. Interestingly the GLIDE score for many of the analogues were better than the GLIDE score of the standard drugs. There was good agreement between the localization of the inhibitor upon docking and from the crystal structure of the protein. Conformational analysis of different docked complexes also shows that residues PHE 117, TYR 225 and TRY 354 for N-myristoyl transferase and PHE 36 for DHFR play an important role in the respective receptor's activity. Docking studies executed by GLIDE has confirmed that the analogues fit well into the binding pocket of the N-myristoyl transferase receptor.
From the results, we may conclude that for successful docking, intermolecular hydrogen bonding and liphophilic interactions between the ligand and the receptor are very important. The main reason for the increase in GLIDE score is due to the penalties for close intra-ligand contacts.

CONCLUSION

Molecular docking studies were performed for all the synthesized furan derivatives on two antifungal target proteins - DHFR of *C. albicans* and N-myristoyl transferase of *C. albicans*. Energy minimization of title compounds was carried out, the protein was optimized and minimized, a 3-dimensional grid was generated at the active site, and molecular docking was carried out using the SP and XP docking modes of Glide module. The docking poses were ranked according to their docking scores (GScore) and their binding energy with the enzyme (Emodel). If the binding energy is less, compound is more active. The results obtained from molecular docking of title compounds with N-myristoyl transferase of *C. albicans* is quite promising. It can be concluded that compound 4e (2,4- dichloro derivative) is predicted to have good antifungal activity. The study suggest that the compounds are specific in binding at the active site of N-myristoyl transferase.

ACKNOWLEDGEMENTS

Authors are thankful to "Pharmacological modelling and research center" Ramaiah University of Applied sciences, for providing the facilities.

REFERENCES

1. Wiederhold NP. Antifungal resistance: current trends and future strategies to combat. Infection and Drug Resistance 2017; 10 (Aug): 249–259.
2. Magill SS, Edwards JR, Bamberg W, Zintars GB, Dumyati G, Kauner MA et al. Multistate point-prevalence survey of health care-associated infections. The New England journal of medicine 2014; 370:1198-1208.
3. "People at Risk for Invasive Candidiasis". cdc. gov. February 13, 2014. http://www.cdc.gov/fungal/diseases/candidiasis/invasive/risk-prevention.html
4. Srinivasan A, Lopez-Ribot JL, Ramasubramanian AK. Overcoming antifungal resistance. Drug Discovery Today: Technologies 2014; 11: 67-71.

5. Perlin DS, Rautemaa-Richardson R, Alastruey-Izquierdo A. The global problem of antifungal resistance: prevalence, mechanisms, and management. The Lancet Infectious Diseases 2017; 17(12): e383 – e392.
6. Banerjee R, Kumar HKS, Banerjee M. Medicinal significance of furan derivatives: A review. International Journal of Review in Life Sciences 2012; 2(1):7-16.
7. Meotti FC, Silva DO, Santos ARS, Zeni G, Rocha JBT, Nogueira CW. Thiophenes and furans derivatives: a new class of potential pharmacological agents. Environmental Toxicology and Pharmacology 2003 Dec; 15(1): 37-44.
8. Southgate R. The synthesis of natural β-lactam antibiotics. Contempory Organic Synthesis 1994; 1(Jan): 417-31.
9. Singh GS. β-Lactams in the New Millennium. Part-I: Monobactams and carbapenems. Mini-Reviews in Medicinal Chemistry 2004 Jan; 4(1):69-92.
10. Jays J, Mohan S, Saravanan J. Synthesis, characterization and antimicrobial screening of some novel furan-azetidinone hybrid compounds. International Journal of Pharma and Bio Sciences 2018 ; 9(Jan): 79-87
11. Prasad KK, Toraskar MP, Kadam VJ. *N-myristoyltransferase*: a novel target. Mini-Reviews in Medicinal Chemistry 2008; 8: 142-149.
12. Zhao C, Ma S. Recent advances in the discovery of *N-myristoyltransferase* inhibitors. ChemMedChem 2014; 9: 2425-2437.
13. Chan JH, Hong JS, Kuyper LF, Baccanari DP, Joyner SS, Tansik RL et.al. Selective Inhibitors of *Candida albicans* Dihydrofolate Reductase: Activity and Selectivity of 5-(Arylthio)-2,4-diaminoquinazolines Journal of Medicinal Chemistry 1995; 38(18) :3608-3616.
14. Whitlow M, Howard AJ, Stewart D, Hardman¶ KD,Kuyper LF, Baccanari DP et al. X-ray Crystallographic Studies of Candida albicans Dihydrofolate Reductase. The Journal Of Biological Chemistry 1997; 272 (48): 30289-30298.
15. Chan JH, Baccanari DP, Tansik RL, Boytos CM, Sharon KR, Brown AD et. al. Synthesis of mino-7,8,9,10-tetrahydropyrido[3,2-f]-quinazolines - inhibitors of *Candida-albicans* dihydrofolate-reductase as potential antifungal agents. Journal of Heterocyclic Chemistry 1997, 34(1): 145-151
16. Schrödinger: LigPrep software, version 2.5. New York, NY: LLC; 2011.
17. Beginning with Maestro. Schrödinger software release. LLC, New York, NY, 2015.
18. Schrödinger: Maestro, version 11. New York, NY: LLC; 2011.
19. Glide, version 6.7, User Manual. Schrödinger, LLC, New York, NY, 2015.
20. Friesner RA, Banks JL, Murphy RB, Halgren TA, Klicic, JJ, Mainz, DT et al. Glide: a new approach for rapid, accurate docking and scoring. 1. Method and assessment of docking accuracy. Journal of Medicinal Chemistry 2004, 47:1739
21. Friesner RA, Murph RB, Repasky MP, Frye LL, Greenwood JR, Halgren, TA, et al. Extra precision glide: docking and scoring incorporating a model of hydrophobic enclosure for protein-ligand complexes. Journal of Medicinal Chemistry 2006; 49: 6177 -96.

Cite this article as:

Judy Jays *et al*. Molecular docking studies of novel furan-azetidinone hybrid compounds as potential antifungal agents. Int. Res. J. Pharm. 2019;10(2):157-160 http://dx.doi.org/10.7897/2230-8407.100260

Source of support: Nil, Conflict of interest: None Declared