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ORIGINAL ARTICLE

Synthesis and structure–activity relationships of novel lincomycin derivatives. Part 4: synthesis of novel lincomycin analogs modified at the 6- and 7-positions and their potent antibacterial activities

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To modify lincomycin (LCM) at the C-6 and the C-7 positions, we firstly prepared various substituted proline intermediates (7, 11–15 and 17). These proline intermediates were coupled with methyl 1-thio-α-lincosamide and tetrakis-*O*-trimethylsilylation followed by selective deprotection of the TMS group at the 7-position gave a wide variety of key intermediates (23–27, 47 and 50). Then, we synthesized a variety of novel LCM analogs modified at the 7-position in application of the Mitsunobu reaction, an S_N2 reaction, and a Pd-catalyzed cross-coupling reaction. Compounds 34 and 35 (1′-*N*H derivatives) exhibited enhanced antibacterial activities against resistant pathogens with *erm* gene compared with the corresponding 1′-*N*-methyl derivatives (3 and 37). On the basis of reported SAR, we modified the 4′-position of LCM derivatives possessing a 5-(2-nitrophenyl)-1,3,4-thiadiazol-2-yl group at the C-7 position. Compound 56 showed significantly potent antibacterial activities against *S. pneumoniae* and *S. pyogenes* with *erm* gene, and its activities against *S. pneumoniae* with *erm* gene were improved compared with those of 34 and 57. Although we synthesized novel analogs by transformation of a C-7 substituent focusing on the 1′-demethyl framework to prepare very potent analogs 73 and 75, it was impossible to generate novel derivatives exhibiting stronger antibacterial activities against *S. pneumoniae* with *erm* gene compared with 56.
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INTRODUCTION

Macrolide antibiotics have been used in the clinical site for bacterial respiratory infections so far. Recently, macrolide-resistant bacteria with *erm* gene have markedly increased.[1–3] Clarithromycin (CAM)[4] and azithromycin[5,6] are not effective enough against resistant pathogens such as *Streptococcus pneumoniae* and *Streptococcus pyogenes* with *erm* gene (Figure 1 and Table 1). Telithromycin (TEL)[7] is effective enough against *S. pneumoniae* with *erm* gene, but has the potential to cause a serious liver damage[8,9] and loss of consciousness.[10,11] Thus, TEL has scarcely been used in Japan. Novel azalides reported by Miura *et al.*[12,13] are also effective against resistant pathogens, but these analogs are still under research process and have not been developed yet. Currently available oral antibiotics are not effective enough against resistant bacteria with *erm* and *mef* genes causing respiratory infections and have some problems in safety or taste in clinical site.

Lincomycin (LCM)[14–17] and clindamycin (CLDM)[18] are effective against pathogens with *mef* gene in clinical isolates, but they are not effective against resistant pathogens with *erm* gene (Figure 1 and Table 1). As an overview, CLDM exhibits the following positive characters: (1) availability in *per os* and intravenous administrations (switch therapy is possible), (2) good distributions to tissue and cells, (3) suppression[19] of toxin production by Streptococcal strains and (4) expected reasonable production cost of LCM/CLDM derivatives. Thus, LCM derivatives might be more clinically valuable than macrolide antibiotics, if they are effective against pathogens with *erm* gene.

Chemical modifications at the C-7 position of LCM were achieved by several research groups.[17,18,20–32] None of them, however, showed effective antibacterial activities against resistant bacteria with *erm* gene. On the other hand, we reported 7(*S*)-thiolincomycin analogs exhibiting antibacterial activities against resistant pathogens with *erm* gene as the first example[33–36] as far as we know. We recently reported 7(*S*)-thiolincomycin analogs, such as compounds **1–3** (Table 1), as the first-generation derivatives in our research.[37] Those compounds possessed weak antibacterial activities against resistant *S. pneumoniae* with *erm* and *mef* genes, but compound **3** exhibited clearly improved activities compared with clarithromycin, azithromycin, LCM and CLDM as shown in Table 1. Recently, we also reported 7-*S*-substituted novel LCM derivatives,[38–41] and some of them showed stronger activities than compound **3** did.

Chemical modification of a proline moiety at the C-6 position of LCM and CLDM was performed by several research groups.[17,24,42–48]

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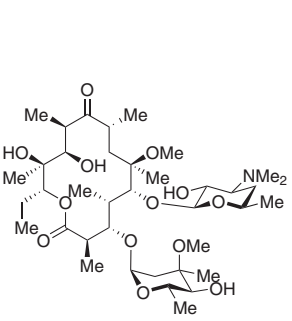
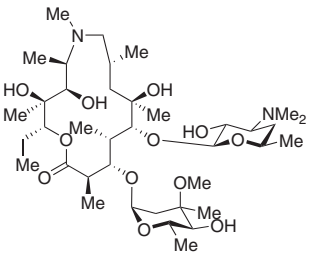


Clarithromycin (CAM)



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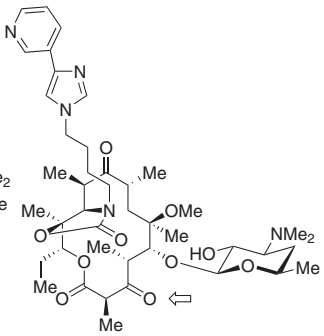
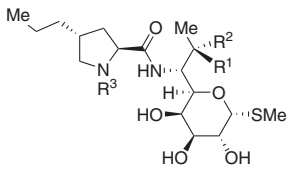
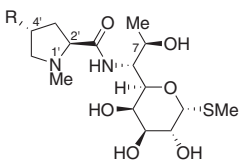


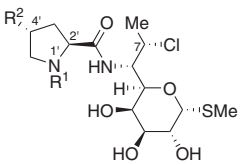
Telithromycin (TEL)



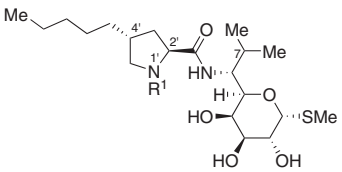
Lincomycin (LCM): R^1 = OH, R^2 = H, R^3 = Me
1'-Demethyllincomycin: R^1 = OH, R^2 = H, R^3 = H
Clindamycin (CLDM): R^1 = H, R^2 = Cl, R^3 = Me
1'-Demethylclindamycin: R^1 = H, R^2 = Cl, R^3 = H



4'-Substituted lincomycins: R = H, Me to ^{n}octyl
4'-Depropyl-4'-ethoxylincomycin: R = ethoxy
4'-Depropyl-4'-ethoxycarbonylmethyllincomycin: R = ethoxycarbonylmethyl
4'-Depropyl-4'-*p*-toluenesulfonyloxylincomycin: R = *p*-toluensulfonyloxy



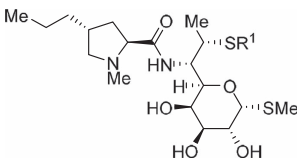
4'-Depropyl-4'-^{n}pentylclindamycin: R^1 = Me, R^2 = ^{n}pentyl
1'-Demethyl-4'-depropyl-4'-^{n}pentylclindamycin: R^1 = H, R^2 = ^{n}pentyl

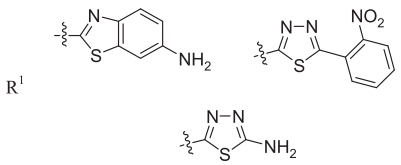


VIC105403: R^1 = H
VIC105404: R^1 = 2-hydroxyethyl
VIC105405: R^1 = 2-(*R*)-hydroxypropyl
VIC105446: R^1 = 2-(*S*)-hydroxypropyl

Figure 1 Chemical structures of clarithromycin, azithromycin, telithromycin, lincomycin, clindamycin and a variety of reported analogs of lincomycin or clindamycin.

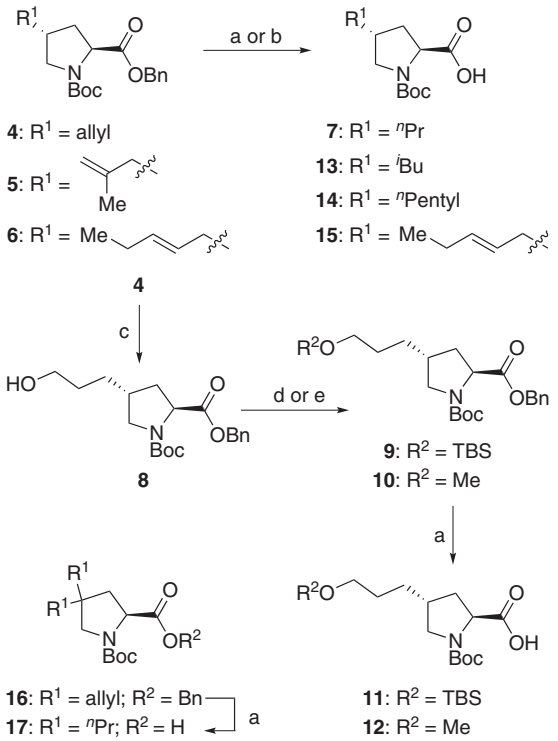
Table 1 Anti-bacterial activities (MIC, μg ml^{-1}) of CAM, AZM, LCM, CLDM, and previously reported LCM derivatives (1–3)





Test organism[a]	*Characteristics*[b]	*CAM*	*AZM*	*LCM*	*CLDM*	***1***	***2***	***3***
Streptococcus pneumoniae DP1 type I	Susceptible	0.03	0.06	1	0.06	0.06	0.03	0.015
S. pneumoniae-2	Susceptible	0.03	0.03	1	0.12	0.06	0.06	0.015
S. pneumoniae-3	Susceptible	0.015	0.03	0.25	0.06	0.03	0.06	0.03
S. pneumoniae-4	*ermAM* methylase (c)	>128	>128	>128	>128	16	16	4
S. pneumoniae-5	*ermAM* methylase (c)	>128	>128	>128	>128	64	64	8
S. pneumoniae-6	*ermAM* methylase (c)+*mefE*	>128	>128	>128	>128	64	128	16
S. pneumoniae-7	*ermAM* methylase (i)	>128	>128	128	128	16	16	2
S. pneumoniae-8	*ermAM* methylase (i)	>128	>128	128	128	8	16	2
S. pneumoniae-9	*mefE* efflux	0.5	0.5	1	0.12	0.06	0.03	0.015
Streptococcus pyogenes Cook	Susceptible	0.015	0.06	0.12	0.06	0.03	0.03	0.03
S. pyogenes-2	*ermAM* methylase (c)	>128	>128	>128	128	8	8	2
S. pyogenes-3	*mefE* efflux	8	8	0.25	0.12	0.06	0.06	0.03
Haemophilus influenzae	Susceptible	2	0.25	8	16	8	8	16
H. influenzae-2	Susceptible	4	1	16	8	4	4	8
H. influenzae-3	Susceptible	8	2	16	16	32	8	16
H. influenzae-4	⊿acr	0.5	0.5	4	1	0.25	0.25	0.25

Abbreviations: AZM, azithromycin; CAM, clarithromycin; CLDM, clindamycin; LCM, lincomycin.
[a]All strains except standard organisms were clinically isolated.
[b](c), constitutive; (i), inducible.



Scheme 1 Synthesis of proline derivatives. Conditions: (**a**) H_2, Pd/C, MeOH, r.t., 0.5–4.5 h; (**b**) 1 N NaOH, MeOH, r.t., 22 h; (**c**) (1) 9-BBN, THF, 50 °C, 1 h, (2) 1 N NaOH, 35% H_2O_2, 0 °C, 2 h; (**d**) TBSCl, imidazole, DMF, r.t., 30 min; (**e**) MeI, NaH, DMF, r.t., 30 min. 9-BBN, 9-borabicyclo[3.3.1] nonane; DMF, dimethylformamide; MeI, methyl iodide; TBSCl, *tert*-butyldimethylsilyl chloride; r.t., room temperature; THF, tetrahydrofuran.

This portion has the following features: (1) Regarding configuration between the 2′- and 4′-positions of a proline ring, an analog with *trans* configuration exhibited more potent antibacterial activity compared with one with *cis* configuration. (2) 4′-Depropyl-3′-propyllincomycin has not been synthesized, but 4′-depropyl-5′-propyllincomycin has already been synthesized. Its antibacterial activity was weaker than that of LCM. (3) 1′-Demethylclindamycin (Figure 1) was two times as active *in vitro* against *Sarcina lutea* as CLDM, but 1′-demethyllincomycin was about one-twentieth as active as LCM (relative potency against *S. lutea*: 1′-demethylclindamycin > CLDM > LCM > 1′-demethyllincomycin = 8 > 4 > 1 > 0.05).[21,42–43] 1′-Demethyl-1′-*N*-ethyllincomycin possessed the same activity as LCM. (4) Regarding SAR of a chain length (H, Me to *n*-octyl) at the 4′-position of the proline moiety for LCM (Figure 1), the *in vitro* antibacterial activities were enhanced until reaching a maximum at the hexyl analog.[21,44] A similar *in vivo* activity was indicated, but maximum effect was exhibited by an *n*-pentyl group. Furthermore, alternative *in vitro* SAR were observed by changing a chain length (Figure 1) for a 4′-alkyl-substituent of CLDM and a 4′-alkyl-substituent of 1′-demethylclindamycin (relative potency: 4′-depropyl-4′-*n*-pentylclindamycin > CLDM, 1′-demethyl-4′-depropyl-4′-*n*-pentylclindamycin > 1′-demethylclindamycin).[21] However, antibacterial activities of those compounds against resistant pathogens with *erm* gene were not disclosed. (5) 4′-Depropyl-4′-ethoxylincomycin had only ~2% antibacterial activities of LCM, and 4′-depropyl-4′-ethoxycarbonylmethyllincomycin and 4′-depropyl-4′-*p*-toluenesulfonyloxylincomycin were essentially inactive in antibacterial testing.[17,21,45]

On the other hand, Vicuron, which was consolidated by Pfizer, reported the following compounds. VIC 105403 (Figure 1),[24] possessing 1′-demethyl, 4′-*n*-pentyl and 7-methyl group, had potent activities compared with CLDM. VIC 105404,[24] possessing 1′-*N*-(2-hydroxyethyl), 4′-*n*-pentyl and 7-methyl group, was already reported to have potent antibacterial activities as VIC 105403. However, those did not show antibacterial activities against resistant pathogens with *erm* gene. Furthermore, VIC 105405 and VIC 105446,[24] having a branched side chain at the 1′-position, showed less antibacterial activities than CLDM. Other VIC compounds possessing a branched side chain, a 3-(pyridin-4-yl)propyl or an *n*-butylthio group at the 4′-position, were already synthesized, but their antibacterial activities were not disclosed in their patent.[46]

According to X-ray crystallographic analysis by Yonath *et al.*,[47] we hypothesized that CLDM had enough three-dimensional space around the 1′-position and it might be able to enhance the antibacterial activities by filling the corresponding space with other functional groups except the Me group. On the basis of the above hypothesis, we designed and synthesized novel 7(*S*)-substituted LCM derivatives modified at the 1′- and/or 4′-position(s) at the proline moiety. Anti-bacterial activities of these novel compounds are also disclosed in this report.

RESULTS AND DISCUSSION

Synthesis of proline derivatives

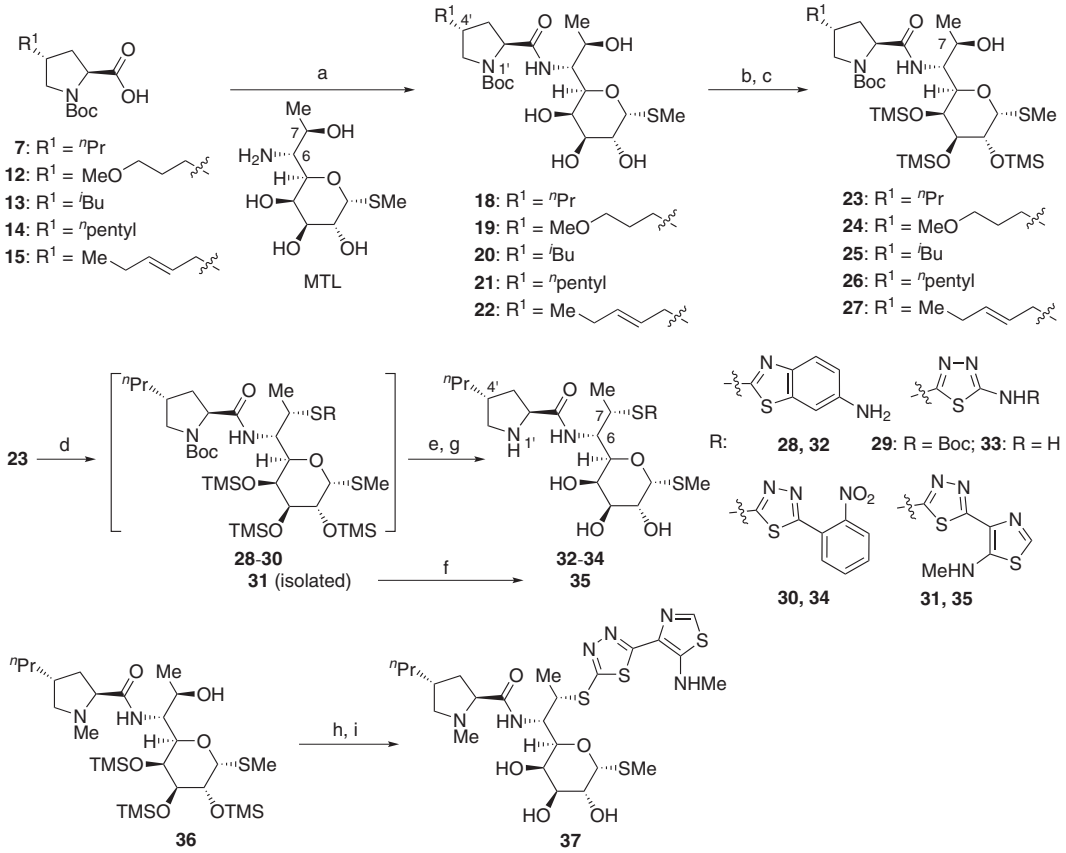
Synthesis of proline derivatives is shown in Scheme 1. Synthetic route for modification of the proline moiety at the 4′-position was already reported by Pedregal *et al.*[48,49] We firstly prepared starting materials (**4–6** and **16**), and compound **8** was synthesized by hydroboration of **4** followed by treatment with *tert*-butyldimethylsilyl chloride (TBSCl) or methyl iodide under the basic condition to give compounds **9** and **10**. The compounds **4–6**, **9**, **10** and **16** were reduced with H_2 in the presence of Pd/C to give the corresponding carboxylic acids (**7**, **11–14** and **17**), respectively. Compound **15** was prepared by hydrolysis of **6**. ^{1}H NMR spectra of compounds **7**, **8**, **10**, **13–15** and **17** were observed as two sets of signals because of a rotamer by the *tert*-butoxycarbonyl (Boc) group.

Synthesis of key intermediates 23–27 and transformation of 23 and 36

Synthesis of key intermediates **23–27** and transformations of **23** and **36** are shown in Scheme 2. Compounds **18–22** were synthesized by condensation of compounds **7** and **12–15** with methyl 1-thio-α-lincosamide (MTL), respectively. Tetra-*O*-trimethylsiliylation of all hydroxyl groups and successive regioselective deprotection of the TMS group at the 7-position gave key intermediates (**23–27**). Furthermore, novel LCM derivatives **32–35** were synthesized via two or three steps from compound **23** by the Mitsunobu reaction at the 7-position and deprotection of the TMS groups and the Boc group at the 1′-position. On the other hand, 1′-*N*-methyl analog **37** was prepared from **36** following the similar procedure as the above. ^{1}H NMR spectra of compounds **18–20** and **23** were observed as two sets of signals because of a rotamer by the Boc group and compounds **25–27** showed broad peaks in ^{1}H NMR spectra. Compounds **32–35** whose Boc groups were deprotected, however, showed a single set of signals and sharp peaks in ^{1}H NMR spectra.

Synthesis of 1′-*N*-modified novel LCM derivatives possessing a substituted 1,3,4-thiadiazol-2-ylthio group at the 7-position

Synthesis of 1′-*N*-modified novel LCM derivatives possessing the substituted 1,3,4-thiadiazol-2-ylthio group at the 7-position is shown in Scheme 3. Desired 1′-*N*-modified derivatives **38**, **39**, **41** and **45** were prepared from **34** or **35** by reductive aminoalkylation. Consequently, the TBS groups of **39** and **41** were removed by TBAF to give compounds **40** and **42**, respectively. Compound **43** was



Scheme 2 Synthesis of key intermediates **23**–**27** and transformation of **23** and **36**. Conditions: (**a**) MTL, DCC, HOBt, DMF, r.t., 3–23 h; (**b**) TMSCl, HMDS, Py, r.t., 1–7 h; (**c**) 6 N AcOH, MeOH, r.t., 0.5–11 h; (**d**) DEAD, PPh_3, HSR (the corresponding heteroaromatic thiols), THF, 0 °C to r.t., 3–18 h; (**e**) 1 N HCl, MeOH, r.t., 0.5–2.5 h; (**f**) 4 N HCl-EtOAc, MeOH, 0 °C to r.t., 4.5 h; (**g**) 4 N HCl-EtOAc, MeOH, r.t., 2–2.5 h; (**h**) DEAD, PPh_3, 5-(5-(methylamino)thiazol-4-yl)-1,3,4-thiadiazole-2-thiol, THF, r.t., 2 h, (**i**) 1 N HCl, MeOH, r.t., 1 h. DCC, dicyclohexylcarbodiimide; DEAD, diethyl azodicarboxylate; DMF, dimethylformamide; HMDS, hexamethyldisilazane; HOBt, 1-hydroxybenzotriazol; MTL, methyl 1-thio-α-lincosamide; PPh_3, triphenylphosphine; Py, pyridine; THF, tetrahydrofuran; TBSCl, *tert*-butyldimethylsilyl chloride; r.t., room temperature.

synthesized in application of (*R*)-2-methyloxirane under the basic condition from **35**. Compound **44** was also prepared by acetic anhydride from **35**.

Synthesis of novel LCM derivatives 48 and 53 possessing a germinal bis-propyl moiety or a 3-(dimethylamino)propyl group at the 4′-position

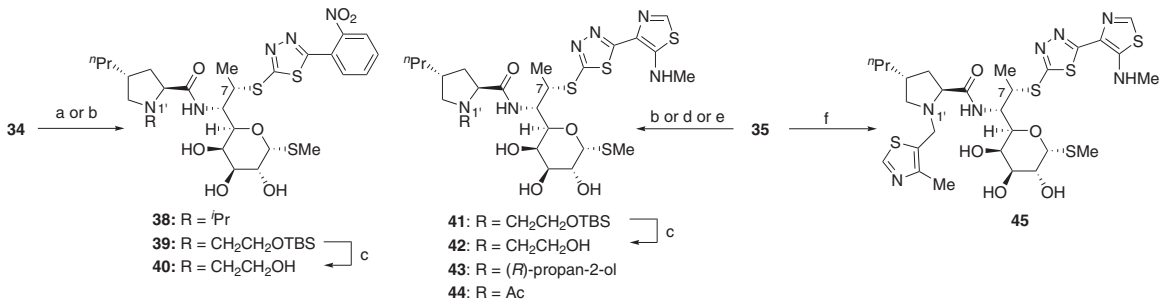
Synthesis of novel LCM derivatives **48** and **53** possessing a germinal bis-propyl moiety and a 3-(dimethylamino)propyl group, respectively, at the 4′-position, and having the 5-(2-nitrophenyl)-1,3,4-thiadiazol-2-ylthio group at the 7-position, is shown in Scheme 4. Key intermediates **47** and **50** were, respectively, synthesized in the application of condensation, tetrakis-*O*-trimethylsilylation, and selective deprotection with the similar procedure to compound **23**. Later, a 7-*S*-substituent was introduced to **47** by the Mitsunobu reaction, and deprotection gave a desired derivative **48**. Compound **50** was transformed to **51** by the Mitsunobu reaction and desilylation. Then, compound **51** was reacted with sodium azide, and an azide group was reduced to an amino group with triphenylphosphine-water. The afforded amino group in **52** was applied with reductive aminoalkylation followed by deprotection of the Boc group under the acidic condition with trifluoroacetic acid (TFA) to give the desired novel derivative **53**. ^{1}H NMR spectra of compounds **47** and **50** were also observed as broad peaks by the influence of a Boc group. Although the final product **53** could be purified as a single molecule, both intermediates **51** and **52** partially included impurity that was not removed by purification steps.

Synthesis of novel LCM derivatives with a variety of 4′-substituents possessing the substituted 1,3,4-thiadiazol-2-yl-thio group at the 7-position

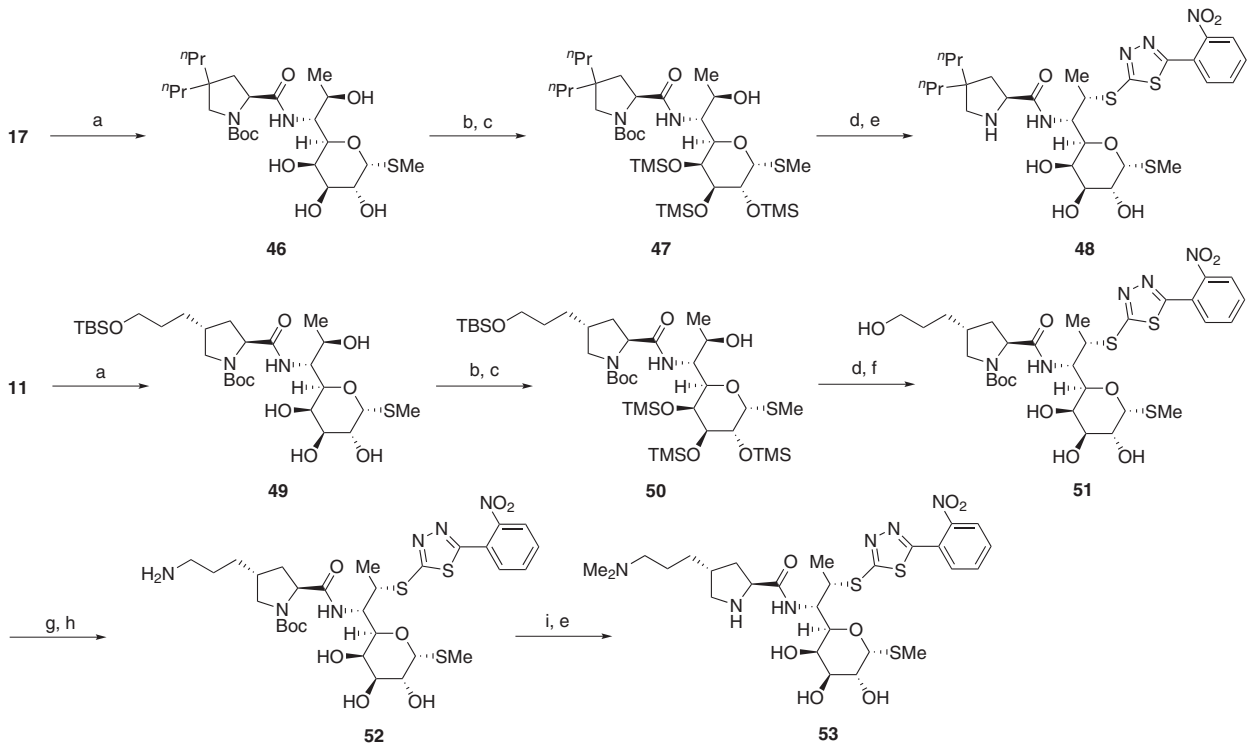
Synthesis of novel LCM derivatives having a 3-methoxypropyl, an *i*-butyl or an *n*-pentyl group at the 4′-position, whose 7-position was the substituted 1,3,4-thiadiazol-2-yl-thio group, is shown in Scheme 5. Compounds **54–56** and **58** were prepared from key intermediates **24–26** by the similar procedure to compound **48**. Reductive aminoalkylation of **56** and **58** afforded the desired compounds **57** and **59**, respectively.

Synthesis of proline-modified novel LCM derivatives possessing a (4-morpholinocarbonyl)phenylthio group at the 7-position

Synthesis of proline-modified novel LCM derivatives possessing a (4-morpholinocarbonyl)phenylthio group at the 7-position is shown in Scheme 6. Because LCM derivatives possessing a (4-morpholinocarbonyl)phenylthio group at the 7-position exhibited improved antibacterial activities, we designed compounds **67–69**. We already reported the methanesulfonyl (Ms) route[37,38] to introduce a phenyl group via sulfur atom at the 7-position. The Ms route was applied to compounds **23** and **27** to give intermediates **60** and **61**, respectively. ^{1}H NMR spectra of compounds **60–62** were observed as two sets of



Scheme 3 Synthesis of 1′-*N*-modified novel LCM derivatives possessing the substituted 1,3,4-thiadiazol-2-ylthio group at the 7-position. Conditions: (**a**) acetone, $NaBH(OAc)_3$, AcOH, $ClCH_2CH_2Cl$, 0 °C to r.t., 15 h; (**b**) 2-(*tert*-butyldimethylsilyloxy)acetaldehyde, $NaBH(OAc)_3$, AcOH, $ClCH_2CH_2Cl$, 0 °C to r.t., 15 h for **39**, r.t., 15 h for **41**; (**c**) TBAF, THF, 0 °C to r.t., 15 h for **40**, 5 h for **42**; (**d**) (*R*)-2-methyloxirane, *i*-Pr_2NEt, MeOH, 0 °C, 16 h; (**e**) Ac_2O, MeOH, 0 °C, 1.5 h; (**f**) 4-methylthiazole-5-carbaldehyde, $NaBH(OAc)_3$, AcOH, MeOH, r.t., 15 h. LCM, lincomycin; TBAF, tetra-*n*-butylammonium fluoride; THF, tetrahydrofuran; r.t., room temperature.



Scheme 4 Synthesis of **48** and **53**. Conditions: (**a**) MTL, DCC, HOBt, DMF, r.t., 14 h; (**b**) TMSCl, HMDS, Py, r.t., 1 h from **46**, 20 min from **49**; (**c**) 6 N AcOH, MeOH, r.t., 1–1.5 h; (**d**) 5-(2-nitrophenyl)-1,3,4-thiadiazole-2-thiol, DEAD, PPh_3, THF, 0 °C to r.t., 6–10 h; (**e**) TFA, 0 °C to r.t., 30 min; (**f**) TBAF, AcOH, THF, r.t., 5 h; (**g**) NaN_3, CBr_4, PPh_3, DMF, r.t. to 50 °C, 2 h; (**h**) PPh_3, H_2O, THF, r.t. to 50 °C, 3 h; (**i**) HCHO, $NaBH(OAc)_3$, AcOH, MeOH, r.t., 20 min. DCC, dicyclohexylcarbodiimide; DEAD, diethyl azodicarboxylate; DMF, dimethylformamide; HMDS, hexamethyldisilazane; HOBt, 1-hydroxybenzotriazol; MTL, methyl 1-thio-α-lincosamide; PPh_3, triphenylphosphine; Py, pyridine; TBAF, tetra-*n*-butylammonium fluoride; TFA, trifluoroacetic acid; THF, tetrahydrofuran; r.t., room temperature.

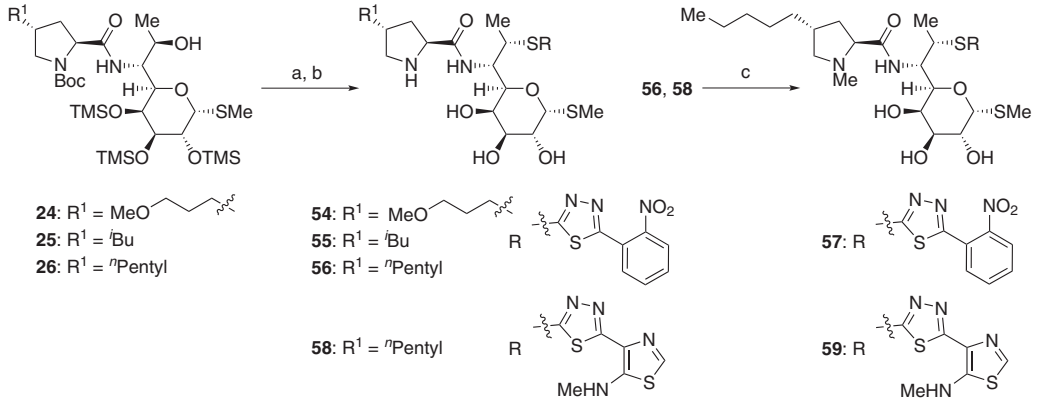
signals because of a rotamer by the Boc group. The desired analogs **67** and **68** were prepared from **60** and **62**, respectively, by (i) hydrolysis under the basic condition, (ii) condensation with morpholine and (iii) deprotection of the Boc group with TFA. Compound **69** was synthesized from **68** by reductive aminoalkylation with HCHO and $NaBH(OAc)_3$ in an acidic condition by AcOH.

Synthesis of 73 and 75

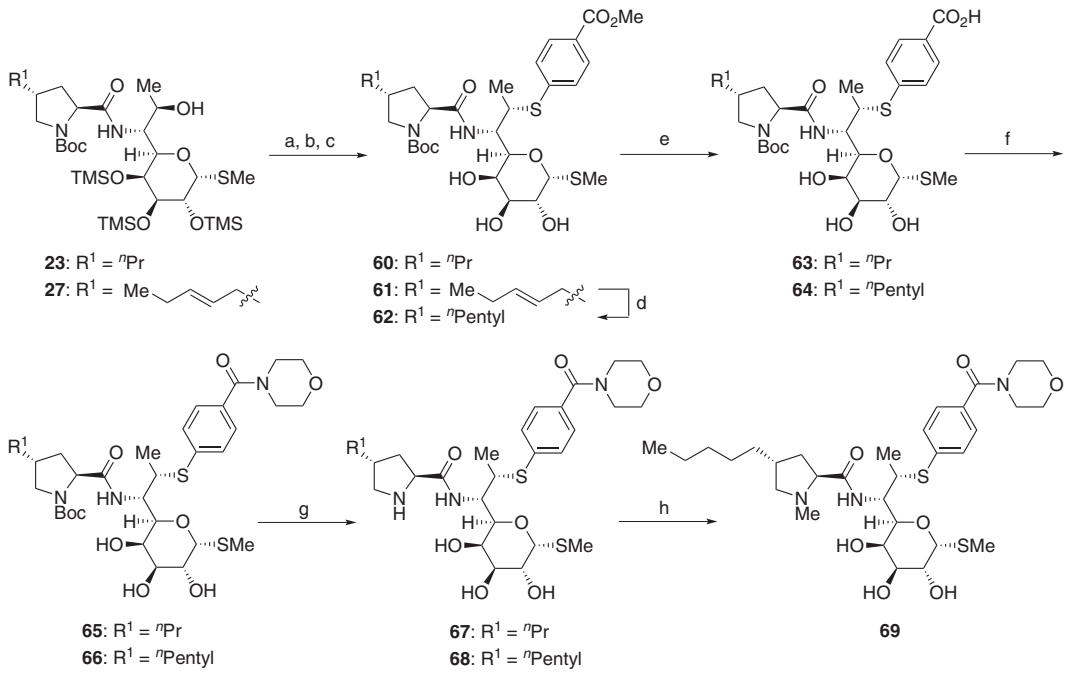
Synthesis of novel LCM derivatives **73** and **75** is shown in Scheme 7. We have already reported a Pd-catalyzed cross-coupling route to introduce an aryl group at the 7-position of 7(*S*)-7-deoxy-7-mercaptlincomycin.[40] A key intermediate **71** was prepared via four steps from **23**, and precursors **72** and **74** were synthesized by Pd-catalyzed cross-coupling reaction of **71**[40,50] with the corresponding bromides. Deprotection of the Boc group afforded the desired analogs **73** and **75**. ^1H NMR spectra of compounds **72** and **74** were observed as two sets of signals because of a rotamer by the Boc group, but both of the final compounds **73** and **75** showed a single set and sharp peaks in NMR spectra.

SAR analysis of 7-*S*-substituted 1′-*NH* LCM derivatives (32–35) and 1′-*N*-Me analog 37

Antibacterial activity of LCM was reduced by 1′-*N*-demethylation, but that of CLDM was enhanced by 1′-*N*-demethylation. Thus, we were



Scheme 5 Synthesis of novel LCM derivatives with a variety of 4′-substituents possessing the substituted 1,3,4-thiadiazol-2-yl-thio group at the 7-position. Conditions: (**a**) DEAD, PPh_3, HSR, THF, 0 °C to r.t., 9.5–24 h; (**b**) TFA, 0 °C to r.t., 15–90 min; (**c**) HCHO, $NaBH(OAc)_3$, AcOH, MeOH, r.t., 1 h. DEAD, diethyl azodicarboxylate; LCM, lincomycin; TFA, trifluoroacetic acid; THF, tetrahydrofuran; r.t., room temperature.

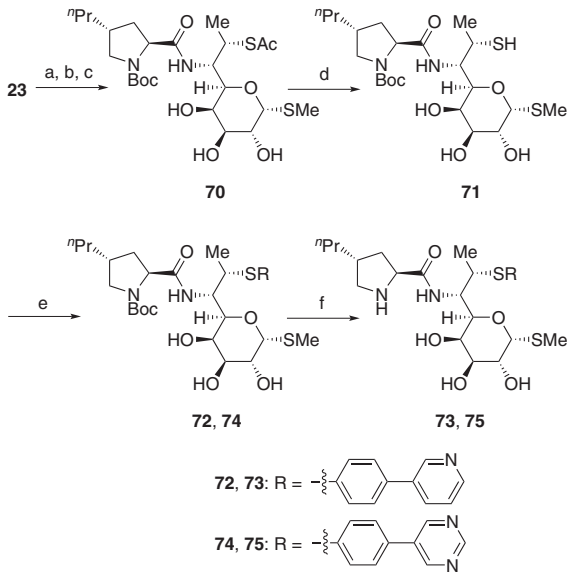


Scheme 6 Synthesis of proline-modified novel LCM derivatives possessing a (4-morpholinocarbonyl)phenylthio group at the 7-position. Conditions: (**a**) MsCl, NEt_3, $CHCl_3$, r.t., 30 min; (**b**) methyl 4-mercaptobenzoate, K_2CO_3, DMF, 80–100 °C, 1–6 h; (**c**) 1 N HCl, MeOH, r.t., 20 min; (**d**) H_2, Pd/C, MeOH, r.t.,15 h; (**e**) 1 N NaOH, MeOH, r.t., 1–7 days; (**f**) morpholine, WSC, HOBt, DMF, r.t. 22–62 h, (**g**) TFA, −15 to 0 °C, 40 min and (**h**) HCHO, AcOH, $NaBH(OAc)_3$, MeOH, 30 min. DMF, dimethylformamide; HOBt, 1-hydroxybenzotriazol; LCM, lincomycin; r.t., room temperature; TFA, trifluoroacetic acid; WSC, water-soluble carbodiimide.

interested in the potency of 1′-*N*-demethyl products of 7(*S*)-substituted LCM derivatives **1–3** as shown in Table 1. Thus, we synthesized 1′-demethyl analogs of **1–3** (**32–34**) and compounds **35** and **37** possessing an alternative 7-substituent. Antibacterial activities of those are shown in Table 2. Among them, 1′-*NH* derivatives **32**, **34** and **35** exhibited improved antibacterial activities against *S. pneumoniae* with *erm* gene compared with the corresponding 1′-*N*-methyl analogs (**1**,**3** and **37**), respectively. Because **34** and **35** especially showed enhanced antibacterial activities against the target pathogens, we found that double modifications at the C-6 and C-7 positions were important to improve antibacterial activities against *S. pneumoniae* with *erm* gene.

SAR analysis of antibacterial activities (MIC, μg ml^{-1}) of 7-*S*-substituted 1′-*N*-modified LCM derivatives (38, 40 and 42–45)

For the purpose of accumulating detail information of SAR at the 1′-position, we synthesized novel LCM derivatives possessing various substituents at the 1′-position. At this point, the 5-(2-nitrophenyl)-1,3,4-thiadiazol-2-yl group and the 4-(5-methylamino-thiazole-4-yl)-1,3,4-thiadiazol-2-yl group were selected as a 7-substituent because of their SAR analysis (Tables 1 and 2). Consequently, compounds **40** and **42**, possessing a 2-hydroxyethyl group at the 1′-position, showed antibacterial activities against target pathogens as shown in Table 3, but we concluded that it was difficult to enhance antibacterial activities against *S. pneumoniae* with *erm* gene by introducing an alternative



Scheme 7 Synthesis of **73** and **75**. Conditions: (**a**) MsCl, NEt_3, CH_2Cl_2, 0 °C, 1 h; (**b**) AcSK, DMF, 60 °C, 10 h; (**c**) 1 N HCl, MeOH, r.t., 1 h; (**d**) NaOMe, MeOH, r.t., 1.5 h; (**e**) 3-(4-bromophenyl)pyridine or 5-(4-bromophenyl)pyrimidine, $Pd_2(DBA)_3$, Xantphos, *i*-Pr_2NEt, dioxane, reflux, 5–6 h; (**f**) TFA, CH_2Cl_2, −20 °C to r.t., 3–5.5 h. DMF, dimethylformamide; r.t., room temperature; TFA, trifluoroacetic acid; Xantphos, 4,5-bis(diphenylphosphino)-9,9-dimethylxanthene, $Pd_2(DBA)_3$, tris(dibenzylideneacetone)dipalladium(0).

substituent at the 1′-position, except a hydrogen atom or a methyl group. These results were closely related to SAR of 1′-*N*-alkyl-1′-demethyllincomycin.[17,21,24] Then, we selected a hydrogen atom and a methyl group at the 1′-position for modification of the proline moiety.

SAR analysis of 7-*S*-substituted 1′-*N*- and 4′-modified LCM derivatives (48 and 53–59) and TEL

To confirm whether 7-*S*-substituted LCM analogs show similar antibacterial spectra to those reported previously, we synthesized 1′-demethyllincomycin derivatives possessing various substituents at the 4′-position. Structures of 7-*S*-substituents followed those in Table 3. As shown in Table 4, compounds **56** and **57**, possessing an *n*-pentyl group instead of an *n*-propyl group at the 4′-position, exhibited strong antibacterial activities against resistant bacteria with *erm* gene. Although antibacterial activities of TEL against *S. pneumoniae* with *erm* gene were stronger than those of **56**, antibacterial activities of **56** against *S. pyogenes* with *erm* gene and resistant bacteria with *mef* gene were remarkably stronger than those of TEL.

SAR analysis of 7-*S*-substituted 1′-*N*- and 4′-modified LCM derivatives with an alternative 7-*S*-substituent (67–69, 73 and 75)

We have reported significant potent antibacterial activities of LCM derivatives possessing a substituted phenyl group at the C-7 position so far.[40] This time, we transformed the proline moiety (the 1′-and 4′-position) of 7-*S*-substituted phenyl derivatives as shown in Table 5, such as 7(*S*)-7-{4-(morphorinocarbonyl)phenylthio}lincomycin (**76**), 7(*S*)-7-{4-(pyridin-3-yl)phenylthio}lincomycin (**77**) and 7(*S*)-7-{4-(pyrimidin-5-yl)phenylthio}lincomycin (**78**). Their antibacterial activities are shown in Table 5. The *n*-pentyl analogs **68** and **69** could not exhibit improved antibacterial activities against *S. pneumoniae* and *S. pyogenes* with *erm* gene compared with **56** and **57**. On the other hand, 1′-*N*H LCM derivatives **73** and **75** with a 4-(pyridin-3-yl)phenyl group and a 4-(pyrimidin-5-yl)phenyl group, respectively, exhibited markedly potent antibacterial activities against *S. pneumoniae* with *erm* gene. We confirmed that combination modification at the C-6 position (the proline moiety) and the C-7 position was important to enhance antibacterial activities against *S. pneumoniae* and *S. pyogenes* with *erm* gene.

CONCLUSION

To modify both the C-6 position (the proline moiety) and the C-7 position of LCM, we firstly prepared various substituted proline intermediates. The intermediates were coupled with MTL to give a wide variety of 1′-*N*-Boc-1′-demethyllincomycin derivatives. The 7-*S*-substituents were introduced as follows. Key intermediates **23–26**, **36**, **47** and **50** were synthesized by the Mitsunobu reaction[37,38] with the corresponding thiol. Other key intermediates **23** and **27** were transformed to 7-*S*-benzoate by an S_N2 reaction[37,38] via Ms derivatives, and the benzoate moiety was finally converted to a 4-(morpholinocarbonyl)phenyl moiety (Scheme 6). 7(*S*)-7-Deoxy-7-thiolincomycin (**71**) was coupled with biaryl bromide under the Pd-catalyzed cross-coupling reaction (Scheme 7).[40,50] Compounds **38**, **40** and **42–45** were also prepared from **34** or **35**. Those methodologies were found to be very practical to synthesize various LCM analogs modified at the C-6 and -7 positions.

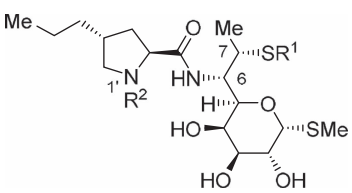
By SAR analysis of combination modification at the 1′- and 7-position, we concluded that it was difficult to enhance antibacterial activities against *S. pneumoniae* with *erm* gene by introducing an alternative substituent at the 1′-position, except a hydrogen atom or a methyl group. Then, we selected a hydrogen atom and a methyl group at the 1′-position for further modification of the proline moiety. We next modified the 4′-position (in the proline moiety) of LCM derivatives possessing a 5-(2-nitrophenyl)-1,3,4-thiadiazol-2-yl group or a 4-(5-methylamino-thiazol-4-yl)-1,3,4-thiadiazol-2-yl group at the C-7 position (Table 4). Compounds **56** and **57**, possessing an *n*-pentyl group instead of an *n*-propyl group at the 4′-position, exhibited strong antibacterial activities against resistant bacteria with *erm* gene. Although antibacterial activities of TEL against *S. pneumoniae* with *erm* gene were stronger than those of **56**, antibacterial activities of **56** against *S. pyogenes* with *erm* gene and resistant bacteria with *mef* gene were remarkably stronger than those of TEL. We found that combination modification at the C-6 position (the proline moiety) and the C-7 position was quite important to enhance antibacterial activities against *S. pneumoniae* and *S. pyogenes* with *erm* gene. The above SAR might be partially related to the polarity or water solubility of a molecule, but we only have limited SAR information. Further combination modification at the C-6 and C-7 positions of LCM analogs is in progress.

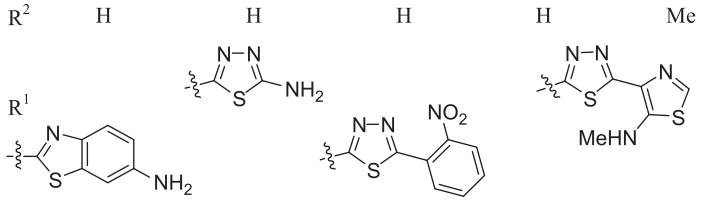
EXPERIMENTAL PROCEDURES

General methods

^1H NMR spectra were measured with a BRUKER Ascend 400 NMR spectrometer (Bruker Corporation, Coventry, UK) for 400 MHz, JEOL JNM-GSX 400 NMR spectrometer (JEOL,Tokyo, Japan) for 400 MHz or a Varian Gemini 300 NMR spectrometer (Varian, Palo Alto, CA, USA) for 300 MHz in $CDCl_3$ or CD_3OD. TMS (0 p.p.m.) in $CDCl_3$ or CD_3OD was used as an internal reference standard. Mass spectra (MS) were obtained on a JEOL JMS-700 mass spectrometer (JEOL) or Agilent Technologies 6530-Q-TOF LC/MS mass spectrometer (Agilent Technologies, Santa Clara, CA, USA). The optical rotations were recorded with Jasco P-2300 digital polarimeter (Jasco Corporation, Tokyo, Japan). Column chromatography was performed with silica gel (Wakogel C200; Wako Pure Chemical Industries, Osaka, Japan). Preparative thin layer chromatography was performed with silica gel (Merck, Darmstadt, Germany; TLC plates Silica gel 60 F254). All organic extracts were dried over

Table 2 Anti-bacterial activities (MIC, μg ml^{-1}) of 7-*S*-substituted 1′-*N*H LCM derivatives (32–35) and 1′-*N*-Me analog 37





Test organism[a]	*Characteristics*[b]	***32***	***33***	***34***	***35***	***37***
Streptococcus pneumoniae DP1 type I	Susceptible	0.06	0.5	0.03	0.12	0.12
S. pneumoniae-2	Susceptible	0.12	0.5	0.03	0.12	0.12
S. pneumoniae-3	Susceptible	0.03	0.5	0.03	0.12	0.12
S. pneumoniae-4	*ermAM* methylase (c)	8	128	1	1	2
S. pneumoniae-5	*ermAM* methylase (c)	64	>128	2	2	4
S. pneumoniae-6	*ermAM* methylase (c)+*mefE*	64	>128	8	8	16
S. pneumoniae-7	*ermAM* methylase (i)	8	128	1	2	4
S. pneumoniae-8	*ermAM* methylase (i)	8	128	1	2	2
S. pneumoniae-9	*mefE* efflux	0.06	0.5	0.015	0.12	0.06
Streptococcus pyogenes Cook	Susceptible	0.03	0.5	0.03	0.12	0.12
S. pyogenes-2	*ermAM* methylase (c)	8	32	1	1	2
S. pyogenes-3	*mefE* efflux	0.12	0.5	0.06	0.12	0.12
Haemophilus influenzae	Susceptible	32	64	16	8	4
H. influenzae-2	Susceptible	32	>128	8	8	4
H. influenzae-3	Susceptible	32	128	16	16	16
H. influenzae-4	⊿acr	0.5	8	0.25	0.25	0.25

Abbreviation: LCM, lincomycin.
[a]All strains except standard organisms were clinically isolated.
[b](c), constitutive; (i), inducible.

anhydrous $MgSO_4$, and the solvent was removed with a rotary evaporator under reduced pressure.

(2*S*,4*R*)-1-*N*-(*tert*-butoxycarbonyl)-4-*n*-propylpyrrolidine-2-carboxylic acid (7)

To a solution of compound **4** (1.0 g, 2.89 mmol) in MeOH (10 ml) was added Pd/C (100 mg), and then vigorously stirred in hydrogen atmosphere at room temperature for 2 h. The mixture was filtrated with celite, and then the solution was concentrated under reduced pressure. The resulting residue was filtrated with Chromatodisc (0.45 μm) (Kurabo Industries, Osaka, Japan). The filtrated solution was concentrated under reduced pressure to obtain the title compound (745 mg, quant) as a colorless solid. FAB-MS *m/z* 258 $(M+H)^+$ as $C_{13}H_{23}NO_4$; ^1H NMR (400 MHz, CD_3OD)) δ 0.70–0.93 (m, 3H), 1.10–1.41 (m, 13H), 1.65–1.87 (m, 1H), 1.93–2.06 (m, 1H), 2.10–2.27 (m, 1H), 2.73–2.89 (m, 1H), 3.43–3.63 (m, 1H) and 4.00–4.25 (m, 1H).

(2*S*,4*R*)-2-benzyl 1-*tert*-butyl 4-(3-hydroxypropyl)pyrrolidine-1,2-dicarboxylate (8)

To a solution of compound **4** (1.03 g, 2.98 mmol) in tetrahydrofuran (THF) (3 ml) was added 0.5 M 9-BBN in THF solution (8.95 ml, 4.47 mmol) and stirred at 50 °C for 1 h. Then, 1N NaOH (4 ml) and 35% H_2O_2 (4 ml) were added to the mixture and stirred at 0 °C for 2 h. The solution was added to the saturated aqueous NaCl. The desired compound was extracted with ethyl acetate, and then the organic phase was dried over Na_2SO_4, filtrated and concentrated under reduced pressure. The resulting residue was purified by silica gel column chromatography (hexane/ethyl acetate = 3/1 to 1/2) to obtain the title compound (937 mg, 87%) as a colorless solid. EI-MS *m/z* 363 $(M)^+$ as $C_{20}H_{29}NO_5$; ^1H NMR (400 MHz, $CDCl_3$) δ 1.31–1.48 (m, 11H), 1.49–1.60 (m, 2H), 1.75–1.92 (m, 1H), 2.05–2.14 (m, 1H), 2.19–2.39 (m, 1H), 2.88–3.05 (m, 1H), 3.57–3.83 (m, 3H), 4.25–4.50 (m, 1H), 4.96–5.32 (m, 2H) and 7.27–7.47 (m, 5H).

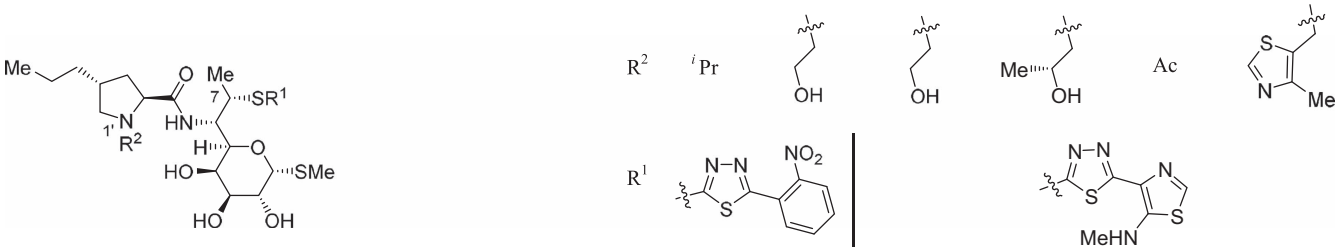
(2*S*,4*R*)-2-Benzyl 1-*tert*-butyl 4-(3-(*tert*-butyldimethylsilyloxy) propyl)pyrrolidine-1,2-dicarboxylate (9)

To a solution of compound **8** (2.80 g, 7.70 mmol) in dimethylformamide (DMF) (15 ml) were added imidazole (1.05 g, 15.4 mmol) and TBSCl (1.74 g, 11.56 mmol), and then stirred at room temperature for 30 min. The mixture was extracted with ethyl acetate and then the organic phase was dried over Na_2SO_4, filtrated and concentrated under reduced pressure. The resulting residue was pumped up to obtain the title compound (3.50 g, crude). The total amount of this compound was used without purification to synthesize **11**.

(2*S*,4*R*)-2-benzyl 1-*tert*-butyl 4-(3-methoxypropyl)pyrrolidine-1,2-dicarboxylate (10)

To a solution of compound **8** (900 mg, 2.48 mmol) in DMF (9 ml) was added 55% NaH in oil (99.2 mg, 3.72 mmol) and stirred at room temperature for 30 min. To the mixture was added methyl iodide (924 μl, 14.86 mmol) and then stirred at room temperature for 1 h. The solution was added to the saturated aqueous NaCl. The desired compound was extracted with ethyl acetate and then the organic phase was dried over Na_2SO_4, filtrated and concentrated under reduced pressure. The resulting residue was purified by silica gel column chromatography (hexane/ethyl acetate = 20/1 to 4/1) to obtain the title compound (240 mg, 26%) as a colorless oil. ESI-MS *m/z* 378 $(M+H)^+$ as $C_{21}H_{31}NO_5$; ^1H NMR (400 MHz, $CDCl_3$) δ 1.33, 1.45 (s x 2, 9H), 1.35–1.50 (m, 2H), 1.50–1.57 (m, 2H), 1.75–1.91 (m, 1H), 2.03–2.13 (m, 1H), 2.18–2.33 (m, 1H), 2.90–3.04 (m, 1H), 3.31 (s, 3H), 3.25–3.40 (m, 2H), 3.59–3.80 (m, 1H), 4.25–4.47 (m, 1H), 5.03–5.29 (m, 2H) and 7.29–7.42 (m, 5H).

Table 3 Anti-bacterial activities (MIC, μg ml^{-1}) of 7-*S*-substituted 1′-*N*-modified LCM derivatives (38, 40 and 42–45)



Test organism[a]	*Characteristics*[b]	***38***	***40***	***42***	***43***	***44***	***45***
Streptococcus pneumoniae DP1 type I	Susceptible	0.5	0.06	0.12	2	2	1
S. pneumoniae-2	Susceptible	0.5	0.06	0.12	4	2	2
S. pneumoniae-3	Susceptible	0.5	0.06	0.12	2	2	2
S. pneumoniae-4	*ermAM* methylase (c)	128	8	2	>128	64	128
S. pneumoniae-5	*ermAM* methylase (c)	128	8	2	>128	64	128
S. pneumoniae-6	*ermAM* methylase (c)+*mefE*	>128	64	16	>128	128	>128
S. pneumoniae-7	*ermAM* methylase (i)	128	4	2	>128	64	128
S. pneumoniae-8	*ermAM* methylase (i)	128	4	2	>128	64	128
S. pneumoniae-9	*mefE* efflux	0.5	0.03	0.06	1	1	1
Streptococcus pyogenes Cook	Susceptible	0.5	0.03	0.12	2	2	2
S. pyogenes-2	*ermAM* methylase (c)	64	4	1	>128	32	32
S. pyogenes-3	*mefE* efflux	1	0.06	0.12	2	2	2
Haemophilus influenzae	Susceptible	>128	16	4	>128	>128	>128
H. influenzae-2	Susceptible	>128	8	4	>128	>128	>128
H. influenzae-3	Susceptible	>128	32	16	>128	>128	>128
H. influenzae-4	⊿acr	16	0.25	0.25	32	16	8

Abbreviation: LCM, lincomycin.
[a]All strains except standard organisms were clinically isolated.
[b](c), constitutive; (i), inducible.

(2*S*,4*R*)-1-*N*-(*tert*-butoxycarbonyl)-4-(3-(*tert*-butyldimethylsilyloxy) propyl)pyrrolidine-2-carboxylic acid (11)

Compound **9** (3.50 g, crude) in MeOH (50 ml) were treated for 30 min according to the similar procedure as described for the preparation of **7** to afford **11** (3.02 g, crude). The total amount of this compound was used to synthesize **49**.

(2*S*,4*R*)-1-*N*-(*tert*-butoxycarbonyl)-4-(3-methoxypropyl) pyrrolidine-2-carboxylic acid (12)

Compound **10** (200 mg, 0.530 mmol) in MeOH (2 ml) were treated for 1 h according to the similar procedure as described for the preparation of **7** to afford **12** (152 mg, crude). The total amount of this compound was used without purification to synthesize **19**.

(2*S*,4*R*)-1-*N*-(*tert*-butoxycarbonyl)-4-*i*-butylpyrrolidine-2-carboxylic acid (13)

Compound **5** (195.3 mg, 0.54 mmol) in MeOH (2 ml) were treated for 30 min according to the similar procedure as described for the preparation of **7** to afford **13** (141.1 mg, 96%) as an off-white solid. ESI-MS *m/z* 272 $(M+H)^+$ as $C_{14}H_{25}NO_4$; ^{1}H NMR (400 MHz, CD_3OD) δ 0.90 (d, *J*=6.7 Hz, 3H), 0.92 (d, *J*=6.7 Hz, 3H), 1.23–1.35 (m, 2H), 1.41, 1.45 (s x 2, 9H), 1.51–1.64 (m, 1H), 1.76–1.95 (m, 1H), 2.07–2.15 (m, 1H), 2.28–2.44 (m, 1H), 2.85–3.00 (m, 1H), 3.57–3.74 (m, 1H) and 4.18–4.32 (m, 1H).

(2*S*,4*R*)-1-*N*-(*tert*-butoxycarbonyl)-4-*n*-pentylpyrrolidine-2-carboxylic acid (14)

Compound **6** (1.69 g, 4.53 mmol) in MeOH (20 ml) were treated for 2 h according to the similar procedure as described for the preparation of **7** to afford **14** (1.16 g, 89.5%) as a colorless solid. FAB-MS *m/z* 286 $(M+H)^+$ as $C_{15}H_{27}NO_4$; ^{1}H NMR (400 MHz, CD_3OD) δ 0.80–0.98 (m, 3H), 1.20–1.51 (m, 8H), 1.42, 1.46 (s x 2, 9H), 1.78–1.98 (m, 1H), 2.10 (ddd, *J*=12.7, 6.2, 2.1 Hz, 1H), 2.19–2.36 (m, 1H), 2.85–3.01 (m, 1H), 3.57–3.75 (m, 1H), 4.17–4.33 (m, 1H).

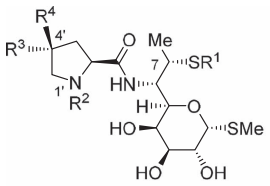
(2*S*,4*R*)-1-*N*-(*tert*-butoxycarbonyl)-4-((*E*)-pent-2-enyl)pyrrolidine-2-carboxylic acid (15)

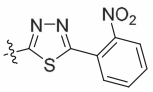
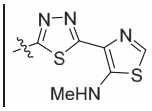
To a solution of compound **6** (2.0 g, 5.79 mmol) in MeOH (20 ml) was added 1 M aqueous NaOH (20 ml) and stirred at room temperature for 22 h. The mixture was diluted with H_2O and Et_2O and washed by Et_2O. Aqueous layer was added to the saturated aqueous citric acid, extracted with ethyl acetate and then the organic phase was washed with H_2O, dried over Na_2SO_4, filtrated and concentrated under reduced pressure to obtain the title compound (1.55 g, 94.5%) as a colorless solid. FAB-MS *m/z* 284 $(M+H)^+$ as $C_{15}H_{25}NO_4$; ^{1}H NMR (400 MHz, CD_3OD) δ 0.89–1.05 (m, 3H), 1.41 (s, 6H), 1.45 (s, 3H), 1.85–2.15 (m, 6H), 2.23–2.39 (m, 1H), 2.92–3.09 (m, 1H), 3.52–3.68 (m, 1H), 4.18–4.31 (m, 1H), 5.30–5.44 (m, 1H) and 5.46–5.60 (m, 1H).

2(*S*)-1-*N*-(*tert*-butoxycarbonyl)-4,4-di-*n*-propylpyrrolidine-2-carboxylic acid (17)

Compound **16** (2.0 g, 5.19 mmol) in MeOH (23 ml) were treated for 4.5 h according to the similar procedure as described for the preparation of **7** to

Table 4 Anti-bacterial activities (MIC, μg ml^{-1}) of 7-*S*-substituted 1′-*N*- and 4′-modified LCM derivatives (48 and 53–59) and TEL



R^4	^{n}Pr	H	H	H	H	H	H	H	
R^3	^{n}Pr	3-NMe$_2$-Pr	3-MeO-Pr	^{i}Bu	^{n}Pentyl	^{n}Pentyl	^{n}Pentyl	^{n}Pentyl	
R^2	H	H	H	H	H	Me	H	Me	
R^1									

Test organism[a]	*Characteristics*[b]	***48***	***53***	***54***	***55***	***56***	***57***	***58***	***59***	*TEL*
Streptococcus pneumoniae DP1 type I	Susceptible	16	128	0.06	0.06	≦0.008	≦0.008	0.03	0.03	≦0.008
S. pneumoniae-2	Susceptible	16	128	0.06	0.06	≦0.008	0.015	0.06	0.06	≦0.008
S. pneumoniae-3	Susceptible	16	32	0.06	0.03	≦0.008	≦0.008	NT	0.015	≦0.008
S. pneumoniae-4	*ermAM* methylase (c)	64	>128	32	32	0.5	1	1	2	0.5
S. pneumoniae-5	*ermAM* methylase (c)	64	>128	16	32	0.5	1	2	4	2
S. pneumoniae-6	*ermAM* methylase (c) + *mefE*	64	>128	64	128	2	4	4	8	1
S. pneumoniae-7	*ermAM* methylase (i)	64	>128	8	8	NT	NT	NT	NT	0.03
S. pneumoniae-8	*ermAM* methylase (i)	64	>128	8	8	0.5	0.5	1	2	0.03
S. pneumoniae-9	*mefE* efflux	16	64	0.06	0.03	≦0.008	≦0.008	0.06	0.015	0.06
Streptococcus pyogenes Cook	Susceptible	2	8	0.06	0.03	0.015	0.015	0.12	0.06	≦0.008
S. pyogenes-2	*ermAM* methylase (c)	64	>128	8	8	0.5	0.5	0.5	0.5	16
S. pyogenes-3	*mefE* efflux	8	8	0.12	0.06	0.015	0.03	0.06	0.06	0.25
Haemophilus influenzae	Susceptible	>128	>128	128	64	8	8	16	8	0.5
H. influenzae-2	Susceptible	128	>128	128	32	8	4	8	4	2
H. influenzae-3	Susceptible	>128	>128	>128	128	16	8	16	8	1
H. influenzae-4	⊿acr	32	>128	2	2	0.25	0.06	0.25	0.12	0.25

Abbreviations: LCM, lincomycin; NT, not tested; TEL, telithromycin.
[a]All strains except standard organisms were clinically isolated.
[b](c): constitutive; (i): inducible.

afford **17** (1.55 g, quant) as an off-white oil. ESI-MS *m/z* 300 (M+H)$^+$ as $C_{16}H_{29}NO_4$; ^{1}H NMR (400 MHz, CD_3OD) δ 0.91 (t, *J*=6.9 Hz, 3H), 0.93 (t, *J*=6.9 Hz, 3H), 1.17–1.48 (m, 8H), 1.42, 1.45 (s x 2, 9H), 1.64–1.75 (m, 1H), 2.11–2.24 (m, 1H), 3.10 (d, *J*=10.6 Hz, 1H), 3.35–3.42 (m, 1H), 4.01–4.25 (m, 1H).

1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyllincomycin (18)

To a solution of compound **7** (745.0 mg, 2.90 mmol) in DMF (27 ml) were added 1-hydroxybenzotriazole (469.4 mg, 3.47 mmol), *N*,*N*′-dicyclohexylcarbodiimide (716.8 mg, 3.47 mmol) and MTL (1.10 g, 4.34 mmol) and stirred at room temperature for 15 h. The mixture was added to H_2O and ethyl acetate. The desired compound was extracted with ethyl acetate, and then the organic phase was washed with H_2O, dried over Na_2SO_4, filtrated and concentrated under reduced pressure to obtain the title compound. The total amount of this compound was used without purification to synthesize **23**. For the qualified analytical purpose, the above crude compound **18** was purified by column chromatography (ethyl acetate only) to obtain the title compound as a colorless solid. FAB-MS *m/z* 493 (M+H)$^+$ as $C_{22}H_{40}N_2O_8S$; ^{1}H NMR (400 MHz, CD_3OD) δ 0.78–0.93 (m, 3H), 1.05–1.45 (m, 7H), 1.35, 1.38 (s x 2, 9H), 1.59–1.81 (m, 1H), 1.88–2.09 (m, 1H), 1.97 (s, 3H), 2.23–2.43 (m, 1H), 2.83 (br t, *J*=10.1 Hz, 1H), 3.43–3.63 (m, 2H), 3.64–3.82 (m, 1H), 3.84–4.09 (m, 3H), 4.10–4.19 (m, 1H), 4.21–4.39 (m, 1H) and 5.14 (br d, *J*=5.4 Hz, 1H).

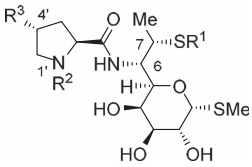
1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-4′-depropyl-4′-(3-methoxypropyl)lincomycin (19)

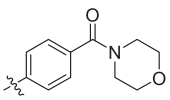
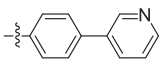
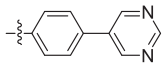
To a solution of compound **12** (152.3 mg, 0.53 mmol) in DMF (1.5 ml) were added 1-hydroxybenzotriazole (93.0 mg, 0.689 mmol), *N*,*N*′-dicyclohexylcarbodiimide (142.0 mg, 0.689 mmol) and MTL (174.0 mg, 0.689 mmol) and stirred at room temperature for 4.5 h. The mixture was added to H_2O and ethyl acetate. The desired compound was extracted with ethyl acetate, and then the organic phase was washed with H_2O, dried over Na_2SO_4, filtrated and concentrated under reduced pressure. The resulting residue was purified by silica gel column chromatography (hexane/ethyl acetate=1/1 to ethyl acetate only to ethyl acetate/MeOH=24/1) to obtain the title compound (245.0 mg, 88.5% in two steps from **10**) as a colorless solid. ESI-MS *m/z* 523 (M+H)$^+$ as $C_{23}H_{42}N_2O_9S$; ^{1}H NMR (400 MHz, CD_3OD) δ 1.15–1.29 (m, 3H), 1.44, 1.46 (s x 2, 9H), 1.36–1.66 (m, 4H), 1.73–1.88 (m, 1H), 2.06, 2.07 (s x 2, 3H), 2.09–2.17 (m, 1H), 2.33–2.50 (m, 1H), 2.93 (br t, *J*=9.9 Hz, 1H), 3.32 (s, 3H), 3.40 (t, *J*=6.3 Hz, 2H), 3.53–3.96 (m, 3H), 3.98–4.18 (m, 3H), 4.20–4.26 (m, 1H), 4.28–4.48 (m, 1H) and 5.24 (d, *J*=5.6 Hz, 1H).

1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-4′-depropyl-4′-*i*-butyllincomycin (20)

To a solution of compound **13** (57.9 mg, 0.21 mmol) in DMF (1.0 ml) were added 1-hydroxybenzotriazole (43.2 mg, 0.32 mmol), *N*,*N*′-dicyclohexylcarbodiimide (61.4 g, 0.32 mmol) and MTL (81.1 mg, 0.32 mmol) and stirred at room temperature for 1 h. The mixture was added to the saturated aqueous $NaHCO_3$ and ethyl acetate. The desired compound was extracted with ethyl acetate, and then the organic phase was dried over Na_2SO_4, filtrated and concentrated under reduced pressure. The resulting residue was purified by silica gel column chromatography (chloroform/MeOH=10/1) to obtain the title compound (100.2 mg, 92.7%) as a colorless solid. FAB-MS *m/z* 507 (M+H)$^+$ as $C_{23}H_{42}N_2O_8S$; ^{1}H NMR (400 MHz, CD_3OD) δ 0.87–0.98 (m, 6H),

Table 5 Anti-bacterial activities (MIC, μg ml^{-1}) of 7-*S*-substituted 1′-*N*- and 4′-modified LCM derivatives with an alternative 7-*S*-substituent (67–69, 73 and 75)



	R^3	^{n}Pr		^{n}Pentyl		^{n}Pr		^{n}Pr	
	R^2	H	Me	H	Me	H	Me	H	Me
	R^1								
Test organism[a]	*Characteristics*[b]	***67***	***76***[c]	***68***	***69***	***73***	***77***[c]	***75***	***78***[c]
Streptococcus pneumoniae DP1 type I	Susceptible	0.25	0.06	NT	NT	0.015	≦0.008	≦0.008	≦0.008
S. pneumoniae-2	Susceptible	0.25	0.06	0.12	0.06	0.015	≦0.008	≦0.008	≦0.008
S. pneumoniae-3	Susceptible	0.25	0.06	0.25	0.12	0.03	0.015	≦0.008	≦0.008
S. pneumoniae-4	*ermAM* methylase (c)	16	8	4	4	1	0.5	0.5	0.5
S. pneumoniae-5	*ermAM* methylase (c)	16	2	8	4	2	1	NT	1
S. pneumoniae-6	*ermAM* methylase (c)+*mefE*	64	8	16	16	4	2	2	2
S. pneumoniae-7	*ermAM* methylase (i)	4	2	2	1	0.5	0.25	0.25	0.25
S. pneumoniae-8	*ermAM* methylase (i)	8	1	0.5	0.5	0.5	0.25	0.25	0.25
S. pneumoniae-9	*mefE* efflux	0.25	0.03	0.06	0.015	≦0.008	≦0.008	≦0.008	≦0.008
Streptococcus pyogenes Cook	Susceptible	0.12	0.06	0.12	0.12	0.03	0.015	≦0.008	≦0.008
S. pyogenes-2	*ermAM* methylase (c)	4	4	0.5	0.5	0.5	0.5	0.5	0.5
S. pyogenes-3	*mefE* efflux	0.25	0.06	0.12	0.06	0.03	0.015	≦0.008	0.015
Haemophilus influenzae	Susceptible	32	4	32	8	8	4	8	4
H. influenzae-2	Susceptible	64	4	32	8	8	2	4	2
H. influenzae-3	Susceptible	128	16	32	16	16	16	8	8
H. influenzae-4	⊿acr	2	0.25	1	0.12	0.25	0.06	0.12	0.06

Abbreviations: LCM, lincomycin; NT, not tested.
[a]All strains except standard organisms were clinically isolated.
[b](c), constitutive; (i), inducible.
[c]Previously reported.

1.16–1.32 (m, 6H), 1.44, 1.47 (s x 2, 9H), 1.51–1.66 (m, 1H), 1.70–1.86 (m, 1H), 2.05, 2.06 (s x 2, 3H), 2.07–2.15 (m, 1H), 2.39–2.57 (m, 1H), 2.90 (t, J = 10.1 Hz, 1H), 3.55–3.71 (m, 1H), 3.72–3.88 (m, 1H), 3.98–4.19 (m, 2H), 4.23 (d, J = 8.8 Hz, 1H), 4.32–4.51 (m, 1H) and 5.23 (d, J = 5.4 Hz, 1H).

1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-4′-depropyl-4′-*n*-pentyllincomycin (21)

Compound **14** (1.16 g, 4.05 mmol), 1-hydroxybenzotriazole (820 mg, 6.07 mmol), *N*,*N*′-dicyclohexylcarbodiimide (1.25 g, 6.07 mmol) and MTL (1.54 g, 6.07 mmol) in DMF (15.0 ml) were treated for 23 h according to the similar procedure as described for the preparation of **18** to afford **21**. The total amount of this compound was used without purification to synthesize **26**.

1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-4′-depropyl-4′-{(*E*)-pent-2-enyl}lincomycin (22)

Compound **15** (1.55 g, 5.47 mmol), 1-hydroxybenzotriazole (1.11 g, 8.21 mmol), *N*,*N*′-dicyclohexylcarbodiimide (1.69 g, 8.21 mmol) and MTL (2.08 mg, 8.21 mmol) in DMF (23 ml) were treated for 3 h according to the similar procedure as described for the preparation of **18** to afford **22**. The total amount of this compound was used without purification to synthesize **27**.

1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-2,3,4-tris-*O*-(trimethylsilyl)lincomycin (23)

To a solution of compound **18** (crude) in pyridine (15 ml) were added trimethylchlorosilane (1.85 ml, 14.5 mmol) and hexamethyldisilazane (3.03 ml, 14.5 mmol) and stirred at room temperature for 30 min, and then the solution was added to the saturated aqueous $NaHCO_3$. The desired compound was extracted with ethyl acetate, washed with H_2O and then the organic phase was dried over Na_2SO_4, filtrated and concentrated under reduced pressure. To the resulting residue were added methanol (16.4 ml) and 6 N acetic acid (0.87 ml), and stirred at room temperature for 11 h. The mixture was added to the saturated aqueous $NaHCO_3$ and concentrated under reduced pressure to remove MeOH. The desired compound was extracted with ethyl acetate, and then the organic phase was dried over Na_2SO_4, filtrated and concentrated under reduced pressure. The resulting residue was purified by silica gel column chromatography (hexane/ethyl acetate = 2/1 to 1/2) to obtain the title compound (1.45 g, 70.6% in three steps from **7**) as a colorless solid. ESI-MS *m*/*z* 709 $(M+H)^+$ as $C_{31}H_{64}N_2O_8SSi_3$; ^{1}H NMR (400 MHz, CD_3OD) δ 0.12–0.26 (m, 27H), 0.85–1.00 (m, 3H), 1.07–1.25 (m, 3H), 1.26–1.41 (m, 4H), 1.44, 1.46 (s x 2, 9H), 1.66–1.93 (m, 1H), 2.03, 2.05 (s x 2, 3H), 1.98–2.42 (m, 2H), 2.88–3.02 (m, 1H), 3.54–4.40 (m, 8H) and 5.18 (d, J = 5.4 Hz, 1H).

1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-4′-depropyl-4′-(3-methoxypropyl)-2,3,4-tris-*O*-(trimethylsilyl)lincomycin (24)

Compound **19** (290 mg, 0.56 mmol), trimethylchlorosilane (355 μl, 2.77 mmol) and hexamethyldisilazane (581 μl, 2.77 mmol) in pyridine (1.0 ml) were treated for 1.0 h according to the similar procedure as described for the preparation of **23**, and then the crude compound and 6 N acetic acid (167 μl) in methanol (3.1 ml) were treated for 30 min according to the similar procedure as described for the preparation of **23** to afford **24** (282 mg, 68.7% in two steps from **19**) as a colorless solid. ESI-MS *m*/*z* 739 $(M+H)^+$ as $C_{32}H_{66}N_2O_9SSi_3$.

1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-4′-depropyl-4′-*i*-butyl-2,3,4-tris-*O*-(trimethylsilyl)lincomycin (25)

Compound **20** (652 mg, 1.63 mmol), trimethylchlorosilane (1.02 ml, 8.02 mmol) and hexamethyldisilazane (1.68 ml, 8.02 mmol) in pyridine (3.5 ml) were treated for 1 h according to the similar procedure as described for the preparation of **23**, and then the crude compound and 6 N acetic acid (480 µl) in methanol (9 ml) were treated for 30 min according to the similar procedure as described for the preparation of **23** to afford **25** (946 mg, 79.8% in two steps from **20**) as a colorless solid. FAB-MS *m/z* 723 $(M+H)^+$ as $C_{32}H_{66}N_2O_8SSi_3$; ^{1}H NMR (400 MHz, $CDCl_3$) δ 0.11–0.21 (m, 27H), 0.90 (d, *J*=6.6 Hz, 6H), 1.05–1.33 (m, 5H), 1.49 (s, 9H), 1.50–1.61 (m, 2H), 2.07 (s, 3H), 2.13–2.57 (m, 1H), 2.72–3.13 (m, 1H), 3.40–3.82 (m, 3H), 3.94–4.19 (m, 3H), 4.22–4.40 (m, 2H) and 5.19 (d, *J*=5.6 Hz, 1H).

1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-4′-depropyl-4′-*n*-pentyl-2,3,4-tris-*O*-(trimethylsilyl)lincomycin (26)

Compound **21** (crude), trimethylchlorosilane (2.6 ml, 20.3 mmol) and hexamethyldisilazane (4.24 ml, 20.3 mmol) in pyridine (10.0 ml) were treated for 1 h according to the similar procedure as described for the preparation of **23**, and then the crude compound and 6 N acetic acid (1.21 ml) in methanol (23 ml) were treated for 2.0 h according to the similar procedure as described for the preparation of **23** to afford **26** (2.21 g, 74% in three steps from **14**) as a colorless solid. FAB-MS *m/z* 737 $(M+H)^+$ as $C_{33}H_{68}N_2O_8SSi_3$; ^{1}H NMR (400 MHz, $CDCl_3$) δ 0.07–0.25 (m, 27H), 0.78–0.97 (m, 3H), 1.05–1.42 (m, 11H), 1.48 (s, 9H), 2.07 (s, 3H), 2.10–3.20 (m, 4H), 3.40–3.90 (m, 3H), 3.92–4.20 (m, 3H), 4.23–4.47 (m, 2H) and 5.19 (br d, *J*=5.4 Hz, 1H).

1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-4′-depropyl-4′-{(*E*)-pent-2-enyl}-2,3,4-tris-*O*-(trimethylsilyl)lincomycin (27)

Compound **22** (crude), trimethylchlorosilane (3.50 ml, 27.4 mmol) and hexamethyldisilazane (5.70 ml, 27.4 mmol) in pyridine (10 ml) were treated for 1 h according to the similar procedure as described for the preparation of **23** and then, the crude compound and 6 N acetic acid (1.64 ml) in methanol (31 ml) were treated for 1.0 h according to the similar procedure as described for the preparation of **23** to afford **27** (3.29 g, 81.8% in three steps from **15**) as a colorless solid. ^{1}H NMR (400 MHz, $CDCl_3$) δ 0.02–0.28 (m, 27H), 0.96 (t, *J*=7.4 Hz, 3H), 1.08–1.25 (m, 3H), 1.48 (s, 9H), 1.89–2.51 (m, 6H), 2.02 (s, 3H), 2.66–3.26 (m, 2H), 3.40–3.64 (m, 2H), 3.68–4.19 (m, 4H), 4.22–4.50 (m, 2H), 5.19 (br d, *J*=5.1 Hz, 1H), 5.26–5.39 (m, 1H) and 5.43–5.59 (m, 1H).

7(*S*)-1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-7-deoxy-7-[5-{5-(methylamino)thiazol-4-yl}-1,3,4-thiadiazol-2-ylthio]-2,3,4-tris-*O*-(trimethylsilyl)lincomycin (31)

To a solution of compound **23** (200 mg, 0.28 mmol) in THF (2 ml) at 0 °C were added triphenylphosphine (110.9 mg, 0.42 mmol), diethylazodicarboxylate (77 µl, 0.42 mmol), 5-{5-(methylamino)thiazol-4-yl}-1,3,4-thiadiazole-2-thiol (100.7 mg, 0.44 mmol) and stirred at room temperature for 18 h. The solution was purified by preparative TLC (hexane/ethyl acetate=2/1) to obtain the title compound as an off-white solid (88.8 mg, 34.2%). FAB-MS *m/z* 921 $(M+H)^+$ as $C_{37}H_{68}N_6O_7S_4Si_3$; ^{1}H NMR (400 MHz, $CDCl_3$) δ 0.00–0.25 (m, 27H), 0.80–1.00 (m, 3H), 1.08–1.67 (m, 16H), 1.74–2.99 (m, 7H), 3.02–3.22 (m, 3H), 3.43–3.90 (m, 3H), 3.98–4.50 (m, 4H), 4.60–4.94 (m, 1H), 5.20 (br d, *J*=5.4 Hz, 1H) and 7.85–8.00 (br s, 1H).

7(*S*)-7-(6-aminobenzothiazol-2-ylthio)-7-deoxy-1′-demethyllincomycin (32)

To a solution of compound **23** (200 mg, 0.28 mmol) in THF (2 ml) at 0 °C were added triphenylphosphine (110.9 mg, 0.42 mmol), diethylazodicarboxylate (77 µl, 0.42 mmol), 6-aminobenzothiazole-2-thiol (79.7 mg, 0.44 mmol) and stirred at room temperature for 4 h. To the solution was added 1 N HCl (1 ml)–MeOH (1 ml) and stirred at room temperature for 30 min. The solution was added to the saturated aqueous $NaHCO_3$. The desired compound was extracted with ethyl acetate, and then the organic phase was dried over Na_2SO_4, filtrated and concentrated under reduced pressure. The resulting residue was purified by preparative TLC ($CHCl_3$/MeOH/28% aq. NH_4OH=9/2/0.2) to obtain 7(*S*)-7-(6-aminobenzothiazol-2-yl)thio-1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-7-deoxy-2,3,4-tris-*O*-(trimethylsilyl)lincomycin (197.2 mg, crude).

To the solution of this intermediate in MeOH (2 ml) was added 4 N HCl-ethyl acetate (2.5 ml) and stirred at room temperature for 2 h. The solution was concentrated under reduced pressure. The resulting residue was purified by silica gel column chromatography ($CHCl_3$/MeOH/28% aq. NH_4OH=9/2/0.2) to obtain the title compound (96.4 mg, 61.4% in three steps from **23**) as an off-white solid. $[\alpha]_D^{26}$+92.1° (*c* 2.49, MeOH); ESI-MS *m/z* 557 $(M+H)^+$ as $C_{24}H_{36}N_4O_5S_3$; TOF-ESI-HRMS $(M+H)^+$ calcd. for $C_{24}H_{36}N_4O_5S_3$: 557.1926, found: 557.1920; ^{1}H NMR (400 MHz, CD_3OD) δ 0.86–0.96 (m, 3H), 1.25–1.40 (m, 4H), 1.49 (d, *J*=6.9 Hz, 3H), 1.69–1.82 (m, 1H), 1.93 (s, 3H), 1.96–2.13 (m, 2H), 2.52 (dd, *J*=10.4, 8.1 Hz, 1H), 3.20 (dd, *J*=10.4, 6.9 Hz, 1H), 3.58 (dd, *J*=10.2, 3.2 Hz, 1H), 3.80–3.87 (m, 2H), 4.11 (dd, *J*=10.2, 5.6 Hz, 1H), 4.27 (dq, *J*=6.9, 2.7 Hz, 1H), 4.39 (br dd, *J*=10.0, 0.9 Hz, 1H), 4.57 (dd, *J*=10.0, 2.7 Hz, 1H), 5.26 (d, *J*=5.6 Hz, 1H), 6.85 (dd, *J*=8.7, 2.1 Hz, 1H), 7.08 (d, *J*=2.1 Hz, 1H) and 7.59 (d, *J*=8.7 Hz, 1H).

7(*S*)-7-(5-amino-1,3,4-thiadiazol-2-ylthio)-7-deoxy-1′-demethyllincomycin (33)

To a solution of compound **23** (200 mg, 0.28 mmol) in THF (2 ml) at 0 °C were added triphenylphosphine (138.0 mg, 0.53 mmol), diethylazodicarboxylate (96 µl, 0.53 mmol), 5-(*tert*-butoxycarbonylamino)-1,3,4-thiadiazole-2-thiol (126.9 mg, 0.54 mmol) and stirred at room temperature for 3 h. To the solution was added 1 N HCl (1 ml)–MeOH (1 ml), and stirred at room temperature for 50 min. The solution was added to the saturated aqueous $NaHCO_3$. The desired compound was extracted with ethyl acetate, and then the organic phase was dried over Na_2SO_4, filtrated and concentrated under reduced pressure. The resulting residue was purified by preparative TLC ($CHCl_3$/MeOH/28% aq. NH_4OH=9/2/0.2) to obtain 7(*S*)-7-[5-{(*tert*-butoxycarbonyl)amino}-1,3,4-thiadiazol-2-ylthio]-1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-7-deoxylincomycin as a colorless solid (146.6 mg, 73.4%). ^{1}H NMR (400 MHz, CD_3OD) δ 0.85–0.98 (m, 3H), 1.25–1.60 (m, 25 H), 1.80–1.97 (m, 1H), 1.98–2.17 (m, 1H), 2.06, 2.10 (s x 2, 3H), 2.22–2.40 (m, 1H), 2.89–3.05 (m, 1H), 3.50–3.60 (m, 1H), 3.69–3.82 (m, 1H), 3.84–4.00 (m, 1H), 4.02–4.20 (m, 2H), 4.26–4.42 (m, 2H), 4.45–4.60 (m, 1H) and 5.27 (br d, *J*=5.4 Hz, 1H).

To the solution of this intermediate (146.6 mg, 0.21 mmol) in MeOH (1.4 ml) was added 4 N HCl-ethyl acetate (1.7 ml), and stirred at room temperature for 2 h. The solution was concentrated under reduced pressure. The resulting residue was purified by preparative TLC ($CHCl_3$/MeOH/28% aq. NH_4OH=9/2/0.2) to obtain the title compound (25.4 mg, 24.2%) as a colorless solid. $[\alpha]_D^{25}$ +101° (*c* 0.33, MeOH); ESI-MS *m/z* 508 $(M+H)^+$ as $C_{19}H_{33}N_5O_5S_3$; TOF-ESI-HRMS $(M+H)^+$ calcd. for $C_{19}H_{33}N_5O_5S_3$: 508.1722, found: 508.1719; ^{1}H NMR (400 MHz, CD_3OD) δ 0.89–0.98 (m, 3H), 1.33–1.45 (m, 4H), 1.37 (d, *J*=7.0 Hz, 3H), 1.84–1.95 (m, 1H), 2.03–2.18 (m, 2H), 2.11 (s, 3H), 2.66 (dd, *J*=10.6, 8.3 Hz, 1H), 3.30–3.36 (m, 1H), 3.56 (dd, *J*=10.2, 3.2 Hz, 1H), 3.81 (br dd, *J*=3.2, 0.7 Hz, 1H), 3.96 (dq, *J*=7.0, 2.6 Hz, 1H), 4.00 (dd, *J*=9.3, 4.0 Hz, 1H), 4.09 (dd, *J*=10.2, 5.6 Hz, 1H), 4.37 (br dd, *J*=10.0, 0.7 Hz, 1H), 4.52 (dd, *J*=10.0, 2.6 Hz, 1H) and 5.27 (d, *J*=5.6 Hz, 1H).

7(*S*)-1′-demethyl-7-deoxy-7-{5-(2-nitrophenyl)-1,3,4-thiadiazol-2-ylthio}lincomycin (34)

To a solution of compound **23** (200 mg, 0.28 mmol) in THF (2 ml) at 0 °C were added triphenylphosphine (110.9 mg, 0.42 mmol), diethylazodicarboxylate (77 µl, 0.42 mmol), 5-(2-nitrophenyl)-1,3,4-thiadiazole-2-thiol (104.6 mg, 0.44 mmol), and stirred at room temperature for 7 h. The solution was added to the saturated aqueous $NaHCO_3$. The desired compound was extracted with ethyl acetate, and then the organic phase was dried over Na_2SO_4, filtrated and concentrated under reduced pressure. To the resulting residue was added MeOH (4 ml)–1N HCl (1 ml) and stirred at room temperature for 2.5 h. The solution was added to the saturated aqueous $NaHCO_3$. The desired compound was extracted with ethyl acetate, and then the organic phase was dried over Na_2SO_4, filtrated and concentrated under reduced pressure. The resulting residue was purified by preparative TLC ($CHCl_3$/MeOH/28% aq. NH_4OH=9/2/0.2) to obtain 7(*S*)-1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-7-deoxy-7-{5-(2-nitrophenyl)-1,3,4-thiadiazol-2-ylthio}lincomycin (164.9 mg as crude). To

the solution of this crude compound (63.9 mg) in MeOH (0.6 ml) was added 4 N HCl-ethyl acetate (0.75 ml), and stirred at room temperature for 2.5 h. The solution was concentrated under reduced pressure. The resulting residue was purified by preparative TLC ($CHCl_3$/MeOH/28% aq. NH_4OH = 9/2/0.2) to obtain the title compound (13.0 mg, 19.4% in three steps from **23**) as a colorless solid. $[\alpha]_D^{26}$ +37.1° (*c* 0.21, MeOH); ESI-MS *m/z* 614 $(M+H)^+$ as $C_{25}H_{35}N_5O_7S_3$; TOF-ESI-HRMS $(M+H)^+$ calcd. for $C_{25}H_{35}N_5O_7S_3$: 614.1777, found: 614.1778; ^1H NMR (400 MHz, CD_3OD) δ 0.83–0.99 (m, 3H), 1.30–1.45 (m, 4H), 1.56 (d, *J* = 7.0 Hz, 3H), 1.88–1.98 (m, 1H), 1.99 (s, 3H), 2.07–2.25 (m, 2H), 2.69 (br dd, *J* = 10.6, 8.4 Hz, 1H), 3.32–3.42 (m, 1H), 3.56 (dd, *J* = 10.2, 3.2 Hz, 1H), 3.82–3.88 (m, 1H), 4.00–4.10 (m, 1H), 4.10 (dd, *J* = 10.2, 5.6 Hz, 1H), 4.36–4.46 (m, 1H), 4.47 (dq, *J* = 7.0, 2.5 Hz, 1H), 4.66 (dd, *J* = 10.0, 2.5 Hz, 1H), 5.28 (d, *J* = 5.6 Hz, 1H), 7.74–7.87 (m, 3H) and 8.05–8.15 (m, 1H).

7(*S*)-1′-demethyl-7-deoxy-7-[5-{5-(methylamino)thiazol-4-yl}-1,3,4-thiadiazol-2-ylthio]lincomycin (35)

To the solution of compound **31** (88.8 mg 0.096 mmol) in MeOH (0.5 ml) was added 4 N HCl-ethyl acetate (0.79 ml), stirred at 0 °C for 1 h and then stirred at room temperature for 3.5 h. The solution was concentrated under reduced pressure. The resulting residue was purified by preparative TLC ($CHCl_3$/MeOH/28% aq. NH_4OH = 9/2/0.2) to obtain the title compound (39.8 mg, 68.3%) as an off-white solid. $[\alpha]_D^{26}$ +69.5° (*c* 0.60, MeOH); ESI-MS *m/z* 605 $(M+H)^+$ as $C_{23}H_{36}N_6O_5S_4$; TOF-ESI-HRMS $(M+H)^+$ calcd. for $C_{23}H_{36}N_6O_5S_4$: 605.1708, found: 605.1706; ^1H NMR (400 MHz, CD_3OD) δ 0.86–0.95 (m, 3H), 1.29–1.41 (m, 4H), 1.49 (d, *J* = 6.9 Hz, 3H), 1.78–1.87 (m, 1H), 1.98–2.14 (m, 2H), 2.01 (s, 3H), 2.56 (dd, *J* = 10.5, 8.1 Hz, 1H), 3.11 (s, 3H), 3.25 (dd, *J* = 10.5, 7.0 Hz, 1H), 3.56 (dd, *J* = 10.3, 3.2 Hz, 1H), 3.82 (br dd, *J* = 3.2, 0.9 Hz, 1H), 3.86 (dd, *J* = 9.2, 3.9 Hz, 1H), 4.09 (dd, *J* = 10.3, 5.6 Hz, 1H), 4.27 (dq, *J* = 6.9, 2.7 Hz, 1H), 4.39 (br dd, *J* = 10.0, 0.9 Hz, 1H), 4.59 (dd, *J* = 10.0, 2.7 Hz, 1H), 5.26 (d, *J* = 5.6 Hz, 1H) and 8.12 (s, 1H).

7(*S*)-7-deoxy-7-[5-{5-(methylamino)thiazol-4-yl}-1,3,4-thiadiazol-2-ylthio]lincomycin (37)

Compound **36** (240 mg, 0.39 mmol), triphenylphosphine (150.0 mg, 0.57 mmol), diethylazodicarboxylate (100 µl, 0.64 mmol) and 5-{5-(methylamino)thiazol-4-yl}-1,3,4-thiadiazole-2-thiol (150.0 mg, 0.65 mmol) in THF (5 ml) were treated for 2 h, and then to the solution was added 1N HCl (0.5 ml)–MeOH (5 ml) and stirred at room temperature for 1 h. The solution was then added to the saturated aqueous $NaHCO_3$. The desired compound was extracted with ethyl acetate, and then the organic phase was dried over Na_2SO_4, filtrated and concentrated under reduced pressure. The resulting residue was purified by preparative TLC ($CHCl_3$/MeOH/28% aq. NH_4OH = 9/2/0.2) to obtain the title compound **37** (88.2 mg, 37% in two steps) as an off-white solid. $[\alpha]_D^{26}$ +125° (*c* 0.89, MeOH); ESI-MS *m/z* 619 $(M+H)^+$ as $C_{24}H_{38}N_6O_5S_4$; TOF-ESI-HRMS $(M+H)^+$ calcd. for $C_{24}H_{38}N_6O_5S_4$: 619.1865, found: 619.1860; ^1H NMR (400 MHz, CD_3OD) δ 0.82–0.98 (m, 3H), 1.21–1.39 (m, 4H), 1.53 (d, *J* = 6.9 Hz, 3H), 1.78–1.89 (m, 1H), 1.92–2.08 (m, 2H), 2.02 (s, 3H), 2.09–2.25 (m, 1H), 2.35 (s, 3H), 3.00 (dd, *J* = 10.5, 5.0 Hz, 1H), 3.13 (s, 3H), 3.19 (dd, *J* = 8.5, 6.2 Hz, 1H), 3.59 (dd, *J* = 10.2, 3.2 Hz, 1H), 3.77–3.85 (m, 1H), 4.11 (dd, *J* = 10.2, 5.6 Hz, 1H), 4.27 (dq, *J* = 6.9, 3.0 Hz, 1H), 4.44 (br dd, *J* = 9.8, 0.5 Hz, 1H), 4.57 (dd, *J* = 9.8, 3.0 Hz, 1H), 5.27 (d, *J* = 5.6 Hz, 1H) and 8.13 (s, 1H).

7(*S*)-1′-demethyl-7-deoxy-7-{5-(2-nitrophenyl)-1,3,4-thiadiazol-2-yl}thio-1′-*N*-*i*-propyllincomycin (38)

To a solution of compound **34** (30.5 mg, 0.05 mmol) in 1,2-dichloroethane (1 ml) at 0 °C were added acetone (40 µl, 0.054 mmol), AcOH (one drop) and $NaBH(OAc)_3$ (21.7 mg, 0.10 mmol) and stirred at room temperature for 15 h. The mixture was concentrated under reduced pressure. The resulting residue was purified by preparative TLC ($CHCl_3$/MeOH/28% aq NH_4OH = 10/1/0.1) and then by LH-20 ($CHCl_3$/MeOH = 1/1) to obtain the title compound (20.1 mg, 61%) as a colorless solid. $[\alpha]_D^{26}$ +76.8° (*c* 0.59, MeOH); ESI-MS *m/z* 656 $(M+H)^+$ as $C_{28}H_{41}N_5O_7S_3$; TOF-ESI-HRMS $(M+H)^+$ calcd. for $C_{28}H_{41}N_5O_7S_3$: 656.2246, found: 656.2243; ^1H NMR (400 MHz, CD_3OD) δ 0.86–0.98 (m, 3H), 1.06–1.16 (m, 6H), 1.28–1.42 (m, 4H), 1.58 (d, *J* = 6.9 Hz, 3H), 1.69–1.81 (m, 1H), 1.99 (s, 3H), 1.97–2.07 (m, 1H), 2.07–2.20 (m, 1H), 2.20–2.31 (m, 1H), 2.76–2.90 (m, 1H), 3.26–3.34 (m, 1H), 3.39–3.52 (m, 1H), 3.57 (dd, *J* = 10.2, 3.2 Hz, 1H), 3.82 (br dd, *J* = 3.2, 0.8 Hz, 1H), 4.10 (dd, *J* = 10.2, 5.6 Hz, 1H), 4.40 (br dd, *J* = 9.3, 0.8 Hz, 1H), 4.51 (dq, *J* = 6.9, 3.4 Hz, 1H), 4.59 (dd, *J* = 9.3, 3.4 Hz, 1H), 5.26 (d, *J* = 5.6 Hz, 1H), 7.74–7.86 (m, 3H) and 8.06–8.12 (m, 1H).

7(*S*)-1′-*N*-{2-(*tert*-butyldimethylsilyloxy)ethyl}-1′-demethyl-7-deoxy-7-{5-(2-nitrophenyl)-1,3,4-thiadiazol-2-ylthio}lincomycin (39)

Compound **34** (49.2 mg, 0.08 mmol), 2-(*tert*-butyldimethylsilyloxy)acetaldehyde (23 µl, 0.12 mmol), AcOH (one drop) and $NaBH(OAc)_3$ (34.2 mg, 0.16 mmol) in 1,2-dichloroethane (1 ml) were treated at 0 °C for 15 h according to the similar procedure as described for the preparation of **38** to afford **39** (26.9 mg, 44.0%) as a colorless solid. FAB-MS *m/z* 772 $(M+H)^+$ as $C_{33}H_{53}N_5O_8S_3Si$; ^1H NMR (400 MHz, CD_3OD) δ 0.07 (s, 3H), 0.08 (s, 3H), 0.82–0.98 (m, 12H), 1.25–1.42 (m, 4H), 1.59 (d, *J* = 6.8 Hz, 1H), 1.68–1.87 (m, 1H), 1.94–2.05 (m, 1H), 1.97 (s, 3H), 2.07–2.21 (m, 2H), 2.54–2.92 (m, 2H), 3.37–3.44 (m, 1H), 3.56 (dd, *J* = 10.2, 3.2 Hz, 1H), 3.71–3.92 (m, 4H), 4.10 (dd, *J* = 10.2, 5.6 Hz, 1H), 4.35–4.44 (m, 1H), 4.47–4.56 (m, 1H), 4.58–4.65 (m, 1H), 5.26 (d, *J* = 5.6 Hz, 1H), 7.74–7.86 (m, 3H) and 8.06–8.13 (m, 1H).

7(*S*)-1′-demethyl-7-deoxy-1′-*N*-(2-hydroxyethyl)-7-{5-(2-nitrophenyl)-1,3,4-thiadiazol-2-ylthio}lincomycin (40)

To a solution of compound **39** (26.9 mg, 0.035 mmol) in THF (0.5 ml) at 0 °C were added 1M THF solution of *tetra-n*-butyl ammonium fluoride (100 µl, 0.10 mmol) and stirred at room temperature for 15 h. The mixture was concentrated under reduced pressure. The resulting residue was purified by preparative TLC ($CHCl_3$/MeOH/28% aq NH_4OH = 10/1/0.1) to obtain the title compound (16.5 mg, 72%) as a colorless solid. $[\alpha]_D^{25}$ +70.5° (*c* 0.22, MeOH); ESI-MS *m/z* 658 $(M+H)^+$ as $C_{27}H_{39}N_5O_8S_3$; TOF-ESI-HRMS $(M+H)^+$ calcd. for $C_{27}H_{39}N_5O_8S_3$: 658.2039, found: 658.2044; ^1H NMR (400 MHz, CD_3OD) δ 0.87–0.96 (m, 3H), 1.29–1.43 (m, 4H), 1.60 (d, *J* = 6.9 Hz, 3H), 1.79–1.93 (m, 1H), 1.99 (s, 3H), 2.02–2.10 (m, 1H), 2.10–2.23 (m, 2H), 2.64–2.75 (m, 1H), 2.83–2.96 (m, 1H), 3.31–3.40 (m, 1H), 3.40–3.50 (m, 1H), 3.56 (dd, *J* = 10.3, 3.2 Hz, 1H), 3.63–3.77 (m, 2H), 3.83 (dd, *J* = 3.2, 0.8 Hz, 1H), 4.10 (dd, *J* = 10.3, 5.6 Hz, 1H), 4.45 (br dd, *J* = 9.9, 0.8 Hz, 1H), 4.51 (dq, *J* = 6.9, 2.9 Hz, 1H), 4.63 (dd, *J* = 9.9, 2.9 Hz, 1H), 5.26 (d, *J* = 5.6 Hz, 1H), 7.74–7.86 (m, 3H) and 8.06–8.12 (m, 1H).

7(*S*)-1′-*N*-{2-(*tert*-butyldimethylsilyloxy)ethyl}-1′-demethyl-7-deoxy-7-[5-{5-(methylamino)thiazol-4-yl}-1,3,4-thiadiazol-2-ylthio]lincomycin (41)

Compound **35** (74.3 mg, 0.12 mmol), 2-(*tert*-butyldimethylsilyloxy)acetaldehyde (34 µl, 0.18 mmol), AcOH (one drop) and $NaBH(OAc)_3$ (51.0 mg, 0.24 mmol) in 1,2-dichloroethane (1 ml) were treated at room temperature according to the similar procedure as described for the preparation of **38** to afford **41** (54.8 mg, 60%) as a colorless solid. FAB-MS *m/z* 763 $(M+H)^+$ as $C_{31}H_{54}N_6O_6S_4Si$; ^1H NMR (400 MHz, CD_3OD) δ 0.05 (s, 3H), 0.06 (s, 3H), 0.78–1.00 (m, 12H), 1.23–1.42 (m, 4H), 1.54 (d, *J* = 7.1 Hz, 3H), 1.72–1.86 (m, 1H), 1.92–2.05 (m, 1H), 1.99 (s, 3H), 2.08–2.22 (m, 2H), 2.50–2.87 (m, 2H), 3.12 (s, 3H), 3.25–3.30 (m, 1H), 3.36–3.44 (m, 1H), 3.52–3.60 (m, 1H), 3.68–3.88 (m, 3H), 4.10 (dd, *J* = 10.2, 5.6 Hz, 1H), 4.30–4.44 (m, 2H), 4.54–4.62 (m, 1H), 5.26 (d, *J* = 5.6 Hz, 1H) and 8.13 (s, 1H).

7(*S*)-1′-demethyl-7-deoxy-1′-*N*-(2-hydroxylethyl)-7-[5-{5-(methylamino)thiazol-4-yl}-1,3,4-thiadiazol-2-ylthio]lincomycin (42)

Compound **41** (54.8 mg, 0.072 mmol) and 1 M THF solution of *tetra-n*-butyl ammonium fluoride (200 µl, 0.20 mmol) in THF (1.0 ml) were treated at 0 °C for 1 h and then treated at room temperature for 5 h according to the similar procedure as described for the preparation of **40** to afford **42** (31.3 mg, 67.0%) as a colorless solid. $[\alpha]_D^{25}$ +51.1° (*c* 0.27, MeOH); ESI-MS *m/z* 649 $(M+H)^+$ as $C_{25}H_{40}N_6O_6S_4$; TOF-ESI-HRMS $(M+H)^+$ calcd. for $C_{25}H_{40}N_6O_6S_4$: 649.1970, found: 649.1973; ^1H NMR (400 MHz, CD_3OD) δ 0.88–0.96 (m, 3H), 1.30–1.40

(m, 4H), 1.54 (d, $J=6.9$ Hz, 3H), 1.79–1.90 (m, 1H), 1.97–2.08 (m, 1H), 2.00 (s, 3H), 2.09–2.21 (m, 2H), 2.62–2.72 (m, 1H), 2.83–2.93 (m, 1H), 3.11 (s, 3H), 3.30–3.37 (m, 1H), 3.39–3.45 (m, 1H), 3.56 (dd, $J=10.2$, 3.2 Hz, 1H), 3.65–3.74 (m, 2H), 3.81 (br dd, $J=3.2$, 0.8 Hz, 1H), 4.09 (dd, $J=10.2$, 5.6 Hz, 1H), 4.33 (dq, $J=6.9$, 2.8 Hz, 1H), 4.43 (br dd, $J=9.8$, 0.8 Hz, 1H), 4.59 (dd, $J=9.8$, 2.8 Hz, 1H), 5.25 (d, $J=5.6$ Hz, 1H) and 8.11 (s, 1H).

7(*S*)-1′-demethyl-7-deoxy-1′-*N*-{2(*R*)-hydroxypropyl}-7-[5-{5-(methylamino)thiazol-4-yl}-1,3,4-thiadiazol-2-ylthio]lincomycin (43)

To a solution of compound **35** (28.7 mg, 0.048 mmol) and *N,N*-diisopropylethylamine (10.0 µl, 0.057 mmol) in MeOH (1 ml) at 0 °C was added (*R*)-2-methyloxirane (0.30 ml, 4.3 mmol) and stirred at 0 °C for 96 h. The mixture was concentrated under reduced pressure. The resulting residue was purified by preparative TLC ($CHCl_3$/MeOH/28% aq. $NH_4OH=10/1/0.1$) to obtain the title compound (10.6 mg, 33%) as a colorless solid. $[\alpha]_D^{26}$ +29.2° (*c* 0.15, MeOH); ESI-MS *m/z* 663 $(M+H)^+$ as $C_{26}H_{42}N_6O_6S_4$; TOF-ESI-HRMS $(M+H)^+$ calcd. for $C_{26}H_{42}N_6O_6S_4$: 663.2127, found: 663.2127; ^{1}H NMR (400 MHz, CD_3OD) δ 0.87–0.96 (m, 3H), 1.16 (d, $J=6.2$ Hz, 3H), 1.27–1.42 (m, 4H), 1.55 (d, $J=6.9$ Hz, 1H), 1.78–1.87 (m, 1H), 1.98–2.27 (m, 3H), 1.99 (s, 3H), 2.43–2.55 (m, 1H), 2.56–2.66 (m, 1H), 3.11 (s, 3H), 3.39–3.46 (m, 1H), 3.55 (dd, $J=10.3$, 3.2 Hz, 1H), 3.83 (br dd, $J=3.2$, 0.9 Hz, 1H), 3.86–3.97 (m, 1H), 4.10 (dd, $J=10.3$, 5.7 Hz, 1H), 4.33 (dq, $J=6.9$, 2.9 Hz, 1H), 4.42–4.48 (m, 1H), 4.52–4.59 (m, 1H), 4.61 (dd, $J=9.9$, 2.9 Hz, 1H), 5.25 (d, $J=5.7$ Hz, 1H) and 8.11 (s, 1H).

7(*S*)-1′-*N*-acetyl-1′-demethyl-7-deoxy-7-[5-{5-(methylamino) thiazol-4-yl}-1,3,4-thiadiazol-2-ylthio]lincomycin (44)

To a solution of compound **35** (30.2 mg, 0.050 mmol) in MeOH (0.5 ml) at 0 °C was added acetic anhydride (7.0 µl, 0.074 mmol) and stirred at 0 °C for 1.5 h. The mixture was concentrated under reduced pressure. The resulting residue was purified by preparative TLC ($CHCl_3$/MeOH/28% aq. $NH_4OH=10/1/0.1$) to obtain the title compound (10.8 mg, 33%) as an off-white solid. $[\alpha]_D^{25}$ +56.5° (*c* 1.25, MeOH); ESI-MS *m/z* 647 $(M+H)^+$ as $C_{25}H_{38}N_6O_6S_4$; TOF-ESI-HRMS $(M+H)^+$ calcd. for $C_{25}H_{38}N_6O_6S_4$: 647.1814, found: 647.1807; ^{1}H NMR (400 MHz, CD_3OD) δ 0.87–0.98 (m, 3H), 1.25–1.42 (m, 4H), 1.49 (d, $J=7.0$ Hz, 3H), 1.82–1.95 (m, 1H), 1.98 (s, 3H), 2.01–2.08 (m, 1H), 2.09 (s, 3H), 2.35–2.50 (m, 1H), 3.12 (s, 3H), 3.14–3.20 (m, 1H), 3.56 (dd, $J=10.2$, 3.3 Hz, 1H), 3.79–3.87 (m, 1H), 3.99 (br dd, $J=3.3$, 1.0 Hz, 1H), 4.09 (dd, $J=10.2$, 5.6 Hz, 1H), 4.27 (dq, $J=7.0$, 2.4 Hz, 1H), 4.36–4.43 (m, 1H), 4.48 (dd, $J=8.8$, 2.7 Hz, 1H), 4.61 (dd, $J=10.0$, 2.4 Hz, 1H) and 8.12 (s, 1H).

7(*S*)-1′-demethyl-7-deoxy-7-[5-{5-(methylamino)thiazol-4-yl}-1,3,4-thiadiazol-2-ylthio]-1′-*N*-(4-methylthiazol-5-ylmethyl)lincomycin (45)

Compound **35** (24.0 mg, 0.04 mmol), 4-methylthiazole-5-carbaldehyde (16.2 mg, 0.13 mmol), AcOH (one drop) and $NaBH(OAc)_3$ (25.5 mg, 0.12 mmol) in MeOH (0.5 ml) were treated at room temperature for 15 h according to the similar procedure as described for the preparation of **38** to afford **45** (6.4 mg, 22.0%) as a colorless solid. $[\alpha]_D^{26}$ +26° (*c* 0.05, MeOH); ESI-MS *m/z* 716 $(M+H)^+$ as $C_{28}H_{41}N_7O_5S_5$; TOF-ESI-HRMS $(M+H)^+$ calcd. for $C_{28}H_{41}N_7O_5S_5$: 716.1851, found: 716.1854; ^{1}H NMR (400 MHz, CD_3OD) δ 0.75–0.86 (m, 3H), 1.15–1.26 (m, 4H), 1.42 (d, $J=6.8$ Hz, 3H), 1.68–1.78 (m, 1H), 1.89–2.15 (m, 3H), 1.94 (s, 3H), 2.27 (s, 3H), 3.00 (s, 3H), 3.09 (dd, $J=8.2$, 6.2 Hz, 1H), 3.24–3.31 (m, 1H), 3.50 (dd, $J=10.2$, 3.2 Hz, 1H), 3.72 (d, $J=14.3$ Hz, 1H), 3.79 (br dd, $J=3.2$, 1.0 Hz, 1H), 3.85 (d, $J=14.3$ Hz, 1H), 4.01 (dd, $J=10.2$, 5.5 Hz, 1H), 4.13–4.21 (m, 1H), 4.28–4.34 (m, 1H), 4.44 (dd, $J=8.8$, 4.6 Hz, 1H), 5.16 (d, $J=5.5$ Hz, 1H, 8.03 (s, 1H) and 8.69 (s, 1H).

1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-4′-propyllincomycin (46)

Compound **17** (1.55 g, 5.19 mmol), 1-hydroxybenzotriazole (1.05 g, 7.78 mmol), *N,N*′-dicyclohexylcarbodiimide (1.61 g, 7.78 mmol) and MTL (1.97 g, 7.78 mmol) in DMF (15.0 ml) were treated for 14 h according to the similar procedure as described for the preparation of **18** to afford **46**. The total amount of this compound was used without purification to synthesize **47**. For the qualified analytical purpose, the above crude **46** was purified by column chromatography (ethyl acetate only) to obtain the title compound as a colorless solid. ESI-MS *m/z* 535 $(M+H)^+$ as $C_{25}H_{46}N_2O_8S$; ^{1}H NMR (400 MHz, CD_3OD) δ 0.76–0.90 (m, 6H), 1.04–1.32 (m, 11H), 1.35, 1.36 (s x 2, 9H), 1.57–1.72 (m, 1H), 1.97, 1.99 (s x 2, 3H), 2.00–2.15 (m, 1H), 3.03 (br t, $J=10.4$ Hz, 1H), 3.29–3.56 (m, 2H), 3.67–3.90 (m, 1H), 3.91–4.36 (m, 5H) and 5.15 (br d, $J=5.4$ Hz, 1H).

1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-4′-propyl-2,3,4-tris-*O*-(trimethylsilyl)lincomycin (47)

Compound **46** (crude), trimethylchlorosilane (3.32 ml, 25.9 mmol) and hexamethyldisilazane (5.44 ml, 25.9 mmol) in pyridine (5.0 ml) were treated for 1 h according to the similar procedure as described for the preparation of **23** and then the crude compound and 6 N acetic acid (1.55 ml) in methanol (29.4 ml) were treated for 1 h according to the similar procedure as described for the preparation of **23** to afford **47** (1.66 g, 41.6% in three steps from **17**) as a colorless solid. ESI-MS *m/z* 751 $(M+H)^+$ as $C_{34}H_{70}N_2O_8SSi_3$; ^{1}H NMR (400 MHz, $CDCl_3$) δ 0.14 (s, 18H), 0.18 (s, 9H), 0.82–0.98 (m, 6H), 1.08–1.35 (m, 11H), 1.46 (s, 9H), 1.80–2.35 (m, 1H), 2.06 (s, 3H), 2.66–3.30 (m, 2H), 3.36–4.41 (m, 8H) and 5.13–5.24 (m, 1H).

7(*S*)-1′-demethyl-7-deoxy-4′-*n*-propyl-7-{5-(2-nitrophenyl)-1,3,4-thiadiazol-2-ylthio}lincomycin (48)

To a solution of compound **47** (400 mg, 0.53 mmol) in THF (4 ml) at 0°C were added triphenylphosphine (209.6 mg, 0.80 mmol), diethylazodicarboxylate (0.15 ml, 0.80 mmol), 5-(2-nitrophenyl)-1,3,4-thiadiazole-2-thiol (296.7 mg, 1.24 mmol) and stirred at room temperature for 6 h. The solution was purified by preparative TLC (hexane/ethyl acetate = 2/1) to obtain 7(*S*)-1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-7-deoxy-4′-*n*-propyl-7-{5-(2-nitrophenyl)-1,3,4-thiadiazol-2-ylthio}-2,3,4-tris-*O*-(trimethylsilyl)lincomycin (235.4 mg with unseparable impurity). To this crude compound (235.4 mg 0.24 mmol) was added 2,2,2-trifluoroacetic acid (1 ml) at 0 °C and stirred at room temperature for 30 min. The solution was concentrated under reduced pressure. The resulting residue was purified by preparative TLC ($CHCl_3$/MeOH/28% aq $NH_4OH=9/2/0.2$) to obtain the title compound (57.3 mg, 36.1% in 2 steps from **47**) as a colorless solid. $[\alpha]_D^{26}$ +91.0° (*c* 0.40, MeOH); ESI-MS *m/z* 656 $(M+H)^+$ as $C_{28}H_{41}N_5O_7S_3$; TOF-ESI-HRMS $(M+H)^+$ calcd. for $C_{28}H_{41}N_5O_7S_3$: 656.2246, found: 656.2239; ^{1}H NMR (400 MHz, CD_3OD) δ 0.86–0.97 (m, 6H), 1.22–1.42 (m, 8H), 1.56 (d, $J=6.8$ Hz, 3H), 1.63 (dd, $J=13.0$, 8.2 Hz, 1H), 2.00 (s, 3H), 2.11–2.19 (m, 1H), 2.79–2.88 (m, 2H), 3.56 (dd, $J=10.2$, 3.2 Hz, 1H), 3.80–3.86 (m, 1H), 3.93 (t, $J=8.4$ Hz, 1H), 4.10 (dd, $J=10.2$, 5.6 Hz, 1H), 4.38–4.43 (m, 1H), 4.45 (dq, $J=6.8$, 2.7 Hz, 1H), 4.64 (dd, $J=10.0$, 2.7 Hz, 1H), 5.27 (d, $J=5.6$ Hz, 1H), 7.75–7.86 (m, 3H) and 8.07–8.13 (m, 1H).

1′-*N*-(*tert*-butoxycarbonyl)-4′-{3-(*tert*-butyldimethylsilyloxy) propyl}-1′-demethyl-4′-depropyllincomycin (49)

Compound **11** (3.02, crude), 1-hydroxybenzotriazole (1.35 g, 10.01 mmol), *N, N*′-dicyclohexylcarbodiimide (2.07 g, 10.01 mmol) and MTL (2.54 g, 10.01 mmol) in DMF (15.0 ml) were treated for 13 h according to the similar procedure as described for the preparation of **19** to afford **49** (3.0 g, 62.5% in 3 steps from **8**) as a colorless solid. ESI-MS *m/z* 623 $(M+H)^+$ as $C_{28}H_{54}N_2O_9SSi$; ^{1}H NMR (400 MHz, CD_3OD) δ 0.06 (s, 6H), 0.90 (s, 9H), 1.15–1.27 (m, 3H), 1.37–1.64 (m, 4H), 1.44, 1.47 (s x 2, 9H), 1.74–1.91 (m, 1H), 2.06, 2.07 (s x 2, 3H), 2.09–2.18 (m, 1H), 2.30–2.45 (m, 1H), 2.94 (br t, $J=10.0$ Hz, 1H), 3.53–3.72 (m, 4H), 3.74–3.93 (m, 1H), 3.98–4.17 (m, 3H), 4.24 (br dd, $J=8.9$, 1.3 Hz, 1H), 4.27–4.46 (m, 1H) and 5.24 (d, $J=5.4$ Hz, 1H).

1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-4′-depropyl-4′-{03-(*tert*-butyldimethylsilyloxy)propyl}- 2,3,4-tris-*O*-(trimethylsilyl) lincomycin (50)

Compound **49** (3.0 g, 4.82 mmol), trimethylchlorosilane (3.08 ml, 24.1 mmol) and hexamethyldisilazane (5.05 ml, 24.1 mmol) in pyridine (10 ml) were treated for 20 min according to the similar procedure as described for the preparation of **23** and then the crude compound and 6 N acetic acid (1.45 ml) in methanol (27 ml) were treated for 80 min according to the similar procedure

as described for the preparation of **23** to afford **50** (3.02 g, 74.8% in two steps from **49**) as a colorless solid. ESI-MS *m/z* 839 $(M+H)^+$ as $C_{37}H_{78}N_2O_9SSi_4$; ^1H NMR (400 MHz, $CDCl_3$) δ 0.04 (s, 6H), 0.07–0.20 (m, 27H), 0.89 (s, 9H), 1.05–1.22 (m, 3H), 1.35–1.90 (m, 5H), 1.48 (s, 9H), 2.07 (s, 3H), 2.11–3.25 (m, 3H), 3.44–3.66 (m, 4H), 3.70–3.89 (m, 1H), 3.91–4.18 (m, 3H), 4.19–4.42 (m, 2H) and 5.19 (br d, *J*=5.6 Hz, 1H).

7(*S*)-1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-7-deoxy-4′-depropyl-4′-(3-hydroxypropyl)-7-{5-(2-nitrophenyl)-1,3,4-thiadiazol-2-ylthio} lincomycin (51)

To a solution of compound **50** (2.87 g, 3.41 mmol) in THF (15 ml) at 0 °C were added triphenylphosphine (1.34 g, 5.12 mmol), diethylazodicarboxylate (932 µl, 5.12 mmol) and 5-(2-nitrophenyl)-1,3,4-thiadiazole-2-thiol (1.26 g, 5.29 mmol) and stirred at room temperature for 10 h. The solution was substituted by toluene, purified by silica gel column chromatography (hexane to hexane/ethyl acetate=4/1) to obtain 1′-*N*-(*tert*-butoxycarbonyl)-4′-{3-(*tert*-butyldimethylsilyloxy)propyl}-1′-demethyl-7-deoxy-4′-depropyl-7-{5-(2-nitrophenyl)-1,3,4-thiadiazol-2-ylthio}-2,3,4-tris-*O*-(trimethylsilyl)lincomycin (2.62 g, crude). To this crude compound (2.62 g) was added 1 M THF solution of *tetra-n*-butyl ammonium fluoride (14.8 ml, 14.8 mmol) and acetic acid (0.848 ml, 14.8 mmol) and stirred at room temperature for 5 h. The mixture was diluted with brine and ethyl acetate, extracted with ethyl acetate, and then the organic phase was dried over Na_2SO_4, filtrated and concentrated under reduced pressure. The resulting residue was purified by silica gel column chromatography (hexane/ethyl acetate=1/1 to ethyl acetate only to ethyl acetate/MeOH=10/1) to obtain the title compound (1.74 g with unseparable impurity (96% in two steps as reference yield)). FAB-MS *m/z* 752 $(M+Na)^+$ as $C_{30}H_{43}N_5O_{10}S_3$.

7(*S*)-4′-(3-aminopropyl)-1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-7-deoxy-4′-depropyl-7-{5-(2-nitrophenyl)-1,3,4-thiadiazol-2-ylthio} lincomycin (52)

To a solution of compound **51** (500 mg, 0.685 mmol), sodium azide (223 mg, 3.43 mmol) and triphenylphosphine (359.3 mg, 1.37 mmol) in DMF (7 ml) was added tetrabromomethane (454.3 mg, 1.37 mmol) and stirred at 50 °C for 2 h. The mixture was diluted with brine and ethyl acetate, extracted with ethyl acetate and then the organic phase was dried over Na_2SO_4, filtrated and concentrated under reduced pressure. The resulting residue was purified by silica gel column chromatography (chloroform/MeOH=10/1) to obtain 7(*S*)-4′-(3-azidopropyl)-1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-7-deoxy-4′-depropyl-7-{5-(2-nitrophenyl)-1,3,4-thiadiazol-2-ylthio}lincomycin (468 mg with unseparable impurity (91% as reference yield)). To a solution of this crude compound (239 mg, 0.317 mmol) in THF (3 ml) was added triphenylphosphine (249.0 mg, 0.95 mmol), stirred at room temperature for 1 h, and then H_2O was added and stirred at 50 °C for 2 h. The mixture was diluted with brine, ethyl acetate and 1 N HCl (400 µl), washed with ethyl acetate and then the aqueous phase was added to the saturated aqueous $NaHCO_3$, extracted with $CHCl_3$, dried over Na_2SO_4, filtrated and concentrated under reduced pressure to obtain the title compound (229 mg with unseparable impurity (99% as reference yield)).

7(*S*)-1′-demethyl-7-deoxy-4′-depropyl-4′-(3-dimethylaminopropyl)-7-{5-(2-nitrophenyl)-1,3,4-thiadiazol-2-ylthio}lincomycin (53)

Compound **52** (69.0 mg, 0.093 mmol), 36% aqueous formaldehyde (23.1 µl, 0.28 mmol), AcOH (79.5 µl, 1.39 mmol) and $NaBH(OAc)_3$ (294.3 mg, 1.39 mmol) in MeOH (1 ml) were treated at room temperature for 20 min according to the similar procedure as described for the preparation of **38** to afford 7(*S*)-1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-7-deoxy-4′-depropyl-4′-(3-dimethylaminopropyl)-7-{5-(2-nitrophenyl)-1,3,4-thiadiazol-2-ylthio}lincomycin (75.0 mg, crude). To this crude compound (75.0 mg) was added 2,2,2-trifluoroacetic acid (1 ml) at 0 °C and stirred at room temperature for 30 min. The solution was concentrated under reduced pressure. The resulting residue was purified by preparative TLC ($CHCl_3$/MeOH/28% aq. NH_4OH=9/4/0.4) to obtain the title compound (35.0 mg, 57.6% in two steps from **52**) as a colorless solid. $[\alpha]_D^{25}$ +83.7° (*c* 0.45, MeOH); ESI-MS m/z 657 $(M+H)^+$ as $C_{27}H_{40}N_6O_7S_3$; TOF-ESI-HRMS $(M+H)^+$ calcd. for $C_{27}H_{40}N_6O_7S_3$: 657.2199, found: 657.2193; ^1H NMR (400 MHz, CD_3OD) δ 1.34–1.44 (m, 2H), 1.48–1.64 (m, 2H), 1.57 (d, *J*=7.0 Hz, 3H), 1.76–1.88 (m, 1H), 2.01 (s, 3H), 2.04–2.13 (m, 2H), 2.34 (s, 6 H), 2.46 (t, *J*=7.8 Hz, 2H), 2.56 (dd, *J*=10.3, 7.8 Hz, 1H), 3.24 (dd, *J*=10.3, 6.8 Hz, 1H), 3.56 (dd, *J*=10.2, 3.3 Hz, 1H), 3.81 (dd, *J*=9.3, 3.9 Hz, 1H), 3.83 (br dd, *J*=3.3, 0.8 Hz, 1H), 4.10 (dd, *J*=10.2, 5.6 Hz, 1H), 4.41 (br dd, *J*=9.9, 0.8 Hz, 1H), 4.46 (dq, *J*=7.0, 2.8 Hz, 1H), 4.62 (dd, *J*=9.9, 2.8 Hz, 1H), 5.28 (d, *J*=5.6 Hz, 1H), 7.75–7.87 (m, 3H) and 8.06–8.15 (m, 1H).

7(*S*)-1′-demethyl-7-deoxy-4′-depropyl-4′-(3-methoxypropyl)-7-{5-(2-nitrophenyl)-1,3,4-thiadiazol-2-ylthio}lincomycin (54)

Compound **24** (266.0 mg, 0.36 mmol), triphenylphosphine (142.0 mg, 0.54 mmol), diethylazodicarboxylate (98 µl, 0.54 mmol) and 5-(2-nitrophenyl)-1,3,4-thiadiazole-2-thiol (132.0 mg, 0.56 mmol) in THF (2 ml) were treated for 18 h according to the similar procedure as described for the preparation of **31** to afford 7(*S*)-1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-7-deoxy-4′-depropyl-4′-(3-methoxypropyl)-7-{5-(2-nitrophenyl)-1,3,4-thiadiazol-2-ylthio}-2,3,4-tris-*O*-(trimethylsilyl)lincomycin (325.0 mg as crude). To this crude compound (325.0 mg) was added 2,2,2-trifluoroacetic acid (1 ml) at 0 °C and stirred at room temperature for 15 min. The solution was concentrated under reduced pressure. The resulting residue was purified by preparative TLC ($CHCl_3$/MeOH/28% aq. NH_4OH=9/2/0.2) to obtain the title compound (112.0 mg, 48.3% in two steps from **24**) as a colorless solid. $[\alpha]_D^{24}$ +82.6° (*c* 0.45, MeOH); ESI-MS *m/z* 644 $(M+H)^+$ as $C_{26}H_{37}N_5O_8S_3$; TOF-ESI-HRMS $(M+H)^+$ calcd. for $C_{26}H_{37}N_5O_8S_3$: 644.1883, found: 644.1880; ^1H NMR (400 MHz,CD_3OD) δ 1.40–1.52 (m, 2H), 1.54–1.65 (m, 2H), 1.57 (d, *J*=6.9 Hz, 3H), 1.89–1.97 (m, 1H), 2.01 (s, 3H), 2.09–2.23 (m, 2H), 2.68 (dd, *J*=10.7, 8.0 Hz, 1H), 3.30 (s, 3H), 3.32–3.36 (m, 1H), 3.39 (t, *J*=6.2, 2H), 3.57 (dd, *J*=10.2, 3.2 Hz, 1H), 3.85 (br dd, *J*=3.2, 0.8 Hz, 1H), 4.02 (dd, *J*=9.1, 4.1 Hz, 1H), 4.11 (dd, *J*=10.2, 5.6 Hz, 1H), 4.42 (br dd, *J*=10.0, 0.8 Hz, 1H), 4.47 (dq, *J*=6.9, 2.7 Hz, 1H), 4.66 (dd, *J*=10.0, 2.7 Hz, 1H), 5.28 (d, *J*=5.6 Hz, 1H), 7.76–7.87 (m, 3H) and 8.06–8.14 (m, 1H).

7(*S*)-4′-*i*-butyl-1′-demethyl-7-deoxy-4′-depropyl-7-(5-(2-nitrophenyl)-1,3,4-thiadiazol-2-yl)thiolincomycin (55)

Compound **25** (204.0 mg, 0.28 mmol), triphenylphosphine (109.1 mg, 0.42 mmol), diethylazodicarboxylate (75.8 µl, 0.42 mmol) and 5-(2-nitrophenyl)-1,3,4-thiadiazole-2-thiol (103.0 mg, 0.43 mmol) in THF (2 ml) were treated for 14.5 h according to the similar procedure as described for the preparation of **31** to afford 7(*S*)-1′-*N*-(*tert*-butoxycarbonyl)-4′-*i*-butyl-1′-demethyl-7-deoxy-4′-depropyl-7-{5-(2-nitrophenyl)-1,3,4-thiadiazol-2-ylthio}-2,3,4-tris-*O*-(trimethylsilyl)lincomycin (170.0 mg as crude). To this crude compound (170.0 mg) was added 2,2,2-trifluoroacetic acid (1 ml) at 0 °C and stirred at room temperature for 30 min. The solution was concentrated under reduced pressure. The resulting residue was purified by preparative TLC ($CHCl_3$/MeOH/28% aq. NH_4OH=9/2/0.2) to obtain the title compound (39.0 mg, 22.5% in two steps from **25**) as a colorless solid. $[\alpha]_D^{24}$ +85.8° (*c* 0.54, MeOH); ESI-MS *m/z* 628 $(M+H)^+$ as $C_{26}H_{37}N_5O_7S_3$; TOF-ESI-HRMS $(M+H)^+$ calcd. for $C_{26}H_{37}N_5O_7S_3$: 628.1933, found: 628.1936; ^1H NMR (400 MHz, CD_3OD) δ 0.90 (d, *J*=5.3 Hz, 3H), 0.91 (d, *J*=5.4 Hz, 3H), 1.23–1.34 (m, 2H), 1.53–1.64 (m, 1H), 1.57 (d, *J*=6.8 Hz, 3H), 1.81–1.92 (m, 1H), 2.00 (s, 3H), 2.04–2.15 (m, 1H), 2.15–2.27 (m, 1H), 2.58 (dd, *J*=10.4, 8.7 Hz, 1H), 3.26–3.30 (m, 1H), 3.56 (dd, *J*=10.2, 3.2 Hz, 1H), 3.84 (br dd, *J*=3.2, 0.8 Hz, 1H), 3.92 (dd, *J*=9.2, 4.3 Hz, 1H), 4.10 (dd, *J*=10.2, 5.6 Hz, 1H), 4.41 (br dd, *J*=10.0, 0.8 Hz, 1H), 4.47 (dq, *J*=6.8, 2.8 Hz, 1H), 4.65 (dd, *J*=10.0, 2.8 Hz, 1H), 5.28 (d, *J*=5.6 Hz, 1H), 7.76–7.87 (m, 3H) and 8.08–8.13 (m, 1H).

7(*S*)-1′-demethyl-7-deoxy-4′-depropyl-7-{5-(2-nitrophenyl)-1,3,4-thiadiazol-2-ylthio}-4′-*n*-pentyllincomycin (56)

Compound **26** (400 mg, 0.54 mmol), triphenylphosphine (213.5 mg, 0.81 mmol), diethylazodicarboxylate (150 µl, 0.81 mmol) and 5-(2-nitrophenyl)-1,3,4-thiadiazole-2-thiol (201.4 mg, 0.84 mmol) in THF (4 ml) were treated for 24 h according to the similar procedure as described for the preparation of **31** to afford 7(*S*)-1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-7-

deoxy-4′-depropyl-7-{5-(2-nitrophenyl)-1,3,4-thiadiazol-2-ylthio}-4′-*n*-pentyl-2,3,4-tris-*O*-(trimethylsilyl)lincomycin (374.0 mg as crude). To this crude compound (374 mg) was added 2,2,2-trifluoroacetic acid (1 ml) at 0 °C and stirred at room temperature for 30 min. The solution was concentrated under reduced pressure. The resulting residue was purified by preparative TLC ($CHCl_3$/MeOH/28% aq. NH_4OH = 9/2/0.2) to obtain the title compound (72.6 mg, 20.9% in two steps from **26**) as a colorless solid. $[\alpha]_D^{25}$ +34.3° (*c* 0.10, MeOH); ESI-MS *m/z* 642 $(M+H)^+$ as $C_{27}H_{39}N_5O_7S_3$; TOF-ESI-HRMS $(M+H)^+$ calcd. for $C_{27}H_{39}N_5O_7S_3$: 642.2090, found: 642.2083; ^{1}H NMR (400 MHz, CD_3OD) δ 0.80–0.95 (m, 3H), 1.21–1.43 (m, 8H), 1.57 (d, *J* = 7.0 Hz, 3H), 1.81–1.95 (m, 1H), 2.02–2.14 (m, 2H), 2.09 (s, 3H), 2.62 (dd, *J* = 10.4, 7.9 Hz, 1H), 3.27 (dd, *J* = 10.5, 6.8 Hz, 1H), 3.61 (dd, *J* = 10.2, 3.2 Hz, 1H), 3.83 (br dd, *J* = 3.2, 0.9 Hz, 1H), 3.91 (dd, *J* = 9.1, 4.2 Hz, 1H), 4.14 (dd, *J* = 10.2, 5.6 Hz, 1H), 4.34–4.40 (m, 1H), 4.42 (dq, *J* = 7.0, 2.9 Hz, 1H), 4.57 (dd, *J* = 10.0, 2.9 Hz, 1H), 5.31 (d, *J* = 5.6 Hz, 1H), 7.71–7.87 (m, 3H) and 8.07–8.14 (m, 1H).

7(*S*)-7-deoxy-4′-depropyl-7-{5-(2-nitrophenyl)-1,3,4-thiadiazol-2-ylthio}-4′-*n*-pentyllincomycin (57)

Compound **56** (56.1 mg, 0.087 mmol), 36% aqueous formaldehyde (22 μl, 0.26 mmol), AcOH (30 μl, 0.52 mmol) and $NaBH(OAc)_3$ (55.5 mg, 0.26 mmol) in MeOH (0.6 ml) were treated at room temperature for 1 h according to the similar procedure as described for the preparation of **38** to afford **57** (49.1 mg, 85.7%) as a colorless solid. $[\alpha]_D^{25}$ +91.6° (*c* 1.16, MeOH); ESI-MS *m/z* 656 $(M+H)^+$ as $C_{28}H_{41}N_5O_7S_3$; TOF-ESI-HRMS $(M+H)^+$ calcd. for $C_{28}H_{41}N_5O_7S_3$: 656.2246, found: 656.2238; ^{1}H NMR (400 MHz, CD_3OD) δ 0.83–0.92 (m, 3H), 1.21–1.42 (m, 8H), 1.57 (d, *J* = 7.0 Hz, 3H), 1.78–1.91 (m, 1H), 1.96–2.10 (m, 2H), 2.01 (s, 3H), 2.12–2.27 (m, 1H), 2.39 (s, 3H), 3.02 (dd, *J* = 10.5, 5.1 Hz, 1H), 3.23–3.29 (m, 1H), 3.57 (dd, *J* = 10.2, 3.2 Hz, 1H), 3.81 (dd, *J* = 3.2, 0.7 Hz, 1H), 4.11 (dd, *J* = 10.2, 5.6 Hz, 1H), 4.39–4.49 (m, 1H), 4.60 (dd, *J* = 9.8, 3.2 Hz, 1H), 5.27 (d, *J* = 5.6, 1H), 7.75–7.85 (m, 3H) and 8.06–8.11 (m, 1H).

7(*S*)-1′-demethyl-7-deoxy-4′-depropyl-7-[5-{5-(methylamino)thiazol-4-yl}-1,3,4-thiadiazol-2-ylthio]-4′-*n*-pentyllincomycin (58)

Compound **26** (400 mg, 0.54 mmol), triphenylphosphine (213.5 mg, 0.81 mmol), diethylazodicarboxylate (150 μl, 0.81 mmol) and 5-{5-(methylamino)thiazol-4-yl}-1,3,4-thiadiazole-2-thiol (193.9 mg, 0.84 mmol) in THF (4 ml) were treated for 24 h according to the similar procedure as described for the preparation of **31** to afford 7(*S*)-1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-7-deoxy-4′-depropyl-7-[5-{5-(methylamino)thiazol-4-yl}-1,3,4-thiadiazol-2-ylthio]-4′-*n*-pentyl-2,3,4-tris-*O*-(trimethylsilyl)lincomycin (109.8 mg, 21.3%). To the compound (109.8 mg, 0.12 mmol) was added 2,2,2-trifluoroacetic acid (1 ml) at 0 °C and stirred at room temperature for 30 min. The solution was concentrated under reduced pressure. The resulting residue was purified by preparative TLC ($CHCl_3$/MeOH/28% aq. NH_4OH = 9/2/0.2) to obtain the title compound (59.0 mg, 80.6%) as a colorless solid. $[\alpha]_D^{25}$ +48.5° (*c* 0.22, MeOH); ESI-MS *m/z* 633 $(M+H)^+$ as $C_{25}H_{40}N_6O_5S_4$; TOF-ESI-HRMS $(M+H)^+$ calcd. for $C_{25}H_{40}N_6O_5S_4$: 633.2021, found: 633.2021; ^{1}H NMR (400 MHz, CD_3OD) δ 0.83–0.94 (m, 3H), 1.21–1.43 (m, 8H), 1.50 (d, *J* = 6.8 Hz, 3H), 1.80–1.91 (m, 1H), 2.02 (s, 3H), 2.02–2.13 (m, 2H), 2.59 (dd, *J* = 10.5, 8.0 Hz, 1H), 3.12 (s, 3H), 3.24–3.30 (m, 1H), 3.58 (dd, *J* = 10.2, 3.2 Hz, 1H), 3.85 (br dd, *J* = 3.2, 0.7 Hz, 1H), 3.93 (dd, *J* = 9.1, 4.0 Hz, 1H), 4.11 (dd, *J* = 10.2, 5.6 Hz, 1H), 4.29 (dq, *J* = 6.8, 2.7 Hz, 1H), 4.37–4.44 (m, 1H), 4.61 (dd, *J* = 10.0, 2.7 Hz, 1H), 5.28 (d, *J* = 5.6 Hz, 1H) and 8.13 (s, 1H).

7(*S*)-7-deoxy-4′-depropyl-7-[5-{5-(methylamino)thiazol-4-yl}-1,3,4-thiadiazol-2-ylthio]-4′-*n*-pentyllincomycin (59)

Compound **58** (35.8 mg, 0.057 mmol), 36% aqueous formaldehyde (14 μl, 0.17 mmol), AcOH (20 μl, 0.34 mmol) and $NaBH(OAc)_3$ (36.0 mg, 0.17 mmol) in MeOH (0.5 ml) were treated at room temperature for 1 h according to the similar procedure as described for the preparation of **38** to afford **59** (29.4 mg, 80.3%) as an off-white solid. $[\alpha]_D^{25}$ +64.5° (*c* 0.30, MeOH); ESI-MS *m/z* 647 $(M+H)^+$ as $C_{26}H_{42}N_6O_5S_4$; TOF-ESI-HRMS $(M+H)^+$ calcd. for $C_{26}H_{42}N_6O_5S_4$: 647.2178, found: 647.2176; ^{1}H NMR (400 MHz, CD_3OD) δ 0.84–0.93 (m, 3H), 1.19–1.39 (m, 8H), 1.54 (d, *J* = 6.9 Hz, 3H), 1.80–1.92 (m, 1H), 2.02 (s, 3H), 1.96–2.21 (m, 3H), 2.40 (s, 3H), 3.06–3.14 (m, 1H), 3.12 (s, 3H), 3.21 (dd, *J* = 7.9, 5.5 Hz, 1H), 3.61 (dd, *J* = 10.2, 3.1 Hz, 1H), 3.84 (dd, *J* = 3.1, 0.6 Hz, 1H), 4.12 (dd, *J* = 10.2, 5.6 Hz, 1H), 4.28 (dq, *J* = 6.9, 2.9 Hz, 1H), 4.43–4.44 (m, 1H), 4.59 (dd, *J* = 10.0, 2.9 Hz, 1H), 5.28 (d, *J* = 5.6 Hz, 1H) and 8.14 (s, 1H).

7(*S*)-1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-7-deoxy-7-{4-(methoxycarbonyl)phenylthio}lincomycin (60)

To a solution of compound **23** (5.0 g, 7.05 mmol) in $CHCl_3$ (22 ml) were added Et_3N (2.45 ml, 17.6 mmol), methanesulfonyl chloride (1.1 ml, 14.1 mmol) and stirred at room temperature for 30 min. The mixture was added to saturated aqueous $NaHCO_3$, extracted with $CHCl_3$, dried over Na_2SO_4 and concentrated under reduced pressure. To a solution of crude compound in DMF (50 ml) were added K_2CO_3 (2.92 g, 21.2 mmol) and methyl 4-mercaptobenzoate (2.37 g, 14.1 mmol), stirred at 100 °C for 6 h and concentrated under reduced pressure. The resulting residue in MeOH (50 ml) was added to 1 N HCl (100 ml) and stirred at room temperature for 20 min. The mixture was added to the saturated aqueous $NaHCO_3$, then extracted with ethyl acetate, dried over Na_2SO_4, filtrated and concentrated under reduced pressure. The resulting residue was purified by silica gel column chromatography ($CHCl_3$/MeOH/28% aq. NH_4OH = 20/1/0.1) to obtain the title compound as a colorless solid (1.25 g, 27.6% in three steps from **23**). ESI-MS (*m/z*) 643 $(M+H)^+$ as $C_{30}H_{46}N_2O_9S_2$; ^{1}H NMR (400 MHz, CD_3OD) δ 0.86–1.00 (m, 3H), 1.25–1.65 (m, 16H), 1.45, 1.48 (s x 2, 9H), 1.73–1.99 (m, 1H), 1.79, 1.85 (s x 2, 3H), 2.07–2.20 (m, 1H), 2.22–2.42 (m, 1H), 2.92–3.01 (m, 1H), 3.53–3.61 (m, 1H), 3.63–3.74 (m, 1H), 3.88 (s, 3H), 3.90–4.15 (m, 3H), 4.28–4.50 (m, 2H), 4.54–4.68 (m, 1H), 5.24 (d, *J* = 5.4 Hz, 1H), 7.39–7.46 (m, 2H) and 7.89–7.96 (m, 2H).

7(*S*)-1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-7-deoxy-4′-depropyl-7-(4-(methoxycarbonyl)phenyl)thio-4′-{(*E*)-pent-2-enyl}lincomycin (61)

Compound **27** (1.45 g, 1.97 mmol), Et_3N (0.69 ml, 4.93 mmol) and methanesulfonyl chloride (0.31 ml, 3.93 mmol) in $CHCl_3$ (6.1 ml) were treated at room temperature for 30 min according to the similar procedure as described for the preparation of **60** to afford 1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-4′-depropyl-7-*O*-methanesulfonyl-4′-{(*E*)-pent-2-enyl}-2,3,4-tris-*O*-(trimethylsilyl)lincomycin. To a solution of crude compound were added K_2CO_3 (1.23 g, 8.87 mmol) and methyl 4-mercaptobenzoate (0.99 g, 5.91 mmol) in DMF (13.8 ml) and treated at 80 °C for 1 h. The resulting residue and 1 N HCl (14 ml) in MeOH (14 ml) were treated at room temperature for 20 min according to the similar procedure as described for the preparation of **60** to afford **61** (1.21 g, 92% in three steps from **27**) as a colorless solid. EI-MS *m/z* 668 $(M)^+$ as $C_{32}H_{48}N_2O_9S_2$; ^{1}H NMR (400 MHz, CD_3OD) δ 0.95 (t, *J* = 7.4 Hz, 3H), 1.33–1.44 (m, 3H), 1.45, 1.48 (s x 2, 9H), 1.80, 1.85 (s x 2, 3H), 1.87–2.17 (m, 6H), 2.26–2.40 (m, 1H), 3.01–3.09 (m, 1H), 3.51–3.67 (m, 2H), 3.88 (s, 3H), 3.91–4.12 (m, 3H), 4.27–4.46 (m, 2H), 4.53–4.64 (m, 1H), 5.24 (d, *J* = 5.6 Hz, 1H), 5.31–5.45 (m, 1H), 5.47–5.59 (m, 1H), 7.36–7.44 (m, 2H) and 7.86–7.95 (m, 2H).

7(*S*)-1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-7-deoxy-4′-depropyl-7-{4-(methoxycarbonyl)phenylthio}-4′-*n*-pentyllincomycin (62)

Compound **61** (105 mg, 0.16 mmol) and Pd/C (100 mg) in MeOH (2 ml) were treated for 15 h according to the similar procedure as described for the preparation of **7** to afford **62** (97 mg, 92.1%) as a colorless solid. ESI-MS *m/z* 671 $(M+H)^+$ as $C_{32}H_{50}N_2O_9S_2$; ^{1}H NMR (400 MHz, CD_3OD) δ 0.18–0.92 (m, 3H), 1.15–1.52 (m, 11H), 1.44, 1.47 (s x 2, 9H), 1.71–1.93 (m, 1H), 1.78, 1.83 (s x 2, 3H), 2.05–2.18 (m, 1H), 2.20–2.36 (m, 1H), 2.95 (br t, *J* = 10.0 Hz, 1H), 3.52–3.61 (m, 1H), 3.61–3.73 (m, 1H), 3.87 (s, 3H), 3.91–4.03 (m, 2H), 4.03–4.15 (m, 1H), 4.28–4.47 (m, 2H), 4.53–4.65 (m, 1H), 5.25 (d, *J* = 5.6 Hz, 1H), 7.35–7.47 (m, 2H) and 7.85–7.95 (m, 2H).

7(*S*)-1′-*N*-(*tert*-butoxycarbonyl)-7-{4-(carboxyl)phenylthio}-1′-demethyl-7-deoxylincomycin (63)

To a solution of compound **60** (761 mg, 1.18 mmol) in MeOH (20 ml) was added 1 N NaOH (1.78 ml, 1.78 mmol) and stirred at room temperature for 7 days. The mixture was added 1 N HCl (pH = 3), extracted with $CHCl_3$, dried over Na_2SO_4 and concentrated under reduced pressure to obtain the title compound (705 mg, 94.7%) as an off-white solid. ESI-MS *m/z* 629 $(M+H)^+$ as $C_{29}H_{44}N_2O_9S_2$.

7(*S*)-1′-*N*-(*tert*-butoxycarbonyl)-7-{4-(carboxyl)phenylthio}-1′-demethyl-7-deoxy-4′-depropyl-4′-*n*-pentyllincomycin (64)

Compound **62** (97 mg, 0.15 mmol) and 1 N NaOH (0.55 ml) in MeOH (1.1 ml) were treated for 18 h according to the similar procedure as described for the preparation of **63** to afford **64** (91.3 mg, 96.1%) as a colorless solid. FAB-MS *m/z* 695 $(M+K)^+$ as $C_{31}H_{48}N_2O_9S_2$; ^{1}H NMR (400 MHz, CD_3OD) δ 0.82–0.92 (m, 3H), 1.20–1.45 (m, 11H), 1.46, 1.48 (s x 2, 9H), 1.75–1.95 (m, 1H), 1.81, 1.86 (s x 2, 3H), 2.09–2.19 (m, 1H), 2.21–2.37 (m, 1H), 2.90–3.02 (m, 1H), 3.54–3.62 (m, 1H), 3.63–3.75 (m, 1H), 3.90–4.05 (m, 2H), 4.05–4.15 (m, 1H), 4.30–4.49 (m, 2H), 4.53–4.67 (m, 1H), 5.27 (d, *J* = 5.6 Hz, 1H), 7.37–7.45 (m, 2H) and 7.89–7.97 (m, 2H).

7(*S*)-1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-7-deoxy-7-{4-(morpholinocarbonyl)phenylthio}lincomycin (65)

To a solution of compound **63** (200 mg, 0.32 mmol), 1-hydroxybenzotriazole (64.5 mg, 0.48 mmol) and 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide HCl salt (91.5 mg, 0.48 mmol) in DMF (2 ml) was added to morpholine (42 µl, 0.48 mmol) and stirred at room temperature for 22 h. The mixture was added to saturated aqueous $NaHCO_3$, extracted with ethyl acetate, dried over Na_2SO_4 and concentrated under reduced pressure. The resulting residue was purified by preparative TLC ($CHCl_3$/MeOH/28% aq. NH_4OH = 9/2/0.2) to obtain the title compound (215.0 mg, 96.8%) as a colorless solid. ESI-MS *m/z* 698 $(M+H)^+$ as $C_{33}H_{51}N_3O_9S_2$; ^{1}H NMR (400 MHz, CD_3OD) δ 0.86–0.97 (m, 3H), 1.25–1.45 (m, 7H), 1.46, 1.48 (s x 2, 9H), 1.78–1.95 (m, 1H), 1.88, 1.91 (s x 2, 3H), 2.06–2.18 (m, 1H), 2.24–2.40 (m, 1H), 2.92–2.99 (m, 1H), 3.37–3.85 (m, 10H), 3.88–3.99 (m, 2H), 4.05–4.14 (m, 1H), 4.30–4.45 (m, 2H), 4.52–4.63 (m, 1H), 5.26 (d, *J* = 5.6 Hz, 1H), 7.35–7.42 (m, 2H) and 7.43–7.48 (m, 2H).

7(*S*)-1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-7-deoxy-4′-depropyl-7-{4-(morpholinocarbonyl)phenylthio}-4′-*n*-pentyllincomycin (66)

Compound **64** (91.3 mg, 0.14 mmol), 1-hydroxybenzotriazole (28.1 mg, 0.21 mmol) and 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide HCl salt (40.0 mg, 0.21 mmol) and morpholine (18 µl, 0.21 mmol) in DMF (1 ml) were treated for 62 h according to the similar procedure as described for the preparation of **65** to afford **66** (82.0 mg, 81.3%) as a colorless solid. FAB-MS *m/z* 726 $(M+H)^+$ as $C_{35}H_{55}N_3O_9S_2$; ^{1}H NMR (400 MHz, CD_3OD) δ 0.82–0.94 (m, 3H), 1.21–1.42 (m, 11H), 1.46, 1.48 (s x 2, 9H), 1.77–1.95 (m, 1H), 1.87, 1.91 (s x 2, 3H), 2.08–2.18 (m, 1H), 2.20–2.38 (m, 1H), 2.90–3.01 (m, 1H), 3.38–3.84 (m, 10H), 3.87–4.01 (m, 2H), 4.05–4.14 (m, 1H), 4.29–4.46 (m, 2H), 4.52–4.63 (m, 1H), 5.26 (d, *J* = 5.6 Hz, 1H), 7.35–7.42 (m, 2H) and 7.42–7.48 (m, 2H).

7(*S*)-1′-demethyl-7-deoxy-7-{4-(morpholinocarbonyl)phenylthio}lincomycin (67)

To the compound **65** (215 mg, 0.32 mmol) was added 2,2,2-trifluoroacetic acid (2 ml) at 0 °C and stirred for 40 min. The solution was concentrated under reduced pressure. The resulting residue was purified by preparative TLC ($CHCl_3$/MeOH/28% aq. NH_4OH = 9/2/0.2) to obtain the title compound (104.0 mg, 56.5%) as a colorless solid. $[\alpha]_D^{25}$ +70.5° (*c* 0.42, MeOH); ESI-MS *m/z* 598 $(M+H)^+$ as $C_{28}H_{43}N_3O_7S_2$; TOF-ESI-HRMS $(M+H)^+$ calcd. for $C_{28}H_{43}N_3O_7S_2$: 598.2621, found: 598.2623; ^{1}H NMR (400 MHz, CD_3OD) δ 0.88–0.97 (m, 3H), 1.30–1.44 (m, 4H), 1.33 (d, *J* = 7.0 Hz, 3H), 1.80–1.93 (m, 1H), 1.90 (s, 3H), 2.05–2.18 (m, 2H), 2.65 (dd, *J* = 10.4, 8.2 Hz, 1H), 3.26–3.32 (m, 1H), 3.35–3.56 (m, 2H), 3.56 (dd, *J* = 10.2, 3.3 Hz, 1H), 3.52–3.85 (m, 6H), 3.79 (br dd, *J* = 3.3, 0.8 Hz, 1), 3.90–3.98 (m, 2H), 4.08 (dd, *J* = 10.2, 5.6 Hz, 1H), 4.37 (br dd, *J* = 9.9, 0.8 Hz, 1H), 4.54 (dd, *J* = 9.9, 2.7 Hz, 1H), 5.25 (d, *J* = 5.6 Hz, 1H), 7.35–7.41 (m, 2H) and 7.43–7.49 (m, 2H).

7(*S*)-1′-demethyl-7-deoxy-4′-depropyl-7-{4-(morpholinocarbonyl)phenylthio}-4′-*n*-pentyllincomycin (68)

Compound **66** (82.0 mg, 0.11 mmol) and 2,2,2-trifluoroacetic acid (1 ml) were treated at −15 to 0 °C for 40 min according to the similar procedure as described for the preparation of **67** to afford **68** (57.0 mg, 80.6%) as a colorless solid. $[\alpha]_D^{26}$ +74.0° (*c* 0.51, MeOH); ESI-MS *m/z* 626 $(M+H)^+$ as $C_{30}H_{47}N_3O_7S_2$; TOF-ESI-HRMS $(M+H)^+$ calcd. for $C_{30}H_{47}N_3O_7S_2$: 626.2934, found: 626.2924; ^{1}H NMR (400 MHz, CD_3OD) δ 0.83–0.93 (m, 3H), 1.23–1.44 (m, 8H), 1.33 (d, *J* = 6.9 Hz, 3H), 1.77–1.88 (m, 1H), 1.90 (s, 3H), 1.99–2.15 (m, 2H), 2.61 (dd, *J* = 10.4, 8.2 Hz, 1H), 3.24 (dd, *J* = 10.4, 7.0 Hz, 1H), 3.36–3.57 (m, 2H), 3.56 (dd, *J* = 10.2, 3.2 Hz, 1H), 3.57–3.84 (m, 6H), 3.78 (br dd, *J* = 3.2, 0.7 Hz, 1H), 3.88 (dd, *J* = 9.5, 4.0 Hz, 1H), 3.95 (dq, *J* = 6.9, 2.6 Hz, 1H), 4.08 (dd, *J* = 10.2, 5.6 Hz, 1H), 4.36 (br dd, *J* = 9.9, 0.7 Hz, 1H), 4.53 (dd, *J* = 9.9, 2.6 Hz, 1H), 5.25 (d, *J* = 5.6 Hz, 1H), 7.34–7.41 (m, 2H) and 7.42–7.50 (m, 2H).

7(*S*)-7-deoxy-4′-depropyl-7-{4-(morpholinocarbonyl)phenylthio}-4′-*n*-pentyllincomycin (69)

Compound **68** (31.0 mg, 0.050 mmol), 36% aqueous formaldehyde (12 µl, 0.15 mmol), AcOH (17 µl, 0.30 mmol) and $NaBH(OAc)_3$ (31.6 mg, 0.15 mmol) in MeOH (0.5 ml) were treated at room temperature for 30 min according to the similar procedure as described for the preparation of **38** to afford **69** (31.0 mg, 97.8%) as a colorless solid. $[\alpha]_D^{26}$ +68° (*c* 0.12, MeOH); ESI-MS *m/z* 640 $(M+H)^+$ as $C_{31}H_{49}N_3O_7S_2$; TOF-ESI-HRMS $(M+H)^+$ calcd. for $C_{31}H_{49}N_3O_7S_2$: 640.3090, found: 640.3080; ^{1}H NMR (400 MHz, CD_3OD) δ 0.85–0.94 (m, 3H), 1.23–1.43 (m, 8H), 1.35 (d, *J* = 6.8 Hz, 3H), 1.81–1.92 (m, 1H), 1.91 (s, 3H), 2.03 (ddd, *J* = 13.0, 7.8, 5.0 Hz, 1H), 2.08–2.25 (m, 2H), 2.44 (s, 3H), 3.06 (dd, *J* = 10.5, 4.9 Hz, 1H), 3.25–3.30 (m, 1H), 3.38–3.84 (m, 9H), 3.58 (dd, *J* = 10.2, 3.2 Hz, 1H), 3.97 (dq, *J* = 6.8, 2.6 Hz, 1H), 4.10 (dd, *J* = 10.2, 5.6 Hz, 1H), 4.33–4.39 (m, 1H), 4.51 (dd, *J* = 9.7, 2.6 Hz, 1H), 5.26 (d, *J* = 5.6 Hz, 1H), 7.37–7.42 (m, 2H) and 7.45–7.50 (m, 2H).

7(*S*)-7-acetylthio-1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-7-deoxylincomycin (70)

To a solution of compound **23** (500.3 mg, 0.71 mmol) in CH_2Cl_2 (10 ml) were added Et_3N (0.25 ml, 1.77 mmol) and methanesulfonyl chloride (0.11 ml, 1.39 mmol), stirred at 0 °C for 1 h. The mixture was added to saturated aqueous NH_4Cl, extracted with ethyl acetate, washed with 25% brine, dried over Na_2SO_4 and concentrated under reduced pressure. To a solution of this crude compound in DMF (8 ml) was added AcSK (501.7 mg, 4.39 mmol), stirred at 60°C for 10 h. The mixture was added to the saturated aqueous $NaHCO_3$, then extracted with ethyl acetate, dried over Na_2SO_4, filtrated and concentrated under reduced pressure. The resulting residue was purified by silica gel column chromatography (hexane/ethyl acetate = 10/1 to 2/1) to obtain 7(*S*)-7-acetylthio-1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-7-deoxy-2,3,4-tris-*O*-(trimethylsilyl)lincomycin as a colorless solid (217.8 mg, 40.2% in two steps from **23**). To a solution of this intermediate in MeOH (2.2 ml) was added 1 N HCl (2.2 ml) and stirred at room temperature for 1 h. The mixture was added to saturated aqueous $NaHCO_3$, extracted with ethyl acetate, dried over Na_2SO_4 and concentrated under reduced pressure. The resulting residue was purified by silica gel column chromatography (chloroform/MeOH = 40/1) to obtain the title compound (137 mg, 87.6%) as a colorless solid. ESI-MS *m/z* 551 $(M+H)^+$ as $C_{24}H_{42}N_2O_8S_2$; ^{1}H NMR (400 MHz, CD_3OD) δ 0.86–0.98 (m, 3H), 1.26–1.41 (m, 7H), 1.46 (s, 9H), 1.76–1.96 (m, 1H), 2.01, 2.03 (s, 3H), 1.95–2.17 (m, 1H), 2.20–2.38 (m, 1H), 2.32 (s, 3H), 2.95 (t, *J* = 9.8 Hz, 1H), 3.51 (dd, *J* = 10.2, 3.3 Hz, 1H), 3.66 (dd, *J* = 10.0, 7.6 Hz, 1H), 3.81–4.01 (m, 2H), 4.01–4.12 (m, 1H), 4.13–4.37 (m, 2H), 4.41–4.50 (m, 1H) and 5.21 (d, *J* = 5.5 Hz, 1H).

7(*S*)-1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-7-deoxy-7-mercaptolincomycin (71)

To a solution of compound **70** (137 mg, 0.25 mmol) in MeOH (1.5 ml) was added sodium methoxide (43.2 mg, 0.76 mmol) and stirred at room temperature for 1.5 h. The mixture was diluted with 8% aqueous $NaHCO_3$, extracted with ethyl acetate, dried over Na_2SO_4 and concentrated under reduced pressure. The resulting residue was purified by silica gel column chromatography ($CHCl_3/CH_3OH = 40/1$ to 10/1) to obtain the title compound (120.3 mg, 95.1%) as a colorless solid. FAB-MS *m/z* 509 $(M+H)^+$ as $C_{22}H_{40}N_2O_7S_2$; ^1H NMR (400 MHz, CD_3OD) δ 0.87–0.99 (m, 3H), 1.23–1.42 (m, 7H), 1.47 (s, 9H), 1.80–1.98 (m, 1H), 2.05–2.15 (m, 1H), 2.15 (s, 3H), 2.20–2.39 (m, 1H), 2.97 (br t, *J* = 9.6 Hz, 1H), 3.39–3.58 (m, 1H), 3.54 (dd, *J* = 10.2, 3.0 Hz, 1H), 3.66 (dd, *J* = 10.2, 7.4 Hz, 1H), 3.80–3.90 (m, 1H), 4.02–4.18 (m, 2H), 4.26–4.46 (m, 2H) and 5.25 (d, *J* = 5.5 Hz, 1H).

7(*S*)-1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-7-deoxy-7-{(4-(pyridin-3-yl)phenylthio}lincomycin (72)

To a solution of 3-(4-bromophenyl)pyridine (75.4 mg, 0.32 mmol), 4,5-bis (diphenylphosphino)-9,9-dimethylxanthene (13.4 mg, 0.023 mmol) and tris (dibenzylideneacetone)dipalladium(0) ($Pd_2(dba)_3$) (12.3 mg, 13.4 μmol) in 1,4-dioxane (1 ml) were added to compound **71** (120.3 mg, 0.24 mmol) and *N*,*N*-diisopropylethylamine (82 μl, 0.47 mmol) and refluxed for 5 h. The mixture was filtrated by either Chromatodisc (0.45 μm) (Kurabo Industries) or celite, concentrated under reduced pressure. The resulting residue was purified by silica gel column chromatography ($CHCl_3$/MeOH/28% aq. $NH_4OH = 10/1/0.1$) to obtain the title compound as an off-white solid (125.9 mg, 80.4%). FAB-MS *m/z* 662 $(M+H)^+$ as $C_{33}H_{47}N_3O_7S_2$; ^1H NMR (400 MHz, CD_3OD) δ 0.87–1.00 (m, 3H), 1.21–1.43 (m, 7H), 1.48 (s, 9H), 1.77–1.95 (m, 1H), 1.94, 1.97 (s x 2, 3H), 2.05–2.21 (m, 1H), 2.22–2.44 (m, 1H), 2.97 (t, *J* = 9.6 Hz, 1H), 3.59 (dd, *J* = 10.0, 3.2 Hz, 1H), 3.68 (br dd, *J* = 9.8, 8.1 Hz, 1H), 3.85–4.01 (m, 2H), 4.05–4.16 (m, 1H), 4.28–4.41 (m, 1H), 4.41–4.50 (m, 1H), 4.51–4.64 (m, 1H), 5.27 (d, *J* = 5.5 Hz, 1H), 7.47–7.57 (m, 3H), 7.59–7.69 (m, 2H), 8.05–8.14 (m, 1H), 8.51 (dd, *J* = 4.8, 1.5 Hz, 1H) and 8.78–8.83 (m, 1H).

7(*S*)-1′-demethyl-7-deoxy-7-{(4-(pyridin-3-yl)phenylthio}lincomycin (73)

Compound **72** (125.9 mg, 0.19 mmol) and 2,2,2-trifluoroacetic acid (0.29 ml) in CH_2Cl_2 (2.5 ml) were treated at − 20 °C for 10 min, and then treated at 0 °C for 3 h according to the similar procedure as described for the preparation of **67** to afford **73** (99.1 mg, 92.7%) as a colorless solid. $[\alpha]_D^{25}$ +91.4° (*c* 0.74, MeOH); ESI-MS *m/z* 562 $(M+H)^+$ as $C_{28}H_{39}N_3O_5S_2$; TOF-ESI-HRMS $(M+H)^+$ calcd. for $C_{28}H_{39}N_3O_5S_2$: 562.2409, found: 562.2407; ^1H NMR (400 MHz, CD_3OD) δ 0.86–0.97 (m, 3H), 1.30–1.42 (m, 4H), 1.33 (d, *J* = 6.9 Hz, 3H), 1.77–1.87 (m, 1H), 1.97 (s, 3H), 2.02–2.13 (m, 2H), 2.59 (dd, *J* = 10.3, 8.0 Hz, 1H), 3.22 (dd, *J* = 10.3, 7.0 Hz, 1H), 3.58 (dd, *J* = 10.2, 3.3 Hz, 1H), 3.79 (br dd, *J* = 3.3, 0.9 Hz, 1H), 3.86 (dd, *J* = 9.3, 3.7 Hz, 1H), 3.93 (dq, *J* = 6.9, 2.6 Hz, 1H), 4.10 (dd, *J* = 10.2, 5.6 Hz, 1H), 4.36–4.42 (m, 1H), 4.51 (dd, *J* = 9.9, 2.6 Hz, 1H), 5.27 (d, *J* = 5.6 Hz, 1H), 7.51 (ddd, *J* = 8.0, 4.9, 0.9 Hz, 1H), 7.51–7.58 (m, 2H), 7.60–7.68 (m, 2H), 8.07 (ddd, *J* = 8.0, 2.4, 1.6 Hz, 1H), 8.51 (dd, *J* = 4.9, 1.6 Hz, 1H) and 8.79 (dd, *J* = 2.4, 0.9 Hz, 1H).

7(*S*)-1′-*N*-(*tert*-butoxycarbonyl)-1′-demethyl-7-deoxy-7-{4-(pyrimidin-5-yl)phenylthio}lincomycin (74)

Compound **71** (116.4 mg, 0.23 mmol), 5-(4-bromophenyl)pyrimidine (107.3 mg, 0.46 mmol), 4,5-bis(diphenylphosphino)-9,9-dimethylxanthene (14.2 mg, 0.024 mmol), $Pd_2(dba)_3$ (11.8 mg, 0.013 mmol) and *N*,*N*-diisopropylethylamine (79.5 μl, 0.46 mmol) in 1,4-dioxane (1.5 ml) were treated for 6 h according to the similar procedure as described for the preparation of **72** to afford **74** (117.9 mg, 77.7%) as a colorless solid. FAB-MS *m/z* 663 $(M+H)^+$ as $C_{32}H_{46}N_4O_7S_2$; ^1H NMR (400 MHz, CD_3OD) δ 0.86–1.00 (m, 3H), 1.25–1.43 (m, 7H), 1.48, 1.47 (s x 2, 9H), 1.80–1.95 (m, 1H), 1.92, 1.95 (s x 2, 3H), 2.04–2.20 (m, 1H), 2.21–2.44 (m, 1H), 2.97 (t, *J* = 9.6 Hz, 1H), 3.56–3.75 (m, 2H), 3.88–4.03 (m, 2H), 4.06–4.19 (m, 1H), 4.29–4.42 (m, 1H), 4.46 (d, *J* = 9.6 Hz, 1H), 4.54–4.67 (m, 1H), 5.28 (d, *J* = 5.5 Hz, 1H), 7.50–7.59 (m, 2H), 7.65–7.73 (m, 2H), 9.07 (s, 2H), 9.13 (s, 1H).

7(*S*)-1′-demethyl-7-deoxy-7-{4-(pyrimidin-5-yl)phenylthio}lincomycin (75)

Compound **74** (117.9 mg, 0.18 mmol) and 2,2,2-trifluoroacetic acid (0.27 ml) in CH_2Cl_2 (2.5 ml) were treated at − 20 °C for 20 min, and then treated room temperature for 5 h according to the similar procedure as described for the preparation of **73** to afford **75** (82.2 mg, 82.1%) as a colorless solid. $[\alpha]_D^{25}$ +91.7° (*c* 0.33, MeOH); ESI-MS *m/z* 563 $(M+H)^+$ as $C_{27}H_{38}N_4O_5S_2$; TOF-ESI-HRMS $(M+H)^+$ calcd. for $C_{27}H_{38}N_4O_5S_2$: 563.2362, found: 563.2356; ^1H NMR (400 MHz, CD_3OD) δ 0.78–0.88 (m, 3H), 1.20–1.32 (m, 4H), 1.25 (d, *J* = 6.9 Hz, 3H), 1.68–1.78 (m, 1H), 1.86 (s, 3H), 1.92–2.05 (m, 2H), 2.55 (dd, *J* = 10.2, 8.0 Hz, 1H), 3.12 (dd, *J* = 10.2, 6.7 Hz, 1H), 3.48 (dd, *J* = 10.2, 3.3 Hz, 1H), 3.69 (br dd, *J* = 3.3, 0.7 Hz, 1H), 3.75 (dd, *J* = 9.1, 3.4 Hz, 1H), 3.87 (dq, *J* = 6.9, 2.6 Hz, 1H), 4.01 (dd, *J* = 10.2, 5.5 Hz, 1H), 4.27–4.32 (m, 1H), 4.43 (dd, *J* = 9.9, 2.6 Hz, 1H), 5.18 (d, *J* = 5.5 Hz, 1H), 7.43–7.50 (m, 2H), 7.55–7.65 (m, 2H), 8.97 (s, 2H), 9.03 (s, 1H).

In vitro antibacterial activity

MIC (μg ml^{-1}) was determined by the agar dilution method, which was described in Clinical and Laboratory Standards Institute (M07-06 in 2003). Test strains of *S. pneumoniae* and *S. pyogenes* were subjected to seed culture using brain heart infusion agar (Becton Dickinson and Company, Tokyo, Japan) and 5% defibrinated horse blood. Test strains of *H. influenzae* were subjected to seed culture using sensitivity disk agar-N 'Nissui' (Nissui, Tokyo, Japan), 5% defibrinated horse blood, 5 μg ml^{-1} Hemin and 15 μg ml^{-1} nicotinamide adenine dinucleotide. A 5 μl portion of cell suspension of the test strains having about 10^6 colony-forming unit per ml was inoculated into sensitivity disk agar-N 'Nissui' supplemented with 5% defibrinated horse blood, 5 μg ml^{-1} Hemin and 15 μg ml^{-1} nicotinamide adenine dinucleotide, and incubated at 37 °C for 18–22 h. Then, the MIC was measured.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

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