HETEROCYCLES, Vol. 90, No. 2, 2015, pp. 1179 - 1195. © 2015 The Japan Institute of Heterocyclic Chemistry
Received, 28th July, 2014, Accepted, 14th August, 2014, Published online, 20th August, 2014
DOI: 10.3987/COM-14-S(K)93

TOTAL SYNTHESIS OF MAREMYCINS A AND D_1 USING CHIRAL AND CYCLIC NITRONE WITH (*E*)-3-ETHYLIDENE-1-METHYLINDOLIN-2-ONE

Tohru Ueda, Mitsuhide Inada, Nobuyoshi Morita, and Osamu Tamura*

Showa Pharmaceutical University, Machida, Tokyo 194-8543, Japan
tamura@ac.shoyaku.ac.jp

This paper is dedicated to Prof. Dr. Isao Kuwajima, Professor Emeritus of Tokyo Institute of Technology, on the occasion of his 77th birthday.

Abstract – Total syntheses of maremycin A (**4**) and maremycin D_1 (**8**) were described, featuring 1,3-dipolar cycloaddition of a chiral cyclic nitrone **15** with (*E*)-3-ethylidene-1-methylindolin-2-one (**13**). The cycloaddition was reversible, especially at high temperature in the presence of a Lewis acid or in a solvent possessing a high acceptor number. One of the cycloadducts was efficiently led to maremycin A (**4**) and maremycin D_1 (**8**). High optical purity of **4** was confirmed by chiral HPLC comparison with *ent*-**4** prepared from *ent*-**15** and **13**.

INTRODUCTION

β-Methyltryptophan (**1**) and related structures are important components of many natural products, in which both stereochemistries of the methyl group can be found. For example, indolmycin (**2**) and FR900452 (**3**) possess opposite stereochemistry (Figure 1).[1]

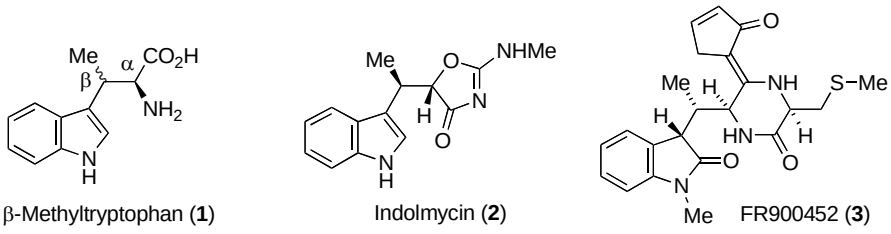


Figure 1. β-Methyltryptophan and related natural products

Maremycins A (**4**) and B (**5**) are diketopiperazine alkaloids that also include β-methyltryptophan featuring 3-hydroxyindolin-2-one[2] structure, and their stereochemistries were tentatively proposed to be as depicted in Figure 2 on the basis of molecular mechanics calculations and spectroscopic data.[3] An inseparable 1:1 mixture of maremycins C_1 (**6**) and C_2 (**7**) was obtained from *Streptomyces* sp. GT 051237, together with an inseparable 3:1 mixture of maremycins D_1 (**8**) and D_2 (**9**).[4] The stereochemistries of **6** and **7** were deduced by comparison of the NMR spectroscopic data with those of **5**, while the stereostructures of **8** and **9** were assigned by comparison of the spectra with those of **4** and **5**. Therefore, confirmation of the stereostructures of maremycins A (**4**) and B (**5**) is important to establish the stereochemistries of maremycin family members. Several years ago, we accomplished the first synthesis of maremycins A (**4**) and D_1 (**8**) and confirmed their stereostructures to be as depicted in Figure 2.[5] Since then, several total syntheses of maremycins have been reported.[6] We now present a full account of our syntheses of **4** and **8**, together with mechanistic considerations, a synthesis of the enantiomer of **4**, and determination of the optical purity of the compounds.

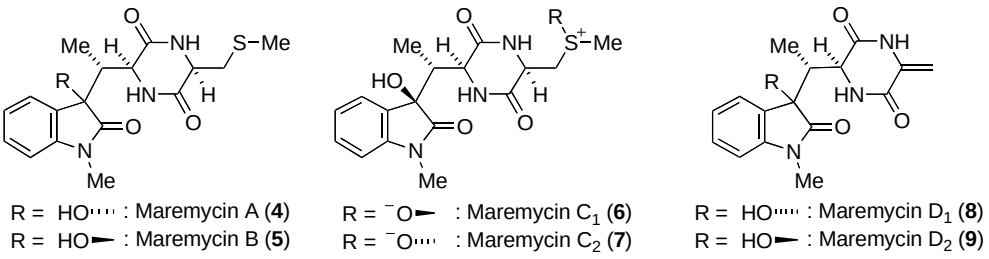
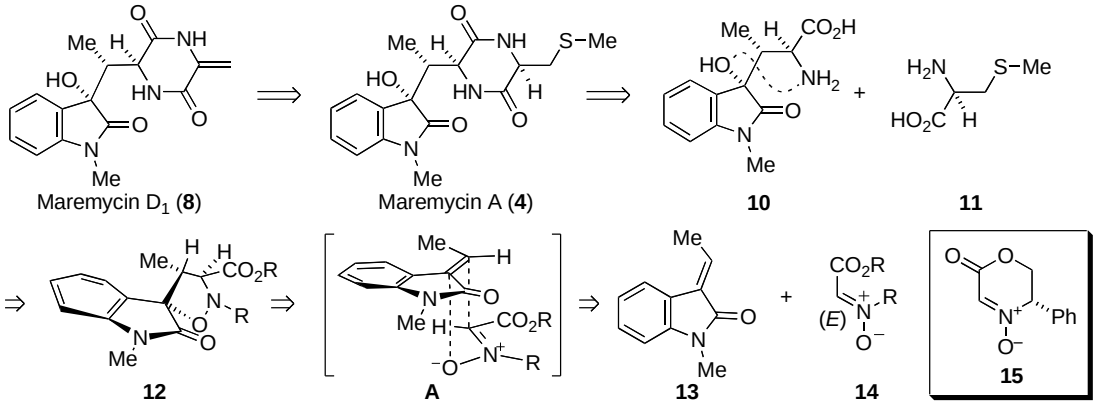


Figure 2. Structures of maremycins

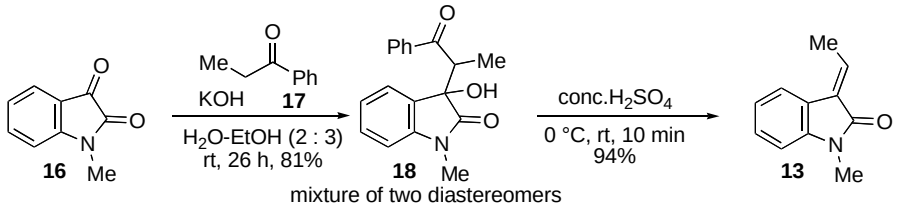
RESULTS AND DISCUSSION

The retrosynthetic analysis of maremycins A (**4**) and D_1 (**8**) is shown in Scheme 1. The exomethylene of **8** can be obtained by elimination of methylsulfide of **4** via its sulfoxide. The diketopiperazine of **4** is constructed from two amino acids, β-methyl amino acid **10** and *S*-methyl-L-cysteine (**11**). Connection of oxygen and the amino group in amino acid **10** would afford spiro-isoxazolidine **12**, which can be obtained by cycloaddition of 3-ethylidene-indolinone **13** with (*E*)-*C*-alkoxycarbonyl nitrone **14** via transition state **A**.[7-9] As an equivalent of **14**, we chose chiral cyclic nitrone **15**.[10-12]

The synthesis began with preparation of indolinone **13** according to a known method (Scheme 2).[13] Aldol reaction of *N*-methylisatin (**16**) with propiophenone (**17**) in the presence of potassium hydroxide gave a 1:1 diastereomeric mixture of aldol adducts **18**. Without separation, the aldol adducts **18** were treated with sulfuric acid to afford (*E*)-3-ethylidene-1-methylindolin-2-one (**13**).

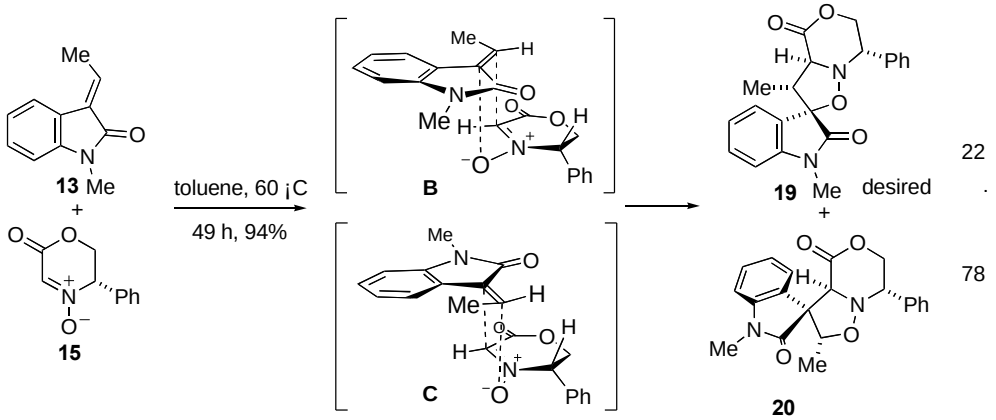


Scheme 1. Retrosynthetic analysis of maremycins A (**4**) and D_1 (**8**)



Scheme 2. Preparation of (*E*)-3-ethylidene-1-methylindolin-2-one (**13**)

The crucial cycloaddition of ethylidene-indolinone **13** with nitrone **15** was next examined. Heating a solution of indolinone **13** (1 equiv) and **15** nitrone (1 equiv) in toluene at 60 °C for two days resulted in cycloaddition reaction to afford a 22:78 mixture of **19** and **20** in 94% yield (Scheme 3).



Scheme 3. Cycloaddition of indolinone **13** with nitrone **15** in toluene

The structure of **19** was established by X-ray crystallography after recrystallization from hexane-AcOEt, whereas that of **20** was deduced on the basis of detailed NMR studies, including NOE experiments (Figure 3). The crystallography of **19** revealed that the minor product **19** was the desired cycloadduct

possessing the required three contiguous stereogenic centers for preparation of non-proteinogenic amino acid **10**, and indicated that **19** was formed via transition state **B**. The NMR studies of **20** showed that the major compound was not the stereoisomer of **19**, surprisingly, but a regio isomer of **19** generated via transition state **C**. It should be noted that both cycloadducts were formed by endo-addition from the less hindered face of nitrone **15**.

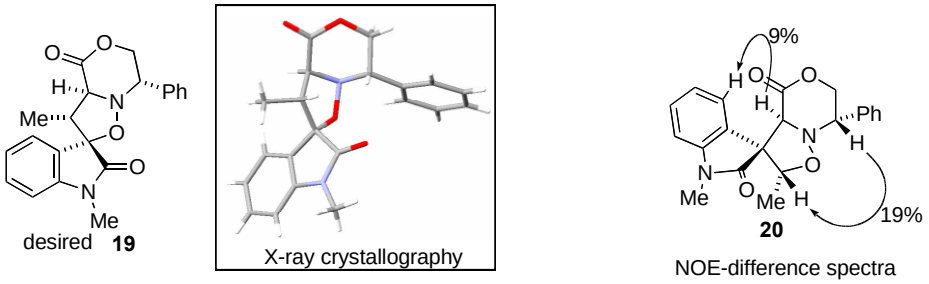


Figure 3. Structure determination of cycloadducts **19** and **20**

Unfortunately, the major product was the undesired regio isomer **20**. In order to improved the yield of **19**, we next examined the use of a Lewis acid such as $MgBr_2$•OEt_2,[14] $ZnBr_2$,[15] $Zn(OTf)_2$,[16] $Cu(OTf)_2$,[17] or $Ti(O^iPr)_2Cl_2$.[18] However, undesired isomer **20** was always obtained predominantly (**19**:**20** = 33:67 – 4:96), although the total yield of the cycloadducts **19** and **20** was very high (94-100%). For example, in the case of $Ti(O^iPr)_2Cl_2$, a 4:96 mixture of **19** and **20** was obtained with 100% conversion.

To shorten the reaction time, neat conditions at various reaction temperatures were investigated (Table 1, entries 1-3). Unexpectedly, it was found that higher reaction temperature gave a higher ratio of cycloadduct **20**. Thus, reaction of nitrone **15** with **13** at -25 °C afforded a 47:53 mixture of **19** and **20** (entry 1), whereas reaction at 60 °C gave a 22:78 mixture (entry 3). Since a kinetically controlled reaction should exhibit a higher ratio at lower temperature, these results imply that the present cycloaddition at high temperature might involve a thermodynamically controlled equilibrium between **19** and **20** via cycloreversion[19] of the cycloadducts **19** and **20** to the starting **13** and **15**. The effect of solvent was next examined (entries 4-9). When polar solvents such as methanol and acetonitrile were employed, cycloadduct **20** was predominant (entries 4 and 5). Use of tetrahydrofuran resulted in equal formation of **19** and **20** (entry 6). Since the reactions in entries 4-6 did not go to completion, 3 equiv. of **13** was used for the reaction at 50 °C (entry 7). Interestingly, cycloaddition of **13** and **15** smoothly proceeded in hexane, giving rise to a 1:1 mixture of cycloadducts **19** and **20** (entry 8). In hexane, elevated temperature did not change the ratio of the cycloadducts (entry 9). Since the synthesis of maremycins would require large-scale reaction, neat conditions (entry 2) appeared to be convenient in terms of the distribution of products as well as the solubility of the starting materials in hexane. In fact, reaction of **13** and **15** on a 30 mmol scale under neat conditions furnished a 48:52 mixture of **19** and **20**.

Next, the possibility of cycloreversion of cycloadducts **19** and **20** to the starting dipolarophile **13** and nitrone **15** was investigated. We previously found that nitrone **15** reacted with 10 equiv of methyl methacrylate (**21**) to exclusively afford cycloadduct **22** in quantitative yield (Scheme 4).[11] Cycloadduct **19**, on heating with methacrylate **21** in toluene at 100 °C for 1 h, released 3-ethylidene-indolin-2-one **13** and afforded cycloadduct **22** of methacrylate **21** in quantitative yield. Similar treatment of cycloadduct **20** with **21** provided 3-ethylidene-indolin-2-one **13** (45%) and cycloadduct **22** (45%), along with recovery of **20** (55%).

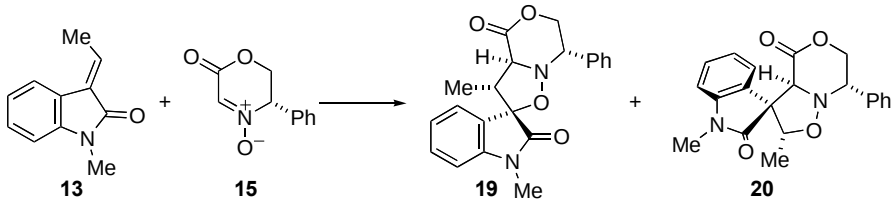
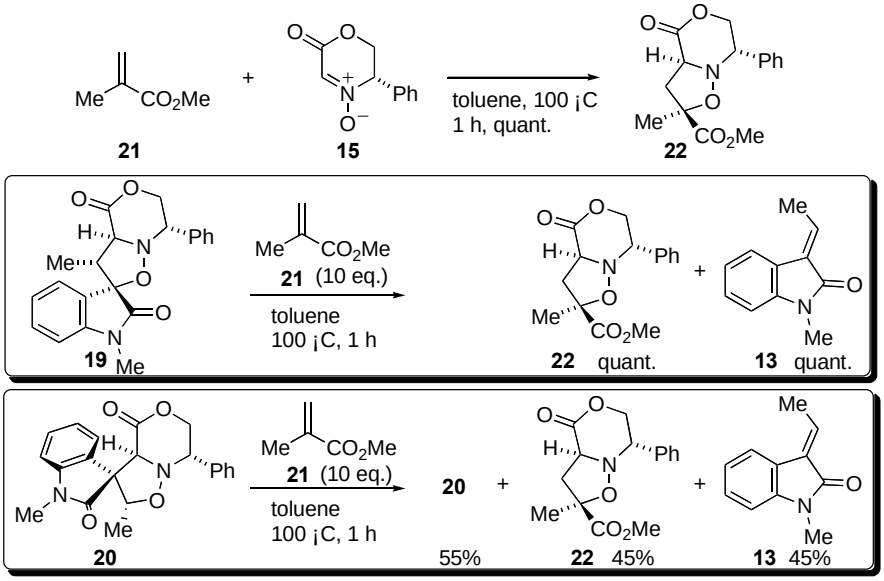


Table 1. Cycloaddition of **13** with **15**

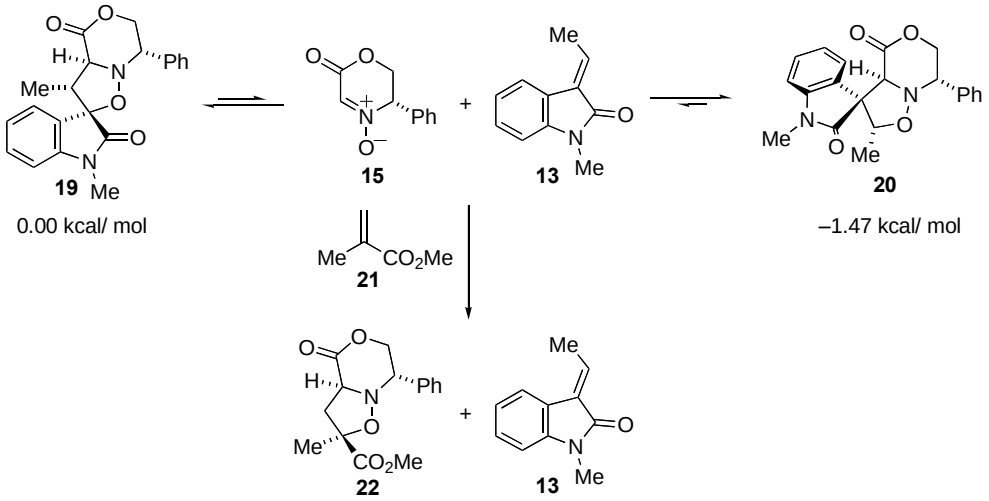
entry	solvent	ε[c]	AN[d]	temp. (¡C)	time (h)	yield (%)[e]	**19**: **20**[e]
1[a]	neat	—	—	-25	11	40	47 : 53
2[a]	neat	—	—	rt	11	93	45 : 55
3[a]	neat	—	—	60	11	96	22 : 78
4[a]	MeOH	32.7	41.3	rt	11	55	26 : 74
5[a]	MeCN	38	18.9	rt	11	46	42 : 58
6[a]	THF	7.6	8.0	rt	11	54	50 : 50
7[b]	THF	7.6	8.0	50	24	quant.	47 : 53
8[b]	hexane	1.9	0.0	rt	8	quant.	50 : 50
9[b]	hexane	1.9	0.0	50	2.5	quant.	50 : 50

[a] Nitrone **15** (1 eq.) and ethylidene-indolinone **13** (1 eq.) were used.
[b] Nitrone **15** (1 eq.) and ethylidene-indolinone **13** (3 eq.) were used.
[c] Dielectronic constants.
[d] Acceptor numbers.
[e] The yields and ratios of products **19** and **20** were determined by ^{1}H-NMR spectra.



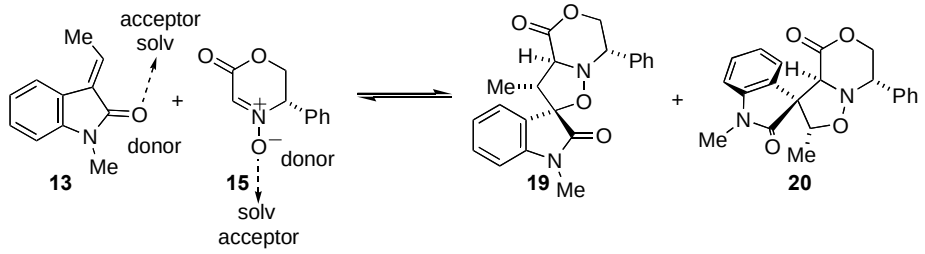
Scheme 4. Crossover experiments of cycloadducts **19** and **20** with methyl methacrylate (**21**)

Formation of **22** from **19** or **20** would involve regeneration of nitrone **15** and indolinone **13**. Thus, heating **19** or **20** would cause cycloreversion to indolinone **13** and nitrone **15**, which, in turn, could undergo re-cycloaddition with methacrylate **21** to provide **22** (Scheme 5). The results shown in Scheme 4 also suggest that cycloadduct **20** may be thermodynamically more stable than cycloadduct **19** because cycloadduct **20** exhibited lower cycloreversion reactivity on heating than did **19**. In fact, 6-31G** calculation indicated that **20** is more stable by 1.47 kcal/mol than **19**. Thus, in the reaction of nitrone **15** and indolinone **13** at low temperature, the reaction rate for the formation of cycloadduct **19** would be as fast as that for **20** (Table 1, entry 1). However, cycloadducts **19** and **20** may exist in equilibrium via the starting nitrone **15** and indolinone **13** at high temperature, and hence formation of more stable **20** is dominant (Table 1, entry 3). We planned to convert regioisomer **20** to the desired cycloadduct **19** via the equilibrium. When a mixture of regioisomer **20** and ethylidene-indolinone **13** was heated in toluene, cycloreversion-re-cycloaddition occurred to give the desired cycloaddition product **19** in 10% yield, accompanied with recovery of regioisomer **20** in 87% yield and ethylidene-indolinone **13** (90%). Thus, the yield of the desired cycloadduct **19** can be improved by recycling this reaction.



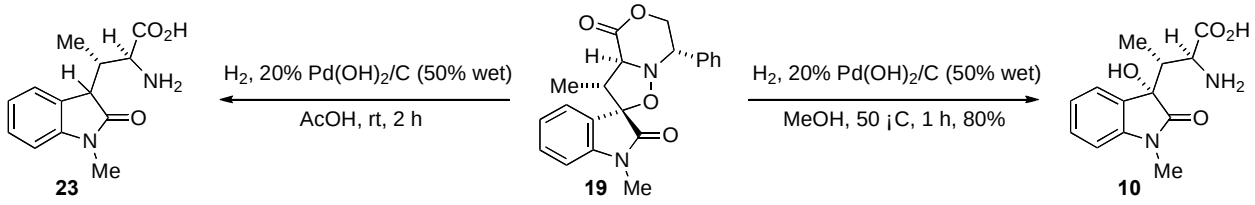
Scheme 5. Equilibrium between **19** and **20** via cycloreversion to **15** and **13**

Table 1 shows dielectronic constants (ε) and acceptor numbers (AN)[20] of solvents used for the cycloaddition. Of the two parameters, AN values seemed to better match the distribution of the two isomers **19** and **20**. Thus, reaction in methanol, having a high AN, afforded a high ratio of cycloadduct **20**, whereas reaction in hexane, with a low AN (0), gave a 1:1 ratio of cycloadducts **19** and **20**. In a solvent of high AN, donating groups of the starting materials **13** and **15** would interact with the solvent to stabilize the left side of the equilibrium, favoring cycloreversion to afford the more stable cycloadduct **20** as the major product through the equilibrium (Scheme 6). Use of a Lewis acid would also stabilize the left side of the equilibrium to yield **20** predominantly.



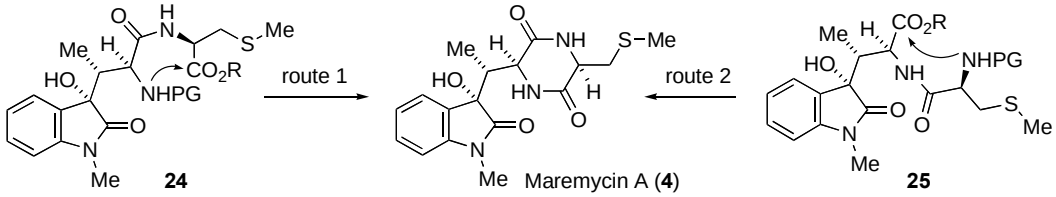
Scheme 6. Interaction of **13** or **15** with a solvent having high AN

The next task was hydrogenolysis of cycloadduct **19** with retention of the hydroxyl group at the benzylic position (Scheme 7). Stirring a mixture of **19** and 20%$Pd(OH)_2$ in acetic acid under an atmosphere of hydrogen at room temperature to give amino acid **23** with loss of the benzylic hydroxy group. After experimentation, we finally found that hydrogenolysis in methanol at 50 °C furnished the desired amino acid **10**.



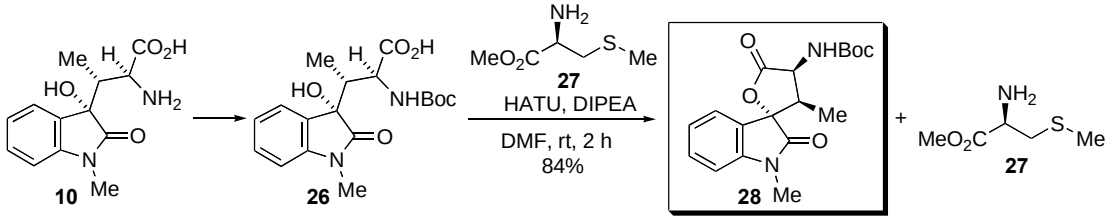
Scheme 7. Hydrogenolysis of **19**

To construct the diketopiperazine framework of maremycin A, two routes could be considered (Scheme 8): one is the route via amino acid **24** (route 1) and the other is that via **25** (route 2).



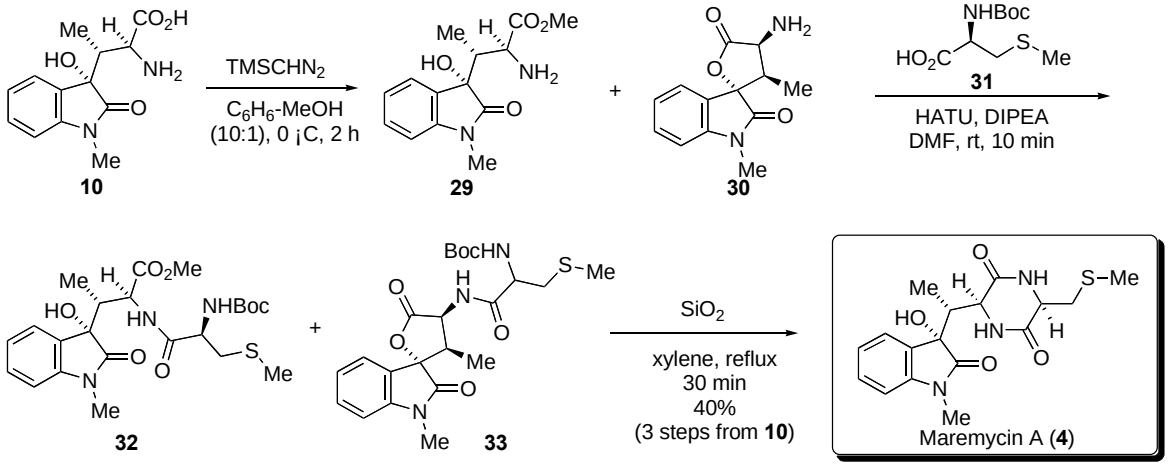
Scheme 8. Possible two routes to maremycin A (**4**) from amino acid **10**

We first examined route 1. Treatment of *N*-protected amino acid **26** derived from **10** with HATU and *S*-methylcysteine methyl ester (**27**)[21] in the presence of DIPEA unfortunately resulted in cyclization to gave lactone **28** in place of the desired dipeptide **24** (PG = Boc, R = Me).



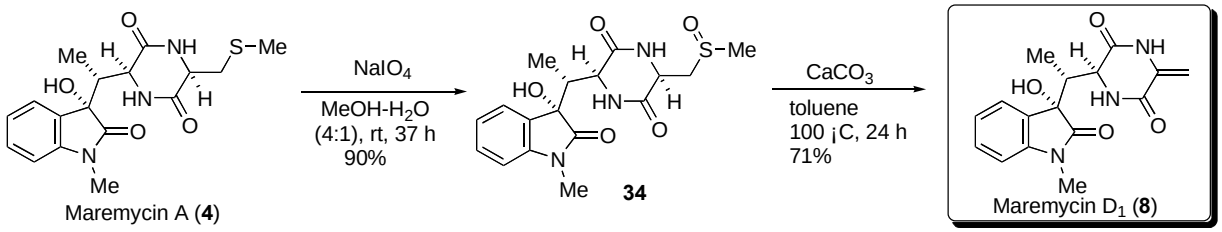
Scheme 9. Attempt to prepare amino acid **24**

Next, route 2 was investigated (Scheme 10). Amino acid was exposed to trimethylsilyldiazomethane to yield ester **29** along with a small amount of lactone **30**. Without separation, **29** and **30** were condensed with *S*-methylcysteine derivative **31**[22] to afford a mixture **32** and **33**. Synthesis of maremycin A (**4**) was accomplished in 40% yield from amino acid **10** by heating of the mixture of **32** and **33** with silica gel in refluxing xylene via removal of the Boc group, followed by formation of the diketopiperazine ring.



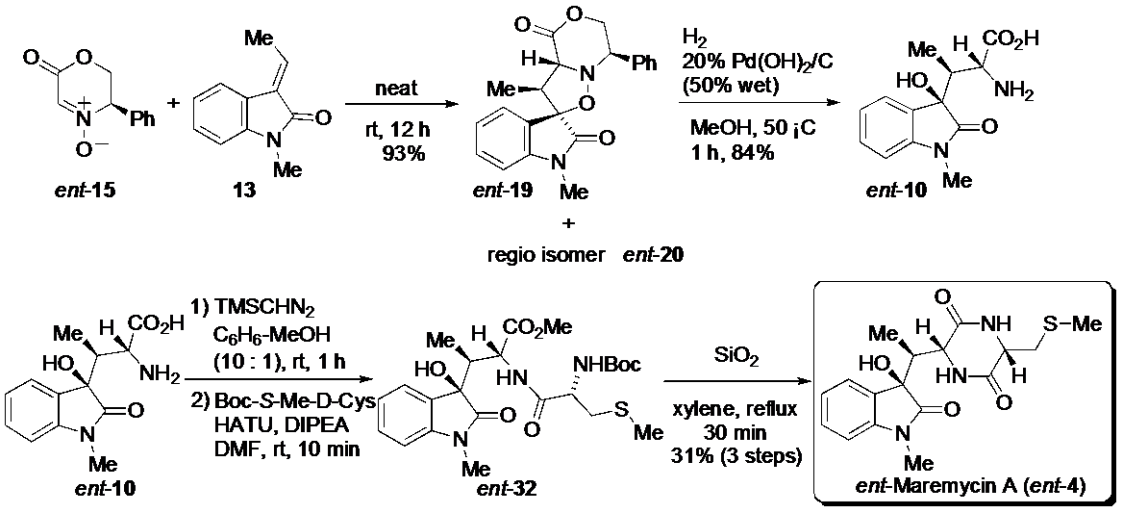
Scheme 10. Synthesis of maremycin A

Fortunately, transformation from maremycin A (**4**) to maremycin D_1 (**8**) could be conducted without difficulty (Scheme 11). The sulfide group of **4** was oxidized with sodium periodate to give sulfoxide **34** as a mixture of diastereomers, which, on heating in toluene with calcium carbonate, underwent *syn*-elimination to afford maremycin D_1 (**8**).



Scheme 11. Synthesis of maremycin D_1 (**8**)

The final task was confirmation of the optical purity of the synthesized maremycin. For this purpose, we synthesized *ent*-maremycin A (*ent*-**4**) from nitrone *ent*-**15**, indolinone **13**, and *S*-methyl-D-cysteine according to the method used for the synthesis of natural **4** (Scheme 12). With both enantiomers of maremycin A in hand, chiral HPLC analysis was conducted. The analyses of racemates prepared from both enantiomers, compound **4**, and *ent*-**4** clearly showed that both enantiomers had an optical purity of >99 %ee.



Scheme 12. Synthesis of *ent*-maremycin A (*ent*-**4**)

In conclusion, we have accomplished the first synthesis of maremycins A (**4**) and D_1 (**8**), featuring cycloaddition of cyclic nitrone **15** with (*E*)-3-ethylidene-1-methylindolin-2-one (**13**). This also conclusively determined the stereochemistries of the natural products **4** and **8**. In addition, **8** was fully characterized for the first time. Mechanistically, it was demonstrated that cycloreversion of the cycloadducts **19** and **20** influenced the distribution of **19** and **20**, as did the acceptor number of the solvent used for the cycloaddition. This short-step synthesis clearly shows the usefulness of nitrone cycloaddition of **15** in the synthesis of natural products containing unusual amino acids.

EXPERIMENTAL

General: Melting points are uncorrected. Infrared (IR) spectra were recorded with a Shimadzu FTIR-8200A spectrometer, and ^{1}H NMR and ^{13}C NMR spectra were recorded with JEOL JNM-AL300 spectrometer, with tetramethylsilane as an internal standard ($CDCl_3$ solution). Chemical shifts are recorded in ppm, and coupling constants (*J*) in Hz. Mass spectra were recorded on JEOL JMS-D300 and HX110 spectrometers. Elemental analyses were performed on an Amco Flash EA 1112 instrument. Merck silica gel 60 (1.09385) and Merck silica gel 60 F254 were used for column chromatography and

thin layer chromatography (TLC), respectively.

3-Hydroxy-1-methyl-3-(1-methyl-2-oxo-2-phenylethyl)-1,3-dihydro-2*H*-indol-2-one (18)

Potassium hydroxide (372 mg, 6.6 mmol) was dissolved in H_2O-EtOH (2:3, 200 mL). *N*-Methylisatin (**16**) (9.88 g, 59.5 mmol) and propiophenone (**17**) (12.0 mL, 90.2 mmol) were added to the KOH solution (140 mL, 4.6 mmol) prepared above. The mixture was stirred at room temperature for 6 h, and then the mixture was concentrated under reduced pressure. The residue was chromatographed on silica gel with CH_2Cl_2-AcOEt (5:1) to give **18** (14.5 g, 81%) as a ca 3:1 mixture of diastereomers. This material was used for the next step without further purification.

Major diastereomer: ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.26 (3H, d, *J* = 7.1 Hz), 3.20 (3H, s), 4.02 (1H, q, *J* = 7.1 Hz), 4.34 (1H, s), 6.84 (1H, br d, *J* = 7.9 Hz), 7.09 (1H, td, *J* = 7.7, 0.9 Hz), 7.34 (1H, td, *J* = 7.7, 1.3 Hz), 7.42-7.51 (2H, m), 7.54-7.62 (2H, m), 7.93-7.99 (2H, m).

Minor diastereomer: ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.60 (3H, d, *J* = 6.8 Hz), 3.13 (3H, s), 3.93 (1H, q, *J* = 7.2 Hz), 4.15 (1H, s), 6.82 (1H, dd, *J* = 8.2, 0.5 Hz), 6.98 (1H, td, *J* = 7.5, 0.9 Hz), 7.26-7.34 (2H, m), 7.38-7.46 (2H, m), 7.55 (1H, tt, *J* = 7.3 1.5 Hz), 7.77-7.83 (2H, m).

(3*E*)-3-Ethylidene-1-methyl-1,3-dihydro-2*H*-indol-2-one (13)

Compound **18** (14.5 g, 49.2 mmol) was added to concentrated H_2SO_4 (400 mL) at 0 °C, and then the mixture was stirred at room temperature for 10 min. The mixture was poured into ice water, and the whole was extracted with $CHCl_3$. The combined organic phases were washed with a saturated aqueous solution of $NaHCO_3$ and brine, and dried over $MgSO_4$. The solvent was removed by rotary evaporation and the crude product was purified by column chromatography on silica gel with hexane-AcOEt (3:1) to give **13** (7.97 g, 94%) as a yellow solid. ^{1}H NMR (300 MHz, $CDCl_3$) δ 2.28 (3H, d, *J* = 7.6 Hz), 3.24 (3H, s), 6.83 (1H, br d, *J* = 7.8 Hz), 7.05 (1H, td, *J* = 7.6, 1.0 Hz), 7.13 (1H, q, *J* = 7.6 Hz), 7.28 (1H, td, *J* = 7.8, 1.0 Hz), 7.58 (1H, br d, *J* = 7.6 Hz). Spectral data were identical with the literature values.[13]

(3*S*,3'*R*,3a'*S*,7'*S*)-1,3'-Dimethyl-7'-phenyl-6',7'-dihydro-3'*H*-spiro[indole-3,2'-isoxazolo[3,2-*c*][1,4]-oxazine]-2,4'(1*H*,3a'*H*)-dione (19), (2'*R*,3*S*,3a'*S*,7'*S*)-1,2'-dimethyl-7'-phenyl-6',7'-dihydrospiro[indole-3,3'-isoxazolo[3,2-*c*][1,4]oxazine]-2,4'(1*H*,3a'*H*)-dione (20) and their enantiomers *ent*-19 and *ent*-20

(a) A solution of nitrone **15** (99.5 mg, 0.521 mmol) and **13** (90.0 mg, 0.520 mmol) in toluene (5 mL) was heated at 60 °C for 49 h and then concentrated under reduced pressure to give a 22:78 mixture (181 mg, 94%). Chromatography on silica gel (CH_2Cl_2-MeCN, 20 : 1) gave **19** and **20**.

19: mp 142.5-143.5 °C (hexane-AcOEt); $[\alpha]_D^{27}$ +125.2 (*c* 1.00, $CHCl_3$); IR (KBr) 3444, 1746, 1736, 1614, 1497, 1472, 1458, 1375, 1352, 1298, 1227, 1043, 768, 760, 696 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$) δ 1.07 (3H, d, *J* = 7.0 Hz), 3.13 (3H, s), 3.44 (1H, dq, *J* = 11.7, 7.0 Hz), 3.89-4.26 (3H, m), 4.97 (1H, dd, *J* = 9.7, 5.1 Hz), 6.76 (1H, d, *J* = 7.9 Hz), 7.06 (1H, td *J* = 7.6, 0.9 Hz), 7.24-7.39 (5H, m), 7.55 (2H, d, *J* = 6.8 Hz), ^1H NMR (300 MHz, C_6D_6) δ 0.91 (3H, d, *J* = 6.9 Hz), 2.26 (3H, s), 3.38 (1H, dq, *J* = 11.6, 6.8 Hz), 3.75 (1H, dd, *J* = 11.6, 4.1 Hz), 3.83 (1H, dd, *J* = 11.5, 10.8 Hz), 4.14 (1H, d, *J* = 11.9 Hz), 4.97 (1H, dd, *J* = 10.8, 4.0 Hz), 6.00 (1H, d, *J* = 7.7 Hz), 6.72 (1H, td, *J* = 7.5, 1.0 Hz), 6.85 (1H, d, *J* = 7.2 Hz), 6.90 (1H, dd, *J* = 7.7, 1.4 Hz), 7.03 (1H, dt, *J* = 6.1, 1.4 Hz) 7.58-7.64 (2H, m); ^{13}C NMR (75 MHz, $CDCl_3$) δ 13.3, 26.7, 49.0, 62.5, 70.2, 70.6, 87.7, 108.7, 123.0, 125.5, 125.8, 128.3, 128.5, 128.7, 130.4, 135.3, 143.8, 167.6, 175.8; HRMS *m/z* calcd for $C_{21}H_{20}N_2O_4$, 364.1423; found, 364.1415. *Anal.* Calcd for $C_{21}H_{20}N_2O_4$: C, 69.22; H, 5.53; N, 7.69. Found: C, 69.49; H, 5.50; N, 7.58.

20: mp 136.0-140.0 °C (hexane-AcOEt); $[\alpha]_D^{24}$ +160.2 (*c* 1.00, $CHCl_3$); IR (KBr) 3442, 1751, 1713, 1612, 1495, 1472, 1458, 1375, 1231, 1094, 756 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$) δ 0.92 (3H, d, *J* = 6.2 Hz), 3.25 (3H, s), 4.27 (1H, dd, *J* = 11.4, 10.6 Hz), 4.43 (1H, dd, *J* = 11.5, 3.4 Hz), 4.71 (1H, q, *J* = 6.2 Hz), 4.82 (1H, s), 5.19 (1H, dd, *J* = 10.5, 3.3 Hz), 6.91 (1H, dd, *J* = 8.3, 1.0 Hz), 7.15 (1H, td, *J* = 7.6, 1.0 Hz), 7.32-7.47 (5H, m), 7.51-7.59 (2H, m), ^{13}C NMR (75 MHz, $CDCl_3$) δ 14.3, 26.7, 58.7, 67.5, 71.1, 74.7, 79.9, 109.0, 123.0, 124.4, 127.9, 128.2, 128.7, 128.9, 129.2, 134.9, 143.6, 167.1, 176.0; HRMS *m/z* calcd for $C_{21}H_{20}N_2O_4$, 364.1423; found, 364.1423.

(b) Nitrone **15** (6.07 g, 31.8 mmol) and **13** (5.50 g, 31.8 mmol) were dissolved in CH_2Cl_2 (10 mL). The mixture was concentrated under reduced pressure below 10 °C, and the residue was kept room temperature for 33 h. The ^1H NMR spectrum of the mixture showed a 48:52 mixture of **19** and **20**. Flash chromatography (CH_2Cl_2-MeCN, 20:1) afforded pure **19** and **20**, whose ^1H NMR spectra were identical with those obtained in (a).

(c) A solution of nitrone **15** (3.0 mg, 0.016 mmol) and **13** (8.2 mg, 0.047 mmol) in hexane (0.5 mL) was heated at 50 °C for 2.5 h, then concentrated. The ^1H NMR spectrum of the residue showed completion of the reaction and formation of a 1:1 mixture of **19** and **20**.

(d) According to procedure (b), reaction of *ent*-**15** (507 mg, 2.66 mmol) with **13** (503 mg, 2.90 mg) afforded a 47:53 mixture of *ent*-**19** and *ent*-**20** (901 mg, 93%). Pure samples were obtained by chromatography.

ent-**19**: mp 141.5-143.0 °C (hexane-AcOEt); $[\alpha]_D^{27}$ = -127.7 (c 1.0, $CHCl_3$).

ent-**20**: mp 135.0-139.0 °C (hexane-AcOEt); $[\alpha]_D^{27}$ = -156.5 (c 1.0, $CHCl_3$).

Transformation of 20 to 19

A solution of **20** (25.0 mg, 0.0686 mmol) and **13** (35.7 mg, 0.206 mmol) in toluene (1.0 mL) was heated

at 100 °C for 1 h. After concentration, the mixture was chromatographed to give **19** (2.6 mg, 10%) along with recovery of **20** (21.8 mg, 87%) and **13** (32.2 mg, 90%).

(2*S*,3*R*)-2-Amino-3-[(3*S*)-3-hydroxy-1-methyl-2-oxo-2,3-dihydro-1*H*-indol-3-yl]butanoic acid (10) and its enantiomer *ent*-10

(a) A mixture of **19** (1.00 g, 2.74 mmol) and 20% $Pd(OH)_2/C$ (50% wet, 1.00 g) in MeOH (200 mL) was stirred at 50 °C under an atmosphere of hydrogen for 1 h. The mixture was filtered and the filtrate was concentrated under reduced pressure to give a residue, which was triturated with CH_2Cl_2 to afford **10** (580 mg, 80%) as a colorless powder. mp 159.5-160.0 °C; $[\alpha]_D^{28}$ -32.8 (*c* 0.20, MeOH); IR (KBr) 3446, 3128, 3059, 2975, 2935, 2890, 2361, 1709, 1614, 1500, 1495, 1420, 1377, 1354, 1323, 1261, 1219, 1124, 1097, 1086, 758, 692, 540 cm^{-1}; ^1H NMR (300 MHz, CD_3OD) δ 0.91 (3H, d, *J* = 7.1 Hz), 2.25 (1H, quin, *J* = 7.0 Hz), 3.20 (3H, s), 4.41 (1H, d, *J* = 6.0 Hz), 7.03 (1H, d, *J* = 7.7 Hz), 7.15 (1H, td *J* = 7.5, 0.7 Hz), 7.40 (1H, td, *J* = 7.7, 1.1 Hz), 7.51 (1H, d, *J* = 7.5 Hz), ^{13}C NMR (75 MHz, CD_3OD) δ 11.2, 26.5, 41.4, 56.5, 79.7, 110.1, 124.1, 126.3, 130.6, 131.1, 144.4, 173.1, 179.9, HRMS *m/z* calcd for $C_{13}H_{16}N_2O_4$, 264.1110, found, 264.1093.

(b) According to procedure (a), hydrogenolysis of *ent*-**19** (300 mg, 0.824 mmol) with 20% $Pd(OH)_2/C$ (50% wet, 301 mg) in MeOH (100 mL) afforded *ent*-**10** (184 mg, 84%). mp 160.0-160.5 °C; $[\alpha]_D^{23}$ -33.0 (*c* 0.20, MeOH).

Methyl (2*S*,3*R*)-2-Amino-3-[(3*S*)-3-hydroxy-1-methyl-2-oxo-2,3-dihydro-1*H*-indol-3-yl]butanoate (29) and lactone 30, their enantiomers *ent*-29 and *ent*-30

(a) To a stirred solution of **10** (400 mg, 1.52 mmol) in benzene-MeOH (10:1, 15 mL) was added a 2.0 M solution of $TMSCHN_2$ in hexane (7.5 mL, 15 mmol) at 0 °C, and the mixture was stirred at the same temperature for 2 h. The mixture was concentrated under reduced pressure, and then the residue was diluted with CH_2Cl_2, washed successively with water and brine, dried (Na_2SO_4), and concentrated to give **29** (420 mg, quant) that contained a small amount of **30**.

29: ^1H NMR (300 MHz, $CDCl_3$) δ 0.51 (3H, d, *J* = 7.0 Hz), 2.52 (1H, dq, *J* = 10.3, 6.6 Hz), 3.18 (3H, s), 3.72 (3H, s), 3.89 (1H, d, *J* = 10.3 Hz), 6.83 (1H, d, *J* = 7.7 Hz), 7.10 (1H, t, *J* = 7.3 Hz), 7.2-7.4 (2H, m).

30: ^1H NMR (300 MHz, $CDCl_3$) δ 1.13 (3H, d, *J* = 7.3 Hz), 2.87 (1H, quin, *J* = 7.7 Hz), 3.20 (3H, s), 4.87 (1H, d, *J* = 7.7 Hz), 6.87 (1H, dd, *J* = 8.3, 0.9 Hz), 7.11 (1H, td, *J* = 7.6, 0.7 Hz), 7.37-7.45 (2H, m).

This material was used for the next step without further purification.

(b) According to a procedure similar to that described in (a), *ent*-**29** (166 mg, 98%) containing a small amount of *ent*-**30** was obtained from *ent*-**10** (162 mg, 0.613 mg).

This material was used for the next step without further purification.

(2*S*,3*R*)-2-{[*N*-(*tert*-Butoxycarbonyl)-*S*-methyl-L-cysteinyl]amino}-3-[(3*S*)-3-hydroxy-1-methyl-2-oxo-2,3-dihydro-1*H*-indol-3-yl] butanoate 32 and lactone 33, their enantiomers *ent*-32 and *ent*-33

(a) 2-(1*H*-7-Azabenzotriazol-1-yl)-1,1,3,3-tetramethyluronium hexafluorophosphate methanaminium (HATU, 651 mg, 1.66 mmol) was added to a stirred solution of **29** prepared above (418 mg, 1.50 mmol), **31** (394 mg, 1.65 mmol), and diisopropylethylamine (0.30 mL, 1.74 mmol) in DMF (15 mL) at 0 °C, and the mixture was stirred at room temperature for 10 min. After concentration, the residue was diluted with CH_2Cl_2, washed with 10% aqueous HCl, a saturated aqueous solution of $NaHCO_3$ and brine, dried (Na_2SO_4), and concentrated to give **32** (691 mg, 92%) containing a small amount of **33** as a syrup.

32: ^{1}H NMR (300 MHz, $CDCl_3$) δ 0.87 (3H, d, *J* = 7.0 Hz), 1.43 (9H, s), 2.17 (3H, s), 2.70 (1H, quin, *J* = 7.2 Hz), 3.19 (3H, s), 3.69 (3H, s), 3.99 (1H, br s), 4.25 (1H, q, *J* = 7.2 Hz), 4.67 (1H, br s), 5.39 (1H, d, *J* = 7.3 Hz), 6.84 (1H, d, *J* = 7.9 Hz), 7.10 (1H, td, *J* = 7.2 Hz), 7.35 (1H, td, *J* = 7.8, 1.2 Hz), 7.47 (1H, br d, *J* = 7.5 Hz).

33: ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.03 (3H, d, *J* = 7.3 Hz), 1.46 (9H, s), 2.18 (3H, s), 2.87 (1H, dd, *J* = 13.8, 6.7 Hz), 2.97 (1H, dd, *J* = 13.9, 6.2 Hz), 3.20 (3H, s), 3.14-3.27 (1H, m), 4.31 (1H, q, *J* = 6.7 Hz), 5.33 (1H, br s), 5.77 (1H, dd, *J* = 7.8, 5.6 Hz), 6.88 (1H, d, *J* = 7.7 Hz), 6.99 (1H, br d, *J* = 5.0 Hz), 7.31 (1H, dq, *J* = 7.5, 0.6 Hz), 7.42 (1H, td, *J* = 7.8, 1.3 Hz).

This material was used for the next step without further purification.

(b) According to a procedure similar to that described in (a), *ent*-**32** (262 mg, quant.) containing a small amount of *ent*-**33** was obtained from *ent*-**29** (157 mg, 0.566 mg) and *ent*-**31** (124 mg, 0.528 mmol).

This material was used for the next step without further purification.

(3*S*,6*R*)-3-{(1*R*)-1-[(3*S*)-3-Hydroxy-1-methyl-2-oxo-2,3-dihydro-1*H*-indol-3-yl]ethyl}-6-[(methylsulfanyl)methyl]piperazine-2,5-dione, maremycin A (4) and its enantiomer *ent*-4

(a) A mixture of **32** prepared above (688 mg, 1.39 mmol) and silica gel (3.45 g) in xylene (20 mL) was heated at reflux for 30 min. The solvent was evaporated, and the residue was subjected to column chromatography on silica gel ($CHCl_3$ then $CHCl_3$-MeOH, 40:1) to give maremycin A (**4**) (220 mg, 40% from **10**) as a colorless solid. mp 229.0-229.5 °C (*lit*.[3] 229 °C); $[\alpha]_D^{25}$ -110.2 (*c* 0.15, MeOH), [*lit*.[3] $[\alpha]_D^{25}$ -120.95 (*c* 0.21, MeOH); IR (KBr) 3433, 2920, 1707, 1670, 1614, 1472, 1458, 1375, 1261, 1213, 1126, 1097, 976, 758, 692, 540 cm^{-1}; ^{1}H NMR (300 MHz, DMSO-*d*$_6$) δ 1.11 (3H, d, *J* = 7.3 Hz), 2.04 (1H, dd, *J* = 7.3, 2.2 Hz), 2.08 (3H, s), 2.79 (1H, dd, *J* = 13.9, 4.0 Hz), 2.97 (1H, dd, *J* = 13.9, 3.7 Hz), 3.09 (3H, s), 4.27 (1H, m), 4.88 (1H, br s), 7.00 (1H, d, *J* = 7.5Hz), 7.04 (1H, ddd, *J* = 7.5, 7.5, 1.0 Hz), 7.29-7.40 (2H, m), 7.59 (1H, br s), 7.92 (1H, br s), 8.64 (1H, br s), ^{13}C NMR (75 MHz, DMSO-*d*$_6$) δ 8.3, 16.4, 26.0, 36.4, 43.1, 53.6, 54.3, 76.4, 108.6, 121.9, 125.1, 129.2, 130.7, 143.1, 165.7, 168.1, 187.1, HRMS *m/z* calcd for $C_{17}H_{21}N_3O_4S$, 363.1253, found, 363.1258. These data were identical with reported

values.[3]

(b) According to a procedure similar to that described in (a), *ent*-**4** (70 mg, 31% from *ent*-**10**) was obtained from *ent*-**32** (252 mg, 0.566 mg) and silica gel (1.26 g).

ent-**4**: mp 225.0-226.0 °C; $[\alpha]_D^{22}$ +99.4 (*c* 0.10, MeOH).

(c) HPLC analysis. Analytical conditions were as follows: column, DAICEL Chiralcel OJ-H; mobile phase, hexane-EtOH (1:5); flow rate, 0.25 mL/min; detector, UV (254 nm). Retention times for racemic **4**: 18.2 min (50%, **4**), 21.7 min (50%, *ent*-**4**). Retention time for **4**: 18.4 min (100%). Retention time for *ent*-**4**: 21.7 min (100%).

(3*S*,6*R*)-3-{(1*R*)-1-[(3*S*)-3-Hydroxy-1-methyl-2-oxo-2,3-dihydro-1*H*-indol-3-yl]ethyl}-6-[(methylsulfinyl)methyl]piperazine-2,5-dione (34)

To a solution of maremycin A (**4**, 46.0 mg, 0.126 mmol) in MeOH (20 mL) was added a solution of $NaIO_4$ (41.3 mg, 0.193 mmol) in water (4.0 mL) at room temperature, and the mixture was stirred at the same temperature for 17 h. To the mixture was added an additional solution of $NaIO_4$ (20.3 mg, 0.093 mmol) in water (2.0 mL), and stirring was continued for 20 h. After concentration under reduced pressure, the crude material was chromatographed on silica gel (CH_2Cl_2-MeOH, 10:1) to give **34** (43.2 mg, 90%) as a 1:1 mixture of diastereomers.

^{1}H NMR (300 MHz, DMSO-d_6) δ 0.95 (3H, d, *J* = 7.0 Hz), 1.00 (3H, d, *J* = 7.1 Hz), 2.17-2.32 (2H, m), 2.62 (6H, s), 2.97-3.29 (5H, m), 3.09 (3H, s), 3.10 (3H, s), 4.42-4.50 (2H, m), 4.56 (1H, br s), 4.61 (1H, br s), 7.00 (2H, br d, *J* = 7.9 Hz), 7.05 (2H, br t, *J* = 8.0 Hz), 7.11 (1H, s), 7.15 (1H, s), 8.09 (1H, s), 8.20 (1H, s), 8.33 (1H, s), 8.56 (1H, s). This material was used for the next step without further purification.

(3*S*)-3-{(1*R*)-1-[(3*S*)-3-Hydroxy-1-methyl-2-oxo-2,3-dihydro-1*H*-indol-3-yl]ethyl}-6-methylidenepiperazine-2,5-dione, maremycin D_1 (8)

A mixture of **34** (40.8 mg, 0.107 mmol) and $CaCO_3$ (11.9 mg, 0.119 mmol) in toluene (2.0 mL) was stirred at 100 °C for 24 h, and then concentrated under reduced pressure. The crude material was chromatographed on silica gel (CH_2Cl_2-MeOH, 10:1) to give recovered maremycin A *S*-oxide (22.3 mg, 55%) and maremycin D_1 (**5**) [10.9 mg, 33.7% (71% based on recovered starting material)], mp 224.0-226.0 °C (decomp.). $[\alpha]_D^{22}$ -40.8 (*c* 0.1, MeOH); IR (KBr) 3310, 3209, 2920, 1708 cm^{-1}; ^{1}H NMR (300 MHz, DMSO-d_6) δ 1.00 (3H, d, *J* = 7.3 Hz), 2.57 (1H, dq, *J* = 7.7, 2.2 Hz), 3.04 (3H, s), 4.14 (1H, br s), 4.65 (1H, s), 5.12 (1H, s), 6.24 (1H, br s), 6.91 (1H, d, *J* = 7.7 Hz), 7.00 (1H, td, *J* = 7.7, 0.7 Hz), 7.29, (1H, td, *J* = 7.7, 1.5 Hz), 7.35 (1H, dt, *J* = 7.4, 0.6 Hz), 8.27 (1H, br s), 10.40 (1H, br s); ^{13}C NMR (75 MHz, DMSO-d_6) δ 10.0, 25.8, 44.4, 56.9, 75.9, 98.8, 108.2, 121.7, 124.9, 129.1, 129.7, 134.4, 143.5, 158.0, 165.2, 176.5; HRMS *m/z* calcd for $C_{16}H_{17}N_3O_4$, 315.1219, found 315.1217. The ^{1}H and ^{13}C NMR data were identical reported values.[4]

ACKNOWLEDGEMENTS

This work was supported in part by "Platform for Drug Discovery, Informatics, and Structural Life Science from the Ministry of Education, Culture, Sports, Science and Technology, Japan," "the Takeda Science Foundation," "Research Foundation for Pharmaceutical Sciences, the Houan-sha," "MEXT-Supported Program for the Strategic Research Foundation at Private Universities (2013-2017)," and "Grants-in-Aid for Scientific Research (C) from JSPS."

REFERENCES AND NOTES

1. Y. Zou, Q. Fang, H. Yin, Z. Liang, D. Kong, L. Bai, Z. Deng, and S. Lin, *Angew. Chem. Int. Ed.*, 2013, **52**, 12951.
2. For a review on 3-substituted-3-hydroxy-2-oxindoles, see: S. Peddibhotla, *Curr. Bioact. Compd.*, 2009, **5**, 20.
3. W. Balk-Bindseil, E. Helmke, H. Weyland, and H. Laatsch, *Liebigs Ann. Chem.*, 1995, 1291.
4. Y. Tang, I. Sattler, R. Thiericke, and S. Grabley, *Eur. J. Org. Chem.*, 2001, 261.
5. T. Ueda, M. Inada, I. Okamoto, N. Morita, and O. Tamura, *Org. Lett.*, 2008, **10**, 2043.
6. G. Bergonzini and P. Melchiorre, *Angew. Chem. Int. Ed.*, 2012, **51**, 971; Y. Liu, L. Zhang, and Y. Jia, *Tetrahedron Lett.*, 2012, **53**, 684; J. Dugal-Tessier, E. A. O'Bryan, T. B. H. Schroeder, D. T. Cohen, and K. A. Scheidt, *Angew. Chem. Int. Ed.*, 2012, **51**, 4963.
7. Unlike azomethine ylides or nitrile oxides, a nitrone rarely undergoes cycloaddition with a 2-oxoindolin-3-ylidene. For known examples, see: R. Raunak, V. Kumar, S. Mukherjee, P. Poonam, A. K. Prasad, C. E. Olsen, S. J. C. Schäffer, S. K. Sharma, A. C. Watterson, W. Erringtone, and V. S. Parmar, *Tetrahedron*, 2005, **61**, 5687; S. Malhotra, S. Balwani, A. Dhawan, Raunak, Y. Kumar, B. K. Singh, C. E. Olsen, A. K. Prasad, V. S. Parmar, and B. Ghosh, *Med. Chem. Commun.*, 2012, **3**, 1536; Y. Shi, A. Lin, H. Mao, Z. Mao, W. Li, H. Hu, C. Zhu, and Y. Cheng, *Chem. Eur. J.* 2013, **19**, 1914.
8. For recent examples of cycloaddition of azomethine ylides with 2-oxoindolin-3-ylidenes, see: L. Shu, Z. Li, C. Gu, and D. Fishlock, *Org. Process Res. Dev.*, 2013, **17**, 247; A. Dandia, A. K. Jain, A. K. Laxkar, and D. S. Bhati, *Tetrahedron*, 2013, **69**, 2062; Y. Arun, G. Bhaskar, C. Balachandran, S. Ignacimuthu, and P. T. Perumal, *Bioorg. Med. Chem. Lett.*, 2013, **23**, 1839; L. Wang, X.-M. Shi, W.-P. Dong, L.-P. Zhu, and R. Wang, *Chem. Commun.*, 2013, **49**, 3458; W. Sun, G. Zhu, C. Wu, G. Li, L. Hong, and R. Wang, *Angew. Chem. Int. Ed.*, 2013, **52**, 8633; J. Day, M. Uroos, R. A. Castledine, W. Lewis, B. McKeever-Abbas, and J. Dowden, *Org. Biomol. Chem.*, 2013, **11**, 6502; J.-A. Xiao, H.-G. Zhang, S. Liang, J.-W. Ren, H. Yang, and X.-Q. Chen, *J. Org. Chem.*, 2013, **78**, 11577; S. Lanka, S. Thennarasu, and P. T. Perumal, *Tetrahedron Lett.*, 2014, **55**, 2585.

9. For selected examples of cycloaddition of azomethine ylides with 2-oxoindolin-3-ylidenes, see: A. Singh and G. P. Roth, *Org. Lett*., 2011, **13**, 2118; A. Singh and G. P. Roth, *Tetrahedron Lett*., 2012, **53**, 4889; A. V. Velikorodov, V. A. Ionova, O. V. Degtyarev, and L. T. Sukhenko, *Pharm. Chem. J.*, 2013, **46**, 715; C. J. A. Ribeiro, J. D. Amaral, C. M. P. Rodrigues, R. Moreira, and M. M. M. Santos, *Bioorg. Med. Chem*., 2014, **22**, 577.
10. O. Tamura, K. Gotanda, R. Terashima, M. Kikuchi, T. Miyawaki, and M. Sakamoto, *Chem. Commun*., 1996, 1861; O. Tamura, T. Kuroki, Y. Sakai, J. Takizawa, J. Yoshino, Y. Morita, N. Mita, K. Gotanda, and M. Sakamoto, *Tetrahedron Lett.*, 1999, **40**, 895; O. Tamura, S. Yoshida, H. Sugita, N. Mita, Y. Uyama, N. Morita, M. Ishiguro, T. K Kawasaki, H. Ishibashi, and M. Sakamoto, *Synlett*, 2000, 1553; O. Tamura, K. Gotanda, J. Yoshino, Y. Morita, R. Terashima, M. Kikuchi, T. Miyawaki, N. Mita, M. Yamashita, H. Ishibashi, and M. Sakamoto, *J. Org. Chem.*, 2000, **65**, 8544; O. Tamura, T. Shiro, A. Toyao, and H. Ishibashi, *Chem. Commun.*, 2003, 2678.
11. O. Tamura, T. Shiro, M. Ogasawara, A. Toyao, and H. Ishibashi, *J. Org. Chem.*, 2005, **70**, 4569.
12. For reviews, see: O. Tamura, *Synth. Org. Chem. Jpn*., 2010, **68**, 1272; O. Tamura, 'Methods and Applications of Cycloaddition Reactions in Organic Synthesis: Geometry-Controlled Cycloaddition of *C*-Alkoxycarbonyl Nitrones: Synthetic Studies on Nonproteinogenic Amino Acids,' ed. by N. Nishiwaki, John Wiley & Sons, Inc., Hoboken, 2014, pp. 151-174.
13. P. López-Alvarado and C. Avendaño, *Synthesis*, 2002, 104.
14. S. Kanemasa, T. Tsuruoka, and E. Wada, *Tetrahedron Lett*., 1993, **34**, 87; S. Kanemasa and T. Tsuruoka, *Chem. Lett*., 1995, 49; R. Hanselmann, J. Zhou, P. Ma, and P. N. Confalone, *J. Org. Chem*., 2003, **68**, 8739.
15. S. Kanemasa, T. Tsuruoka, and E. Wada, *Tetrahedron Lett*., 1993, **34**, 87; T. Saito, T. Yamada, S. Miyazaki, and T. Otani, *Tetrahedron Lett*., 2004, **45**, 9581; B. Dugovic, L. Fisera, M. K. Cyranski, C. Hametner, N. Pronayova, and M. Obranec, *Helv. Chim. Acta*, 2005, **88**, 1432.
16. P. Merino, T. Tejero, M. Laguna, E. Cerrada, A. Moreno, and J. A. Lopez, *Org. Biomol. Chem*., 2003, **1**, 2336; Q. Zhao, F. Han, and D. L. Romero, *J. Org. Chem*., 2002, **67**, 3317.
17. K. B. Jensen, R. G. Hazell, and K. A. Jørgensen, *J. Org. Chem*., 1999, **64**, 2353.
18. P. Bayon, P. De March, M. Figueredo, and J. Font, *Tetrahedron*, 1998, **54**, 15691.
19. For examples of cycloreversion of nitrone-cycloadducts, see: G. M. Williams, S. D. Roughley, J. E. Davies, and A. B. Holmes, *J. Am. Chem. Soc.*, 1999, **121**, 4900; F. Pisaneschi, F. M. Cordero, and A. Brandi, *Synlett,* 2003, 1889; J. L. Garcia Ruano, A. Fraile, A. M. Martin Castro, and M. R. Martin, *J. Org. Chem.*, 2005, **70**, 8825; J. Garcia Ruano, J. F. Soriano, A. Fraile, M. R. Martin, and A. Nunez, *J. Sulfur Chem*., 2013, **34**, 17; A. J. Hodges, J. P. Adams, A. D. Bond, A. B. Holmes, N. J. Press, S. D. Roughley, J. H. Ryan, S. Saubern, C. J. Smith, M. D. Turnbullf, and A. F. Newton, *Org. Biomol.*

Chem., 2012, **10**, 8963.

20. Gutmann *et al.* defined "Acceptor Number," a quantitative empirical parameter for the Lewis acidities of solvents, based on the ^{31}P-NMR chemical shift of $Et_3P{=}O$ in the solvent, see: U. Mayer, V. Gutmann, and W. Gerger, *Monatsh. Chem*., 1975, **106**, 1235; V. Gutmann, 'The Donor-Acceptor Approach to Molecular Interactions,' Plenum Press, New York, 1978.
21. C. C. G. Scully and P. J. Rutledge, *Tetrahedron*, 2010, **66**, 5653.
22. H. Ikeuchi, M. E. Meyer, Y. Ding, J. Hiratake, and N. G. J. Richards, *Bioorg. Med. Chem*., 2009, **17**, 6641.