Bioorganic Chemistry 90 (2019) 103093

Contents lists available at ScienceDirect

Bioorganic Chemistry

journal homepage: www.elsevier.com/locate/bioorg

Amino acids conjugated quinazolinone-Schiff's bases as potential antimicrobial agents: Synthesis, SAR and molecular docking studies

K.P. Rakesh[a], H.K. Kumara[b], B.J. Ullas[a], J. Shivakumara[a], D. Channe Gowda[a,*]

[a] Department of Studies in Chemistry, University of Mysore, Manasagangotri, Mysore 570 006, Karnataka, India
[b] Department of Chemistry, KLE Society's Jagadguru Tontadarya College, Gadag-582101, Karnataka, India

ARTICLE INFO

Keywords:
Quinazolinone
Amino acids
Schiff's bases
Antimicrobial
Docking studies

ABSTRACT

A series of amino acids conjugated quinazolinone-Schiff's bases were synthesized and characterized by analytical and spectroscopic methods. All the synthesized analogues (**8–43**) and the intermediates (**1–7**) were screened for their *in vitro* antibacterial and antifungal activities. In antimicrobial activity, compounds **12–16**, **21–25**, **30–34** and **39–43** showed excellent antibacterial activity which is better than the antibacterial standard Streptomycin. Compounds **15**, **23–25**, **30–34**, **36** and **38–43** showed excellent antifungal activities which is more active than the reference antifungal drug Bavistin. Further, to understand the correlation of biological activity with that of drug-receptor interaction, molecular docking was performed on active site of*glucosamine-6-Phosphate (GlcN-6-P) synthase* (PDB ID: 2VF5) which showed good binding profile. Molecular docking studies and Preliminary structure-activity (SAR) relationship revealed that the tryptophan and phenylalanine conjugated quinazolinones with electron donating groups (OH and OCH_3) were found to be excellent antimicrobial activities which is better than the glycine and alanine conjugated derivatives. This may be explained by the contribution of aromaticity and hydrophobicity of amino acids. Among the series, compounds **41** and **43** showed the highest docking scores for antimicrobial activity. The conjugation plays a major role in improving the biological activities of those compounds.

1. Introduction

The damage of bacterial and fungal infections has increased hugely in recent years [1]. Infectious diseases caused by bacterial pathogens have become a main public health problem due to the extensive occurrence of drug resistance. Resistance to antimicrobial agents has increased health concerns and resulted in mortality and morbidity from treatment failures [2]. The global hazard of infectious diseases has been aggravated by the emergence of bacterial resistance to antibiotics. Such antibiotic resistance has inspired the urgent need for the design of new antibiotics with different modes of action [3–7]. Several bacterial strains causing infectious diseases which seemed to be in control are once again causing death every year due to the absence of an appropriate antibiotic drug [8]. Unfortunately, the development of new antibiotics has failed to keep pace with the development of drug-resistance over the past few decades [9]. Thus, there is a critical global healthcare crisis, which requires the urgent development of more effective antibiotics. In particular, attention has focused on the Gram positive organism *Staphylococcus aureus* because many strains of this organism are now resistant against clinically useful antibiotics like methicillin and vancomycin [10].

Staphylococcus aureus is one of the main human pathogen cause's soft external infections to harsh life-threatening persistent infections to the human world ensuing in important morbidity and mortality [11–16]. A potential approach to overcome this resistance problem is to design new and innovative agents with a completely different mode of action so that no cross-resistance with the present therapeutically can occur. In the past few decades, the number of efforts has been made in the medicinal chemistry through synthetic tailoring in a combinatorial fashion, to generate a large set of analogues as core scaffolds. Although the tremendous approaches have been fruitful, no new major class of antibiotics were invented between 1962 and 2000 [17]. Therefore, to come up with new effective therapeutic agents, there is a need for aggressive efforts and it is imperative to discover novel synthetic entities for the microbial target is a big challenge to the medicinal chemistry [18]. The overall clinical experiences reveal that the single-targeted drugs will not be effective agents to the biological system even if they have good inhibitory activity against specific target [19].

The growing interest in heterocyclic compounds is basically because of their raised biological activity and also they make possible

* Corresponding author.
E-mail address: dchannegowda@yahoo.co.in (D. Channe Gowda).

https://doi.org/10.1016/j.bioorg.2019.103093
Received 8 April 2019; Received in revised form 24 June 2019; Accepted 26 June 2019
Available online 27 June 2019
0045-2068/

development of novel materials with unique properties. One very interesting and promising class of heterocycle is the series of quinazolinone and its derivatives. This small and rigid heterocyclic backbone could act on various pharmacological targets. Especially, quinazolinone nucleus could be found in a broad range of biologically active compounds displaying antimicrobial [20–23], antioxidants [24], anticancer [25], anticonvulsant [26], antiulcer [27], analgesic [28], immunotropic activities [29] and are also known to act as thymidylate synthase [30], poly(ADP-ribose) polymerase [31], anti-breast cancer [32], aromatase inhibitor [33,34], and COX inhibitors [35].

Extensive work has been reported on the conjugation of different amino acids/peptides to various biologically active moieties [36–40] which reveals that conjugation plays a leading position in exerting the activity. Also, involving amino acids/peptides in drugs makes them low toxic, ample bioavailability and permeability, modest potency and good metabolic and pharmacokinetic properties [41]. In a continuous effort to develop novel antimicrobial agents with improved results, the present work involves the synthesis of amino acids conjugated quinazolinones-Schiff's bases and evaluated for their *in vitro* antimicrobial activities. In addition, in this work, we have also conducted molecular docking studies of the compounds to correlate them with their antimicrobial activities.

2. Results and discussion

2.1. Chemistry

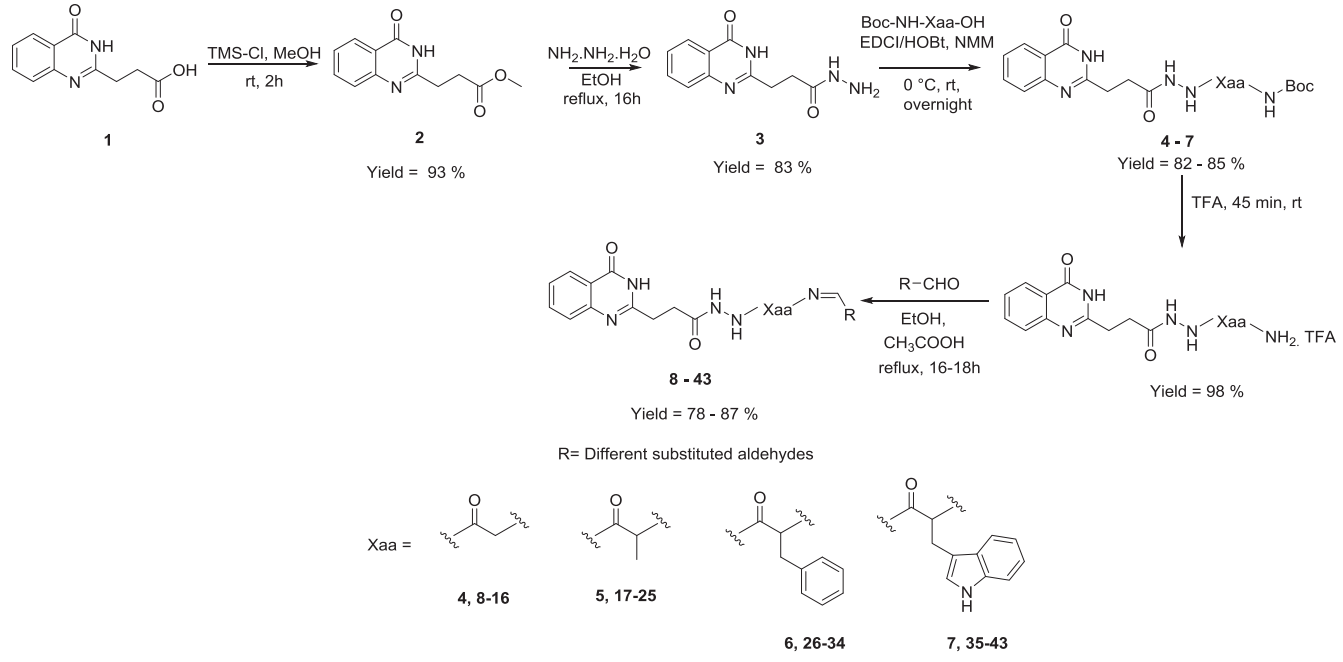
The heterocycle3-(4-oxo-3,4-dihydroquinozolin-2-yl)propanoic acid (**1**) was synthesized following literature methods [42–44]. This was further methylated using trimethylsilyl chloride (TMS-Cl) and methanol at room temperature, which upon reaction with excess of hydrazine hydrate afforded the corresponding quinazolinone hydrazides (**3**). These hydrazides were conjugated to Boc protected amino acids using *N*-(3-dimethylaminopropyl)-*N′*-ethylcarbodiimide (EDCI), 1-hydroxybenzotriazole (HOBt) as coupling agent and *N*-methylmorpholine (NMM) as base. Boc group of the conjugates was removed using trifluoroacetic acid (TFA) and reacted with various substituted aldehydes in presence of catalytic amount of glacial acetic acid to obtain amino acids conjugated quinazolinone-Schiff's base derivatives (Scheme 1). The formation of methyl esters (**2**) was confirmed by the appearance of a singlet at 3.68 δ for $-OCH_3$ and absence of COOH proton peak at 12.25 δ in ^{1}H NMR spectrum. In IR spectra, bands at 3310 and 3217 cm^{-1} for NH_2-NH-groups indicates the conversion of methyl esters into hydrazides. The stretching frequencies appeared at 1630–1644 cm^{-1} (CO) and 3300 cm^{-1} (NH) in IR spectra and the peak appeared at $\delta \sim 11.20$ as a singlet (NH) confirms the conjugation. The formation of Schiff's bases were confirmed by the presence of absorption at 1612–1630 for imines i.e., −N═CH− in IR spectra. All the derivatives were obtained in high yields. The structures of all the newly synthesized compounds including intermediates were confirmed by IR, ^{1}H NMR, ^{13}C NMR and mass spectral analysis. The NMR and mass data were found to be in good agreement with the structures assigned.

2.2. Antimicrobial activity

The proficiency of synthesized amino acids conjugated quinazolinone-Schiff's bases were evaluated for their *in vitro* antibacterial activity against one strain of gram positive bacteria like *S. aureus* and one gram negative bacteria like *E. coli* followed by antifungal studies against *A. niger* and *F. oxysporum* by agar well diffusion method. Zone of inhibition (mm) values are presented in the Table 1. The standard drugs streptomycin and bavistin were used as standards for antibacterial and antifungal activities respectively.

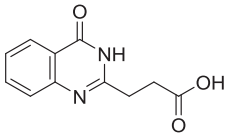
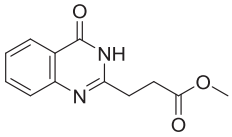
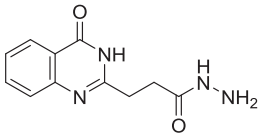
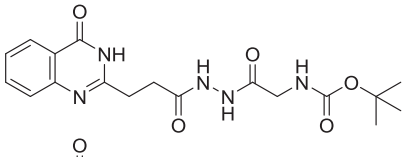
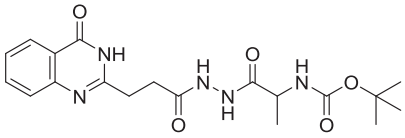
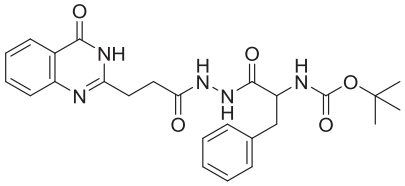
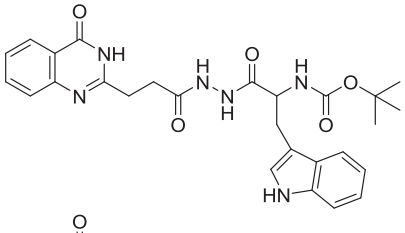
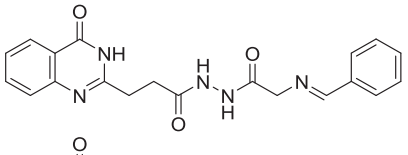
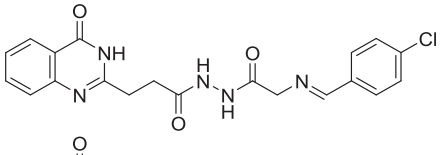
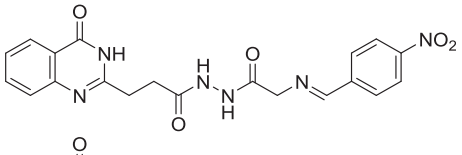
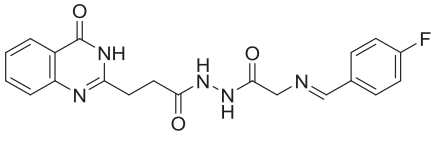
Quinazolinone acid (**1**), ester (**2**) and its hydrazides (**3**) showed very less antimicrobial property whereas amino acid conjugated quinazolinone derivatives (**4–7**) showed enhanced in the antimicrobial properties compared with its counterpart [45]. A drastic improvement in the activity was observed when **4–7** were converted into its corresponding Schiff's bases. The nature of the substituent's present on the phenyl ring affected the biological activity of the compounds to a greater extent.

The compounds showed significant effects on the growth of the tested bacterial and fungal strains. The compounds **12–16**, **21–25**, **30–34** and **39–43** showed excellent antibacterial activity compared to standard streptomycin and compounds **15**, **23–25**, **30–34**, **36**, and **38–43** showed excellent antifungal activity compared to standard bavistin. The phenyl alanine (**32–34**) and tryptophan (**41–43**) conjugated analogues are more potent than glycine (**14–16**) and alanine (**23–25**) conjugated derivatives. This could be due to the presence of electron



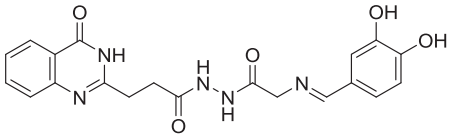
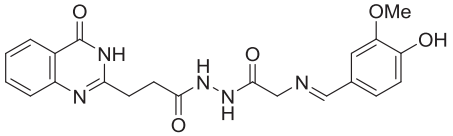
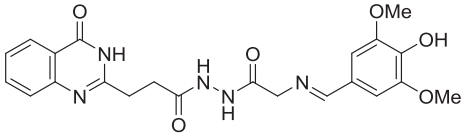
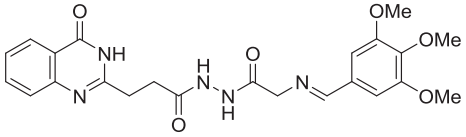
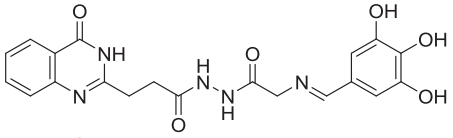
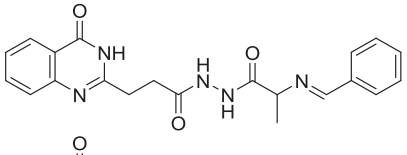
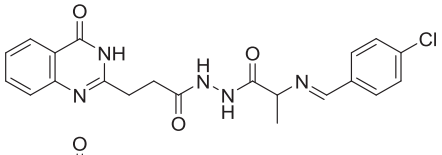
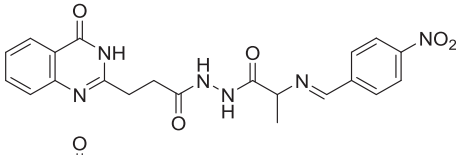
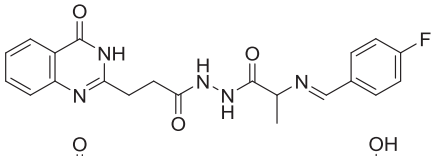
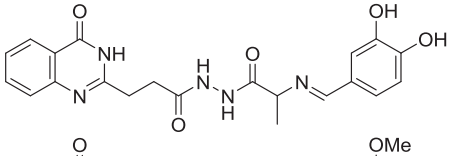
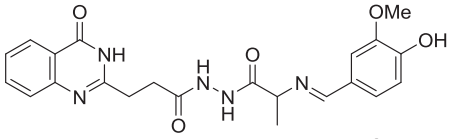
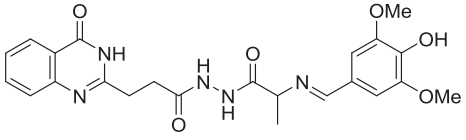
Scheme 1. Synthesis of the title compounds (**4–43**).

Table 1
Antimicrobial activity of the synthesized amino acids conjugated quinazolinone Schiff's base derivatives.

Sl. No	Structure	Antibacterial activity[a]		Antifungal activity[a]	
		S. aureus	*E. coli*	*A. niger*	*F. oxysporum*
1		04 ± 0.12	03 ± 0.30	02 ± 0.22	04 ± 0.11
2		02 ± 0.31	0 ± 0	01 ± 0.31	0 ± 0
3		01 ± 0.14	03 ± 0.16	02 ± 0.10	03 ± 0.12
4		06 ± 0.33	04 ± 0.17	05 ± 0.09	08 ± 0.15
5		05 ± 0.11	06 ± 0.13	07 ± 0.20	06 ± 0.15
6		09 ± 0.44	10 ± 0.19	07 ± 0.35	09 ± 0.31
7		09 ± 0.22	11 ± 0.17	10 ± 0.09	09 ± 0.23
8		08 ± 0.14	10 ± 0.11	04 ± 0.20	03 ± 0.11
9		07 ± 0.21	09 ± 0.15	07 ± 0.14	06 ± 0.14
10		08 ± 0.17	10 ± 0.22	12 ± 0.19	09 ± 0.11
11		10 ± 0.18	09 ± 0.26	10 ± 0.17	12 ± 0.17

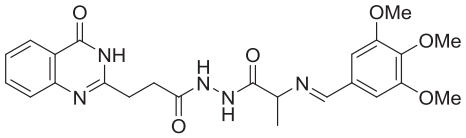
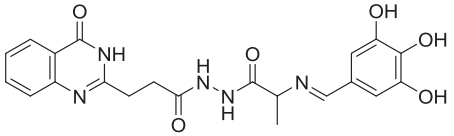
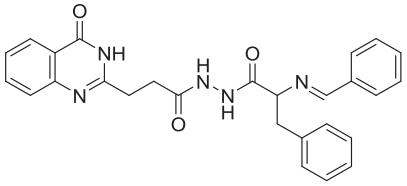
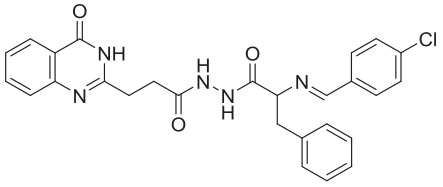
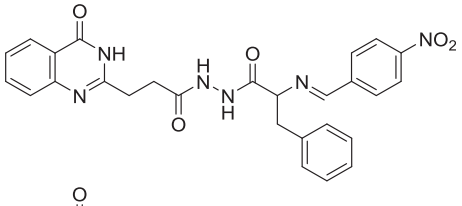
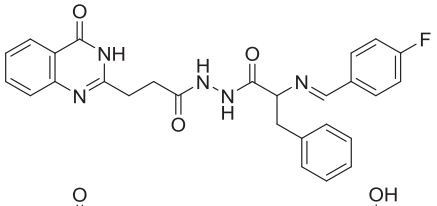
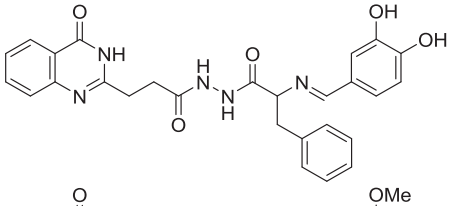
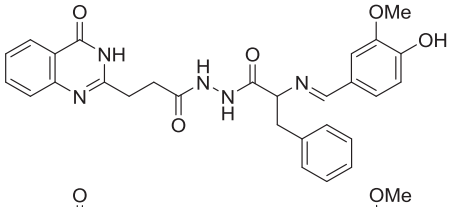
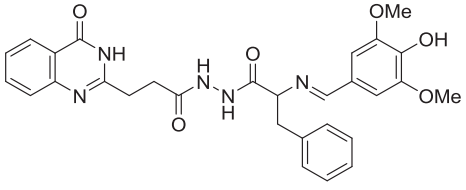
(*continued on next page*)

Table 1 (*continued*)

Sl. No	Structure	Antibacterial activity[a]		Antifungal activity[a]	
		S. aureus	*E. coli*	*A. niger*	*F. oxysporum*
12		13 ± 0.48	14 ± 0.14	10 ± 0.17	09 ± 0.30
13		14 ± 0.10	14 ± 0.15	13 ± 0.18	12 ± 0.15
14		15 ± 0.23	16 ± 0.20	12 ± 0.17	10 ± 0.11
15		17 ± 0.04	15 ± 0.11	15 ± 0.40	14 ± 0.31
16		16 ± 0.27	17 ± 0.28	10 ± 0.22	12 ± 0.20
17		10 ± 0.21	09 ± 0.18	09 ± 0.17	08 ± 0.14
18		09 ± 0.13	10 ± 0.16	13 ± 0.14	12 ± 0.19
19		08 ± 0.10	07 ± 0.13	11 ± 0.14	09 ± 0.13
20		07 ± 0.12	10 ± 0.14	07 ± 0.15	06 ± 0.30
21		18 ± 0.23	15 ± 0.21	14 ± 0.15	10 ± 0.14
22		15 ± 0.16	14 ± 0.11	12 ± 0.16	11 ± 0.31
23		16 ± 0.17	17 ± 0.21	16 ± 0.14	17 ± 0.24

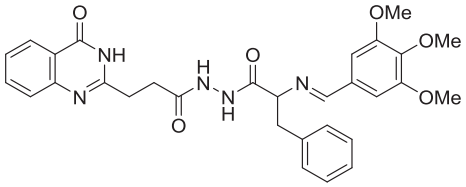
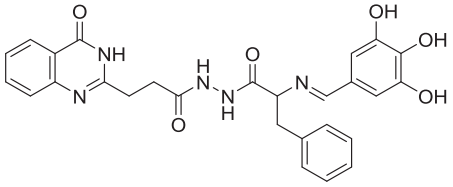
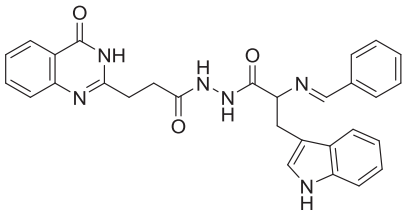
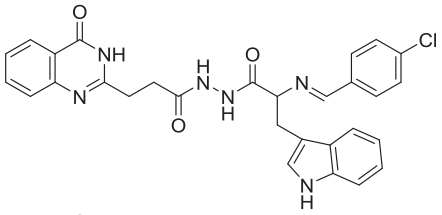
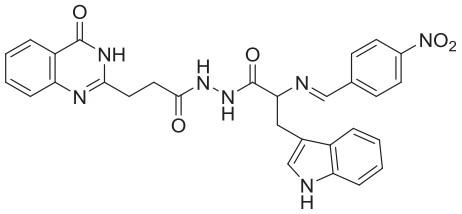
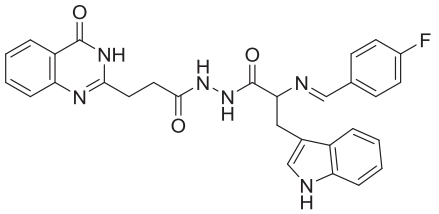
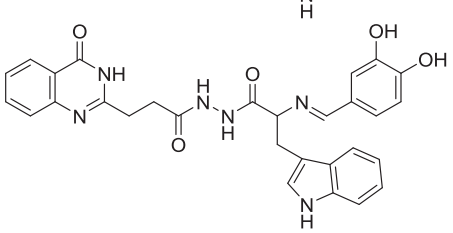
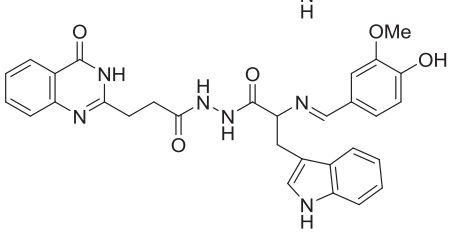
(*continued on next page*)

Table 1 (*continued*)

Sl. No	Structure	Antibacterial activity[a]		Antifungal activity[a]	
		S. aureus	*E. coli*	*A. niger*	*F. oxysporum*
24		15 ± 0.14	16 ± 0.26	15 ± 0.18	14 ± 0.17
25		18 ± 0.23	16 ± 0.24	17 ± 0.12	15 ± 0.48
26		12 ± 0.40	14 ± 0.33	08 ± 0.34	06 ± 0.17
27		10 ± 0.14	12 ± 0.21	16 ± 0.17	18 ± 0.16
28		09 ± 0.16	11 ± 0.18	10 ± 0.21	13 ± 0.17
29		12 ± 0.23	14 ± 012	15 ± 0.24	11 ± 0.23
30		17 ± 0.31	19 ± 0.44	20 ± 0.23	18 ± 0.27
31		15 ± 0.21	18 ± 0.23	14 ± 0.16	17 ± 0.11
32		20 ± 0.12	18 ± 0.24	19 ± 0.15	20 ± 0.32

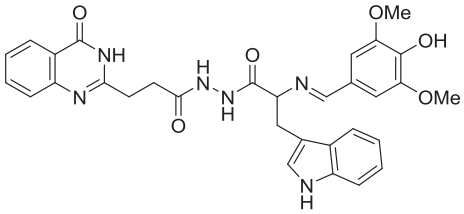
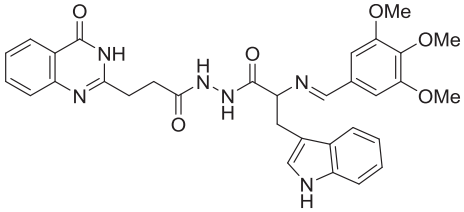
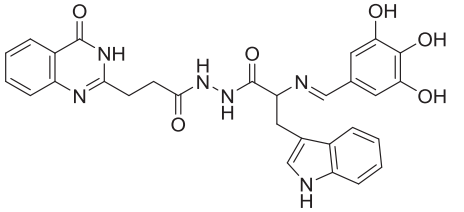
(*continued on next page*)

Table 1 (*continued*)

Sl. No	Structure	Antibacterial activity[a]		Antifungal activity[a]	
		S. aureus	*E. coli*	*A. niger*	*F. oxysporum*
33		23 ± 0.27	20 ± 0.21	22 ± 0.16	24 ± 0.26
34		21 ± 0.34	23 ± 0.27	23 ± 0.19	26 ± 0.24
35		10 ± 0.17	14 ± 0.23	11 ± 0.20	13 ± 0.14
36		09 ± 0.14	12 ± 0.15	16 ± 0.31	18 ± 0.12
37		10 ± 0.25	13 ± 0.26	14 ± 0.21	10 ± 0.10
38		11 ± 0.29	10 ± 0.15	18 ± 0.32	15 ± 0.20
39		18 ± 0.32	17 ± 0.16	20 ± 0.42	17 ± 0.16
40		17 ± 0.19	16 ± 0.14	16 ± 0.14	17 ± 0.16

(*continued on next page*)

Table 1 (*continued*)

Sl. No	Structure	Antibacterial activity[a]		Antifungal activity[a]	
		S. aureus	*E. coli*	*A. niger*	*F. oxysporum*
41		20 ± 0.30	21 ± 0.20	17 ± 0.14	19 ± 0.22
42		25 ± 0.22	22 ± 0.14	24 ± 0.14	26 ± 0.32
43		24 ± 0.22	21 ± 0.10	28 ± 0.15	23 ± 0.45
Std	**Streptomycin**	**10 ± 0.23**	**12 ± 0.21**	**–**	**–**
Std	**Bavistin**	**–**	**–**	**10 ± 0.21**	**13 ± 0.14**

[a] Vaues are mean of three determinations, the ranges of which are < 5% of the mean in all cases. Doxorubicin used as standard.

donating groups (OH and OCH_3) and phenylalanine and tryptophan amino acids are aromatic, more hydrophobic and acid base character of tryptophan indole nucleus. The number of hydroxyl and methoxy groups increase in the phenyl ring of the Schiff's bases the antimicrobial efficiency also increases [46]. The compounds **9–11**, **18–20**, **27–29**, **36-38** bearing electron withdrawing groups (F, Cl and NO_2) on phenyl ring of the Schiff's bases showed least antimicrobial properties. Among the glycine, alanine and phenylalanine conjugates, tryptophan conjugates shows most potent due to the presence of aromaticity, hydrophobicity, light unstable and stabilizing the amphiphilic structure is necessary for antimicrobial activity [47,48].

2.3. Molecular docking studies:

GlcN-6-P synthase is the member of amidotransferase subfamily of enzymes. It, catalysing the first committed step in a pathway leading to the eventual formation of uridine 5′-diphospho-*N*-acetyl-D-glucosamine (UDP-GlcNAc), is an essential building block of bacterial and fungal cell wall. UDP-GlcNAc is an important point of metabolic control in biosynthesis of amino sugar-containing macromolecules [49,50]. Hence, molecular docking was conducted with active site of *GlcN-6-P Synthase* (PDB ID: 2VF5). It showed good binding interaction with surrounded amino acid residues. The docking score and the interacting amino acids are tabulated in Table 2. Most of the synthesized compounds exhibited well established bond with one or more amino acids in the receptor active pocket of 2VF5 protein. The potentiality of the compounds as antimicrobial agents was determined based on docking score. Ligand **41** formed hydrogen bond interaction with Gly301, Ser303, Ala602 and Gln348 which is vital for a substrate to binding with 2VF5. Hence it clearly represents that ligand **41** (Fig. 1) could be a better bio-conjugate molecule against bacterial targeting 2VF5. Ligand **43** showed hydrogen bond interaction with Ser347, Ser349, Gln348, Lys603, Glu488 and Ala602 with a favorable energy pose (Fig. 2). Microbial infections are very closely related; hence we checked the potency of the ligands for antibacterial activities.

3. Conclusion

In short, we designed and synthesized a novel series of amino acids conjugated quinazolinone-Schiff's base analogues were screened for their *in vitro* antibacterial and antifungal activities. The compounds **14–16**, **21–25**, **30–34** and **36–43** showed good antimicrobial activities. To study the SAR, hydrophobic and aromatic amino acids like tryptophan and phenylalanine containing electron donating (OH and OCH_3) groups on the phenyl ring are most favour the antimicrobial activities and electron withdrawing (F, Cl and NO_2) groups showed least antimicrobial activity. Overall, the work is found to be highly versatile, which may be use lead to the development of new therapeutic agents useful in fighting diseases caused by microbes.

4. Experimental

4.1. General instrumentation and chemicals

All Boc-amino acids, EDCI, HOBt and TFA were purchased from Advanced Chem. Tech. (Louisville, Kentucky, USA). All the amino acids used except glycine were of L-configuration unless otherwise mentioned. NMM was purchased from Sigma Chemical Co. (St. Louis, MO). All chemicals and reagents were obtained from Aldrich (USA), Spectrochem Pvt. Ltd. (India) and Rankem Pvt. Ltd. (India) and were used without further purification. Progress of the reaction was monitored by TLC using silica gel coated on glass plates with solvent system comprising chloroform/methanol/acetic acid in the ratio 98:02:03 (R_f^a)/95:05:03 (R_f^b) and the compounds on TLC plates were detected by iodine vapors. Melting points were determined on a Superfit melting point apparatus (India) and are uncorrected. FT-IR was performed using a Jasco spectrometer (Japan) using nujol media. ^{1}H NMR (400 MHz) and ^{13}C NMR (100 MHz) spectra were recorded on a Agilent

Table 2
Antimicrobial docking studies with 2VF5 protein.

Sl. No	Docking score	Glide gscore	Glide energy	XP HBond
1	−5.116	−5.18	−44.38	−1.751
2	−5.15	−5.189	−32.126	−0.711
3	−4.932	−4.975	−41.213	−2.049
4	−7.667	−7.71	−57.64	−2.59
5	−7.68	−7.723	−51.197	−1.719
6	−5.446	−5.489	−53.586	−0.598
7	−5.357	−5.4	−54.217	0
8	−8.555	−8.624	−57.567	−2.199
9	−7.148	−7.213	−62.084	−2.211
10	−7.962	−8.026	−60.496	−1.911
11	−7.045	−7.11	−53.512	−1.636
12	−8.627	−8.691	−61.774	−4.65
13	−7.325	−7.389	−61.343	−2.533
14	−5.684	−5.748	−59.297	−1.576
15	−5.903	−5.988	−62.3	−1.771
16	−8.125	−8.2	−62.552	−4.502
17	−7.686	−7.754	−56.178	−1.8
18	−7.113	−8.461	−62.52	−2.729
19	−7.853	−7.917	−60.939	−3.237
20	−8.174	−8.238	−56.022	−2.978
21	**−10.504**	**−10.568**	**−63.718**	**−2.527**
22	−7.811	−7.875	−61.234	−2.255
23	−6.797	−6.862	−62.047	−1.115
24	−7.432	−8.801	−63.269	−3.012
25	−9.075	−9.149	−60.654	−3.01
26	−4.732	−4.796	−55.762	−1.654
27	−4.601	−4.665	−56.71	−1.435
28	−5.996	−6.06	−57.927	−0.35
29	−4.048	−4.112	−49.116	−0.568
30	−6.342	−6.406	−66.769	−1.8
31	−6.574	−6.638	−57.933	−2.666
32	−6.577	−6.642	−67.296	−1.63
33	−5.029	−5.1	−55.821	−1.442
34	−7.177	−7.251	−56.72	−2.717
35	−6.761	−6.826	−60.335	−1.112
36	−6.325	−6.389	−59.718	−0.14
37	−4.474	−4.517	−49.126	0
38	−6.112	−7.46	−67.846	−1.64
39	−8.286	−8.35	−72.022	−2.898
40	**−9.184**	**−9.048**	**−63.929**	**−3.716**
41	**−9.968**	**−9.933**	**−67.673**	**−2.907**
42	−7.315	−8.667	−73.167	−1.809
43	**−8.962**	**−11.37**	**−67.352**	**−4.382**
Streptomycin	**−7.744**	**−7.917**	**−45.403**	**−4.854**
Bavistin	**−5.953**	**−6.14**	**−42.862**	**−3.231**

Technologies (USA) using DMSO (d_6) as solvent and the chemical shifts are reported as parts per million (δ ppm) using TMS as an internal standard. High resolution mass spectroscopic analysis was performed on a BrukerMicroTOF QII mass spectrometer in positive mode.

4.2. Synthesis

4.2.1. Synthesis of methyl 3-(4-oxo-3,4-dihydroquinazolin-2-yl) propanoate

To a solution of QZN (0.02 mol, 4.36 g) dissolved in methanol (40 mL), trimethylsilyl chloride (0.02 mol, 3.80 mL) was added slowly. The reaction mixture was stirred for 4 hrs to complete the reaction (monitored by TLC). The solvent was removed under reduced pressure and the resultant precipitate was washed with ice cold water and filtered to yield the desired product **2**.

4.2.2. Synthesis of 3-(4-oxo-3,4-dihydroquinazolin-2-yl)propane hydrazide

To a solution of **2** (0.015 mol, 4.02 g) dissolved in ethanol (40 mL), hydrazine hydrate (0.020 mol, 0.97 mL) was added. The reaction mixture was refluxed for 16 hrs for completion of the reaction (monitored by TLC). The solvent was removed under reduced pressure and cooled by adding ice cold water. The resulting precipitate was filtered, washed with cold water and recrystallized from ethanol to get the desired compound **3**.

4.2.3. General procedure for the conjugation of Boc-amino acids to 3-(4-oxo-3,4-dihydroquinazolin-2-yl)propanoic acid (QZN)

Boc-amino acids (1 mmol) dissolved in dimethyl formamide (DMF) separately (10 mL/g of compound) and cooled to 0 °C was added NMM (1 mmol). EDCI (1 mmol) was added under stirring while maintaining the temperature at 0 °C and stirred for 15 min. HOBt (1 mmol) in DMF (2 mL) was added. The reaction mixture was stirred for an additional 10 min and a pre-cooled solution of 3-(4-oxo-3,4-dihydroquinazolin-2-yl)propanoic acid (QZN) (1 mmol) and NMM (1 mmol) in DMF was added slowly. After 20 min, pH of the solution was adjusted to 8 by the addition of NMM and the reaction mixture was stirred overnight at room temperature. DMF was removed under reduced pressure and the residue was poured into about 100 mL ice-cold 90% saturated $KHCO_3$ solution and stirred for 30 min. The precipitated product was taken into $CHCl_3$ and washed sequentially with 5% $NaHCO_3$ solution (2 × 20 mL), water (2 × 20 mL), 0.1 N cold HCl solution (2 × 20 mL) and finally brine (2 × 20 mL). The $CHCl_3$ layer was dried over anhydrous Na_2SO_4 and the solvent was removed under reduced pressure. The products so

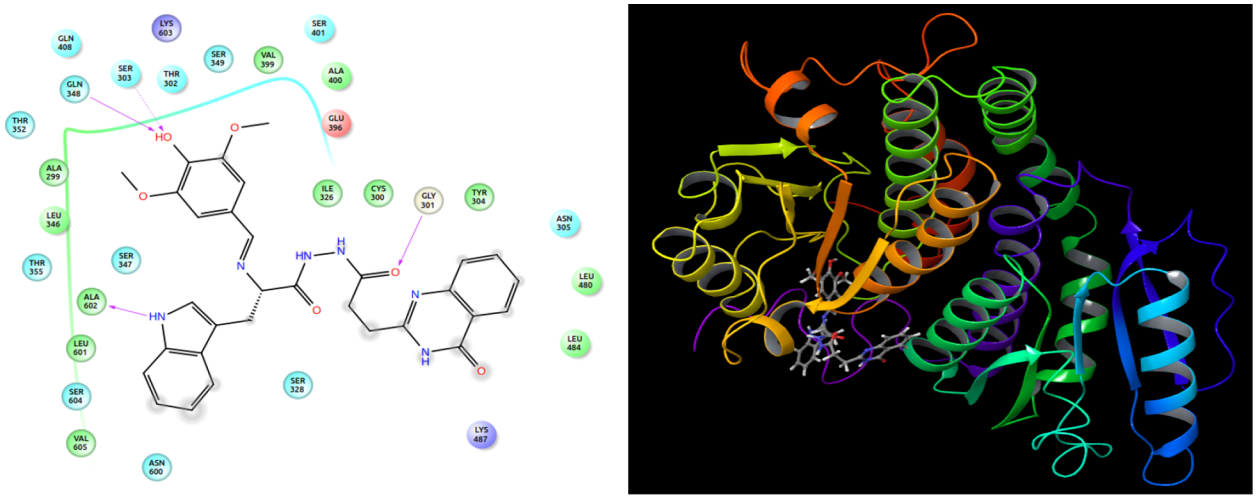


Fig. 1. 2D and 3D image of compound 41 with 2VF5 protein.

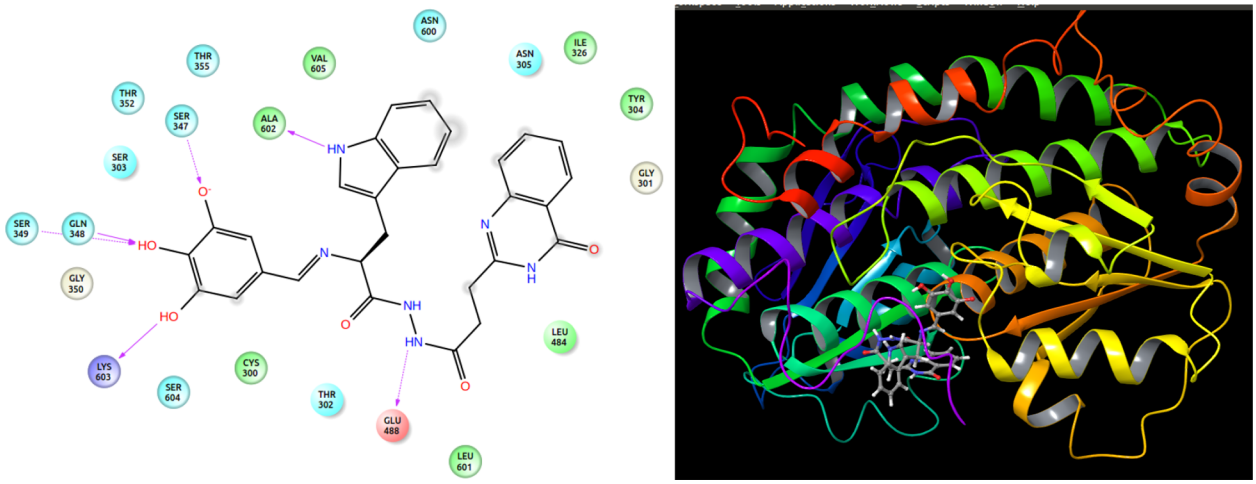


Fig. 2. 2D and 3D image of compound 43 with 2VF5 protein.

obtained were recrystallized from ether/petroleum ether to get desired products **4–7**.

4.2.4. Deblocking of Boc group

QZN-Xaa-Boc (1 mmol) was stirred with 2.0 mL of TFA for 45 min at room temperature. After completion of the reaction monitored by TLC, reaction mixture was concentrated at high vacuum to get QZN-Xaa-H. TFA which was then triturated with dry ether, filtered and dried.

*4.2.5. Procedure for the synthesis of Schiff's bases (**8–43**)*

QZN-Xaa-H. TFA (1 mmol) were dissolved in minimum quantity of DMF and then neutralized with NMM as a base, after the neutralization ethanol (10 mL/g of compound) was added and treated with appropriate aldehydes (1 mmol) in the presence of catalytic amount of glacial acetic acid. The reaction mixtures were refluxed for 16hrs and the completion of reaction was monitored by TLC. After completion of the reaction, the solvent was removed under reduced pressure and cooled by adding ice cold water. The resulting precipitate was filtered, washed with water and recrystallized from ethanol to obtain the desired amino acids derived quinazolinone Schiff's bases **(8–43)**.

*4.3. 3-(4-oxo-3,4-dihydroquinazolin-2-yl)propanoic acid (**1**)*

Yield 86%, white solid, R_f^a = 0.40, R_f^b = 0.48, m.p. 206–207 °C, (lit 205–206 °C), MS *m/z*: 219.1080, IR KBr (cm^{-1}): 1630, 1768, 2910, 3510, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ*: 12.16 (2H, s, NH& COOH), 8.05–7.40 (4H, m, Ar-H), 2.83–2.78 (2H, t, CH_2), 2.72–2.69 (2H, t, CH_2);^{13}C NMR (DMSO-d_6, 100 MHz) *δ*: 173.9, 162.0, 156.7, 146.7, 134.6, 127.2, 126.4, 126.1, 121.3, 30.2, 29.5.

*4.3.1. Methyl 3-(4-oxo-3,4-dihydroquinazolin-2-yl)propanoate (**2**)*

Yield 93%, white solid, R_f^a = 0.66, R_f^b = 0.70, m.p. 184–185 °C, IR KBr (cm^{-1}): 1627, 1770, 2945, MS *m/z*: 219.1245, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ*: 11.31 (1H, s, Het-NH), 8.15–7.59 (4H, m, Ar-H), 3.61 (3H, s, OCH_3), 3.11–3.08 (2H, t, CH_2), 3.00–2.97 (2H, t, CH_2); ^{13}C NMR (DMSO-d_6, 500 MHz) *δ*: 173.0, 168.0, 156.2, 146.7, 133.7, 129.2, 126.3, 126.1, 121.3, 51.8, 30.3, 29.1.

*4.3.2. 3-(4-Oxo-3,4-dihydroquinazolin-2-yl)propanehydrazide (**3**)*

Yield 83%, white solid, R_f^a = 0.32, R_f^b = 0.36, m.p. 220–221 °C, MS *m/z*: 247.1264, IR KBr (cm^{-1}): 1640, 1778, 1940, 3312, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ*: 11.20 (1H, s, Het-NH), 8.99 (1H, s, NH), 8.02–7.38 (4H, m, ArH), 4.14 (2H, s, NH_2), 2.79–2.77 (2H, t, CH_2), 2.46–2.48 (2H, t, CH_2), ^{13}C NMR (DMSO-d_6, 100 MHz) *δ*: 173.2, 167.0, 156.7, 146.1, 133.6, 128.2, 127.1, 126.8, 121.2, 30.1, 29.4.

*4.3.3. Tert-butyl (2-oxo-2-(2-(3-(4-oxo-3,4-dihydroquinazolin-2-yl) propanoyl)hydrazinyl)ethyl)carbamate (**4**)*

Yield 85%, white solid. R_f^a = 0.52, R_f^b = 0.59, m.p. 220–221 °C, MS *m/z*: (M+1) 390.2564, IR KBr (cm^{-1}): 1633, 1780, 2947, 3214, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ*: 11.10 (1H, s, Het-NH), 10.70 (1H, s, NH), 8.05–7.17 (4H, m, ArH), 6.73–6.72 (1H, d, NH), 3.90–3.89 (2H, s, $^{\alpha}CH_2$), 3.87 (1H, s, NH), 2.92–2.91 (2H, t, CH_2), 2.63–2.61 (2H, t, CH_2), 1.30 (9H, s, Boc-H), ^{13}C NMR (DMSO-d_6, 100 MHz) *δ*: 173.2, 171.1, 161.1, 156.7, 156.1, 148.1, 133.1, 127.4, 126.9, 126.1, 120.1, 78.8, 48.1, 30.1, 29.4, 24.3.

*4.3.4. Tert-butyl (1-oxo-1-(2-(3-(4-oxo-3,4-dihydroquinazolin-2-yl) propanoyl)hydrazinyl)propan-2-yl)carbamate (**5**)*

Yield 82%, white solid, R_f^a = 0.49, R_f^b = 0.58, m.p. 156–157 °C, MS *m/z*, (M+1): 404.1548, IR KBr (cm^{-1}): 1630, 1748, 2917, 3312, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ*: 11.10 (1H, s, Het-NH), 8.01–7.31 (4H, m, ArH), 6.91–6.89 (2H, d, 2NH), 3.98–3.94 (1H, m, $^{\alpha}CH$), 3.41 (1H, s, NH), 2.80–2.77 (2H, t, CH_2), 2.59–2.56 (2H, t, CH_2), 1.33 (9H, s, Boc-H), 1.22–1.20 (3H, d, CH_3), ^{13}C NMR (DMSO-d_6, 100 MHz) *δ*: 171.2, 170.0, 164.4, 159.6, 155.3, 149.7, 133.7, 126.7, 126.1, 125.4, 121.3, 78.4, 48.8, 31.3, 30.7, 28.6, 18.7.

*4.3.5. Tert-butyl (1-oxo-1-(2-(3-(4-oxo-3,4-dihydroquinazolin-2-yl) propanoyl)hydrazinyl)-3-phenylpropan-2-yl)carbamate (**6**)*

Yield 82%, white solid, brown solid, R_f^a = 0.50, R_f^b = 0.63, m.p. 180–182 °C, MS *m/z*, (M+1): 480.1659, IR KBr (cm^{-1}): 1640, 1785, 2917, 3216, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ*: 10.71 (1H, s, Het-NH), 8.05–7.12 (9H, m, ArH), 6.90–6.92 (1H, d, NH), 4.22–4.17 (1H, m, $^{\alpha}CH$), 3.31 (3H, s, NH), 3.01–2.96 (d, 2H, d, $^{\beta}CH_2$), 2.88–2.84 (2H, m, CH_2), 2.67–2.66 (2H, t, CH_2), 1.28 (9H, s, Boc-H), ^{13}C NMR (DMSO-d_6, 100 MHz) *δ*: 171.4, 170.2, 164.4, 159.6, 156.7, 148.1, 137.1, 133.1, 128.7, 127.6, 127.1, 126.9, 126.1, 125.1, 121.1, 78.1, 58.4, 38.1, 30.1, 29.9, 25.7.

*4.3.6. Tert-butyl (3-(1H-indol-3-yl)-1-oxo-1-(2-(3-(4-oxo-3,4-dihydroquinazolin-2-yl)propanoyl)hydrazinyl)propan-2-yl)carbamate (**7**)*

Yield 84%, reddish solid, R_f^a = 0.54, R_f^b = 0.63, m.p. 184–186 °C, MS *m/z*, (M+1): 519.2365, IR KBr (cm^{-1}): 1640, 1772, 2942, 3220, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ*: 11.23 (1H, s, Het-NH), 10.7 (1H, s, Indole-NH), 8.14–7.03 (8H, m, ArH), 6.72–6.71 (1H, d, NH), 4.95–4.94 (1H, m, $^{\alpha}CH$), 3.41–3.40 (2H, d, $^{\beta}CH_2$), 3.68 (2H, s, NH), 3.12–3.10 (2H, m, CH_2), 2.70–2.68 (2H, t, CH_2), 1.29 (9H, s, Boc-H), ^{13}C NMR (DMSO-d_6, 100 MHz) *δ*: 171.1, 170.7, 163.7, 159.1, 156.1, 148.7, 136.7, 133.4, 128.1, 127.1, 126.4, 126.0, 124.1, 122.1, 121.6, 119.2, 118.1, 114.1, 108.7, 79.0, 60.1, 31.1, 29.1, 28.40, 24.3.

*4.3.7. N-(2-(Benzylideamino)acetyl)-3-(4-oxo-3,4-dihydroquinazolin-2-yl)propanehydrazide (**8**)*

Yield 82%, white solid, R_f^a = 0.57, R_f^b = 0.68, m.p. 167–168 °C, MS *m/z*, (M + 1): 378.2315, IR KBr (cm^{-1}): 1612, 1620, 1763, 2917, 3220, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ*: 12.18 (1H, s, Het-NH), 11.46 (1H, s, NH), 8.19–8.18 (1H, d, NH), 8.05–7.37 (9H, m, ArH), 7.96 (1H, s, - N = CH), 4.22 (2H, s, $^{\alpha}CH_2$), 2.86–2.82 (2H, t, CH_2), 2.71–2.70 (2H, t, CH_2), ^{13}C NMR (DMSO-d_6, 100 MHz) *δ*:171.9, 170.7, 162.0, 157.1, 147.0, 143.8, 134.6, 134.5, 130.2, 129.2, 128.7, 127.4, 126.3, 126.1, 121.3, 60.0, 31.7, 30.2.

4.3.8. N-(2-((4-Chlorobenzylidene)amino)acetyl)-(3-(4-oxo-3,4-dihydroquinazolin-2-yl)propanehydrazide (9)

Yield 88%, white solid, R_f^a = 0.45, R_f^b = 0.60, m.p. 188–191 °C, MS *m/z*, (M + 1): 412.2316, IR KBr (cm^{-1}): 1609, 1628, 1760, 2930, 3226, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ*: 11.13 (1H, s, Het-NH), 10.80 (1H, s, NH), 8.14–7.16 (8H, m, ArH), 7.84 (1H, s, CH), 6.72–6.71 (1H, d, NH), 4.52–4.51 (2H, m, $^{\alpha}CH_2$), 8.87–2.85 (2H, t, CH_2), 2.66–2.64 (2H, t, CH_2), ^{13}C NMR (DMSO-d_6, 100 MHz) *δ*: 171.4, 170.2, 163.1, 160.7, 156.1, 148.1, 137.1, 135.1, 133.7, 131.1, S129.4, 127.9, 127.0, 126.8, 120.1, 58.7, 30.9, 29.8.

*4.3.9. N′-(2-((4-Nitrobenzylidene)amino)acetyl)-(3-(4-oxo-3,4-dihydroquinazolin-2-yl)propanehydrazide (**10**)*

Yield 83%, yellow solid, R_f^a = 0.57, R_f^b = 0.63, m.p. 179–181 °C, MS *m/z* (M + 1): 423.6231, IR KBr (cm^{-1}): 1619, 1642, 1778, 2943, 3246, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ*: 11.09 (s, 1H, Het-NH), 8.09–8.10 (m, 8H, Ar-H), 7.80 (s, 1H, - N = CH), 6.72–6.73 (d, 1H, NH), 4.52–4.53 (m, 2H, $^{\alpha}CH_2$), 3.70 (s, 2H, NH), 3.10–3.12 (m, 2H, CH_2), 2.66–2.68 (t, 2H, CH_2), ^{13}C NMR (DMSO-d_6, 100 MHz) *δ* ppm: 171.1, 170.3, 161.9, 160.0, 156.1, 149.4, 148.3, 143.0, 133.7, 130.2, 127.4, 126.7, 126.6, 125.4, 121.45, 60.9, 30.3, 29.2.

*4.3.10. N-(2-((4-Fluorobenzylidene)amino)acetyl)-(3-(4-oxo-3,4-dihydroquinazolin-2-yl)propanehydrazide (**11**)*

Yield 83%, brown solid, R_f^a = 0.50, R_f^b = 0.58, m.p. 166–167 °C, MS *m/z*, (M + 1): 396.2364, IR KBr (cm^{-1}): 1614, 1635, 1778, 2946, 3226, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ*: 11.20 (1H, s, Het-NH), 10.27 (1H, s, NH), 8.07–7.26 (8H, m, ArH), 7.84 (s, 1H, CH), 6.71–6.72 (1H, d, NH), 4.52–4.50 (2H, m, $^{\alpha}CH_2$), 2.81–2.80 (2H, t, CH_2), 2.56–2.54 (2H, t, CH_2), ^{13}C NMR (DMSO-d_6, 100 MHz) *δ*: 172.2, 171.1, 165.1, 161.7, 156.1, 148.1, 144.2, 135.1, 132.1, 129.2, 128.7, 127.6, 126.4, 120.9, 116.3, 60.2, 31.2, 29.7.

*4.3.11. N′-(2-((3,4-Dihydroxybenzylidene)amino)acetyl)-(3-(4-oxo-3,4-dihydroquinazolin-2-yl)propanehydrazide (**12**)*

Yield 77%, white solid, R_f^a = 0.37, R_f^b = 0.43, m.p. 186–188 °C, MS *m/z*, (M + 1): 410.2315, IR KBr (cm^{-1}): 1621, 1624, 1784, 2914, 3256, 3561, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ*: 11.10 (1H, s, Het-NH), 10.88 (1H, s, NH), 9.85 (2H, s, OH), 8.03–7.10 (7H, m, Ar-H), 7.78 (s, 1H, −N═CH), 6.90–6.88 (1H, d, NH), 4.52–4.51 (2H, s, $^{\alpha}CH_2$), 2.90–2.88 (2H, t, CH_2), 2.66–2.64 (2H, t, CH_2), ^{13}C NMR (DMSO-d_6, 500 MHz) *δ* ppm: 172.8, 170.8, 161.7, 156.6, 149.8, 148.5, 147.3, 143.9, 134.7, 133.8, 127.2, 126.7, 126.2, 123.9, 121.0, 117.8, 115.5, 59.2, 30.9, 29.2.

*4.3.12. N′-(2-((4-Hydroxy-3-methoxybenzylidene)amino)acetyl)-(3-(4-oxo-3,4-dihydroquinazolin-2-yl)propanehydrazide (**13**)*

Yield 76%, white solid, R_f^a = 0.54, R_f^b = 0.61, m.p. 155–157 °C, MS *m/z*, (M + 1): 424.2364, IR KBr (cm^{-1}): 1616, 1636, 1780, 2940, 3316, 3564, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ*: 11.13 (1H, s, Het-NH), 10.22 (1H, s, NH), 9.12 (1H, s, OH), 8.10–7.03 (7H, m, Ar-H), 7.89 (s, 1H, −N═CH), 6.71–6.72 (1H, d, NH), 4.53–4.52 (2H, s, $^{\alpha}CH_2$), 3.72 (3H, s, −OCH_3) 2.89–2.87 (2H, t, CH_2), 2.60–2.58 (2H, t, CH_2), ^{13}C NMR (DMSO-d_6, 100 MHz) *δ*: 173.2, 171.8, 161.4, 156.1, 153.6, 148.1, 146.5, 144.6, 135.7, 133.4, 128.2, 127.7, 126.2, 122.4, 120.8, 116.8, 112.5, 60.2, 52.7, 30.2, 29.8.

*4.3.13. N-(2-((4-Hydroxy-3,5-dimethoxybenzylidene)amino)acetyl)-(3-(4-oxo-3,4-dihydroquinazolin-2-yl)propanehydrazide (**14**)*

Yield 84%, white solid, R_f^a = 0.53, R_f^b = 0.70 m.p. 186–188 °C, MS *m/z*, (M + 1): 454.2364, IR KBr (cm^{-1}): 1626, 1640, 1783, 2954, 3217, 3567, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ*: 11.30 (1H, s, Het-NH), 10.50 (1H, s, NH), 8.95 (1H, s, OH), 8.09–7.01 (6H, m, Ar-H), 7.98 (1H, s, - N = CH), 6.50–6.49 (1H, d, NH), 4.52–4.51 (2H, s, $^{\alpha}CH_2$), 3.71 (6H, s, $2OCH_3$), 2.80–2.78 (2H, t, CH_2), 2.62–2.60 (2H, t, CH_2), ^{13}C NMR (DMSO-d_6, 100 MHz) *δ*: 174.2, 172.8, 161.5, 156.3, 148.7, 148.1, 143.7, 139.7, 135.5, 133.8, 127.7, 126.8, 126.4, 120.5, 104.9, 59.8, 52.8, 30.5, 29.2.

*4.3.14. 3-(4-Oxo-3,4-dihydroquinazolin-2-yl)-N-(2-((3,4,5-trimethoxybenzylidene)amino)acetyl)propanehydrazide (**15**)*

Yield 82%, white solid, R_f^a = 0.56, R_f^b = 0.65, m.p. 190–192 °C, MS *m/z*, (M + 1): 468.2164, IR KBr (cm^{-1}): 1614, 1637, 1783, 2941, 3229, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ*: 11.15 (s, 1H, Het-NH), 9.60 (1H, s, NH), 8.13–6.92 (6H, m, Ar-H), 7.79 (1H, s, −N═CH), 6.69–6.68 (1H, d, NH), 4.52–4.50 (2H, m, $^{\alpha}CH_2$), 3.81 (9H, s, $3OCH_3$), 2.90–2.88 (2H, m, CH_2), 2.63–2.61 (2H, t, CH_2), ^{13}C NMR (DMSO-d_6, 100 MHz) *δ*: 171.19, 170.87, 163.10, 160.14, 156.20, 152.17, 148.70, 142.08, 138.16, 133.70, 128.14, 127.17, 126.90, 121.31, 104.60, 60.80, 59.80, 56.44, 30.14, 29.17.

*4.3.15. 3-(4-Oxo-3,4-dihydroquinazolin-2-yl)-N-(2-((3,4,5-trihydroxybenzylidene)amino)acetyl)propanehydrazide (**16**)*

Yield 76%, white solid, R_f^a = 0.33, R_f^b = 0.41, m.p. 210–211 °C, MS *m/z*, (M + 1): 426.3216, IR KBr (cm^{-1}): 16139, 1620, 1758, 2940, 3245, 3641, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ*: 11.25 (1H, s, Het-NH), 9.20 (1H, s, NH), 9.85 (2H, s, 2OH), 8.95 (IH, s, OH), 8.10–6.72 (6H, m, Ar-H), 7.79 (1H, s, - N = CH), 6.68–6.69 (1H, d, NH), 4.52–4.51 (2H, s, $^{\alpha}CH_2$), 2.78–2.76 (2H, t, CH_2), 2.60–2.58 (2H, t, CH_2), ^{13}C NMR (DMSO-d_6, 100 MHz) *δ* ppm: 173.2, 171.7, 161.8, 156.9, 148.7, 146.5, 144.7, 139.5, 135.8, 133.8, 128.8, 127.1, 126.5, 120.8, 109.2, 59.8, 30.8, 29.6.

*4.3.16. 2-(Benzylideneamino)-N-(3-(4-oxo-3,4-dihydroquinazolin-2-yl) propanoyl)propanehydrazide (**17**)*

Yield 80%, white solid, R_f^a = 0.53, R_f^b = 0.60, m.p. 178–179 °C, MS *m/z*, (M + 1): 392.1645, IR KBr (cm^{-1}): 1616, 1633, 1777, 2912, 3238, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ*: 11.16 (1H, s, Het-NH), 9.90 (1H, s, NH), 8.03–7.36 (9H, m, Ar-H), 7.88 (1H, s, −N═CH), 6.71–6.70 (1H, d, NH), 4.21–4.20 (1H, m, $^{\alpha}CH$), 2.90–2.88 (2H, t, CH_2), 2.60–2.58 (2H, t, CH_2), 1.39–1.38 (3H, d, $^{\beta}CH_3$), ^{13}C NMR (DMSO-d_6, 100 MHz) *δ*: 173.8, 171.6, 161.3, 156.7, 148.5, 143.9, 136.7, 133.5, 130.5, 129.5, 128.6, 127.1, 126.9, 126.2, 120.9, 69.1, 30.7, 29.8, 19.1.

*4.3.17. 2-((4-Chlorobenzylidene)amino)-N–3-(4-oxo-3,4-dihydroquinazolin-2-yl)propanoyl)propanehydrazide (**18**)*

Yield 81%, white solid, R_f^a = 0.54, R_f^b = 0.65, m.p. 197–198 °C, MS *m/z*, (M + 1):426.3164, IR KBr (cm^{-1}): 1614, 1636, 1778, 2938, 3260, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ*: 11.17 (1H, s, Het-NH), 9.90 (1H, s, NH), 8.10–6.88 (8H, m, Ar-H), 7.80 (1H, s, CH), 6.71–6.70 (d, 1H, NH), 4.77–4.75 (1H, m, $^{\alpha}CH$), 2.85–2.83 (2H, m, CH_2), 2.68–2.66 (2H, t, CH_2), 1.39–1.38 (3H, s, $^{\beta}CH_3$), ^{13}C NMR (DMSO-d_6, 100 MHz) *δ*: 171.70, 170.11, 163.01, 160.87, 156.24, 148.14, 136.70, 134.17, 133.07, 129.14, 127.16, 127.01, 126.14, 121.19, 70.81, 30.17, 29.81, 18.67.

*4.3.18. 2-((4-Nitrobenzylidene)amino)-N-(3-(4-oxo-3,4-dihydroquinazolin-2-yl)propanoyl)propanehydrazide (**19**)*

Yield 86%, yellow solid, R_f^a = 0.52, R_f^b = 0.58, m.p. 185–186 °C, MS *m/z*. (M + 1): 437.2364, IR KBr (cm^{-1}): 1627, 1629, 1785, 2964, 3306, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ*: 11.14 (s, 1H, Het-NH), 8.50

(1H, s, NH), 8.12–6.86 (8H, m, Ar-H), 7.73 (1H, s, −N═CH), 6.63–6.61 (1H, d, NH), 4.71–4.70 (1H, m, $^{\alpha}$CH), 2.92–2.90 (2H, m, CH_2), 2.65–2.63 (2H, t, CH_2), 1.40–1.38 (3H, s, $^{\beta}CH_3$), ^{13}C NMR (DMSO-d_6, 500 MHz) *δ*: 171.16, 170.88, 163.71, 160.70, 156.23, 149.14, 148.28, 143.70, 133.70, 130.14, 128.70, 126.77, 126.01, 124.11, 121.10, 70.14, 30.16, 29.317, 18.70.

4.3.19. 2-((4-Fluorobenzylidene)amino)-N–3-(4-oxo-3,4-dihydroquinazolin-2-yl)propanoyl)propanehydrazide ***(20)***

Yield 81%, brown solid, R_f^a = 0.47, R_f^b = 0.56, m.p. 147–149 °C, MS *m/z*, (M + 1): 410.3648, IR KBr (cm^{-1}): 1607, 1642, 1781, 2940, 3213, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ*: 11.12 (1H, s, Het-NH), 9.56 (1H, s, NH), 8.03–7.24 (8H, m, Ar-H), 7.89 (1H, s, −N═CH), 6.68–6.67 (1H, d, NH), 4.22–4.21 (1H, m, $^{\alpha}$CH), 3.87–3.86 (2H, t, CH_2), 2.61–2.59 (2H, t, CH_2), 1.40–1.39 (3H, d, $^{\beta}CH_3$), ^{13}C NMR (DMSO-d_6, 100 MHz) *δ*: 172.7, 170.5, 165.6, 161.4, 156.2, 148.9, 143.6, 134.7, 131.6, 130.5, 127.2, 127.1, 126.8, 120.5, 115.3, 68.7, 30.7, 29.5, 17.9.

4.3.20. 2-((3,4-Dihydroxybenzylidene)amino)-N-(3-(4-oxo-3,4-dihydroquinazolin-2-yl)propanoyl)propanehydrazide ***(21)***

Yield 85%, white solid, R_f^a = 0.46, R_f^b = 0.51, m.p. 170–171 °C, MS *m/z*, (M + 1): 424.2346, IR KBr (cm^{-1}): 1605, 1632, 1771, 2939, 3214, 3561, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ*: 11.28 (1H, s, Het-NH), 9.50 (1H, s, NH), 8.59 (2H, s, 2OH), 8.11–6.68 (7H, m, Ar-H), 7.85 (1H, s, −N═CH), 6.63–6.62 (1H, d, NH), 4.26–4.25 (1H, m, $^{\alpha}$CH), 2.80–2.78 (2H, t, CH_2), 2.60–2.58 (2H, t, CH_2), 1.36–1.35 (3H, d, $^{\beta}CH_3$), ^{13}C NMR (DMSO-d_6, 100 MHz) *δ* ppm: 171.8, 170.6, 161.9, 156.2, 150.7, 148.7, 147.5, 144.2, 133.8, 133.1, 127.2, 126.8, 126.2, 124.2, 120.9, 118.2, 117.0, 68.1, 30.2, 29.3, 17.9.

4.3.21. 2-((4-Hydroxy-3-methoxybenzylidene)amino)-N-(3-(4-oxo-3,4-dihydroquinazolin-2-yl)propanoyl)propanehydrazide ***(22)***

Yield 85%, white solid, R_f^a = 0.53, R_f^b = 0.61, m.p. 175–176 °C, MS *m/z*, (M + 1): 438.4231, IR KBr (cm^{-1}): 1607, 1636, 1786, 2915, 3239, 3564, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ*: 11.16 (1H, s, Het-NH), 9.89 (1H, s, NH), 8.59 (1H, s, OH), 8.09–6.88 (7H, m, Ar-H), 7.95 (1H, s, −N═CH), 6.55–6.54 (1H, d, NH), 4.28–4.27 (1H, m, $^{\alpha}$CH), 3.71 (3H, s, OCH_3), 2.84–2.83 (2H, t, CH_2), 2.79–2.77 (2H, t, CH_2), 1.37–1.36 (3H, d, $^{\beta}CH_3$), ^{13}C NMR (DMSO-d_6, 100 MHz) *δ*: 172.3, 171.3, 161.8, 156.4, 152.6, 149.4, 147.1, 144.2, 134.5, 133.7, 128.1, 127.7, 126.2, 122.8, 120.7, 118.8, 111.7, 69.8, 52.6, 30.8, 29.7, 17.9.

4.3.22. 2-((4-Hydroxy-3,5-dimethoxybenzylidene)amino)-N-(3-(4-oxo-3,4-dihydroquinazolin-2-yl)propanoyl)propanehydrazide ***(23)***

Yield 79%, brown solid, R_f^a = 0.49, R_f^b = 0.57, m.p. 193–194 °C, MS *m/z*, (M + 1): 468.3264, IR KBr (cm^{-1}): 1612, 1636, 1784, 2905, 3236, 3564, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ*: 11.32 (1H, s, Het-NH), 9.90 (1H, s, NH), 8.55 (1H, s, OH), 8.06–7.10 (6H, m, Ar-H), 7.88 (1H, s, −N═CH), 6.60–6.59 (1H, d, NH), 4.26–4.25 (1H, m, $^{\alpha}$CH), 3.82 (6H, s, $2OCH_3$), 2.86–2.84 (2H, t, CH_2), 2.59–2.57 (2H, t, CH_2), 1.38–1.37 (3H, d, $^{\beta}CH_3$), ^{13}C NMR (DMSO-d_6, 100 MHz) *δ* ppm: 172.7, 171.6, 162.0, 156.9, 148.8, 148.2, 143.9, 140.7, 135.7, 133.6, 127.8, 126.9, 126.1, 120.5, 104.7, 68.2, 52.7, 30.3, 29.8, 18.3.

4.3.23. 3-(4-Oxo-3,4-dihydroquinazolin-2-yl)-N-(2-((3,4,5-trimethoxybenzylidene)amino)propanoyl)propanehydrazide ***(24)***

Yield 78%, white solid, R_f^a = 0.59, R_f^b = 0.64, m.p. 210–212 °C, MS *m/z*, (M + 1): 482.3645, IR KBr (cm^{-1}): 1626, 1634, 1786, 2943, 3304, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ*: 11.19 (s, 1H, Het-NH), 9.52 (1H, s, NH), 8.14–6.90 (6H, m, Ar-H), 7.71 (s, 1H, −N═CH), 6.61–6.59 (1H, d, NH), 4.73–4.72 (2H, m, $^{\alpha}$CH), 3.83 (9H. s, $3OCH_3$), 2.87–2.85 (2H, t, CH_2), 2.62–2.60 (2H, t, CH_2), 1.39–1.37 (3H, d, 3H, $^{\beta}CH_3$), ^{13}C NMR (DMSO-d_6, 100 MHz) *δ*: 171.90, 170.27, 162.14, 160.88, 156.23, 153.27, 148.14, 142.14, 135.14, 133.19, 127.23, 126.92, 126.17, 121.14, 104.78, 70.07, 60.81, 56.44, 30.22, 29.18, 18.66.

4.3.24. 3-(4-Oxo-3,4-dihydroquinazolin-2-yl)-N-(2-((3,4,5-trihydroxybenzylidene)amino)propanoyl)propanehydrazide ***(25)***

Yield 85%, white solid, R_f^a = 0.34, R_f^b = 0.42, m.p. 169–170 °C, MS *m/z*, (M + 1): 440.1364, IR KBr (cm^{-1}): 1601, 1624, 1745, 2964, 3289, 3610, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ*: 11.22 (1H, s, Het-NH), 10.60 (1H, s, NH), 9.10 (2H, s, 2OH), 8.89 (1H, s, OH), 8.08–6.72 (7H, m, Ar-H), 7.89 (1H, s, −N═CH), 6.56–6.54 (1H, d, NH), 4.23–4.22 (1H, m, $^{\alpha}$CH), 2.79–2.77 (2H, t, CH_2), 2.2.56–2.54 (2H, t, CH_2), 1.37–1.36 (3H, d, $^{\beta}CH_3$), ^{13}C NMR (DMSO-d_6, 100 MHz) *δ* ppm: 171.7, 170.6, 161.3, 156.8, 147.9, 146.5, 144.2, 139.7, 136.2, 133.5, 128.2, 127.1, 126.5, 120.4, 109.6, 67.9, 30.8, 29.4, 18.2.

4.3.25. 2-(Benzylideneamino)-N-(3-(4-oxo-3,4-dihydroquinazolin-2-yl) propanoyl)-3-phenylpropanehydrazide ***(26)***

Yield 85%, white solid, R_f^a = 0.52, R_f^b = 0.60, m.p. 181–182 °C, MS *m/z*, (M + 1): 468.2368, IR KBr (cm^{-1}): 1613, 1633, 1786, 2951, 3260, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ* ppm: 11.10 (1H, s, Het-NH), 10.23 (1H, s, NH), 8.07–7.26 (14H, m, Ar-H), 7.89 (1H, s, −N═CH), 6.71–6.70 (1H, d, NH), 4.46–4.48 (1H, m, $^{\alpha}$CH), 3.30–3.29 (2H, t, $^{\beta}CH_2$), 2.88–2.86 (2H, t, CH_2), 2.69–2.67 (2H, t, CH_2), ^{13}C NMR (DMSO-d_6, 100 MHz) *δ* ppm: 172.8, 171.0, 161.7, 156.4, 148.3, 143.7, 138.2, 136.5, 133.8, 130.4, 129.9, 128.7, 128.4, 127.9, 127.2, 126.9, 126.4, 125.3, 120.3, 74.6, 38.6, 30.7, 29.8.

4.3.26. 2-((4-Chlorobenzylidene)amino)-N-(3-(4-oxo-3,4-dihydroquinazolin-2-yl)propanoyl)-3-phenylpropanehydrazide ***(27)***

Yield 81%, white solid, R_f^a = 0.49, R_f^b = 0.60, m.p. 194–196 °C, MS *m/z*, (M + 1): 502.6521, IR KBr (cm^{-1}): 1611, 1626, 1776, 2938, 3229, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ*: 11.23 (1H, s, Het-NH), 10.53 (1H, s, NH), 8.04–7.12 (13H, m, Ar-H), 7.83 (1H, s, −N═CH), 6.70–6.69 (1H, d, NH), 4.42–4.41 (1H, m, $^{\alpha}$CH), 3.31–3.29 (2H, t, $^{\beta}CH_2$), 2.79–2.77 (2H, t, CH_2), 2.68–2.67 (2H, t, CH_2), ^{13}C NMR (DMSO-d_6, 100 MHz) *δ* ppm: 172.8, 171.6, 161.9, 156.4, 148.7, 144.6, 137.8, 136.6, 134.9, 133.2, 130.7, 129.6, 128.4, 127.7, 127.2, 126.5, 126.2, 125.1, 120.7, 72.8, 38.6, 30.4, 29.6.

4.3.27. 2-((4-Nitrobenzylidene)amino)-N-(3-(4-oxo-3,4-dihydroquinazolin-2-yl)propanoyl)-3-phenylpropanehydrazide ***(28)***

Yield 84%, yellow solid, R_f^a = 0.58, R_f^b = 0.64, m.p. 201–202 °C, MS *m/z*, (M + 1): 513.2364, IR KBr (cm^{-1}): 1620, 1630, 1745, 2962, 3231, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ*: 11.32 (1H, s, Het-NH), 9.80 (1H, s, NH), 8.13–7.12 (13H, m, Ar-H), 7.85 (1H, s, −N═CH), 6.62–6.61 (1H, d, NH), 4.47–4.45 (1H, m, $^{\alpha}$CH), 3.27–3.26 (2H, t, $^{\beta}CH_2$), 2.80–2.78 (2H, t, CH_2), 2.62–2.61 (2H, t, CH_2); ^{13}C NMR (DMSO-d_6, 100 MHz) *δ*: 172.7, 170.4, 161.3, 156.4, 151.7, 148.4, 144.7, 137.9, 136.8, 133.4, 130.6, 128.4, 127.9, 127.8, 126.6, 126.3, 125.5, 124.6, 120.7, 73.1, 38.4, 30.4, 29.8.

4.3.28. 2-((4-Fluorobenzylidene)amino)-N-(3-(4-oxo-3,4-dihydroquinazolin-2-yl)propanoyl)-3-phenylpropanehydrazide ***(29)***

Yield 82%, brown solid, R_f^a = 0.51, R_f^b = 0.59, m.p. 161–163 °C, MS *m/z*, (M + 1): 486.3246, IR KBr (cm^{-1}): 1614, 1632, 1775, 2952, 3214, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ*: 11.28 (1H, s, Het-NH), 9.25 (1H, s, NH), 8.17–6.79 (14H, m, Ar-H& −N═CH), 5.98–5.91 (1H, d, NH), 4.04–3.99 (1H, m, $^{\alpha}$CH), 3.22–3.18 (2H, t, $^{\beta}CH_2$), 2.97–2.93 (2H, t, CH_2), 2.89–2.86 (2H, t, CH_2); ^{13}C NMR (DMSO-d_6, 100 MHz) *δ*: 172.6, 171.3, 167.7, 161.7, 157.9, 148.2, 144.8, 138.3, 134.9, 132.7, 129.5, 129.2, 128.5 127.7, 126.7, 126.2, 120.1, 121.4, 116.0, 72.7, 37.4, 31.7, 29.7.

4.3.29. 2-((3,4-Dihydroxybenzylidene)amino)-N-(3-(4-oxo-3,4-dihydroquinazolin-2-yl)propanoyl)-3-phenylpropanehydrazide ***(30)***

Yield 78%, white solid, R_f^a = 0.42, R_f^b = 0.47, m.p. 175–177 °C, MS *m/z*, (M + 1): 500.3216, IR KBr (cm^{-1}): 1614, 1633, 1785, 2941, 3262, 3561, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ*: 12.15 (1H, s, Het-NH), 11.33–11.15 (1H, d, NH), 9.20 (2H, s, 2OH), 8.42–6.73 (12H, m, Ar-H),

7.96 (1H, s, −N═CH), 6.75–6.73 (1H, d, NH), 4.54–4.49 (1H, m, $^{\alpha}$CH), 3.00–2.97 (2H, t, $^{\beta}CH_2$), 2.84–2.78 (2H, t, CH_2), 2.64–2.61 (2H, t, CH_2), ^{13}C NMR (DMSO-d_6, 100 MHz) δ: 172.3, 171.4, 161.2, 156.5, 150.4, 148.7, 147.3, 144.3, 137.8, 134.4, 133.2, 129.7, 128.3, 127.1, 129.6, 126.5, 125.4, 123.4, 120.6, 117.3, 116.0, 73.4, 37.6, 30.7, 28.6.

*4.3.30. 2-((4-Hydroxy-3-methoxybenzylidene)amino)-N-(3-(4-oxo-3,4-dihydroquinazolin-2-yl)propanoyl)-3-phenylpropanehydrazide **(31)***

Yield 83%, white solid, R_f^a = 0.42, R_f^b = 0.51, m.p. 177–178 °C, MS *m/z*, (M+1): 514.6521, IR KBr (cm^{-1}): 1606, 1632, 1745, 2956, 3269, 3561, ^{1}H NMR (DMSO-d_6, 500 MHz) δ: 11.18 (1H, s, Het-NH), 9.76 (1H, s, NH), 8.80 (1H, s, OH), 8.10–7.08 (12H, m, Ar-H), 7.89 (1H, s, −N═CH), 6.60–6.59 (1H, d, NH), 4.44–4.43 (1H, m, $^{\alpha}$CH), 3.78 (3H, s, OCH_3), 3.26–3.24 (2H, t, $^{\beta}CH_2$), 2.83–2.81 (2H, t, CH_2), 2.66–2.64 (2H, t, CH_2), ^{13}C NMR (DMSO-d_6, 500 MHz) δ: 172.6, 171.4, 161.6, 156.3, 151.4, 150.3, 148.6, 144.3, 137.8, 135.5, 133.6, 129.7, 128.1, 127.2, 126.6, 126.1, 125.4, 123.3, 120.4, 117.7, 112.9, 73.2, 52.7, 37.6, 30.4, 28.9.

*4.3.31. 2-(4-Hydroxy-3,5-dimethoxybenzylidene)amino)-N-(3-(4-oxo-3,4-dihydroquinazolin-2-yl)propanoyl)-3-phenylpropanehydrazide **(32)***

Yield 78%, white solid, R_f^a = 0.47, R_f^b = 0.55, m.p. 160–161 °C, MS *m/z*, (M+1): 544.3218, IR KBr (cm^{-1}): 1618, 1626, 1790, 2920, 3326, 3562, ^{1}H NMR (DMSO-d_6, 500 MHz) δ: 11.09 (1H, s, Het-NH), 9.89 (1H, s, NH), 8.80 (2H, s, 2OH), 8.11–7.05 (11*H*, m, Ar-H), 7.82 (1H, s, −N═CH), 6.62–6.61 (1H, d, NH), 4.43–4.42 (1H, m, $^{\alpha}$CH), 3.81 (6H, s, $2OCH_3$), 3.27–3.25 (2H, t, $^{\beta}CH_2$), 2.89–2.87 (2H, t, CH_2), 2.69–2.67 (2H, t, CH_2), ^{13}C NMR (DMSO-d_6, 500 MHz) δ: 172.8, 170.7, 161.3, 156.4, 149.7, 148.7, 144.0, 139.5, 137.3, 135.2, 134.3, 128.8, 127.9, 127.8, 126.9, 126.3, 125.7, 120.4, 104.7, 73.0, 52.8, 37.4, 30.6, 28.7.

*4.3.32. 3-(4-Oxo-3,4-dihydroquinazolin-2-yl)propanoyl)-3-phenyl-2-((3,4,5-trimethoxybenzylidene)amino)propanehydrazide **(33)***

Yield 87%, brown solid, R_f^a = 0.56, R_f^b = 0.67, m.p. 204–206 °C, MS *m/z*, (M+1): 558.6213, IR KBr (cm^{-1}): 1614, 1633, 1745, 2941, 3289, ^{1}H NMR (DMSO-d_6, 500 MHz) δ: 11.21 (1H, s, Het-NH), 9.90 (1H, s, NH), 8.08–7.15 (11*H*, m, Ar-H), 7.86 (1H, s, −N═CH), 6.60–6.58 (1H, d, NH), 4.40–4.38 (1H, m, $^{\alpha}$CH), 3.81 (9H, s, $3OCH_3$), 3.22–3.20 (2H, t, $^{\beta}CH_2$), 2.86–2.83 (2H, t, CH_2), 2.60–2.58 (2H, t, CH_2), ^{13}C NMR (DMSO-d_6, 100 MHz) δ: 172.3, 170.4, 162.0, 156.7, 154.3, 148.6, 143.4, 142.1, 137.3, 134.6, 133.5, 128.6, 127.8, 127.7, 126.9, 126.4, 125.9, 120.7, 104.7, 73.6, 60.9, 52.7, 37.7, 30.4, 28.6.

*4.3.33. 3-(4-Oxo-3,4-dihydroquinazolin-2-yl)propanoyl)-3-phenyl-2-((3,4,5-trihydroxybenzylidene)amino)propanehydrazide **(34)***

Yield 83%, white solid, R_f^a = 0.32, R_f^b = 0.37, m.p. 210–211 °C, MS *m/z*, (M+1): 516.2348, IR KBr (cm^{-1}): 1614, 1636, 1786, 2915, 3219, 3591, ^{1}H NMR (DMSO-d_6, 500 MHz) δ: 11.19 (1H, s, Het-NH), 9.92 (1H, s, NH), 9.30 (2H, s, 2OH), 8.47 (1H, s, OH), 8.08–6.60 (11*H*, m, Ar-H), 7.81 (1H, s, −N═CH), 6.67–6.66 (1H, d, NH), 4.40–4.39 (1H, m, $^{\alpha}$CH), 3.22–3.20 (2H, t, $^{\beta}CH_2$), 2.82–2.80 (2H, t, CH_2), 2.59–2.57 (2H, t, CH_2), ^{13}C NMR (DMSO-d_6, 100 MHz) δ ppm: 172.1, 171.6, 161.3, 156.7, 147.9, 146.4, 143.6, 139.2, 137.2, 135.4, 133.3, 129.1, 128.2, 127.4, 127.0, 126.4, 125.5, 120.4, 108.6, 73.5, 37.4, 30.6, 28.6.

*4.3.34. 2-(Benzylideneamino)-3-(1H-indol-3-yl)-N-(3-(4-oxo-3,4-dihydroquinazolin-2-yl)propanoyl)propanehydrazide **(35)***

Yield 80%, white solid, R_f^a = 0.57, R_f^b = 0.69, m.p. 192–193 °C, MS *m/z*, (M+1): 507.2348, IR KBr (cm^{-1}): 1610, 1629, 1774, 2950, 3226, 3310, ^{1}H NMR (DMSO-d_6, 500 MHz) δ: 11.31 (1H, s, Het-NH), 10.24 (1H, s, Indole-NH), 9.92 (1H, s, NH), 8.08–6.62 (14H, m, Ar-H), 7.87(1H, s, −N═CH), 6.60–6.59 (1H, d, NH), 4.46–4.45 (1H, m, $^{\alpha}$CH), 3.18–3.17 (2H, t, CH_2), 3.14–3.12 (2H, t, $^{\beta}CH_2$), 2.80–2.79 (2H, t, CH_2), ^{13}C NMR (DMSO-d_6, 100 MHz) δ: 173.3, 171.8, 162.1, 156.4, 147.9, 143.5, 138.8, 136.7, 133.5, 131.4, 129.8, 128.8, 127.4, 127.3, 126.6, 126.2, 123.4, 122.2, 121.7, 119.4, 115.6, 112.2, 111.7, 73.1, 34.4, 30.7, 28.6.

*4.3.35. 2-((4-Chlorobenzylidene)amino)-3-(1H-indol-3-yl)-N-(3-(4-oxo-3,4-dihydroquinazolin-2-yl)propanoyl)propanehydrazide **(36)***

Yield 79%, white solid, R_f^a = 0.56, R_f^b = 0.63, m.p. 190–191 °C, MS *m/z*, (M+1): 541.2364, IR KBr (cm^{-1}): 1609, 1626, 1756, 2945, 3246, 3312, ^{1}H NMR (DMSO-d_6, 500 MHz) δ: 11.19 (1H, s, Het-NH), 10.20 (1H, s, Indole-NH), 9.98(1H, s, NH), 8.10–6.80(13H, m, ArH), 7.80(1H, s, CH), 6.55–6.54 (1H, d, NH), 4.47–4.46 (1H, m, $^{\alpha}$CH), 3.14–3.13 (2H, t, CH_2), 3.10–3.08 (2H, t, $^{\beta}CH_2$), 2.82–2.80 (2H, t, CH_2); ^{13}C NMR (DMSO-d_6, 100 MHz) δ: 172.7, 171.4, 162.7, 156.3, 148.2, 143.3, 137.1, 136.5, 134.3, 133.6, 130.4, 129.1, 128.7, 127.4, 127.1, 126.4, 123.5, 122.8, 121.6, 119.3, 118.7, 112.4, 111.3, 72.8, 35.7, 30.6, 28.4.

*4.3.36. 2-((4-Nitrobenzylidene)amino)-3-(1H-indol-3-yl)-N-(3-(4-oxo-3,4-dihydroquinazolin-2-yl)propanoyl)propanehydrazide **(37)***

Yield 81%, yellow solid, R_f^a = 0.55, R_f^b = 0.61, m.p. 184–186 °C, MS *m/z*, (M+1): 552.6245, IR KBr (cm^{-1}): 1605, 1631, 1788, 2915, 3219, 3267, ^{1}H NMR (DMSO-d_6, 500 MHz) δ: 11.12 (1H, s, Het-NH), 10.26 (1H, s, Indole-NH), 9.56(1H, s, NH), 8.06–6.90 (13H, m, ArH), 7.88 (1H, s, CH), 6.62–6.61 (1H, d, NH), 4.43–4.42 (1H, m, $^{\alpha}$CH), 3.15–3.13 (2H, t, CH_2), 3.12–3.10 (2H, t, $^{\beta}CH_2$), 2.81–2.79 (2H, t, CH_2); ^{13}C NMR (DMSO-d_6, 500 MHz) δ: 172.1, 170.6, 161.3, 156.3, 151.7, 147.8, 144.3, 139.7, 137.6, 133.2, 130.1, 127.8, 127.4, 126.6, 126.2, 124.7, 123.6, 122.0, 121.8, 119.2, 118.4, 112.7, 111.4, 73.6, 34.8, 30.6, 28.1.

*4.3.37. 2-((4-Fluorobenzylidene)amino)-3-(1H-indol-3-yl)-N-(3-(4-oxo-3,4-dihydroquinazolin-2-yl)propanoyl)propanehydrazide **(38)***

Yield 77%, brown solid, R_f^a = 0.46, R_f^b = 0.54, m.p. 157–158 °C, MS *m/z*, (M+1): 525.1648, IR KBr (cm^{-1}): 1610, 1636, 1788, 2945, 3266, ^{1}H NMR (DMSO-d_6, 500 MHz) δ: 11.21 (1H, s, Het-NH), 10.35 (1H, s, Indole-NH), 9.59(1H, s, NH), 8.10–6.96 (13H, m, ArH), 7.89 (1H, s, CH), 6.60–6.59 (1H, d, NH), 4.42–4.40 (1H, m, $^{\alpha}$CH), 3.14–3.12 (2H, t, CH_2), 3.08–3.07 (2H, t, $^{\beta}CH_2$), 2.86–2.84 (2H, t, CH_2); ^{13}C NMR (DMSO-d_6, 100 MHz) δ ppm: 171.7, 170.6, 166.6, 162.3, 156.3, 147.9, 144.5, 137.7, 133.5, 132.4, 130.6, 128.7, 127.1, 126.8, 126.4, 123.6, 122.2, 120.4, 119.6, 118.4, 115.6, 112.8, 111.3, 73.6, 34.5, 30.8, 28.4.

*4.3.38. 2-((3,4-Dihydroxybenzylidene)amino)-3-(1H-indol-3-yl)-N-(3-(4-oxo-3,4-dihydroquinazolin-2-yl)propanoyl)propanehydrazide **(39)***

Yield 83%, white solid, R_f^a = 0.42, R_f^b = 0.49, m.p. 182–184 °C, MS *m/z*, (M+1): 539.6231, IR KBr (cm^{-1}): 1616, 1639, 1785, 2962, 3226, 3542, ^{1}H NMR (DMSO-d_6, 500 MHz) δ: 11.28 (1H, s, Het-NH), 10.10 (1H, s, Indole-NH), 9.80 (1H, s, NH), 8.40 (2H, s, 2OH), 8.10–6.70 (12H, m, ArH), 7.76 (1H, s, CH), 6.52–6.50 (1H, d, NH), 4.40–4.38 (1H, m, $^{\alpha}$CH), 3.19–3.18 (2H, t, CH_2), 3.10–3.08 (2H, t, $^{\beta}CH_2$), 2.85–2.83 (2H, t, CH_2), ^{13}C NMR (DMSO-d_6, 500 MHz) δ: 172.8, 171.7, 161.4, 156.3, 150.7, 149.5, 148.7, 143.3, 136.4, 135.4, 133.2, 128.7, 127.4, 126.5, 126.1, 124.1, 123.3, 122.6, 120.5, 119.4, 118.7, 117.4, 116.3, 112.1, 111.4, 73.2, 34.5, 30.7, 28.6.

*4.3.39. 2-((4-Hydroxy-3-methoxybenzylidene)amino)-3-(1H-indol-3-yl)-N-(3-(4-oxo-3,4-dihydroquinazolin-2-yl)propanoyl)propanehydrazide **(40)***

Yield 81%, white solid, R_f^a = 0.43, R_f^b = 0.50, m.p. 179–181 °C, MS *m/z*, (M+1): 553.4268, IR KBr (cm^{-1}): 1609, 1616, 1784, 2940, 3246, 3564, ^{1}H NMR (DMSO-d_6, 500 MHz) δ: 11.24 (1H, s, Het-NH), 10.28 (1H, s, Indole-NH), 9.91 (1H, s, NH), 8.2 (1H, s, OH), 8.06–6.86 (12H, m, ArH), 7.80 (1H, s, CH), 6.70–6.68 (1H, d, NH), 4.45–4.44 (1H, m, $^{\alpha}$CH), 3.77 (3H, s, OCH3), 3.20–3.19 (2H, t, CH_2), 3.07–3.05 (2H, t, $^{\beta}CH_2$), 2.68–2.65 (2H, t, CH_2), ^{13}C NMR (DMSO-d_6, 500 MHz) δ: 172.7, 171.4, 161.6, 156.3, 149.6, 148.3, 144.6, 137.7, 134.5, 133.3, 128.7, 127.4, 126.9, 126.8, 123.5, 123.0, 122.4, 121.8, 119.7, 118.4, 117.1, 112.6, 111.4, 111.2, 73.6, 52.4, 33.8, 30.5, 28.6.

*4.3.40. 2-((4-Hydroxy-3,5-dimethoxybenzylidene)amino)-3-(1H-indol-3-yl)-N-(3-(4-oxo-3,4-dihydroquinazolin-2-yl)propanoyl)propanehydrazide (**41**)*

Yield 86%, white solid, R_f^a = 0.43, R_f^b = 0.51, m.p. 193–195 °C, MS *m/z*, (M + 1): 583.1264, IR KBr (cm^{-1}): 1609, 1639, 1785, 2936, 3226, 3312, 3549, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ*: 11.22 (1H, s, Het-NH), 10.33 (1H, s, Indole-NH), 9.82 (1H, s, NH), 8.20 (1H, s, OH), 8.06–6.80 (11*H*, m, ArH), 7.82 (1H, s, CH), 6.68–6.67 (1H, d, NH), 4.44–4.43 (1H, m, $^{\alpha}$CH), 3.77 (6H, s, 2OCH_3), 3.16–3.14 (2H, t, CH_2), 3.05–3.03 (2H, t, $^{\beta}CH_2$), 2.70–2.68 (2H, t, CH_2), ^{13}C NMR (DMSO-d_6, 100 MHz) *δ*: 172.7, 171.7, 161.8, 156.3, 149.5, 148.6, 144.5, 140.7, 137.8, 135.6, 133.5, 128.6, 127.1, 126.9, 126.5, 123.9, 122.8, 121.7, 119.5, 118.7, 112.1, 111.7, 104.8, 73.6, 52.8, 35.1, 30.8, 28.7.

*4.3.41. 3-(1H-indol-3-yl)-N-(3-(4-oxo-3,4-dihydroquinazolin-2-yl) propanoyl)-2-((3,4,5-trimethoxybenzylidene)amino)propanehydrazide (**42**)*

Yield 83%, R_f^a = 0.58, R_f^b = 0.64, m.p. 176–177 °C, IR KBr (cm^{-1}): MS *m/z*: 597.1258, IR KBr (cm^{-1}): 1614, 1628, 1783, 2942, 3226, 3312, ^{1}H NMR (DMSO-d_6) *δ* ppm: 11.20 (1H, s, Het-NH), 10.18 (1H, s, Indole-NH), 9.90 (1H, s, NH), 8.10–6.96 (11*H*, m, ArH), 7.88 (1H, s, CH), 6.60–6.58 (1H, d, NH), 4.40–4.38 (1H, m, $^{\alpha}$CH), 3.81 (9H, s, 3OCH_3), 3.19–3.17 (2H, t, CH_2), 3.10–3.09 (2H, t, $^{\beta}CH_2$), 2.71–2.69 (2H, t, CH_2), ^{13}C NMR (DMSO-d_6, 100 MHz) *δ*: 173.6, 170.5, 161.4, 156.7, 152.4, 148.4, 143.3, 142.7, 136.4, 135.1, 133.4, 128.2, 127.5, 126.8, 126.4, 123.4, 121.3, 120.9, 119.4, 118.0, 112.4, 111.7, 104.4, 75.3, 60.8, 56.7, 37.2, 30.6, 28.6.

*4.3.42. 3-(1H-indol-3-yl)-N-(3-(4-oxo-3,4-dihydroquinazolin-2-yl) propanoyl)-2-((3,4,5-trihydroxybenzylidene)amino)propanehydrazide (**43**)*

Yield 80%, brown solid, R_f^a = 0.35, R_f^b = 0.39, m.p. 177–179 °C, MS *m/z*, (M + 1): 555.3648, IR KBr (cm^{-1}): 1606, 1636, 1781, 2943, 3214, 3526, ^{1}H NMR (DMSO-d_6, 500 MHz) *δ*: 11.25 (1H, s, Het-NH), 10.18 (1H, s, Indole-NH), 9.90 (1H, s, NH), 8.59 (2H, s, 2OH), 8.40 (1H, s, OH), 8.11–6.90 (11*H*, m, ArH), 7.86 (1H, s, CH), 6.61–6.59 (1H, d, NH), 4.46–4.45 (1H, m, $^{\alpha}$CH), 3.17–3.16 (2H, t, CH_2), 3.11–3.10 (2H, t, $^{\beta}CH_2$), 2.70–2.69 (2H, t, CH_2), ^{13}C NMR (DMSO-d_6, 100 MHz) *δ*: 172.5, 170.6, 161.8, 156.4, 148.7, 146.3, 144.3, 138.4, 135.3, 134.7, 133.5, 128.3, 127.4, 126.9, 126.1, 123.4, 121.4, 120.5, 119.7, 118.5, 111.7, 110.9, 109.8, 75.6, 37.6, 30.4, 28.6.

4.4. Biological assays

4.4.1. In vitro antimicrobial assay

4.4.1.1. Antibacterial assay. In vitro antibacterial activity was evaluated against human pathogens of Gram negative organisms namely *L. basillus* and Gram positive bacteria namely *E. coli* by agar well diffusion method [51]. The microorganisms were inoculated in to the sterilized nutrient broth and maintained at 37 °C for 24 h. On the day of testing, bacteria were subcultured separately into 25 mL of sterilized nutrient broth. Inoculated subcultured broths were kept at room temperature for the growth of inoculums. Each test compounds (**1–43**) and standard drug (Streptomycin) of 10 mg was dissolved in 10 mL of DMSO to get a concentration of 1 mg/mL and further diluted to get a final concentration of 30 μg/mL. About 15–20 mL of molten nutrient agar was poured into each of the sterile plates. With the help of cork borer of 6 mm diameter, the cups were punched and scooped out of the set agar and the plates were inoculated with the suspension of particular organism by spread plate technique. The cups of inoculated plates were then filled with 0.1 mL of the test solution, streptomycin solution and DMSO (negative control). The plates were allowed to stay for 24 h at 37 °C and zone of inhibition (mm) was then measured.

4.4.1.2. Antifungal assay. In vitro antifungal activity was evaluated against two fungal species namely *A. niger* and *F. oxysporum* by agar well diffusion method [52]. The fungal strains were subcultured separately into 25 mL of sterilized nutrient broth and incubated for one day to obtain the inoculums. Each test compounds (**1–43**) and standard drug (Bavistin) of 10 mg was dissolved in 10 mL of DMSO to get a concentration of 1 mg/mL and further diluted to get a final concentration of 30 μg/mL. Molten media of Sabouraud agar of 10–15 mL was poured into the petri plates and allowed to solidify. Fungal subculture was inoculated on the solidified media. With the help of 6 mm cork borer, the cups were punched and scooped out of the set agar. The cups of inoculated plates were then filled with 0.1 mL of the test solution, bavistin solution and DMSO (negative control). The plates were allowed to stay for 3 days at room temperature and zone of inhibition (mm) was then measured.

5. Molecular docking studies

Maestro 9.3.5 version of the Schrodinger software suite, 2011 was used to obtain binding interaction of molecules with targeted site. The 3D crystallographic structure of proteins (PDB ID: 2VF5) was retrieved from Protein Data Bank (www.rcsb.org/pdb). The lowest energy states of ligand were achieved using LigPrep program and it was optimized by force field OPLS-2005 (Optimized Potential for Liquid Simulations). The protein structures were pre-processed, modified and refined by Protein Preparation Wizard. Further, it was minimized by OPLS-2005 force field. The protein and ligand interaction performed by generation of receptor gridin the target site of protein by GLIDE. Depending on the extent of docking, the scores were produced (docking score) which will determine the best fitted ligand to target protein [50].

Acknowledgements

We gratefully acknowledge UGC for the award of UGC-BSR fellowship to DCG, Centre with Potential for Excellence in a Particular Area (CPEPA), University with Potential of Excellence (UPE) and Department of Science and Technology-Promotion of University Research and Scientific Excellence (DST-PURSE), UGC, New Delhi, India for the financial assistance.

Appendix A. Supplementary material

Supplementary data to this article can be found online at https://doi.org/10.1016/j.bioorg.2019.103093.

References:

[1] G.F. Zha, J. Leng, N. Darshini, T. Shubhavathi, H.K. Vivek, A.M. Asiri, H.M. Marwani, K.P. Rakesh, N. Mallesha, H.L. Qin, Bioorg. Med. Chem. Lett. 27 (2017) 3148–3155.
[2] K.P. Rakesh, H.K. Vivek, H.M. Manukumar, C.S. Shantharam, S.N.A. Bukhari, H.L. Qin, M.B. Sridhara, RSC Adv. 8 (2018) 5473–5483.
[3] K. Bush, P. Courvalin, G. Dantas, J. Davies, B. Eisenstein, P. Huovinen, et al., Nat. Rev. Microbiol. 9 (2011) 894–896.
[4] S.B. Levy, B. Marshall, Nat. Med. 10 (2004) S122–S129.
[5] C. Ghosh, J. Haldar, Chem. Med. Chem. 10 (2015) 1606–1624.
[6] C. Li, M.B. Sridhara, K.P. Rakesh, H.K. Vivek, H.M. Manukumar, C.S. Shantharam, H.L. Qin, Bioorg. Chem. 81 (2018) 389–395.
[7] L. Ravindar, S.N.A. Bukhari, K.P. Rakesh, H.M. Manukumar, H.K. Vivek, N. Mallesha, Z.Z. Xie, H.L. Qin, Bioorg. Chem. 81 (2018) 107–118.
[8] S.M. Lim, S.A.R. Webb, Anaesthesia 60 (2005) 887–902.
[9] H.W. Boucher, G.H. Talbot, D.K. Benjamin, J. Bradley, R.J. Guidos, R.N. Jones, et al., Clin. Infect. Dis. 56 (2013) 1685.
[10] H. Grundmann, M. Aires-de-Sousa, J. Boyce, E. Tiemersma, Lancet 368 (2006) 874–885.
[11] Y.H.E. Mohammed, H.M. Manukumar, K.P. Rakesh, C.S. Karthik, P. Mallu, H.L. Qin, Microbial Pathogenesis 123 (2018) 339–347.
[12] X. Zhang, H.M. Manukumar, K.P. Rakesh, C.S. Karthik, H.S. Nagendra Prasad, S. Nanjunda Swamy, et al., Microbial Pathogenesis 123 (2018) 275–284.
[13] X. Zhang, H.M. Manukumar, K.P. Rakesh, H.L. Qin, Microbiol. Res. 212–213 (2018) 59–66.
[14] K.P. Rakesh, H.M. Manukumar, S. Srivastava, Xing Chen, Sihui Long, C.S. Karthik, P. Mallu, H.L. Qin, ACS Comb. Sci. 20 (2018) 681–693.
[15] H.M. Manukumar, B. Chandrasekhar, K.P. Rakesh, A.P. Ananda, M. Nandhini,

P. Lalitha, S. Sumathi, H.L. Qin, S. Umesha, Med. Chem. Commun. 8 (2017) 2181–2194.
[16] G.F. Zha, S.M. Wang, K.P. Rakesh, S.N.A. Bukhari, H.M. Manukumar, H.K. Vivek, N. Mallesha, H.L. Qin, Eur. J. Med. Chem. 162 (2019) 364–377.
[17] M.A. Fischbach, C.T. Walsh, Science 325 (2009) 1089–1093.
[18] D.M. Livermore, J. Antimicrob. Chemother. 66 (2011) 1941–1944.
[19] A.D.W. Boran, R. Iyengar, Curr. Opin. Discovery Devel. 13 (2010) 297–309.
[20] K.P. Rakesh, R. Suhas, H.M. Manukumar, S. Chanda, D.C. Gowda, Eur. J. Chem. 6 (2015) 254–260.
[21] M. Dinari, F. Gharahi, P. Asadi, J. Mole. Struct. 1156 (2018) 43–50.
[22] G. Khodarahmi, P. Asadi, F. Hassanzadeh, E. Khodarahmi, J. Res. Med. Sci. 20 (2015) 1094–1104.
[23] P. Asadi, G. Khodarahmi, A. Jahanian-Najafabadi, L. Saghaie, F. Hassanzadeh, Iran. J. Basic Med. Sci. 20 (2017) 975–989.
[24] K.P. Rakesh, H.M. Manukumar, D.C. Gowda, Bioorg. Med. Chem. Lett. 25 (2015) 1072–1077.
[25] K.P. Rakesh, H.K. Kumara, H.M. Manukumar, D.C. Gowda, Bioorg. Chem. 87 (2019) 252–264.
[26] M.A. Aziza, M.K. Ibrahim, A.G. El-Helpy, Al-Azhar, J. Pharm. Sci. 14 (1994) 193–201.
[27] K.P. Rakesh, C.S. Shantharam, H.M. Manukumar, Bioorg. Chem. 68 (2016) 1–8.
[28] K. Terashima, H. Shimamura, A. Kawase, Y. Tanaka, T. Tanimura, T. Kamisaki, et al., Chem. Pharm. Bull. 43 (1995) 2021–2023.
[29] A. Gursoy, N. Karali, Farmaco. 50 (1995) 857–866.
[30] D.J. Baek, Y.K. Park, H.L. Heo, M.H. Lee, Z.Y. Yang, M.H. Chio, Bioorg. Med. Chem. Lett. 8 (1998) 3287–3290.
[31] R.J. Griffin, S. Srinivasan, K. Bowman, A.H. Calvert, N.J. Curtin, D.R. Neweli, L.C. Pemberton, B.T. Golding, J. Med. Chem. 41 (1998) 5247–5256.
[32] P. Asadi, G. Khodarahmi, H. Farrokhpour, F. Hassanzadeh, L. Saghaei, Res. Pharm. Sci. 12 (2017) 233–240.
[33] G. Khodarahmi, P. Asadi, H. Farrokhpour, F. Hassanzadeh, M. Dinari, RSC Adv. 5 (2015) 58055–58064.
[34] G. Razafimamonjison, J.M.L. Pock Tsy, M. Randriamiarinarivo, P. Ramanoelina, J. Rasoarahona, F. Fawbush, P. Danthu, Chem. Biodivers. 14 (2017) e1600411.
[35] H.M. Revankar, S.N.A. Bukhari, G.B. Kumar, H.L. Qin, Bioorg. Chem 71 (2017) 146–159.
[36] K.P. Rakesh, R. Suhas, D.C. Gowda, Inter. J. Pep. Res. Ther. 25 (2018) 227–234.
[37] K.P. Rakesh, R. Suhas, J. Shivakumar, D. ChanneGowda, Russian J, Bioorg. Chem. 44 (2018) 158–164.
[38] S.M. Wang, C. Zhao, X. Zhang, H.L. Qin, Org. Biomole. Chem. 17 (2019) 4087–4101.
[39] M. Wang, K.P. Rakesh, J. Leng, W.Y. Fang, L. Ravindar, D.C. Gowda, H.L. Qin, Bioorg. Chem. 76 (2018) 113–129.
[40] Xing Chen, Jing Leng, K.P. Rakesh, N. Darshini, T. Shubhavathi, H.K. Vivek, N. Mallesha, Hua-Li Qin, Chem. Comm. 8 (2017) 1706–1719.
[41] T.R. Gadek, J.B. Nicholas, Biochem. Phar. Macol. 65 (2003) 1–8.
[42] B.M. Santhosh, P.A. Narasimha, J. Org. Chem. 66 (2001) 9040.
[43] A. Wohlrab, R. Lamer, M.S. VanNieuwenhze, J. Am. Chem. Soc. 129 (2007) 4177.
[44] J. Fournier, C. Bruneau, H. Dixneuf, S. Lécolier, J. Org. Chem. 56 (1991) 4458.
[45] H.K. Kumara, R. Suhas, D.M. SuyogaVardhan, M. Shobha, D.C. Gowda, RSC Adv. 8 (2018) 10644–10653.
[46] T. Izumiya, T. Kato, H. Aoyagi, M. Waki, M. Kondo, Synthetic Aspects of Biologically Active Cyclic Peptides, Gramicidin S and Tyrocidines, Kodansha, Tokyo, 1979.
[47] H. Khandelia, Y.N. Kaznessis, J. Phys. Chem. 111 (2007) 242–250.
[48] S. Milewski, Biochim. Biophys. Acta 1597 (2002) 173–192.
[49] A. Teplyakov, G. Obmolova, M.A. Badet-denisot, B. Badet, Protein Sci. 8 (1999) 596–602.
[50] H.K. Kumara, R. Suhas, D.M. Suyoga Vardhan, J. Shiva Kumar, D.C. Gowda, Med. Chem. Res. 27 (2018) 1504–1516.
[51] C. Perez, M. Paul, P. Bazerque, Acta. Biol. Med. Exp. 15 (1990) 113–115.
[52] I. Singh, V.P. Singh, Phytomorphology 50 (2000) 151–157.