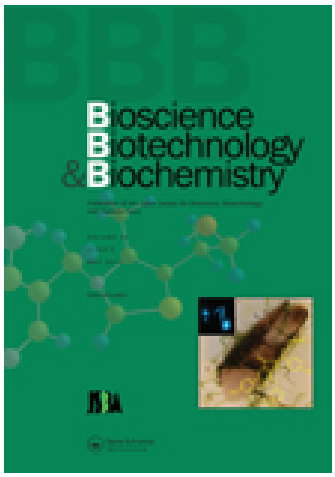

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Facile synthesis of the cyclohexane fragment of enacloxins, a series of antibiotics isolated from Frateuria sp. W-315

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Note

Facile synthesis of the cyclohexane fragment of enacloxins, a series of antibiotics isolated from *Frateuria* sp. W-315

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An efficient and good yield synthesis of the cyclohexane moiety of enacyloxins, a series of antibiotics isolated from *Frateuria* sp. W-315, was achieved from D-quinic acid using a successive Barton–McCombie deoxygenation.

Key words: enacyloxin; antibiotics; Barton–McCombie deoxygenation; D-quinic acid

Enacyloxins (ENXs) are unique polyhydroxy-polyenic and yellow-colored antibiotics produced by *Frateuria* sp. W-315 in a Czapek-Dox medium spent by *Neurospora crassa*.[1)] ENXs show antibiotic activity against Gram-positive and Gram-negative bacteria, but inactive for yeast and fungi.[1,2)] Its mode of action was considered to be an inhibition of peptide biosynthesis by hindering the release of EF-Tu GDP from the ribosome.[3)] Furthermore, ENXs have attracted considerable attention because of the inhibitory activity toward organelle protein synthesis in *Plasmodium falciparum*.[4)] The whole stereochemistry of ENXs [ENX IVa (**1**)] was elucidated by our synthetic[5–7)] and spectroscopic studies,[8)] and Parmeggiani's X-ray crystallographic analysis of the *Escherichia coli* EF-Tu/guanylyl iminodiphosphate-ENX IIa (**2**) complex.[3)] In spite of these unique properties, synthetic study other than by our group has not been reported yet. On the continuation of our chemical works on ENXs,[5–7,9,10)] we attempted to improve the synthesis of the cyclohexane moiety **3**. Although our previous synthesis had led us to determine its (1*S*,3*R*,4*S*) stereochemistry, it suffered from the long synthetic sequence from tri-*O*-acetyl-D-glucal and non-stereoselective intramolecular alkylation.[5)] Synthetic trial via *cis*-dihydroxylation of 3-cyclohexenecarboxylate derivatives resulted in formation of diastereomeric mixtures (Furukawa H and Kiyota H, unpublished results) (see Scheme 1).

Our new synthesis of the cyclohexane moiety **3** started with D-quinic acid (**4**) because of its stereochemical similarity.[11)] According to published procedures, acid-catalyzed reaction of **4** in acetone led to form lactone ring as well as isopropylidene acetal in high yield (**5**).[12)] Then radical-mediated deoxygenation of the tertiary hydroxy group was achieved via imidazole-type thiocarbamate[13)] with retention of the C1 configuration to give the known lactone **6** in 72% yield.[14)] Ring-opening of the lactone ring of **6** was performed using sodium methoxide to give methyl ester **7** in 86%. The second two-step reductive deoxygenation of **7** afforded **8** in 93% yield. Then hydrolysis of the acetal group in **8** with aqueous trifluoroacetic acid gave the target cyclohexane moiety **3** of ENXs in 87% yield. The overall yield was 55% in 7 steps from D-quinic acid (**4**). To confirm the stereochemistry, **3** was converted to the corresponding dibenzoate **9**.[5)] The physicochemical and spectroscopic data of **9** are identical with those reported by us.[5)]

In summary, the short, efficient, and facile synthesis of the cyclohexane moiety of enacycloxins, a series of polyhydroxy-polyenic antibiotics produced by *Frateuria* sp. W-315, was achieved using Barton–McCombie deoxygenation as the key steps from D-quinic acid (see Scheme 2).

Experimental

Optical rotation values were measured by a Horiba Sepa-300 polarimeter. FT-IR spectra were recorded as films by a Jasco 4100 spectrometer (ATR, Zn-Se). ^{1}H and ^{13}C NMR spectra were recorded with Agilent NMR System 600 (600 MHz for ^{1}H) and 400-MR (400 MHz for ^{1}H and 100 MHz for ^{13}C) spectrometers in $CDCl_3$ with tetramethylsilane (δ_H 0 ppm) and $CHCl_3$ (δ_C 77 ppm) as internal standards. Mass spectra (FAB) were recorded with a Jeol JMS-700 spectrometer. Merck silica gel 60 (70–230 mesh) was used for column chromatography.

(1S,3R,4R,5R)-3,4-O-Isopropylidenedioxycyclohexane-1,5-carbolactone (6). To a suspension of **5** (500 mg, 2.3 mmol) in dry 1,2-dichloroethane (6.7 mL) was added thiocarbonyldiimidazole (530 mg, 2.9 mmol, 1.3 eq) and

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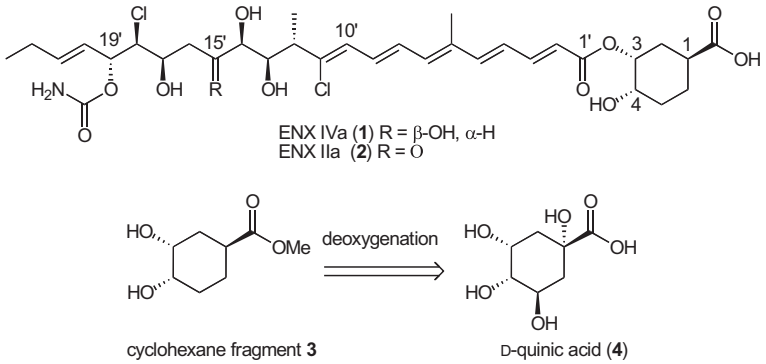
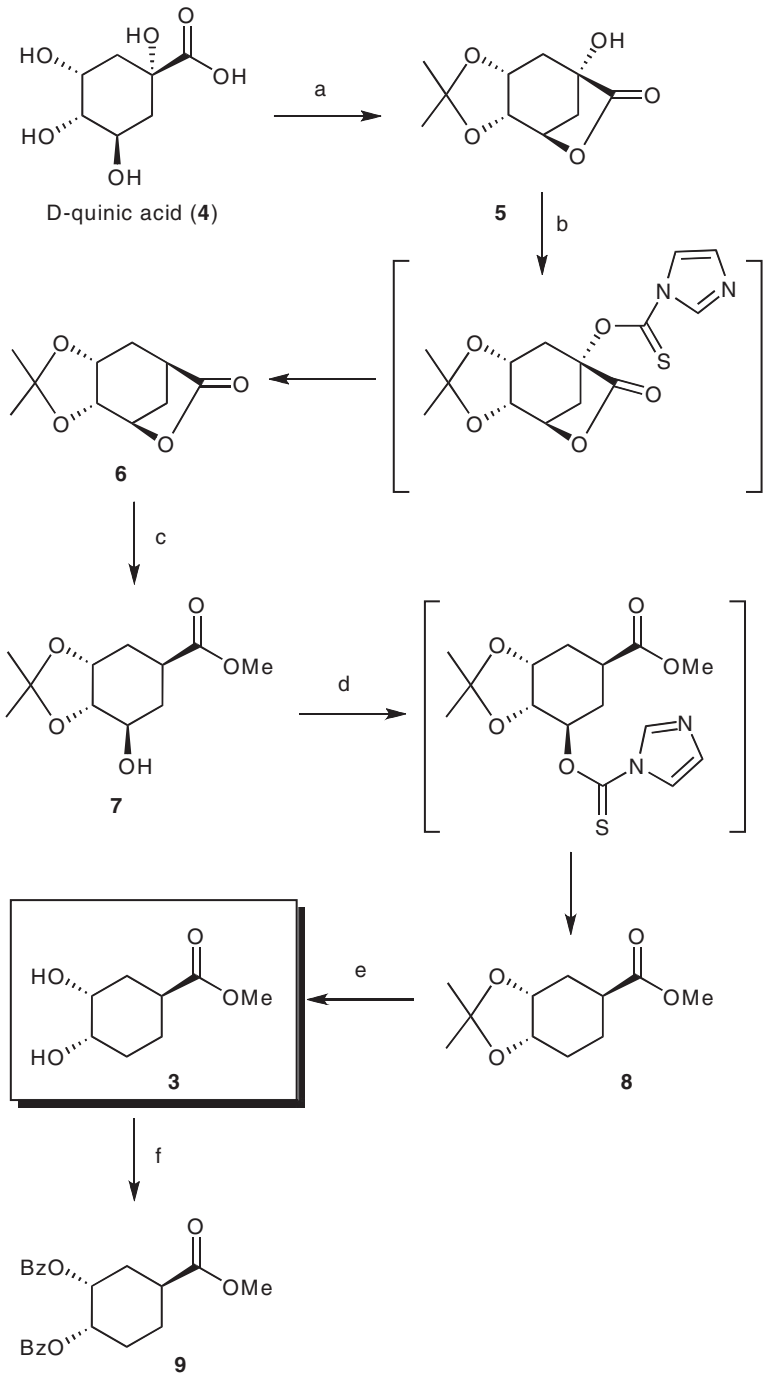


Scheme 1. Enacyloxin IIa (**1**), IVa (**2**), and retrosynthesis of the cyclohexane fragment **3** from D-quinic acid (**4**).



Scheme 2. Synthesis of **3**. *Reagents and conditions*: (a) Sanchez-Abella's procedure;[12] (b) (i) $Im_2C=S$, imidazole, $ClCH_2CH_2Cl$, reflux; (ii) Bu_3SnH, AIBN, xylene, reflux (72%); (c) NaOMe, MeOH, 20 °C (86%); (d) (i) $Im_2C=S$, imidazole, $ClCH_2CH_2Cl$, reflux; (ii) Bu_3SnH, AIBN, xylene, reflux (93%); (e) TFA, H_2O, 0 °C (87%); (f) see lit.[5] (37%).

imidazole (240 mg, 3.5 mmol, 1.5 eq) at 20 °C and the reaction mixture was stirred under reflux for 3 h after being cooled to 20 °C, the reaction mixture was concentrated and diluted by 50% of 2-butanone in acetonitrile. The dilute was concentrated to give crude thiocarbamate as an orange amorphous solid.

N_2 gas was passed through a dry xylene (86 mL) solution of the crude thiocarbamate and AIBN (16 mg, 0.092 mmol, 0.04 eq) for 5 min. To this was added tributyltin hydride (1.3 mL, 5.0 mmol, 2.2 eq) and the mixture was stirred under reflux for 1.5 h, and then cooled to 20 °C. The solvent was evaporated under reduced pressure, and the residue was purified by silica gel column chromatography (hexane-diethyl ether, 15:1–6:1) and recrystallization (hexane-diethyl ether) to give **6** (323 mg, 1.7 mmol, 72%) as a white amorphous solid; mp 95–96 °C [Lit.:[14] 96–98 °C (hexane-diethyl ether), $[\alpha]_D^{22}$ −36.3° (*c* 2.2, $CHCl_3$) [Lit.:[14] −36.2° (*c* 2.2, $CHCl_3$)].

The spectroscopic data were in good agreement with those reported.[14]

*Methyl (1*S*,3*R*,4*S*,5*R*)-5-Hydroxy-3,4-*O*-isopropylidendioxycyclohexanecarboxylate (7)*. To a solution of lactone **6** (380 mg, 1.9 mmol) in anhydrous methanol (4.6 mL) was added dropwise a solution of sodium methoxide (57 mg, 2.5 mmol) in anhydrous methanol (8.5 mL) over 30 min at 0 °C, and the mixture was neutralized with AcOH. Then, to this was added an aqueous saturated solution of $NaHCO_3$ and the mixture was extracted with CH_2Cl_2. The combined organic fraction was dried over Na_2SO_4, and concentrated under reduced pressure. The residue was purified by silica gel column chromatography (hexane-ethyl acetate, 5:1) to give **7** (380 mg, 1.6 mmol, 86%) as a colorless oil; $[\alpha]_D^{24}$ −71.7° (*c* 1.0, $CHCl_3$). IR ν_{max} cm^{-1}: 3466 (s, O–H), 2952 (m), 1730 (s, C=O), 1240 (m, C–O), 1055 (m, C–O). NMR (400 MHz): δ_H 1.37 (6 H, s, *gem*-CH_3), 1.49 (1 H, dd, *J* = 13.4, 8.0 Hz, H-6), 1.88 (1 H, ddd, *J* = 15.3, 12.0, 3.8 Hz, H-2), 2.12 (1 H, dd, *J* = 13.4, 4.0 Hz, H-6), 2.35 (1 H, ddd, *J* = 15.3, 6.0, 1.9 Hz, H-2), 2.74 (1 H, dddd, *J* = 12.0, 8.0, 4.0, 3.0 Hz, H-1), 3.70 (3 H, s, OCH_3), 3.75 (1 H, m, H-5), 3.84 (1 H, dd, *J* = 12.4, 6.0 Hz, H-4), 4.37 (1 H, m, H-3). NMR (100 MHz): δ_C 25.99, 28.17, 28.78, 32.60, 36.03, 51.91 (OMe), 71.44, 72.98, 107.89 (Me_2C), 175.20 (C-1). MS: *m/z*: 95, 155, 173 $[M+H-CO_2Me]^+$, 215 $[M+H-OH]^+$, 231 $[M+H]^+$. HRMS *m/z* ($[M+H]^+$): Calcd. for $C_{11}H_{19}O_5$: 231.1233; Found: 231.1232.

*Methyl (1*S*,3*R*,4*S*)-3,4-*O*-Isopropylidenedioxycyclohexanecarboxylate (8)*. To a suspension of **7** (380 mg, 1.6 mmol) in dry 1,2-dichloroethane (7.8 mL) was added thiocarbonyldiimidazole (380 g, 2.1 mmol, 1.3 eq) and imidazole (220 mg, 3.2 mmol, 2 eq) at 20 °C and the reaction mixture was stirred under reflux for 3 h. After being cooled to 20 °C, the reaction mixture was concentrated under reduced pressure and diluted by 50% of 2-butanone in acetonitrile. The dilute was concentrated under reduced pressure to give crude thiocarbamate as an orange amorphous solid.

N_2 gas was passed through a dry toluene (31 mL) solution of the crude thiocarbamate and AIBN (5.5 mg, 0.033 mmol, 0.02 eq) for 5 min. To this was added tributyltin hydride (0.49 mL, 1.8 mmol, 1.1 eq) and the mixture was stirred under reflux for 1.5 h, and then cooled to 20 °C. The solvent was evaporated under reduced pressure, and the residue was purified by silica gel column chromatography (hexane-diethyl ether, 10:1) to give **8** (320 mg, 1.5 mmol, 93%) as a colorless oil; $[\alpha]_D^{23}$ −10.3° (*c* 2.0, $CHCl_3$). IR ν_{max} cm^{-1}: 2953 (m), 1730 (s, C=O), 1217 (m, C–O), 1040 (m, C–O). NMR (400 MHz): δ_H 0.92 (2 H, t, *J* = 7.3 Hz, H-6), 1.34 (6 H, s, *gem*-CH_3), 1.45 (1 H, m, H-5), 1,65 (1 H, m, H-5), 1.85 (1 H, ddd, *J* = 15.3, 11.3, 4.1 Hz, H-2), 2.19 (1 H, ddd, *J* = 15.3, 4.1, 3.5 Hz, H-2), 2.68 (1 H, m, H-1), 3.68 (3 H, s, OCH_3), 4.11 (1 H, dd, *J* = 11.8, 5.0 Hz, H-4), 4.30 (1 H, ddd, *J* = 5.0, 3.5, 4.1 Hz, H-3). NMR (100 MHz): δ_C 23.51, 25.88, 27.80, 28.04, 29.13, 36.37, 51.69 (OMe), 72.40, 73.02, 107.93 (Me_2C), 175.94 (C-1). MS *m/z*: 175, 177, 179, 185 $[M+H-OMe]^+$, 199 $[M+H-Me]^+$, 216 $[M+H]^+$. HRMS *m/z* ($[M+H]^+$): Calcd. for $C_{11}H_{19}O_4$: 215.1283; Found: 215.1285.

*Methyl (1*S*,3*R*,4*S*)-3,4-Dihydroxycyclohexanecarboxylate (3)*. Cooled TFA/H_2O (1:1) solution (2.0 mL) was added to **8** (130 mg, 0.57 mmol) at 0 °C and the mixture was stirred for 1 h at 0 °C. Then the reaction mixture was allowed to warm to 20 °C and concentrated under reduced pressure. The residue was purified by silica gel column chromatography (hexane-ethyl acetate, 1:1) to give **3** (86 mg, 0.49 mmol, 87%) as a colorless oil; $[\alpha]_D^{23}$ –1.8° (*c* 1.0, $CHCl_3$). IR ν_{max} cm^{-1}: 3419 (s, O–H), 2950 (m), 2361 (s), 1725 (s, C=O), 1212 (m, C–O), 1065 (m, C–O). NMR (600 MHz): δ_H 1.49 (1 H, ddd, *J* = 16.8, 13.3, 11.2 Hz, H-6), 1.68 (1 H, ddd, *J* = 13.0, 11.4, 2.4 Hz, H-2), 1.73 (1 H, m, H-5), 1.98 (1 H, ddd, *J* = 16.8, 4.0, 2.4 Hz, H-6), 2.14 (1 H, ddd, 13.3, 13.0, 4.1 Hz, H-5), 2.18 (1 H, m, H-2), 2.72 (1 H, dddd, *J* = 15.1, 11.4, 11.2, 4.0 Hz, H-1), 3.65 (1 H, m, H-4), 3.67 (3 H, s, OCH_3), 4.01 (1 H, m, H-3). NMR (100 MHz): δ_C 26.04, 27.61, 32.95, 36.38, 51.72 (OMe), 68.56, 70.73, 176.18 (C=O). MS *m/z*: 97, 125, 137, 154, 175 $[M+H]^+$. HRMS *m/z* ($[M+H]^+$): Calcd. for $C_8H_{15}O_4$: 175.0970; Found: 175.0971.

*Methyl (1*S*,3*R*,4*S*)-3,4-Dibenzoyloxycyclohexanecarboxylate (9)*. **9** was obtained as a colorless oil in 37% from **3** according to our reported procedure; $[\alpha]_D^{22}$ −30.2° (*c* 0.54, $CHCl_3$). [Lit.:[5] −36.8° (*c* 0.54, $CHCl_3$)].

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