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Synthesis and antibacterial activity of novel ketolides with 11,12-sulfur contained aryl alkyl side chains

Xiao-zhuo Chen, Peng Xu, Lu Liu, Dan Zheng, Ping-sheng Lei*

Key Laboratory of Bioactivity Substance and Resources Utilization of Chinese Herbal Medicine, Ministry of Education, Institute of Materia Medica, Chinese Academy of Medical Sciences & Peking Union Medical College, Beijing 100050, China

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ABSTRACT

A novel series of ketolides with 11,12-sulfur contained aryl alkyl side chains were synthesized and evaluated for their antibacterial activity. These ketolides exhibited potent activity against key macrolide sensitive and resistant respiratory pathogens. The newly synthesized **9a**, **9e**, **9k** and **9n** showed a similar antimicrobial spectrum and comparable activity to telithromycin, the commercial ketolide antibacterial.

1. Introduction

Macrolide antibiotics have been used safely and effectively for treating community-acquired respiratory tract infections since 1950s. In recent years, with intensive emergence of antibacterial resistance against antibiotics [1], great efforts have been made to find new structures which are potent against resistant pathogens. The typical third generation macrolide [2], telithromycin for example, has been launched to the market [3]. Another ketolide, cethromycin, has been approved by FDA and now is in late-stage development for the treatment of community acquired pneumonia and infections with biodefense pathogens [4]. The advantage of ketolides is that they cover macrolide sensitive and resistant strains and are poor inducers of the *erm* resistance mechanism as well as *mef* efflux-mediated resistance [5,6]. However, with the use of telithromycin, some safety concerns emerged [7]. So new drug developments are crucially needed (Fig. 1).

Both telithromycin and cethromycin comprise the same key structure features: a 3-keto group and an 11,12-carbamate functionality. In addition, they have a proper aryl alkyl side chain which can interact with nucleotide A752 in domain II of the 23S rRNA [8]. In recent years, many ketolides have been synthesized in order to find macrolides with improved activity and low toxicity. For instance, the fluoroketolide antibiotic **CEM-101** (OPT-1068; Cempra Pharmaceuticals) is to enter into phase II investigation in 2010 [9]. The azetidinyl ketolides **Thomas V-6s** synthesized by Pfizer, Inc. with low hepatotoxicity has come into phase Iclinical trials [10]. The molecule of **Thomas V-6s** showed that a tiny change in the 11,12-side chain made notable decrease of hepatotoxicity.

In our previous work [11], the introduction of a sulfur atom to the 11,12-cyclic carbamate side chains retained the antibacterial activity against erythromycin sensitive and resistant test strains. Compared to telithromycin, the 14-membered ketolide with imidazo[4,5-b]pyridinyl sulfur contained alkyl side chain **Xu-15a** demonstrated improved *in vitro* activity against erythromycin susceptible and resistant strains including *Streptococcus pneumoniae* (Ery-S) and *Streptococcus pyogenes* (Ery-R).

For sulfur atom is slightly larger than carbon atom and the electron density is also higher, inserting a sulfur atom to carbon chains could slightly adjust the length and nucleophilicity of the side chain in macrolides. Hunziker et al. [12] developed a series of novel ketolides with a fused five-membered lactone ring and S-alkyl or aryl substituted α-thio acetic acid side chain. These sulfur contained ketolide products exhibited high activity against a representative set of erythromycin sensitive and resistant strains.

* Corresponding author. Institute of Materia Medica, Peking Union Medical College & Chinese Academy of Medical Sciences, Room 408, East building, No. 1 Xia Xuan Wu district, Beijing 100050, PR China. Tel.: +86 10 63162006; fax: +86 10 63017757.
E-mail address: lei@imm.ac.cn (P.-s. Lei).

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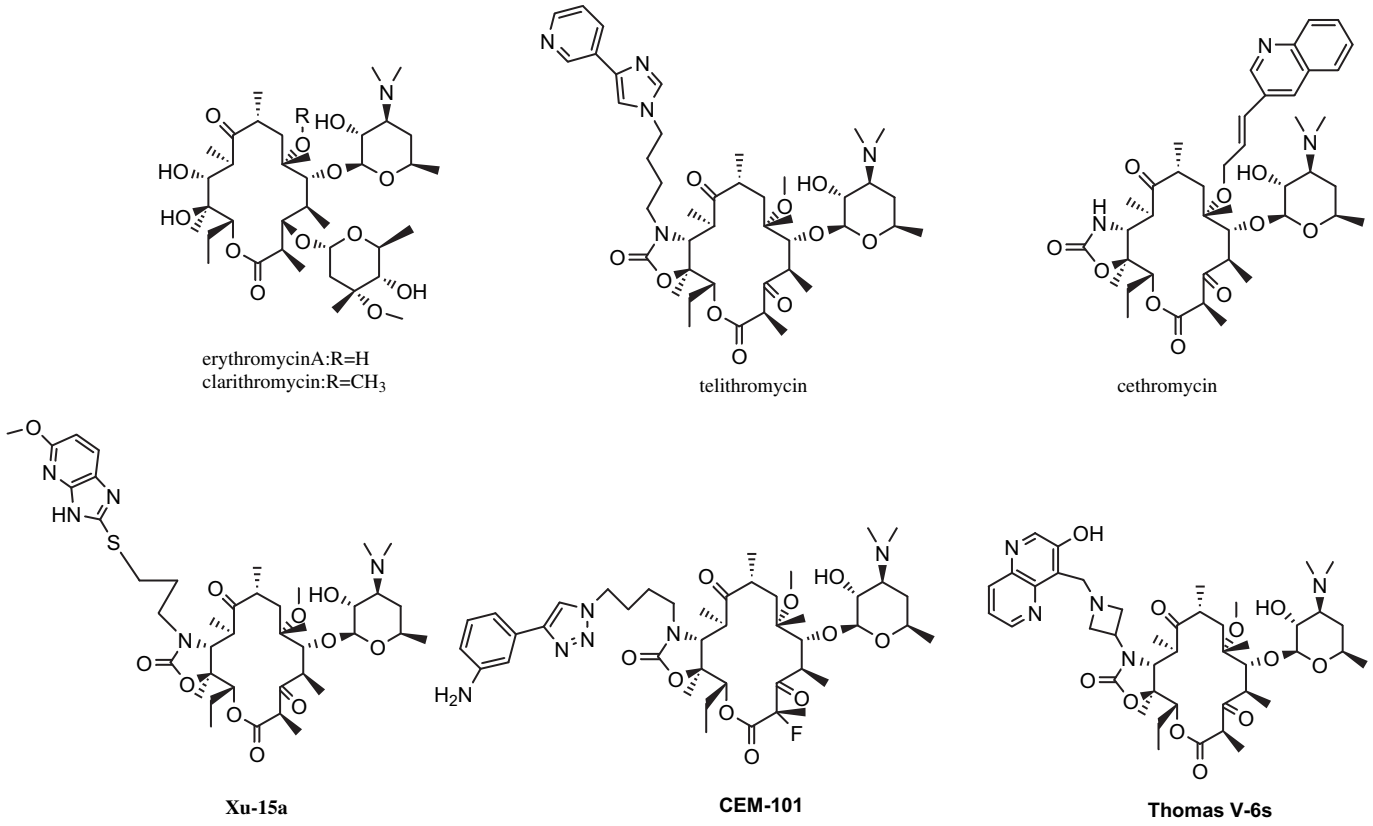


Fig. 1. Structures of representative macrolides.

The best compound showed a similar antimicrobial spectrum and comparable *in vitro* activity to telithromycin.

It is reported that, the hepatotoxicity can be decreased by increasing protonation or diminishing the hydrophobicity of the erythromycin structure [13]. It is presumed that by varying the side chain with different protonation contained heterocyclics, novel ketolides with improved antibacterial activity or lower hepatotoxicity could be found.

We sought to further optimize the 11,12-cyclic carbamate sulfur contained aryl alkyl side chains and to introduce several new heterocyclic groups in the side chain. In this report, a novel series of ketolides were described with the aim of finding new macrolide antibiotics with higher antibacterial activity and/or improved pharmacokinetics.

2. Results and discussion

2.1. Chemistry

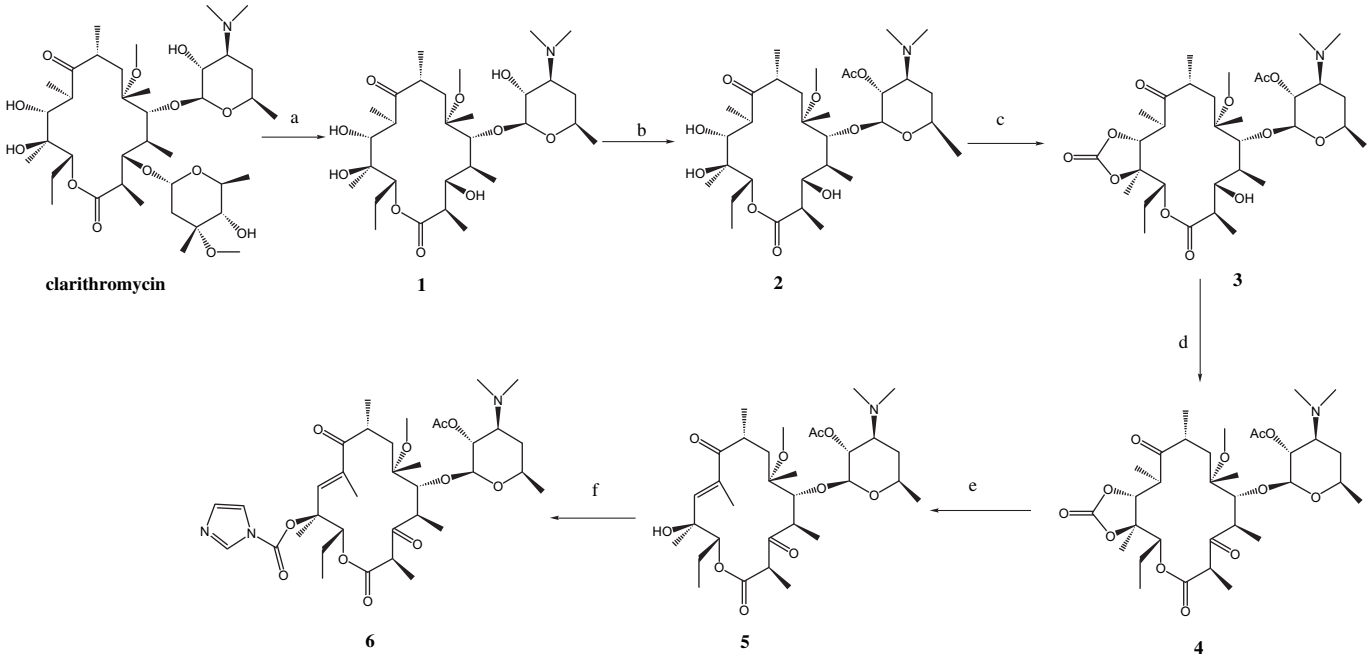
The synthesis of **6**, an important intermediate of ketolides, started from commercially available clarithromycin (Scheme 1). Decladinose on the 3-position to form the 3-hydroxy group **1** in a yield of 96%, followed by acetylation of the desosamine to form **2** in a yield of 86%. Formation of 11,12-carbonate **3** was carried out with trichloromethyl chloroformate in a mixture of CH_2Cl_2 and pyridine at 0 °C in 75% yield. The Swern Oxidation was carried to convert the 3-hydroxy group to the carbonyl group **4** in a yield of 76%. The double bond on the 10,11-positions was formed underwent smoothly elimination by treating **4** with 1,8-Diazabicyclo [5.4.0] undec-7-ene (DBU) in acetone to afford **5** in a yield of 81%. 1,1′-carbonyldiimidazole (CDI) was used to transform of the 11-hydroxyl to the acyl imidazole **6** in a yield of 82%. The structure of this key intermediate was characterized by its ^{1}H NMR, ^{13}C NMR and HRMS. In our work, simple isolation and purification procedures were used in the last two steps. For instance, the product of **5** was recrystallized from ethyl acetate as a white solid without chromatography [14]. In the preparation of **6**, the great quantity of solvent (DMF) was not easy to remove under vacuum. The reaction mixture was poured into ice water after the reaction was completed. The crude product of **6** was obtained as white solid, which was separated easily by filtration.

The synthesis of sulfur contained aryl alkyl side chains was depicted in Scheme 2 using the method published previously [15]. A series of heterocyclic with a mercaptan substituent were treated with one equivalent of N-(3-bromopropyl) phthalimide in DMF at 90 °C to afford products **7a**–**r** with a yield of 70–99%. After removing the phthalimide group of **7a**–**r** in a solution of 85% aqueous hydrazine in ethanol, a series of free amines **8a**–**r** were obtained.

The 11,12-cyclic carbamate ketolides with sulfur contained aryl alkyl side chains **9a**–**o** were obtained from the reaction of **6** with excessive **8a**–**o** in 10% aqueous acetonitrile at 30–40 °C [16]. The acetyl group was removed to afford **9a**–**o** by refluxing in CH_3OH without any additional base (Scheme 3).

Some uncyclized *O*-carbamate ketolides with sulfur contained aryl alkyl side chains **10a**–**c** were also obtained under this procedure. The HRMS showed the same molecular weight as the 11,12-cyclic carbamate products. But the ^{1}H NMR signals for the 11-H of **10a**–**c** appeared at around 6.76 ppm, while it was shifted upfield to 3.54 ppm in cyclized compounds. The low field signal indicated the existence of 10,11-ene. A broad single signal for the N–H was found at about 5.50 ppm. And the ^{13}C NMR signals for C-9 were about 10 ppm higher than that in cyclized compounds, which were in accord with the shift of α, β-unsaturated ketones (Scheme 4).

Considering the base-catalyzed mechanism of Michael reaction, the uncyclized O-carbamates **10a**–**c** were treated with sodium hydride in THF. Unfortunately, no cyclic carbamate was formed.



Scheme 1. Preparation of compound **6**.

2.2. Pharmacology

The obtained compounds **9a**–**o** and **10a**–**c** were tested against a panel of representative respiratory tract pathogens together with telithromycin and clarithromycin as references (Table 1 and **2**). Various macrolide sensitive and resistant strains were included in order to evaluate the activity of this novel series of macrolide derivatives.

ATCC29213 is a methicillin-sensitivity *Staphylococcus aurous* (MSSA). ANS46 (SCCmecIII) is methicillin-resistant *Staphylococcus aurous* (MRSA). 07C134 and 07T202 are methicillin-sensitivity *Staphylococcus epidermidis* (MSSE). 07R066 and 07A006 are methicillin-resistant *Staphylococcus epidermidis* (MRSE). ATCC49619 and 07H252 are erythromycin-sensitivity *Streptococcus pneumoniae* (ESSP). 07D201 and 07P390 are erythromycin-resistant *Streptococcus pneumoniae* (ERSP), 07D201 was encoded by both *ermB* and *mef* genes while 07P390 was encoded by only *ermB* gene. 07U084 and 07U086 are erythromycin-sensitivity *Streptococcus pyogenes* (ESSPy); 03-233 and 01-533 are erythromycin-resistant *Streptococcus pyogenes* (ERSPy), which are encoded by the *ermB* and *mef* genes, respectively. ATCC49247, 07O114, 07O339 and 07O394 are *Haemophilus influenzae* strains which were sensitive to erythromycin.

All the strains chosen in this test were supplied by the Ministry of Health National Antimicrobial Resistance Investigation Net (MOHNARIN, China). The *in vitro* antibacterial activity was reported as minimum inhibitory concentrations (MICs), which was determined by the broth microdilution method as recommended by the NCCLS [17].

All the tested ketolides with 11,12-cyclic carbamate sulfur contained side chains showed potent antibacterial activity against MSSA strain ATCC29213.

It is interesting that, against the methicillin-resistant strains of *S. epidermidis* 07R066 and 07A006, all the tested compounds showed potent activity, while against the methicillin-sensitivity strain of *S. epidermidis* 07T202, all the tested compound showed weak antibacterial activity. This could explain by the different resistant mechanisms of bacteria to methicillin and macrolides. It is worth notice that strain 07A006 was resistant to clarithromycin (MIC 8 mg/L), but sensitive to telithromycin and all the synthesized ketolides. **9a**, **9e**, **9f**, **9k** and **9n** showed excellent activity which were on equality with telithromycin against 07A006.

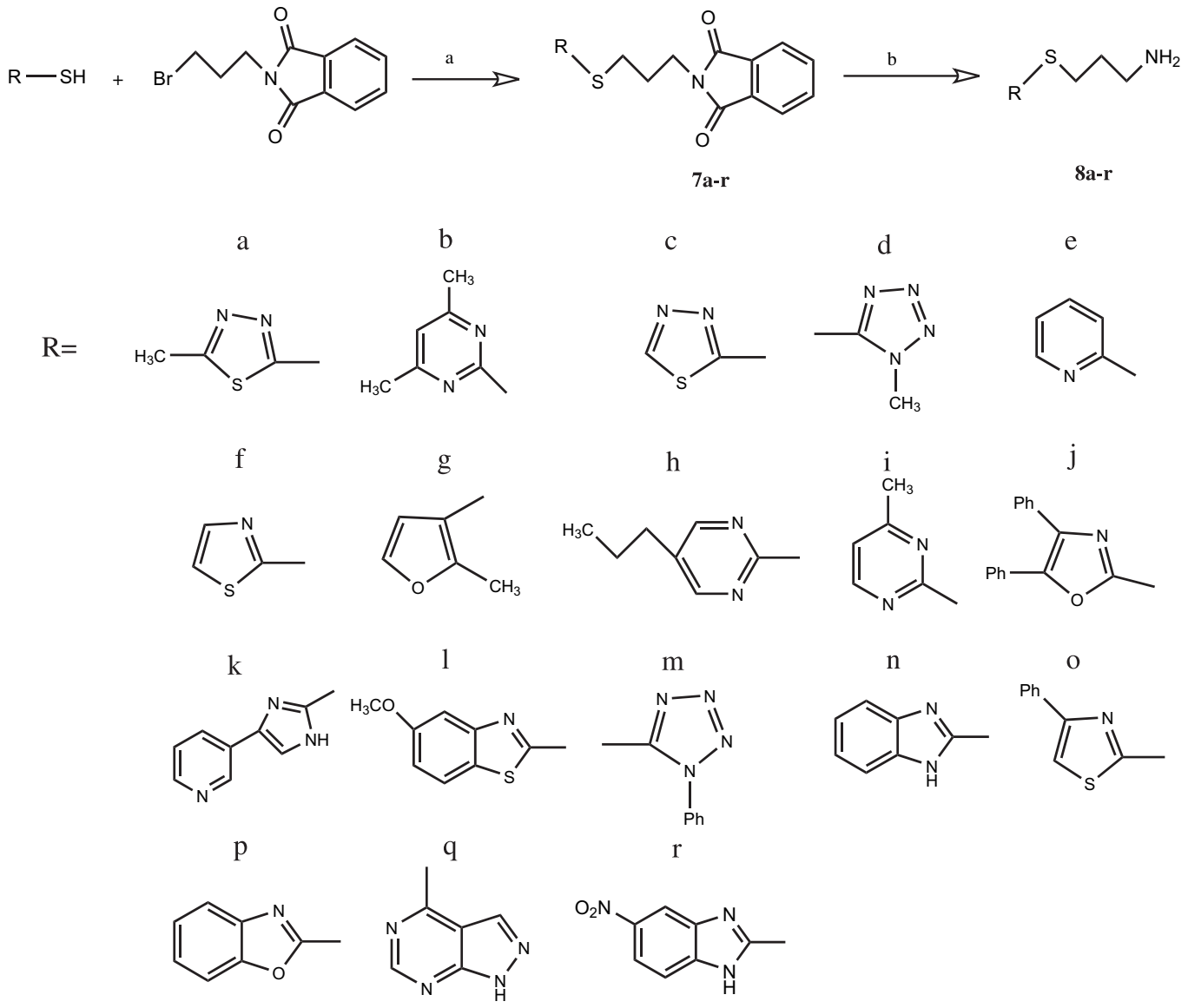
Against erythromycin-sensitivity *S. pyogenes* 07U084 and 07U086, the synthesized ketolides also showed excellent antibacterial activity. The MIC of **9a**, **9e**, **9f**, **9n** were 4 folds lower compared to telithromycin, **9k** was 2 times more potent than telithromycin. Compared to telithromycin, compound **9k** demonstrated improved (2 folds lower) activity against erythromycin-resistant *S. pyogenes* (ERSPy) of 03-233 and 01-533. All the tested compounds showed potent activity (MIC$<$4 mg/L) against strain 01-533 which was encoded by *mef*-gene.

All the tested compounds showed excellent activities against erythromycin-sensitivity *S. pneumoniae* ATCC49619, but against 07H252 which was also sensitive to clarithromycin, the ketolides we prepared showed weak activities except compound **9a** ($\leq$0.016 mg/L). Against erythromycin-resistant *S. pneumoniae* 07D201 and 07P390, the tested compounds showed improved activity compared to clarithromycin, **9k** has nearly equal activity with telithromycin.

All the compounds were tested against four *H. influenzae* strains. **9a**, **9c**, **9f**, **9k**, **9n** showed equal or improved activity compared to telithromycin.

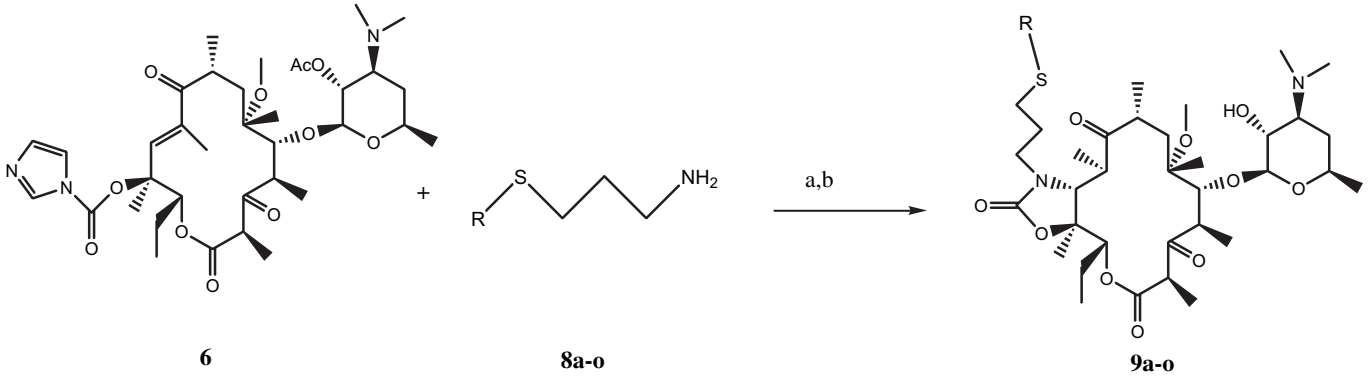
The antibacterial activity of uncyclized *O*-carbamate ketolides **10a**–**c** was weak. This phenomena confirmed that the 11,12-cyclic carbamate was a very important factor for the improvement of the macrolide antibacterial activity. This 5-member ring could increase the rigidity of the ketolide conformation. The carbamate group could form a network of hydrophobic interactions with domain V of 23S rRNA; And N2 of the carbamate group could form a hydrogen bond with O4 of U2588DR (U2609EC) in domain V of 23S rRNA. These interactions seem to be the main contributors for the enhancement of the binding of ketolides to the ribosome [18].

From Tables 1 and 2, some structure–activity relationships could be summarized. First at all, among the various ketolide derivatives,

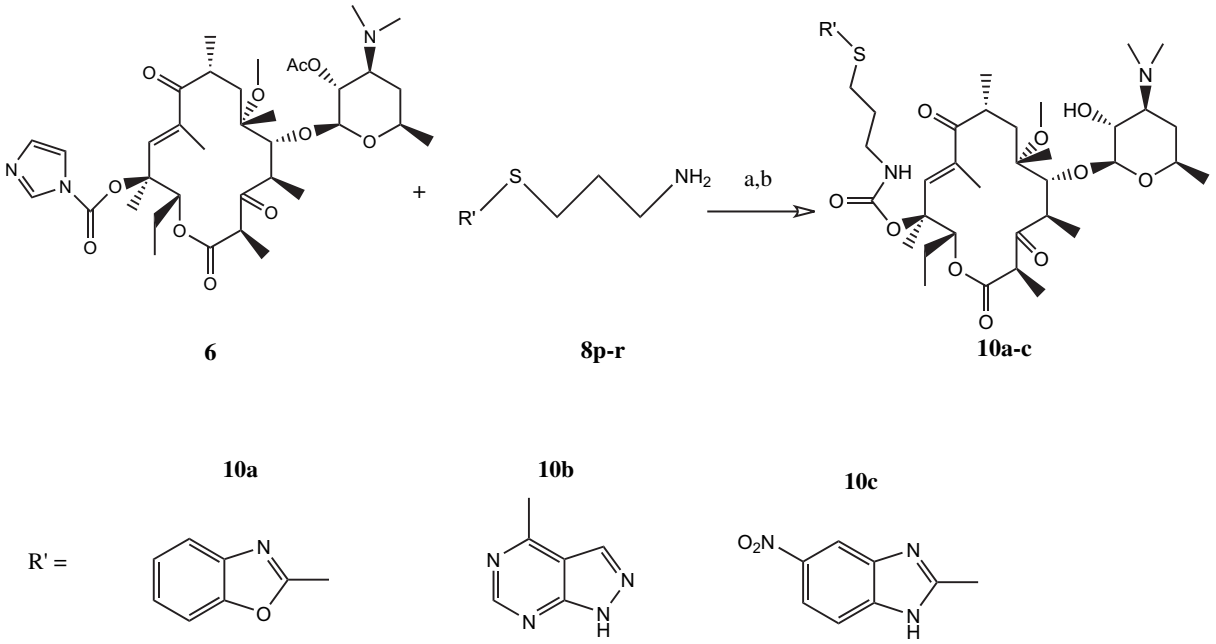


Scheme 2. Preparation of compounds **8a**–**r**.

9k and **9n** exhibited the most potent activity against both susceptible and resistant bacteria. It was revealed that introduction of sulfur atom to the 11,12-cyclic carbamate side chains did make sense for further research in macrolide antibiotics. Then, too many nitrogen atoms resulted in no improvement of activity. For instance, there are four nitrogen atoms contained in the aryl alkyl side chains of **9d** and **9m**, both of which showed weaker activity against susceptible and resistant bacteria. The last, compared **9j** with **9g**,



Scheme 3. Preparation of compounds **9a**–**o**.



Scheme 4. Preparation of compounds **10a**–**c**.

Table 1
In vitro antibacterial activity [MICs (mg/L)] of synthesized ketolides against selected respiratory pathogens(1).

	S. aureus		*S. epidermidis*				*S. pyogenes*			
	ATCC29213	ANS46	07C134	07T202	07R066	07A006	07U084	07U086	03-233	01-533
9a	0.25	>32	0.125	>32	2	0.125	≤0.016	0.031	>32	1
9b	0.5	>32	0.5	>32	0.5	0.5	0.125	0.125	>32	2
9c	0.25	>16	0.25	>16	0.5	0.25	0.062	0.062	>16	1
9d	0.25	>32	0.25	>32	>32	1	0.062	0.062	>32	1
9e	0.125	>16	0.125	>16	0.125	0.125	0.016	0.016	nt	nt
9f	0.125	>32	0.125	>32	0.062	0.125	≤0.016	≤0.016	>32	0.5
9g	0.25	>16	0.25	>16	0.25	0.25	0.125	0.125	>16	0.5
9h	0.5	>16	0.5	>16	0.5	1	0.125	0.125	nt	nt
9i	0.5	>32	0.25	>32	0.25	0.5	0.125	0.125	2	2
9j	4	>16	4	>16	4	2	0.5	0.5	nt	nt
9k	0.125	>32	0.125	32	0.125	0.125	0.031	0.031	1	0.125
9l	0.5	>16	0.25	16	0.25	0.5	0.062	0.062	nt	nt
9m	0.25	>32	0.5	>32	0.25	0.5	0.125	0.125	>32	2
9n	0.125	>16	0.062	>16	0.062	0.125	0.016	0.016	nt	nt
9o	0.5	32	0.5	32	0.5	0.5	0.25	0.25	32	1
10a	4	>16	4	>16	4	4	2	2	>16	>16
10b	0.5	>16	0.5	>16	0.5	0.5	0.25	0.25	>16	8
10c	1	>16	1	>16	1	1	0.125	0.125	nt	nt
Cla	0.25	>32	0.25	>32	0.125	8	0.031	0.031	>32	2
Teli	0.125	>16	0.125	>16	0.125	0.125	0.062	0.062	2	0.25

adding two phenyls to the heterocyclic made activity against both susceptible and resistant bacteria significantly descend. The similar trend could be seen from the comparison between **9o** and **9f**.

3. Conclusion

In summary, a series of ketolides with 11,12-sulfur contained aryl alkyl side chains were synthesized. Some of the newly prepared ketolides displayed excellent *in vitro* antimicrobial activity against macrolide sensitive and resistant pathogens. Against erythromycin sensitivity and resistant *S. pyogenes*, erythromycin-sensitivity *S. pneumoniaes*, erythromycin-sensitivity *H. influenzaes*, **9n** and **9k** showed better antibacterial activity than telithromycin. **9a**, **9e**, **9k** and **9n** have *in vitro* antibacterial spectrum similar to telithromycin and are potent against *erm* and *mef*-gene containing resistant strains. In summary, the novel class of ketolides presented in our laboratory emerged as a valuable lead series that might be useful in the fight against macrolide resistance pathogens. These ketolides are promising candidates for further efficacy evaluation.

4. Experimental section

4.1. General chemistry methods

All NMR spectra were recorded on Mercury-300 spectrometers in $CDCl_3$ or DMSO. The chemical shifts are reported impart per million (ppm) with tetramethylsilane (TMS) as an internal standard. HRMS experiments were performed using an Aglient 1100 series LC/MSD TOF. Analytical thin layer chromatography (TLC) was carried out on TLC plates silica gel $HSGF_{254}$ percolated by Branch of Qingdao Haiyang Chemical Plant. Chromatography was performed with silica gel H (HG/T2354-92). The major chemicals were

Table 2
In vitro antibacterial activity [MICs (mg/L)] of synthesized ketolides against selected respiratory pathogens (2).[a]

	S. pneumoniae				*H. influenzae*			
	ATCC49619	07H252	07D201	07P390	ATCC49247	07O114	07O339	07O394
9a	≤0.016	≤0.016	16	0.125	2	2	2	2
9b	0.062	2	>32	1	4	8	4	8
9c	≤0.016	2	>16	0.5	2	2	2	2
9d	0.031	8	>32	0.5	8	8	8	8
9e	0.016	nt	nt	nt	8	8	4	8
9f	0.031	0.5	>32	0.25	4	4	4	4
9g	0.031	1	>16	0.25	16	8	16	16
9h	0.125	nt	nt	nt	16	16	16	16
9i	0.062	8	>32	0.25	8	8	8	8
9j	1	nt	nt	nt	>16	>16	>16	>16
9k	0.062	0.25	4	0.031	2	2	2	2
9l	0.125	nt	nt	nt	16	16	16	16
9m	0.125	1	>32	0.5	16	16	32	32
9n	0.016	nt	nt	nt	4	2	2	2
9o	0.25	0.5	8	0.25	16	32	32	32
10a	2	16	>16	>16	>16	>16	>16	>16
10b	0.5	8	>16	16	>16	>16	>16	>16
10c	0.125	nt	nt	nt	16	16	>16	>16
Cla	0.031	0.062	>32	2	4	4	8	8
Teli	0.031	0.062	1	0.031	4	4	4	4

[a] 'nt' = not test.

purchased from Alfa Chemical Corporation. All other chemicals were analytical grade. All reactions were performed under argon unless otherwise noted.

Clarithromycin for antibacterial testing was obtained from HUAYI Pharmaceutical Co., China. Bacterial strains were from the American Type Culture Collection (ATCC) as indicated in Tables 1 and 2, or were clinical isolates from the Ministry of Health national antimicrobial resistance investigation net, China (MOHNARIN, China).

4.2. General procedure for compounds **7a**–**r**

To a suspension of mercaptan substituent heterocyclic in DMF, one equivalent of N-(3-bromopropyl) phthalimide and K_2CO_3 (1 equivalent) were added. The reaction was stirred for 6 h at 90 °C before cooled to room temperature. After evaporation of the DMF under vacuum, the residue was diluted with methylene chloride, washed with water and brine, and dried over $MgSO_4$. After evaporation of the solution, the crude products were purified by column chromatography eluting with 2.5:1 petroleum ether/acetone to afford the desired products as yellow or white solid.

4.2.1. 2-(3-(5-Methyl-1,3,4-thiadiazole-2-thio) propyl) 1H-isoindole-1,3(2H)-dione (**7a**)

Yield: 99%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.384 (m, 2H), 7.714 (m, 2H), 3.846 (t, J = 6.9 Hz, 2H), 3.334 (t, J = 7.2 Hz, 2H), 2.702 (s, 3H), 2.195 (m, 2H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 168.31, 164.95, 134.03, 132.00, 123.31, 36.66, 31.16, 28.34, 15.60. HRMS (ESI) m/z found 320.0537, calcd for $C_{14}H_{14}N_3O_2S_2$ $(M+H)^+$ 320.0527.

4.2.2. 2-(3-(4,6-Dimethylpyrimidine-2-thio) propyl) 1H-isoindole-1,3(2H)-dione (**7b**)

Yield: 98%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.832 (m, 2H), 7.702 (m, 2H), 6.634 (s, 1H), 3.828 (t, J = 6.9 Hz, 2H), 3.147 (t, J = 7.8 Hz, 2H), 2.314 (s, 6H), 2.124 (m, 2H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 168.31, 166.92, 133.87, 132.10, 123.17, 115.56, 37.24, 28.75, 27.87, 23.73. HRMS (ESI) m/z found 328.1107, calcd for $C_{17}H_{18}N_3O_2S$ $(M+H)^+$ 328.1120.

4.2.3. 2-(3-(1,3,4-Thiadiazole-2-thio) propyl) 1H-isoindole-1,3(2H)-dione (**7c**)

Yield: 80%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 8.988 (s, 1H), 7.847 (m, 2H), 7.721 (m, 2H), 3.867 (t, J = 6.9 Hz, 2H), 3.415 (t, J = 6.9 Hz, 2H), 2.234 (m, 2H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 168.32, 151.30, 134.04, 131.98, 123.33, 36.63, 31.32, 28.28. HRMS (ESI) m/z found 306.0351, calcd for $C_{13}H_{12}N_3O_2S_2$ $(M+H)^+$ 306.0365.

4.2.4. 2-(3-(1-Methyltetrazole-5-thio) propyl) 1H-isoindole-1,3(2H)-dione (**7d**)

Yield: 88%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.848 (m, 2H), 7.732 (m, 2H), 3.920 (s, 3H), 3.862 (t, J = 6.9 Hz, 2H), 3.380 (t, J = 6.9 Hz, 2H), 2.215 (m, 2H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 168.35, 153.91, 134.12, 131.92, 123.34, 36.26, 33.36, 30.65, 28.46. HRMS (ESI) m/z found 304.0852, calcd for $C_{13}H_{14}N_5O_2S$ $(M+H)^+$ 304.0863.

4.2.5. 2-(3-(Pyridine-2-thio) propyl) 1H-isoindole-1,3(2H)-dione (**7e**)

Yield: 99%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 8.276 (dt, J = 5.1, 0.9 Hz, 1H), 7.822 (dd, J = 5.4, 3.0 Hz, 2H), 7.691 (dd, J = 5.4, 3.0 Hz, 2H), 7.416 (t, J = 7.8 Hz, 1H), 7.120 (dd, J = 7.8, 0.9 Hz, 1H), 6.907 (dd, J = 7.8, 5.1 Hz, 1H), 3.821 (t, J = 6.6 Hz, 2H), 3.189 (t, J = 6.6 Hz, 2H), 2.084 (m, 2H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 168.28, 158.40, 149.30, 135.73, 133.84, 132.06, 123.13, 122.30, 119.27, 37.04, 28.60, 26.98. HRMS (ESI) m/z found 299.0959, calcd for $C_{16}H_{15}N_2O_2S$ $(M+H)^+$ 299.0916.

4.2.6. 2-(3-(Thiazole-2-thio) propyl) 1H-isoindole-1,3(2H)-dione (**7f**)

Yield: 93%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.842 (m, 2H), 7.715 (m, 2H), 7.595 (d, J = 3.3 Hz, 1H), 7.192 (d, J = 3.3 Hz, 1H), 3.838 (t, J = 6.9 Hz, 2H), 3.262 (t, J = 6.9 Hz, 2H), 2.154 (m, 2H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 168.31, 142.42, 133.99, 132.03, 123.28, 118.94, 36.77, 31.65, 28.46. HRMS (ESI) m/z found 305.0411, calcd for $C_{14}H_{13}N_2O_2S_2$ $(M+H)^+$ 305.0418.

4.2.7. 2-(3-(2-Methylfurane-3-thio) propyl) 1H-isoindole-1,3(2H)-dione (**7g**)

Yield: 80%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.828 (m, 2H), 7.701 (m, 2H), 7.243 (d, J = 1.5 Hz, 1H), 6.352 (d, J = 1.5 Hz, 1H), 3.779 (t, J = 6.9 Hz, 2H), 2.636 (t, J = 7.2 Hz, 2H), 2.335 (s, 3H), 1.885 (m, 2H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 168.26, 155.18, 140.57, 133.92, 132.03, 123.19, 114.99, 36.84, 33.21, 28.36, 11.75. HRMS (ESI) m/z found 302.0797, calcd for $C_{16}H_{16}N_3S$ $(M+H)^+$ 302.0845.

*4.2.8. 2-(3-(5-n-Propylpyrimidine-2-thio) propyl) 1H-isoindole-1,3 (2H)-dione (**7h**)*

Yield: 99%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 8.262(s, 2H), 7.844 (dd, J = 5.4, 3.0 Hz, 2H), 7.712 (dd, J = 5.4, 3.0 Hz, 2H), 3.839 (t, J = 6.9 Hz, 2H), 3.157 (t, J = 6.9 Hz, 2H), 2.462 (t, J = 7.2 Hz, 2H), 2.132 (m, 2H), 1.596 (m, 2H), 0.935 (t, 3H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 168.37, 157.16, 133.92, 132.12, 130.07, 123.20, 37.18, 31.74, 28.62, 28.02, 23.90, 13.47. HRMS (ESI) *m*/*z* found 342.1265, calcd for $C_{18}H_{20}N_3O_2S$ $(M+H)^+$ 342.1271.

*4.2.9. 2-(3-(4-Methylpyrimidine-2-thio) propyl) 1H-isoindole-1,3 (2H)-dione (**7i**)*

Yield: 96%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 8.251 (d, J = 4.8 Hz, 1H), 7.835 (dd, J = 5.4, 3.3 Hz, 2H), 7.704 (dd, J = 5.4, 3.3 Hz, 2H), 6.773 (d, J = 4.8 Hz, 1H), 3.832 (t, J = 6.6 Hz, 2H), 3.153 (t, J = 6.6 Hz, 2H), 2.379 (s, 3H), 2.128 (m, 2H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 171.25, 168.31, 167.50, 156.62, 133.89, 132.09, 123.17, 116.08, 37.18, 28.62, 27.93, 23.97. HRMS (ESI) *m*/*z* found 314.0958, calcd for $C_{16}H_{16}N_3O_2S$ $(M+H)^+$ 314.0958.

*4.2.10. 2-(3-(4,5-Diphenyloxazole-2-thio) propyl) 1H-isoindole-1,3 (2H)-dione (**7j**)*

Yield: 98%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.836 (dd, J = 5.4, 3.0 Hz, 2H), 7.704 (dd, J = 5.4, 3.0 Hz, 2H), 7.632 (d, J = 2.1 Hz, 1H), 7.607 (d, J = 2.1 Hz, 1H), 7.553 (d, J = 2.1 Hz, 1H), 7.527 (d, J = 2.1 Hz, 1H), 7.330 (m, 6H), 3.891 (t, J = 6.9 Hz, 2H), 3.292 (t, J = 6.9 Hz, 2H), 2.266 (m, 2H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 168.31, 158.80, 147.12, 136.27, 133.96, 132.00, 128.61, 128.50, 128.16, 127.86, 126.30, 123.26, 36.74, 29.81, 28.83. HRMS (ESI) *m*/*z* found 441.1265, calcd for $C_{26}H_{21}N_2O_3S$ $(M+H)^+$ 441.1267.

*4.2.11. 2-(3-(4-(3-Pyridine)-1H-imidazole-2-thio) propyl) 1H-isoindole-1,3(2H)-dione (**7k**)*

Yield: 84%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 8.955 (d, J = 1.5 Hz, 1H), 8.470 (dd, J = 4.8, 1.5 Hz, 1H), 8.087 (d, J = 7.8 Hz, 1H), 7.839 (dd, J = 5.4, 3.3 Hz, 2H), 7.718 (dd, J = 5.4, 3.3 Hz, 2H), 7.461 (s, 1H), 7.299 (dd, J = 5.1, 7.8 Hz, 1H), 3.946 (t, J = 7.2 Hz, 2H), 3.038 (t, J = 7.2 Hz, 2H), 1.977 (m, 2H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 168.64, 147.69, 146.08, 134.19, 132.29, 131.90, 123.65, 123.39, 36.03, 32.71, 28.19. HRMS (ESI) *m*/*z* found 365.1074, calcd for $C_{19}H_{17}N_4O_2S$ $(M+H)^+$ 365.1072.

*4.2.12. 2-(3-(5-Methoxybenzothiazole-2-thio) propyl) 1H-isoindole-1,3(2H)-dione (**7l**)*

Yield: 99%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.840 (dd, J = 5.4, 3.0 Hz, 2H), 7.710 (dd, J = 5.4, 3.0 Hz, 2H), 7.553 (d, J = 9.0 Hz, 1H), 7.275 (d, J = 2.4 Hz, 1H), 6.910 (dd, J = 9.0, 2.4 Hz, 1H), 3.872 (t, J = 6.9 Hz, 2H), 3.843 (s, 3H), 3.371 (t, J = 6.9 Hz, 2H), 2.232 (m, 2H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 168.28, 167.42, 158.83, 154.26, 133.96, 131.98, 126.76, 123.25, 121.07, 113.89, 104.46, 55.56, 36.77, 30.61, 28.51. HRMS (ESI) *m*/*z* found 385.0668, calcd for $C_{19}H_{17}N_2O_3S_2$ $(M+H)^+$ 385.0675.

*4.2.13. 2-(3-(1-Phenyl-tetrazole-5-thio) propyl) 1H-isoindole-1,3 (2H)-dione (**7m**)*

Yield: 88%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.838 (m, 2H), 7.720 (m, 2H), 7.552 (m, 5H), 3.856 (t, J = 6.6 Hz, 2H), 3.425 (t, J = 7.2 Hz, 2H), 2.249 (m, 2H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 168.34, 134.07, 131.92, 130.09, 129.77, 123.81, 123.31, 36.38, 30.60, 28.31. HRMS (ESI) *m*/*z* found 366.1020, calcd for $C_{18}H_{16}N_5O_2S$ $(M+H)^+$ 366.1025.

*4.2.14. 2-(3-(Benzoxazole-2-thio) propyl) 1H-isoindole-1,3(2H)-dione (**7n**)*

Yield: 70%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.798 (dd, J = 5.4, 3.0 Hz, 2H), 7.686 (dd, J = 5.4, 3.0 Hz, 2H), 7.478 (dd, J = 6.0, 3.0 Hz, 2H), 7.167 (dd, J = 6.0, 3.0 Hz, 2H), 3.862 (t, J = 6.6 Hz, 2H), 3.323 (t, J = 6.6 Hz, 2H), 2.135 (m, 2H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 168.44, 149.33, 133.99, 131.90, 123.25, 122.30, 114.12, 36.45, 30.23, 28.55. HRMS (ESI) *m*/*z* found 338.0961, calcd for $C_{18}H_{16}N_3O_2S$ $(M+H)^+$ 338.0963.

*4.2.15. 2-(3-(4-Phenylthiazole-2-thio) propyl) 1H-isoindole-1,3 (2H)-dione (**7o**)*

Yield: 97%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.696 (m, 4H), 7.562 (m, 2H), 7.224 (d, J = 7.5 Hz, 1H), 7.170 (d, J = 5.1 Hz, 2H), 7.113 (m, 1H), 3.733 (t, J = 6.9 Hz, 2H), 3.185 (t, J = 6.9 Hz, 2H), 2.098 (m, 2H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 168.29, 163.99, 155.26, 133.96, 132.00, 128.64, 128.07, 126.23, 123.26, 112.35, 36.86, 31.61, 28.57. HRMS (ESI) *m*/*z* found 381.0717, calcd for $C_{20}H_{17}N_2O_2S_2$ $(M+H)^+$ 381.0726.

*4.2.16. 2-(3-(Benzoxazole-2-thio) propyl) 1H-isoindole-1,3(2H)-dione (**7p**)*

Yield: 82%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.863 (m, 2H), 7.736 (m, 2H), 7.488 (dd, J = 6.9, 1.8 Hz, 1H), 7.408 (dd, J = 6.9, 1.8 Hz, 1H), 7.229 (m, 2H), 3.895 (t, J = 6.6 Hz, 2H), 3.342 (t, J = 7.2 Hz, 2H), 2.266 (m, 2H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 168.37, 164.54, 151.87, 141.87, 134.04, 132.04, 124.21, 123.81, 123.34, 118.35, 109.77, 36.67, 29.47, 28.68. HRMS (ESI) *m*/*z* found 339.0815, calcd for $C_{18}H_{15}N_2O_3S$ $(M+H)^+$ 339.0803.

*4.2.17. 2-(3-(Pyrazolo [3,4d] pyrimidine-4-thio) propyl) 1H-isoindole-1,3(2H)-dione (**7q**)*

Yield: 70%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 8.649 (s, 1H), 8.123 (s, 1H), 7.866 (dd, J = 5.4, 3.0 Hz, 2H), 7.735 (dd, J = 5.4, 3.0 Hz, 2H), 3.891 (t, J = 6.9 Hz, 2H) 3.442 (t, J = 6.9 Hz, 2H), 2.212 (m, 2H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 168.43, 165.91, 153.64, 151.67, 134.07, 132.97, 132.03, 123.31, 36.83, 28.55, 26.28. HRMS (ESI) *m*/*z* found 340.0865, calcd for $C_{16}H_{14}N_5O_2S$ $(M+H)^+$ 340.0862.

*4.2.18. 2-(3-(5-Nitrobenzimidazole-2-thio) propyl) 1H-isoindole-1, 3(2H)-dione (**7r**)*

Yield: 80%. ^{1}H NMR (300 MHz, DMSO): δ 13.236 (s, 1H), 8.013 (d, J = 8.7 Hz, 1H), 7.881 (m, 4H), 7.475 (d, J = 8.7 Hz, 1H), 3.722 (t, J = 6.6 Hz, 2H), 3.354 (t, J = 6.6 Hz, 2H), 2.029 (m, 2H). ^{13}C NMR (75 MHz, DMSO): δ 168.11, 134.36, 131.77, 123.02, 117.37, 54.90, 36.33, 30.69, 28.56. HRMS (ESI) *m*/*z* found 383.0814, calcd for $C_{18}H_{15}N_4O_4S$ $(M+H)^+$ 383.0814.

*4.3. General procedure for compounds **8a**–**r***

To a stirred suspension of compounds **7a**–**r** in dry ethanol, 2 equiv. of 85% aqueous hydrazine was added. After refluxing for 4 h, the solvent was concentrated. The mixture was diluted with ethyl acetate, washed with 2 mol/L NaOH aqueous, water and brine, dried over $MgSO_4$. After evaporation of the solvent, the crude products were used for the next step without isolation.

*4.4. General procedure for compounds **9a**–**o** and **10a**–**c***

Macrolide intermediate **6** and 5 equiv. of amino side chains **8a**–**r** were dissolved in 10% aqueous acetonitrile, the reaction was stirred at 30 °C until **6** disappeared by TLC. The mixture was diluted with CH_2Cl_2, washed with 5% KH_2PO_4 aqueous, water and brine, and dried over $MgSO_4$. After evaporation of the solvent, the products were purified by column chromatography eluting with 3:1 petroleum ether/acetone to afford products with 2′-OAc as white foam. After refluxing with CH_3OH, purified by column chromatography eluting with 1:1 petroleum ether/acetone, the desired products were obtained as white foam.

*4.4.1. 11,12-Didehydro-3-O-descladinosyl-6-O-methyl-3-oxo-12,11-(oxycarbonyl (3-(5-methyl-1,3,4-thiadiazole-2-mercapto) propyl) imino) erythromycin A (**9a**)*

Yield: 30%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 4.917 (d, J = 9.3 Hz, 1H), 4.274 (d, J = 7.2 Hz, 1H), 4.228 (d, J = 8.4 Hz, 1H), 3.831 (q, J = 6.6 Hz, 1H), 3.754 (m, 2H), 3.566 (s, 1H), 3.534 (d, J = 3.9 Hz, 1H), 3.483 (s, 1H), 3.331 (m, 2H), 3.115 (m, 3H), 2.708 (s, 3H), 2.623 (s, 3H), 2.450 (m, 1H), 2.555 (s, 6H), 2.165 (s, 1H), 1.457 (s, 3H), 1.148 (d, J = 6.6 Hz, 3H), 1.000 (d, J = 6.6 Hz, 3H), 0.851 (t, J = 7.2 Hz, 3H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 215.97, 203.70, 169.52, 165.46, 164.69, 157.15, 103.96, 82.22, 79.58, 78.14, 70.35, 69.62, 65.88, 60.60, 51.18, 49.89, 47.54, 44.87, 42.66, 40.21, 39.54, 39.01, 31.64, 28.15, 26.73, 22.26, 21.16, 19.74, 18.33, 15.71, 15.57, 14.61, 14.37, 13.85, 10.39. HRMS (ESI): m/z found 785.3879, calcd for $C_{37}H_{61}N_4O_{10}S_2$ $(M+H)^+$ 785.3824.

*4.4.2. 11,12-Didehydro-3-O-descladinosyl-6-O-methyl-3-oxo-12,11-(oxycarbonyl (3-(4, 6-dimethylpyrimidine-2-mercapto) propyl) imino) erythromycin A (**9b**)*

Yield: 34%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 6.635 (s, 1H), 4.930 (d, J = 10.5 Hz, 1H), 4.271(d, J = 7.2 Hz, 1H), 4.212 (d, J = 8.4 Hz, 1H), 3.817 (q, J = 6.6 Hz, 1H), 3.713 (m, 2H), 3.582 (s, 1H), 3.519 (m, 2H), 3.169 (m, 3H), 2.603 (s, 3H), 2.370 (s, 6H), 2.265 (s, 6H), 1.483 (s, 3H), 1.139 (d, J = 6.9 Hz, 3H), 0.999 (d, J = 6.6 Hz, 3H), 0.832 (t, J = 7.5 Hz, 3H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 215.78, 203.84, 171.07, 169.36, 166.81, 157.06, 115.38, 103.83, 82.10, 79.47, 78.03, 70.28, 69.52, 65.88, 60.64, 51.15, 49.82, 47.51, 44.85, 43.18, 40.21, 39.45, 39.00, 28.17, 28.02, 26.99, 23.79, 22.22, 21.13, 19.69, 18.31, 15.74, 14.59, 14.33, 13.85, 10.37. HRMS (ESI): m/z found 793.4402, calcd for $C_{40}H_{65}N_4O_{10}S$ $(M+H)^+$ 793.4416.

*4.4.3. 11,12-Didehydro-3-O-descladinosyl-6-O-methyl-3-oxo-12,11-(oxycarbonyl (3-(1, 3,4-thiadiazole-2-mercapto) propyl) imino) erythromycin A (**9c**)*

Yield: 30%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 8.969 (s, 1H), 4.915 (dd, J = 10.8, 1.8 Hz, 1H), 4.262 (d, J = 6.9 Hz, 1H), 4.216 (d, J = 8.7 Hz, 1H), 3.788 (m, 3H), 3.561 (s, 1H), 3.521 (m, 1H), 3.406 (t, J = 7.2 Hz, 2H), 2.618 (s, 3H), 2.434 (m, 1H), 2.247 (s, 6H), 2.155 (d, J = 2.4 Hz, 1H), 2.103 (t, J = 7.5 Hz, 1H), 1.449 (s, 3H), 1.138 (d, J = 6.9 Hz, 3H), 0.990 (d, J = 6.6 Hz, 3H), 0.839 (t, J = 7.2 Hz, 3H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 216.05, 203.67, 169.54, 157.15, 151.07, 103.88, 82.22, 79.56, 78.10, 77.17, 70.27, 69.55, 65.80, 60.50, 51.13, 49.85, 47.53, 44.82, 42.57, 40.17, 39.47, 38.95, 31.67, 28.10, 26.55, 22.17, 21.12, 19.70, 18.28, 15.73, 14.56, 14.31, 13.81, 10.33. HRMS (ESI): m/z found 771.3668, calcd for $C_{36}H_{59}N_4O_{10}S_2$ $(M+H)^+$ 771.3673.

*4.4.4. 11,12-Didehydro-3-O-descladinosyl-6-O-methyl-3-oxo-12,11-(oxycarbonyl (3-(1-methyltetrazole-5-mercapto) propyl) imino) erythromycin A (**9d**)*

Yield: 40%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 4.905(d, J = 9.0 Hz, 1H), 4.280 (d, J = 7.5 Hz, 1H), 4.218 (d, J = 8.4 Hz, 1H), 3.909 (s, 3H), 3.824 (m, 1H), 3.755 (m, 2H), 3.543 (s, 1H), 3.394 (m, 2H), 3.215 (m, 1H), 3.080 (m, 2H), 2.562 (s, 3H), 2.333 (s, 6H), 2.073 (q, 1H), 1.453 (s, 3H), 1.143 (d, J = 7.2 Hz, 3H), 0.972 (d, J = 6.6 Hz, 3H), 0.846 (t, J = 7.2 Hz, 3H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 216.08, 203.67, 169.64, 157.21, 103.70, 82.28, 79.55, 78.11, 77.23, 70.16, 69.34, 66.03, 60.45, 51.14, 49.83, 47.50, 44.84, 42.17, 40.23, 39.45, 38.95, 33.36, 30.94, 28.54, 26.83, 22.19, 21.09, 19.70, 18.31, 15.73, 14.59, 14.34, 13.81, 10.33. HRMS (ESI): m/z found 769.4150, calcd for $C_{36}H_{61}N_6O_{10}S$ $(M+H)^+$ 769.4170.

*4.4.5. 11,12-Didehydro-3-O-descladinosyl-6-O-methyl-3-oxo-12,11-(oxycarbonyl (3-(pyridine-2-mercapto) propyl) imino) erythromycin A (**9e**)*

Yield: 50%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 8.378 (d, J = 4.5 Hz, 1H), 7.423 (td, J = 8.1, 1.8 Hz, 1H), 7.135 (d, J = 8.1 Hz, 1H), 6.920 (td, J = 4.5, 1.8 Hz, 1H), 4.930 (dd, J = 10.2, 1.8 Hz, 1H), 4.304 (d, J = 6.9 Hz, 1H), 4.211 (d, J = 8.4 Hz, 1H), 3.813 (q, J = 6.9 Hz, 1H), 3.727 (t, J = 8.4 Hz, 2H), 3.581 (s, 1H), 3.548 (m, 1H), 3.281 (q, 1H), 3.175 (t, 2H), 3.083 (m, 2H), 2.600 (s, 3H), 2.429 (s, 6H), 1.450 (s, 3H), 1.329 (d, J = 6.6 Hz, 3H), 1.145 (d, J = 6.9 Hz, 3H), 0.995 (d, J = 6.6 Hz, 3H), 0.826 (t, J = 7.2 Hz, 3H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 215.84, 203.87, 169.36, 157.06, 149.35, 135.66, 122.06, 119.08, 103.39, 82.08, 79.38, 77.99, 77.23, 70.01, 68.97, 66.10, 60.61, 55.60, 51.14, 49.76, 47.39, 44.81, 42.96, 40.22, 39.38, 39.01,29.07, 27.30, 26.98, 22.23, 21.01, 19.66, 18.27, 15.67, 14.63, 14.34, 13.81, 10.33. HRMS (ESI) m/z found 764.4120, calcd for $C_{39}H_{62}N_3O_{10}S$ $(M+H)^+$ 764.4150.

*4.4.6. 11,12-Didehydro-3-O-descladinosyl-6-O-methyl-3-oxo-12,11-(oxycarbonyl (3-(thiazole-2-mercapto) propyl) imino) erythromycin A (**9f**)*

Yield: 48%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.616 (d, J = 3.3 Hz, 1H), 7.163 (d, J = 3.3 Hz, 1H), 4.911 (dd, J = 10.5, 2.1 Hz, 1H), 4.269 (d, J = 7.2 Hz, 1H), 4.211 (d, J = 8.7 Hz, 1H), 3.817 (q, J = 6.6 Hz, 1H), 3.732 (m, 2H), 3.561 (s, 1H), 3.535 (m, 2H), 2.602 (s, 3H), 2.283 (s, 6H), 1.513 (s, 3H), 1.137 (d, J = 7.2 Hz, 3H), 0.988 (d, J = 6.6 Hz, 3H), 0.831 (t, J = 7.5 Hz, 3H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 215.98, 203.73, 169.42, 164.83, 157.09, 142.62, 118.52, 103.77, 82.16, 79.52, 78.05, 77.17, 70.21, 69.43, 65.89, 60.56, 51.14, 49.80, 47.51, 44.82, 42.73, 40.20, 39.47, 38.96, 31.74, 29.21, 28.31, 26.81, 22.20, 21.11, 19.69, 18.28, 15.75, 14.60, 14.33, 13.82, 10.34. HRMS (ESI) m/z found 770.3732, calcd for $C_{37}H_{60}N_3O_{10}S_2$ $(M+H)^+$ 770.3720.

*4.4.7. 11, 12-Didehydro-3-O-descladinosyl-6-O-methyl-3-oxo-12,11-(oxycarbonyl (3-(2-methylfurane-3-mercapto) propyl) imino) erythromycin A (**9g**)*

Yield: 52%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.230 (d, J = 1.5 Hz, 1H), 6.362 (d, J = 1.5 Hz, 1H), 4.881 (d, J = 9.0 Hz, 1H), 4.275 (d, J = 7.2 Hz, 1H), 4.224 (d, J = 8.4 Hz, 1H), 3.826 (q, J = 6.6 Hz, 1H), 3.681 (m, 2H), 3.540 (s, 1H), 3.506 (m, 1H), 2.584 (s, 3H), 2.320 (s, 3H), 2.272 (s, 6H), 1.444 (s, 3H), 1.142 (d, J = 6.9 Hz, 3H), 0.983 (d, J = 6.6 Hz, 3H), 0.829 (t, J = 7.2 Hz, 3H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 216.05, 203.78, 169.39, 157.06, 155.00, 140.37, 115.07, 103.82, 82.07, 79.49, 78.04, 77.23, 70.25, 69.50, 65.88, 60.44, 51.14, 49.76, 47.46, 44.88, 42.55, 40.19, 39.47, 38.97, 33.40, 28.22, 27.32, 22.22, 21.14, 19.69, 18.34, 15.68, 14.63, 14.34, 13.85, 11.73, 10.38. HRMS (ESI) m/z found 767.4152, calcd for $C_{39}H_{63}N_2O_{11}S$ $(M+H)^+$ 767.4147.

*4.4.8. 11,12-Didehydro-3-O-descladinosyl-6-O-methyl-3-oxo-12,11-(oxycarbonyl (3-(5-n-propylpyrimidine-2-mercapto) propyl) imino) erythromycin A (**9h**)*

Yield: 35%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 8.315 (s, 2H), 4.946 (d, J = 9.0 Hz, 1H), 4.276 (d, J = 7.5 Hz, 1H), 4.219 (d, J = 8.4 Hz, 1H), 4.107 (q, J = 7.5 Hz, 1H), 3.819 (q, J = 6.9 Hz, 1H), 3.720 (t, J = 6.9 Hz, 2H), 3.593 (s, 1H), 3.522 (m, 1H), 2.636 (s, 3H), 2.477 (t, J = 7.5 Hz, 3H), 2.266 (s, 6H), 1.450 (s, 3H), 1.142 (d, J = 6.9 Hz, 3H), 1.001 (d, J = 7.2 Hz, 3H), 0.944 (t, J = 7.2 Hz, 3H), 0.829 (t, J = 7.2 Hz, 3H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 215.84, 203.83, 169.33, 157.10, 129.75, 103.85, 82.11, 79.49, 78.04, 70.28, 69.52, 65.86, 60.67, 53.75, 51.16, 49.82, 47.54, 44.86, 43.10, 40.20, 39.47, 39.01, 31.81, 29.65, 29.23, 28.17, 26.94, 23.94, 22.25, 21.14, 19.70, 18.30, 15.75, 14.60, 14.31, 13.84, 13.49, 10.34. HRMS (ESI) m/z found 807.4570, calcd for $C_{41}H_{67}N_4O_{10}S$ $(M+H)^+$ 807.4578.

*4.4.9. 11,12-Didehydro-3-O-descladinosyl-6-O-methyl-3-oxo-12,11-(oxycarbonyl (3-(4-methylpyrimidine-2-mercapto) propyl)imino) erythromycin A (**9i**)*

Yield: 36%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 8.300 (d, J = 5.1 Hz, 1H), 6.760 (d, J = 5.1 Hz, 1H), 4.917 (dd, J = 9.0, 1.8 Hz, 1H), 4.266 (d, J = 7.2 Hz, 1H), 4.194 (d, J = 9.0 Hz, 1H), 3.803 (q, J = 6.9 Hz, 1H), 3.700 (m, 2H), 3.565 (s, 1H), 3.518 (m, 1H), 3.349 (q, J = 6.9 Hz, 1H),

2.598 (s, 3H), 2.406 (s, 3H), 2.312 (s, 6H), 1.970 (s, 3H), 1.433 (s, 3H), 1.127 (d, $J = 6.9$ Hz, 3H), 0.981 (d, $J = 6.9$ Hz, 3H), 0.812 (t, $J = 7.2$ Hz, 3H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 215.88, 203.84, 169.33, 167.31, 156.63, 115.91, 103.64, 82.11, 79.40, 77.98, 70.13, 69.28, 65.89, 60.59, 51.10, 49.74, 47.48, 44.81, 43.04, 40.18, 39.38, 29.27, 28.49, 28.02, 27.75, 26.84, 23.99, 22.17, 21.06, 19.66, 18.24, 15.78, 14.56, 14.30, 13.81, 10.31. HRMS (ESI): *m/z* found 779.4274, calcd for $C_{39}H_{63}N_4O_{10}S$ $(M + H)^+$ 779.4259.

4.4.10. 11,12-Didehydro-3-O-descladinosyl-6-O-methyl-3-oxo-12,11-(oxycarbonyl (3-(4,5-diphenyloxazole-2-mercapto) propyl) imino) erythromycin A (**9j**)

Yield: 17%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.655 (d, $J = 6.6$ Hz, 2H), 7.556 (d, $J = 6.6$ Hz, 2H), 7.359 (m, 6H), 4.949 (dd, $J = 8.7$, 1.2 Hz, 1H), 4.267 (d, $J = 7.2$ Hz, 1H), 4.209 (d, $J = 8.4$ Hz, 1H), 3.810 (m, 2H), 3.594 (s, 1H), 3.525 (m, 1H), 3.300 (t, $J = 7.2$ Hz, 2H), 2.624 (s, 3H), 2.257 (s, 6H), 1.460 (s, 3H), 1.336 (d, $J = 6.9$ Hz, 3H), 1.146 (d, $J = 6.9$ Hz, 3H), 1.013 (d, $J = 7.2$ Hz, 3H), 0.823 (t, $J = 7.2$ Hz, 3H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 215.92, 203.71, 169.51, 159.08, 157.17, 146.93, 136.44, 132.28, 129.55, 128.81, 128.54, 128.44, 128.24, 128.02, 126.34, 125.75, 103.88, 82.23, 79.54, 78.08, 77.19, 70.31, 69.56, 65.85, 60.66, 53.78, 51.17, 49.89, 47.49, 45.81, 44.86, 42.83, 40.21, 39.51, 39.00, 31.71, 29.90, 29.67, 29.25, 28.15, 27.16, 22.23, 21.15, 19.70, 18.34, 15.67, 14.60, 14.34, 13.87, 10.34. HRMS (ESI) *m/z* found 906.4543, calcd for $C_{49}H_{68}N_3O_{11}S$ $(M + H)^+$ 906.4569.

4.4.11. 11, 12-Didehydro-3-O-descladinosyl-6-O-methyl-3-oxo-12,11-(oxycarbonyl(3-(4-(3-pyridine)-1H-imidazole-2-mercapto) propyl) imino)erythromycin A (**9k**)

Yield: 42%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 11.201 (br s, 1H), 8.942 (s, 1H), 8.441 (d, $J = 4.5$ Hz, 1H), 8.111 (s, 1H), 7.439 (s, 1H), 7.284 (dd, $J = 7.5$, 3.3 Hz, 1H), 4.901 (d, $J = 9.6$ Hz, 1H), 4.264 (d, $J = 7.2$ Hz, 1H), 4.203 (d, $J = 9.0$ Hz, 1H), 3.848 (m, 3H), 3.583 (s, 1H), 3.519 (m, 1H), 2.856 (s, 3H), 2.278 (s, 6H), 1.468 (s, 3H), 1.111 (d, $J = 6.9$ Hz, 3H), 1.029 (d, $J = 6.6$ Hz, 3H), 0.833 (t, $J = 7.2$ Hz, 3H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 216.72, 203.54, 169.91, 157.78, 147.64, 146.25, 123.55, 103.85, 82.85, 79.46, 78.16, 77.18, 70.24, 69.49, 65.84, 61.20, 51.14, 49.69, 47.63, 45.88, 44.96, 42.46, 40.15, 39.53, 38.90, 33.35, 29.64, 29.23, 28.28, 27.03, 22.19, 21.12, 19.67, 18.21, 15.90, 14.30, 13.93, 10.38. HRMS (ESI) *m/z* found 830.4305, calcd for $C_{42}H_{64}N_5O_{10}S$ $(M + H)^+$ 830.4368.

4.4.12. 11,12-Didehydro-3-O-descladinosyl-6-O-methyl-3-oxo-12,11-(oxycarbonyl(3-(5-methoxybenzothiazole-2-mercapto) propyl) imino) erythromycin A (**9l**)

Yield: 41%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.557 (d, $J = 9.0$ Hz, 1H), 7.376 (d, $J = 2.1$ Hz, 1H), 6.898 (dd, $J = 9.0$, 2.1 Hz, 1H), 4.928 (d, $J = 9.3$ Hz, 1H), 4.262 (d, $J = 7.2$ Hz, 1H), 4.202 (d, $J = 9.0$ Hz, 1H), 3.857 (s, 3H), 3.807 (m, 2H), 3.581 (s, 1H), 3.521 (m, 1H), 2.601 (s, 3H), 2.257 (s, 6H), 2.161 (s, 3H), 1.456 (s, 3H), 1.330 (d, $J = 6.9$ Hz, 3H), 1.145 (d, $J = 6.9$ Hz, 3H), 1.003 (d, $J = 6.9$ Hz, 3H), 0.836 (t, $J = 7.2$ Hz, 3H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 215.96, 203.69, 169.48, 158.78, 157.13, 154.52, 126.87, 120.97, 113.74, 104.52, 82.17, 79.55, 78.07, 77.20, 70.27, 69.52, 65.83, 60.55, 55.54, 53.75, 51.14, 49.83, 47.54, 44.82, 42.78, 40.18, 39.48, 38.98, 37.50, 30.86, 29.21, 28.14, 26.87, 22.22, 21.11, 19.66, 18.30, 15.75, 14.60, 14.30, 13.81, 10.34. HRMS (ESI) *m/z* found 850.3962, calcd for $C_{42}H_{64}N_3O_{11}S_2$ $(M + H)^+$ 850.3982.

4.4.13. 11,12-Didehydro-3-O-descladinosyl-6-O-methyl-3-oxo-12,11-(oxycarbonyl (3-(1-phenyl-tetrazole-5-mercapto) propyl) imino) erythromycin A (**9m**)

Yield: 58%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.546 (m, 5H), 4.923 (d, $J = 9.0$ Hz, 1H), 4.276 (d, $J = 7.2$ Hz, 1H), 4.209 (d, $J = 8.7$ Hz, 1H), 3.822 (q, $J = 6.9$ Hz, 1H), 3.720 (m, 2H), 3.548 (s, 1H), 3.206 (dd, $J = 9.9$, 7.5 Hz, 1H), 3.074 (m, 2H), 2.593 (s, 3H), 2.316 (s, 6H), 1.448 (s, 3H), 1.077 (d, $J = 6.3$ Hz, 3H), 0.981 (d, $J = 6.9$ Hz, 3H), 0.839 (t, $J = 7.2$ Hz, 3H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 216.02, 203.69, 169.60, 157.20, 154.28, 133.69, 129.95, 129.69, 123.94, 103.73, 82.27, 79.53, 78.08, 77.18, 70.18, 69.37, 65.95, 60.52, 51.13, 49.80, 47.50, 44.81, 42.32, 40.22, 39.44, 38.93, 30.74, 28.43, 26.62, 22.16, 21.09, 19.69, 18.27, 15.75, 14.57, 14.31, 13.79, 10.30. HRMS (ESI) *m/z* found 831.4322, calcd for $C_{41}H_{63}N_6O_{10}S$ $(M + H)^+$ 831.4326.

4.4.14. 11,12-Didehydro-3-O-descladinosyl-6-O-methyl-3-oxo-12,11-(oxycarbonyl (3-(benzoxazole-2-mercapto) propyl) imino) erythromycin A (**9n**)

Yield: 15%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 10.550 (br s, 1H), 7.169 (dd, $J = 6.0$, 3.0 Hz, 2H), 7.094 (d, $J = 3.0$ Hz, 1H), 4.933 (d, $J = 9.0$ Hz, 1H), 4.262 (d, $J = 7.2$ Hz, 1H), 4.197 (d, $J = 9.0$ Hz, 1H), 3.832 (m, 3H), 3.580 (s, 1H), 3.518 (m, H), 2.566 (s, 3H), 2.444 (m, 1H), 2.255 (s, 6H), 2.052 (q, $J = 6.9$ Hz, 2H), 1.461 (s, 3H), 1.360 (d, $J = 6.9$ Hz, 3H), 1.136 (d, $J = 6.9$ Hz, 3H), 1.007 (d, $J = 6.9$ Hz, 3H), 0.809 (t, $J = 7.5$ Hz, 3H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 216.37, 203.54, 169.86, 157.47, 149.06, 129.51, 125.71, 122.07, 118.53, 103.85, 82.46, 79.46, 78.10, 70.27, 69.55, 65.77, 60.97, 53.74, 51.16, 49.76, 47.59, 44.87, 42.76, 40.15, 39.47, 38.93, 30.86, 29.20, 28.13, 27.24, 22.17, 21.09, 19.64, 18.22, 15.87, 14.53, 14.30, 13.84, 10.33. HRMS (ESI) *m/z* found 803.4263, calcd for $C_{41}H_{63}N_4O_{10}S$ $(M + H)^+$ 803.4259.

4.4.15. 11,12-Didehydro-3-O-descladinosyl-6-O-methyl-3-oxo-12,11-(oxycarbonyl (3-(4-phenylthiazole-2-mercapto) propyl) imino) erythromycin A (**9o**)

Yield: 74%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.880 (d, $J = 7.2$ Hz, 2H), 7.392 (t, $J = 7.8$ Hz, 2H), 7.314 (s, 1H), 7.304 (d, $J = 6.3$ Hz, 1H), 4.930 (d, $J = 8.7$ Hz, 1H), 4.268 (d, $J = 7.2$ Hz, 1H), 4.207 (d, $J = 8.4$ Hz, 1H), 3.803 (m, 3H), 3.583 (s, 1H), 3.530 (m, 2H), 3.323 (t, $J = 7.2$ Hz, 2H), 3.180 (dd, $J = 7.2$, 9.9 Hz, 1H), 3.079 (m, 2H), 2.611 (s, 3H), 2.274 (s, 6H), 1.457 (s, 3H), 1.146 (d, $J = 6.9$ Hz, 3H), 1.006 (d, $J = 7.2$ Hz, 3H), 0.829 (t, $J = 7.2$ Hz, 3H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 215.94, 203.72, 169.47, 164.66, 157.13, 134.15, 128.58, 127.95, 126.33, 112.11, 103.80, 82.16, 79.53, 78.05, 77.18, 70.24, 69.48, 65.87, 60.56, 51.14, 49.85, 47.51, 44.82, 42.83, 40.20, 39.48, 38.98, 31.77, 29.23, 28.22, 26.94, 22.20, 21.12, 19.66, 18.31, 15.72, 14.60, 14.31, 13.84, 10.34. HRMS (ESI) *m/z* found 846.3976, calcd for $C_{43}H_{64}N_3O_{10}S_2$ $(M + H)^+$ 846.4027.

4.4.16. 11,12-Didehydro-3-O-descladinosyl-6-O-methyl-3-oxo-10,11-dehydrate-12-((3-(benzoxazole-2-mercapto)propyl) oxycarbonylimino) erythromycin A (**10a**)

Yield: 25%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.333 (d, $J = 7.5$ Hz, 1H), 7.222 (d, $J = 8.1$ Hz, 1H), 7.138 (t, $J = 7.5$ Hz, 1H), 7.006 (t, $J = 7.5$ Hz, 1H), 6.763 (s, 1H), 5.648 (dd, $J = 9.3$, 2.7 Hz, 1H), 5.501 (s, 1H), 4.234 (d, $J = 7.5$ Hz, 1H), 4.156 (d, $J = 9.3$ Hz, 1H), 3.730 (q, $J = 6.6$ Hz, 1H), 3.497 (s, 3H), 2.895 (t, $J = 6.6$ Hz, 2H), 2.844 (s, 3H), 2.269 (s, 6H), 1.967 (t, $J = 6.3$ Hz, 2H), 1.868 (s, 3H), 1.668 (s, 3H), 1.170 (d, $J = 6.6$ Hz, 3H), 0.901 (t, $J = 7.2$ Hz, 3H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 205.18, 203.38, 168.96, 161.82, 148.46, 142.89, 140.13, 137.41, 123.83, 120.79, 116.26, 108.69, 104.08, 83.41, 78.16, 70.22, 69.40, 65.72, 50.98, 50.50, 47.16, 40.98, 40.20, 38.06, 29.71, 28.36, 27.93, 22.31, 21.04, 20.94, 19.88, 18.45, 14.57, 12.89, 10.41. HRMS (ESI) *m/z* found 804.3999, calcd for $C_{41}H_{62}N_3O_{11}S$ $(M + H)^+$ 804.4099.

4.4.17. 11, 12-Didehydro-3-O-descladinosyl-6-O-methyl-3-oxo-10,11-dehydrate-12-((3-(pyrazolo[3,4d]pyrimidine-4-mercapto) propyl) oxycarbonylimino) erythromycin A (**10b**)

Yield: 31%. ^{1}H NMR (300 MHz, $CDCl_3$): δ 8.381 (s, 1H), 8.075 (s, 1H), 6.771 (s, 1H), 5.650 (dd, $J = 9.9$, 3.3 Hz, 1H), 5.590 (s, 1H), 4.224 (d, $J = 7.2$ Hz, 1H), 4.151 (d, $J = 9.3$ Hz, 1H), 3.733 (m, 1H), 3.671 (m, 1H), 3.477 (m, 2H), 3.047 (t, $J = 8.1$ Hz, 1H), 2.923 (t, $J = 6.6$ Hz, 2H), 2.266 (s, 6H), 1.869 (s, 3H), 1.436 (s, 3H), 0.898 (t, $J = 7.5$ Hz, 3H). ^{13}C

NMR (75 MHz, $CDCl_3$): δ 205.90, 169.24, 155.87, 140.22, 137.69, 135.14, 132.38, 124.96, 103.10, 83.38, 78.34, 69.63, 68.47, 65.92, 63.28, 50.90, 50.32, 47.16, 34.41, 32.14, 31.85, 30.80, 29.29, 28.41, 26.33, 23.35, 22.62, 22.17, 21.09, 20.72, 19.75, 18.73, 14.50, 13.00, 10.41. HRMS (ESI) *m*/*z* found 805.4135, calcd for $C_{39}H_{61}N_6O_{10}S$ $(M+H)^+$ 805.4164.

*4.4.18. 11,12-Didehydro-3-O-descladinosyl-6-O-methyl-3-oxo-10,11-dehydrate-12-((3-(5-nitrobenzimidazole-2-mercapto)propyl) oxycarbonylimino) erythromycin A (**10c**)*

Yield: 20%. ^1H NMR (300 MHz, $CDCl_3$): δ 8.121 (s, 1H), 8.016 (d, $J = 8.7$ Hz, 1H), 7.276 (d, $J = 8.7$ Hz, 1H), 6.727 (s, 1H), 5.861 (br s, 1H), 5.665 (dd, $J = 9.9$, 2.1 Hz, 1H), 4.280 (d, $J = 7.2$ Hz, 1H), 4.150 (d, $J = 9.3$ Hz, 1H), 3.891 (m, 1H), 3.184 (t, $J = 7.5$ Hz, 2H), 3.041 (t, $J = 6.9$ Hz, 1H), 2.629 (s, 3H), 2.265 (s, 6H), 1.152 (d, $J = 6.9$ Hz, 3H), 0.902 (t, $J = 6.0$ Hz, 3H). HRMS (ESI): *m*/*z* found 848.4071, calcd for $C_{41}H_{62}N_5O_{12}S$ $(M+H)^+$ 848.4110.

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