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Synthesis and antibacterial evaluation of novel 11-*O*-aralkylcarbamoyl-3-*O*-descladinosylclarithromycin derivatives

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ABSTRACT

A series of novel 11-*O*-aralkylcarbamoyl-3-*O*-descladinosylclarithromycin derivatives were designed, synthesized and evaluated for their *in vitro* antibacterial activity. The results showed that the majority of the target compounds displayed potent activity against erythromycin-susceptible *S. pyogenes*, erythromycin-resistant *S. pneumoniae* A22072 expressing the *mef* gene and *S. pneumoniae* AB11 expressing the *mef* and *erm* genes. Besides, most of the target compounds exhibited moderate activity against erythromycin-susceptible *S. aureus* ATCC25923 and *B. subtilis* ATCC9372. In particular, compounds **11a**, **11b**, **11c**, **11e**, **11f** and **11h** were found to exert favorable antibacterial activity against erythromycin-susceptible *S. pyogenes* with the MIC values of 0.015–0.125 μg/mL. Furthermore, compounds **10e**, **11a**, **11b** and **11c** showed superior activity against erythromycin-resistant *S. pneumoniae* A22072 with the MIC values of 0.25–0.5 μg/mL. Additionally, compound **11c** was the most effective against all the erythromycin-resistant *S. pneumoniae* strains (A22072, B1 and AB11), exhibiting 8-, 8- and 32-fold more potent activity than clarithromycin, respectively.

Macrolide antibiotics, which are weakly alkaline and lipophilic compounds produced by *Streptomycetes*,[1] have been commonly used for the treatment of upper and lower respiratory tract infections since 1950s because of favorable antibacterial activity, broad antibacterial spectrum and good tolerance.[2] Mechanistic studies demonstrate that macrolides can serve as potent protein synthesis inhibitors by binding to the peptide exit tunnel of the large subunit (50S) of bacterial ribosome, hence interfering with the elongation of nascent polypeptide chains.[3] Erythromycin A (EMA), which belongs to the first-generation macrolides, readily degrades under acidic conditions, leading to the loss of antibacterial activity and the generation of gastrointestinal side effects.[4] Clarithromycin (CAM) and azithromycin (AZM) (Fig. 1), the representatives of the second-generation macrolides, have better acid-stability in the stomach and superior bioavailability, as well as fewer gastrointestinal side effects.

However, more and more inappropriate use of macrolides leads to the increasing drug resistance, which severely threatens the therapeutic effectiveness of macrolides and limits the clinical use of macrolides.[5] Two distinguished mechanisms have accounted for the majority of macrolide resistance: the *erm* gene-mediated base-specific mono- or di-methylation of 23S rRNA, which leads to the MLS_B resistance, and the *mef* gene-mediated membrane protein efflux pump that can limit the steady-state accumulation of drugs by transporting antibiotics out of cytoplasm.[6] Therefore, it's urgent to develop novel macrolide antibiotics against resistant pathogens. An X-ray cocrystal structure research reveals that 3-*O*-cladinose is not a necessary moiety for antibacterial activity,[7] and modification at the C-3 position results in enhanced activity against efflux resistance.[8] Consequently, in the late 1990s, the third generation macrolides known as ketolides[1] and acylides[9] (Fig. 1) were designed to tackle bacterial resistance.

Extensive investigation has been carried out on the chemical modifications of 14-membered macrolides to effectively cope with bacterial resistance over the past years. Compound EP-1553 (Fig. 1), bearing an arylalkyl group at the C-11 position, exhibited good activity against macrolide-resistant bacteria.[10] We reported a 15-membered macrolide azithromycin derivative (**A**) (Fig. 1), which was modified at the C-11 and C-4″ positions, as well as showed remarkably improved anti-resistant activity compared to CAM and AZM.[11] In addition, we designed and synthesized a series of 11-*O*-carbamoyl clarithromycin ketolides (**B**) (Fig. 1), showing comparable activity against susceptible bacteria and improved activity against resistant bacteria in comparison with CAM and AZM.[12] Furthermore, the compound FSM-100573 synthesized by

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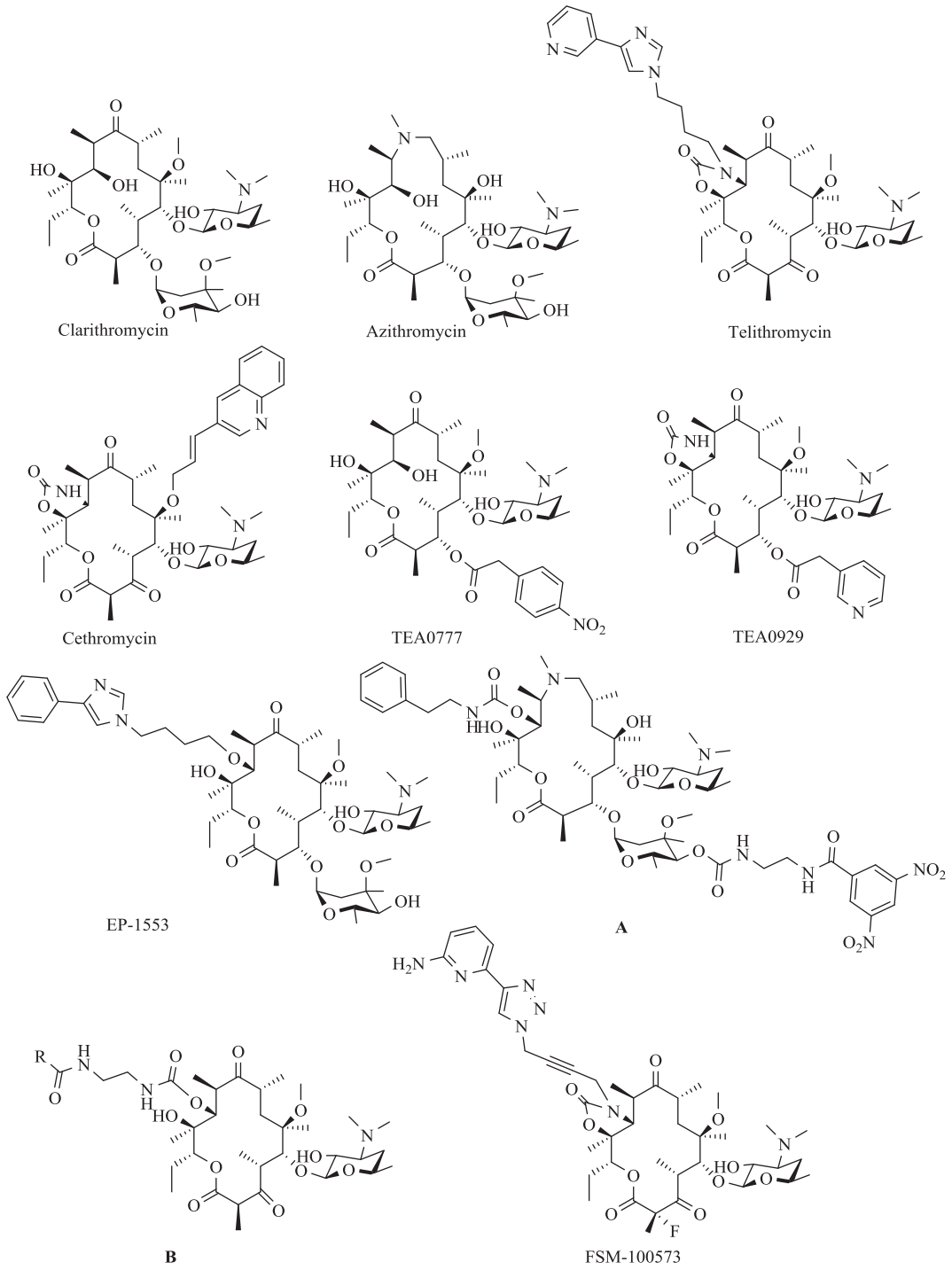
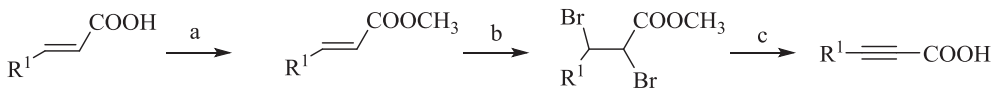
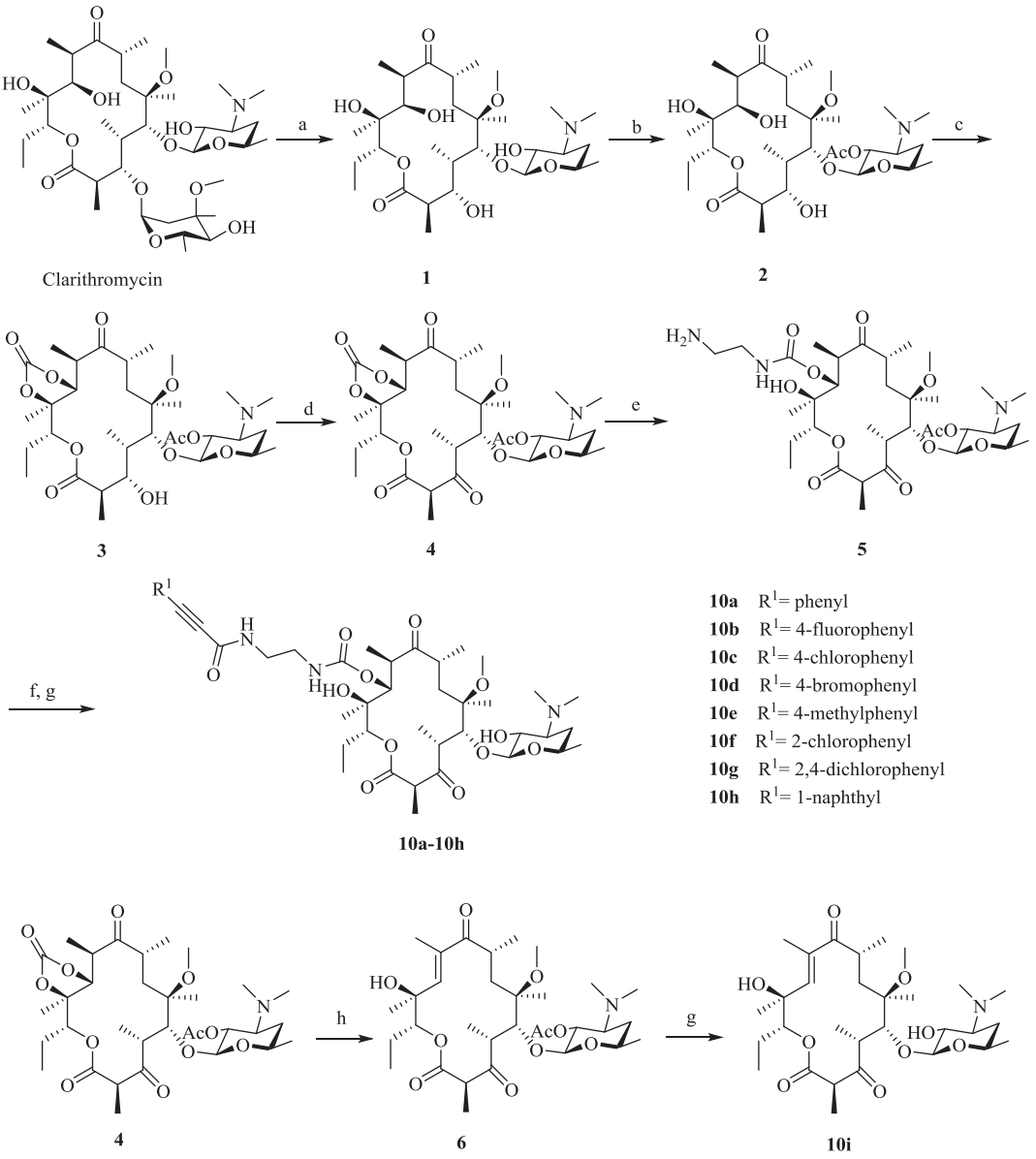


Fig. 1. Structures of clarithromycin, azithromycin, ketolides, acylides, EP-1553, **A**, **B** and FSM-100573.



A1	R^1= phenyl	**A5**	R^1= 4-methylphenyl
A2	R^1= 4-fluorophenyl	**A6**	R^1= 2-chlorophenyl
A3	R^1= 4-chlorophenyl	**A7**	R^1= 2,4-dichlorophenyl
A4	R^1= 4-bromophenyl	**A8**	R^1= naphthyl

Scheme 1. Synthesis of propiolic acids. Reagents and conditions: a) methanol, sulfuric acid, 75 °C, 12 h, 86–100%; b) bromine, CH_2Cl_2, 25 °C, 24 h, 40–100%; c) KOH, ethanol, 85 °C, 36 h, 34–81%.



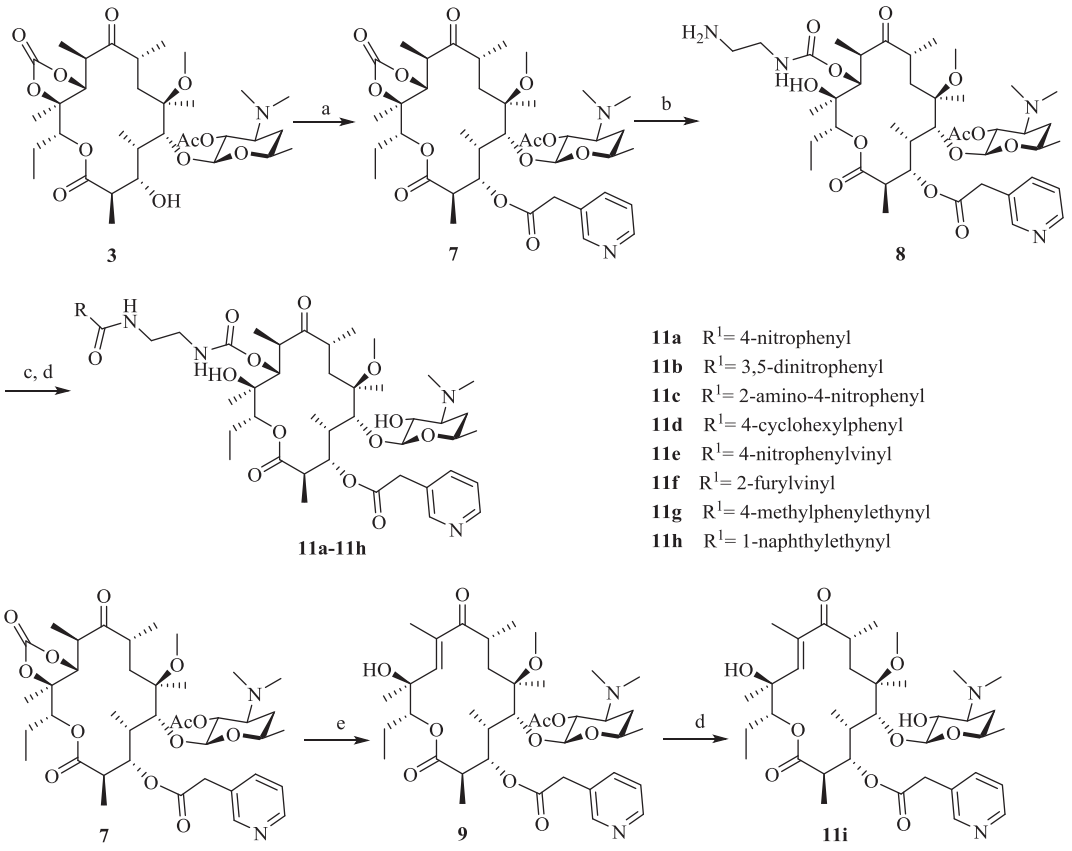
Scheme 2. Synthesis of 11-*O*-aralkylcarbamoyl-3-*O*-descladinosylclarithromycin derivatives (**10a–10i**). Reagents and conditions: a) HCl, H_2O, 25 °C, 3 h, 84%; b) Ac_2O, Et_3N, DCM, 0–25 °C, 36 h, 97%; c) pyridine, BTC, DCM, 0–25 °C, 20 h, 99%; d) PCC, DCM, 25 °C, 24 h, 91%; e) ethylenediamine, pyridine hydrochloride, 15 °C, 6 h, 43%; f) HOBt, DCC, THF, 0–25 °C, 10 h, 100%; g) CH_3OH, 55 °C, 24 h, 36–55%; h) ethylenediamine, 25 °C, 6 h, 21%.

the convergent assembly of simple chemical building blocks was found to displayed superior potencies against Gram-positive and Gram-negative bacteria.[13]

On the basis of the consideration detailed above, we designed and synthesized a series of novel 11-*O*-aralkylcarbamoyl-3-*O*-descladinosylclarithromycin derivatives to prevent the *erm*- or *mef*-mediated bacterial resistance and broaden their antibacterial spectra. On the one hand, the removal of 3-*O*-cladinose can avoid the induction of bacterial resistance. On the other hand, the 11-*O*-arylalkylcarbamoyl side chain is beneficial to bind with A752 in domain II through additional forces such as hydrogen bonding, π-π stacking or van der Waals interactions to improve the antibacterial activity. Besides, we hope that the introductions of propynylamide group and 3-pyridyl acetate group are able to enhance the antibacterial activity.

The general synthetic method for the propiolic acids is shown in Scheme 1. The reaction of different acrylic acids gave corresponding methyl acrylates in the presence of methanol and concentrated sulfuric acid. Then, they were dibrominated by Br_2. Finally, the corresponding propiolic acids (**A1–A8**) were prepared in the presence of KOH and ethanol.

The synthetic method for 11-*O*-aralkylcarbamoyl-3-*O*-descladinosylclarithromycin derivatives is shown in Schemes 2 and 3 .[14,15] Clarithromycin was used as the starting material for the synthesis of the target compounds, which was treated with 36% hydrochloric acid to generate the 3-descladinosylclarithromycin (**1**). Then, it was coupled with acetic anhydride in the presence of triethylamine to give the acetyl product (**2**) that was subsequently reacted with bis(trichloromethyl)carbonate (BTC) to afford the 11,12-cyclic carbonate (**3**) in the presence of pyridine. After that, the 3-hydroxy group of **3** was converted into 3-ketone to obtain the ketone (**4**) under pyridinium chlorochromate (PCC) oxidation, which was treated with ethylenediamine and pyridine hydrochloride at 15 °C to give 2′-*O*-acetyl-11-*O*-(aminoethyl)carbamoyl-3-*O*-descladinosyl-3-ketoclarithromycin (**5**). The structure of **5** was confirmed by ^1H and ^{13}C NMR spectra which displayed in the Supplementary material in detail and further HMBC spectrum (correlation of CH-11 (δ 4.67)/CO (δ 156.12)) indicated that the ethylenediamine side chain was linked to the C-11 position. Finally, the target compounds (**10a–10h**) were prepared by coupling **5** with the



Scheme 3. Synthesis of 11-*O*-aralkylcarbamoyl-3-*O*-descladinosylclarithromycin derivatives (**11a–11i**). Reagents and conditions: a) 3-pyridylacetic acid hydrochloride, pyridine, DCC, DMAP, CH_2Cl_2, 0–25 °C, 7 h, 85%; b) ethylenediamine, pyridine hydrochloride, 15 °C, 6 h, 46%; c) HOBt, DCC, THF, 0–25 °C, 10 h, 96–100%; d) CH_3OH, 55 °C, 24 h, 46–67%; e) ethylenediamine, 25 °C, 6 h, 21%.

corresponding propiolic acids (**A1**-**A8**) in the presence of 1-hydroxybenzotrizole (HOBt) and dicyclohexylcarbodiimide (DCC), followed by methanolysis. Besides, treatment of **4** treated with ethylenediamine at 25 °C gave 2′-*O*-acetyl-10,11-dehydration-3-*O*-descladinosyl-3-keto-clarithromycin (**6**), and the target compound (**10i**) was prepared after deprotection in methanol.[16]

3-Hydroxy group of **3** was converted into 3-pyridyl acetate product (**7**) under 3-pyridylacetic acid hydrochloride, DCC, DMAP and pyridine, which was treated with ethylenediamine and pyridine hydrochloride to give 11-O-(aminoethyl)carbamoyl intermediate (**8**). After that, the target compounds (**11a–11h**) were prepared by coupling **8** with the selected acids in the presence of HOBt and DCC, followed by methanolysis. In addition, **7** was treated with ethylenediamine at 25 °C to give 2′-*O*-acetyl-10,11-anhydro-3-*O*-descladinosyl-3-*O*-(2-(3′-pyridyl)acetyl) clarithromycin (**9**), and the target compound (**11i**) was prepared after deprotection in methanol.[17]

All the 11-*O*-aralkylcarbamoyl-3-*O*-descladinosylclarithromycin derivatives (**10a–10i** and **11a–11i**), as well as CAM and AZM as references, were tested for their *in vitro* antibacterial activity. The minimal inhibitory concentration (MIC) values of antibacterial activity *in vitro* were determined with the application of the standard broth microdilution method recommended by NCCLS. The tested strains included five susceptible strains of *Staphylococcus aureus* ATCC25923 (erythromycin-susceptible strain), *Streptococcus pyogenes* (erythromycin-susceptible strain isolated clinically), *Bacillus subtilis* ATCC9372 (penicillin-susceptible strain), *Escherichia coli* ATCC25922 (penicillin-susceptible strain) and *Pseudomonas aeruginosa* ATCC27853 (penicillin-susceptible strain), and five resistant strains of *Staphylococcus aureus* ATCC43300 (methicillin-resistant strain), *Streptococcus pneumoniae* A22072 (erythromycin-resistant strain expressing the *mef* gene), *Streptococcus pneumoniae* B1 (erythromycin-resistant strain expressing the *erm* gene), *Streptococcus pneumoniae* AB11 (erythromycin-resistant strain expressing the *erm* and *mef* genes) and *Streptococcus pyogenes* R1 (erythromycin-resistant strain isolated clinically). The MIC results are shown in Table 1.

Compared to comparators AZM and CAM, the 11-*O*-aralkylcarbamoyl-3-*O*-descladinosylclarithromycin derivatives (**10a–10h**) universally exhibited excellent antibacterial activity against *S. pneumoniae* A22072 and *S. pneumoniae* AB11, and good antibacterial activity against *S. pyogenes* and *B. subtilis* ATCC9372, but failed to show improved antibacterial activity against *E. coli* ATCC25922 and *P. aeruginosa* ATCC27853. It was noteworthy that compound **10i** showed almost no antibacterial activity against all tested strains. Surprisingly, the 11-*O*-aralkylcarbamoyl-3-*O*-descladinosylclarithromycin derivatives (**11a–11i**) displayed comparable antibacterial activity against all the five susceptible strains and superior antibacterial activity against all the five resistant strains compared with CAM and AZM.

Among them, compounds **11a**, **11b** and **11c** exhibited the best antibacterial activity against *S. pyogenes* with MIC values of 0.015–0.03 μg/mL, as well as against *S. aureus* ATCC25923 with MIC value of 1 μg/mL. Additionally, compounds **10d**, **10e**, **10g**, **10h**, **11b**, **11c** and **11i** displayed the best antibacterial activity with MIC value of 1 μg/mL against *B. subtilis* ATCC9372. As for *E. coli* ATCC25922 and *P. aeruginosa* ATCC27853, the activity of all the compounds was weak, with a MIC value of only 64 μg/mL. In the inhibition of *S. aureus* ATCC43300, the most active compounds **11c** and **11f** exerted MIC values of 32 μg/mL, 4-fold better than CAM and AZM. Furthermore, compounds **10e**, **11a**, **11b** and **11c** showed the strongest activity against *S. pneumoniae* A22072 with the MIC ≤0.5 μg/mL, showing 4-fold and 8-fold more potent activity than AZM and CAM, respectively. Besides, compound **11c** demonstrated 8-fold better antibacterial activity with MIC value of 16 μg/mL against *S. pneumoniae* B1 than CAM

Table 1
In vitro antibacterial activity of 11-*O*-aralkylcarbamoyl-3-*O*-descladinosylclarithromycin derivatives.

Compound	*S. aureus* ATCC 25923[a]	*E. coli* ATCC 25922[b]	*S. pyogenes*[c]	*P. aeruginosa* ATCC27853[d]	*B. subtilis* ATCC 9372[e]	*S. aureus* ATCC 43300[f]	*S. pneumoniae* B1[g]	*S. pneumoniae* A22072[h]	*S. pneumoniae* AB11[i]	*S. pyogenes* R1[j]
10a	32	> 128	2	> 128	4	128	128	4	128	> 128
10b	16	> 128	1	> 128	2	128	128	2	64	> 128
10c	8	> 128	1	> 128	2	64	128	2	64	128
10d	8	128	1	> 128	1	16	128	2	32	64
10e	8	> 128	0.25	128	1	128	128	0.5	64	> 128
10f	16	> 128	1	> 128	4	128	128	2	128	> 128
10g	16	> 128	1	> 128	1	128	128	2	128	128
10h	16	> 128	0.5	> 128	1	64	128	1	128	64
10i	> 128	> 128	> 16	64	128	> 128	> 128	64	> 128	> 128
11a	1	64	0.03	64	2	128	32	0.5	8	> 128
11b	1	64	0.015	64	1	64	32	0.5	4	128
11c	1	64	0.03	64	1	64	16	0.25	4	128
11d	2	128	0.5	128	4	32	64	1	32	32
11e	8	128	0.125	128	4	64	32	1	8	> 128
11f	16	128	0.125	> 128	8	32	64	1	16	> 128
11g	8	> 128	0.25	> 128	2	128	64	1	32	> 128
11h	4	> 128	0.125	> 128	2	64	32	1	16	32
11i	1	128	0.25	128	1	128	32	1	16	128
AZM	0.25	16	0.03	16	0.25	> 128	> 128	4	> 128	> 128
CAM	0.25	64	0.03	64	0.25	> 128	> 128	2	128	> 128

[a] *S. aureus* ATCC25923: erythromycin-susceptible strain.
[b] *E. coli* ATCC25922: penicillin-susceptible strain.
[c] *S. pyogenes*: erythromycin-susceptible strain isolated clinically.
[d] *P. aeruginosa* ATCC27853: penicillin-susceptible strain.
[e] *B. subtilis* ATCC9372: erythromycin-susceptible strain.
[f] *S. aureus* ATCC43300: methicillin-resistant strain.
[g] *S. pneumoniae* B1: erythromycin-resistant strain encoded by the *erm*B gene.
[h] *S. pneumoniae* A22072: erythromycin-resistant strain encoded by the *mef*A gene.
[i] *S. pneumoniae* AB11: erythromycin-resistant strain encoded by the *erm*B and *mef*A genes.
[j] *S. pyogenes* R1: erythromycin-resistant strain isolated clinically.

and AZM. For *S. pneumoniae* AB11, the MIC values of **11b** and **11c** reached 4 μg/mL, 32-fold better than CAM and AZM. In addition, compounds **11d** and **11h** displayed the best antibacterial activity against *S. pyogenes* R1 with MIC value of 32 μg/mL, 4-fold better than CAM and AZM.

Generally, in comparison with acrylamide-linked compounds[12] synthesized before, the propynylamide-linked compounds **10a–10i** exhibited more potent antibacterial activity. Besides, compounds **11a** bearing a phenyl side chain of six atoms distance from the *O*-11 position, exerted better antibacterial activity against susceptible strains than compounds **11e** which correspondingly had the same phenyl side chain of eight atoms distance from the *O*-11 position. In particular, among all the synthesized target compounds, compounds **10d**, **11a**, **11b**, **11c** and **11e**, bearing the side chains of 4-bromophenyl groups or nitrophenyl groups, were the most active against all the types of tested bacteria. Compounds **11d** and **11h**, bearing bigger groups at the end of side chains, showed excellent antibacterial activity against resistant strains including *S. aureus* ATCC43300 and *S. pyogenes* R1. In addition, in the case of same C-11 side chains, compounds **11g**, **11h** and **11i** demonstrated more potent activity against both susceptible and resistant bacteria than compounds **10e**, **10h** and **10i**. Furthermore, compound **10i** almost lost antibacterial activity, while compound **11i** displayed superior antibacterial activity.

In summary, the synthesized target compounds showed comparable activity against susceptible bacteria and improved activity against resistant bacteria in comparison with CAM and AZM. In addition, most of the compounds exhibited favorable activity against Gram-positive bacteria but weak activity against Gram-negative bacteria. The results indicated that the introduction of the arylalkyl group into the C-11 position of CAM was favorable for the potency of compounds against some susceptible and resistant strains. Furthermore, modification of the C-11 and C-3 positions of CAM at the same time could greatly improve their activity against resistant strains. We hypothesized that the substituents at the C-11 position could interact directly with the nucleotide A752 and the removal of cladinose enhanced activity against efflux resistance at the same time, resulting in excellent antibacterial activity.

From the data mentioned above, we could summarize their SARs as follows. Introduction of the novel aralkylcarbamoyl side chains into the C-11 position could greatly improve activity against erythromycin-resistant *S. pneumoniae* compared with CAM and AZM due to the additional binding interaction with A752. The propynylamide-linked compounds were more potent than the compounds linked via arylamide groups, suggesting that appropriate linking groups were beneficial to enhance antibacterial activity. In addition, proper length of side chain could reach the chloramphenicol binding site in bacterial ribosomes, thereby inhibiting peptide formation, so that proper length was favorable to increase the antibacterial activity while the too long or short side chains tended to decrease the antibacterial activity. Besides, the electron-withdrawing groups on the benzene ring of the side chains were good for the improvement of the antibacterial activity, and compounds with bulkier groups at the end of side chains exhibited higher antibacterial activity, indicating that the terminal group might affect the hydrogen bonding, π-π stacking or the electrostatic interaction between the compounds and the bacterial ribosomes. Moreover, in the case of same C-11 side chains, compounds with the C-3-hydroxyl into C-3-pyridyl acetate displayed more potent activity against susceptible and resistant bacteria than compounds with the C-3 ketone. Furthermore, compound **10i** with the C-3 ketone and dehydration at the C-10,11 lost antibacterial activity, possibly due to that these compounds cannot bind with bacterial ribosome well, thus affecting the antibacterial activity.

In a word, a series of novel 11-*O*-aralkylcarbamoyl-3-*O*-descladinosylclarithromycin derivatives were designed, synthesized and evaluated for their *in vitro* antibacterial activity against various Gram-

positive and Gram-negative bacteria. Among them, a majority of the target compounds exerted potent activity against erythromycin-susceptible *S. pyogenes* and penicillin-susceptible *B. subtilis* ATCC9372, and significantly improved activity against the three phenotypes of resistant *S. pneumoniae* compared to CAM and AZM. In particular, compounds **11b** and **11c** possessed the best antibacterial activity against both sensitive and resistant strains including *S. pyogenes* (MIC $\leq$0.03 μg/mL), *S. aureus* ATCC25923 (MIC = 1 μg/mL), *B. subtilis* ATCC9372 (MIC = 1 μg/mL), *S. pneumoniae* A22072 (MIC $\leq$0.5 μg/mL), *S. pneumoniae* B1 (MIC $\leq$32 μg/mL) and *S. pneumoniae* AB11 (MIC = 4 μg/mL). Compounds **10d** (MIC = 16 μg/mL) displayed potent activity against *S. aureus* ATCC43300, showing 8-fold higher activity than CAM and AZM. Besides, compounds **11d** and **11h** (MIC = 32 μg/mL) exhibited the strongest antibacterial activity against susceptible *S. pyogenes* R1, 4-fold better than CAM and AZM. More importantly, the results demonstrated that the modification of both C-11 and C-3 positions of clarithromycin could dramatically enhance the antibacterial activity. Further optimization of clarithromycin derivatives with potent antibacterial activity should continue to be investigated.

Acknowledgments

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A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.bmcl.2018.06.006.

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16. The representative experimental procedure and analytical data of 11-*O*-(2-(3-(4-methylphenyl)propionamido)ethyl)carbamoyl-3-*O*-descladinosyl-3-ketoclarithromycin (**10e**). To a solution of 4-methylphenypropiolic acid (80 mg, 0.50 mmol) and HOBt (85 mg, 0.63 mmol) in THF (6.0 mL) was stirred at 0 °C for 30 min. Afterwards, DCC (130 mg, 0.63 mmol) was added to the solution. The resulting solution was allowed to stir at 0 °C for 6 h and then added **5** (300 mg, 0.42 mmol). The reaction mixture was stirred for 4 h at room temperature and evaporated in vacuum to dryness. The residue was dissolved in CH_2Cl_2 (15 mL) and then filtered. The resulting solution was washed by saturated $NaHCO_3$ solution and brine. The organic layers was combined and dried over anhydrous Na_2SO_4, filtered, and concentrated in vacuum to afford crude product. The above crude product was dissolved in methanol, and stirred for 24 h at 55 °C. The reaction solution was subsequently concentrated to provide the crude product. The above crude product was purified by flash column chromatography (dichloromethane/methanol, 30:1) to afford the desired product **10e** (151 mg, 37%) as white solids. mp 128–131 °C, R_f = 0.51 (dichloromethane/methanol, 10:1); ^{1}H NMR (400 MHz, $CDCl_3$, δ ppm): 7.42 (d, *J* = 8.1 Hz, 2H), 7.16 (d, *J* = 7.8 Hz, 2H), 5.01 (d, *J* = 11.1 Hz, 1H), 4.79 (s, 1H), 4.40 (d, *J* = 7.1 Hz, 1H), 4.11 (d, *J* = 10.2 Hz, 1H), 3.75 (d, *J* = 6.9 Hz, 2H), 3.55 (s, 1H), 3.45 (s, 1H), 3.26 (s, 1H), 2.81 (s, 3H), 2.38 (s, 3H), 2.34 (s, 1H), 2.18 (d, *J* = 6.3 Hz, 1H), 1.98 (s, 1H), 1.93 (s, 1H), 1.85 (s, 1H), 1.66 (q, *J* = 6.7 Hz, 3H), 1.48 (s, 3H), 1.32 (d, *J* = 7.2 Hz, 3H), 1.27 (t, *J* = 6.2 Hz, 15H), 1.23-1.19 (m, 6H), 0.89 (d, *J* = 7.2 Hz, 3H), 0.88-0.85 (m, 3H), 0.83 (s, 3H); ^{13}C NMR (150MHz, DMSO-d_6, *δ* ppm): 213.76, 212.75, 169.94, 156.48, 153.21, 140.79, 132.45, 130.75, 130.03, 129.59, 128.30, 117.22, 107.36, 104.33, 86.76, 84.05, 82.53, 79.81, 78.18, 70.65, 68.80, 64.97, 63.47, 52.28, 49.30, 47.63, 46.04, 30.25, 29.10, 26.78, 23.48, 23.08, 21.60, 21.54, 21.31, 19.55, 18.91, 16.14, 16.04, 15.08, 14.47, 11.67, 11.49; MS (ESI) *m/z* calcd. for $C_{43}H_{65}N_3O_{12}$ 815.5; found $[M+H]^+$ 817.5.
17. The representative experimental procedure and analytical data of 11-*O*-(2-(2-amino-4-nitrobenzamido)ethyl)carbamoyl-3-*O*-descladinosyl-3-*O*-(2-(3′-pyridyl)acetyl)clarithromycin (**11c**). To a solution of 2-amino-4-nitrobenzoic acid (80 mg, 0.44 mmol) and HOBt (73 mg, 0.54 mmol) in THF (6.0 mL) was stirred at 0 °C for 30 min. Afterwards, DCC (111 mg, 0.54 mmol) was added to the solution. The resulting solution was allowed to stir at 0 °C for 6 h and then added **8** (300 mg, 0.36 mmol). The reaction mixture was stirred for 4 h at room temperature and evaporated in vacuum to dryness. The residue was dissolved in CH_2Cl_2 (15 mL) and then filtered. The resulting solution was washed by saturated $NaHCO_3$ solution and brine. The organic layer was combined and dried over anhydrous Na_2SO_4, filtered, and concentrated in vacuum to afford crude product. The above crude product was dissolved in methanol, and stirred for 24 h at 55 °C. The reaction solution was subsequently concentrated to provide the crude product. The above crude product was purified by flash column chromatography (dichloromethane/methanol, 30:1) to afford the desired product **11c** (211 mg, 50%) as yellow solids. yield 50%, mp 127–130 °C, R_f = 0.23 (dichloromethane/methanol, 10:1); ^{1}H NMR (400 MHz, DMSO-d_6, δ ppm): 8.67 (t, *J* = 5.6 Hz, 1H), 8.51 (t, *J* = 2.5 Hz, 1H), 8.48 (d, *J* = 4.7 Hz, 1H), 8.02 (dd, *J* = 9.2, 2.9 Hz, 1H), 7.78 (d, *J* = 8.1 Hz, 2H), 7.72 (d, *J* = 8.4 Hz, 1H), 7.38 (td, *J* = 7.8, 4.7 Hz, 1H), 7.30-7.21 (m, 1H), 6.79 (d, *J* = 9.3 Hz, 2H), 5.50 (d, *J* = 7.9 Hz, 1H), 5.35 (d, *J* = 8.8 Hz, 1H), 4.95 (t, *J* = 8.9 Hz, 1H), 4.68 (dd, *J* = 8.5, 3.9 Hz, 1H), 4.39 (d, *J* = 7.6 Hz, 1H), 4.04 (s, 1H), 3.83 (s, 1H), 3.68-3.62 (m, 1H), 3.59-3.49 (m, 2H), 3.21 (d, *J* = 6.5 Hz, 2H), 3.06 (s, 1H), 3.00 (s, 3H), 2.35 (d, *J* = 3.8 Hz, 5H), 2.32 (s, 2H), 1.35 (s, 1H), 1.31 (s, 2H), 1.25-1.18 (m, 6H), 1.07 (s, 3H), 1.05 (s, 3H), 1.00 (d, *J* = 6.4 Hz, 6H), 0.95 (d, *J* = 7.2 Hz, 6H), 0.90 (d, *J* = 6.2 Hz, 3H), 0.87 (d, *J* = 7.1 Hz, 6H); ^{13}C NMR (150 MHz, DMSO-d_6, *δ* ppm): 202.89, 175.07, 170.54, 167.86, 156.91, 155.82, 150.94, 148.58, 137.60, 135.36, 127.84, 126.31, 123.84, 118.91, 116.31, 113.23, 85.28, 81.53, 77.82, 71.73, 70.34, 68.77, 67.62, 65.19, 63.11, 48.81, 47.66, 46.69, 46.36, 43.50, 37.95, 37.40, 36.71, 30.82, 30.47, 27.34, 23.96, 21.86, 21.41, 20.46, 19.12, 18.03, 16.53, 13.43, 12.30, 11.95, 11.50; MS (ESI) *m/z* calcd. for $C_{47}H_{70}N_6O_{15}$ 958.5; found $[M+H]^+$ 959.7, $[M+2H]^{2+}/2$ 480.6.