

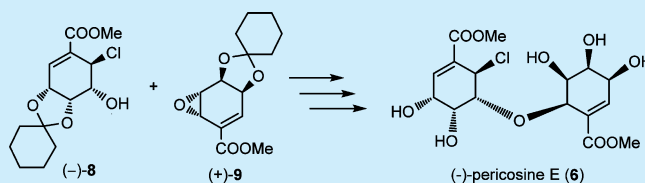
Synthesis of Marine Natural Product (–)-Pericosine E

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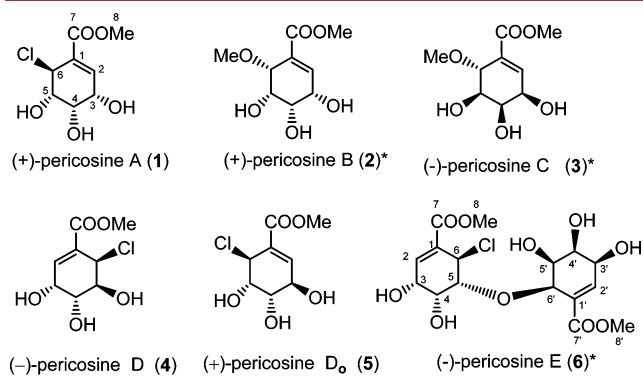
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Supporting Information

ABSTRACT: The first synthesis of (–)-pericosine E (**6**), a metabolite of the *Periconia byssoides* OUPS-N133 isolated originally from the sea hare *Aplysia kurodai*, has been achieved. Efficient and regioselective synthetic procedures for the synthesis of key intermediates, *anti*- and *syn*-epoxides **9** and **10**, were developed using an *anti*-epoxidation of diene **12** with TFDO and a bromohydrination of **12** with NBS in CH₃CN/H₂O (2:3), respectively. In addition, comparison of the specific optical rotations between synthetic **6** and natural **6** elucidated that the naturally preferred enantiomer of pericosine E had the same absolute configuration as (–)-**6** synthesized from chlorohydrin (–)-**8** and *anti*-epoxide (+)-**9**.



The identification of possible drug candidates from marine sources has been developing progressively over recent decades.^{1,2} However, the traditional natural product research style of isolating biologically active compounds from whole marine animals or plants has the potential of destroying large areas of natural habitat. An alternative approach for the discovery of biologically active molecules from marine animals or plants has emerged recently.^{3,4} To this end, Numata and co-workers reported the isolation and structural determination of pericosines A–E (**1**–**6**), metabolites of the fungus *Periconia byssoides* OUPS-N133, which were originally isolated from the sea hare *Aplysia kurodai*.^{5–7} All of these compounds, with the exception of pericosine E (**6**), have unique C₇-cyclohexenoid structures containing multifunctional groups on the six-membered ring, as shown in Figure 1. The absolute configurations in Figure 1 represent the preferential enantiomers that occur in Nature. It is worth noting that pericosines **2**, **3**, and **6** exist as enantiomeric mixtures in Nature.^{6,8}



*existing as enantiomeric mixture in Nature

Figure 1. Structures of naturally occurring pericosines.

Pericosine A (**1**) has previously shown remarkable physiological activities: (i) in vitro growth inhibition of breast cancer cell line HBC-5 and the central nervous system cell line SNB-75; (ii) inhibition against the protein kinase epidermal growth factor receptor; (iii) topoisomerase II inhibitory activity.⁶ Thus, research into the total synthesis of pericosines has received much attention in recent years.^{9–20}

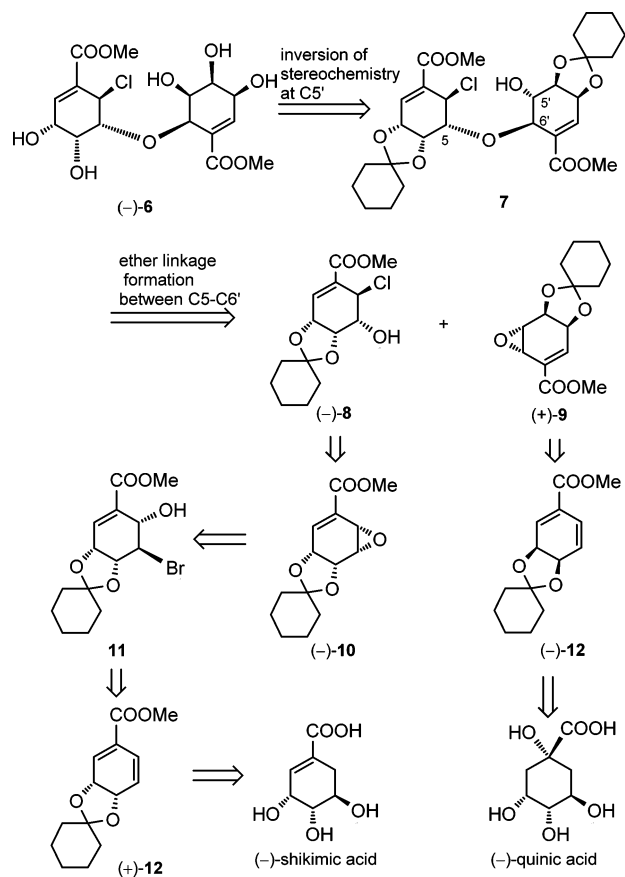
Of particular interest to us, pericosine E (**6**) is a unique natural product. It appears to be formed by connection of (–)-**1** and (+)-**2**, with the opposite enantiomer of the natural pericosine A forming the right half of **6**, while the left half adopts the intrinsic configuration of pericosine B (Figure 1). As the chemistry of **6** is complicated, the synthesis presents an exciting new challenge toward a compound with potentially valuable biological activity (ED₅₀ value of **6**: 15.5 μg/mL in in vitro growth inhibition activity against murine P388 cell line⁶).

If a synthetic route were established, it would allow the biological activity of **6** to be determined and improve the understanding of the biosynthetic route toward **6**. Herein, we disclose the first stereoselective total synthesis of (–)-**6** which clarifies the absolute configuration of the naturally occurring preferential enantiomer.

We envisioned the retrosynthesis of **6** as summarized in Scheme 1. In our previous synthetic studies on pericosine A [(+)-**1**] and C [(+)- or (–)-**3**],^{20,21} asymmetric synthesis of the bromohydrin (–)-**11** afforded only a low yield of the desired product. Moreover, in our work toward (–)-pericosine D (**4**), the low regioselectivity (ca. 1:1) in the epoxidation of (+)-**12** using *m*CPBA led to an inseparable mixture of epoxide (–)-**9** and its regioisomer.^{18,22} Thus, a more efficient and regioselective synthesis of the enantiomerically pure intermediates

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Scheme 1. Retrosynthetic Strategy of Pericosine E (6)



(+)-11 and (+)-9 was essential for the synthesis of (-)-pericosine E (6) (Scheme 1).

Initially, the synthesis of (-)-8 was undertaken, as illustrated in Scheme 2. (-)-Shikimic acid was converted to alcohol 13 under microwave irradiation (MW; 160 °C, 30 min) in 92% yield. Triflate 14 derived from 13 was treated with CsOAc in DMF to afford diene (+)-12 in two steps (78%).

The bromohydration of (+)-12 with NBS was examined in detail using various solvent systems and substrate concentrations within a fixed reaction time (20 h), as summarized in Table 1. The best selectivity for desired product (+)-11 was achieved using an acetonitrile–water (2:1) solvent system, with isomer 15 and byproduct 16 also formed within the reaction (Scheme 2 and Table 1, entries 1–5 and 7). The regio- and stereochemistry of byproduct 16 was established using ^1H – ^1H COSY, NOESY, HSQC, and HMBC experiments (see Supporting Information). From these results, acetonitrile–water was chosen as the optimal solvent system for the bromohydration. The ratio of acetonitrile to water was further explored (entries 6–10), using a 5 mg/mL concentration of (+)-12, with 2:3 acetonitrile–water giving the best selectivity (entry 9). The concentration of (+)-12 was investigated but did not improve the selectivity of the desired product (entries 11 and 12). Finally, it was found that a 4 h reaction time gave almost the same result compared to 20 h (entry 9 and 13). Using the optimized conditions, the three-step conversion of alcohol 13 to (+)-11 was achieved in 65% yield without purification. The bromohydrin obtained was treated with LHMDS in THF at –78 °C to afford (-)-10 in 83% yield,

Scheme 2. Synthesis of Chlorohydrin (-)-8 from (-)-Shikimic Acid

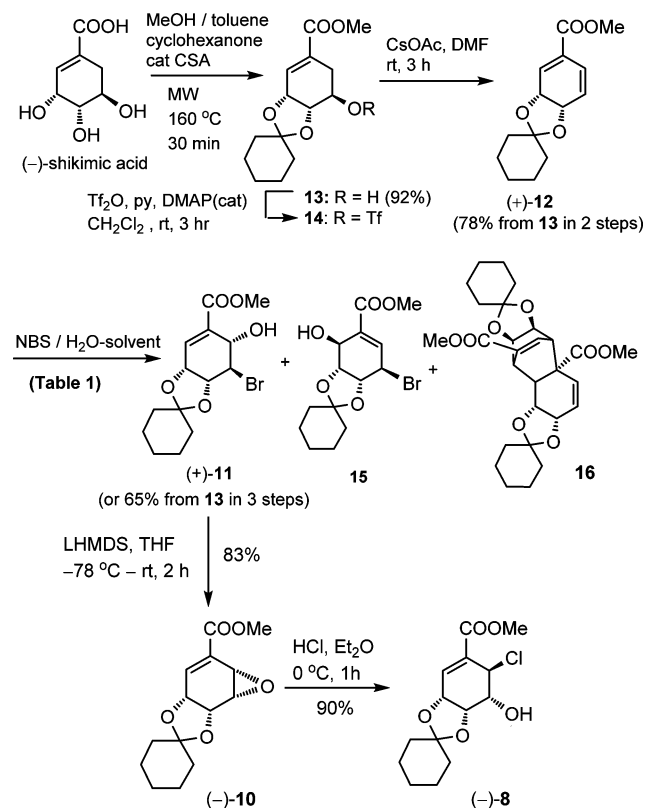


Table 1. Bromohydration of (+)-12 to (+)-11^{20,21}

entry ^a	concn of 12 (mg/mL)	solvent	product ratio ^b (%)		
			(+)-11	15	16
1	5	dioxane–H ₂ O (2:1)	36	49	15
2	5	<i>t</i> BuOH–H ₂ O (2:1)	43	27	30
3	5	DMSO–H ₂ O (2:1)	insoluble		
4	5	acetone–H ₂ O (2:1)	72	12	16
5 ^c	5	THF–H ₂ O (2:1)	33	46	
6	5	MeCN–H ₂ O (4:1)	38	45	17
7	5	MeCN–H ₂ O (2:1)	76	8	16
8	5	MeCN–H ₂ O (1:1)	74	13	13
9	5	MeCN–H ₂ O (2:3)	80	7	13
10	5	MeCN–H ₂ O (1:2)	75	12.5	12.5
11	2.5	MeCN–H ₂ O (2:3)	74	9	17
12	7.5	MeCN–H ₂ O (2:3)	75	11	14
13	5	MeCN–H ₂ O (2:3)	80	8	12

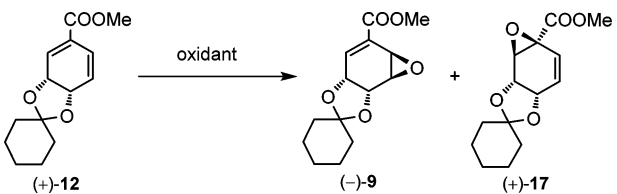
^aReaction time in all entries was set to 20 h except entry 13 (4 h).

^bRatios were determined by analysis of ^1H NMR spectra of crude reaction mixtures. ^cRecovered diene 12 at a ratio of 21%.

which was then converted to (-)-8 by treatment with HCl in 90% yield.

Next, the formation of epoxy intermediate (+)-9 was explored using readily accessible diene (+)-12, the enantiomer of intermediate (-)-12 for pericosine D synthesis (Table 2). It is worth noting that both enantiomers of 12 can be accessed from (-)-quinic acid²³ or (-)-shikimic acid. Oxidation of 12

Table 2. Examination of the Epoxidation of (+)-12



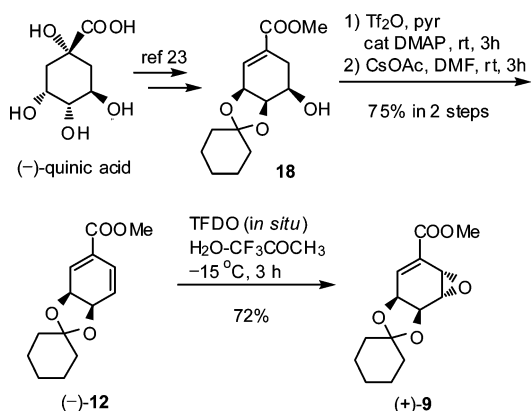
entry	oxidant	solvent	reaction time (h)	temp (°C)	product yield ^a (%)	
					9	17
1 ^b	<i>m</i> CPBA	CH ₂ Cl ₂	10	40	35	35
2 ^c	DMDO ^d	H ₂ O–acetone	10	0 to rt	24	18
3	TFDO ^d	H ₂ O–CF ₃ COCH ₃	3	0 to rt	57.4	3.3
4	TFDO ^d	H ₂ O–CF ₃ COCH ₃	3	0	65	0
5	TFDO ^d	H ₂ O–CF ₃ COCH ₃	3	–15	72	0

^aYields were calculated as combined yield from ¹H NMR spectrum of crude products. ^bFrom ref 18. ^cExperiment was carried out with the procedure described in ref 8. ^dPrepared in situ.

using dimethyldioxirane (DMDO) gave an improved 4:3 ratio of (–)-9 and (+)-17 (entry 2) compared to oxidation using *m*CPBA (entry 1).¹⁸ Furthermore, oxidation of 12 with methyl(trifluoromethyl)dioxirane (TFDO)^{24,25} provided a 16:1 mixture of 9 and 17 in 61% combined yield (entry 3). Performing the reaction at 0 °C for 3 h afforded only 9 in 65% isolated yield (entry 4). Two explanations for better selectivity with TFDO against DMDO are plausible. Bulkiness of the trifluoromethyl group in TFDO might inhibit oxidation of the double bond bearing a methoxycarbonyl group in 12, or increased electrophilicity of TFDO from inductive effect of the trifluoromethyl group might accelerate the reaction to another double bond, which does not bear an electron-withdrawing methoxycarbonyl group in 12. Finally, reduction of the reaction temperature to –15 °C increased the yield of (–)-9 to 72% (entry 5).

Next, the synthesis of *anti*-epoxide (+)-9 was carried out, as shown in Scheme 3. Starting from (–)-quinic acid, known

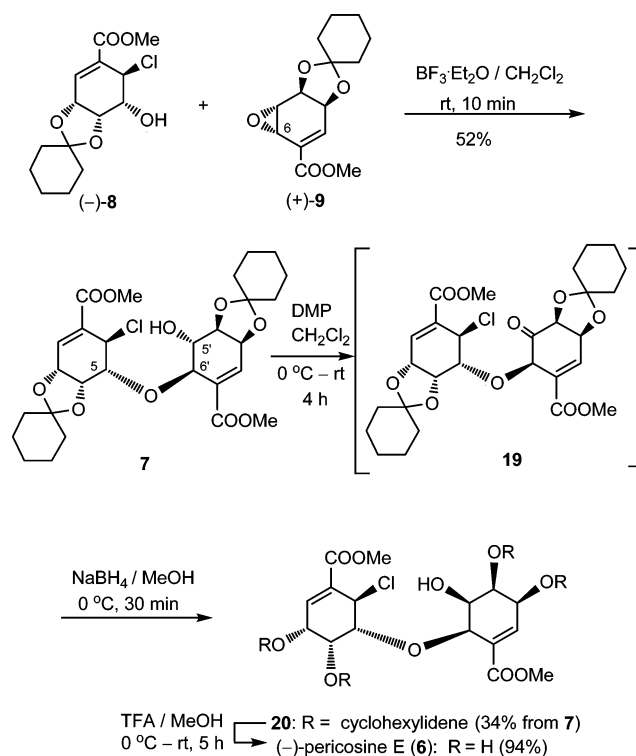
Scheme 3. Synthesis of (+)-9 from (–)-Quinic Acid



alcohol 18²³ was converted to cyclohexadiene (–)-12 in two steps according to a literature procedure in 75% yield.²⁰ Using the optimized epoxidation conditions, (–)-12 was successfully oxidized to (+)-9 in 72% yield.

With required molecules (–)-8 and (+)-9 in hand, attention turned to the synthesis of (–)-pericosine E (6) (Scheme 4). Ether linkage formation between (–)-8 and (+)-9 was carried out by treatment with BF₃–Et₂O (0.1 equiv) in CH₂Cl₂ at room temperature, affording condensation product 7 in 52%

Scheme 4. Synthesis of (–)-Pericosine E (6) from (–)-8 and (+)-9



isolated yield (see Table SI-1 in the Supporting Information on the model studies of the coupling reaction). The regioselective outcome of the nucleophilic epoxide opening was determined to be at C6 of (+)-9, on the basis of 600 MHz ¹H–¹H COSY analysis of 7 in CDCl₃. The NOESY cross-peak H6'/H4' suggested the configuration at C6' in 7, as shown in Scheme 3. An oxidation–reduction sequence was applied for inversion of the remaining C5'-hydroxyl group of 7. Alcohol 7 was treated with Dess-Martin periodinane to give a crude inseparable mixture of ketone 19 and an undefined compound. Subsequently, the crude mixture including 19 was reduced with NaBH₄ to afford the desired epimeric alcohol 20 in 34% yield from 7. The cross-peak H3'/H5' in the NOESY spectrum of 20 in acetone-*d*₆ (600 MHz) confirmed the configuration of a newly generated stereocenter at C5'. Finally, treatment of 20

with trifluoroacetic acid in methanol completed the total synthesis of **6**. Spectral data obtained for product **6** were consistent with those of the naturally occurring product except for the specific rotation.⁶ The specific rotation for synthesized **6** was $[\alpha]_D -68.3$, whereas that for reported natural **6** is -31.5 . This indicates that the absolute configuration of the naturally dominant enantiomer of (–)-**6** is assigned to (3*R*,4*R*,5*R*,6*R*)-methyl 6-chloro-3,4-dihydroxy-5-[(1*R*,4*S*,5*S*,6*S*)-4,5,6-trihydroxy-2-(methoxycarbonyl)cyclohex-2-en-1-yl]oxy)cyclohex-1-enecarboxylate. Accordingly, the ratio of (–)-**6**/(+)-**6** in natural pericosine E would roughly correlate to 3:1.

In conclusion, the total synthesis of pericosine E has been achieved for the first time and allowed the elucidation of the absolute configuration of the naturally preferred pericosine E as (–)-**6**. Regioselectivities in the synthesis of epoxides **9** and **10**, which are the key intermediates in pericosine synthesis, were efficiently improved. Bromohydration of cyclohexadiene **12** in the CH₃CN–H₂O (2:3) solvent system was proved to be the optimum conditions for the formation of bromohydrin **11**, which was subsequently converted to epoxide **10**. Epoxidation of **12** with TFDO formed in situ afforded epoxide **9** with complete regiocontrol. We believe that the synthesis could be scaled up to meet the demand in the search of novel bioactive analogues of pericosine E.

■ ASSOCIATED CONTENT

● Supporting Information

Experimental procedures and ¹H and ¹³C NMR spectra for new compounds described therein. This material is available free of charge via the Internet at <http://pubs.acs.org>.

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Notes

The authors declare no competing financial interest.

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