European Journal of Medicinal Chemistry 44 (2009) 3903–3908

Contents lists available at ScienceDirect

European Journal of Medicinal Chemistry

journal homepage: http://www.elsevier.com/locate/ejmech

Original article

Synthesis, structure and structure–activity relationship analysis of 3-*tert*-butoxycarbonyl-2-arylthiazolidine-4-carboxylic acid derivatives as potential antibacterial agents

Zhong-Cheng Song, Gao-Yuan Ma, Peng-Cheng Lv, Huan-Qiu Li, Zhu-Ping Xiao, Hai-Liang Zhu*

Institute of Functional Biomolecules, State Key Laboratory of Pharmaceutical Biotechnology, Nanjing University, Nanjing 210093, People's Republic of China

ARTICLE INFO

Article history:
Received 3 June 2008
Received in revised form
28 March 2009
Accepted 2 April 2009
Available online 14 April 2009

Keywords:
Thiazolidine
Antibacterial activity
P. aeruginosa
Structure–activity relationship (SAR)

ABSTRACT

Nine 2-arylthiazolidine-4-carboxylic acid derivatives and nine 3-*tert*-butoxycarbonyl-2-arylthiazolidine-4-carboxylic acid derivatives were synthesized to screen for their antibacterial activities. Compounds **5**, **14**–**18** were first reported. Their chemical structures were clearly determined by ^{1}H NMR, ^{13}C NMR, ESI mass spectra and elemental analyses, coupled with one selected single-crystal structure. All the compounds were assayed for antibacterial activities against two Gram-positive bacterial strains (*Bacillus subtilis* ATCC 6633 and *Staphylococcus aureus* ATCC 6538) and two Gram-negative bacterial strains (*Escherichia coli* ATCC 35218 and *Pseudomonas aeruginosa* ATCC 13525) by MTT method. Most of the 3-*tert*-butoxycarbonyl-2-arylthiazolidine-4-carboxylic acid derivatives exhibited better antibacterial activities against the four bacterial strains than relative 2-arylthiazolidine-4-carboxylic acid derivatives. Compound (2*RS*,4*R*)-3-(*tert*-butoxycarbonyl)-2-(5-fluoro-2-hydroxyphenyl)thiazolidine-4-carboxylic acid (**14**) showed powerful antibacterial activities against *P. aeruginosa* with IC_{50} value of 0.195 μg/mL, which was superior to the positive controls Penicillin G and Kanamycin B, respectively. On the basis of the biological results, structure–activity relationships were discussed.

1. Introduction

Currently, heterocyclic compounds have been extensively studied due to their important properties and applications. Among these compounds, thiazole and thiazolidine derivatives have become especially noteworthy in recent years [1,2]. Additionally, thiazole and thiazolidine moieties have played an increasingly important role in many different types of chemical structures, generating by structure modification a series of compounds with broad-spectrum bioactivities [3]. Moreover, thiazolidine derivatives can be antibacterial agents to cure patients infected by some pathogenic bacteria [4–7]. In previous studies [8,9], it was reported that 2-substituted 3-*tert*-butoxycarbonyl-thiazolidine-(4*R*)-carboxylic acid (TBTCA) can protect against the hepatotoxicity elicited by high doses of acetaminophen in mice. Thus, it is very significant to study on these types of thiazolidine derivatives. However, to the best of our knowledge, it has not been reported that thiazolidine derivatives like TBTCA had better antibacterial activities against the four bacteria, Gram-positive (*Bacillus subtilis* ATCC 6633 and *Staphylococcus aureus* ATCC 6538) and Gram-negative (*Pseudomonas aeruginosa* ATCC 13525 and *Escherichia coli* ATCC 35218), which often caused infection in burned patients. Herein, various TBTCA derivatives with modifications were synthesized in order to find new antibacterial agents. Our current work highlights antibacterial activity evaluation of a series of synthesized TBTCA against *B. subtilis* as well as similarity comparison of antibacterial activities against *S. aureus*, *P. aeruginosa* and *E. coli* strains. Furthermore, the minimal inhibitory concentration (MIC) of these TBTCA derivatives with antibacterial activities against the above four bacteria and structure–activity relationship (SAR) were also discussed.

2. Results and discussion

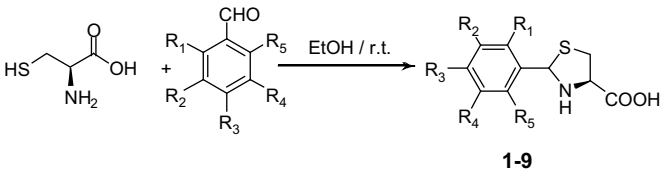
2.1. Chemistry

Nine 2-arylthiazolidine-4-carboxylic acid derivatives were obtained by reaction of L-cysteine with various aldehydes, as described in Scheme 1 [3,10,11].

Initial SAR studies were performed by modification of the parent compound to determine if any of the subunits displayed antibacterial activity. Based on this consideration, we made related modifications to TCA as shown in Fig. 1.

* Corresponding author. Tel.: +86 25 8359 2572; fax: +86 25 8359 2672.
E-mail address: zhuhl@nju.edu.cn (H.-L. Zhu).

0223-5234/$ – see front matter © 2009 Elsevier Masson SAS. All rights reserved.
doi:10.1016/j.ejmech.2009.04.014



Scheme 1. Syntheses of substituted 2-arylthiazolidine-4-carboxylic acid (TCA) **1–9**.

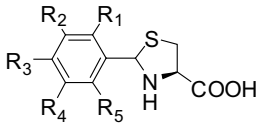
In order to improve the solubility, we have also carried out the next step 3-*tert*-butoxycarbonyl reaction. The corresponding nine TBTCA were obtained by the reaction of *tert*-butyloxycarbonyl anhydride (BOC_2O) with compounds **1–9**, as described in Scheme 2 [12] and Fig. 2.

2.2. Crystal structure of '2*R*, 4R' isomer of two TBTCA (*12*)

The '2*R*, 4*R*' isomer of TBTCA (**12**) was characterized by single-crystal X-ray determinations. It crystallizes in the monoclinic space group $P2_1$, and the crystallographic data are presented in Table 1. The selected bond distances, bond angles and torsion angles are given in Table 2. As shown in Table 2, the bond lengths of O2═C5 (1.233(6) Å) and O3═C6 (1.193(6) Å) conform to the values for carbon–oxygen double bonds. Generally, a typical C–O single bond is around 1.40–1.42 Å. However, the short bond C6–O4 (1.315(7) Å) in the carboxyl group comes from the resonance with C6–O3 (1.193(6) Å) and the very long bond C4–O1 (1.482(7) Å) might come from a steric hindrance with the bulky *tert*-butyl-group. The C–S (1.802(6)–1.844(5) Å) single bonds are in the normal ranges. A normal C–N-single bond is around 1.45 Å, so N–C7 (1.446(6) Å) and N–C9 (1.474(6) Å) might be considered as normal single bonds; however, N–C5 (1.360(6) Å) is significantly shortened due to the interaction with the two C5–O bonds. The C5–N–C7, C5–N–C9 and C7–N–C9 bond angles are close to 120°, while the N–C(9)–S and C(8)–S–C(9) bond angles are 103.0(3) and 90.7(3)°. The C(5)–N–C(9)–C(10), C(5)–N–C(9)–S and C(8)–S–C(9)–C(10) torsion angles in the molecule are 58.4(6), 179.3(4) and 155.1(4)°, respectively. The presence of the S atom allowed the unambiguous assignment of the absolute configuration on the basis of the refined Flack parameter value (−0.09 (19)) [13–15]. Molecular structure (thermal ellipsoids at the 35% probability level) of '2*R*, 4*R*' isomer of compound **12** shown in Fig. 3 revealed the 2*R*, 4*R* absolute configuration. In the crystal structure of the compound, molecules are linked through intermolecular hydrogen bonds of O4–H4A···O2 and C9–H9A···O3 (Table 3), forming chains running along the *a* axis. The chains are further linked through intermolecular hydrogen bonds of C16–H16A···O3 at the *b* axis, forming layers parallel to the *ab* plane, as shown in Fig. 4.

2.3. Biological activity

All the TCA and TBTCA derivatives were screened for antibacterial activities *in vitro* against two Gram-positive bacterial strains *B. subtilis* and *S. aureus*, and two Gram-negative bacterial strains *E. coli* and *P. aeruginosa* by MTT method [16,17]. The IC_{50}s of the TCA derivatives against the four bacteria are presented in Tables 4 and 5.



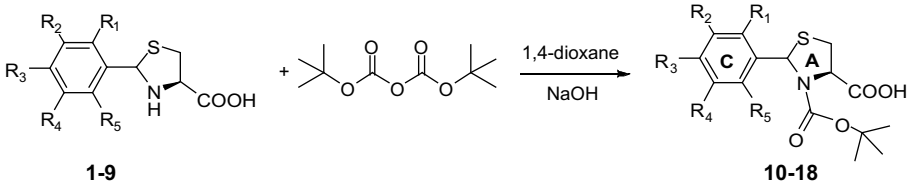
	R1	R2	R3	R4	R5
1	H	H	H	H	H
2	H	H	OH	H	H
3	H	H	OMe	H	H
4	H	H	Br	H	H
5	OH	H	H	F	H
6	OH	H	H	Cl	H
7	OH	H	H	Br	H
8	OH	Br	H	Br	H
9	H	$C(CH_3)_3$	OH	$C(CH_3)_3$	H

Fig. 1. Chemical structures of TCA **1–9**.

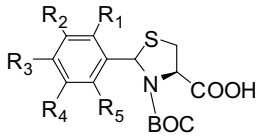
It can be seen from Table 4 that many of these TCA derivatives were very effective and selective for antibacterial activities *in vitro* against the four bacteria used in the biological experiments. As shown in Table 4, the synthesized thiazolidine derivative (compound **5**) exhibited excellent high selective antibacterial activity against *P. aeruginosa* with IC_{50} as low as 1.5625 μg/mL, however, exhibited low antibacterial activities against other three bacteria (*B. subtilis*, *S. aureus* and *E. coli*). Furthermore, inspection of the chemical structure of the synthesized TCA derivatives (Fig. 1) with phenyl functional group part suggested that they could be divided into three types by the number of substituents: a single substituent, two substituents and three substituents. Initial SAR studies were performed by modification of the parent compound to determine how the substituents of the subunits affected the antibacterial activities. Compounds bearing two substituents exhibited higher antibacterial activities than the compounds containing a single substituent or three substituents, as shown in Table 4.

It can be found that the results of antibacterial activities of TCA derivatives bearing two substituents against the four bacteria were very selective and complicated. According to the experiment results (Table 4), the antibacterial activity against the four bacteria is in accordance with the order F > Cl > Br this order at the 5-position on the phenyl group.

It can be found that all TBTCA derivatives exhibited excellent results of antibacterial activities (Table 5) compared with those of



Scheme 2. Syntheses of TBTCA **10–18**.



10-18

	R1	R2	R3	R4	R5
10	H	H	H	H	H
11	H	H	OH	H	H
12	H	H	OMe	H	H
13	H	H	Br	H	H
14	OH	H	H	F	H
15	OH	H	H	Cl	H
16	OH	H	H	Br	H
17	OH	Br	H	Br	H
18	H	$C(CH_3)_3$	OH	$C(CH_3)_3$	H

Fig. 2. Chemical structures of TBTCA **10**–**18**.

TCA derivatives (Table 4). This may be due to the introduction of *tert*-butyloxycarbonyl, destructing the intermolecular hydrogen bond of the original carboxylic acid, making an increasing solubility of the compounds and resulting in the increasing antibacterial activity. Compound **14** exhibited higher antibacterial activity against *P. aeruginosa* with IC_{50} value of 0.195 μg/mL, which may be attributed to the electron-withdrawing inductive effect and the unique biological activity of F atom substituent. Because of the stronger bond energy of C–F (477 KJ/mol) compared with C–H (398 KJ/mol) and smaller radius (van der Waals radius nearly to H atom radius), F atom often links the vulnerable attacked metabolism site to prevent the metabolism of the bacteria.

It is well known that *P. aeruginosa* is a Gram-negative, aerobic, rod-shaped bacterium with unipolar motility [18,19] that grows in soil, marshes, and coastal marine habitats, as well as on plant and animal tissues [20], and is commonly isolated from clinical specimens. An opportunistic human pathogen [21–24], *P. aeruginosa* infection occurring in any part of the human body and organizations, is common in parts of burns or trauma, ear, cornea, urethra

Table 1
Crystal structure data for '2*R*, 4*R*' isomer of compound **12**.

Compound	'2*R*, 4*R*' isomer of **12**
Formula	$C_{16}H_{21}NO_5S$
M_r	339.40
Crystal size (mm^3)	0.40 × 0.30 × 0.20
Crystal system	Monoclinic
Space group	$P2_1$
a (Å)	6.490(2)
b (Å)	11.019(2)
c (Å)	12.296(3)
α (°C)	90.00
β (°C)	95.53(3)
γ (°C)	90.00
V ($Å^3$)	875.2(3)
Z	2
D_c (g/cm^3)	1.288
μ (mm^{-1})	0.208
F (000)	360
θ Range (°)	1.66/25.96
Index range (*h*, *k*, *l*)	−7/7, 0/13, 0/15
Reflections collected/unique	1961/1798
$R_{(int)}$	0.0144
Observed reflections $I > 2\sigma(I)$	1498
Min. and max. transmission	0.9595 and 0.9213
Data/restraints/parameters	1798/1/202
Goodness-of-fit on F^2	1.029
R_1, wR_2 [$I > 2\sigma(I)$]	0.0549/0.1329
R_1, wR_2	0.0691/0.1483
Absolute structure parameter	−0.09(19)
Large diff. peak and hole (e $Å^{-3}$)	0.226/−0.244

Table 2
Selected bond lengths (Å), bond angles (°) and torsion angles(°) for '2*R*, 4*R*' isomer of compound **12**.

S1–C(8)	1.802(6)	O(3)–C(6)	1.193(6)
S1–C(9)	1.844(5)	O(4)–C(6)	1.315(7)
N–C(5)	1.360(6)	O(5)–C(13)	1.355(8)
N–C(7)	1.446(6)	O(5)–C(16)	1.400(9)
N–C(9)	1.474(6)	C(6)–C(7)	1.515(7)
O(1)–C(5)	1.321(7)	C(7)–C(8)	1.525(7)
O(1)–C(4)	1.482(7)	C(9)–C(10)	1.499(8)
O(2)–C(5)	1.233(6)		
C(8)–S–C(9)	90.7(3)	O(1)–C(5)–N	110.7(4)
C(5)–N–C(7)	122.8(4)	O(3)–C(6)–O(4)	123.2(5)
C(5)–N–C(9)	118.8(4)	O(3)–C(6)–C(7)	122.4(5)
C(7)–N–C(9)	117.8(4)	O(4)–C(6)–C(7)	114.3(4)
C(5)–O(1)–C(4)	121.5(5)	N–C(7)–C(6)	114.8(4)
O(1)–C(4)–C(2)	109.0(6)	N–C(7)–C(8)	105.6(4)
O(1)–C(4)–C(1)	110.8(5)	C(6)–C(7)–C(8)	110.6(4)
O(1)–C(4)–C(3)	101.2(5)	C(7)–C(8)–S	104.0(4)
C(13)–O(5)–C(16)	118.0(6)	N–C(9)–C(10)	117.8(4)
O(2)–C(5)–O(1)	126.0(5)	N–C(9)–S	103.0(3)
O(2)–C(5)–N	123.3(5)	C(10)–C(9)–S	109.7(3)
C(4)–O(1)–C(5)–N	177.1(5)	C(6)–C(7)–C(8)–S	−84.8(5)
C(7)–N–C(5)–O(2)	−175.7(5)	C(9)–S–C(8)–C(7)	−40.5(4)
C(9)–N–C(5)–O(2)	−5.0(8)	C(5)–N–C(9)–C(10)	58.4(6)
C(7)–N–C(5)–O(1)	4.7(7)	C(7)–N–C(9)–C(10)	−130.4(5)
C(9)–N–C(5)–O(1)	175.4(4)	C(5)–N–C(9)–S	179.3(4)
C(5)–N–C(7)–C(6)	−86.8(6)	C(7)–N–C(9)–S	−9.6(5)
C(9)–N–C(7)–C(6)	102.4(5)	C(8)–S–C(9)–N	28.8(4)
C(5)–N–C(7)–C(8)	151.1(5)	C(8)–S–C(9)–C(10)	155.1(4)
C(9)–N–C(7)–C(8)	−19.8(6)	N–C(9)–C(10)–C(15)	−141.6(5)
O(3)–C(6)–C(7)–N	−169.7(5)	S–C(9)–C(10)–C(15)	101.1(5)
O(4)–C(6)–C(7)–N	10.9(6)	N–C(9)–C(10)–C(11)	42.6(7)
N–C(7)–C(8)–S	40.0(5)	S–C(9)–C(10)–C(11)	−74.8(5)

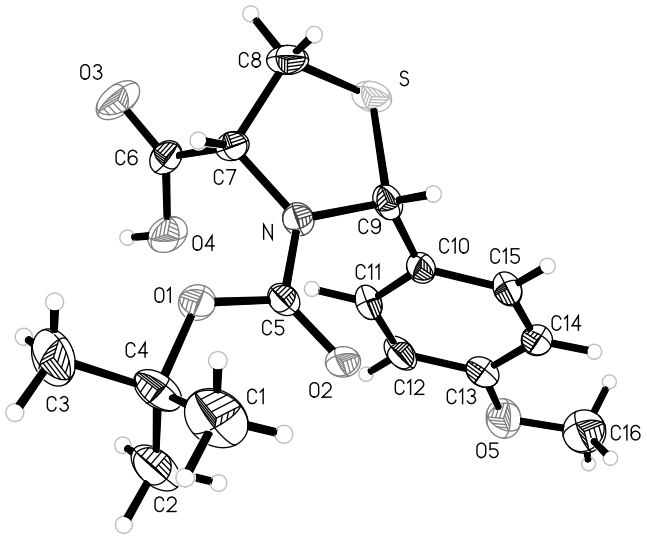


Fig. 3. Molecular structure (thermal ellipsoids at the 35% probability level) of '2*R*, 4*R*' isomer of compound **12**.

Table 3
Hydrogen Bonds for '2*R*, 4*R*' isomer of compound **12** ((Å) and (°)).

D–H···A	d(D–H)	d(H···A)	d(D···A)	<(DHA)
O(4)–H(4A)···O(2)[a]	0.82	1.81	2.629(5)	178.9
C(1)–H(1B)···O(2)	0.96	2.39	2.976(9)	119.3
C(2)–H(2A)···O(2)	0.96	2.49	3.060(8)	117.7
C(8)–H(8B)···O(3)	0.97	2.53	2.908(7)	102.9
C(9)–H(9A)···O(3)[b]	0.98	2.50	3.303(7)	139.2
C(11)–H(11A)···O(4)	0.93	2.55	3.421(7)	155.5
C(16)–H(16A)···O(3)[c]	0.96	2.48	3.031(10)	116.7

Symmetry transformations used to generate equivalent atoms:
[a] $x+1, y, z$.
[b] $x-1, y, z$.
[c] $x-1, y-1, z$.

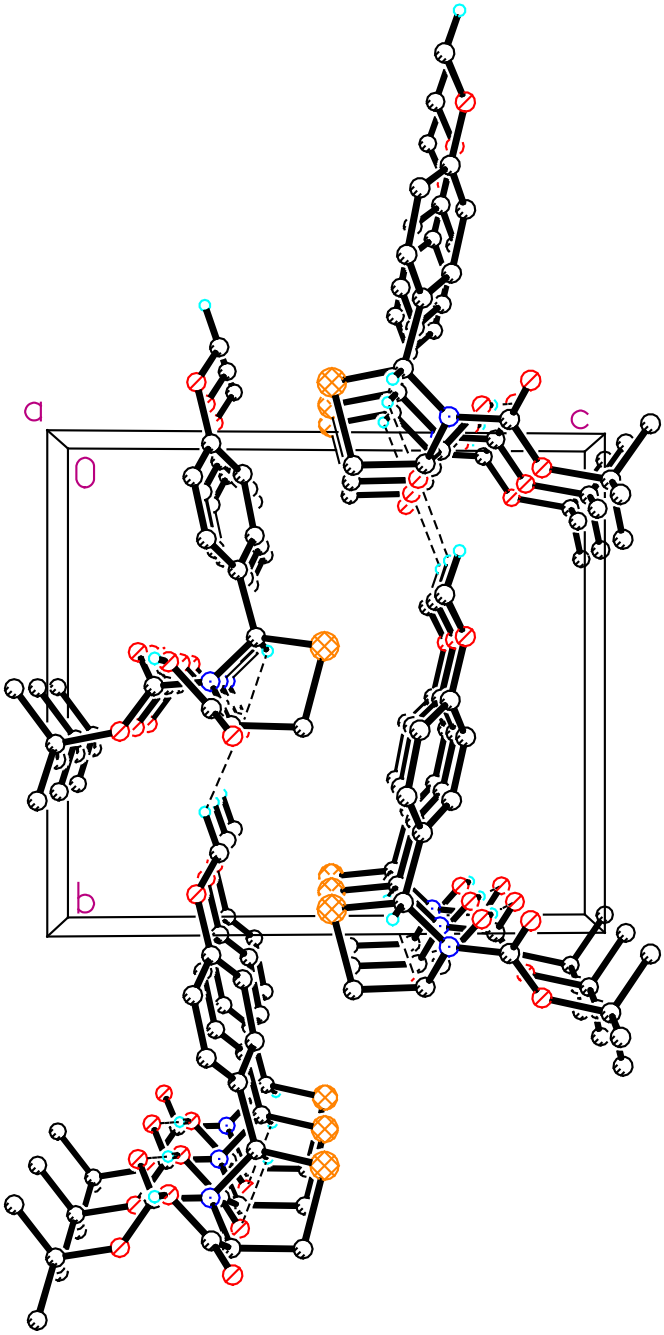


Fig. 4. Packing diagram of '2*R*, 4*R*' isomer of compound **12**, viewed along from a axis, showing the intermolecular hydrogen-bonding scheme (dashed lines).

Table 4
IC_{50} values of the synthesized TCA **1**–**9** against the four bacteria.

Compound	IC_{50}s (μg/mL)			
	Gram-positive		Gram-negative	
	B. subtilis	*S. aureus*	*P. aeruginosa*	*E. coli*
1	3.125	12.5	>50	25
2	25	>50	25	>50
3	12.5	25	>50	>50
4	25	6.25	6.25	12.5
5	3.125	6.25	1.5625	12.5
6	12.5	6.25	3.125	12.5
7	12.5	12.5	6.25	25
8	12.5	>50	25	12.5
9	25	12.5	25	25
Kanamycin G	0.39	1.562	3.125	3.125
Penicillin B	1.562	1.562	6.25	6.25

Table 5
IC_{50} values of the synthesized TBTCA **10**–**18** against the four bacteria.

Compound	IC_{50}s (μg/mL)			
	Gram-positive		Gram-negative	
	B. subtilis	*S. aureus*	*P. aeruginosa*	*E. coli*
10	3.125	0.78	0.78	0.39
11	0.39	3.125	0.39	0.78
12	3.125	1.562	0.39	0.78
13	3.125	3.125	0.39	0.39
14	0.39	0.39	0.195	0.78
15	1.562	1.562	0.39	1.562
16	1.562	1.562	0.78	1.562
17	3.125	1.562	0.39	0.78
18	0.39	1.562	0.39	0.69
Kanamycin G	0.39	1.562	3.125	3.125
Penicillin B	1.562	1.562	6.25	6.25

and respiratory tract and can cause endocarditis, gastroenteritis, septicemia or empyema. The most widely used medicines to treat this infection are broad-spectrum cephalosporins such as cefepime (CFPM) [25], cefozopran (CZOP) [26], and ceftazidime (CAZ) [27,28] or quinolones such as ciprofloxacin [29].

However, there are few reports showing that thiazolidine derivatives may be used as antibacterial agents to cure patients who were infected by *P. aeruginosa*. It is very interesting to find that compound **14** exhibited the highest antibacterial activity against *P. aeruginosa* with the lowest IC_{50} value of 0.195 μg/mL. Therefore, it is suggested that one of our synthesized TBTCA derivatives (compound **14**) could be potential antibacterial agents against *P. aeruginosa*.

3. Conclusions

In summary, a series of TCA and TBTCA derivatives were synthesized and their antibacterial activities were also tested against *B. subtilis*, *S. aureus*, *P. aeruginosa*, and *E. coli*. Based on the analysis and evaluation of antibacterial activities, compound (2*RS*,4*R*)-3-(*tert*-butoxycarbonyl)-2-(5-fluoro-2-hydroxyphenyl) thiazolidine-4-carboxylic acid (**14**) exhibited higher selective antibacterial activities against pathogenic bacterium *P. aeruginosa* than positive controls Kanamycin G and Penicillin B did. Hence, it may be as potential therapeutic utilities if further structural optimization and reorganization are performed. Future efforts shall be aimed at synthesis and evaluation of pure individual stereoisomer of the most promising thiazolidine derivatives discussed above. Our understanding of the antibacterial activity of thiazolidine derivatives against *P. aeruginosa* is still limited, and further investigation is in progress.

4. Experiments

4.1. Crystallographic studies

X-ray single-crystal diffraction data for '2*R*, 4*R*' isomer of compound **12** were collected on a Nonius CAD4 diffractometer equipped with graphite-monochromatized Mo Kα ($\lambda = 0.71073$ Å) radiation. The program CAD4 software was used for data collection and cell refinement. Data reduction was solved by XCAD4 program. Structure was solved by direct methods using the SHELXS program of the SHELXTL package and refined by full-matrix least-squares methods with SHELXL [30]. All non-hydrogen atoms of '2*R*, 4*R*' isomer of compound **12** were refined with anisotropic thermal parameters. All hydrogen atoms were placed in geometrically idealized positions and constrained to ride on their parent atoms.

4.2. Biological assay

The antibacterial activities of the synthesized compounds were tested against *B. subtilis*, *E. coli*, *P. aeruginosa* and *S. aureus* using MH medium (Muellere–Hinton medium: casein hydrolysate 17.5 g, soluble starch 1.5 g, beef extract 1000 mL). The IC_{50}s of the tested compounds were determined by a colorimetric method using the dye MTT. A stock solution of the synthesized compound (50 μg/mL) in DMSO was prepared and graded quantities of the tested compounds were incorporated in specified quantity of sterilized liquid medium (MH medium for antibacterial activity). A specified quantity of the medium containing the compound was poured into microtitration plates. Suspension of the microorganism was prepared to contain approximately 10^5 cfu/mL and applied to microtitration plates with serially diluted compounds in DMSO to be tested and incubated at 37 °C for 24 h. After the IC_{50}s were visually determined on each of the microtitration plates, 50 mL of PBS phosphate buffered saline 0.01 mol/L, pH 7.4, $Na_2HPO_4 \cdot 12H_2O$ (2.9 g), KH_2PO_4 (0.2 g), NaCl (8.0 g), KCl (0.2 g), distilled water (1000 mL) containing 2 mg of MTT/mL was added to each well. Incubation was continued at room temperature for 4–5 h. The content of each well was removed, and 100 mL of isopropanol containing 5% 1 mol/L HCl was added to extract the dye. After 12 h of incubation at room temperature, the optical density (OD) was measured with a microplate reader at 570 nm[5]. The observed IC_{50}s are presented in Tables 4 and 5.

4.3. Chemistry syntheses

All the reagents and solvents used were of analytical reagent grade or were purified by standard methods before use. Melting points (uncorrected) were determined on an XT4 MP apparatus (Taike Corp., Beijing, China). ESI mass spectra were obtained on a Mariner System 5304 mass spectrometer, and ^{1}H NMR spectra were recorded at DPX300 in DMSO-d_6 on a Bruker spectrometer. Elemental analyses were performed on a CHN-O-Rapid instrument and were within ± 0.4% of the theoretical values.

4.3.1. General procedure for the preparation of TCA derivatives **1**–**9**

A mixture of L-cysteine (0.121 g, 1.0 mmol) and appropriate aldehyde (1.0 mmol) in ethanol (25 mL) was stirred at room temperature for 8 h, and the solid separated was collected, washed with diethyl ether and dried to obtain **1**–**9**.

*4.3.1.1. (2RS,4R)-2-Phenylthiazolidine-4-carboxylic acid (**1**).* Obtained as white solids (0.196 g, 94%). ^{1}H NMR (DMSO-d_6) δ 7.24–7.53 (m, 5H), 5.67 (s, 0.6H), 5.50 (s, 0.4H), 4.22 (dd, J = 6.9, 4.5 Hz, 0.6H), 3.90 (dd, J = 8.7, 7.2 Hz, 0.4H), 3.27–3.40 (m, 1H), 3.04–3.16 (m, 1H); MS (ESI) m/z 208 (M-1). Anal. Calcd. for $C_{10}H_{11}NO_2S$: C, 57.39; H, 5.30; N, 6.69. Found: C, 57.62; H, 5.28; N, 6.71.

*4.3.1.2. (2RS,4R)-2-(4-Hydroxyphenyl)thiazolidine-4-carboxylic acid (**2**).* Obtained as white solids (0.209 g, 93%). ^{1}H NMR (DMSO-d_6) δ 6.71–6.77 (m, 2H), 7.24–7.33(m, 2H), 5.53 (s, 0.5H), 5.40 (s, 0.5H), 4.27 (dd, J = 4.02 Hz, 4.02 Hz, 0.5H), 3.85 (dd, J = 7.32 Hz, 7.32 Hz, 0.5H), 3.26–3.43 (m, 1H), 2.96–3.18 (m, 1H); MS (ESI) m/z 224 (M-1). Anal. Calcd. for $C_{10}H_{11}NO_3S$: C, 53.32; H, 4.92; N, 6.22. Found: C, 53.50; H, 4.94; N, 6.20.

*4.3.1.3. (2RS,4R)-2-(4-Methoxyphenyl)thiazolidine-4-carboxylic acid (**3**).* Obtained as white solids (0.220 g, 92%). ^{1}H NMR (DMSO-d_6) δ 7.35–7.45 (m, 2H), 6.87–6.94(m, 2H), 5.60 (s, 0.5H), 5.45 (s, 0.5H), 4.24 (dd, J = 4.05 Hz, 4.38 Hz, 0.5H), 3.86 (dd, J = 8.40 Hz, 7.32 Hz, 0.5H), 3.28–3.42 (m, 1H), 2.77–2.92 (m, 1H); MS (ESI) m/z 238 (M-1). Anal. Calcd. for $C_{11}H_{13}NO_3S$: C, 55.21; H, 5.48; N, 5.85. Found: C, 55.43; H, 5.50; N, 5.83.

*4.3.1.4. (2RS,4R)-2-(4-Bromophenyl)thiazolidine-4-carboxylic acid (**4**).* Obtained as white solids (0.245 g, 85%). ^{1}H NMR (DMSO-d_6) δ 7.53–7.58 (m, 2H), 7.38–7.50(m, 2H), 5.68 (s, 0.6H), 5.50 (s, 0.4H), 4.17 (dd, J = 5.19 Hz, 6.75 Hz, 0.5H), 3.90 (dd, J = 6.99 Hz, 8.70 Hz, 0.5H), 3.10–3.12 (m, 1H), 3.05–3.08 (m, 1H); MS (ESI) m/z 286 (M-1). Anal. Calcd. for $C_{10}H_{10}BrNO_2S$: C, 41.68; H, 3.50; N, 4.86. Found: C, 41.51; H, 3.52; N, 4.88.

*4.3.1.5. (2RS,4R)-2-(5-Fluoro-2-hydroxyphenyl)thiazolidine-4-carboxylic acid (**5**).* Obtained as white solids (0.218 g, 90%). ^{1}H NMR (DMSO-d_6) δ 6.74–6.83 (m, 1H), 6.87–7.01 (m, 1H), δ 7.10–7.41 (m, 1H), 5.80 (s, 0.6H), 5.63 (s, 0.4H), 4.20 (dd, J = 5.85 Hz, 6.21 Hz, 0.6H), 3.86 (dd, J = 6.75 Hz, 6.78 Hz, 0.4H), 3.19–3.24 (m, 1H), 2.96–3.04 (m, 1H); MS (ESI) m/z 242 (M-1). Anal. Calcd for $C_{10}H_{10}FNO_3S$: C, 49.37; H, 4.14; N, 5.76. Found: C, 49.56; H, 4.12; N, 5.74.

*4.3.1.6. (2RS,4R)-2-(5-Chloro-2-hydroxyphenyl)thiazolidine-4-carboxylic acid (**6**).* Obtained as white solids (0.230 g, 88%). ^{1}H NMR (DMSO-d_6) δ 7.33–7.50 (m, 1H), δ 7.10–7.20 (m, 1H), δ 6.79–6.85 (m, 1H), 5.80 (s, 0.6H), 5.63 (s, 0.4H), 4.18 (dd, J = 6.03 Hz, 6.03 Hz, 0.6H), 3.87 (dd, J = 6.93 Hz, 6.78 Hz, 0.4H), 3.19–3.25 (m, 1H), 2.97–3.03 (m, 1H); MS (ESI) m/z 258 (M-1). Anal. Calcd. for $C_{10}H_{10}ClNO_3S$: C, 46.25; H, 3.88; N, 5.39. Found: C, 46.43; H, 3.86; N, 5.41.

*4.3.1.7. (2RS,4R)-2-(5-Bromo-2-hydroxyphenyl)thiazolidine-4-carboxylic acid (**7**).* Obtained as white solids (0.278 g, 91%). ^{1}H NMR (DMSO-d_6) δ 7.44–7.60 (m, 1H), δ 7.20–7.30 (m, 1H), δ 6.72–6.78 (m, 1H), 5.80 (s, 0.6H), 5.63 (s, 0.4H), 4.16 (dd, J = 6.03 Hz, 6.21 Hz, 0.6H), 3.85 (dd, J = 6.96 Hz, 6.78 Hz, 0.4H), 3.18–3.24 (m, 1H), 2.96–3.02 (m, 1H); MS (ESI) m/z 302 (M-1). Anal. Calcd. for $C_{10}H_{10}BrNO_3S$: C, 39.49; H, 3.31; N, 4.61. Found: C, 39.65; H, 3.33; N, 4.59.

*4.3.1.8. (2RS,4R)-2-(3,5-Dibromo-2-hydroxyphenyl)thiazolidine-4-carboxylic acid (**8**).* Obtained as white solids (0.623 g, 75%). ^{1}H NMR (DMSO-d_6) δ 7.41–7.68 (m, 2H), 5.88 (s, 0.6H), 5.70 (s, 0.4H), 4.12–4.17 (m, 0.4H), 3.96–4.05 (m, 0.6H), 2.94–3.03 (m, 1H), 2.73–2.82 (m, 1H); MS (ESI) m/z 380 (M-1). Anal. Calcd. for $C_{10}H_9Br_2NO_3S$: C, 31.35; H, 2.37; N, 3.66. Found: C, 31.48; H, 2.38; N, 3.64.

*4.3.1.9. (2RS,4R)-2-(3,5-Di-tert-butyl-4-hydroxyphenyl)thiazolidine-4-carboxylic acid (**9**).* Obtained as white solids (0.273 g, 81%). ^{1}H NMR (DMSO-d_6) δ 6.93–7.25 (m, 2H), 5.51 (s, 0.6H), 5.40 (s, 0.4H), 4.29 (dd, J = 3.90 Hz, 3.75 Hz, 0.5H), 3.84 (dd, J = 7.53 Hz, 8.16 Hz, 0.5H), 3.14–3.19 (m, 1H), 2.73–2.92 (m, 1H), 1.41 (s, 18H); MS (ESI) m/z 336 (M-1). Anal. Calcd. for $C_{18}H_{27}NO_3S$: C, 64.06; H, 8.06; N, 4.15. Found: C, 64.27; H, 8.09; N, 4.13.

4.3.2. General procedure for the preparation of TBTCA derivatives ***10–18***

A mixture of TCA (1.0 mmol) and appropriate NaOH (10%, 1.0 mmol) in dioxane (25 mL) was stirred at ice-water temperature for 2 h. BOC_2O (1.0 mmol) was added and stirred at ice-water temperature for 1 h and then room temperature for 5 h. Most of the solvent was extracted and appropriate amount of water was added to adjust to neutral pH values. Ethyl acetate was added and extracted (50 mL × 3), and washed with appropriate saturated aqueous solution of common salt, and dried with anhydrous magnesium sulphate. Solvent was extracted to dry to obtain whiter solids **10**–**18**.

*4.3.2.1. (2RS,4R)-3-(tert-Butoxycarbonyl)-2-phenylthiazolidine-4-carboxylic acid (**10**).* Obtained as white solids (0.283 g, 92%). ^{1}H NMR (DMSO-d_6) δ 7.64–7.66 (m, 2H), δ 7.26–7.35 (m, 3H), 6.13 (s, 0.5H), 5.97 (s, 0.5H), 4.73 (s, 0.5H), 4.59 (s, 0.5H), 3.44–3.50 (m, 1H), 2.09–2.21 (m, 1H), 1.35 (s, 4.5H), 1.12 (s, 4.5H); MS (ESI) *m*/*z* 308 (M-1). Anal. Calcd. for $C_{15}H_{19}NO_4S$: C, 58.23; H, 6.19; N, 4.53. Found: C, 58.05; H, 6.15; N, 4.57.

*4.3.2.2. (2RS,4R)-3-(tert-Butoxycarbonyl)-2-(4-hydroxyphenyl)thiazolidine-4-carboxylic acid (**11**).* Obtained as white solids (0.301 g, 86%). ^{1}H NMR (DMSO-d_6) δ 9.36 (s, 1H) 7.43 (d, 2H), 6.70 (d, 2H), 5.87–6.02 (m, 1H), 4.56–4.69 (m, 1H), 3.44 (dd, J = 6.90 Hz, 6.90 Hz, 1H), 3.12–3.17 (m, 1H), 1.39 (s, 4.5H), 1.15 (s, 4.5H); MS (ESI) *m*/*z* 324 (M-1). Anal. Calcd. for $C_{15}H_{19}NO_5S$: C, 55.37; H, 5.89; N, 4.30. Found: C, 55.62; H, 6.85; N, 4.32.

*4.3.2.3. (2RS,4R)-3-(tert-Butoxycarbonyl)-2-(4-methoxyphenyl)thiazolidine-4-carboxylic acid (**12**).* Obtained as white solids (0.324 g, 96%). ^{1}H NMR (DMSO-d_6) δ 7.54–7.57 (d, 2H), δ 6.85–6.87 (d, 2H), 5.91–6.06 (m, 1H), 4.54–4.68 (m, 1H), 3.73 (s, 3H), 3.39–3.49 (m, 1H), 3.12–3.18 (m, 1H), 1.35 (s, 4.5H), 1.13 (s, 4.5H); MS (ESI) *m*/*z* 338 (M-1). Anal. Calcd. for $C_{16}H_{21}NO_5S$: C, 56.62; H, 6.24; N, 4.13. Found: C, 56.90; H, 6.21; N, 4.17.

*4.3.2.4. (2RS,4R)-2-(4-Bromophenyl)-3-(tert-butoxycarbonyl)thiazolidine-4-carboxylic acid (**13**).* Obtained as white solids (0.366 g, 95%). ^{1}H NMR (DMSO-d_6) δ 7.41–7.47 (m, 4H), 5.90–5.92 (m, 1H), 4.90–4.92 (m, 1H), 3.31–3.37 (m, 2H), 1.25 (s, 9H); MS (ESI) *m*/*z* 338 (M-1). Anal. Calcd. for $C_{15}H_{18}BrNO_4S$: C, 46.40; H, 4.67; N, 3.61. Found: C, 46.72; H, 4.49; N, 3.66.

*4.3.2.5. (2RS,4R)-3-(tert-Butoxycarbonyl)-2-(5-fluoro-2-hydroxyphenyl)thiazolidine-4-carboxylic acid (**14**).* Obtained as white solids (0.319 g, 93%). ^{1}H NMR (DMSO-d_6) δ 9.83–9.87 (b, 1H), 7.75–7.79 (m, 1H), 6.85–6.92 (m, 1H), 6.73–6.77 (m, 1H), 6.09 (s, 1H), 4.48–4.59 (m, 1H), 3.44 (dd, J = 6.6 Hz, 6.3 Hz, 1H), 3.01–3.12 (m, 1H), 1.37 (s, 4.5H); 1.16 (s, 4.5H); MS (ESI) *m*/*z* 342 (M-1). Anal. Calcd. for $C_{15}H_{18}FNO_5S$: C, 52.47; H, 5.28; N, 4.08. Found: C, 52.11; H, 5.25; N, 4.12.

*4.3.2.6. (2RS,4R)-3-(tert-Butoxycarbonyl)-2-(5-chloro-2-hydroxyphenyl)thiazolidine-4-carboxylic acid (**15**).* Obtained as white solids (0.312 g, 87%). ^{1}H NMR (DMSO-d_6) δ 10.11 (b, 1H), 7.96 (s, 1H), 7.12 (dd, J = 2.7 Hz, 2.4 Hz, 1H), 6.80 (s, 1H), 4.50–4.63 (m, 1H), 3.41–3.47 (m, 1H), 2.01–2.13 (m, 1H), 1.37 (s, 4.5H); 1.16 (s, 4.5H); MS (ESI) *m*/*z* 358 (M-1). Anal. Calcd. for $C_{15}H_{18}ClNO_5S$: C, 50.07; H, 5.04; N, 3.89. Found: C, 50.32; H, 5.01; N, 3.92.

*4.3.2.7. (2RS,4R)-2-(5-Bromo-2-hydroxyphenyl)-3-(*tert*-butoxycarbonyl)thiazolidine-4-carboxylic acid (**16**).* Obtained as white solids (0.370 g, 92%). ^{1}H NMR (DMSO-d_6) δ 7.95–8.09 (m, 1H), 7.21 (dd, J = 2.70 Hz, 2.40 Hz, 1H), 6.70 (s, 1H), 6.02–6.05 (m, 1H), 4.46–4.54 (m, 1H), 4.54 (s, 0.5H), 4.46 (s, 0.5H), 3.07–3.11 (m, 1H), 1.36 (s, 4.5H), 1.13 (s, 4.5H); MS (ESI) *m*/*z* 402 (M-1). Anal. Calcd. for $C_{15}H_{18}BrNO_5S$: C, 44.56; H, 4.49; N, 3.46. Found: C, 44.38; H, 4.47; N, 3.49.

*4.3.2.8. (2RS,4R)-3-(tert-Butoxycarbonyl)-2-(3,5-dibromo-4-hydroxyphenyl)thiazolidine-4-carboxylic acid (**17**).* Obtained as white solids (0.429 g, 89%). ^{1}H NMR (DMSO-d_6) δ 7.38–7.51 (m, 2H), 5.72–5.88 (m, 1H), 4.01–4.15 (m, 1H), 2.85–3.10 (m, 1H), 2.65–2.79 (m, 1H); 1.27 (s, 4.5H), 1.10 (s, 4.5H); MS (ESI) *m*/*z* 480 (M-1). Anal. Calcd. for $C_{15}H_{17}Br_2NO_5S$: C, 37.29; H, 3.55; N, 2.90. Found: C, 37.02; H, 3.57; N, 2.87.

*4.3.2.9. (2RS,4R)-3-(tert-Butoxycarbonyl)-2-(3,5-di-*tert*-butyl-4-hydroxyphenyl)thiazolidine-4-carboxylic acid (**18**).* Obtained as white solids (0.381 g, 87%). ^{1}H NMR (DMSO-d_6) δ 6.85–7.52 (m, 2H), 5.41–5.56 (m, 1H), 3.97–4.26 (m, 2H), 3.07–3.25 (m, 1H), 2.58–2.79 (m, 1H), 1.32 (s, 18H); 1.25 (s, 4.5H), 1.19 (s, 4.5H); MS (ESI) *m*/*z* 436 (M-1). Anal. Calcd. for $C_{23}H_{35}NO_5S$: C, 63.13; H, 8.06; N, 3.20. Found: C, 63.41; H, 8.04; N, 3.23.

Acknowledgements

The work was financed by grants (Project 30772627) from National Natural Science Foundation of China.

References

[1] B. Jan, O. Barbara, K.M. Jan, O. Andrzej, Eur. J. Med. Chem. 42 (2007) 993–1003.
[2] V. Sriramurthy, G.A. Barcan, O. Kwon, J. Am. Chem. Soc. 129 (2007) 12928–12929.
[3] V. Gududuru, E. Hurh, J.T. Dalton, D.D. Miller, J. Med. Chem. 48 (2005) 2584–2588.
[4] B. Hartzoulakis, S. Rossiter, H. Gill, B. O'Hara, E. Steinke, P.J. Gane, R. Hurtado-Guerrero, J.M. Leiper, P. Vallance, J.M. Rustc, D.L. Selwood, Bioorg. Med. Chem. Lett. 17 (2007) 3953–3956.
[5] Y. Hiraiwa, T. Usui, Y. Akiyama, K. Maebashi, N. Minowa, D. Ikeda, Bioorg. Med. Chem. Lett. 17 (2007) 3540–3543.
[6] M. Tuncbilek, N. Altanlar, Arch. Pharm. Chem. Life Sci. 339 (2006) 213–216.
[7] J. Wrobel, J. Jetter, W. Kao, J. Rogers, L. Di, J. Chi, M.C. Peréz, G.C. Chen, E.S. Shen, Bioorg. Med. Chem. 14 (2006) 5729–5741.
[8] H.T. Nagasawa, D.J.W. Goon, R.T. Zera, D.L. Yuzon, J. Med. Chem. 25 (1982) 489–491.
[9] H.T. Nagasawa, D.J.W. Goon, W.P. Muldoon, R.T. Zera, J. Med. Chem. 27 (1984) 591–596.
[10] M. Seki, Y. Mori, M. Hatsuda, S. Yamada, J. Org. Chem. 67 (2002) 5527–5536.
[11] A. Ei'ichi, N. Koichiro, S. Akihiko, N. Jeffrey-Tri, H. Koushi, H. Yoshio, N. Shingo, K. Tooru, H. Yoshio, K. Yoshiaki, Bioorg. Med. Chem. Lett. 17 (2007) 4213–4217.
[12] N.A. Noureldin, J.W. Bellegarde, Synthesis 6 (1999) 943–946.
[13] H.D. Flack, Acta Crystallogr., Sect. A 39 (1983) 876–881.
[14] J. Zhang, S. Chen, T. Wu, P. Feng, X. Bu, J. Am. Chem. Soc. 130 (2008) 12882–12883.
[15] D.E. Williams, J. Davies, B.O. Patrick, H. Bottriell, T. Tarling, M. Roberge, R.J. Andersen, Org. Lett. 10 (2008) 3501–3504.
[16] L. Shi, H.M. Ge, S.H. Tan, H.Q. Li, Y.C. Song, H.L. Zhu, Eur. J. Med. Chem. 42 (2007) 558–564.
[17] G. Turan-Zitouni, J.A. Fehrentz, P. Chevallet, J. Martinez, Z.A. Kaplancıklı, A. Özdemir, M. Arslanyolu, M.T. Yıldız, Arch. Pharm. Chem. Life Sci. 340 (2007) 310–314.
[18] N.R. Pace, Science 276 (1997) 734–740.
[19] C.R. Woese, Microbiol. Rev. 51 (1987) 221–271.
[20] *P. aeruginosa* Info—(http://www.pseudomonas.com).
[21] J.R. Govan, V. Deretic, Microbiol. Rev. 60 (1996) 539–574.
[22] A. Erik, Science 312 (2006) 1526–1530.
[23] T.L. Vincent, S.S. Roger, T. Burkhard, L. Stephen, Infect. Immun. 73 (2005) 1695–1705.
[24] V. Mantareva, V. Kussovski, I. Angelov, E. Borisova, L. Avramov, G. Schnurpfeil, D. Wöhrle, Bioorg. Med. Chem. 15 (2007) 4829–4835.
[25] T. Naito, S. Aburaki, H. Kamachi, Y. Narita, J. Okumura, H. Kawaguchi, J. Antibiot. 39 (1986) 1092–1107.
[26] A. Miyake, Y. Yoshimura, Y. Yamaoka, T. Nishimura, N. Hashimoto, A. Imada, J. Antibiot. 45 (1992) 485–499.
[27] L. Verbist, J. Verhaegen, Antimicrob. Agents Chemother. 17 (1980) 807–812.
[28] C.H. O'callaghan, P. Acred, P.B. Harper, D.M. Ryan, S.M. Kirby, S.M. Harding, Antimicrob. Agents Chemother. 17 (1980) 876–883.
[29] R. Wise, J.M. Andrews, L.J. Edwards, Antimicrob. Agents Chemother. 23 (1983) 559–564.
[30] G.M. Sheldrick, SHELXTL-97: Program for Crystal Structure Solution and Refinement, University of Göttingen, Göttingen, Germany, 1997.