SYNTHESIS, SPECTROSCOPIC CHARACTERIZATIONS AND MOLECULAR MODELING STUDIES OF 2-(2-(2,2,2-TRIFLUOROETHOXY)-5-SUBSTITUTEDPHENYL)-5-(3-SUBSTITUTEDPHENYL)-1,3,4-OXADIAZOLE DERIVATIVES

C. Rajalakshmi[1], N. Santhi[2]
Department of Chemistry, Government Arts College, Chidambaram, Tamil Nadu

ABSTRACT

In the present study, synthesis and antimicrobial activity of 2,5- disubstituted 1,3,4-oxadiazole derivatives 3a-g are described. The structures of the newly synthesized compounds were confirmed by FTIR, ^{1}H NMR and ^{13}C NMR. In-vitro antibacterial activity was evaluated against staphylococcus aureus bacterial strain for all target compounds. In order to findout the affinity to bacterial proteins docking study were performed for seven synthesized derivatives, two antibiotic drug and co-crytallised ligands with different mechanism of action DNA gyrase B and Methylthioadenosine / S-Adenosylhomocysteine Nucleosidase (MTAN) using AutoDock 4.2. The in vitro antibacterial activity results and docking studies indicated that the synthesized compounds have potential antibacterial activity and can be further optimized as privileged scaffolds to design and develop potent antibacterial drugs.

Keywords: 1,3,4-Oxadiazole , synthesis, docking, DNA Gyras, MTAN, antibacterial activity

INTRODUCTION

Increasing number of multi-drug resistant microbial pathogens and emerging infectious diseases are the major challenging problems for the clinicians. There is an urgent need to develop new class of potent antimicrobial agents. Heterocyclic compounds play an important role in designing of a new class of structural entities of medicinal importance with new mechanisms of action.Heterocyclic compounds particularly 5-membered rings acquired importance because of their versatile pharmacological and biological activities [1]. In particular, 1,3,4-oxadiazoles have been used as 'privileged' scaffolds to produce substances of interest in numerous therapeutic areas. Differently substituted 1,3,4-oxadiazoles have been found to exhibit anti-inflammatory, antimicrobial, antitubercular, antidepressant, antimalarial, anti-HIV, and anticancer activities [2- 5]. 1,3,4-Oxadiazoles are well known bioisosteres of amides and esters [6], that can contribute significant pharmacokinetic property due to presence of N=C-O linkage in oxadiazole nucleus which increases the lipophilicity that influence the ability of drug to reach the target by transmembrane diffusion and show remarkable biological activity. Often, these molecules are used as pharmacophores due to their ability to bind with target proteins through hydrogen bond formation [7].

The several examples of compounds containing the 1,3,4-oxadiazole unit currently used in clinical medicine are: Raltegravir , an antiretroviral drug [8] and Zibotentanan anticancer agent. Oxadiazole nucleus is present in antihypertensive drugs such as tiodazosin and nesapidil and antibiotics such as furamizole.

DNA gyrase is a type II DNA topoisomerase that introduces negative supercoils into DNA in an adenosine-triphosphate (ATP)-dependent reaction[9]. Gyrase exists only in prokaryotes making it an attractive target of antibacterial drugs[10]. Gyrase was found to contain two subunits GyrA and GyrB, with the A protein being responsible

for DNA cleavage and rejoining, whereas the B protein contains the ATP-binding site and catalyzes ATP hydrolysis, providing the driving force for supercoiling of DNA2

One potential drug target that has been identified is enzyme 5'-methylthioadenosine/S adenosylhomocysteine (MTA/SAH) nucleosidase[11] an enzyme present in most bacterial species but absent in humans. MTA/SAH nucleosidase cleaves the glycosidic linkage of the nucleosides to generate the thiosugars 5-methylthioribose (MTR) or S-ribosylhomocysteine (SRH) and the purine adenine[12]. Nucleosidase inhibitors could exert antimicrobial actions by one or more mechanisms and have been explored as a target for the design of the broad-spectrum antibiotics against Gram-positive and Gram-negative bacteria by several laboratories[13-14]. Thus, inhibition of the nucleosidase could work by promoting a build-up of MTA and SAH to growth-inhibitory levels. In addition, the loss of salvage of purine and methionine components may suppress bacterial growth by nutrient limitation, and because it is energetically expensive to make methionine de novo

Considering these perspectives and our ongoing efforts to develop therapeutically active agent using a hybrid pharmacophore approach, we decided to incorporate proved biologically active moieties into single hybrid system and investigated their antibacterial activity against both staphylococcus aureus bacteria. Since both moieties are enabled to inhibit DNA gyrase and MTAN, we also decided to explore the interaction of this hybrid pharmacophore with the active site of DNA gyrase and MTAN. The main objective of our present study was to identify effective inhibitors of the DNA gyrase enzyme and MTAN by utilising its structural similarity to the commercially available antibiotic drug and its co crystallized ligand molecuyles. By combining computational and experimental approaches, for active compounds, antibacterial activity on Staphylococcus aureus was determined.

Material and Methods

Chemistry

Thin layer chromatography (TLC) was used to check the progress and completion of the reaction using silica gel G as an adsorbent (stationary phase) and ethyl acetate and hexane as mobile phase. Open glass capillaries were used to determine the melting point on popular melting point apparatus and were uncorrected. H^1 and ^{13}C nuclear magnetic resonance (H^1 NMR & ^{13}CNMR) spectra were recorded on Bruker Avance II 400 NMR spectrometer (400 MHz) at 298K, in appropriate deuterated solvent. Chemical shift were reported as δ (ppm) relative to tetramethyl silane (TMS) as internal standard. Infrared spectra (IR) were recorded as KBr pellet on Agilent Resolutions Pro FT-IR spectrometer.

Docking studies

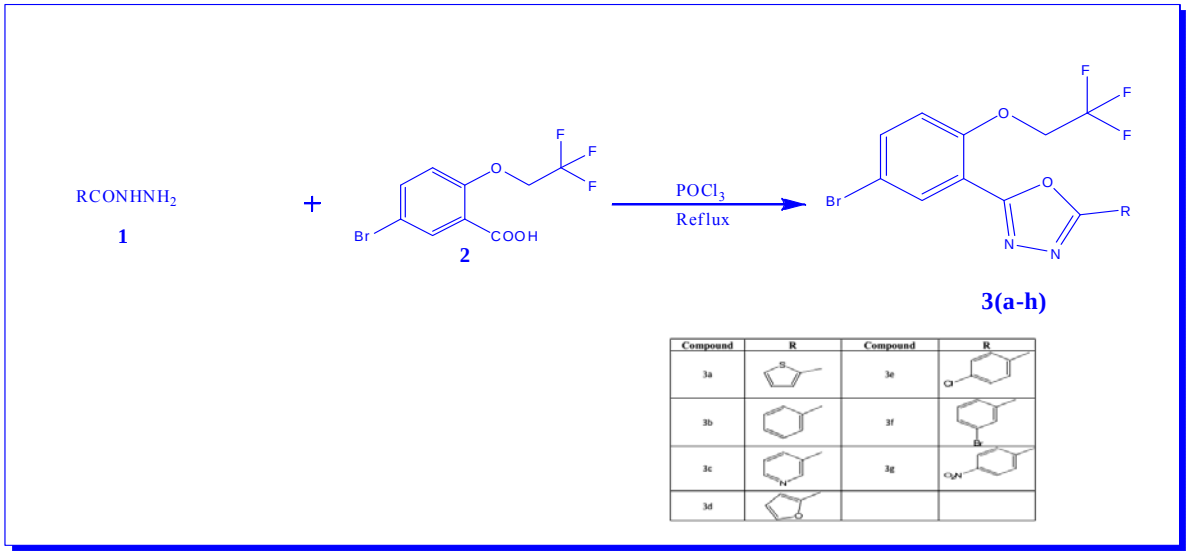
X-ray crystal structures of DNA gyrase B (PDB ID: 3G7B) and 5'-Methylthioadenosine/S-adenosylhomocysteine Nucleosidase [MTAN] (PDB ID: 3BL6) were retrieved from the Protein Data Bank [15]. The docking studies were performed using crystal structure of DNA Gyrase of S.aureus (PDB ID: 3U2D retrieved from the Protein Data Bank. To prepare the receptor for docking studies, cocrystallized ligand and water molecules were removed, while polar hydrogen atoms and Kollman-united charges were included to the DNA Gyrase receptor molecules. The necessary pdb and pdbqt files of ligands and S.aureus DNA Gyrase receptor were prepared using AutoDock 4.2 software[16]. The study was carried out using the usual docking protocol applied for AutoDock Vina in PyRx 0.8 software[17]. The docking results were analyzed using Discovery Studio 4.0 (Accelrys, Inc. San Diego, CA 92121, USA).

Anibacterial activity

All the synthesized 1,3,4-oxadiazole derivatives(**3a-g**) were tested for their anti-bacterial activity against a bacterial strain, namely, Staphylococcus aureus by paper disc diffusion method with different concentrations of the solutions prepared in Dimethyl sulfoxide (DMSO). The reason of choosing DMSO for antibacterial studies was that it has no effect on the above mentioned bacterial strains[18]. Nutrient agar was used as the culture medium for the growth of bacterial colony that was prepared by using peptone (3.0 g), NaCl (3.0 g), Yeast (1.5 g), Agar (6.0 g) in 300 mL of distilled water with pH at 7.0 The prepared medium is autoclaved at 15 pa for 20 minutes and kept at 120ºC for 30

minutes to sterilize the media. This media was then poured into petridishes slowly in laminar flow environment, allowed to solidify and kept at 30˚C for 24 hrs[19]. The bacterial strains were inoculated by spreading in peptidases and its temperature is maintained at 30˚C for 24 hr. Using paper disc (8 mm) in nutrient agar culture medium, different concentrations (50, 100, 150 µg/mL) of the newly synthesized 1,3,4-oxadiazole (4a-e) were loaded through bacteria free micro pipettes. The anti-bacterial activity was determined by measuring the zone of inhibition in millimeters and compared with standard drug Amoxillin and Cefixime.

Experimental



Scheme 1: Synthesis of 2-(5-bromo-2-(2,2,2-trifluoroethoxy)phenyl)-5-substituted-1,3,4-oxadiazole (3a–g)

General Procedure for the Synthesis of 2-(5-bromo-2-(2,2,2-trifluoroethoxy)phenyl)-5-substituted-1,3,4-oxadiazole (3a–g).

Aryl hydrazide 1 was dissolved in phosphorous oxychloride (5 mL) and to it equimolar amount of 5-bromo-2-(2,2,2-trifluoroethoxy)benzoic acid was added. The reaction mixture, after refluxing for 6-7 hours, was cooled to room temperature and poured onto crushed ice. On neutralization of the contents with sodium bicarbonate solution (20%), a solid mass separated out. This was filtered and washed with water. It was crystallized by using methanol to give 3a-g. The corresponding R for 6a–h is given in scheme 1

2-(5-bromo-2-(2,2,2-trifluoroethoxy)phenyl)-5-(thiophen-2-yl)-1,3,4-oxadiazole **(3a)**. Yield 72%, white powder, mp (∘C): 240–242; IR (cm^{-1}): 3114 (CHarom), 2925 (CHaliph), 1602 (C=N), 1065 (C–O–C), 1169 (C-F); ^{1}H NMR ($CDCl_3$, 400 MHz, ppm): 4.64 (s, 2H), 7.10-8.24 (m, 2H, aromatic protons); ^{13}C NMR ($CDCl_3$); 158.8 (C-5), 165.2(C-9), 163.4 (C-12), 57.4 (C-15).

2-(5-bromo-2-(2,2,2-trifluoroethoxy)phenyl)-5-phenyl-1,3,4-oxadiazole **(3b)**. Yield 76%, white powder, mp (∘C): 240–242; IR (cm^{-1}): 3072 (CHarom), 2921 (CHaliph), 1586 (C=N), 1063 (C–O–C), 1174 (C-F); ^{1}H NMR ($CDCl_3$, 400 MHz, ppm): 4.77 (s, 2H), 6.94-8.16 (m, 2H, aromatic protons); ^{13}C NMR ($CDCl_3$); 154.5 (C-5), 165.3(C-9), 161.7(C-12), 67.7 (C-15).

2-(5-bromo-2-(2,2,2-trifluoroethoxy)phenyl)-5-(pyridin-4-yl)-1,3,4-oxadiazole **(3c)**. Yield 68%, white powder, mp (∘C): 240–242; IR (cm^{-1}): 3097 (CHarom), 2922 (CHaliph), 1595 (C=N), 1058 (C–O–C), 1185 (C-F); ^{1}H NMR ($CDCl_3$, 400 MHz, ppm): 4.50 (s, 2H), 6.87-8.76 (m, 2H, aromatic protons); ^{13}C NMR ($CDCl_3$); 151.4 (C-5), 164.8(C-9), 159.1 (C-12), 57.6 (C-15).

2-(5-bromo-2-(2,2,2-trifluoroethoxy)phenyl)-5-(furan-2-yl)-1,3,4-oxadiazole **(3d)**. Yield 75%, white powder, mp (∘C): 240–242; IR (cm^{-1}): 3091 (CHarom), 2922 (CHaliph), 1525 (C=N), 1080(C–O–C), 1166 (C-F); ^{1}H NMR ($CDCl_3$, 400 MHz, ppm): 4.51 (s, 2H), 6.63-8.31 (m, 2H, aromatic protons); ^{13}C

NMR ($CDCl_3$); 158.0 (C-5), 164.8(C-9), 161.5 (C-12), 67.2 (C-15).

2-(5-bromo-2-(2,2,2-trifluoroethoxy)phenyl)-5-(4-chlorophenyl)-1,3,4-oxadiazole **(3e)**. Yield 75%, white powder, mp (∘C): 240–242; IR (cm^{-1}): 3076 (CHarom), 2946 (CHaliph), 1545 (C=N), 1059 (C–O–C), 1178 (C-F); ^1H NMR ($CDCl_3$, 400 MHz, ppm): 4.53 (s, 2H), 6.93-8.34(m, 2H, aromatic protons); ^{13}C NMR ($CDCl_3$); 154.0 (C-5), 164.5(C-9), 163.9(C-12), 67.5 (C-15).

2-(5-bromo-2-(2,2,2-trifluoroethoxy)phenyl)-5-(3-bromophenyl)-1,3,4-oxadiazole **(3f)**. Yield 78%, white powder, mp (∘C): 240–242; IR (cm^{-1}): 3070 (CHarom), 2922 (CHaliph), 1603 (C=N), 1070(C–O–C), 1187 (C-F); ^1H NMR ($CDCl_3$, 400 MHz, ppm): 4.51 (s, 2H), 6.93-8.33(m, 2H, aromatic protons); ^{13}C NMR ($CDCl_3$); 154.5 (C-5), 164.0(C-9), 162.0 (C-12), 67.1 (C-15).

2-(5-bromo-2-(2,2,2-trifluoroethoxy)phenyl)-5-(4-nitrophenyl)-1,3,4-oxadiazole **(3g)**. Yield 75%, white powder, mp (∘C): 240–242; IR (cm^{-1}): 3098 (CHarom), 2922 (CHaliph), 1584 (C=N), 1065 (C–O–C), 1192 (C-F); ^1H NMR ($CDCl_3$, 400 MHz, ppm): 4.54 (s, 2H), 6.94-8.43 (m, 2H, aromatic protons); ^{13}C NMR ($CDCl_3$); 154.5 (C-5), 163.6(C-9), 161.7 (C-12), 67.5 (C-15).

Docking Sudy

The three dimensional x-ray structures of DNA gyrase B (PDB ID: 3G7B) and MTAN (3BL6) of S.aureus, retrieved from the Protein Data Bank are shown in Figures 1 along with its active site residues. DNA gyrase B and MTAN were solved in complex with its respective inhibitors namely, methyl ((5-(4-(4-hydroxypiperidin-1-yl)-2-phenylthiazol-5-yl)-1H-pyrazol-3yl)methyl) carbamate and Formycin A.

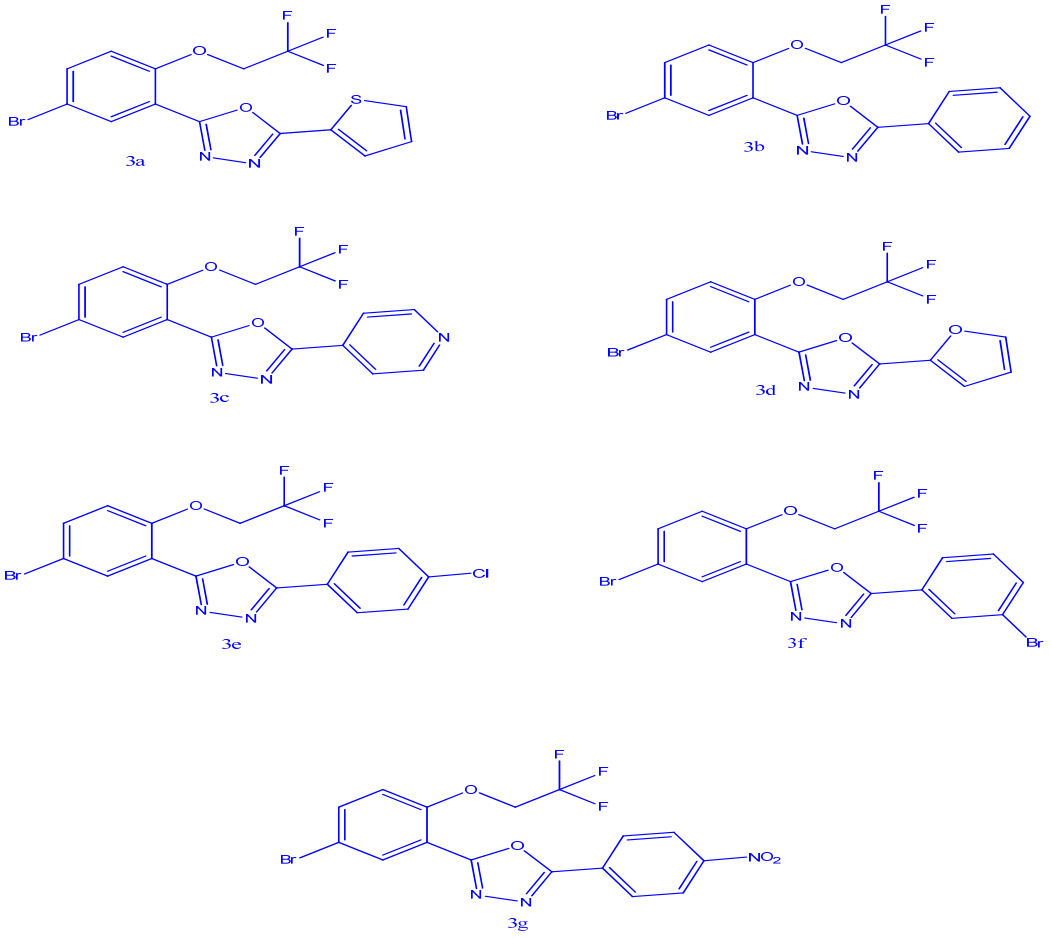


Figure 1: Schematic representation of structures of 1,3,4-Oxadiazole derivatives (3a-g)

These complex structures reveal essential interactions between the inhibitor and the protein and these interactions are taken as the reference for the 1,3,4-Oxadiazole derivative(3a-g). The co-crystallized ligand are forms hydrogen bond interaction with the residues GLY 85, ARG 144 and ARG 84 (Figure 1) which are present within the ATP binding pocket. The ligand is also further stabilized by a number of hydrophobic contacts with the residues ILE 175, ILE 51, ILE 86, GLU 58 and PRO 87. The co-crystallized ligand of MTAN forms several hydrogen bonds with the residues of SER 75, ASP 196, MET 172, VAL 170, GLU 11 and GLU 173.

The seven 1,3,4-Oxadiazole derivatives (3a-g) shown in Figure 1 were taken for docking studies. These compounds are synthesized and their structures have been determined by IR,^{1}H and^{13}CNMR spectroscopy. The docking studies clearly reveal that some of these compounds bind efficiently to the enzymes of S.aureus. Binding score of autodock 4.2 varies between -7.2 to -8.3 for compounds 3a-g tested for DNA gyrase B and -7.1 to -7.5 for MTAN (Table 4) Out of the seven 1,3,4-Oxadiazole derivatives analyzed, compound 3g forms the best interaction with DNA gyrase B and compound 3f forms the best interaction with MTAN respectively.

The compound 3g has the highest binding score of -8.3. The fluorine and oxgen atom on oxadiazole compound forms hydrogen bond with the hydrogen atom of ASN 54, ARG 144 of DNA gyrase B (Figure 2). Compound 3e having a binding score of -7.9 makes hydrogen bonds with the active site residue SER 55 and GLU 58 of DNA gyrase B (Figure 2). Re-docking of the inhibitor from the co-crystallized complex structure (Figure 2) of DNA gyrase B resulted in a binding score of -8.1 which is comparable to the scores found for compound 3g and 3e (Table 1). The re-docked conformation of co-crystallized ligand (Figure 2) resembles the conformation of the 1,3,4-oxadiazole derivative (compound 3g and 3e respectively).

Table 1: Binding sores of the 1,3,4-Oxadiazole compounds with DNA gyrase B and MTAN of S.aureus

DNA gyrase B	
COMPOUNDS	**BINDING SCORE**
3a	-7.2
3b	-7.4
3c	-7.6
3d	-7.2
3e	-7.9
3f	-7.6
3g	-8.3
Amoxillin	-6.7
Cefixime	-7.0
Co crystallized Ligand	-8.1
MTAN	
3a	-7.1
3b	-7.2
3c	-7.4
3d	-7.1
3e	-7.3
3f	-7.5
3g	-7.4
Amoxillin	-6.9
Cefixime	-6.5
Co crystallized Ligand	-6.7

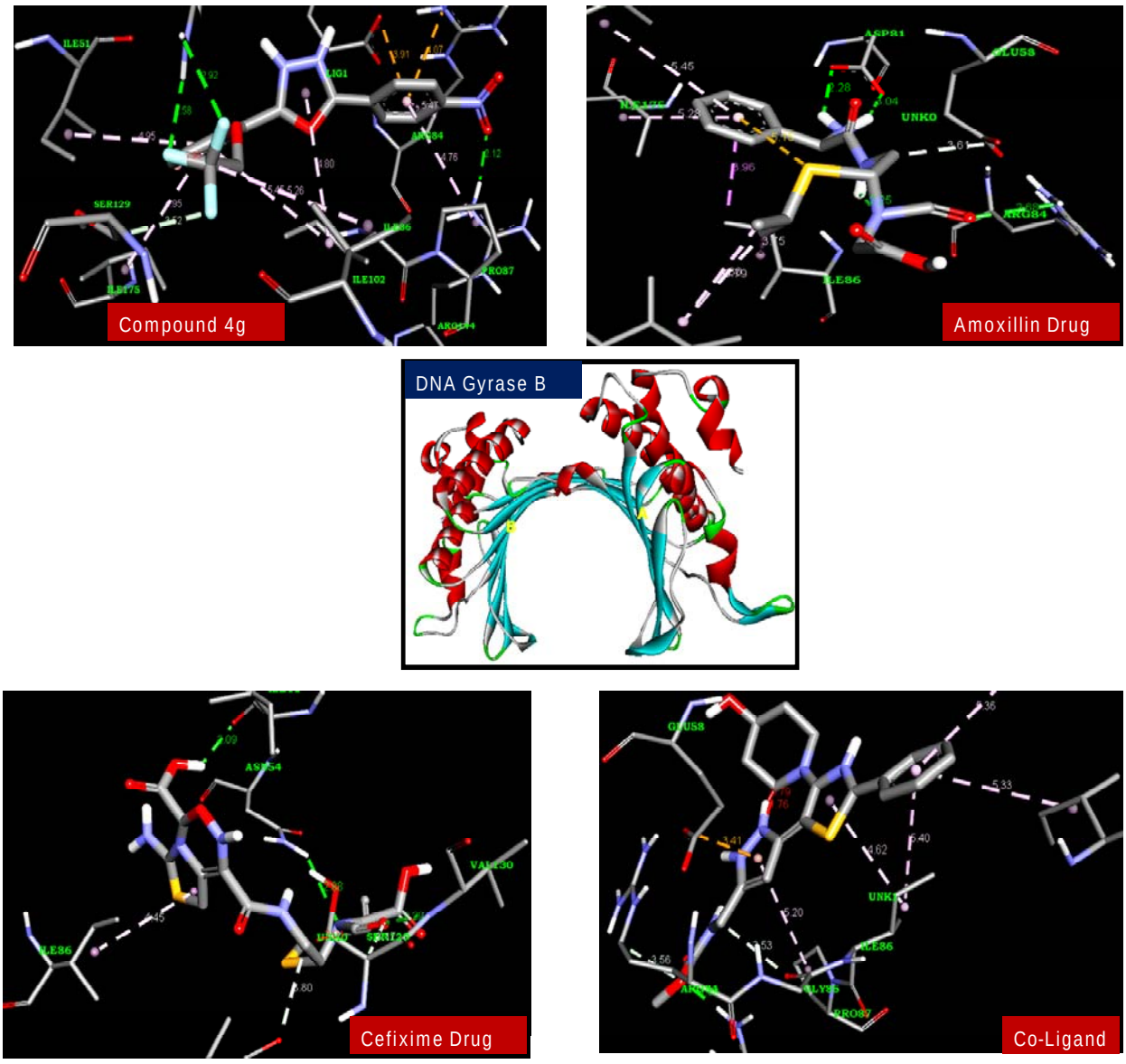


Figure:2 DNA gyrase B of S.aureus, Binding pose of Amoxillin with DNA gyrase B, Binding pose of Cefixime with DNA gyrase B , Binding pose of co-crystal structure ligand with DNA gyrase B and Binding pose of 1,3,4-Oxadiazole derivatives(3a-g) with DNA gyrase

We also analyzed the interaction and energy profile of DNA gyrase B with the already available drug Amoxillin and cefixime (Figure 2). The docking of Amoxillin and cefixime with DNA Gyrase B of S.aureus resulted in a binding score of -6.7 and -7.0, which is less binding energy than compound 3g,3c and 3e.

The Compound 3f, 3c and 3g form the best interactions with MTAN of S.aureus. The Compound 3f form hydrogen bonds with GLN 119 of MTAN with the binding score of –7.5. Apart from this, hydrophobic interactions are observed between the compound 3f with VAL 186, ASN 185, PRO 187, CYS 181, PHE 188, ALA 178, TYR 182 and Compound 3c hydrogen bonded with SER 121, GLN 119. which also contributes to the stability of this complex. The Compound 3g form hydrogen bonds with GLN 119 of MTAN with the binding score of –7.4

Table: 2 Interaction pattern of 1,3,4-Oxadiazole derivatives with DNA Gyrase B protein

COMPOUNDS	Hydrogen Bond Interaction	Other Interaction
3a	-	ASN 54, ASP 57, ILE 86 , ASP 81
3b	-	ASN 54, ASP 57, ILE 86 , ASP 81
3c		ASN 54, ILE 51, ILE 86 , ASP 81,ILE 175, GLU 58
3d	-	ASN 54, GLU 58, ILE 86, ASP 81
3e	SER 55, GLU 58	ASN 54,ILE 102, ILE 86, ARG 84, PRO 87
3f		ASN 54, ASP 81, ILE 86, PRO 87
3g	ASN 54, ARG 144	ASN 54, ASP 81, ILE 86, ARG 84
Amoxillin	GLU 58, ASP 81, ARG 84	ILE 175, ILE 51, ILE 102
Cifixime	ILE 51, ILE 102, ASN 54, VAL 130, SER 129	ILE 86
Co-crystallized ligand	GLY 85, ARG 144, ARG 84	ILE 175, ILE 51, ILE 86, GLU 58, PRO 87

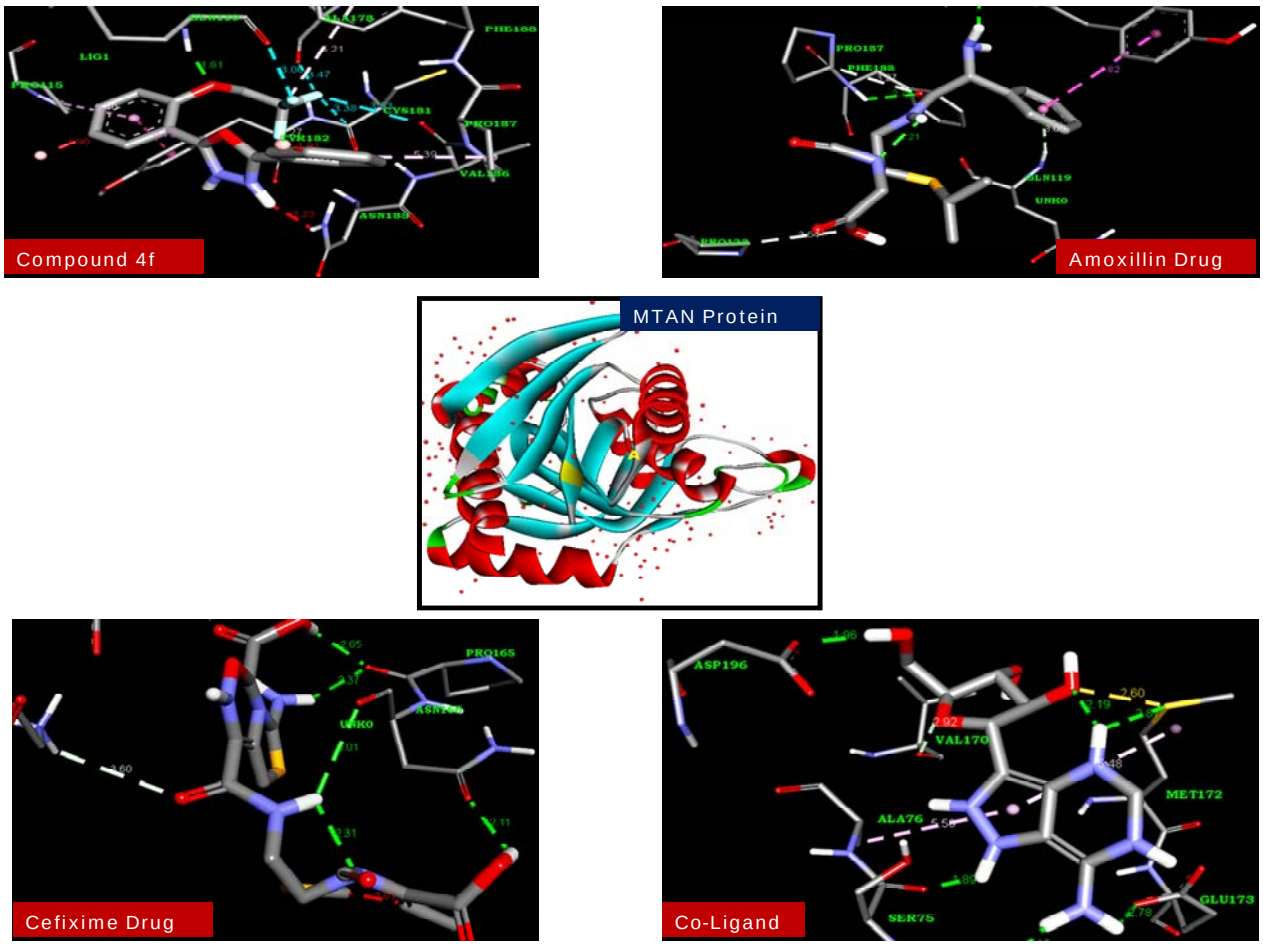


Figure:3 DNA gyrase B of S.aureus, Binding pose of Amoxillin with MTAN , Binding pose of Cefixime with MTAN, Binding pose of co-crystal structure ligand with MTAN and Binding pose of 1,3,4-Oxadiazole derivatives(3a-g) with MTAN

Table: 3 Interaction pattern of 1,3,4-Oxadiazole derivatives(3a-g) with MTAN protein

COMPOUNDS	Hydrogen Bond Interaction	Other Interaction
3a	GLN 119	VAL 186, ASN 185, PRO 187, CYS 181, PHE 188, ALA 178, TYR 182, PRO 115
3b		ASN 185, PRO 187, CYS 181, PHE 188, ALA 178, TYR 182, PRO 115
3c	GLN 119, SER 121	VAL 186, ASN 185, PRO 187,PHE 188, TYR 182
3d	CYS 181	VAL 186, ASN 185, PRO 187, PHE 188, ALA 178, TYR 182, PRO 115,GLN 119
3e	GLN 119	ASN 185, PRO 187, PHE 188, ALA 178, TYR 182, PRO 115
3f	GLN 119	VAL 186, ASN 185, PRO 187, CYS 181, PHE 188, ALA 178, TYR 182
3g	GLN 119	ASN 185, PRO 187, PHE 188, ALA 178, TYR 182, PRO 115
Amoxillin	GLN 119, CYS 181, PRO 187, PHE 188, PRO 123	TYR 182
Cifixime	PRO 165, ASP 80, ASN 166, LYS 140	
Co-crystallized ligand	SER 75, ASP 196, MET 172, VAL 170, GLU 11, GLU 173	ALA 76

The MTAN Compound 3c complex is mostly stabilized by hydrophobic and van der Waals interactions resulting in a binding score of -7.4. In order compare the binding mode of 1,3,4-Oxadiazole derivative with the already established inhibitor for this enzyme, Formycin A. The docking results showed that our compounds also bind with the MTAN of S.aureus in a almost similar way as like the Formycin A. (binding score –6.7).

We also analyzed the interaction and energy profile of MTAN with the already available drug Amoxillin and cefixime (Figure 3). The docking of Amoxillin and cefixime with MTAN of S.aureus resulted in a binding score of -6.9 and -6.5, which is less binding score results found for compound 3f,3c and 3g.

Antibacterial activity

Table:4 Minimum inhibitory concentrations (MIC) of chalcone imine derivatives (3a-g) against Staphylococcus aureus

Compound	***Streptococcus aureus***		
	Zone of inhibition (mm)		
	50µg/ml	**100** µg/ml	**150** µg/ml
3a	12	18	18
3b	14	18	20
3c	09	14	16
3d	11	14	16
3e	15	17	20
3f	12	17	21

3g	17	22	24
Amoxillin (100µg/ml)		28	
Cefixime(100) µg/ml		26	
Control	-	-	-

All the synthesized compounds were studied for their antibacterial activity against clinically isolated Gram-positive strains S. aureus using disc diffusion method. The minimum inhibitory concentration (MIC) values were calculated by comparison to amoxicillin and cefixime as the reference bacterial drug and they are shown in Table 4. Compounds 3a-3e have shown antibacterial activity with MIC value in the range of 100 µg/mL against Gram-positive S. aureus. Compounds 3g and 3e have shown antibacterial activity with MIC value in the range of 50µg/mL against Gram-positive S. aureus. GyrB has been proposed as the main binding target for antibacterial activity[20]. Molecular docking described an explanation for the mechanism for synthesized derivatives antibacterial activities by illustrating the interaction between 1,3,4-Oxadiazole with GyrB and MTAN. From the docking study we predicted that 1,3,4-Oxadiazole analogues (3g-e) possess better antibacterial activity equal to the standard drugs by having good binding affinity with target protein and it could be used as potential drug as antimicrobial. Amongst all the docked compounds, compound 3g,3c and 3e shows good binding affinity and interaction with DNA gyrase and MTAN enzymes with reference to amoxicillin and cefixime. It was found that the predicted docking results using autodock 4 Software were quite accurate after comparing it with the actual antimicrobial activity.

Conclusion

We have demonstrated a highly efficient green catalytic approach for the one-pot synthesis of 1,3,4-Oxadiazole derivatives. Docking analysis of all the seven 1,3,4-Oxadiazole derivatives (3a-g), two drugs Amoxillin and Cefixime, co-crystallised ligands of DNA Gyrase B and MTAN with the protein targets. The docking studies with the structure of 1,3,4-Oxadiazole derivatives show that the compound 3g,3c and 3e being small and compact molecule are located deep into the active site of the MTAN and DNA gyrase B of S.aureus. In order to compare 1,3,4-Oxadiazole derivatives for its efficiency standard drugs amoxillin, cefixime and co crystallized ligands methyl ((5-(4-(4-hydroxypiperidin-1-yl)-2-phenylthiazol-5-yl)-1H-pyrazol-3-yl)methyl)carbamate and Formycin A is included in our docking studies and show almost equal binding energy with synthesized derivatives. The anti-bacterial activity of synthesized novel 1,3,4-Oxadiazole derivatives were effectively screened against Gram positive S. aureus bacterial strain. Most of these compounds show moderate antibacterial activity comparable with to marketable compounds. The zone of inhibition of tested compounds shows, the 1,3,4-Oxadiazole derivative 3g,3c and 3e encompass potent bio-activities against bacterial strain.

Reference

1. S. Bajaj, V. Asati, J. Singh, P. P. Roy, Eur. J. Med. Chem. 97 **(**2015) 124-141.
2. S. Rapolu, M. Alla, V. R. Bommena, R. Murthy, N. Jain, V. M. Bommareddy, M. R. Bommineni, Eur. J. Med. Chem. 66 **(**2013) 91-100.
3. R. A. Rane, P. Bangalore, S. D. Borhade, P. K. Khandare, Eur. J. Med. Chem. 70 (2013) 49-58.
4. P. Singh, P. K. Sharma, J. K. Sharma, A. Upadhyay, N. Kumar, Org. Med. Chem. Lett. 2 (2012) 1-10.
5. G. G. Ladani, M. P. Patel, New J. Chem. 39 (2015) 9848-9857.
6. S. Borg, G. Estenne-Bouhtou, K. Luthman, I. Csoregh, W. Hesselink, U. Hacksell, J. Org. Chem. 60 **(**1995) 3112-3120.
7. H. Lai, D. Dou, S. Aravapalli, T. Teramoto, G. H. Lushington, T. M. Mwania, K. R. Alliston, D. M. Eichhorn, R. Padmanabhan, W. C. Groutas, Bioorg. Med. Chem. 21 **(**2013) 102-113
8. Somani Rakesh R., Agrawal Anuj G., Kalantri Pushkar P., Gavarkar Pratibha S. and Clerq Erik De, Investigation of 1,3,4-OxadiazoleScaffold as Potentially Active Compounds, *International Journal of Drug Design and Discovery*, 2011; 2(1), 353-36

9. Sutcliffe JA, Gootz TD, Barrett JF (1989) Biochemical characteristics and physiological significance of major DNA topoisomerases. Antimicrob Agents Chemother 33: 2027-2033.
10. Advances In Protein Chemistry, Volume 38 (Google eBook). (1986) Academic Press
11. Sufrin JR, Meshnick SR, Spiess AJ et al. Methionine recycling pathways and antimalarial drug design. Antimicrob Agents Chemother 1995; 39: 2511–5.
12. Duerre JA. A hydrolytic nucleosidase acting on S-adenosylhomocysteine and on 50 methylthioadenosine. J Biol Chem 1962; 237: 3737–41.
13. Gianotti AJ, Tower PA, Sheley JH et al. Selective killing of Klebsiella pneumoniae by 5-trifluoromethylthioribose. Chemotherapeutic exploitation of the enzyme 5-methylthioribose kinase. J Biol Chem 1990; 265: 831–7.
14. Singh V, Lee JE, Nunez S et al. Transition state structure of 50-methylthioadenosine/S-adenosylhomocysteine nucleosidase from Escherichia coli and its similarity to transition state analogues. Biochemistry 2005; 44: 11647–59
15. Morris GM, Huey R, Lindstrom W, Sanner MF, Belew RK, Goodsell DS, et al. AutoDock4 and AutoDockTools4: Automated docking with selective receptor flexibility. J Comput Chem. 2009;30:2785–2791.
16 Trott O, Olson AJ. AutoDock Vina: Improving the speed and accuracy of docking with a new scoring function, efficient optimization, and multithreading. J Comput Chem. 2010;31:455–461.
17 Bernstein FC, Koetzle TF, Williams GJB, Meyer EF, Brice MD, et al. (1977) The Protein Data Bank. A Computer-Based Archival File for Macromolecular Structures. J Mol Biol 112: 535-542.
18 D. Kaushik, S.A. Khan, G. Chawla and S. Kumar, Eur. J. Med. Chem.,45, 3943 (2010).
19 S. Han, F.-F. Zhang, X. Xie and J.-Z. Chen, Eur. J. Med. Chem., 74, 73(2014)
20 Ohemeng K. A. et al. . DNA gyrase inhibitory and antimicrobial activities of some diphenic acid monohydroxamides. J. Med. Chem. 40, 3292–3296 (1997).