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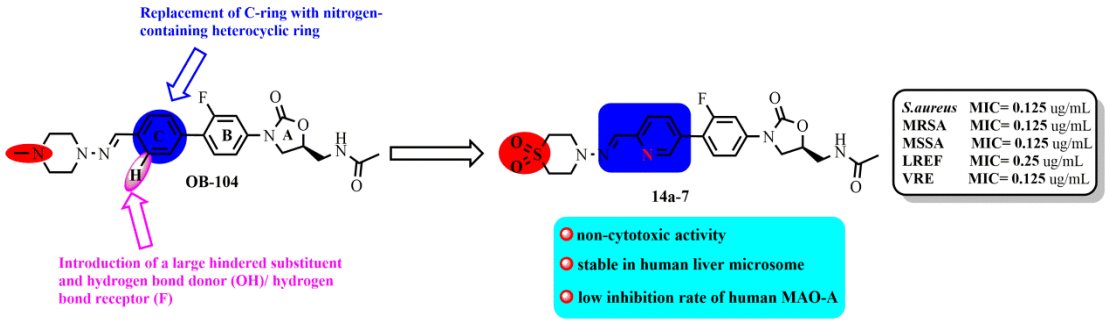

Optimization of biaryloxazolidinone as promising antibacterial agents against antibiotic-susceptible and antibiotic-resistant Gram-positive bacteria

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Abstract:

We previously discovered a series of novel biaryloxazolidinone analogues bearing a hydrazone moiety with potent antibacterial activity. However, the most potent compound **OB-104** exhibited undesirable chemical and metabolic instability. Herein, novel biaryloxazolidinone analogues were designed and synthesized to improve the chemical and metabolic stability. Compounds **6a-1**, **6a-3**, **14a-1**, **14a-3** and **14a-7** showed significant antibacterial activity against the tested Gram-positive bacteria as compared to radezolid and linezolid. Further studies indicated that most of them exhibited improved water solubility and chemical stability. Compound **14a-7** had MIC values of 0.125-0.25 μg/mL against all tested Gram-positive bacteria, and showed excellent antibacterial activity against clinical isolates of antibiotic-susceptible and antibiotic-resistant bacteria. Moreover, it was stable in human liver microsome. From a safety viewpoint, it showed non-cytotoxic activity against hepatic cell and exhibited lower inhibitory activity against human MAO-A compared to linezolid. The potent antibacterial activity and all these improved drug-likeness properties and safety profile suggested that compound **14a-7** might be a promising drug candidate for further investigation.

Keywords: structural optimization; biaryloxazolidinone; antibiotic-resistant infections; Gram-positive bacteria

1. Introduction

The emergence and outbreak of methicillin-resistant *Staphylococcus aureus* (MRSA), vancomycin-resistant *Enterococci* (VRE) and penicillin-resistant *Streptococcus pneumoniae* (PRSP) made antibiotic-resistant infections a critical healthcare concern worldwide [1-3]. Antibiotic-resistant infections resulted in increasing patient mortality and social economic burden. In the United States, there were approximately 19000 people dying each year from MRSA, which is known as the first superbug [4-5]. Finding effective ways to resolve antibiotic-resistant crisis is an urgent task. Karen B *et al.* summarized the approaches for resolving this global health crisis, of which, finding antibiotics with new structural classes or mechanisms was the main concern for drug chemists [6]. Due to developing antibiotics with new mechanisms is a long-term work, we focus our efforts on modifications of old antibiotics with known mechanisms to overcome drug resistance.

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The ribosomes synthesize proteins and the bacterial ribosome is one of the main targets of antibiotics [7]. Many natural, semi-synthetic and synthetic antibiotics bind to bacterial ribosome to inhibit bacterial protein synthesis. In recent years, important progresses on the crystal structures of antibiotics binding to ribosome have been reported, which give drug chemists insight into the mechanism of action of ribosome-targeting antibiotics. And these progresses are crucial for drug chemists to improve the potency and overcome bacterial resistance by further drug optimization [8-9]. Linezolid, the first oxazolidinone drug, was approved in 2000 by FDA for the treatment of antibiotic-resistant Gram-positive infections, including MRSA, PRSP, and VRE [10-11]. It binds to 23S RNA of the 50S ribosomal subunit to inhibit bacterial protein biosynthesis, which belongs to a ribosome-targeting antibiotic [12]. However, bacterial resistance to linezolid has been discovered due to the overuse of linezolid. Resistance to linezolid was mediated by a number of mechanisms, such as alteration of microbial permeability and target mutation [7, 13].

The C-ring of linezolid and many other ribosome-targeting antibiotics, such as chloramphenicol and clindamycin, share an overlapping region on the binding sites of ribosomal RNA, which inspired many research groups to develop oxazolidinone derivatives with C-ring and D-ring to overcome bacterial resistance and to improve the potency [7, 14-15]. Radezolid was developed based on the crystal structures of linezolid binding to ribosomal RNA and it exhibited more potent activity against antibiotic-resistant Gram-positive bacteria than linezolid [16-17]. As reported, oxazolidinone derivatives containing biaryl scaffold showed increased antibacterial potency against Gram-positive bacteria, especially antibiotic-resistant Gram-positive bacteria compared with linezolid [18-22]. These compounds revealed outstanding antibacterial activity by forming π–π stacking or hydrogen bond interactions with the 50S subunit.

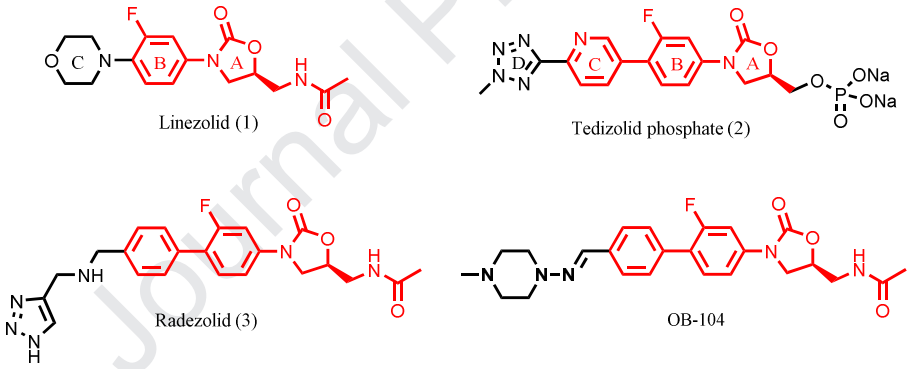


Figure 1. Structures of linezolid and related oxazolidinone analogues.

In our previous work, we discovered compound **OB-104**, a biaryloxazolidinone derivative bearing a hydrazone moiety, showed excellent antibacterial activity against five tested Gram-positive bacteria in *vitro*, which were a 15- to 30-fold increase compared to linezolid in activity against linezolid-resistant *Enterococcus faecalis* [23]. However, compound **OB-104** was unstable under simulated gastric acid condition (pH=1.2) and showed short half-life time (72.85 min) in human liver microsome. Further pharmacokinetic study in rats by intragastric administration revealed that the half-life time ($T_{1/2}$) of **OB-104** was 2.7 h. As shown in **Figure 2**, the metabolites of **OB-104** in rats were further studied by UPLC-QTOF/MS method. Four main metabolites were found in plasma of rats given intragastric administration of **OB-104**. The main metabolite **M-1** was confirmed by MS spectra and the reference substance, which was formed by the hydroxylation of C-ring at C-3 position. At the same time, nitrogen demethylation occurred on the piperazine ring to form metabolite **M-2**. The C=N bond of the hydrazone moiety of **OB-104** was hydrolyzed to the aldehyde metabolite **M-3**, which was further oxidized to the carboxyl metabolite **M-4**. Metabolite **M-1** with hydroxyl group at C-ring attracted our interest, and it was synthesized and its antibacterial activity was evaluated. Remarkably, **M-1** showed significant antibacterial activity against the tested Gram-positive bacteria (data are shown in **Table 1**). As we know, the electronic or sterically hindered effects of

substituents in C-ring may have a large effect on the stability of hydrazone bond. Basing on the structure of **M-1** and other reported biaryloxazolidinone analogues [24-25], we focused on the structural optimization of compound **OB-104** in following strategies, which are shown in **Figure 3**: (I) introduction of a sterically hindered substituent or hydrogen bond donor (OH)/ hydrogen bond receptor (F) at the *ortho*-position of the hydrazone bond on the C-ring (compounds **6a-6e** and **16**). (□) replacement of C-ring with nitrogen-containing heterocyclic ring (compounds **14a-14b** and **15a**), of which, the electron-withdrawing effect of nitrogen atom was expected to improve the chemical stability of hydrazone bond [21].

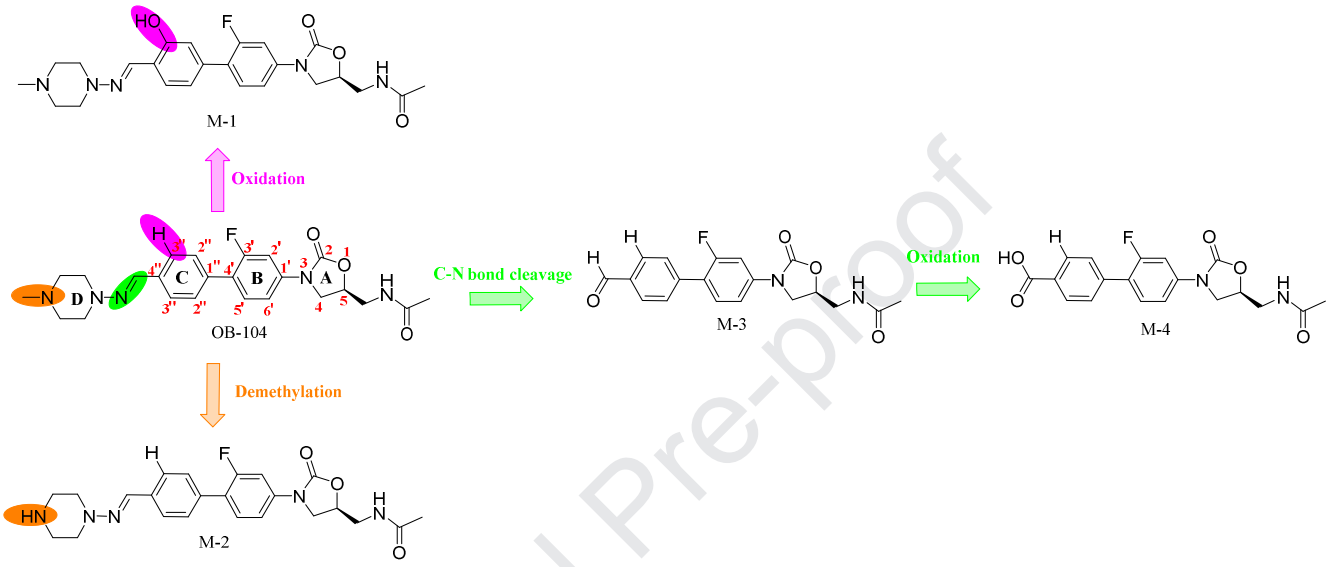


Figure 2. Predicted metabolic pathways of compound **OB-104**.

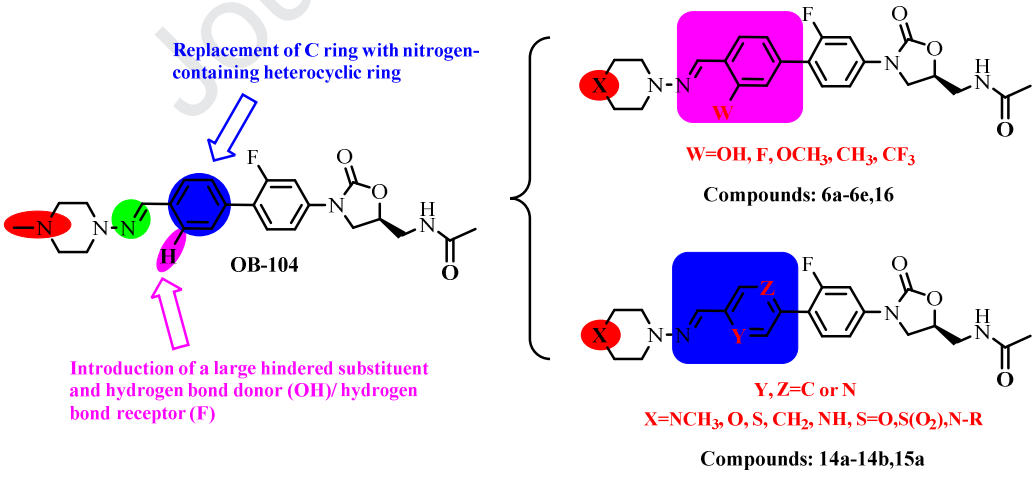
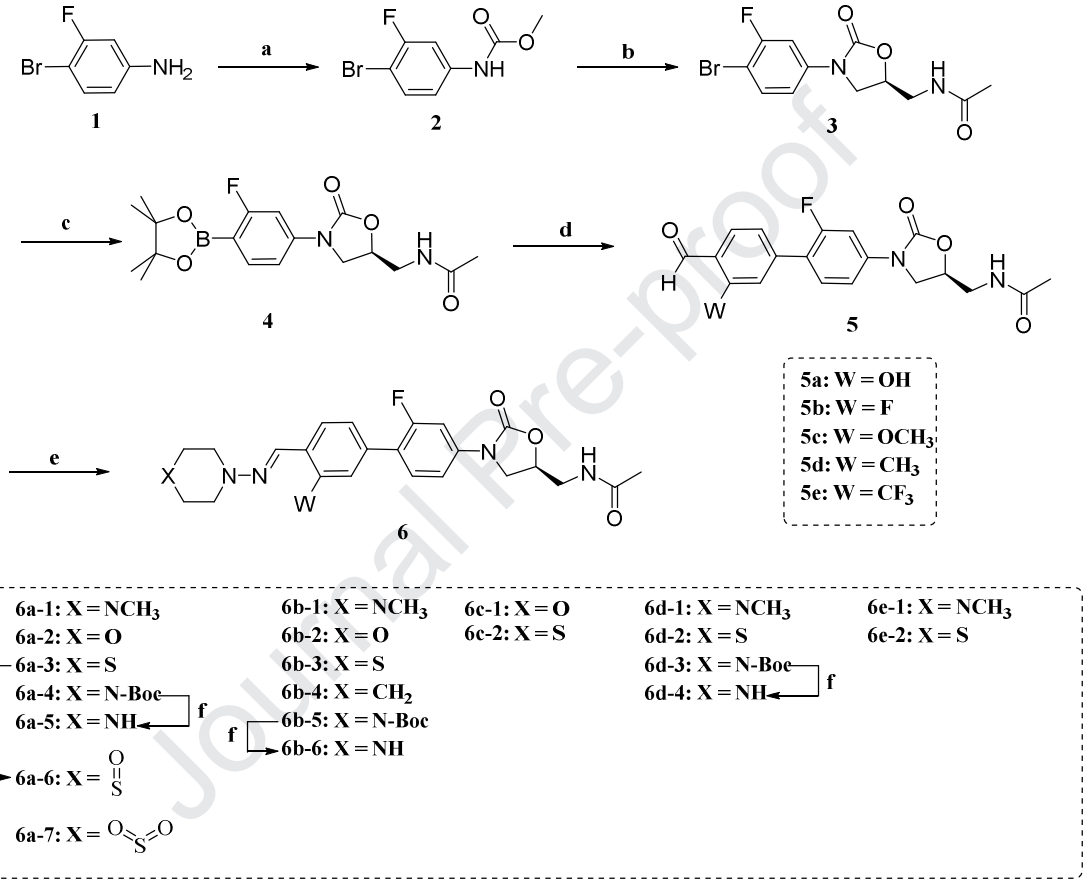


Figure 3. Design strategies for novel biaryloxazolidinone compounds.

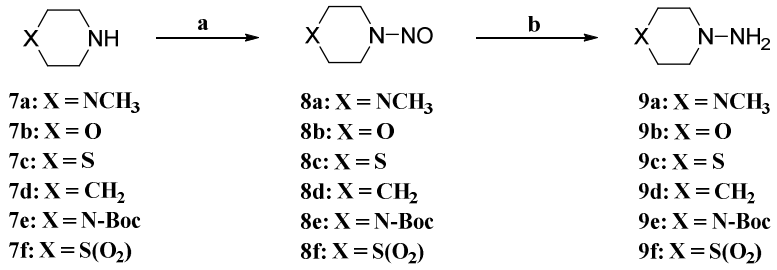
2. Chemistry

The synthesis of target compounds **6** is depicted in **Scheme 1**. 4-Bromo-3-fluoroaniline was acylated with methyl chloroformate to give **2** with a high yield. Intermediate **3** was obtained by cyclization reaction of intermediate **2** with (*S*)-1-((acetylamino)methyl)-2-chloroethyl acetate. Intermediate **4** was prepared by

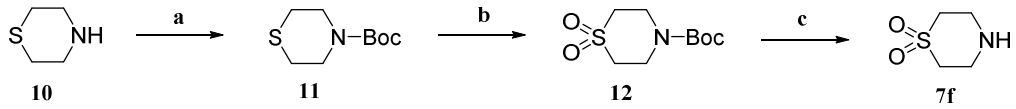
Miyaura-Ishiyama-Hartwig borylation reaction of intermediate **3** with bis(pinacolato)diboron, which was subjected to substituted *p*-bromobenzaldehydes to give the corresponding biaryloxazolidinones **5** by Suzuki coupling. The intermediates **5** were reacted with **9a-9f** in the presence of acetic acid to yield the corresponding target compounds **6** with high yields. The preparation of intermediates **9a-9f** was summarized in **Scheme 2** and **3**, as reported in our previous work [23]. In particular, intermediate **7f** was synthesized through a three-step process from thiomorpholine. Moreover, compound **6a-5** was finally prepared by deprotection of **6a-4** in DCM, and compounds **6b-6** and **6d-4** were prepared according to the same method. Compound **6a-3** was oxidized by 3-chloroperbenzoic acid in DCM to give compound **6a-6.**



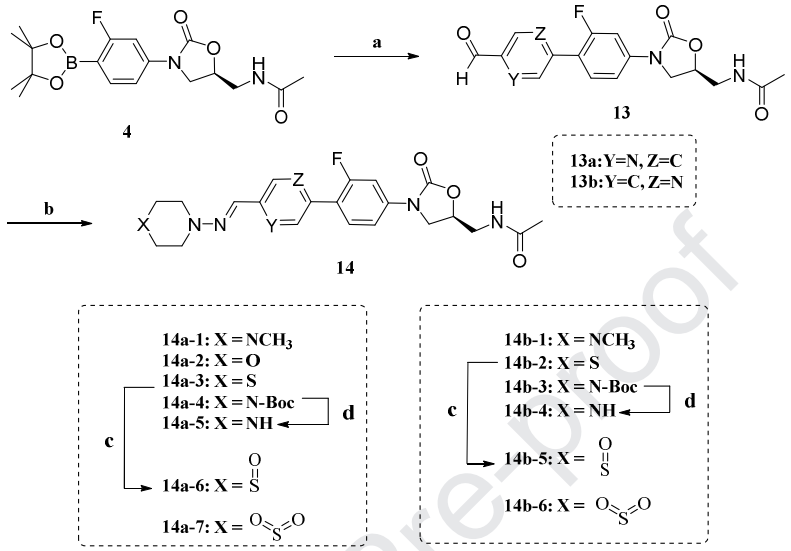
Scheme 1. Reagents and conditions: a: methyl chloroformate, pyridine, DCM, 25 °C, 2 h; b: (*S*)-1-((acetylamino)methyl)-2-chloroethyl acetate, *t*-BuOLi, DMF, CH_3OH, rt, 20 h; c: bis(pinacolato)diboron, $Pd(PPh_3)_2Cl_2$, potassium acetate, 1,4-dioxane, 90 °C, 20 h; d: substituted *p*-bromobenzaldehydes, $Pd(PPh_3)_2Cl_2$, K_2CO_3, DMF/ H_2O, 75 °C, 8-12 h; e: **9a-9f**, acetic acid, EtOH, 78 °C, 2 h. (f) TFA, DCM, 25 °C, 5 h; (g) *m*-CPBA, DCM, 25 °C, 4 h.



Scheme 2. Reagents and conditions: a: $NaNO_2$, HCl, THF/H_2O, 25 °C, 4 h; b: Zn, AcOH/H_2O, 0 °C, 5 h.

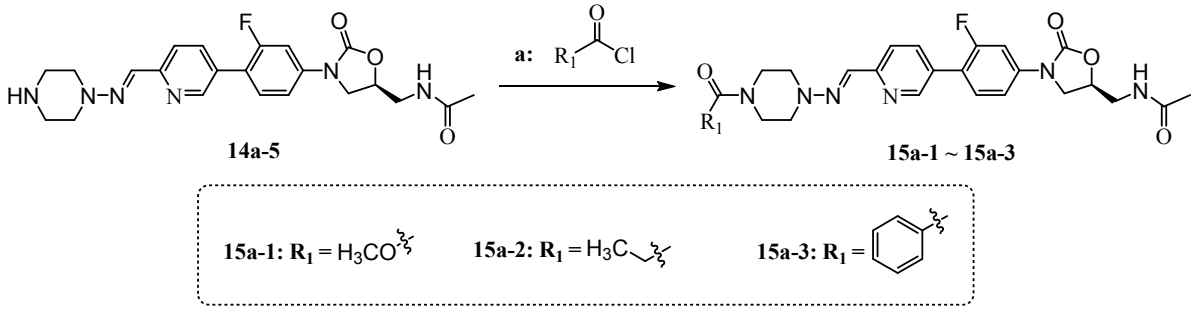


Scheme 3. Reagents and conditions: a: $(Boc)_2O$, TEA, THF, 25 ℃, 5 h; b: H_2O_2(40 %), TFA, AcOH, 50 ℃, 4 h; c: TFA, DCM, 25 ℃, 5 h.

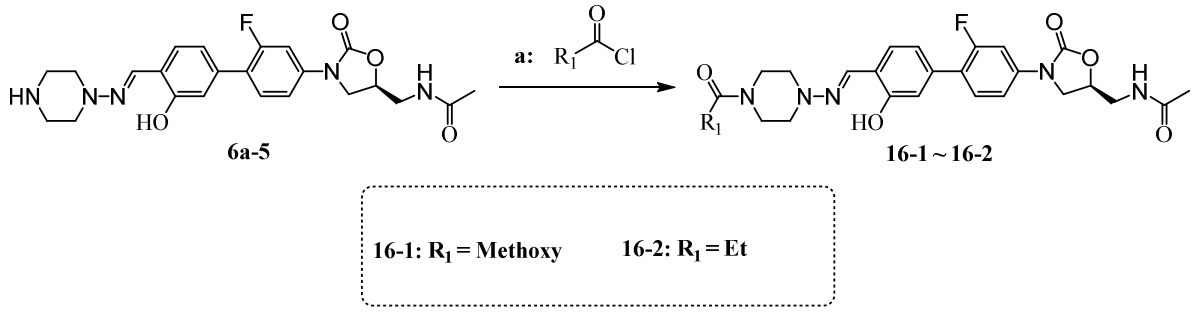


Scheme 4. Reagents and conditions: a: 3-bromo-6-pyridine carboxaldehyde or 6-bromo-3-pyridine carboxaldehyde, $Pd(PPh_3)_2Cl_2$, K_2CO_3, DMF/ H_2O, 75 ℃, 12 h; b: **9a-9f**, EtOH, 78 ℃, 2 h; c: *m*-CPBA, DCM, 25 ℃, 4 h; d: TFA, DCM, 25 ℃, 5 h.

The synthetic route of compounds **14** is depicted in **Scheme 4**. Compounds **14** were synthesized from intermediate **4** and 3-bromo-6-pyridine carboxaldehyde or 6-bromo-3-pyridine carboxaldehyde using the same method for the synthesis of compounds **6**. Moreover, Compounds **15** and **16** were prepared by acylation of **14a-5** and **6a-5** on the terminal piperazinyl ring with acid chloride using DIPEA as a base at room temperature, as shown in **Scheme 5** and **6**.



Scheme 5. Reagents and conditions: a: DIPEA, DMF, 25 ℃, 2 h.



Scheme 6. Reagents and conditions: a: DIPEA, DMF, 25 °C, 2 h.

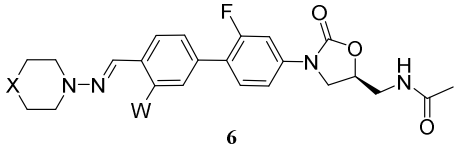
3. Results and discussion

3.1. Evaluation of in vitro antibacterial activity

All the target biaryloxazolidinone compounds were screened for their in *vitro* antibacterial activity against Gram-positive, antibiotic-susceptible and antibiotic-resistant bacteria strains (*S.aureus* ATCC29213, MRSA, MSSA, LREF and VRE), using linezolid and radezolid as positive control. The results of minimum inhibitory concentrations (MICs, μg/mL) determined by the broth liquid microdilution method were summarized in **Tables 1-2**.

As shown in **Table 1**, we firstly synthesized the putative metabolite **M-1** of **OB-104**, namely **6a-1**. It was worth noting that compound **6a-1** showed significantly potent antibacterial activity against five tested Gram-positive bacteria with MIC values of 0.125-0.25 μg/mL, which was more potent than that of linezolid and radezolid. Then a series of biaryloxazolidinone analogues with hydroxyl group (**6a-2** to **6a-7**) at C-ring were synthesized, and compound **6a-3** with terminal thiomorpholine group was the most potent compound, which had the MIC values of 0.125 μg/mL against five tested Gram-positive bacteria. Based on the high activity of compound **6a-3**, hydrogen bond receptor (F) and sterically hindered substituents (OCH_3, CH_3 and CF_3) were introduced into the C-ring at the putative metabolic site to improve the metabolic stability. We speculated that the C-ring substituent at the *ortho*-position of the hydrazone bond is related to the hydrolysis of hydrazone bond. However, most of this series of compounds showed weaker antibacterial activities than **6a-1** and **6a-3**. Compound **6b-3** with fluoro group exhibited 2-fold to 8-fold decrease in antibacterial activity compared to compound **6a-3** with hydroxyl group. Compounds with methoxy, trifluoromethyl or methyl groups respectively showed lower antibacterial activity compared to compounds bearing fluorine or hydroxyl, which indicated that steric hindrance of these substituents can reduce antibacterial activity.

Table 1. In *vitro* antibacterial activity of biaryloxazolidinone analogues.



6

Compounds	X	W	MICs, μg/mL				
			S.aureus[a]	MRSA[b]	MSSA[c]	LREF[d]	VRE[e]
6a-1	NCH_3	OH	0.125	0.125	0.125	0.25	0.25
6a-2	O	OH	0.25	0.25	0.25	0.25	0.25
6a-3	S	OH	0.125	0.125	0.125	0.125	0.125
6a-5	NH	OH	0.5	0.25	0.25	0.25	0.25

6a-6	S=O	OH	0.25	0.125	0.25	0.25	0.25
6a-7	$O=S=O$	OH	0.125	0.25	0.25	0.25	0.25
6b-1	NCH_3	F	1	1	1	1	1
6b-2	O	F	0.25	0.25	0.25	0.25	0.25
6b-3	S	F	0.25	0.25	0.25	1	1
6b-4	CH_2	F	0.25	0.25	0.25	0.5	0.5
6b-6	NH	F	0.5	0.5	0.5	0.5	0.5
6c-1	O	OCH_3	2	2	2	2	2
6c-2	S	OCH_3	2	1	1	1	1
6d-1	NCH_3	CH_3	0.5	0.5	0.5	0.5	0.5
6d-2	S	CH_3	2	1	1	2	1
6d-4	NH	CH_3	0.5	0.5	0.5	0.5	0.5
6e-1	NCH_3	CF_3	2	2	2	2	2
6e-2	S	CF_3	2	2	2	2	2
linezolid			1	1	1	>16	2
radezolid			0.5	0.5	0.5	0.5	0.5

[a] Standard *Staphylococcus aureus* (29213).

[b] Methicillin-resistant *Staphylococcus aureus*.

[c] Methicillin-sensitive *Staphylococcus aureus*.

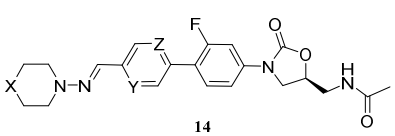
[d] Linezolid-resistant *Enterococcus faecalis*.

[e] Vancomycin-resistant *Enterococcus faecium*.

Compound **6a-3** exhibited MIC values of 0.125 μg/mL against all tested five Gram-positive bacteria, but it showed 2-fold decrease in antibacterial activity compared to **OB-104**, which indicated that the introduction of substituent at *ortho*-position of the hydrazone bond is actually unfavorable for the antibacterial activity. Due to the hydrazone moiety and biaryl scaffold were critical for antibacterial activity, then we explored the impact of C-ring on antibacterial activity by replacing of C-ring with nitrogen-containing heterocyclic ring. We speculated that the electron-withdrawing effect of the nitrogen atom will prevent oxidative metabolism and improve the chemical stability. As illustrated in **Table 2**, when the C-ring was replaced by pydrazin-3-yl group, compound **14a** showed more potent antibacterial activity than compound **14b**, of which, the C-ring was replaced by pyridine-2-yl group. It was worth noting that compounds **14a-1**, **14a-2**, **14a-3** and **14a-7** exhibited MIC values of 0.125-0.25 μg/mL against five tested Gram-positive bacteria, which was 2 to 4-fold higher than radezolid.

Compounds **6a-1** and **14a-1** showed more potent antibacterial activity compared to compounds **6a-5** and **14a-5** respectively, which indicated that substituents on the terminal piperazinyl ring could enhance the antibacterial activity. Then we conducted further structure-activity relationship studies on the substituent of terminal piperazinyl ring (**6a-5** and **14a-5**). Acyl groups were introduced to improve metabolic stability, thus avoiding demethylation. As illustrated in **Table 2**, compound **15a-3** with substituent of benzoyl group exhibited reduced antibacterial activity, while compounds with small substituents showed more potent antibacterial activity compared to compound **15a-3**. Compound **16-2** exhibited 2-fold decrease in antibacterial activity against *S.aureus*, MRSA and MSSA compared to **6a-1**, which indicated that introduction of large steric hindrance acyl group decreased antibacterial activity. Compounds **6a** and **14a** showed significant antibacterial activity and were screened for their antibacterial activity against clinical isolates of Gram-positive bacteria.

Table 2. In *vitro* antibacterial activity of biaryloxazolidinones.



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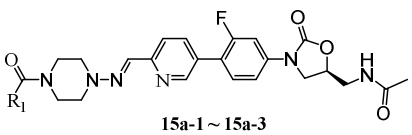
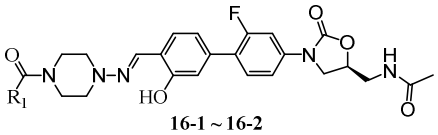


15a-1 ~ 15a-3



16-1 ~ 16-2

Compounds	X/R_1	MICs, μg/mL				
		S.aureus[a]	MRSA[b]	MSSA[c]	LREF[d]	VRE[e]
14a-1	NCH_3	0.125	0.125	0.125	0.25	0.25
14a-2	O	0.125	0.125	0.25	0.25	0.25
14a-3	S	0.125	0.125	0.25	0.25	0.25
14a-5	NH	0.5	0.5	1	0.5	0.5
14a-6	S=O	0.25	0.25	0.5	0.5	0.5
14a-7		0.125	0.125	0.125	0.25	0.125
14b-1	NCH_3	2	1	2	2	2
14b-2	S	1	0.5	2	2	2
14b-4	NH	2	2	2	2	2
14b-5	S=O	2	1	2	2	2
14b-6		2	1	1	1	1
15a-1		1	1	2	1	0.5
15a-2		1	1	1	1	0.5
15a-3		1	1	2	2	1
16-1		0.5	0.5	0.5	0.5	0.5
16-2		0.25	0.25	0.25	0.25	0.25
linezolid		1	1	1	＞16	2
radezolid		0.5	0.5	0.5	0.5	0.5

[a] Standard *Staphylococcus aureus* (29213).

[b] Methicillin-resistant *Staphylococcus aureus*.

[c] Methicillin-sensitive *Staphylococcus aureus*.

[d] Linezolid-resistant *Enterococcus faecalis*.

[e] Vancomycin-resistant *Enterococcus faecium*.

3.2. Evaluation of inhibition of clinical isolates by potent compounds

We further evaluated antibacterial activity of our optimal compounds against four clinical isolates of MRSA (4 isolates), MSSA (4 isolates), LREF (4 isolates) and VRE (4 isolates). These four clinical isolates were antibiotic-susceptible or antibiotic-resistant bacteria, and strain LREF was resistant to linezolid. The results were summarized in **Table 3**. Compounds **6a** with hydroxyl substituent at C-ring and compound **14a** with the C-ring replaced by pyridine significantly decreased the MIC values compared to linezolid. Notably, compound **6a-3** exhibited 4- to 32-fold greater in *vitro* inhibition of four clinical isolates than that of linezolid, with MIC values of 0.125–0.25 μg/mL. And compound **6a-3** was particularly potent against linezolid-resistant *Enterococcus faecalis* (LREF).

Table 3. In *vitro* antibacterial activity against clinical isolates.

Compounds	MICs, μg/mL			
	MRSA[a](4)[b]	MSSA[c](4)	LREF[d](4)	VRE[e](4)

6a-1	0.5-1	0.5-1	1	1
6a-3	0.125-0.25	0.125-0.25	0.125-0.25	0.125-0.25
6a-6	0.5-1	0.5	0.5	0.5
6a-7	1-2	1-2	1-2	1-2
14a-1	2	2	0.25-0.5	0.25-0.5
14a-2	0.25-0.5	0.25-0.5	0.25-0.5	1
14a-3	0.25-0.5	0.25-0.5	0.5	1
14a-7	0.25-0.5	0.25-0.5	0.25-0.5	0.5
16-1	2	2	2	2
16-2	1-2	1-2	1-2	1-2
linezolid	1-4	1-4	>16	1-4

[a] Methicillin-resistant *Staphylococcus aureus*.

[b] Number of bacterial strains tested are given in parentheses.

[c] Methicillin-sensitive *Staphylococcus aureus*.

[d] Vancomycin-resistant *Enterococcus faecium*.

[e] Linezolid-resistant *Enterococcus faecalis*.

3.3. Physicochemical, ADME and drug-likeness properties prediction

Then the predictions of physicochemical, ADME parameters and drug-likeness properties of selected potent compounds were performed using the free SwissADME web tool (http://www.swissadme.ch/), and the results are shown in **Table 4**. No compounds violated Lipinski's rule of five, which indicated that all these compounds are likely to develop into oral drugs. All compounds showed no BBB permeability and high GI values, which indicated that these compounds were difficult to cross the blood-brain barrier to cause CNS adverse effects. High GI values and improved chemical stability suggested that these compounds may show good oral bioavailability. Based on the potent antibacterial activity and improved physicochemical and drug-likeness properties, further studies on cytotoxicity and metabolic stability of these compounds were conducted.

Table 4. *In silico Molecular properties prediction at SwissADME*.

Compounds	MW[a]	Log Po/w[b]	Hbond donor[c]	Hbond acceptor[d]	GI[e]	TPSA (Å2)[f]	Lipinski[g]	logS[h]	BBB permeant[i]
OB-104	453.51	2.66	1	6	High	77.48	0	-4.08	No
6a-1	469.51	2.16	2	7	High	97.71	0	-3.95	No
6a-3	472.53	2.87	2	6	High	119.77	0	-4.36	No
14a-1	454.50	1.86	1	7	High	90.37	0	-3.62	No
14a-2	441.46	1.92	1	7	High	96.36	0	-3.43	No
14a-3	457.52	2.54	1	6	High	112.43	0	-4.04	No
14a-7	489.52	1.73	1	8	High	129.65	0	-3.42	No
radezolid	438.45	2.01	3	7	High	112.24	0	-3.12	No
Linezolid	337.35	1.20	1	5	High	71.11	0	-2.22	No

[a] Molecular weight

[b] Logarithm of compound partition coefficient between n-octanol and water (< 5).

[c] Number of hydrogen bond donor (< 5).

[d] Number of hydrogen bond acceptors (< 10).

[e] Human gastrointestinal absorption.

[f] Topological polar surface area (≤ 140).

[g] Lipinski's rule of five.

[h] Predicted aqueous solubility.

[i] Predicted brain/blood partition coefficient.

3.4. Evaluation of acid stability and water solubility of potent compounds

Compound **OB-104** showed potent antibacterial activity but poor solubility and moderate metabolic stability, which were mainly caused by the hydrazone moiety. Therefore, optimal compounds were selected to evaluate for chemical stability in simulated gastric acid solutions (pH= 1.2) and solubility in water, and the results are shown in **Table 5**. These compounds exhibited excellent chemical stability in gastric acid solutions (pH=1.2) except for compound **6a-1**. Moreover, compounds **14a-1**, **14a-3** and **14a-7** showed improved water solubility compared with compound **OB-104**, which had poor solubility of 11.9 μg/mL. The solubility and chemical stability of these compounds can be significantly improved by optimization strategy at C-ring, which may due to the introduction of hindered and hydrophilic substituent or the change of charge distribution.

Table 5. Water solubility and acid stability of selected potent compounds.

Compounds	% Remaining in simulated gastric acid (pH=1.2) at different times					Solubility[a]
	0h	0.5h	1h	1.5h	2h	(μg/mL)
6a-1	100	69.8	35.0	12.2	5.3	-
6a-3	100	96.0	94.6	82.9	75.9	-
14a-1	100	98.3	96.8	96.2	95.8	63
14a-3	100	99.4	99.0	98.6	98.0	57
14a-7	100	99.7	99.4	99.2	98.7	68
OB-104	100	97.5	40.7	26.3	16.1	11.9

[a] Tested using UV Spectrophotometer (UV-2600).

3.5. In vitro cytotoxicity and ADME Results of selected compounds

Five of the selected potent compounds were evaluated for in *vitro* cytotoxicity and ADME properties, and the results are shown in **Table 6**. All compounds exhibited low cytotoxicity with IC_{50} values more than 25 μM against HepG2 cell, which indicated that these compounds were not toxic to hepatic cells. Furthermore, compound **14a-7** exhibited similar level of metabolic stability with a long half-life time of 409.44 min compared to linezolid, which indicated that this compound was stable in human microsome. However, compound **14a-3** showed a short half-life time of 19.24 min, probably because liver microsomal enzymes can oxidize the thioether moiety of compound **14a-3** to form sulfoxide or sulfone. In addition, compound **14a-1** exhibited moderate metabolic stability.

Table 6. In *vitro* cytotoxicity and ADME Results

Compounds	clogD[a]	HepG2 cytotoxicity	Human liver microsome (HLM)			
		IC_{50} (μM)	$T_{1/2}$ (min)	Cl_{int}	Substrate	Stability[c]

				(mL/min/kg)	remaining (%)[b]	
6a-1	1.14	＞25	-	-	-	-
6a-3	2.01	＞25	-	-	-	-
14a-1	0.95	＞25	101.04	17.20	74.88	moderate metabolism
14a-3	1.81	＞25	19.24	90.34	0.62	no stable
14a-7	0.11	＞25	409.44	4.25	92.99	stable
linezolid	0.64	＞25	∞	0	104.18	stable

[a]Calculated using instant JChem.[b]Substrate remaining were determined in incubations with NADPH after 45 min.[c]$T_{1/2}$ values ($T_{1/2}$ > 120 min)suggested these compounds were stable in liver microsomes; $T_{1/2}$ values ($T_{1/2}$ = 30-120 min) suggested these compounds showed moderate metabolism in liver microsomes; $T_{1/2}$ values ($T_{1/2}$ < 30 min)suggested these compounds were susceptible to metabolism in liver microsomes.

3.6. Evaluation of inhibition of monoamine oxidases A (MAO-A) by selected compounds

We then evaluated the safety profile of selected potent compounds compared with linezolid, which was an inhibitor of human MAO-A and may cause undesired side effect. As shown in **Table 7**, compounds **6a-3**, **6a-7**, **14a-3** and **14a-7** exhibited lower inhibition rates of human MAO-A compared to linezolid. Compounds **6a-6**, **6b-2** and **14a-1** inhibited human MAO-A slightly more strongly than linezolid. Compound **6a-3** exhibited the lowest inhibition rate of human MAO-A, and was more likely to developed into a drug candidate from a safety perspective.

Table 7. MAO-A inhibition of potent compounds

Compounds	**linezolid**	**6a-3**	**6a-6**	**6a-7**	**6b-2**	**14a-1**	**14a-3**	**14a-7**
MAO inhibition (%) (30 μM)	76	62	88	67	80	79	75	72

3.7. Molecular superposition models of 14a-7 and linezolid

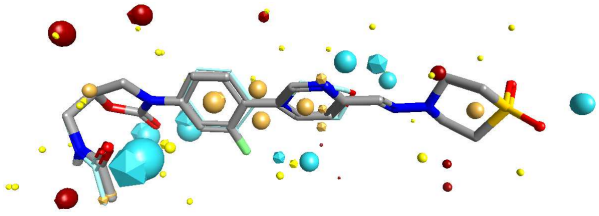


Figure 4. Superposition of compound **14a-7** and linezolid

Field-based molecular superposition of linezolid and compound **14a-7** was performed to predict the action of **14a-7** with ribosome. Obviously the conformations of **14a-7** showed the same field point pattem and perfect overlay with linezolid, which indicated that compound **14a-7** and linezolid may bound in 50S ribosome unit with the same pose. Furthermore, the hydrazone bond and terminal heterocycle may play a key role in the enhanced antibacterial activity.

4. Conclusions

In summary, we described the structural optimization of biaryloxazolidinone analogue **OB-104**, which was reported in our previous work. Compound **OB-104** was unstable in simulated gastric acid conditions (pH= 1.2) and human liver microsome. However, the metabolite **M-1**(**6a-1**) of **OB-104** showed significant antibacterial activity, which prompted us to conduct structural optimization strategies to improve the stability of hydrazone moiety while maintaining or enhancing antibacterial activity. Fortunately, two series of biaryloxazolidinone analogues (**6a-1**, **6a-3**, **14a-1**, **14a-3** and **14a-7**) with hydroxyl substituent at C-ring or the C-ring replaced by pyridine showed significant antibacterial activity against Gram-positive bacteria and clinical isolates as compared to linezolid. Further studies indicated that these compounds showed improved water solubility and chemical stability compared with compound **OB-104**. Compounds **6a-1**, **6a-3**, **14a-1**, **14a-3** and **14a-7** were not toxic to hepatic cells. The antibacterial activity of compound **14a-7** against clinical isolates of selected antibiotic-susceptible and antibiotic-resistant isolates was superior to that of linezolid. Moreover, compound **14a-7** was stable in human microsome, and exhibited lower inhibition rate of human MAO-A than linezolid. The maintaining antibacterial activity and all these improved drug-likeness properties and safety profile suggested that our strategies for structural optimization of biaryloxazolidinone analogue **OB-104** were successful. Further pharmacological studies of these potent compounds are still in progress.

5. Experimental

5.1. Chemistry

All materials were obtained from commercial suppliers and were used without further purification unless stated otherwise. Reactions' time of the compounds were monitored by TLC (silica gel GF254). Column chromatography was run on silica gel (200-300 mesh) from Qingdao Ocean Chemicals (Qingdao, Shandong, China). All melting points were measured with a Büchi Melting Point B-540 apparatus (Büchi Labortechnik, Flawil, Switzerland) and were uncorrected. Mass spectra (MS) were taken in ESI mode on Agilent 1100 LC-MS (Agilent, Palo Alto, CA, U.S.A.). ^{1}H-NMR and ^{13}C-NMR spectra were recorded on Bruker ARX-400, 400 MHz or Bruker ARX-600, 600 MHz spectrometers (Bruker Bioscience, Billerica, MA, USA) with TMS as an internal standard.

*5.1.1. (4-Bromo-3-fluorophenyl)carbamic acid methyl ester (**2**)*

4-Bromo-3-fluoroaniline (50.0 g, 0.26 mol) and pyridine (20.8 g, 0.26 mol) in DCM (250 mL) were added dropwise to a mixture of methyl chloroformate (27.2 g, 0.29 mol) in DCM while maintaining the temperature not more than 5 ℃. Then the reaction mixture was heated to room temperature for 2 h. The reaction mixture was washed with water (150 mL×3) for three times. The organic phase was washed with 3M HCl (200 mL), brine and concentrated in *vacuo* to yield **2** as a white solid in 84.6 % yield. MS (ESI) m/z(%): 248.2 $[M+Na]^{+}$.

*5.1.2. N-(((5S)-3-(4-bromo-3-fluorophenyl)-2-oxo-5-oxazolidinyl)methyl)acetamide (**3**)*

To a well-stirred solution of intermediate **2** (14.8 g, 0.06 mol) and lithium *tert*-butoxide (14.4 g, 0.18 mol) in dry DMF (86 mL) was added methanol (3.84g, 0.12 mol) dropwise over 15 min while maintaining the temperature not more than 5 ℃ under nitrogen and the resulted mixture was stirred at the same temperature for 1.5 h. Then a well-stirred solution of (*S*)-1-((acetylamino)methyl)-2-chloroethyl acetate (22.8 g, 0.18 mol) in DMF (80 mL) was added dropwise to this mixture and the resulted mixture was stirred at 25 ºC for another 20 h. After completion of the reaction monitored by TLC, the mixture was cooled to 0~5 ºC, then a well-stirred solution of ammonium chloride (6.41 g) in water (64 mL) was added while maintaining the temperature below 10 ºC. The resulted mixture was poured into water (800 mL) to give a white precipitate, which was collected by filtration and air-dried to give **3**

as a white solid in 82.5 % yield. MS (ESI) m/z(%): 353.4 $[M+Na]^+$.

*5.1.3. N-(((5S)-3-(3-fluoro-4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenyl)-2-oxo-5-oxazolidinyl)methyl) acetamide (**4**)*

To a well-stirred solution of intermediate **3** (19.8 g, 0.06 mol) and potassium acetate (17.7 g, 0.18 mol) in dry 1,4-dioxane (400 mL) was added bis(pinacolato)diboron (30.5 g, 0.12 mol) and $Pd(PPh_3)_2Cl_2$ (2.1 g, 0.003 mol), then the mixture was stirred at 90 ºC for 20 h under nitrogen . The mixture was cooled to room temperature and filtered through celite. The filtrate was concentrated in *vacuo*, then 20 mL diethyl ether was added to the residue and the mixture was stirred at room temperature for 1 h. The white precipitate was collected by filtration and air-dried to give **4** as a white solid in 75.2 % yield. MS (ESI) m/z(%): 401.3 $[M+Na]^+$.

*5.1.4 (S)-N-(3-(2-fluoro-3'-hydroxy-4'-formylbiphenyl-4-yl)-2-oxo-1,3-oxazolidine-5-yl)acetamide (**5a**)*

To a well-stirred solution of intermediate **4** (2.0 g, 5.29 mmol) and sodium carbonate (2.24 g, 21.16 mmol) in DMF (20 mL) and water (2 mL) was added 4-bromo-2-hydroxybenzaldehyde (1.06 g, 5.29 mmol) and $Pd(PPh_3)_2Cl_2$ (0.18 g, 0.26 mmol), then the mixture was stirred at 75 ºC for 10 h under nitrogen. After completion of the reaction monitored by TLC, water (50 mL) was added and the mixture was extracted with DCM (15 mL×3) for three times. The organic phase was washed with brine and concentrated in *vacuo* to yield **5a** as a white solid in 73 % yield. MS (ESI) m/z(%): 395.0 $[M+Na]^+$. ^{1}H NMR (400 MHz, DMSO) δ 9.55 (s, 1H), 8.26 (t, *J* = 5.6 Hz, 1H), 7.62 – 7.50 (m, 2H), 7.40 (d, *J* = 8.6 Hz, 1H), 7.26 (t, *J* = 7.8 Hz, 1H), 6.95 (d, *J* = 8.7 Hz, 2H), 6.79 (d, *J* = 8.6 Hz, 1H), 4.76 (td, *J* = 11.1, 5.5 Hz, 1H), 4.16 (t, *J* = 9.0 Hz, 1H), 3.78 (dd, *J* = 9.0, 6.5 Hz, 1H), 3.44 (t, *J* = 5.3 Hz, 2H), 1.85 (s, 3H).

*5.1.4.1 (S)-N-(3-(2-fluoro-3'-hydroxy-4'-((4-methylpiperazin-1-yl)iminomethyl)biphenyl-4-yl)-2-oxo-1,3-oxazolidie-5-ylmethyl)acetamide (**6a-1**)*

To a well-stirred solution of intermediate **5a** (2.0 g, 5.29 mmol) in ethanol (10 mL) was added 1-amino-4-methylpiperazine **9a** (0.61 g, 5.29 mmol) and a drop of acetic acid, and the mixture was stirred at 78 ºC for 2 h. The mixture was cooled to room temperature and the resulting solid was collected by filtration and purified by column chromatography to give the target compounds **6a-1** as a yellow solid in 75% yield. M.p.: 228-230 °C; MS (ESI), m/z (%): 470.3 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO) δ 11.44 (s, 1H), 8.27 (t, *J* = 5.7 Hz, 1H), 7.96 (s, 1H), 7.65 – 7.54 (m, 2H), 7.50 – 7.35 (m, 2H), 7.01 (s, 2H), 4.76 (td, *J* = 11.3, 5.4 Hz, 1H), 4.17 (t, *J* = 9.0 Hz, 1H), 3.79 (dd, *J* = 9.0, 6.5 Hz, 1H), 3.44 (t, *J* = 5.4 Hz, 2H), 3.13 (s, 4H), 2.51 (s, 4H), 2.24 (s, 3H), 1.84 (s, 3H).

*5.1.4.2 (S)-N-(3-(2-fluoro-3'-hydroxy-4'-(morpholin-4-yliminomethyl)biphenyl-4-yl)-2-oxo-1,3-oxazolidine-5-ylmethyl)acetamide (**6a-2**)*

Compound **6a-2** was synthesized from intermediate **5a** and **9b** in the same method as described for the synthesis of **6a-1**. Yellow solid; yield: 82 %; M.p.: 253-255 °C; MS (ESI), m/z (%): 457.4 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO) δ 11.33 (s, 1H), 8.27 (t, *J* = 5.7 Hz, 1H), 8.01 (s, 1H), 7.64 – 7.56 (m, 2H), 7.49 – 7.39 (m, 2H), 7.11 – 7.02 (m, 2H), 4.76 (td, *J* = 11.3, 5.4 Hz, 1H), 4.17 (t, *J* = 9.0 Hz, 1H), 3.81 – 3.77 (m, 5H), 3.44 (t, *J* = 5.4 Hz, 2H), 3.18 – 3.10 (m, 4H), 1.85 (s, 3H).

*5.1.4.3 (S)-N-(3-(2-fluoro-3'-hydroxy-4'-(thiomorpholin-4-yliminomethyl)biphenyl-4-yl)-2-oxo-1,3-oxazolidine-5-ylmethyl)acetamide (**6a-3**)*

Compound **6a-3** was synthesized from intermediate **5a** and **9c** in the same method as described for the synthesis of **6a-1**. Yellow solid; yield: 79 %; M.p.: 235-237 °C; MS (ESI), m/z (%): 473.4 $[M+H]^+$. ^{1}H NMR (400 MHz,

DMSO) δ 11.36 (s, 1H), 8.27 (t, *J* = 5.7 Hz, 1H), 8.00 (s, 1H), 7.63 – 7.56 (m, 2H), 7.45 (d, *J* = 8.1 Hz, 1H), 7.41 (dd, *J* = 8.7, 1.7 Hz, 1H), 7.09 – 7.01 (m, 2H), 4.80 – 4.73 (m, 1H), 4.17 (t, *J* = 8.9 Hz, 1H), 3.78 (dd, *J* = 8.9, 6.6 Hz, 1H), 3.52 – 3.48 (m, 4H), 3.44 (t, *J* = 5.2 Hz, 2H), 2.81 – 2.74 (m, 4H), 1.84 (s, 3H).

*5.1.4.4 (S)-N-(3-(2-fluoro-3'-hydroxy-4'-((4-tert-butoxycarbonylpiperazin-1-yl)iminomethyl)biphenyl-4-yl)-2-oxo-1,3-oxazolidine-5-ylmethyl)acetamide (**6a-4**)*

Compound **6a-4** was synthesized from intermediate **5a** and **9e** in the same method as described for the synthesis of **6a-1**. Yellow solid; yield: 85 %.

*5.1.4.5 (S)-N-(3-(2-fluoro-3'-hydroxy-4'-(piperazin-1-yliminomethyl)biphenyl-4-yl)-2-oxo-1,3-oxazolidine-5-ylmethyl)acetamide (**6a-5**)*

Trifluoroacetic acid (5 mL) was added to a well-stirred solution of **6a-4** (0.2 g, 0.36 mmol) in DCM(10 mL) while maintaining the temperature below 5 °C, then the mixture was stirred at 25 ºC for 5 h. After completion of the reaction monitored by TLC, the mixture was concentrated in *vacuo* and then added water (10 mL) to the residue. Sodium carbonate solution was added dropwise to the mixture until pH was 8. The mixture was extracted with DCM (15 mL×3) for three times. The organic phase was washed with brine and concentrated in *vacuo* to yield **6a-5** as a yellow solid in 68 % yield; M.p.: 214-216 °C; MS (ESI), m/z (%): 456.5 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO) δ 11.49 (s, 1H), 8.28 (t, *J* = 5.8 Hz, 1H), 7.96 (s, 1H), 7.63 – 7.56 (m, 2H), 7.45 (d, *J* = 8.1 Hz, 1H), 7.40 (dd, *J* = 8.6, 2.0 Hz,1H), 7.06 (d, *J* = 8.0 Hz, 1H), 7.03 (s, 1H), 4.77 (td, *J* = 11.4, 5.4 Hz, 1H), 4.17 (t, *J* = 9.0 Hz, 1H), 3.79 (dd, *J* = 9.1, 6.5 Hz, 1H), 3.45 – 3.42 (m, 2H), 3.38 (s, 1H), 3.11 – 3.04 (m, 4H), 2.92 (d, *J* = 4.5 Hz, 4H), 1.85 (s, 3H).

*5.1.4.6 (S)-N-(3-(2-fluoro-3'-hydroxy-4'-((1-oxothiomorpholin-4-yl)iminomethyl)biphenyl-4-yl)-2-oxo-1,3-oxazolidin-5-ylmethyl)acetamide (**6a-6**)*

3-Chloroperbenzoic acid (0.08 g, 0.46 mmol) was added to a well-stirred solution of **6a-3** (0.2 g, 0.42 mmol) in DCM (10 mL) while maintaining the temperature below 5 °C, then the mixture was stirred at 25 ºC for 4 h. After completion of the reaction monitored by TLC, 10 mL saturated sodium carbonate solution was added to the mixture and the mixture was extracted with DCM (30 mL×3) for three times. Then the combined organic extract was washed with brine and concentrated in *vacuo*. The residue was purified by column chromatography to give the target compounds **6a-6** as a yellow solid in 71 % yield. M.p.: 205-207 °C; MS (ESI) m/z(%): 511.26 $[M+Na]^+$. ^{1}H NMR (400 MHz, DMSO) δ 11.22 (s, 1H), 8.27 (t, *J* = 5.9 Hz, 1H), 8.09 (s, 1H), 7.63 – 7.56 (m, 2H), 7.48 (d, *J* = 8.1 Hz, 1H), 7.41 (dd, *J* = 8.6, 2.0 Hz, 1H), 7.08 (d, *J* = 8.0 Hz, 1H), 7.04 (s, 1H), 4.76 (td, *J* = 11.3, 5.3 Hz, 1H), 4.17 (t, *J* = 9.0 Hz, 1H), 3.78 (dd, *J* = 9.1, 6.4 Hz, 1H), 3.71 – 3.66 (m, 4H), 3.43 (t, *J* = 5.5 Hz, 2H), 3.04 – 2.96 (m, 2H), 2.80 – 2.77 (m, 2H), 1.84 (s, 3H).

*5.1.4.7 (S)-N-(3-(2-fluoro-3'-hydroxy-4'-((1,1-dioxothiomorpholin-4-yl)iminomethyl)biphenyl-4-yl)-2-oxo-1,3-oxazolidin-5-ylmethyl)acetamide (**6a-7**)*

Compound **6a-7** was synthesized from intermediate **5a** and **9f** in the same method as described for the synthesis of **6a-1**. Yellow solid; yield: 74 %; M.p.: 223-225 °C; MS (ESI), m/z (%): 505.3 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO) δ 11.30 (s, 1H), 8.26 (t, *J* = 5.7 Hz, 1H), 8.00 (s, 1H), 7.64 – 7.55 (m, 2H), 7.47 (d, *J* = 8.1 Hz, 1H), 7.41 (dd, *J* = 8.6, 1.8 Hz, 1H), 7.07 (d, *J* = 8.0 Hz, 1H), 7.03 (s, 1H), 4.76 (td, *J* = 11.3, 5.4 Hz, 1H), 4.17 (t, *J* = 9.0 Hz, 1H), 3.78 (dd, *J* = 9.1, 6.5 Hz, 1H), 3.53 (s, 4H), 3.44 (t, *J* = 5.4 Hz, 2H), 3.12 (s, 4H), 1.84 (s, 3H).

*5.1.5. (S)-N-(3-(2-fluoro-3'-fluoro-4'-formylbiphenyl-4-yl)-2-oxo-1,3-oxazolidine-5-ylmethyl)acetamide (**5b**)*

Compound **5b** was synthesized from intermediate **4** and 2-fluoro-4-bromobenzaldehyde in the same method as described for the synthesis of **5a**. Yellow solid; yield: 71%; MS (ESI) m/z(%): 373.2 $[M-H]^-$.

*5.1.5.1. (S)-N-(3-(2-fluoro-3'-fluoro-4'-((4-methylpiperazin-1-yl)iminomethyl)biphenyl-4-yl)-2-oxo-1,3-oxazolidin-5-ylmethyl)acetamide (**6b-1**)*

Compound **6b-1** was synthesized from intermediate **5b** and **9a** in the same method as described for the synthesis of **6a-1**. Yellow solid; yield: 76 %; M.p.: 205-207 °C; MS (ESI), m/z (%): 472.5 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO) δ 8.27 (t, *J* = 5.2 Hz, 1H), 7.85 (t, *J* = 8.2 Hz, 1H), 7.68 – 7.57 (m, 3H), 7.45 – 7.38 (m, 3H), 4.77 (td, *J* = 11.0, 5.3 Hz, 1H), 4.17 (t, *J* = 8.9 Hz, 1H), 3.79 (dd, *J* = 7.9, 7.1 Hz, 1H), 3.44 (s, 2H), 3.34 (s, 4H), 3.19 (s, 4H), 2.25 (s, 3H), 1.84 (s, 3H).

*5.1.5.2 (S)-N-(3-(2-fluoro-3'-fluoro-4'-(morpholin-4-yliminomethyl)biphenyl-4-yl)-2-oxo-1,3-oxazolidine-5-ylmethyl)acetamide (**6b-2**)*

Compound **6b-2** was synthesized from intermediate **5b** and **9b** in the same manner as described for the synthesis of **6a-1**. Yellow solid; yield: 75%; M.p.: 287-289 °C; MS (ESI), m/z (%): 459.4$[M+H]^+$. ^{1}H NMR (400 MHz, DMSO) δ 8.27 (t, *J* = 5.8 Hz, 1H), 7.86 (t, *J* = 8.2 Hz, 1H), 7.75 (s, 1H), 7.68 – 7.59 (m, 2H), 7.46 – 7.38 (m, 3H), 4.77 (td, *J* = 11.3, 5.3 Hz, 1H), 4.17 (t, *J* = 9.0 Hz, 1H), 3.82 – 3.74 (m, 5H), 3.44 (t, *J* = 5.4 Hz, 2H), 3.21 – 3.15 (m, 4H), 1.84 (s, 3H).

*5.1.5.3 (S)-N-(3-(2-fluoro-3'-fluoro-4'-(thiomorpholin-4-yliminomethyl)biphenyl-4-yl)-2-oxo-1,3-oxazolidine-5-ylmethyl)acetamide (**6b-3**)*

Compound **6b-3** was synthesized from intermediate **5b** and **9c** in the same method as described for the synthesis of **6a-1**. Yellow solid; yield: 82%; M.p.: 220-222 °C; MS (ESI), m/z (%): 475.3 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO) δ 8.26 (t, *J* = 5.8 Hz, 1H), 7.85 (t, *J* = 8.2 Hz, 1H), 7.72 (s, 1H), 7.67 – 7.59 (m, 2H), 7.44 – 7.39 (m, 3H), 4.77 (td, *J* = 11.4, 5.3 Hz, 1H), 4.17 (t, *J* = 9.0 Hz, 1H), 3.79 (dd, *J* = 9.1, 6.4 Hz, 1H), 3.59 – 3.54 (m, 4H), 3.44 (t, *J* = 5.5 Hz, 2H), 2.76 – 2.73 (m, 4H), 1.84 (s, 3H).

*5.1.5.4 (S)-N-(3-(2-fluoro-3'-fluoro-4'-(piperidin-1-yliminomethyl)biphenyl-4-yl)-2-oxo-1,3-oxazolidine-5-ylmethyl)acetamide (**6b-4**)*

Compound **6b-4** was synthesized from intermediate **5b** and **9d** in the same method as described for the synthesis of **6a-1**. Yellow solid; yield: 83%; M.p.: 179-182 °C; MS (ESI), m/z (%): 457.5 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO) δ 8.27 (t, *J* = 5.7 Hz, 1H), 7.85 (t, *J* = 8.2 Hz, 1H), 7.67 – 7.58 (m, 3H), 7.44 – 7.37 (m, 3H), 4.77 (td, *J* = 11.4, 5.5 Hz, 1H), 4.17 (t, *J* = 9.0 Hz, 1H), 3.79 (dd, *J* = 9.0, 6.5 Hz, 1H), 3.44 (t, *J* = 5.4 Hz, 2H), 3.22 – 3.16 (m, 4H), 1.84 (s, 3H), 1.71 – 1.63 (m, 4H), 1.52 (d, *J* = 4.7 Hz, 2H).

*5.1.5.5 (S)-N-(3-(2-fluoro-3'-fluoro-4'-((4-tert-butoxycarbonylpiperazin-1-yl)iminomethyl)biphenyl-4-yl)-2-oxo-1,3-oxazolidine-5-ylmethyl)acetamide (**6b-5**)*

Compound **6b-5** was synthesized from intermediate **5b** and **9e** in the same method as described for the synthesis of **6a-1**. Yellow solid; yield: 80%.

*5.1.5.6 (S)-N-(3-(2-fluoro-3'-fluoro-4'-(piperazin-1-yliminomethyl)biphenyl-4-yl)-2-oxo-1,3-oxazolidine-5-ylmethyl)acetamide (**6b-6**)*

Compound **6b-6** was synthesized from intermediate **6b-5** in the same method as described for the synthesis of

6a-5. Yellow solid; yield: 75%; M.p.: 187-189 °C; MS (ESI) m/z(%):458.3 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO) δ 8.27 (t, *J* = 5.5 Hz, 1H), 8.07 (s, 1H), 7.85 (t, *J* = 8.1 Hz, 1H), 7.71 – 7.58 (m, 3H), 7.42 (d, *J* = 8.6 Hz, 3H), 4.77 (td, *J* = 11.8, 6.0 Hz, 1H), 4.17 (t, *J* = 9.0 Hz, 1H), 3.79 (dd, *J* = 8.2, 7.2 Hz, 1H), 3.46 – 3.42 (m, 2H), 3.14 (s, 4H), 2.91 (s, 4H), 1.84 (s, 3H).

*5.1.6 (S)-N-(3-(2-fluoro-3'-methoxy-4'-formylbiphenyl-4-yl)-2-oxo-1,3-oxazolidine-5-ylmethyl)acetamide (**5c**)*

Compound **5c** was synthesized from intermediate **4** and 4-bromo-2-methoxybenzaldehyde in the same method as described for the synthesis of **5a**. Yellow solid; yield: 75 %; MS (ESI) m/z(%):387.3 $[M+H]^+$.^{1}H NMR (400 MHz, DMSO) δ 10.38 (s, 1H), 8.28 (t, *J* = 5.7 Hz, 1H), 7.78 (d, *J* = 8.0 Hz, 1H), 7.72 (t, *J* = 8.8 Hz, 1H), 7.64 (dd, *J* = 13.7, 2.0 Hz, 1H), 7.46 (dd, *J* = 8.6, 2.0 Hz,1H), 7.36 (s, 1H), 7.27 (d, *J* = 8.0 Hz, 1H), 4.77 (dt, *J* = 11.3, 5.6 Hz, 1H), 4.19 (t, *J* = 9.0 Hz, 1H), 3.99 (s, 3H), 3.80 (dd, *J* = 9.1, 6.5 Hz, 1H), 3.45 (t, *J* = 5.4 Hz, 2H), 1.84 (s, 3H).

*5.1.6.1 (S)-N-(3-(2-fluoro-3'-methoxy-4'-(morpholin-4-yliminomethyl)biphenyl-4-yl)-2-oxo-1,3-oxazolidine-5-ylmethyl)acetamide (**6c-1**)*

Compound **6c-1** was synthesized from intermediate **5c** and **9b** in the same method as described for the synthesis of **6a-1**. Yellow solid; yield: 71 %; M.p.: 179-181 °C; MS (ESI), m/z (%): 471.5 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO) δ 8.28 (t, *J* = 5.6 Hz, 1H), 7.87 (s, 1H), 7.78 (d, *J* = 8.1 Hz, 1H), 7.66 – 7.59 (m, 2H), 7.42 (d, *J* = 8.4 Hz, 1H), 7.17 (s, 1H), 7.13 (d, *J* = 8.0 Hz, 1H), 4.77 (td, *J* = 11.4, 5.5 Hz, 1H), 4.18 (t, *J* = 9.0 Hz, 1H), 3.89 (s, 3H), 3.80 – 3.74 (m, 5H), 3.44 (t, *J* = 5.3 Hz, 2H), 3.16 – 3.06 (m, 4H), 1.85 (s, 3H).

*5.1.6.2 (S)-N-(3-(2-fluoro-3'-methoxy-4'-(thiomorpholin-4-yliminomethyl)biphenyl-4-yl)-2-oxo-1,3-oxazolidine-5-ylmethyl)acetamide (**6c-2**)*

Compound **6c-2** was synthesized from intermediate **5c** and **9c** in the same method as described for the synthesis of **6a-1**. Yellow solid; yield: 76 %; M.p.: 207-209 °C; MS (ESI), m/z (%): 487.5 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO) 8.28 (t, *J* = 5.8 Hz, 1H), 7.85 (s, 1H), 7.78 (d, *J* = 8.1 Hz, 1H), 7.67 – 7.57 (m, 2H), 7.41 (dd, *J* = 8.6, 2.0 Hz, 1H), 7.16 (s, 1H), 7.13 (d, *J* = 8.0 Hz, 1H), 4.77 (td, *J* = 11.4, 5.4 Hz, 1H), 4.17 (t, *J* = 9.0 Hz, 1H), 3.88 (s, 3H), 3.79 (dd, *J* = 9.1, 6.5 Hz, 1H), 3.50 – 3.46 (m, 4H), 3.44 (t, *J* = 5.5 Hz, 2H), 2.78 – 2.72 (m, 4H), 1.85 (s, 3H).

*5.1.7 (S)-N-(3-((2-fluoro-3'-methyl-4'-formyl)biphenyl-4-yl)-2-oxo-1,3-oxazolidine-5-ylmethyl)acetamide (**5d**)*

Compound **5d** was synthesized from intermediate **4** and 4-bromo-2-methylbenzaldehyde in the same method as described for the synthesis of **5a**. Yellow solid; yield: 84 %; MS (ESI) m/z(%):393.2 $[M+H]^+$.

*5.1.7.1 (S)-N-(3-(2-fluoro-3'-methyl-4'-((4-methylpiperazin-1-yl)iminomethyl)biphenyl-4-yl)-2-oxo-1,3-oxazolidin-5-ylmethyl)acetamide (**6d-1**)*

Compound **6d-1** was synthesized from intermediate **5d** and **9a** in the same method as described for the synthesis of **6a-1**. Yellow solid; yield: 76 %; M.p.: 171-174 °C; MS (ESI), m/z (%): 468.3 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO) δ 8.27 (t, *J* = 5.7 Hz, 1H), 7.76 (d, *J* = 7.9 Hz, 2H), 7.63 – 7.54 (m, 2H), 7.40 (dd, *J* = 8.6, 1.9 Hz, 1H), 7.35 (d, *J* = 6.0 Hz, 2H), 4.77 (td, *J* = 11.4, 5.4 Hz, 1H), 4.17 (t, *J* = 9.0 Hz, 1H), 3.79 (dd, *J* = 9.1, 6.5 Hz, 1H), 3.44 (t, *J* = 5.4 Hz, 2H), 3.23 – 3.12 (m, 4H), 2.57 – 2.51 (m, 4H), 2.44 (s, 3H), 2.25 (s, 3H), 1.85 (s, 3H).

*5.1.7.2 (S)-N-(3-(2-fluoro-3'-methyl-4'-(thiomorpholin-4-yliminomethyl)biphenyl-4-yl)-2-oxo-1,3-oxazolidine-5-ylmethyl)acetamide (**6d-2**)*

Compound **6d-2** was synthesized from intermediate **5d** and **9c** in the same method as described for the synthesis of **6a-1**. Yellow solid; yield: 78 %; M.p.: 196-199 °C; MS (ESI), m/z (%): 471.5 $[M+H]^+$. ^{1}H NMR (400 MHz,

DMSO) δ 8.27 (t, *J* = 5.8 Hz, 1H), 7.81 (s, 1H), 7.76 (d, *J* = 8.7 Hz, 1H), 7.63 – 7.55 (m, 2H), 7.40 (dd, *J* = 8.6, 2.0 Hz, 1H), 7.38 – 7.34 (m, *J* = 5.8 Hz, 2H), 4.76 (td, *J* = 11.3, 5.4 Hz, 1H), 4.17 (t, *J* = 9.0 Hz, 1H), 3.79 (dd, *J* = 9.1, 6.5 Hz, 1H), 3.58 – 3.51 (m, 4H), 3.44 (t, *J* = 5.4 Hz, 2H), 2.80 – 2.72 (m, 4H), 2.45 (s, 3H), 1.85 (s, 3H).

*5.1.7.3 (S)-N-(3-(2-fluoro-3'-methyl-4'-((4-tert-butoxycarbonylpiperazin-1-yl)iminomethyl)biphenyl-4-yl)-2-oxo-1,3-oxazolidine-5-ylmethyl)acetamide (**6d-3**)*

Compound **6d-3** was synthesized from intermediate **5d** and **9e** in the same method as described for the synthesis of **6a-1**. Yellow solid; yield: 82 %.

*5.1.7.4 (S)-N-(3-(2-fluoro-3'-methyl-4'-(piperazin-1-yliminomethyl)biphenyl-4-yl)-2-oxo-1,3-oxazolidine-5-ylmethyl)acetamide (**6d-4**)*

Compound **6d-4** was synthesized from intermediate **6d-3** in the same method as described for the synthesis of **6a-5**. White solid; yield: 59 %; M.p.: 202-204 °C; MS (ESI), m/z (%): 454.3 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO) δ 8.92 (s, 1H), 8.29 (t, *J* = 5.6 Hz, 1H), 7.88 (s, 1H), 7.77 (d, *J* = 8.7 Hz, 1H), 7.63 – 7.55 (m, 2H), 7.44 – 7.33 (m, 3H), 4.77 (td, *J* = 11.3, 5.4 Hz, 1H), 4.17 (t, *J* = 9.0 Hz, 1H), 3.80 (dd, *J* = 9.0, 6.5 Hz, 1H), 3.46 – 3.42 (m, 2H), 3.38 (s, 4H), 3.23 (s, 4H), 2.47 (s, 3H), 1.85 (s, 3H).

*5.1.8 (S)-N-(3-((2-fluoro-3'-trifluoromethyl-4'-formyl)biphenyl-4-yl)-2-oxo-1,3-oxazolidine-5-ylmethyl) acetamide (**5e**)*

Compound **5e** was synthesized from intermediate **4** and 4-bromo-2-trifluoromethylbenzaldehyde in the same method as described for the synthesis of **5a**. Yellow solid; yield: 74 %; MS (ESI), m/z (%): 447.2 $[M+H]^+$.

*5.1.8.1 (S)-N-(3-(2-fluoro-3'-trifluoromethyl-4'-((4-methylpiperazin-1-yl)iminomethyl)biphenyl-4-yl)-2-oxo-1,3-oxazolidin-5-ylmethyl)acetamide (**6e-1**)*

Compound **6e-1** was synthesized from intermediate **5e** and **9a** in the same method as described for the synthesis of **6a-1**. Yellow solid; yield: 81 %; M.p.: 198-200 °C; MS (ESI), m/z (%): 522.5 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO) δ 8.28 (s, 1H), 8.12 (d, *J* = 7.9 Hz, 1H), 7.87 – 7.78 (s, 2H), 7.73 – 7.59 (m, 3H), 7.50 – 7.41 (m, 1H), 4.78 (s, 1H), 4.18 (t, *J* = 8.7 Hz, 1H), 3.83 – 3.77 (m, 1H), 3.45 (s, 2H), 3.21 (s, 4H), 2.24 (s, 3H), 1.85 (s, 3H).

*5.1.8.2 (S)-N-(3-(2-fluoro-3'-trifluoromethyl-4'-(thiomorpholin-4-yliminomethyl)biphenyl-4-yl)-2-oxo-1,3-oxazolidin-5-ylmethyl)acetamide (**6e-2**)*

Compound **6e-2** was synthesized from intermediate **5e** and **9c** in the same method as described for the synthesis of **6a-1**. Yellow solid; yield: 76 %; M.p.: 211-213 °C; MS (ESI), m/z (%): 547.3 $[M+Na]^+$. ^{1}H NMR (600 MHz, DMSO) δ 8.27 (t, *J* = 5.8 Hz, 1H), 8.14 (d, *J* = 8.8 Hz, 1H), 7.84 (s, 2H), 7.75 (s, 1H), 7.69 (t, *J* = 8.9 Hz, 1H), 7.64 (dd, *J* = 13.7, 1.7 Hz, 1H), 7.45 (dd, *J* = 8.6, 1.7 Hz, 1H), 4.78 (td, *J* = 11.4, 5.5 Hz, 1H), 4.18 (t, *J* = 9.0 Hz, 1H), 3.83 – 3.77 (m, 5H), 3.45 (t, *J* = 5.4 Hz, 2H), 3.23 – 3.18 (m, 4H), 1.85 (s, 3H).

*5.1.9 (S)-N-(3-(3-fluoro-4-(6-formylpyridin-3-yl)phenyl)-2-oxo-1,3-oxazolidine-5-ylmethyl)acetamide (**13a**)*

Compound **13a** was synthesized from intermediate **4** and 3-bromo-6-pyridinecarboxaldehyde in the same method as described for the synthesis of **5a**. White solid; yield: 85%; MS (ESI), m/z (%): 358.2 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO) δ 10.04 (s, 1H), 9.03 (s, 1H), 8.26 (dd, *J* = 13.1, 7.0 Hz, 2H), 8.04 (d, *J* = 8.1 Hz,1H), 7.78 (t, *J* = 8.8 Hz, 1H), 7.69 (dd, *J* = 13.7, 2.0 Hz, 1H), 7.51 (dd, *J* = 8.6, 2.0 Hz, 1H), 4.79 (td, *J* = 11.3, 5.3 Hz, 1H), 4.20 (t, *J* = 9.0 Hz, 1H), 3.82 (dd, *J* = 9.1, 6.5 Hz, 1H), 3.45 (t, *J* = 5.4 Hz, 2H), 1.85 (s, 3H).

*5.1.9.1 (S)-N-(3-(3-fluoro-4-(6-((4-methylpiperazin-1-yl)iminomethyl)pyridin-3-yl)phenyl)-2-oxo-1,3-oxazolidin -5-ylmethyl)acetamide (**14a-1**)*

Compound **14a-1** was synthesized from intermediate **13a** and **9a** in the same method as described for the synthesis of **6a-1**. Yellow solid; yield: 78%; M.p.: 211-213 °C; MS (ESI), m/z (%): 455.5 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO) δ 8.69 (s, 1H), 8.27 (t, *J* = 5.6 Hz, 1H), 7.94 (d, *J* = 8.5 Hz, 1H), 7.84 (d, *J* = 8.3 Hz, 1H), 7.72 – 7.59 (m, 3H), 7.45 (dd, *J* = 8.5, 1.9 Hz, 1H), 4.78 (td, *J* = 11.4, 5.6 Hz, 1H), 4.19 (t, *J* = 9.0 Hz, 1H), 3.80 (dd, *J* = 9.1, 6.5 Hz, 1H), 3.46 – 3.43 (m, 2H), 3.25 (s, 4H), 2.57 (s, 4H), 2.29 (s, 3H), 1.85 (s, 3H). ^{13}C NMR (101 MHz, DMSO) δ 170.49, 159.65 (d, *J* = 245.4 Hz), 154.68, 154.45, 148.79, 140.33 (d, *J* = 11.4 Hz), 136.55 (d, *J* = 2.8 Hz), 134.45, 131.12 (d, *J* = 4.7 Hz), 129.15, 119.92 (d, *J* = 13.6 Hz), 118.55, 114.59, 106.03 (d, *J* = 28.3 Hz), 72.28, 54.28 (2C), 50.73 (2C), 47.66, 46.02, 41.86, 22.92.

*5.1.9.2 (S)-N-(3-(3-fluoro-4-(6-(morpholin-4-yliminomethyl)pyridin-3-yl)phenyl)-2-oxo-1,3-oxazolidine -5-ylmethyl)acetamide (**14a-2**)*

Compound **14a-2** was synthesized from intermediate **13a** and **9b** in the same method as described for the synthesis of **6a-1**. Yellow solid; yield: 77%; M.p.: 207-209 °C; MS (ESI), m/z (%): 442.4 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO) δ 8.70 (s, 1H), 8.27 (t, *J* = 5.7 Hz, 1H), 7.95 (d, *J* = 8.4 Hz, 1H), 7.85 (d, *J* = 8.3 Hz, 1H), 7.73 – 7.61 (m, 3H), 7.45 (dd, *J* = 8.6, 1.9 Hz, 1H), 4.78 (td, *J* = 11.3, 5.3 Hz, 1H), 4.18 (t, *J* = 9.0 Hz, 1H), 3.82 – 3.78 (m, 5H), 3.45 (t, *J* = 5.4 Hz, 2H), 3.26 – 3.18 (m, 4H), 1.85 (s, 3H).

*5.1.9.3 (S)-N-(3-(3-fluoro-4-(6-(thiomorpholin-4-yliminomethyl)pyridin-3-yl)phenyl)-2-oxo-1,3-oxazolidine -5-ylmethyl)acetamide (**14a-3**)*

Compound **14a-3** was synthesized from intermediate **13a** and **9c** in the same method as described for the synthesis of **6a-1**. Yellow solid; yield: 74%; M.p.: 219-221 °C; MS (ESI), m/z (%): 458.4 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO) δ 8.69 (s, 1H), 8.27 (t, *J* = 5.4 Hz, 1H), 7.93 (d, *J* = 8.3 Hz, 1H), 7.84 (d, *J* = 8.3 Hz, 1H), 7.71 – 7.61 (m, 3H), 7.49 – 7.42 (m, 1H), 4.78 (dd, *J* = 7.8, 5.5 Hz, 1H), 4.18 (t, *J* = 8.9 Hz, 1H), 3.81 (dd, *J* = 8.8, 6.6 Hz, 1H), 3.64 – 3.58 (m, 4H), 3.45 (t, *J* = 5.2 Hz, 2H), 2.81 – 2.70 (m, 4H), 1.86 (s, 3H). ^{13}C NMR (101 MHz, DMSO) δ 170.51, 159.65 (d, *J* = 245.2 Hz), 154.45, 153.87, 148.65, 140.44 (d, *J* = 11.2 Hz), 137.16 (d, *J* = 2.5 Hz), 133.54, 131.10 (d, *J* = 4.7 Hz), 129.41, 119.69 (d, *J* = 13.3 Hz), 119.39, 114.63, 106.04 (d, *J* = 28.3 Hz), 72.30, 52.97 (2C), 47.66, 41.86, 25.74 (2C), 22.91.

*5.1.9.4 (S)-N-(3-(3-fluoro-4-(6-((4-tert-butoxycarbonylpiperazin-1-yl)iminomethyl)pyridin-3-yl)phenyl)-2-oxo-1,3-oxazolidine -5-ylmethyl)acetamide (**14a-4**)*

Compound **14a-4** was synthesized from intermediate **13a** and **9e** in the same method as described for the synthesis of **6a-1**. Yellow solid; yield: 88 %.

*5.1.9.5 (S)-N-(3-(3-fluoro-4-(6-(piperazin-1-yliminomethyl)pyridin-3-yl)phenyl)-2-oxo-1,3-oxazolidine -5-ylmethyl)acetamide (**14a-5**)*

Compound **14a-5** was synthesized from intermediate **14a-4** in the same method as described for the synthesis of **6a-5**. Yellow solid; yield: 72 %; M.p.: 190-192 °C; MS (ESI), m/z (%): 441.7 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO) δ 8.68 (s, 1H), 8.34 (t, *J* = 5.7 Hz, 1H), 7.93 (d, *J* = 8.4 Hz, 1H), 7.83 (d, *J* = 8.4 Hz, 1H), 7.70 – 7.60 (m, 3H), 7.45 (dd, *J* = 8.6, 1.9 Hz, 1H), 4.78 (td, *J* = 11.4, 5.4 Hz, 1H), 4.18 (t, *J* = 9.0 Hz, 1H), 3.82 (dd, *J* = 9.0, 6.5 Hz, 1H), 3.44 (t, *J* = 5.4 Hz, 2H), 3.21 – 3.14 (m, 4H), 2.95 – 2.85 (m, 4H), 1.85 (s, 3H).

*5.1.9.6 (S)-N-(3-(3-fluoro-4-(6-((1-oxothiomorpholin-4-yl)iminomethyl)pyridin-3-yl)phenyl)-2-oxo-1,3-oxazolidin-5-ylmethyl)acetamide (**14a-6**)*

Compound **14a-6** was synthesized from compound **14a-3** in the same method as described for the synthesis of **6a-6**. Yellow solid; yield: 79 %; M.p.: 205-207 °C; MS (ESI), m/z (%): 474.2 $[M+Na]^+$. ^{1}H NMR (400 MHz, DMSO) δ 8.71 (s, 1H), 8.26 (t, *J* = 5.7 Hz, 1H), 7.96 (d, *J* = 8.4 Hz, 1H), 7.88 (d, *J* = 8.4 Hz, 1H), 7.75 (s, 1H), 7.71 – 7.61 (m, 2H), 7.45 (dd, *J* = 8.6, 1.9 Hz, 1H), 4.78 (td, *J* = 11.4, 5.5 Hz, 1H), 4.18 (t, *J* = 9.0 Hz, 1H), 3.85 – 3.74 (m, 5H), 3.44 (t, *J* = 5.4 Hz, 2H), 3.05 – 2.93 (m, 2H), 2.83 – 2.73 (m, 2H), 1.84 (s, 3H).

*5.1.9.7 (S)-N-(3-(3-fluoro-4-(6-((1,1-dioxothiomorpholin-4-yl)iminomethyl)pyridin-3-yl)phenyl)-2-oxo-1,3-oxazolidine-5-ylmethyl)acetamide (**14a-7**)*

Compound **14a-7** was synthesized from intermediate **13a** and **9f** in the same method as described for the synthesis of **6a-1**. Yellow solid; yield: 73 %; M.p.: 229-231 °C; MS (ESI), m/z (%): 490.4 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO) δ 8.74 (s, 1H), 8.26 (t, *J* = 5.7 Hz, 1H), 8.01 (d, *J* = 7.9 Hz, 1H), 7.89 (d, *J* = 8.4 Hz, 1H), 7.79 (s, 1H), 7.72 – 7.62 (m, 2H), 7.46 (dd, *J* = 8.6, 1.9 Hz, 1H), 4.78 (td, *J* = 11.4, 5.4 Hz, 1H), 4.19 (t, *J* = 9.0 Hz, 1H), 3.90 (s, 4H), 3.80 (dd, *J* = 9.1, 6.5 Hz, 1H), 3.44 (t, *J* = 5.4 Hz, 2H), 3.22 (s, 4H), 1.84 (s, 3H). ^{13}C NMR (101 MHz, DMSO) δ 177.19, 170.49, 159.68 (d, *J* = 245.7 Hz), 154.44, 153.67, 148.74, 140.52 (d, *J* = 11.1 Hz), 137.64, 136.95, 131.14, 129.88, 119.65, 114.67, 106.10 (d, *J* = 28.2 Hz), 72.29, 48.93 (2C), 48.76 (2C), 47.71, 41.88, 22.90.

*5.1.10 (S)-N-(3-(3-fluoro-4-(5-formylpyridin-2-yl)phenyl)-2-oxo-1,3-oxazolidine-5-ylmethyl)acetamide (**13b**)*

Compound **13b** was synthesized from intermediate **4** and 2-bromo-5-pyridinecarboxaldehyde in the same method as described for the synthesis of **5a**. White solid; yield: 82 %; MS (ESI), m/z (%): 358.2 $[M+H]^+$.

*5.1.10.1 (S)-N-(3-(3-fluoro-4-(5-((4-methylpiperazin-1-yl)iminomethyl)pyridin-2-yl)phenyl)-2-oxo-1,3-oxazolidin-5-ylmethyl)acetamide (**14b-1**)*

Compound **14b-1** was synthesized from intermediate **13b** and **9a** in the same method as described for the synthesis of **6a-1**. Yellow solid; yield: 72%; M.p.: 226-228 °C; MS (ESI), m/z (%): 455.4 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO) δ 8.85 (d, *J* = 1.8 Hz, 1H), 8.28 (t, *J* = 5.8 Hz, 1H), 8.09 – 8.04 (m, 2H), 7.86 (s, 1H), 7.80 (d, *J* = 7.2 Hz, 1H), 7.63 (dd, *J* = 14.3, 2.1 Hz, 1H), 7.46 (dd, *J* = 8.8, 2.1 Hz, 1H), 4.78 (td, *J* = 11.5, 5.3 Hz, 1H), 4.19 (t, *J* = 9.0 Hz, 1H), 3.83 – 3.70 (m, 5H), 3.45 (t, *J* = 5.4 Hz, 2H), 3.04 – 2.94 (m, 2H), 2.78 (d, *J* = 13.4 Hz, 2H), 1.85 (s, 3H).

*5.1.10.2 (S)-N-(3-(3-fluoro-4-(5-(thiomorpholin-4-yliminomethyl)pyridin-2-yl)phenyl)-2-oxo-1,3-oxazolidine-5-ylmethyl)acetamide (**14b-2**)*

Compound **14b-2** was synthesized from intermediate **13b** and **9c** in the same method as described for the synthesis of **6a-1**. Yellow solid; yield: 79%; M.p.: 232-234 °C; MS (ESI), m/z (%):458.4 $[M+H]^+$。 ^{1}H NMR (400 MHz, DMSO) δ 8.82 (d, *J* = 1.7 Hz, 1H), 8.28 (t, *J* = 5.8 Hz, 1H), 8.11 – 8.00 (m, 2H), 7.79 (d, *J* = 7.2 Hz, 1H), 7.74 (s, 1H), 7.62 (dd, *J* = 14.3, 2.1 Hz, 1H), 7.46 (dd, *J* = 8.8, 2.1 Hz, 1H), 4.78 (td, *J* = 11.5, 5.3 Hz, 1H), 4.18 (t, *J* = 9.0 Hz, 1H), 3.80 (dd, *J* = 9.1, 6.5 Hz, 1H), 3.59 – 3.54 (m, 4H), 3.45 (t, *J* = 5.5 Hz, 2H), 2.77 – 2.73 (m, 4H), 1.85 (s, 3H).

*5.1.10.3 (S)-N-(3-(3-fluoro-4-(5-((4-tert-butoxycarbonylpiperazin-1-yl)iminomethyl)pyridin-2-yl)phenyl)-2-oxo-1,3-oxazolidine -5-ylmethyl)acetamide (**14b-3**)*

Compound **14b-3** was synthesized from intermediate **13b** and **9e** in the same manner as described for the synthesis of **6a-1**. Yellow solid; yield: 80 %.

*5.1.10.4 (S)-N-(3-(3-fluoro-4-(5-(piperazin-1-yliminomethyl)pyridin-2-yl)phenyl)-2-oxo-1,3-oxazolidine-5-ylmethyl)acetamide (**14b-4**)*

Compound **14b-4** was synthesized from intermediate **14b-3** in the same method as described for the synthesis of **6a-5**. Yellow solid; yield: 77 %; M.p.: 250-252 °C; MS (ESI), m/z (%): 441.5 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO) δ 8.85 (d, *J* = 1.7 Hz, 1H), 8.29 (t, *J* = 5.8 Hz, 1H), 8.10 – 8.04 (m, 2H), 7.86 (s, 1H), 7.81 (d, *J* = 7.3 Hz, 1H), 7.63 (dd, *J* = 14.3, 1.9 Hz, 1H), 7.47 (dd, *J* = 8.8, 2.0 Hz, 1H), 4.79 (td, *J* = 11.5, 5.4 Hz, 1H), 4.19 (t, *J* = 9.0 Hz, 1H), 3.86 (s, 4H), 3.81 (dd, *J* = 9.1, 6.6 Hz,1H), 3.46 (t, *J* = 5.4 Hz, 2H), 3.22 (s, 4H), 1.86 (s, 3H).

*5.1.10.5 (S)-N-(3-(3-fluoro-4-(5-((1-oxothiomorpholin-4-yl)iminomethyl)pyridin-2-yl)phenyl)-2-oxo-1,3-oxazolidin-5-ylmethyl)acetamide (**14b-5**)*

Compound **14b-5** was synthesized from compound **14b-2** in the same method as described for the synthesis of **6a-6**. Yellow solid; yield: 70 %. M.p.: 199-201 °C; MS (ESI), m/z (%): 474.4 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO) δ 8.85 (d, *J* = 1.8 Hz, 1H), 8.28 (t, *J* = 5.8 Hz, 1H), 8.09 – 8.04 (m, 2H), 7.86 (s, 1H), 7.80 (d, *J* = 7.2 Hz, 1H), 7.63 (dd, *J* = 14.3, 2.1 Hz, 1H), 7.46 (dd, *J* = 8.8, 2.1 Hz, 1H), 4.78 (td, *J* = 11.5, 5.3 Hz, 1H), 4.19 (t, *J* = 9.0 Hz, 1H), 3.84 – 3.69 (m, 5H), 3.45 (t, *J* = 5.4 Hz, 2H), 3.04 – 2.93 (m, 2H), 2.80 – 3.76 (m, 2H), 1.85 (s, 3H).

*5.1.10.6 (S)-N-(3-(3-fluoro-4-(5-((1,1-dioxothiomorpholin-4-yl)iminomethyl)pyridin-2-yl)phenyl)-2-oxo-1,3-oxazolidine-5-ylmethyl)acetamide (**14b-6**)*

Compound **14b-6** was synthesized from intermediate **13b** and **9f** in the same method as described for the synthesis of **6a-1**. Yellow solid; yield: 79 %; M.p.: 242-244 °C; MS (ESI), m/z (%): 490.3 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO) δ 8.85 (s, 1H), 8.28 (t, *J* = 5.7 Hz, 1H), 8.09 – 8.04 (m, 2H), 7.86 (s, 1H), 7.81 (d, *J* = 7.6 Hz, 1H), 7.63 (dd, *J* = 14.2, 1.7 Hz, 1H), 7.47 (dd, *J* = 8.8, 1.7 Hz, 1H), 4.82 – 4.74 (m, 1H), 4.19 (t, *J* = 9.0 Hz, 1H), 3.85 (s, 4H), 3.81 (dd, *J* = 9.1, 6.8 Hz, 1H), 3.45 (t, *J* = 5.4 Hz, 2H), 3.21 (s, 4H), 1.85 (s, 3H).

*5.1.11.1 (S)-N-(3-(3-fluoro-4-(6-((4-methoxyformylpiperazin-1-yl)iminomethyl)pyridin-3-yl)phenyl)-2-oxo-1,3-oxazolidine-5-ylmethyl)acetamide (**15a-1**)*

Methyl chloroformate (0.043 g, 0.45 mmol) was added dropwise to a well-stirred solution of DIPEA (0.07 g, 0.54 mmol) and compound **14a-5** (0.2 g, 0.45 mmol) in DMF (10 mL), then the mixture was stirred at 25 ºC for 2 h. After completion of the reaction monitored by TLC, water (15 mL) was added and the resulting solid was collected by filtration and purified by column chromatography to give the target intermediate **15a-1** as a white solid in 85 % yield. M.p.: 240-242 °C; MS (ESI), m/z (%): 499.3 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO) δ 8.70 (s, 1H), 8.28 (s, 1H), 7.95 (d, *J* = 7.6 Hz, 1H), 7.85 (d, *J* = 8.2 Hz, 1H), 7.72 – 7.63 (m, 3H), 7.45 (d, *J* = 8.0 Hz, 1H), 4.82 – 4.74 (m, 1H), 4.18 (t, *J* = 8.8 Hz, 1H), 3.80 (dd, *J* = 7.7, 7.1 Hz, 1H), 3.64 (s, 3H), 3.58 (s, 4H), 3.45 (s,2H), 3.24 (s, 4H), 1.85 (s, 3H).

*5.1.11.2 (S)-N-(3-(3-fluoro-4-(6-((4-propionylpiperazin-1-yl)iminomethyl)pyridin-3-yl)phenyl)-2-oxo-1,3-oxazolidine-5-ylmethyl)acetamide (**15a-2**)*

Compound **15a-2** was synthesized from compound **14a-6** and propionyl chloride in the same method as described for the synthesis of **15a-1**. White solid; yield: 82 %; M.p.: 225-228 °C; MS (ESI), m/z (%): 519.4 $[M+Na]^+$. ^{1}H NMR (400 MHz, DMSO) δ 8.70 (s, 1H), 8.28 (s, 1H), 7.95 (d, *J* = 7.7 Hz, 1H), 7.85 (d, *J* = 8.2 Hz, 1H), 7.70 – 7.63 (m, 3H), 7.45 (d, *J* = 8.2 Hz, 1H), 4.78 (s, 1H), 4.18 (t, *J* = 8.7 Hz, 1H), 3.87 – 3.75 (m, 1H), 3.65 (s, 4H), 3.45 (s, 2H), 3.27 (s, 2H), 3.21 (s, 2H), 2.38 (dd, *J* = 14.4, 7.1 Hz, 2H), 1.85 (s, 3H), 1.01 (t, *J* = 7.2 Hz,

3H).

*5.1.11.3 (S)-N-(3-(3-fluoro-4-(6-((4-benzoylpiperazin-1-yl)iminomethyl)pyridin-3-yl)phenyl)-2-oxo-1,3-oxazolidine-5-ylmethyl)acetamide (**15a-3**)*

Compound **15a-3** was synthesized from compound **14a-6** and benzoyl chloride in the same method as described for the synthesis of **15a-1**. White solid; yield: 91 %; M.p.: 201-204 °C; MS (ESI), m/z (%): 545.4 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO) δ 8.71 (s, 1H), 8.28 (t, *J* = 5.6 Hz, 1H), 7.95 (s, 1H), 7.86 (d, *J* = 8.4 Hz, 1H), 7.72 – 7.61 (m, 3H), 7.50 – 7.43 (m, 6H), 4.84 – 4.72 (m, 1H), 4.18 (t, *J* = 8.9 Hz, 1H), 3.93 – 3.70 (m, 3H), 3.47 (dd, *J* = 24.8, 19.5 Hz, 4H), 3.34 – 3.16 (m, 4H), 1.84 (s, 3H).

*5.1.12.1 (S)-N-(3-(2-fluoro-3'-hydroxy-4'-((4-(methoxyformyl)piperazin-1-yl)iminomethyl)biphenyl-4-yl)-2-oxo-1,3-oxazolidin-5-ylmethyl)acetamide (**16-1**)*

Compound **16-1** was synthesized from compound **5b-5** and methyl chloroformate in the same method as described for the synthesis of **15a-1**. White solid; yield: 75 %; M.p.: 208-210 °C; MS (ESI), m/z (%): 514.2 $[M+H]^+$. ^{1}H NMR (400 MHz, DMSO) δ 8.26 (t, *J* = 5.5 Hz, 1H), 7.78 (d, *J* = 8.2 Hz, 1H), 7.67 – 7.59 (m, 3H), 7.51 (d, *J* = 8.2 Hz, 1H), 7.43 (d, *J* = 8.0 Hz, 2H), 4.77 (td, *J* = 11.4, 5.5 Hz, 1H), 4.17 (t, *J* = 8.9 Hz, 1H), 3.84 (s, 3H), 3.82 – 3.76 (m, 1H), 3.56 (s, 4H), 3.44 (t, *J* = 5.2 Hz, 2H), 3.15 (s, 4H), 1.84 (s, 3H).

*5.1.12.2 (S)-N-(3-(2-fluoro-3'-hydroxy-4'-(((4-propionyl)piperazin-1-yl)iminomethyl)biphenyl-4-yl)-2-oxo-1,3-oxazolidin-5-ylmethyl)acetamide (**16-2**)*

Compound **16-2** was synthesized from compound **5b-5** and propionyl chloride in the same method as described for the synthesis of **15a-1**. White solid; yield: 77 %; M.p.: 219-221 °C; MS (ESI), m/z (%): 512.3 $[M+H]^+$.^{1}H NMR (400 MHz, DMSO) δ 11.29 (s, 1H), 8.27 (t, *J* = 5.6 Hz,1H), 8.07 –8.01 (m, 1H), 7.66 – 7.38 (m, 5H), 7.11 – 7.02 (m, 1H), 4.80 – 4.73 (m, 1H), 4.17 (dd, *J* = 9.0, 6.6 Hz, 1H), 3.87 – 3.75 (m, 3H), 3.63 (s, 3H), 3.57 (d, *J* = 4.3 Hz, 4H), 3.44 (t, *J* = 5.1 Hz, 2H), 3.15 (d, *J* = 4.2 Hz, 4H), 1.84 (s, 3H).

5.2. Pharmacology

5.2.1. In vitro antibacterial activity

Isolates were identified on MicroScan Walk Away 96 system and antimicrobial susceptibilities were determined by the broth liquid microdilution method according to the Clinical Laboratory Standards Institute (CLSI) guidelines (4). Minimum inhibitory concentration (MIC) was defined as the lowest antimicrobial concentration that inhibited bacterial growth totally. The conventional antimicrobial agents used here were penicillin, gentamycin, rifampicin, ciprofloxacin, levofloxacin, moxifloxacin, trimethoprim-sulfamethoxazole, clindamycin, erythromycin, linezolid, vancomycin, quinupristin-dalfopristin and tetracyclin. MRSA, VRE or LREF strains were identified according to CLSI guidelines (5). *S.aureus* ATCC29213 was used as the control.

5.2.2. In vitro inhibition assay for MAO-A

The assay to test the inhibition of compounds against human recombinant MAO-A (Active Motif, 31502) enzyme was displayed by Shanghai ChemPartner Co., LTD. using MAO-Glo™ assay kit (Promega Corporation,V1402) with modifications of the instruction manual according to preliminary optimizations. Briefly, dose response curves or indicated concentrations of compounds were made in DMSO. A 200-nL aliquot of the compound solution was transferred into a 384-well plate (Perkin Elmer, 6007299) by Echo® 550 (the final fraction of DMSO was 1%). Compound was incubated with 10-uL of recombinant MAO-A solutions at room temperature

for 15 min, followed by adding 10-uL of luciferin derivative substrate to initiate the reaction. Reactions were incubated for 60 min at room temperature and reporter luciferase detection reagent of 20 μL was added and incubated with each reaction for 20 min to produce luminescence. Relative light units (RLU) were detected using EnVision Multilabel Plate Reader. For 100% inhibition control (Min), 1× assay buffer was used instead of MAO-A enzyme solution. And for no inhibition control (Max), DMSO was used instead of compound DMSO solution. The inhibition percentage in the presence of the compound was calculated according to the equation, Percent inhibition = (Max – ignal)/ (Mx – Min)*100%. Fit the data in GrphaPad Prism V5.0 software to obtain IC_{50} values using equation, Y=Bottom + (Top – Bottom)/(1 + 10^((LogIC_{50} – X)*HillSlope), where Y stands for inhibition percentage and X stands for compound concentration.

5.2.3. Cytotoxicity

The target compounds were screened in HepG2 cells by a standard MTT assay in vitro. HepG2 cells were cultured in RPMI1640 medium supplemented with 10% fetal bovine serum (FBS). Approximately 4×10^4 cells, suspended in MEM medium, were plated onto each well of a 96-well plate and incubated in 5% CO_2 at 37 □ for 24 h. The tested compounds were added to the culture medium at the indicated final concentrations and the cell cultures were continued for 48h. Fresh MTT was added to each well at a final concentration of 0.5 mg/mL and incubated with cells at 37 □ for 4 h. The formazan crystals were dissolved in 150 μL DMSO per each well, and the absorbency at 492 nm (for the absorbance of MTT formazan) and 630 nm (for the reference wavelength) was measured with the ELISA reader. All of the compounds were tested three times in each of the cell lines. The results expressed as IC_{50} (inhibitory concentration of 50%) were the mean ± SD and were calculated by using the Ghaphapad prism 6.02.

5.2.4. Microsomal stability

30 μL of 1.5 μM compounds solution containing 0.75 mg/mL microsomes solution were dispensed to the assay plates designated for different time points (0-, 5-, 15-, 30-, 45-min) on ice. For 0-min, 135 μL of acetonitrile containing internal standard was added to the wells of 0-min plate and afterwards 15 μL of NADPH stock solution (6 mM) was added. Then, pre-incubated all other plates at 37 ºC for 5 min. 15 μL of NADPH stock solution (6 mM) was added to the plates to start the reaction and timing. At 5-min, 15-min, 30-min, and 45-min, 135 μL of acetonitrile containing internal standard was added to the wells of corresponding plates, respectively, to stop the reaction. After quenching, the plates at the vibrator (IKA, MTS 2/4) were shaked for 10 min (600 rpm/min) and then centrifuged at 5594g for 15 min (Thermo Multifuge ×3R). 50 μL of the supernatant from each well was transferred into a 96-well sample plate containing 50 lL of ultra-pure water (Millipore, ZMQS50F01) for LC/MS analysis. All incubations were performed in duplicate. In vitro intrinsic clearance (Cl_{int}) was calculated from half-life ($T_{1/2}$) of the compounds disappearance, which was determined by the slope (k) of log-linear regression analysis from the concentration versus time profiles, i.e., $T_{1/2} = \ln(2)/k$.

5.3. Molecular superposition

The Molecular superposition studies were performed with Forge 3.0 (v10.4.2). The crystal structure of the oxazolidinone antibiotic linezolid bound to the 50S ribosomal subunit (PDB code: 3CPW) was obtained from the RCSB Protein Data Bank (*http://www.rcsb.org/pdb/*). Compound **14a-7** was drawn with Chemdraw. Before molecular superposition, linezolid molecular was obtained from the crystal structure of 3CPW using Discovery Studio 3.0. We selected linezolid molecules as a training set to build the model while the compound **14a-7** served

as test set. Finally, they were docked into the binding site using Discovery Studio 3.0.

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References

[1] R.F. Service, Antibiotics that resist resistance, Science. 270 (1995) 724–727.

[2] B. Spellberg, R. Guidos, D. Gilbert, J. Bradley, H.W. Boucher, W.M. Scheld, J.G. Bartlett, J.J. Edwards, The epidemic of antibioticresistant infections: a call to action for the medical community from the Infectious Diseases Society of America, Clin. Infect. Dis. 46 (2008) 155-164.

[3] Y. Fukuda, New approaches to overcoming bacterial resistance, Drugs Future. 34 (2009) 127-136.

[4] R.M. Klevens, M.A. Morrison, J. Nadle, S. Petit, K. Gershman, S. Ray, L.H. Harrison, R. Lynfield, G. Dumyati, J.M. Townes, A.S. Craig, E.R. Zell, G.E. Fosheim, L.K. McDougal, R.B. Carey, S.K. Fridkin, Invasive methicillin resistant Staphylococcus aureus infections in the United States, JAMA. 298 (2007) 1763-1771.

[5] D.H. Williams, B. Bardsley, The vancomycin group of antibiotics and the fight against resistant bacteria, Angew. Chem. Int. Ed. 38 (1999) 1173-1193.

[6] K. Bush, P. Courvalin, G. Dantas, J. Davies, B. Eisenstein, P. Huovinen, G.A. Jacoby, R. Kishony, B.N. Kreiswirth, E. Kutter, S.A. Lerner, S. Levy, K. Lewis, O. Lomovskaya, J.H. Miller, S. Mobashery, L.J. Piddock, S. Projan, C.M. Thomas, A. Tomasz, P.M. Tulkens, T.R. Walsh, J.D. Watson, J. Witkowski, W. Witte, G. Wright, P. Yeh, H.I. Zgurskaya, Tackling antibiotic resistance, Nat. Rev. Microbiol. 9 (2011) 894-896.

[7] D.N. Wilson, Ribosome-targeting antibiotics and mechanisms of bacterial resistance, Nat. Rev. Microbiol. 12 (2014) 35-48.

[8] J. Poehlsgaard, S. Douthwaite, The bacterial ribosome as a target for antibiotics, Nat. Rev. Microbiol. 3 (2005) 870-881.

[9] T. Tenson, A. Mankin, Antibiotics and the ribosome, Mol. Microbiol. 59 (2006) 1664-1667.

[10] S.J. Brickner, M.R. Barbachyn, D.K. Hutchinson, P.R. Manninen, Linezolid (ZYVOX), the first member of a completely new class of antibacterial agents for treatment of serious gram-positive infections, J. Med. Chem. 51 (2008) 1981-1990.

[11] C.M. Perry, B. Jarvis, Linezolid: a review of its use in the management of serious Gram-positive infections, Drugs. 61 (2001) 525-551.

[12] J.A. Ippolito, Z.F. Kanyo, D. Wang, F.J. Franceschi, P.B. Moore, T.A. Steitz, E.M. Duffy, Crystal structure of oxazolidinone antibiotic linezolid bound to 50S ribosomal subunit, J. Med. Chem. 51 (2008) 3353-3356.

[13] G. Morales, J.J. Picazo, E. Baos, F.J. Candel, A. Arribi, B. Pelaez, R. Andrade, M.A. de la Torre, J. Fereres, M. Sanchez-García, Resistance to linezolid is mediated by the cfr gene in the first report of an outbreak of linezolidresistant Staphylococcus aureus, Clin. Infect. Dis. 50 (2010) 821-825.

[14] F. Franceschi, E.M. Duffy, Structure-based drug design meets the ribosome, Biochem. Pharmacol. 71(2006) 1016-1025.

[15] S.B. Li, X.F. Cheng, Y.H. Zhou, X. Zhen, Sparsomycin-Linezolid Conjugates Can Promote Ribosomal Translocation, Chembiochem. 12(2011) 2801-2806.

[16] J. Zhou, A. Bhattacharjee, S. Chen, Y. Chen, E. Duffy, J. Farmer, J. Goldberg, R. Hanselmann, J.A. Ippolito, R. Lou, A. Orbin, A. Oyelere, J. Salvino, D. Springer, J. Tran, D. Wang, Y. Wu, G. Johnson, Design at the atomic level: design of biaryloxazolidinones as potent orally active antibiotics, Bioorg. Med. Chem. Lett. 18 (2008) 6175-6178.

[17] J. Zhou, A. Bhattacharjee, S. Chen, Y. Chen, E. Duffy, J. Farmer, J. Goldberg, R. Hanselmann, J.A. Ippolito, R. Lou, A. Orbin, A. Oyelere, J. Salvino, D. Springer, J. Tran, D. Wang, Y. Wu, G. Johnson, Design at the atomic level: generation of novel hybrid biaryloxazolidinones as promising new antibiotics, Bioorg. Med. Chem. Lett. 18 (2008) 6179-6183.

[18] F. Reck, F. Zhou, C.J. Eyermann, G. Kern, D. Carcanague, G. Ioannidis, R. Illingworth, G. Poon, M.B. Gravestock, Novel Substituted (Pyridin-3-yl)phenyloxazolidinones: Antibacterial Agents with Reduced Activity against Monoamine Oxidase A and

Increased Solubility, J. Med. Chem. 50 (2007) 4868-4881.

[19] F.E. Boyer, J.V.N.V. Prasad, A.L. Choy, L. Chupak, M.R. Dermyer, Q. Ding, M.D. Huband, W. Jiao, T. Kaneko, V. Khlebnikov, J.Y. Kim, M.S. Lall, S.N. Maiti, N.K. Romero, X. Wu, Synthesis and SAR of novel conformationally-restricted oxazolidinones possessing Gram-positive and fastidious Gram-negative antibacterial activity. Part 1: Substituted pyrazoles, Bioorg. Med. Chem. Lett. 17 (2007) 4694-4698.

[20] T. Komine, A. Kojima, Y. Asahina, T. Saito, H. Takano, T. Shibue, Y. Fukuda, Synthesis and Structure-Activity Relationship Studies of Highly Potent Novel Oxazolidinone Antibacterials, J. Med. Chem. 51 (2008) 6558-6562.

[21] W.B. Im, S.H. Choi, J.Y. Park, S.H. Choi, J. Finn, S.H. Yoon, Discovery of torezolid as a novel 5-hydroxymethyl-oxazolidinone antibacterial agent, Eur. J. Med. Chem. 46 (2011) 1027-1039.

[22] T. Yang, G. Chen, Z. Sang, Y. Liu, X. Yang, Y. Chang, H. Long, W. Ang, J. Tang, Z. Wang, G. Li, S. Yang, J. Zhang, Y. Wei, Y. Luo, Discovery of a teraryl oxazolidinone compound (S)-N-((3-(3-fluoro-4-(4-(pyridin-2-yl)-1H-pyrazol-1-yl)phenyl)-2-oxooxazolidin-5-yl)-methyl)acetamide phosphate as a novel antimicrobial agent with enhanced safety profile and efficacies, J. Med. Chem. 58 (2015) 6389-6409.

[23] Y.C. Wu, X.D. Ding, L. Ding, Y.S. Zhang, L. Cui, L. Sun, W. Li, D. Wang, Y.F. Zhao, Synthesis and antibacterial activity evaluation of novel biaryloxazolidinone analogues containing a hydrazone moiety as promising antibacterial agents, Eur. J. Med. Chem. 158 (2018) 247-258.

[24] H. Suzuki, I. Utsunomiya, K. Shudo, T. Fujimura, M. Tsuji, I. Kato, T. Aoki, A. Ino, T. Iwaki, Potent oxazolidinone antibacterials with heteroaryl C-ring substructure, ACS Med. Chem. Lett. 4 (2013) 1074-1078.

[25] M.S. Deshmukh, N. Jain, Design synthesis, and antibacterial evaluation of oxazolidinones with fused heterocyclic C-ring substructure, ACS Med. Chem. Lett. 8 (2017) 1153-1158.

Highlights

1, Novel biaryloxazolidinone analogues were designed to improve the metabolic stability.

2, Compound **14a-7** exhibited a MIC value of 0.125 μg/mL against *S.aureus*.

3, Compound **14a-7** was stable in human liver microsome.

4, Compound **14a-7** exhibited lower inhibitory activity against human MAO-A compared to linezolid.