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Short communication

Synthesis, characterization, *in vitro* and molecular docking studies of new 2,5-dichloro thienyl substituted thiazole derivatives for antimicrobial properties

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ABSTRACT

A new series of 2-substituted 4-(2,5-dichloro thienyl)-1,3-thiazoles are synthesized by the reaction of 2-bromo-1-(2,5-dichlorothien-3-yl) ethanone with thiourea and substituted thioamides. The newly synthesized compounds **4a**–**e** are characterized by analytical ^{1}H NMR, ^{13}C NMR and mass spectral data. The newly synthesized compounds are screened for antifungal and antibacterial activities. Among them **4a** and **4d** exhibited good antifungal and antibacterial activities. The newly synthesized compounds are subjected to molecular docking studies for the inhibition of the enzyme L-glutamine: D-fructose-6-phosphate amidotransferase [GlcN-6-P] (EC 2.6.1.16) which is a new target for antifungals. Among the five molecules taken for docking studies 2-(8-quinolinyl)-4-(2,5-dichloro thienyl)-1,3-thiazole **4d** shows minimum binding and docking energy and may be considered as good inhibitor of GlcN-6-P synthase.

1. Introduction

Fungal infections are a growing problem in contemporary medicine, yet only a few antifungal agents are used in clinical practice. Although several antifungal agents, such as amphotericin B and the azole class of drugs are currently available there is clearly a critical need for the development of new specific antifungal agents. Heterocycles containing a thiazole ring system are found to exhibit a wide spectrum of biological activities, including antibacterial and antifungal activities. Many thiazole-containing compounds are reported as herbicidal, fungicidal, antitubercular, antiallergic, anti-anaphylactic, antiarthritic, antibiotic, antiviral, anti-inflammatory, analgesic and psychotropic agents [1–13]. Narayana et al. [14–16] synthesized series of new thiazole derivatives and some of them showed excellent antibacterial and antifungal activities. So it was planned to synthesize new thiazole derivatives with 2,5-dichloro thienyl substitution and test them for both *in vitro* and *in silico* antifungal and antibacterial activities. The enzyme, namely L-glutamine: D-fructose-6-phosphate amidotransferase, known under the trivial name of glucosamine-6-phosphate synthase (EC 2.6.1.16), is a new target for antifungals [17–19]. This protein is a complex enzyme. It catalyses a complex reaction involving ammonia transfer from L-glutamine to Fru-6-P, followed by isomerisation of the formed fructosamine-6-phosphate to glucosamine-6-phosphate [20,21]. This reaction is the first step of the pathway leading to the formation of UDP-GlcNAc, a product that is present in all class of organisms, but in fungi and bacteria it is used to build macromolecules necessary for the cell wall assembly, such as chitin and mannoproteins in fungi and peptidoglycan in bacteria.

In mammals, UDP-GlcNAc is utilized for biosynthesis of glycoproteins and mucopolysaccharides. In spite of the fact that glucosamine-6-phosphate synthase is present in all kinds of cells, it may be exploited as a target for potential antifungal drugs and selective toxicity can be achieved [22]. Obviously, glucosamine-6-phosphate, the product of this enzyme, is indispensable for fungi as well as for human cells, yet the consequences of its deficiency in both species are very different. It has been shown that even a short time inactivation of GlcN-6-P synthase is lethal for fungal cells (it induces morphological changes, agglutination and lysis), while in mammals depletion of the aminosugar pool for a short time is not lethal, because of the much longer lifespan of mammalian cells, long half lifetime of GlcN-6-P synthase, and rapid expression of the mammalian gene encoding the enzyme GlcN-6-P synthase [23]. Hence, the molecular docking studies of newly synthesized compounds were carried out and results of such studies are reported.

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2. Results and discussion

2.1. Chemistry

The new series of 2-substituted- 4-(2,5-dichloro thienyl)-1,3-thiazoles **4a**–**e** were synthesized by the reaction of 2-bromo-1-(2,5-dichlorothien-3-yl) ethanone with thiourea and substituted thioamides (Scheme 1). The structures of title compounds were confirmed by analytical, ^{1}H NMR, ^{13}C NMR and ESI-MS mass spectrometry and elemental analysis. The ^{1}H NMR of compound **4a** showed signals at δ 7.13 a singlet for thiophene proton and another singlet at 7.36 for thiazole protons respectively while the signal for amino group appeared with down field shift as a singlet at 7.64 which confirmed the structure of the molecule. It was further supported by recording ^{13}C NMR spectrum and the signals appeared in the spectrum account for all the C-atoms present in the molecule **4a**. The DEPT spectrum showed two signals at 106.81 and 127.95 accounting for thiophene and thiazole CH respectively. LC mass spectrum of the compound **4a** showed molecular ion peaks *m*/*z* 251 [M$^+$], 253 [M+2], 255 [M+4]. This is in agreement with its respective molecular formula $C_7H_4Cl_2N_2S_2$. The ^{1}H NMR was recorded in the DMSO-d_6 (400 MHz) for compound **4b**. The signal appeared in the region δ 7.19–7.44 as a multiplet was due to 2H of 2-fluorophenyl ring, a doublet of doublet resonated at δ 7.44–7.58 ($J = 8.2$ Hz) for 1H, of 2-fluorophenyl moiety and signal at δ 7.69 was appeared as a singlet also due to 1H of thiophene. A triplet at δ 8.27–8.34 could be attributed to another proton on the fluorophenyl ring and a singlet appeared at δ 8.36 was due to thiazole ring proton. The LC mass showed molecular ion peaks at MS (LCMS) *m*/*z* 330.2 [M+], 332.2 [M+2], 334.2 [M+4] further confirmed the formation of new compound **4b**. Similarly, the other newly synthesized compounds were characterized and spectral data are given in the Experimental section.

2.2. Biological evaluation

2.2.1. Antifungal studies

Compounds **4a**–**e** were screened for their antifungal activity against *Aspergillus flavus* (NCIM No. 524), *Aspergillus fumigatus* (NCIM No. 902), *Penicillium marneffei* (recultured) and *Trichophyton mentagrophytes* (recultured). The compounds were dissolved in DMSO and antimicrobial activity was determined by serial plate dilution method [24]. Among the tested compounds, the compound 4-(2,5-dichlorothienyl-3-yl)-2-amino-1,3-thiazole **4a** and 4-(2,5-dichlorothienyl-3-yl)-2-(8-quinolinyl)-1,3-thiazole **4d** have emerged as active against all tested microorganisms, whereas 4-(2,5-dichlorothienyl-3-yl)-2-(2-methoxyphenyl)-1,3-thiazole **4c** showed moderate activity. 4-(2,5-Dichlorothienyl-3-yl)-2-(2-fluorophenyl)-1,3-thiazole **4b** and 4-(2,5-dichlorothienyl-3-yl)-2-(3-pyridinyl)-1,3-thiazole **4e** did not show any activity.

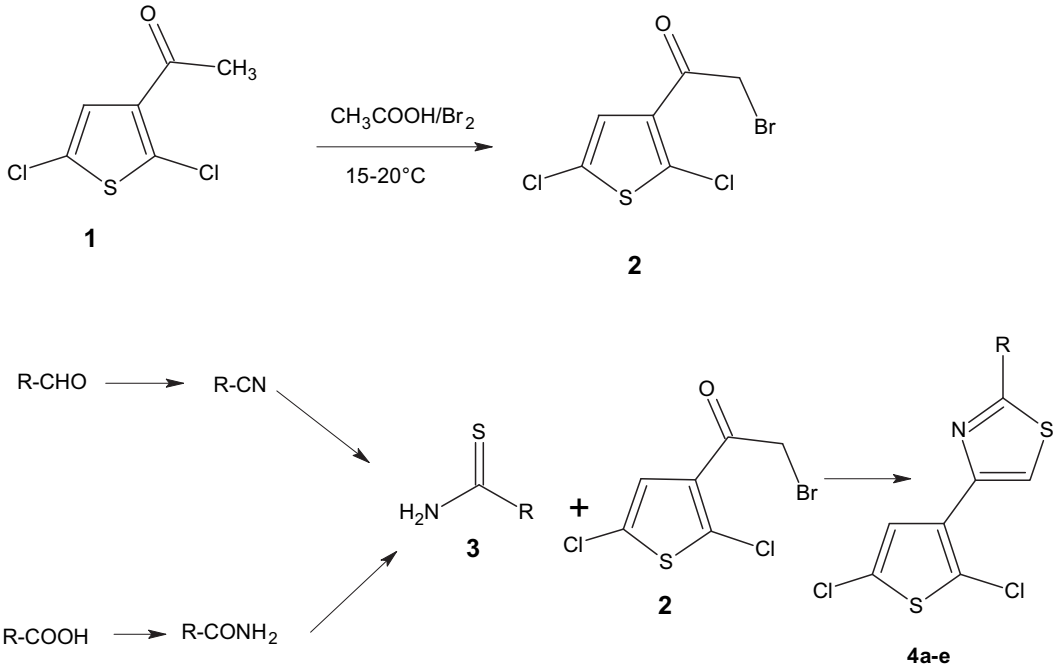
2.2.2. Antibacterial studies

The newly synthesized compounds were also screened for their antibacterial activity against *Escherichia coli*, *Staphyllococcus aureus* (Smith), *Psuedomonus aeruginosa* (Gessard), *and Klebsiella pneumoniae* (Friedlander) bacterial strains by disc diffusion method [25,26]. Among the tested compounds, the compounds **4a** and **4d** have emerged as active against all tested microorganisms and **4c** showed moderate activity whereas **4b** and **4e** did not show any activity.

The enhanced antifungal and antibacterial activities of the **4a** and **4d** could be attributed to the presence of amino and 8-quinolinyl moieties. Quinolone derivatives like ciprofloxacin, nalidixic acids are known antimicrobial drugs. Hence the qunolinyl substitution may be the reason for highest activity of **4d** among the tested compounds. However, based on this promising observation, it is immature to arrive at the conclusion on structure activity aspect of these molecules and further evaluation is needed to use them for clinical use.

2.2.3. Molecular docking studies

With *in vitro* antimicrobial results in hand it is thought worthwhile to do *in silico* studies to support the *in vitro* activity. Automated docking was used to determine the orientation of inhibitors bound in the active site of GlcN-6-P synthase. A Lamarckian genetic algorithm method, implemented in the program AutoDock 3.0, was employed. Fig. 1 shows structure of bacterial glucosamine-6-phosphate synthase (PDB ID 1jxa). The docking of ligand molecules **4a**–**e** with GlcN-6-P synthase reveals that all the inhibitor compounds are



Scheme 1. Preparation of 4-(2,5-dichlorothien-3-yl)-2-amino/substituted aryl-1,3-thiazole **4a**–**e**.

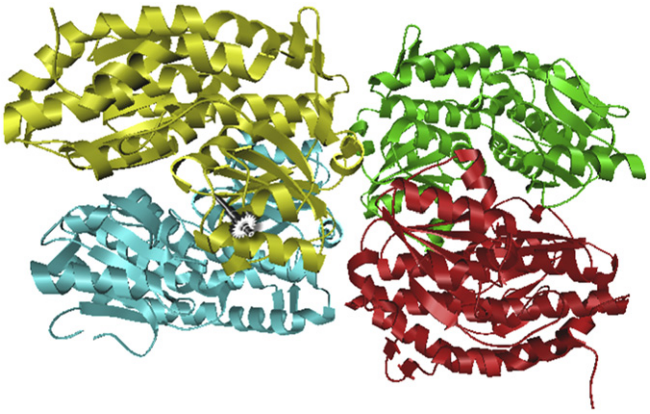


Fig. 1. Structure of bacterial glucosamine-6-phosphate synthase (PDB ID 1jxa).

exhibiting the bonding with one or the other amino acids in the active pockets which is showed in Fig. 2. The ligand molecules were designed and the structure was analyzed using ChemDraw Ultra 6.0. 3D, coordinates were prepared using PRODRG server. The protein structure file (PDB ID: 1gdo) taken from PDB (www.rcsb.org/pdb) was edited by removing the hetero atoms, adding C-terminal oxygen. Fig. 3 shows the *in silico* active pocket prediction of amino acids of protein GlcN-6-P synthase involved in binding with the ligands obtained from PDB sum. For docking calculations, Gasteiger-Marsili partial charges were assigned to the ligands and nonpolar hydrogen atoms were merged. All torsions were allowed to rotate during docking. The Lamarckian genetic algorithm and the pseudo-Solis and Wets methods were applied for minimization, using default parameters. Theoretically all the five molecules showed very good binding energy and docking energy ranging from $-6.29\ kJ\ mol^{-1}$ to $-8.21\ kJ\ mol^{-1}$ and $-7.71\ kJ\ mol^{-1}$ to $-9.18\ kJ\ mol^{-1}$ respectively. Among the five molecules, docking of GlcN-6-P synthase with **4d** revealed that its docking energy and binding energy were $-9.18\ kJ\ mol^{-1}$ and $-8.21\ kJ\ mol^{-1}$ respectively

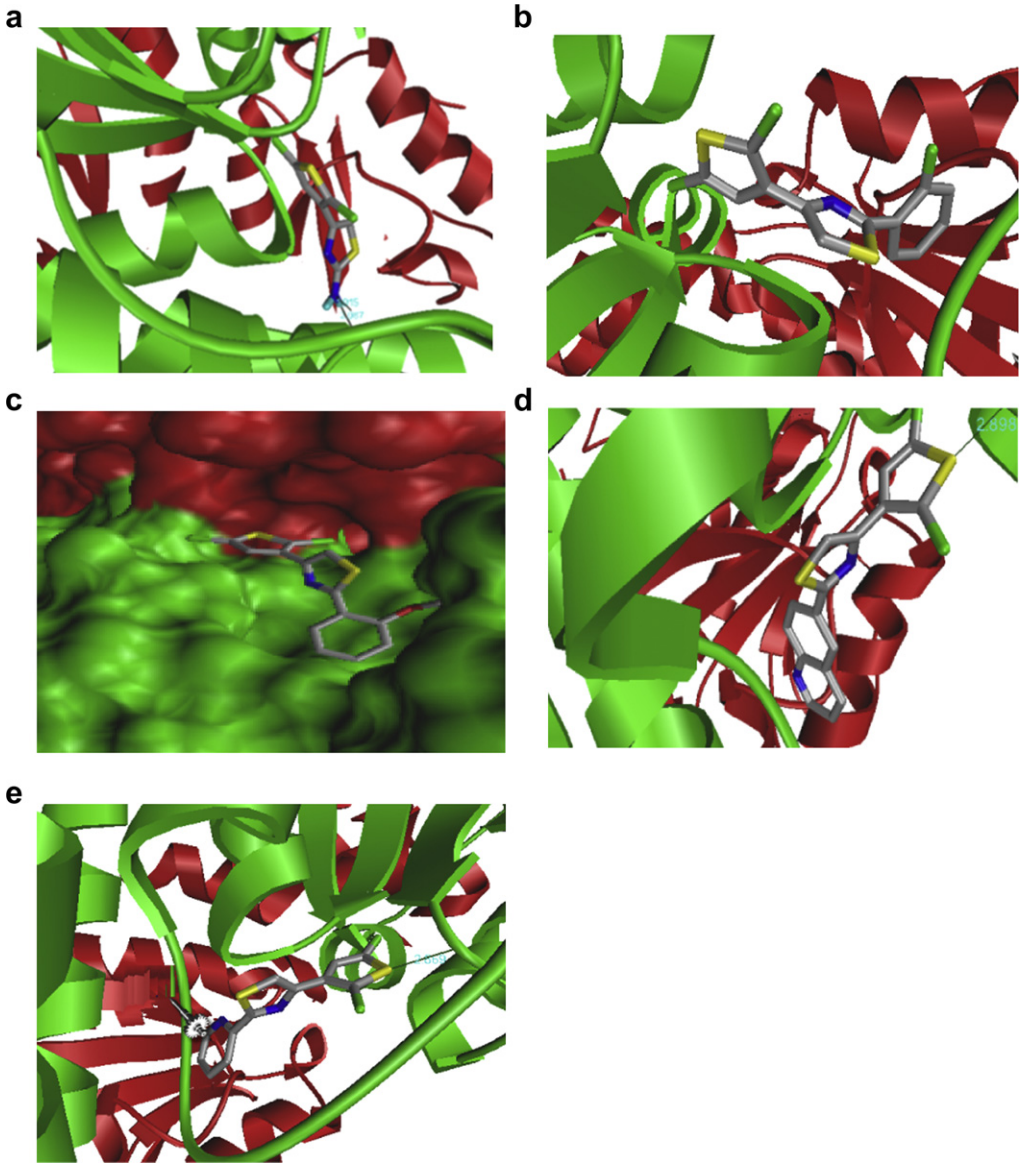


Fig. 2. Interaction of **4a**–**e** with GlcN-6-P. a. Interaction of **4a** with GlcN-6-P. b. Interaction of **4b** with GlcN-6-P. c. Interaction of **4c** with GlcN-6-P. d. Interaction of **4d** with GlcN-6-P. e. Interaction of **4e** with GlcN-6-P.

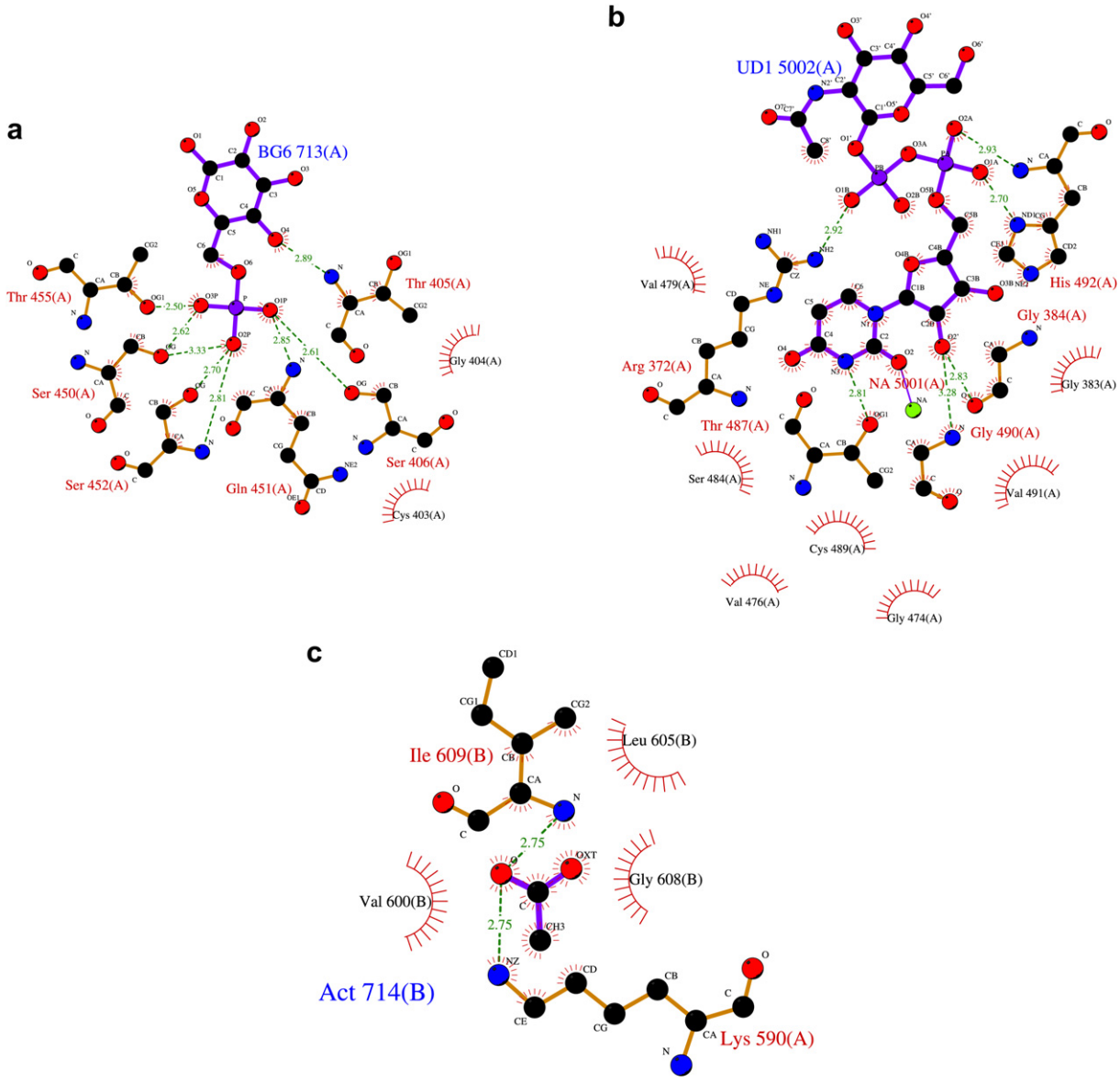


Fig. 3. Ligplot results for GlcN-6-P. a. Showing the binding of ligand BG6 on A-chain amino acids present in an active pocket of GlcN-6-P with eight hydrogen bonds. b. Binding of ligand UDI on A-chain amino acids present in an another active pocket of GlcN-6-P with six hydrogen bonds. c. Showing the binding of ligand ACT on B-chain amino acids present in an active pocket of GlcN-6-P with two hydrogen bonds.

and it may be considered as good inhibitor of GlcN-6-P synthase. In *in vitro* studies also **4d** has emerged as active against all tested microorganisms, so it can be predicted as the activity may be due to inhibition of enzyme GlcN-6-P synthase, which catalyses a complex reaction involving ammonia transfer from L-glutamine to Fru-6-P, followed by isomerisation of the formed fructosamine-6-phosphate to glucosamine-6-phosphate.

3. Conclusion

Five new compounds containing 2,5-dichloro thienyl substituted thiazole ring system, which resemble known antifungal agent N-[4-(4-chlorophenyl)-2-thiazolyl]-salicylamide were synthesized and tested for antimicrobial activities. Among the tested compounds, 4-(2,5-dichlorothienyl-3-yl)-2-(8-quinolinyl)-1,3-thiazole **4d** has emerged as most active against all tested microorganisms. Molecular docking studies also revealed that **4d** has minimum binding and docking energy and may be considered as a good inhibitor of GlcN-6-P. Hence this study has widened the scope of developing these thiazole derivatives as promising antifungal and antibacterial agents.

4. Experimental

4.1. Chemistry

Melting points were recorded on Electrothermal melting point apparatus and are uncorrected. ^{1}H and ^{13}C NMR spectra were recorded on Jeol 400 MHz Perkin Elmer spectrometer at IISc, Bangalore, Karnataka, India. Chemical shifts are shown in δ values (ppm) with tetramethylsilane (TMS) as internal standard. ^{13}C (100 MHz) NMR spectra were recorded for approximately 0.03 M solutions in DMSO-d_6 at 100 MHz with TMS as internal standard. LCMS were obtained using Agilent 1200 series LC and Micromass zQ spectrometer at IISc, Bangalore, Karnataka, India. Column chromatography was performed using a silica gel (230–400 mesh). Elemental analysis was carried out using VARIO EL-III (Elementar

Analysensysteme GmBH) at Department of Chemistry, Mangalore University, Mangalore, Karnataka, India.

All chemicals were purchased from Sigma-Aldrich Co., U.S.A., and all solvents for column chromatography were of reagent grade, and were purchased from commercial sources.

*4.1.1. Procedure for the synthesis 2-bromo-1-(2,5-dichlorothien-3-yl) ethanone (**2**)*

To a solution of 1-(2,5-dichlorothien-3-yl) ethanone (1 equiv, 0.256 mol) in acetic acid (150 ml) at 15–20 °C was added bromine (49 g [16 ml], 1.2 equiv, 0.3072 mol) drop wise over a period of 1 h. Further the reaction was maintained at 20–25 °C till completion (TLC). After completion the mass was quenched to water. The organic layer was separated. The solvent was removed under reduced pressure. Solid obtained was recrystallized in ethanol. Yield: 60 g (85%). Mp: 40–45 °C.

*4.1.2. General procedure for synthesis of substituted thioamides (**3**)*

(i) To a solution of aromatic aldehyde (1 equiv) in formic acid (5 volumes) was added hydroxylamine·HCl (1.5 equiv) and the reaction mass was refluxed for 16 h. The reaction mass was quenched to cold water. Extracted in ethyl acetate and dried over anhydrous sodium sulphate and distilled to give corresponding nitriles. To a solution of nitrile (1 equiv) in ethanol (5 volumes) was added phosphorouspentasulfide (2 equiv) in lots maintaining temperature below 50 °C. After the addition, reaction mixture was refluxed for 3 h and solvent was removed under reduced pressure and quenched into water and extracted in methylene dichloride and dried over anhydrous sodium sulphate and evaporated to isolate the product.

(ii) Carboxylic acid (1 equiv) is refluxed in thionyl chloride (5 volumes) for 2 h under dry conditions. The excess thionyl chloride was distilled off and the obtained acid chloride was quenched in liquor ammonia under cooling. Acetone was added to precipitate the amide.

Table 1
Characterization data of 4-(2,5-dichlorothien-3-yl)-2-amino/substituted aryl-1,3-thiazole **4a**–**e**.

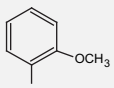
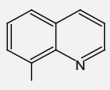
Compound no.	R	Mp	% Yield	Elemental analysis found [calculated]		
				C	H	N
4a	$-NH_2$	200–202	80	33.46 [33.48]	1.63 [1.61]	11.18 [11.15]
4b		180–183	85	47.25 [41.28]	1.79 [1.83]	4.29 [4.24]
4c		175–176	78	49.10 [49.13]	2.66 [2.65]	4.05 [4.09]
4d		218–220	75	52.93 [52.90]	2.25 [2.22]	7.75 [7.71]
4e		190–192	85	46.05 [46.01]	1.90 [1.93]	8.90 [8.94]

Table 2
Antifungal activity of the compounds **4a**–**e** (MIC in μg/mL).

Compounds	*Penicillium marneffei (recultured)*	*Trichophyton mentagrophytes (recultured)*	*Aspergillus flovus*	*Aspergillus fumigatus*
4a	12.5	12.5	6.25	6.25
4b	R	R	R	R
4c	12.5	12.5	100	R
4d	6.25	6.25	6.25	12.5
4e	R	R	R	R
Amphotericin B	<6.25	<6.25	<6.25	<6.25

R = Resistant.

To a solution of amide (1 equiv) in THF was added phosphorous penta sulfide (0.3 equiv) and the reaction mass was refluxed till amide absence. The reaction mass was quenched in cold water and extracted in methylene dichloride. The organic layer was filtered through hyflo and dried over anhydrous sodium sulphate and evaporated to isolate the thioamide.

*4.1.3. General procedure for the synthesis of 4-(2,5-dichlorothien-3-yl) -2-amino/substituted aryl-1,3-thiazole (**4**)*

2-Bromo-1-(2,5-dichlorothien-3-yl) ethanone (**2**) (1 equiv) was dissolved in ethanolic KOH and thioamide (**3**) (1 equiv) was added and the reaction mass was refluxed for 3 h. After the reaction, mass was cooled and the precipitated product was filtered under suction and washed with ethanol and dried. Characterization data are given in Table 1.

*4.1.3.1. 4-(2,5-Dichlorothien-3-yl)-2-amino-1,3-thiazole(**4a**).* ^{1}H NMR (DMSO, 400 MHz): δ 7.13 (s, 1H, thiophene), 7.36 (s, 1H, thiazole), 7.64 (s, 2H, NH_2 proton). ^{13}C NMR (DMSO, 100 MHz): δ 106.81, 122.17, 125.69, 127.96, 130.62, 137.40, 169.0 DEPT (DMSO, 100 MHz) δ 106.81 (thiophene CH), 127.95 (thiazole CH), MS (LCMS): *m*/*z* 251 [M^+], 253 [M+2], 255 [M+4].

*4.1.3.2. 4-(2,5-Dichlorothien-3-yl)-2(2-fluorophenyl)-1,3-thiazole (**4b**).* ^{1}H NMR (DMSO, 400 MHz): δ 7.19–7.44 (m, 2H, 2-fluorophenyl), 7.44–7.589 (dd, J = 8.2 Hz, 1H, 2-fluorophenyl), 7.69(s, 1H, thiophene), 8.27–8.34 (t, 1H, 2-fluorophenyl), 8.36 (s, 1H, thiazole). MS (LCMS): *m*/*z* 330.2 [M^+], 332.2 [M+2], 334.2 [M+4].

*4.1.3.3. 4-(2,5-Dichlorothien-3-yl)-2(2-methoxyphenyl)-1,3-thiazole (**4c**).* ^{1}H NMR (DMSO,400 MHz): δ 7.07–7.14 (t, 1H, 2-methoxyphenyl), 7.25–7.27 (d, J = 8 Hz, 1H, 2-methoxyphenyl), 7.47–7.51 (t, 1H, 2-methoxyphenyl), 7.70 (s, 1H, thiophene), 8.19 (s, 1H, thiazole), 8.38–8.40 (d, J = 8 Hz, 1H, 2-methoxyphenyl), 4.03 (s, 3H, methoxy). ^{13}C NMR (DMSO, 100 MHz): δ 56.43, 112.73, 119.28, 121.34, 121.45, 125.70, 128.29, 128.48, 131.92, 131.91, 146.13, 156.69, 161.12.

DEPT (DMSO, 100 MHz): δ 56.43(OCH_3), 112.73, 119.27, 121.45, 128.29, 128.49, 131.91(CH). MS (LCMS): *m*/*z* 342.1 [M^+], 344.1 [M+2], 346.1[M+4].

Table 3
Antibacterial activity of the compounds **4a**–**e** (MIC in μg/ml).

Compounds	*Staphylococcus aureus*	*Escherichia coli*	*Pseudomonas aeroginosa*	*Klebsiella pneumoniae*
4a	6.25	6.25	12.5	12.5
4b	100	R	R	R
4c	6.25	6.25	12.5	100
4d	6.25	12.5	12.5	6.25
4e	100	R	100	R
Ampicillin	<6.25	<6.25	<6.25	<6.25

R = Resistant.

Table 4
Molecular docking results with Glucosamine-6-phosphate synthase.

Mol. no	Binding energy	Docking energy	Inhibitory constant	Intermol energy	RMS	H-bonds	Bonding
4a	−7.28	−7.71	4.59e-006	−7.59	0.0	2	AF1::DRG1:HAB:G6P:A:GLU591:OE2 AF1::DRG1:NAA:G6P:A:SER503:HN
4b	−7.0	−7.93	7.43e-006	−7.62	0.0	0	==========
4c	−6.87	−7.97	9.17e-006	−7.81	0.0	1	AF3::DRG1:OAK:G6P:A:THR405:HN
4d	−8.21	−9.18	9.57e-007	−8.83	0.0	1	AF4::DRG1:NAK:G6P:A:GLN451:HN
4e	−7.11	−8.02	6.15e-006	−7.73	0.0	1	AF5::DRG1:SAL:G6P:A:SER450:OG

*4.1.3.4. 4-(2,5-Dichlorothien-3-yl)-2-(8-quinolinyl)-1,3-thiazole (**4d**).* ^{1}H NMR (DMSO, 400 MHz): δ 7.92–8.02 (m, 2H, quinoline), 8.20 (s, 1H, thiophene), 8.43–8.45 (d, $J = 8$ Hz, 1H, quinoline), 8.64–8.66 (d, $J = 8$ Hz, 1H, quinoline), 9.03–9.05 (d, $J = 8$ Hz, 1H, quinoline), 9.23 (d, $J = 4$ Hz, 1H, quinoline), 9.44 (s, 1H, thiazole). ^{13}C NMR (DMSO, 100 MHz): δ 122.81, 128.327, 129.28, 133.52, 134.40, 140.14, 144.28, 148.77, 167.99. DEPT (DMSO, 100 MHz): δ 122.81, 128.33, 133.52, 134.41, 144.311, 148.75. MS (LCMS): *m/z* 173(quinoline acid fragment), no molecular ion peak.

*4.1.3.5. 4-(2,5-Dichlorothien-3-yl)-2(3-pyridyl)-1,3-thiazole (**4e**).* ^{1}H NMR (DMSO, 400 MHz): δ 7.81 (s, 1H, thiophene), 8.38–8.42 (t, 1H, pyridine), 8.52(s, 1H, thiazole), 9.04–9.035(d, 1H, $J = 5.84$ Hz, pyridine), 9.28–9.26 (d, 1H $J = 8.3$ Hz, pyridine), 9.73 (s, 1H, pyridine). MS (LCMS): *m/z* 313.22 [M^+], 315.22, [M+2], 317.22 [M+4].

4.2. Antifungal activity

Antifungal activity for newly prepared compounds was screened by serial plate dilution method. Sabourands agar media was prepared by dissolving peptone (1 g), D-glucose (4 g) and agar (2 g) in distilled water (100 ml) and adjusting the pH to 5.7. Normal saline was used to make a suspension of spores of fungal strain for lawning. A loopful of particular fungal strain was transferred to 3 ml saline to get a suspension of corresponding species. A 20 ml of agar media was poured in to each of the petridishes. Excess of suspension was decanted and the plates were dried by placing in an incubator at 37 °C for 1 h. Using an agar punch wells were made on these seeded agar plates and 10 μg/ml of the test compounds in DMSO were added into each well labeled. A control was also prepared for the plates in the same way using solvent DMSO. The petridishes were prepared in triplicate and maintained at 37 °C for 3–4 days. Antifungal activity was determined by measuring the diameter of the inhibition zone. Activity of each compound was compared with Amphotericin B as standard. The minimum inhibitory concentration (MIC) for the Amphotericin B in DMSO was more than 1 μg/ml against the tested species. The MIC values of tested compounds are given in Table 2.

4.3. Antibacterial activity

The newly synthesized thiazoles were screened for their antibacterial activity against bacterial strains by disc diffusion method. The discs measuring 6.25 mm in diameter were punched from Whatman No. 1 filter paper. Batches of 100 discs were dispensed to each screw capped bottles and sterilized by dry heat at 140 °C for an hour. The test compounds were prepared with different concentrations using dimethyl formamide. One millilitre containing 100 times the amount of chemical required in each disc was added to each bottle which contains 100 discs. The discs of each concentration were placed in triplicate in nutrient agar medium seeded with fresh bacteria separately. The incubation was carried out at 37.8 °C for 24 h. Nitrofurazone was used as a standard drug. Solvent and growth controls were kept. The zone of inhibition and minimum inhibitory concentrations [MIC] was noted. The MIC values of tested compounds are given in Table 3.

4.4. Molecular docking studies

The synthesized molecules were subjected for molecular docking by calculating the minimum energy to inhibit the target protein involved in the catalysis of complex reaction involving ammonia transfer from L-glutamine to Fru-6-P, followed by isomerisation of the formed fructosamine-6-phosphate to glucosamine-6-phosphate. The ligands were drawn in ChemDraw Ultra 6.0 assigned with proper 2D orientation (ChemOffice package) and the structure of each ligand was analyzed by using Chem-3D Ultra 6.0 (ChemOffice package) and was checked for the connection error in bond order. ADMET property was achieved through PreADMET server-a web-based application for predicting ADMET data and building drug-like library using *in silico* method. Energy of the molecules was minimized using Dundee PRODRG2 Server. Then the file was opened in SPDB viewer and C-terminal Oxygen was added using fit module property. Active pockets were identified and ligplot of PdbSum provided in the External links of PDB for the proteins was downloaded from PDB. CASTp (Computed Atlas of Surface Topography of proteins) server was used to crosscheck the active pockets on target protein molecules. Autodock V3.0 was used to perform Molecular Docking. The docking results for ligand molecules against glucosamine-6-phosphate synthase [PDB Id: 1jka], showed minimum docking energy, binding energy, inhibition constant, intermolecular energy with 0.0 RMS as documented in Table 4.

Conflict of interest

Authors declare that there are no conflicts of interest.

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