https://doi.org/10.48047/AFJBS.6.14.2024.5161-5176

Design, Synthesis and Exploration of Antibacterial and Antifungal Activity of N'-[2-(Substituted Sulfanyl) Quinazolin-4-yl] Pyridine-4-Carbohydrazide Derivatives.

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Volume 6, Issue 14, Aug 2024

Received: 15 June 2024

Accepted: 25 July 2024

Published: 15 Aug 2024

doi: 10.48047/AFJBS.6.14.2024.5161-5176

ABSTRACT

The emerging resistance to currently available antimicrobial drug demands the synthesis of new agents for microbial infections. In the current research work, a novel series of *N*'-[2-(substituted sulfanyl)quinazolin-4-yl] pyridine-4-carbohydrazide derivatives ($5D_1$-$5D_4$) were synthesized and characterized by IR, ^{1}H NMR, Mass spectra and elemental analysis. All the newly prepared molecules were tested for their antibacterial and antifungal activity against six strains of gram positive and gram-negative bacteria and two strains of fungi using serial plate dilution method. Among the prepared products, N'-{2-[(2-fluorophenyl) sulfanyl] quinazolin-4-yl} pyridine-4-carbohydrazide $5D_1$ was found to exhibit the highest *in vitro* antibacterial and antifungal potential with minimum inhibitory concentration (MIC) values of 3.12, 0.4, 0.8, 12.5, 12.5, 6.25, 0.8 and 1.6 µg/ml against the respective strains. The MIC of compounds $5D_2$ and $5D_4$ were found comparable with fluconazole, making it most potent antimicrobial agents in the series. In the present investigation all the compounds were found to exhibit more inhibition towards gram positive than gram negative bacterial strains. Molecular docking was performed to dock compounds into the ecKAS III binding site, which suggested probable inhibition mechanism. The results revealed the higher potency of compounds $5D_1$, $5D_3$ and $5D_4$ based on glide gscore and binding poses of the molecules as ecKAS III synthase inhibitors.

Keywords: Quinazoline, Molecular docking, Antibacterial activity, Antifungal activity.

INTRODUCTION

Resistance to antimicrobial agents in existing use has been mounting for a great multiplicity of microorganisms and the resistance to multiple drugs is commonly found for several microorganisms.[1] The emergence of antimicrobial resistance is multifaceted and causes severe health problems, because of this discovery or optimization of new antimicrobial agents with lower toxicity and reduced side effects is of paramount significance.[2] Quinazoline is one of the most frequently encountered heterocycle in medicinal chemistry and many of its derivatives are used as medicines and display antibacterial and antifungal[3], anti-HIV[4], anticancer[5], anti-tubercular[6], anti-inflammatory[7], antidepressant[8], anticonvulsant and antihypertensive activities.[9] They also act as powerful inhibitors of epidermal growth factor (EGFR) receptors of tyrosine kinase[10], DNA gyrase inhibitors[11], dihydrofolate reductase enzyme.[12] Among the important pharmacophores responsible for antimicrobial activity, recently2-chloromethyl-3-(*N*-isonicotinamide-yl)-4*H*-quinazolinone[13], (Azetidinyl-3-(isonicotinamide-yl)-6-iodo-quinazolin-4-ones[14] and isoniazid incorporated styryl quinazolinones revealed antimicrobial activity.[15]

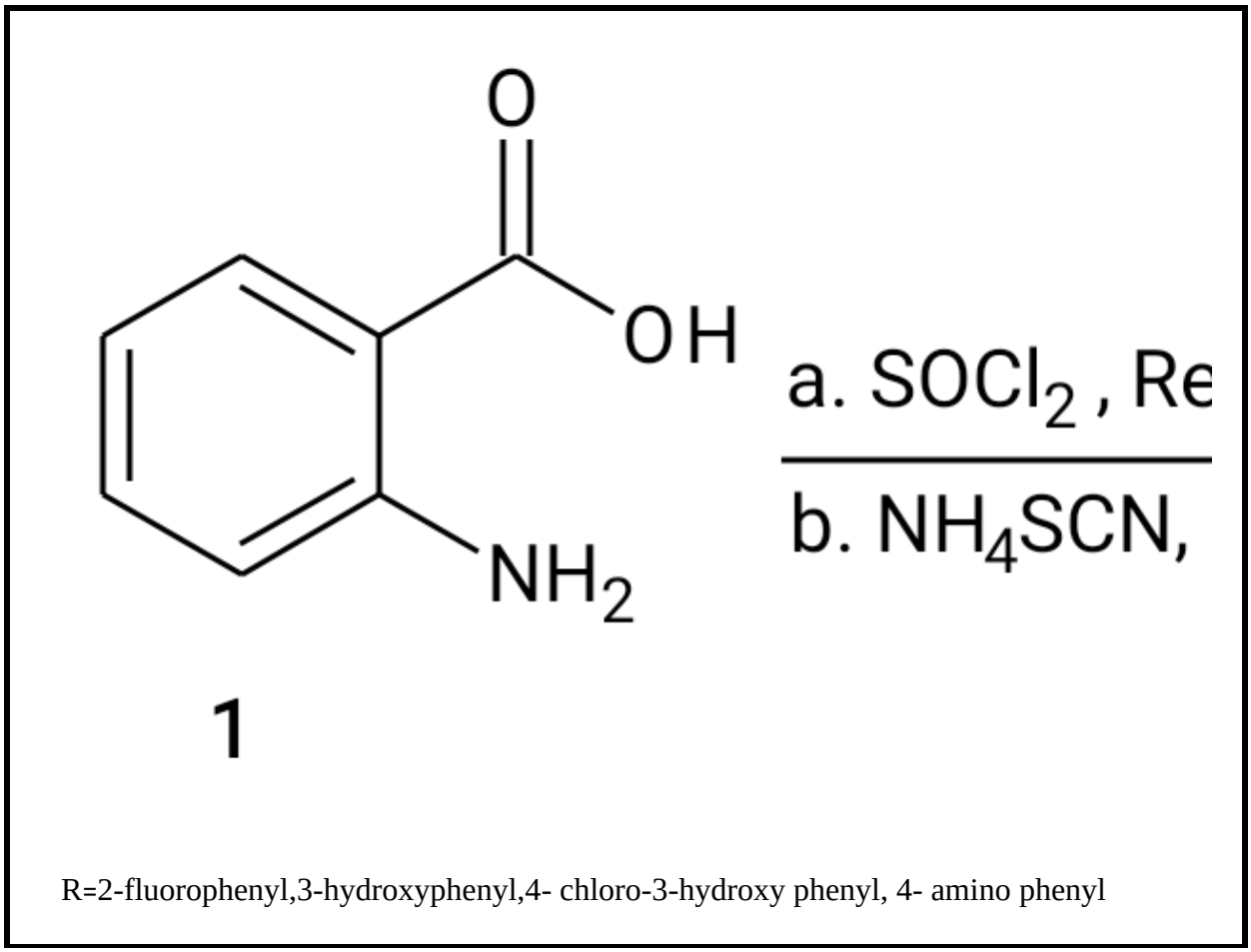
The bacterial fatty acid biosynthesis pathway (FAS) depicts a validated and yet relatively remarkable target for drug discovery. Among the analogous (FAS-II) enzymes the condensing protein, β-ketoacyl-acyl carrier protein synthase (KAS) is attractive target against microbial infections. In bacteria, there appear to be three enzymes designated as KAS I, KAS II and KAS III.[16] The initial C_2-C_4 step of fatty acid biosynthesis is catalysed by KAS III; thereafter, KAS I and KAS II are involved in chain elongation. Notably, KAS III, regulates the fatty acid biosynthesis rate *via* an initiation pathway and its substrate specificity is a key factor in membrane fatty acid composition. This protein has recently been reported as optimistic target against infections caused by multi resistant gram-positive and gram-negative bacteria.[17]RecentlyS-substituted-1,3,4-thiadiazole-thiol and thiazole derivatives were reported as a potent inhibitor of *Escherichia coli* ecKAS III with antimicrobial activity against various bacteria.[18,19]Inspired from these observations, in the present study some novel *N*'-[2-(substituted sulfanyl)quinazolin-4-yl] pyridine-4-carbohydrazide derivatives were synthesized as ecKAS III inhibitors and screened against gram-positive, gram-negative bacteria and pathogenic fungi. Molecular docking was also carried out *in silico* to gain knowledge about the antimicrobial activity of these molecules using the software (Glide, version 9.5, Schrodinger, LLC, New York, NY, 2013). In our study for molecular docking, we have taken β -ketoacyl-acyl carrier protein synthase III (ecKAS III pdb id:

1HNJ) the enzyme from *Escherichia coli* as a receptor responsible for growth of bacteria and our synthesized derivatives as ligands.[20]

MATERIALS AND METHODS

General

All chemicals used were of laboratory grade and procured from Merck and Sigma Aldrich. Melting points were determined by the open tube capillary method and are uncorrected. The solvents used were dried and purified, as and when required. The reaction was monitored using thin-layer chromatography. The spots were observed by exposure to iodine vapour or by UV light. The crude synthesized compounds were purified by column chromatography using silica gel Merck and mobile phase petroleum ether: ethyl acetate (9:1, v/v) as eluent. The crude solid was purified by column chromatography. The IR spectra were obtained on a Shimadzu 8400FT-IR spectrometer (KBr pellets). The ^{1}H NMR spectra were recorded on a Bruker Ascend^{TM} 500 FT-NMR spectrometer using TMS as the internal standard in DMSO-d_6. The mass spectra were recorded on Bruker Daltonik GmbH, Mass spectrometer ESI. Satisfactory analysis for C, H, and N on EuroVector E 3000 elemental analyser was obtained for the compounds within ± 0.4 % of the theoretical values. The following organisms were used in the antimicrobial screening, *Bacillus subtilis*ATCC-60511, *Staphylococcus aureus* ATCC 11632, *Staphylococcus epidermidis* ATCC 155, *Escherichia coli* ATCC 10536, *Klebsiella pneumoniae* ATCC 11298, *Pseudomonas aeruginosa* ATCC 10145, *Candida albicans* ATCC 2091, *Aspergillus niger* ATCC 6275.The synthetic strategy leading to the target compound is illustrated in **Scheme 1**.



Scheme 1: Synthetic route of titled compounds $5D_1$-$5D_4$

General procedure for preparation of titled compounds $5D_1$-$5D_4$.

A mixture of intermediate N'-(2-sulfanylquinazolin-4-yl) pyridine-4-carbohydrazide (4) (0.371g, 0.00125 mole), appropriate halide (0.00125 mol) and tribenzyl ethyl ammonium bromide (0.006 mole) was dissolved in toluene (10 ml) and aqueous potassium hydroxide 0.0605 mole (10 ml). The solution was heated under reflux at 80-85^{o}C for 4-5 hrs. The progress of the reaction was monitored by TLC. After completion of reaction the solvent was removed under vacuum. The separated crude solid was purified by column chromatography using silica gel Merck, mobile phase petroleum ether-ethyl acetate (9:1, v/v) to give the titled compounds in good yield. All the titled compounds were prepared as per the literature procedures with slight modification.[21-25]

N'-{2-[(2-fluorophenyl) sulfanyl] quinazolin-4-yl} pyridine-4-carbohydrazide ($5D_1$)

Light yellow solid, FTIR (KBr, cm^{-1}) υ: 3346.66 (N-H str.), 3027.23 (C-H aromatic),1670.41(C=O of sec. amide), 1565.23(C=N), 1546.57(N-H bending), 1491.59 (C=C), 1368.12 (C-H bending), 1280. 24 (C-N), 1258.16 (N-N),1164.32 (C-S), 1114.89 (C-F).^{1}H NMR (500MHz, DMSO-d6) δ ppm: 8.97 (s, 1H, NH-CO), 6.80-8.70 (m,12H, of Ar-H), 5.92(s, Ar-C-NH); HRMS (ESI), found: 392.0975(M+H).Anal calcd for $C_{20}H_{14}FN_5OS$= C, 61.37;H, 3.61; N, 17.89, Found: C, 61.29; H, 3.52; N,17.78.

N'-{2-[(3-hydroxyphenyl) sulfanyl] quinazolin-4-yl} pyridine-4-carbohydrazide (5D2)

Off white powder; FTIR (KBr, cm^{-1}) υ : 3406.40 (O-H str), 3319.26 (N-H str), 3043.77(C-H aromatic), 1670.41 (C=O of amide), 1590.61 (C=N), 1547.32 (N-H bending), 1460.68(C=C), 1383.01 (C-H bending), 1346.36(C-N), 1249.91(N-N), 1217.12 (C-O), 1145.75 (C-S); ^{1}H NMR (500MHz, DMSO-d_6) δ ppm: 9.40 (s, 1H, NH-CO), 7.32(s, 1H, Ar-OH) 5.80(s, 1H, Ar-C-NH), 6.60-8.02 (m, 12H, Ar-H); HRMS (ESI), found: 390.1224 (M+H). Anal calcd for $C_{20}H_{15}N_5O_2S$: C, 61.68; H, 3.88; N, 17.98. Found: C, 61.57; H, 3.74; N, 17.83.

N'-{2-[(4-chloro-3-hydroxyphenyl) sulfanyl] quinazolin-4-yl} pyridine-4-carbohydrazide (5D3)

Light brown powder; FTIR (KBr, cm^{-1})υ : 3406.40 (O-H str), 3314.78(N-H str), 3037.99 (C-H aromatic), 1670.41 (C=O of amide), 1567.32 (C=N), 1610.61,1468.63 (C=C), 1383.01 (C-H bending), 1346.36 (C-N), 1249.91(N-N),1217.12(C-O), 1145.75 (C-S), 771.56 (C-Cl), 692.47 (oop C-H $_{arm}$); ^{1}H NMR (DMSO-d_6) δ ppm: 11.03 (s, 1H, Ar-C-OH), 8.72 (s, 1H, NH-CO), 7.08-8.60 (m, 11H, Ar-H), 3.40 (s, 1H, Ar-C-NH); Anal calcd for $C_{20}H_{14}ClN_5O_2S$: C, 56.67; H, 3.33; N,16.52. Found: C, 56.58; H, 3.23; N, 16.42.

N'-{2-[(4-aminophenyl) sulfanyl] quinazolin-4-yl} pyridine-4-carbohydrazide (5D4)

Light yellow solid; FTIR (KBr, cm^{-1})υ : 3414.12 (N-H str of NH_2), 3312.34(N-H str), 3037.99 (C-H aromatic), 1670.41(C=O of amide), 1566.25(C=N), 1612.54,1476.36(C=C), 1386.86 (C-H bending), 1327.07(C-N), 1251.84(N-N), 1141.90 (C-S); ^{1}H NMR (500MHz, DMSO-d_6) δ ppm: 9.02(s, 1H, NH-CO), 6.50-8.72(m, 12H, Ar-H), 4.90 (s, 1H, Ar-C-NH), 3.80 (s, 2H, Ar-C-NH_2); HRMS (ESI), found: 389.1164 (M+H). Anal calcd for $C_{20}H_{16}N_6OS$: C, 61.84; H, 4.15; N, 21.63. Found: C, 61.76; H, 3.99; N, 21.53.

***In vitro* Antibacterial and Antifungal screening:**

The synthesized compounds 5D1-5D4 were tested for their *in-vitro* antibacterial activity against three Gram-positive (*Bacillus subtilis*, *Staphylococcus aureus* and *Staphylococcus epidermidis*) and three Gram-negative (*Escherichia coli, Klebsiella pneumoniae* and *Pseudomonas aeruginosa*) bacterial strains and anti-fungal activity of the compounds was assayed against two fungal strains viz. (*Candida albicans* and *Aspergillus niger*) by serial plate dilution method.[26-28]The minimum inhibitory concentration (MIC, µg/ml) of test compounds were visually determined as the lowest concentration of the drug at which there was no visible growth of each strain. The synthesized compounds (10 mg) were dissolved in dimethyl sulfoxide (DMSO,1ml), then diluted in culture medium (Mueller-Hinton Broth for bacteria and Sabouraud Liquid Medium for fungi), further increasing dilutions were done to obtain final concentrations of 0.2, 0.4, 0.8, 1.6, 3.12, 6.25, 12.5, 25, 50 and 100 µg/ml. 20ml of agar media was poured into each petri dish. Excess suspension was decanted and plates were dried by continued placing in an incubator at 37ºC for an hour. Using an agar punch, wells were made on these seeded agar plates and minimum inhibitory concentration of the test compounds in dimethyl sulfoxide were added into each labelled well. A control was also prepared for the plates in the same way using solvent DMSO. The petri dishes were prepared in triplicate and maintained at 37ºC for 3-4 days. Ciprofloxacin and fluconazole were used as standard drugs for antibacterial and antifungal activity respectively, while keeping DMSO as control which did not show inhibition against the tested organisms. Minimum inhibitory concentration (µg/ml) and diameter of the zone of inhibition(mm) was determined for all the synthesized compounds.

In silico study

The molecular docking study was carried out by using the tool, (Glide, version 9.5, Schrodinger, LLC, New York, NY.). The crystal structure of *Escherichia coli* β -ketoacyl-acyl carrier protein synthase III having resolution of 1.46 Å was retrieved from the protein data bank (ecKAS III pdb id: 1HNJ) used as target for docking of synthesized compounds.[29]All molecules were drawn in Maestro and converted to3D conformations. All the possible tautomers and stereo isomers were generated using Epik. Partial atomic charges were also computed by OPLS. The reported crystal structure is a monomer, having only one chain A with the inhibitor bound to it. The protein was prepared using protein preparation wizard and glide energy grids were generated for prepared protein complex. The binding site was defined by a grid box of 20 x 20 x 20 $A^{\circ 3}$ for the receptor

was generated with a default inner box of 10 x 10 x 10 $A^{\circ 3}$, which was centered on the corresponding ligand. The alignment was performed using the protein alignment module (Prime, Schrödinger). Bond orders and formal charges were added for hetero groups and hydrogens were added to all atoms in the system. Water molecules of crystallization beyond 5A° were removed from the complex except in the active site. In the protocol, the prepared ligands were docked against β-ketoacyl-acyl carrier protein synthase III the for the refinement and docking calculations, the default settings as available in the software package were used. Final scoring is then carried out on the energy-minimized poses. The minimized poses are rescored using Schrödinger's proprietary Glide Score (g score) scoring function.[30]

RESULTS AND DISCUSSION

In literature survey, it was observed that few methods have been developed to date for the synthesis of isoniazid incorporated 2-substituted quinazoline derivatives and these include the reaction of anthranilic acid followed by amine insertion. In the present work *N*'-[2-(substituted sulfanyl)quinazolin-4-yl] pyridine-4-carbohydrazide derivatives 5D1-5D4 were synthesized by using commercially available pyridine-4-carbohydrazide and either halogens or electron-withdrawing/electron releasing substituents.

Chemistry

2-sulfanylquinazolin-4(3*H*)-one (2) was prepared from anthranilic acid (1) in thionyl chloride when heated at reflux for 2 hrs, dissolved in acetone and added to a suspension of NH_4SCN with stirring at room temperature. Upon stirring compound (2) in phosphorous oxychloride and N, N'-dimethylaniline under reflux at 108ºC for 3-4 hrs, residue was obtained. It was neutralized to pH 4-5 and further extracted with chloroform, it was removed under reduced pressure to yield 4-chloroquinazoline-2-thiol (3). Refluxing mixture of (3) and pyridine-4-carbohydrazide in glacial acetic acid at 120ºC for 12-14 hrs, gave the corresponding *N*'-(2-sulfanylquinazolin-4-yl) pyridine-4-carbohydrazide (4) in 78-82% yield, reaction was monitored by TLC. The substituted N'-[2-(alky/aryl sulfanyl) quinazolin-4-yl] pyridine-4-carbohydrazide derivatives (5D1-5D4) were obtained by heating a mixture of intermediate *N*'-(2-sulfanylquinazolin-4-yl)pyridine-4-carbohydrazide (4), appropriate halides and tribenzyl ethyl ammonium bromide dissolved in toluene and potassium hydroxide under reflux at 80-85ºC for 4-5 hrs, to get the products in good yields under phase transfer catalysis.

In the IR spectrum of 5D1-5D4 the most characteristic absorption bands observed in the range of 3280.52- 3398.69 $cm^{-1}$for (N-H), C-H aromatic in range of3026.15-3047.63 cm^{-1}, whereas NH-CO band appeared at 1650.68-1689.70 cm^{-1} and the bands of C=N and C-N of aromatic rings in the range of 1590.61-1620.26 and 1286.32-1346.36 cm^{-1} respectively. In the ^{1}H-NMR spectra N-H peaks were observed as singlet for sec. amide at about δ 8.57-9.40 ppm and around 3.40-5.80 ppm for Ar-C-NH. In addition, signal of quinazoline proton H-4 appeared as doublets in the range of 8.013-8.045 ppm, H-3 and H-6 were observed mostly as multiplet in range of 7.762-7.795 ppm and H-5 appeared as triplet of doublet around 7.204-7.582 ppm region. All the other aromatic and aliphatic protons were observed at expected regions. The mass spectrum of compounds showed molecular ion peaks at *m/z* 424.0510 to 338.1939 corresponding to molecular formula. Elemental analysis of these compounds further confirmed the successful formation of these compounds.

Table1: Physical characterization data of N'-{2-[(substituted phenyl) sulfanyl] quinazolin-4-yl} pyridine-4-carbohydrazide(5D Series)

Comp. Code	R	Mol. Formula	Mol. Wt.	m. p^{0}C	% Yield	R_f Value
5D1	2-fluoro	$C_{20}H_{14}FN_5OS$	391.42	162-164 ^{o}C	50	0.67
5D2	3-hydroxy	$C_{20}H_{15}N_5O_2S$	389.43	210-211 ^{o}C	61	0.54
5D3	4-chloro-3-hydroxy	$C_{20}H_{14}ClN_5O_2S$	423.87	251-253 ^{o}C	71	0.79
5D4	4-amino	$C_{20}H_{16}N_6OS$	388.44	225-227^{o}C	68	0.71

***In vitro* Antibacterial and Antifungal screening:**

As per the antibacterial and antifungal data summarized in **Table 2** it was revealed that, compounds 5D1and 5D3 exhibited higher inhibition due to the presence of halogens -F and Cl attached to the aryl ring at C_2 of quinazoline against gram positive *Bacillus subtilis, Staphylococcus aureus, Staphylococcus epidermidis* and gram-negative bacteria *P. aeruginosa* with MIC values between 0.4 to 12.5 μg/ml except *Escherichia coli and K. pneumoniae*. Surprisingly compound 5D2 after substitution of aryl ring with electron donating group-OH showed higher activity against *Staphylococcus aureus* and *Staphylococcus epidermidis* (MIC 0.8 μg/ml) whereas compound 5D4 with -NH_2 group displayed less activity against all gram-positive bacteria and both exhibited least activity with MIC between 50 to 100 μg/ml against all gram-negative bacteria.

The screening data of anti-fungal activity of these series of compounds shows wide range of anti-fungal activity. The compound 5D4 showed moderate to good activity with MIC values between 0.8, 3.12 to 25 µg/ml. Interestingly compounds 5D2 and 5D3 were found to exhibit potent *in-vitro* anti-fungal activity against A. *Niger and C. Albicans* with MIC 0.4 to 1.6 µg/ml. Whereas most potent activity against fungi was exhibited by compound 5D2, which is equipotent to a standard drug fluconazole. on the other hand, compound 5D2 and 5D3 has emerged as most potent antifungal agents, this might be due to the increased lipophilicity or with favourable steric hinderance. It is interesting to note that a minor alteration in the molecular configuration of investigated compounds may have a pronounced effect on antibacterial and antifungal screening.

Table 2. *In vitro* Antibacterial and Antifungal screening of the synthesized compounds 5D1-5D4

Comp. Code	**R**	***In vitro* activity-zone of inhibition in mm (MIC in µg/ml)**							
		Gram positive bacteria			**Gram negative bacteria**			**Fungi**	
		***B.s.* ATCC 60511**	***S.a.* ATCC 11632**	***S. e.* ATCC 155**	***E. c.* ATCC 10536**	***K. p.* ATCC 11298**	***P.a.* ATCC 10145**	***C.a.* ATCC 2091**	***A. n.* ATCC 6275**
5D1	2-fluoro phenyl	17 (3.12)	23 (0.4)	21 (0.8)	15 (12.5)	16 (12.5)	17 (6.25)	22 (0.8)	20 (1.6)
5D2	3-hydroxy phenyl	13 (25)	19 (0.8)	20 (0.8)	14 (25)	10 (50)	11 (50)	23 (0.4)	22 (0.8)
5D3	4-chloro-3-hydroxy phenyl	23 (0.4)	15 (6.25)	16 (6.25)	12 (25)	11 (50)	12 (50)	17 (1.6)	23 (0.4)
5D4	4-amino Phenyl	8 (100)	11 (50)	19 (0.8)	9 (100)	10 (50)	11 (50)	10 (25)	18 (3.12)
Ciprofloxacin		24 (0.4)	25 (0.4)	25 (0.2)	21 (0.2)	22 (0.8)	21 (<4)	-	-

Fluconazole		-	-	-	-	-	-	25 (16)	26 (8)
DMSO	-	-	-	-	-	-	-	-	-

B.c.-Bacillus subtilis, S.a.-Staphylococcus aureus, S.e.-Staphylococcus epidermidis, E.c. Escherichia coli, K.p.-Klebsiella pneumoniae, P.a.-Pseudomonas aeruginosa, C.a.-Candida albicans, A. n.- Aspergillus niger. (-) indicates no inhibition zone.

The relative zone of inhibition of compounds 5D1-5D4 shown graphically in **Fig. 1.**

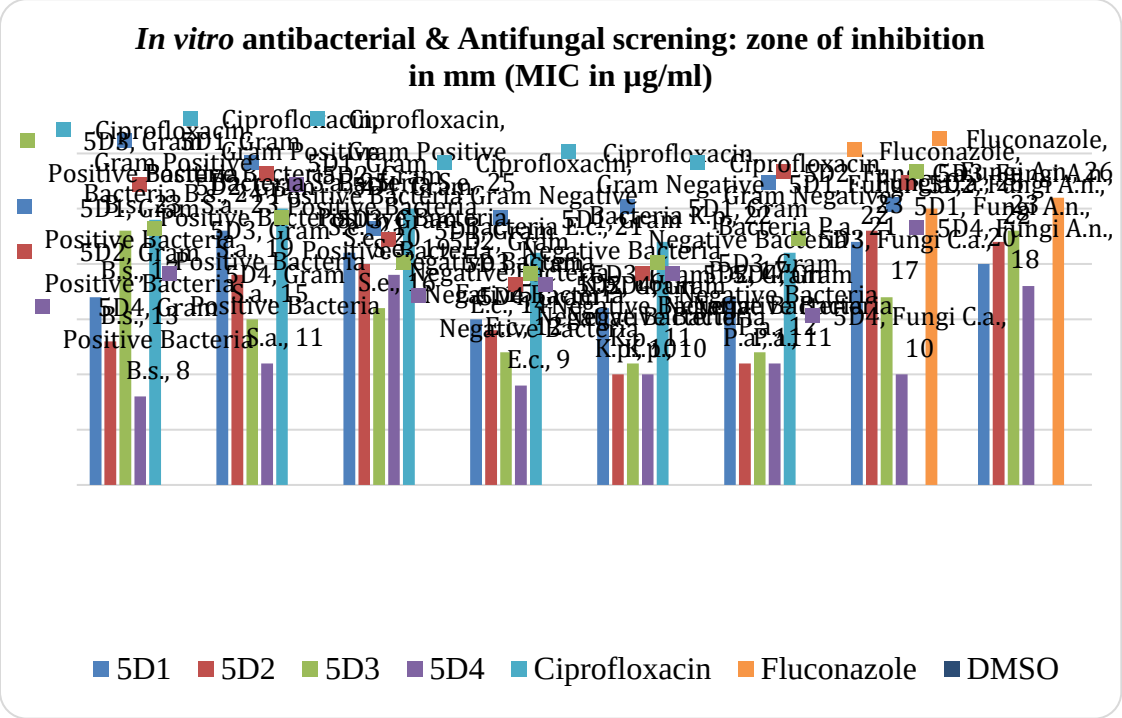


Fig.1: Zone of inhibition of synthesized derivatives (5D1-5D4) ciprofloxacin and Fluconazole

Molecular Docking study

Here, we investigated the binding affinities of synthesized compounds 5D1-5D4 into ecKAS III. The protein structure file (PDB ID: 1HNJ) taken from PDB (www.rcsb.org/pdb) was edited by removing the hetero atoms, adding C-terminal oxygen also shows the *in silico* active pocket prediction of amino acids of protein ecKAS III synthase involved in binding with the ligands obtained from PDB sum. The active site of ecKAS III was defined based on the centre and radius of the binding substrate in X-ray structure of enzyme complexed with CoA or inhibitor towards optimization of the aforementioned compounds of the promising antimicrobial activities.[31] The binding affinity was evaluated by glide gscore, glide energy and hydrogen bonding. The

compounds which revealed the highest binding affinities, that is, lowest glide gscore, within KAS III and the hydrogen bond interactions into the target macromolecule are represented in **Table 3**. Many of these derivatives exhibited weak π-π stacking and one or two hydrogen bonds between N, O, H of series 5D1-5D4 and different amino acids of the target ecKAS III including Thr 28, Arg 151 and Gly 209.

Table 3. Molecular docking results of 5D1 and 5D2 against β-Ketoacyl carrier protein.

Compound Code	**Glide emodel**	**Glide score**	**No. of hydrogen bond and distance in A^{o}**
5D1	-62.379	-6.349	02 O (5D1) H-N (Thr 28): 2.18 N (5D1) H-O (Arg 151): 2.63
5D3	-75.843	-7.717	03 O (5D3) H-N (Thr 28): 1.88 N (5D3) H-O (Arg 151): 2.48 N (5D3) H-O (Gly 209):1.70

After analysing the different docking interactions of ligands, the compounds namely 5D2and 5D3 showed fairly better interaction with ecKAS III with the lowest glide gscore value than the other molecules. Theoretically all the molecules showed very good glide gscore and glide emodel ranging from -5.280 to -7.717 and -52.370 to -75.843 kJ/mol, respectively.

In in-vitro study, among these molecules, compound 5D1 have emerged as active against all tested gram positive and gram-negative bacterial strains, whereas antifungal evaluation revealed the compound 5D3 has most potent against fungi indicating that the docking method was most appropriate for clarifying the binding mode of this novel series of compounds as good inhibitor of InhA, as illustrated in **Fig. 2 & 3.** So, it can be predicted as the activity may be due to inhibition of enzyme ecKAS III synthase.

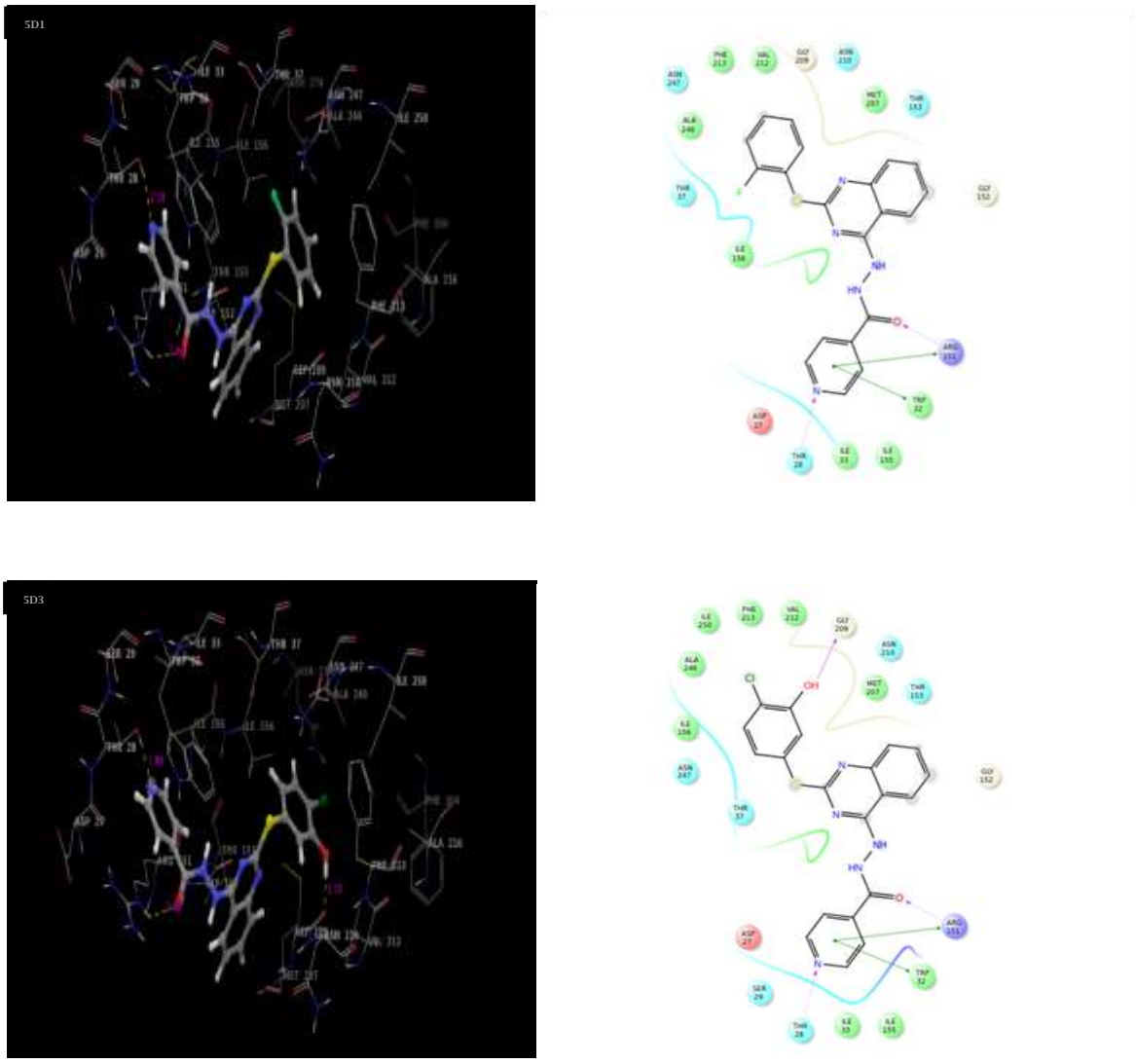


Fig. 2 & 3. Compound 5D1 and 5D3 bound into ecKASIII receptor site via hydrophobic interactions and hydrophilic binding by hydrogen bond between compound and amino acids.

CONCLUSION

As a result of docking and antimicrobial evaluation, a few conclusions could be made, some of the newly synthesized compounds 5D1-5D4 exhibited promising antibacterial activity against *E. coli, P. aeruginosa, S. aureus* strains, while antifungal activity against *C. albicans, A. niger*. Compounds 5D1 possess excellent activity against both gram positive and gram negative bacterial and compound 5D3 against fungal species. It seems that the halogens at para position was very significant for enhancing activity against gram positive and gram-negative bacterial species as well

as hydroxy group at meta position for fungal species, which could be promising agents. Molecular docking studies also revealed that 5D1, 5D2, and 5D3have minimum glide energy and glide score and may be considered as a good ecKAS III inhibitors. Thus by analyzing these data *N*'-[2-(substituted sulfanyl) quinazolin-4-yl] pyridine-4-carbohydrazide derivatives can be considered as potent inhibitor against the enzyme ecKAS III synthase. Hence this study has widened the scope of developing these derivatives as promising antibacterial and antifungal agents.

ACKNOWLEDGEMENT

The authors would like to express their gratitude and thanks to Principal, Maratha Mandal's Dental College, Hospital and Research Centre, Belgaum, India, for testing the antibacterial and antifungal activity of our new series of compounds. The authors also thanks to NMIMS, School of Pharmacy, Shirpur for the valuable guidance and permission to use the software package of Maestro, version 9.5, Schrodinger, LLC, New York, USA.

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