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Improved Syntheses of Morinol C and D by Employing Mizoroki-Heck Reaction and Their Cytotoxic and Antimicrobial Activities

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Improved syntheses of optically pure (−)- and (+)-morinol C, and (−)- and (+)-morinol D were achieved by employing the Mizoroki-Heck reaction to construct the cinnamyl moiety. The protective group of the alkene substrate affected the yield of this key reaction. The reaction with a combination of the acetate-protected olefin and 4-methoxyphenylboronic acid gave the best result producing morinol C and D. All stereoisomers of morinol C and D showed cytotoxic activity, with (*R*,*R*)-morinol C showing the highest antibacterial activity.

Key words: morinol C and D; neolignan; Mizoroki-Heck reaction

Morinol C (**1** and **2**) and D (**3** and **4**) (Fig. 1) were isolated from *Morina chinensis* as an enantiomeric mixtures.[1)] Although this plant source has been used as a traditional Chinese medicine, the effect of **1**–**4** has not been elucidated. These compounds have a characteristic phenylpropanoid bonding system. Since many biological activities of the phenylpropanoid compounds have been reported,[2,3)] biological research into **1**–**4** is an interesting subject. The development of an effective method for synthesizing optically pure compounds is important to pursue into the biological activity of these compounds. Our previous study of the first syntheses of **1**–**4**[4)] employed the Horner-Wadsworth-Emmons reaction to furnish the cinnamyl moiety, giving **1**–**4** by 7–11 steps in 5–9% overall yields. A more effective synthetic method by employing the Mizoroki-Heck reaction to construct the cinnamyl structure is described in this article. The antimicrobial and cytotoxic activities are also discussed.

Results and Discussion

Syntheses of morinol C and D

The optimum conditions for the Mizoroki-Heck reaction of **5**–**8** were examined to achieve an efficient synthesis of **1**. Substrates **5**–**8** were prepared by the reported method with a small modification.[4)] The reaction with diol **5** has not so far given the coupling compounds under the conditions we examined.[5–10)] We checked the effect of the protective groups by selecting pivaloyl ester **6**, acetate **7**, and silyl ether **8** as the substrates. Table 1 shows the results. The highest yield (97% yield) of *trans* olefin **10** was observed, when 4-methoxyphenylboronic acid, $Cu(OAc)_2$, LiOAc, and $Pd(OAc)_2$ in DMF[6)] were used with acetate substrate **7** (entry 2). The concentration of the substrate between 1 mM and 5 mM did not affect the yield. The reaction with pivalate **6** under the same condition gave **9** in 76% yield (entry 1), while the reaction with triisopropylsilyl ether **8** gave **11** in only 32% yield (entry 6). The other reaction conditions for alkene coupling using the protective substrates are shown in the table. The best substrates for the Mizoroki-Heck reaction were 4-methoxyphenylboronic acid and acetate. The hydrolysis of coupling compound **10** gave **1** in 83% yield. Compound **2** was obtained from an enantiomer of **7** and compounds **3** and **4** were respectively obtained from the diastereomeric acetate **12**[11)] and its enantiomer (Scheme 1). The enantiomeric excess was determined to be more than 99% by HPLC with a chiral column.

This study enable us to improve the synthetic route to morinol C and D by employing the Mizoroki-Heck reaction through four steps to give 46%–63% overall yields.

Biological activity

Synthesized stereoisomers **1**–**4** were evaluated by a WTS-8 assay for their cytotoxic activity toward U266 cells. All compounds showed almost same level of cytotoxic activity at 30 μmol (Fig. 2). A cytotoxic activity test of the γ-butyrolactone type of lignan, (−)-arctigenin,[2)] has shown a decrease in living cells to 30% at 30 μmol. It was found that the stereochemistry of morinol C and D had no affect on the cytotoxic activity. The antibacterial activity was also evaluated (Table 2). Two gram-positive bacteria (*B. subtillus* and *L. denitrificans*) and one gram-negative bacterium (*S. choleraesuis*) were sensitive to these compounds. Stereoisomer **2** in particular showed the highest activity, giving an MIC value of 6.3 mM against *S. choleraesuis*. This activity is weaker than that of the butane type of lignan, (+)-guaiaretic acid[2)] (MIC of 3.1 mM) and the antibiotic, valinomycin (MIC of 6.5 μM). Although some lignans have shown antifungal activity,[12–14)] no antifungal activity of **1**–**4** against phytopathogenic fungi was apparent. The biological activities of all the stereoisomers of **1**–**4**, which are new neolignans, were examined for the first time in this study.

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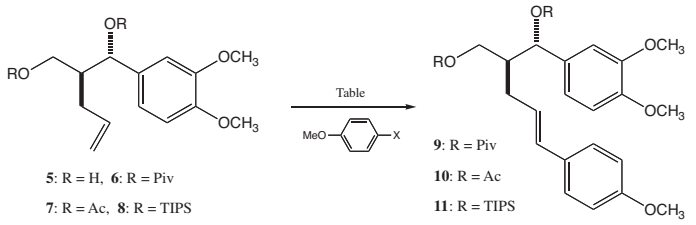
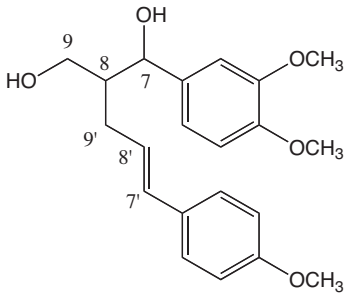
Table 1. Coupling Reaction Using Substrates **6–8**



Entry	Substrate	X	Reagents	Temperature (°C)	Time (h)	Product (Yield, %)
1	**6**	$B(OH)_2$	$Cu(OAc)_2$, LiOAc, $Pd(OAc)_2$/DMF	100	2	**9** (76)
2	**7**	$B(OH)_2$ (1.2 eq.)	$Cu(OAc)_2$ (2.0 eq.), LiOAc (3.0 eq.), $Pd(OAc)_2$ (0.1 eq.)/DMF	100	2	**10** (97)
3	**7**	$B(OH)_2$ (1.2 eq.)	$Cu(OAc)_2$ (2.0 eq.), LiOAc (3.0 eq.), $Pd(OAc)_2$ (0.01 eq.)/DMF	100	2	**10** (78)
3	**7**	$B(OH)_2$ (1.2 eq.)	$Cu(OAc)_2$ (1.0 eq.), LiOAc (1.5 eq.), $Pd(OAc)_2$ (0.1 eq.)/DMF	100	2	**10** (93)
4	**7**	$B(OH)_2$ (1.2 eq.)	$Cu(OAc)_2$ (2.0 eq.), LiOAc (3.0 eq.), $Pd(OAc)_2$ (0.1 eq.)/DMF	80	2	**10** (64)
5	**7**	$B(OH)_2$ (1.2 eq.)	$Cu(OAc)_2$ (2.0 eq.), LiOAc (3.0 eq.), $Pd(OAc)_2$ (0.1 eq.)/DMF	60	2	**10** (52)
6	**8**	$B(OH)_2$	$Cu(OAc)_2$, LiOAc, $Pd(OAc)_2$/DMF	100	2	**11** (32)
7	**6**	I	$Pd(OAc)_2/CH_3CN$	80	16	**9** (0)
8	**7**	I	$Pd(OAc)_2/CH_3CN$	80	16	**10** (21)
9	**8**	I	$Pd(OAc)_2/CH_3CN$	80	16	**11** (11, Z isomer, 6)
10	**6**	Br	$PdCl_2(PPh_3)_2$, Et_3N/DMF	90	4	**9** (0)
11	**7**	Br	$PdCl_2(PPh_3)_2$, Et_3N/DMF	90	4	**10** (7)
12	**8**	Br	$PdCl_2(PPh_3)_2$, Et_3N/DMF	90	4	**11** (0)
13	**6**	$CH{=}CH_2$	2nd Grubbs/CH_2Cl_2	40	9	**9** (30)
14	**7**	$CH{=}CH_2$	2nd Grubbs/CH_2Cl_2	40	7	**10** (34)
15	**7**	$CH{=}CH_2$	2nd Grubbs/toluene	40	7	**10** (40)
16	**8**	$CH{=}CH_2$	2nd Grubbs/CH_2Cl_2	40	7	**11** (0)
17	**8**	$CH{=}CH_2$	2nd Grubbs/toluene	40	7	**11** (25)



(7*S*,8*S*)-Morinol C (**1**)

(7*R*,8*R*)-Morinol C (**2**)

(7*S*,8*R*)-Morinol D (**3**)

(7*R*,8*S*)-Morinol D (**4**)

Fig. 1. (7*S*,8*S*)- and (7*R*,8*R*)-Morinol C and (7*S*,8*R*)- and (7*R*,8*S*)-Morinol D.

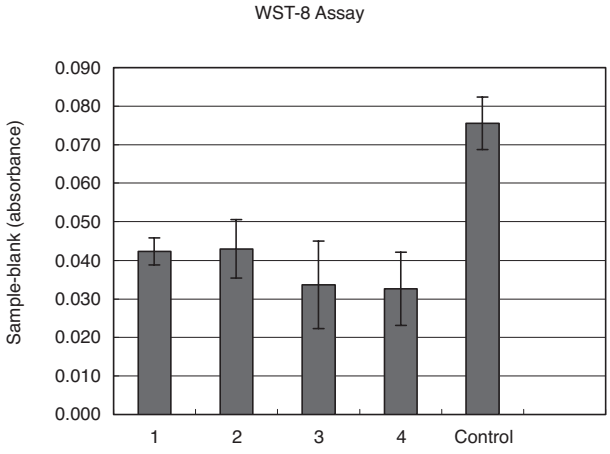
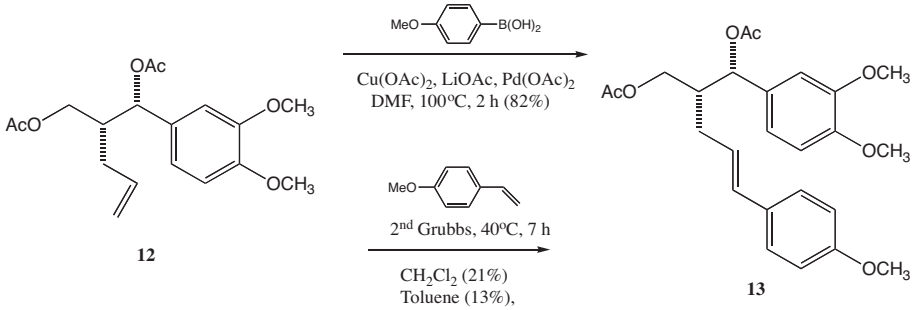


Fig. 2. Cytotoxic Activity of **1–4** by Using U266 Cell and WST-8 Assay at 30 μmol.



Scheme 1. Mizoroki-Heck Reaction Using Substrate **12**.

Table 2. Antibacterial Activity of **1**–**4** (MIC, mM)

	1	**2**	**3**	**4**
Bucillus subtilis	50	50	>50	50
Listeria denitrificans	50	25	25	50
Salmonella choleraesuis	50	6.3	25	25

Experimental

General experimental procedures. Melting point (mp) data are uncorrected. NMR data were measured by a JNM-EX400 spectrometer, using TMS as a standard (0 ppm), MS data were measured with a JMS-MS700V spectrometer, and optical rotation values were evaluated with a Jasco P-2100 polarimeter. Elemental analyses were carried out with a Yanako MT-5 CHN coder, and the silica gel used was Wakogel C-300 (Wako, 200–300 mesh).

*(1S,2S)-2-Allyl-1-(3,4-dimethoxyphenyl)-1,3-propanediol (**5**).* To a solution of (*S*)-4-benzyl-3-{(2*R*)-2-[(*S*)-(3,4-dimethoxyphenyl)(hydroxy)methyl]-4-pentenoyl}-2-oxazolidinone[4] (7.30 g, 17.2 mmol) and MeOH (1.50 ml) in THF (45 ml) was added a solution of $LiBH_4$ (1.63 g, 74.8 mmol) in THF (100 ml) at 0 °C. After stirring at 0 °C for 1 h, 1 M aq. NaOH (80 ml) solution was added. The organic solution was separated, washed with brine, and dried ($NaSO_4$). Concentration followed by silica gel column chromatography (EtOAc/hexane = 1/1) gave diol **5** (3.49 g, 13.8 mmol, 80%) as colorless crystals, mp 74–75 °C, $[\alpha]_D^{20} = -40$ (*c* 0.22, $CHCl_3$). ^{1}H-NMR ($CDCl_3$) δ: 1.91–1.95 (2H, m), 2.02 (1H, m), 3.00 (1H, t, $J = 6.6$ Hz), 3.21 (1H, d, $J = 4.6$ Hz), 3.69 (1H, m), 3.84 (1H, m), 3.87 (3H, s), 3.89 (3H, s), 4.64 (1H, dd, $J = 9.2$, 4.6 Hz), 5.00 (1H, d, $J = 15.1$ Hz), 5.01 (1H, d, $J = 13.1$ Hz), 5.70 (1H, m), 6.83 (1H, d, $J = 8.1$ Hz), 6.87 (1H, dd, $J = 8.1$, 1.7 Hz), 6.90 (1H, d, $J = 1.7$ Hz). ^{13}C-NMR ($CDCl_3$) δ: 33.0, 46.2, 55.9, 64.4, 78.4, 109.4, 110.9, 116.7, 118.9, 135.9, 136.1, 148.5, 149.1. *Anal*. Found: C, 66.36; H, 7.86%. Calcd. for $C_{14}H_{20}O_4$: C, 66.65%; H, 7.99%. (1*R*,2*R*)-**5**, 80% yield, $[\alpha]_D^{20} = +40$ (*c* 1.0, $CHCl_3$).

*(1S,2S)-2-Allyl-1-(3,4-dimethoxyphenyl)trimethylene dipivalate (**6**).* To an ice-cooled solution of diol **5** (0.55 g, 2.2 mmol) in pyridine (4.0 ml) was added PivCl (0.50 ml, 4.1 mmol). The resulting reaction mixture was stirred at room temperature for 1 h before additions of EtOAc and H_2O. The organic solution was separated, washed with 6 M aq. HCl solution, sat. aq. $NaHCO_3$ solution, and brine, and dried (Na_2SO_4). Concentration followed by silica gel column chromatography (EtOAc/hexane = 1/7) gave dipivalate **6** (0.66 g, 1.6 mmol, 73%) as a colorless oil, $[\alpha]_D^{20} = -30$ (*c* 0.74, $CHCl_3$). ^{1}H-NMR ($CDCl_3$) δ: 1.20 (9H, s), 1.22 (9H, s), 1.98–2.10 (2H, m), 2.27 (1H, m), 3.87 (3H, s), 3.88 (3H, s), 4.10 (1H, dd, $J = 11.2$, 4.1 Hz), 4.21 (1H, dd, $J = 11.2$, 5.2 Hz), 4.99 (1H, d, $J = 17.0$ Hz), 5.04 (1H, d, $J = 10.2$ Hz), 5.67 (1H, d, $J = 8.2$ Hz), 5.70 (1H, m), 6.82 (1H, s), 6.83 (1H, d, $J = 8.2$ Hz), 6.87 (1H, d, $J = 8.2$ Hz). ^{13}C-NMR ($CDCl_3$) δ: 27.0, 27.2, 32.0, 38.8, 38.9, 43.0, 55.8, 62.2, 74.7, 109.8, 110.9, 117.4, 119.4, 131.4, 135.2, 148.7, 148.9, 176.9, 178.3. EIMS *m*/*z* (%) 420 (M^+, 39), 57 (100). HREIMS *m*/*z* M^+: calcd. for $C_{24}H_{36}O_6$, 420.2511; found, 420.2513.

*(1S,2S)-2-Allyl-1-(3,4-dimethoxyphenyl)trimethylene diacetate (**7**).* After a solution of diol **5** (0.46 g, 1.8 mmol) in pyridine (3.0 ml) and Ac_2O (3.0 ml) was stirred at room temperature for 5 h, ice was added. The mixture was extracted with EtOAc. The organic solution was separated, washed with 6 M aq. HCl solution, sat. aq. $NaHCO_3$ solution, and brine, and dried (Na_2SO_4). Concentration followed by silica gel column chromatography (EtOAc/hexane = 1/3) gave diacetate **7** (0.43 g, 1.3 mmol, 72%) as a colorless oil, $[\alpha]_D^{20} = -36$ (*c* 1.2, $CHCl_3$). ^{1}H-NMR ($CDCl_3$) δ: 1.92–2.06 (2H, m), 2.06 (6H, s), 2.26 (1H, m), 3.87 (3H, s), 3.89 (3H, s), 4.08 (1H, dd, $J = 11.1$, 4.3 Hz), 4.26 (1H, dd, $J = 11.1$, 5.0 Hz), 4.99 (1H, d, $J = 17.3$ Hz), 5.03 (1H, d, $J = 10.2$ Hz), 5.71 (1H, d, $J = 8.0$ Hz), 5.70 (1H, m), 6.83–6.90 (3H, m). ^{13}C-NMR ($CDCl_3$) δ: 20.8, 21.1, 32.0, 42.5, 55.8, 55.9, 62.4, 75.1, 110.1, 110.9, 117.4, 119.6, 131.0, 134.9, 148.8, 148.9, 169.9, 170.9. *Anal*. Found: C, 63.99; H, 7.21%. Calcd. for $C_{18}H_{24}O_6$: C, 64.27%; H, 7.19%. (1*R*,2*R*)-**7**, 79% yield, $[\alpha]_D^{20} = +36$ (*c* 1.0, $CHCl_3$).

*(4S,5S)-5-(3,4-Dimethoxyphenyl)-5-triisopropylsilyloxy-4-(triisopropylsilyloxy)methyl-1-pentene (**8**).* To an ice-cooled solution of diol **5** (0.23 g, 0.91 mmol) and 2,6-lutidine (0.41 ml, 3.5 mmol) in CH_2Cl_2 (80 ml) was added TIPSOTf (0.60 ml, 2.2 mmol). The resulting reaction solution was stirred at room temperature for 2 h before addition of sat. aq. $NaHCO_3$ solution. The organic solution was separated, washed with sat. aq. $CuSO_4$ solution and sat. aq. $NaHCO_3$ solution, and dried (Na_2SO_4). Concentration followed by silica gel column chromatography (5%EtOAc/hexane) gave diTIPS ether **8** (0.34 g, 0.60 mmol, 66%) as a colorless oil, $[\alpha]_D^{20} = -13$ (*c* 1.3, $CHCl_3$). ($[\alpha]_D^{20} = -14$ (*c* 1.0, $CHCl_3$) in the literature).[4] The NMR data agreed with those in the literature.[4]

*(1S,2S)-1-(3,4-Dimethoxyphenyl)-2-[(*E*)-3-(4-methoxyphenyl)-2-propen-1-yl]trimethylene dipivalate (**9**).* Method A: A reaction mixture of dipivalate **6** (50 mg, 0.12 mmol), 4-methoxyphenylboronic acid (22 mg, 0.14 mmol), $Cu(OAc)_2$ (43 mg, 0.24 mmol), LiOAc (24 mg, 0.36 mmol), and $Pd(OAc)_2$ (3.0 mg, 0.013 mmol) in DMF (0.50 ml) was stirred at 100 °C for 2 h before additions of EtOAc and sat. aq. NH_4Cl solution. The organic solution was separated, washed with brine, and dried (Na_2SO_4). Concentration followed by silica gel column chromatography (EtOAc/hexane 1/3) gave **9** (48 mg, 0.091 mmol, 76%). Method E: A reaction mixture of dipivalate **6** (50 mg, 0.12 mmol), 4-methoxystyrene (65 mg, 0.48 mmol), and 2nd Grubbs catalyst (10 mg, 0.012 mmol) in CH_2Cl_2 (12 ml) was stirred at 40 °C for 7 h before concentration. The residue was applied to silica gel column chromatography (EtOAc/hexane = 1/7) to give **9** (19 mg, 0.036 mmol, 30%). **9**: colorless oil, $[\alpha]_D^{20} = -8$ (*c* 0.4, $CHCl_3$). ^{1}H-NMR ($CDCl_3$) δ: 1.20 (9H, s), 1.23 (9H, s), 2.10–2.22 (2H, m), 2.34 (1H, m), 3.79 (3H, s), 3.87 (6H, s), 4.14 (1H, dd, $J = 11.2$, 4.0 Hz), 4.25 (1H, dd, $J = 11.2$, 5.3 Hz), 5.70 (1H, d, $J = 8.1$ Hz), 5.93 (1H, m), 6.24 (1H, d, $J = 15.8$ Hz), 6.82 (2H, d, $J = 8.6$ Hz), 6.85 (1H, d, $J = 2.3$ Hz), 6.88 (1H, dd, $J = 8.8$, 2.3 Hz), 7.21 (2H, d, $J = 8.6$ Hz), 7.43 (1H, d, $J = 8.8$ Hz). ^{13}C-NMR ($CDCl_3$) δ: 27.1, 27.2, 31.3, 38.8, 38.9, 43.7, 55.3, 55.85, 55.88, 62.6, 74.9, 110.0, 111.0, 113.9, 114.1, 119.6, 124.5, 127.1, 127.4, 130.0, 131.5, 131.9, 148.8, 148.9, 158.9, 176.9. EIMS *m*/*z* (%): 526 (M^+, 28), 322 (100), 291 (63), 147 (90). HREIMS *m*/*z* M^+: calcd. for $C_{31}H_{42}O_7$, 526.2931; found, 526.2932.

*(1S,2S)-1-(3,4-Dimethoxyphenyl)-2-[(*E*)-3-(4-methoxyphenyl)-2-propen-1-yl]trimethylene diacetate (**10**).* Method A: 97% yield, Method B: A reaction mixture of diacetate **7** (50 mg, 0.15 mmol), 1-iodo-4-methoxybenzene (40 mg, 0.17 mmol), $Pd(OAc)_2$ (5.0 mg, 0.022 mmol), and Et_3N (0.20 ml, 1.4 mmol) in MeCN (2.0 ml) was stirred at 85 °C for 16 h before additions of H_2O and EtOAc. The organic solution was separated, washed with brine, and dried (Na_2SO_4). Concentration followed by silica gel column chromatography (EtOAc/hexane = 1/3) gave **10** (14 mg, 0.032 mmol, 21%). Method C: A reaction mixture of diacetate **7** (50 mg, 0.15 mmol), 1-bromo-4-methoxybenzene (30 mg, 0.16 mmol), $PdCl_2(PPh_3)_2$ (10 mg, 0.014 mmol), and Et_3N (70 µl, 0.50 mmol) in DMF (0.30 ml) was stirred at 80 °C for 4 h before additions of H_2O and EtOAc. The organic solution was separated, washed with brine, and dried (Na_2SO_4). Concentration followed by silica gel column chromatography (EtOAc/hexane = 1/3) gave **10** (5.0 mg, 0.011 mmol, 7%). Method E: CH_2Cl_2 was used at 40 °C, 34% yield. Toluene was used at 40 °C, 40% yield. **10**: colorless oil, $[\alpha]_D^{20} = -8$ (*c* 0.9, $CHCl_3$). ^{1}H-NMR ($CDCl_3$) δ: 2.06 (6H, s), 2.08–2.20 (2H, m), 2.34 (1H, m), 3.79 (3H, s), 3.87 (3H, s), 3.88 (3H, s), 4.13 (1H, dd, $J = 11.2$, 4.4 Hz), 4.30 (1H, dd, $J = 11.2$, 5.3 Hz), 5.74 (1H, d, $J = 8.3$ Hz), 5.93 (1H, m), 6.24 (1H, d, $J = 15.8$ Hz), 6.74–6.85 (4H, m), 6.91 (1H, dd, $J = 8.3$, 1.9 Hz), 7.22 (2H, d, $J = 8.7$ Hz). ^{13}C-NMR ($CDCl_3$) δ: 20.8, 21.1, 31.3, 43.1, 55.80, 55.84, 55.9, 62.7, 75.3, 110.2, 110.3, 110.9, 113.8, 119.7, 124.3, 127.0, 127.1, 130.0, 131.1, 131.8, 148.8, 148.9, 158.9, 169.9, 171.0. EIMS *m*/*z* (%): 442 (M^+, 48), 322 (93), 167 (100), 147 (81), 121 (50). HREIMS *m*/*z* M^+: calcd. for $C_{25}H_{30}O_7$, 442.1992; found, 442.1992. (1*R*,2*R*)-**10**: method A, 86% yield, $[\alpha]_D^{20} = +5$ (*c* 0.7, $CHCl_3$).

*(*E*)-(4S,5S)-5-(3,4-Dimethoxyphenyl)-1-(4-methoxyphenyl)-5-(triisopropylsilyloxy)-4-(triisopropylsilyloxy)methyl-1-pentene (**11**).* Method A: 32% yield. Method B: 11% yield: Z-isomer, 6% yield. Method E: 25% yield by using toluene. **11**: colorless oil, $[\alpha]_D^{20} = +15$ (*c* 0.23,

$CHCl_3$). ($[\alpha]_D^{20} = +24$ (*c* 1.4, $CHCl_3$) in the literature).[4] NMR data agreed with those in the literature.[4] *Z*-isomer: colorless oil, $[\alpha]_D^{20} = -42$ (*c* 0.10, $CHCl_3$). ^{1}H-NMR ($CDCl_3$) δ: 0.95–1.00 (42H, m), 1.00–1.08 (1H, overlapped), 1.62 (1H, dd, $J = 13.9$, 11.6 Hz), 2.20 (1H, m), 3.11 (1H, dd, $J = 10.5$, 10.5 Hz), 3.59 (1H, dd, $J = 10.5$, 4.8 Hz), 3.81 (3H, s), 3.87 (3H, s), 3.90 (3H, s), 4.77 (1H, s), 5.12 (1H, s), 5.28 (1H, d, $J = 3.8$ Hz), 6.80–6.83 (3H, m), 6.92 (1H, d, $J = 9.9$ Hz), 6.98 (1H, s), 7.36 (2H, d, $J = 8.7$ Hz). ^{13}C-NMR ($CDCl_3$) δ: 11.9, 12.3, 18.0, 18.1, 31.0, 46.4, 55.3, 55.7, 55.8, 63.0, 72.8, 110.1, 110.6, 112.2, 113.5, 119.6, 127.7, 133.2, 134.4, 146.8, 147.7, 148.1, 159.1. FABMS *m/z* (%) 671 [$(M + H)^+$, 0.4]. HRFABMS *m/z* $(M + H)^+$: calcd. for $C_{39}H_{67}O_5Si_2$, 671.4528; found, 671.4531.

*(7S,8S)-Morinol C (**1**).* A reaction solution of diacetate **10** (0.21 g, 0.47 mmol) in EtOH (3.0 ml) and 1 M aq. NaOH solution (3.0 ml) was stirred at room temperature for 24 h before additions of $CHCl_3$ and H_2O. The organic solution was separated and dried (Na_2SO_4). Concentration followed by silica gel column chromatography (EtOAc/hexane = 1/1) gave **1** (0.14 g, 0.39 mmol, 83%) as a colorless oil, $[\alpha]_D^{20} = +43$ (*c* 0.72, $CHCl_3$). ($[\alpha]_D^{20} = +48$ (*c* 0.50, $CHCl_3$) in the literature).[4] The NMR spectral agreed with those in the literature.[4] >99%ee (DAICEL chiral column AD-H, detected at 271 nm, 1 ml/min, *iso*-PrOH:hexane = 1:5, t_R 19 min).

*(7R,8R)-Morinol C (**2**).* The NMR data agreed with those for (7*S*,8*S*)-morinol C, 82% yield, $[\alpha]_D^{20} = -43$ (*c* 0.33, $CHCl_3$). ($[\alpha]_D^{20} = -48$ (*c* 0.30, $CHCl_3$) in the literature).[4] The NMR spectral data also agreed with those in the literature. >99%ee (DAICEL chiral column AD-H, detected at 271 nm, 1 ml/min, *iso*-PrOH:hexane = 1:5, t_R 17 min).

*(1S,2R)-2-Allyl-1-(3,4-dimethoxyphenyl)trimethylene diacetate (**12**).* The title compound was obtained from (*S*)-4-benzyl-3-{(2*S*)-2-[(*S*)-(3,4-dimethoxyphenyl)(hydroxy)methyl]-4-pentenoyl}-2-oxazolidinone[11] by the same method as that described for the synthesis of **5** and acetylation in 84% yield through 2 steps as a colorless oil, $[\alpha]_D^{20} = -35$ (*c* 1.1, $CHCl_3$). ^{1}H-NMR ($CDCl_3$) δ: 2.02 (3H, s), 2.09 (3H, s), 2.15–2.33 (3H, m), 3.78 (1H, dd, $J = 11.3$, 4.6 Hz), 3.86 (3H, s), 3.88 (3H, s), 4.02 (1H, dd, $J = 11.3$, 5.3 Hz), 5.06 (1H, d, $J = 14.2$ Hz), 5.07 (1H, d, $J = 12.4$ Hz), 5.71–5.81 (1H, m), 5.76 (1H, d, $J = 7.2$ Hz), 6.80–6.87 (3H, m). ^{13}C-NMR ($CDCl_3$) δ: 20.7, 21.1, 31.7, 42.7, 55.78, 55.81, 63.1, 75.3, 109.7, 110.9, 117.2, 119.2, 131.0, 135.4, 148.7, 148.9, 169.9, 170.7. *Anal*. Found: C, 63.98; H, 7.16%. Calcd. for $C_{18}H_{24}O_6$: C, 64.27%; H, 7.19%. (1*R*,2*S*)-**12**: 88% yield, $[\alpha]_D^{20} = +36$ (*c* 1.0, $CHCl_3$).

*(1S,2R)-1-(3,4-Dimethoxyphenyl)-2-[(E)-3-(4-methoxyphenyl)-2-propen-1-yl]trimethylene diacetate (**13**).* Method A: 82% yield. Method E: by using CH_2Cl_2, 21% yield; by using toluene, 13% yield. **13**: colorless oil, $[\alpha]_D^{20} = -20$ (*c* 0.21, $CHCl_3$). ^{1}H-NMR ($CDCl_3$) δ: 2.02 (3H, s), 2.09 (3H, s), 2.25–2.38 (2H, m), 2.42 (1H, m), 3.80 (3H, s), 3.80–3.88 (1H, overlapped), 3.87 (3H, s), 3.88 (3H, s), 4.07 (1H, dd, $J = 11.3$, 5.0 Hz), 5.80 (1H, d, $J = 6.8$ Hz), 6.00 (1H, m), 6.34 (1H, d, $J = 15.6$ Hz), 6.82–6.88 (5H, m), 7.26 (2H, d, $J = 8.7$ Hz). ^{13}C-NMR ($CDCl_3$) δ: 20.8, 21.2, 31.1, 43.4, 55.3, 55.86, 55.90, 63.4, 75.5, 109.8, 111.0, 113.9, 119.3, 124.9, 127.1, 130.1, 131.1, 131.6, 148.8, 149.0, 158.9, 170.1, 170.8. EIMS *m/z* (%) 442 (M^+, 42), 322 (100), 291 (55), 167 (64), 151 (82), 147 (68), 121 (50). HREIMS *m/z* M^+: calcd. for $C_{25}H_{30}O_7$, 442.1992; found, 442.1990. (1*R*,2*S*)-**13**: method A, 86% yield, $[\alpha]_D^{20} = +23$ (*c* 0.54, $CHCl_3$).

*(7S,8R)-Morinol D (**3**).* The title compound was obtained from **13** by the same method as that described for **1** as a colorless oil in 96% yield, $[\alpha]_D^{20} = +14$ (*c* 0.33, $CHCl_3$). ($[\alpha]_D^{20} = +22$ (*c* 0.50, $CHCl_3$) in the literature).[4] The NMR spectral data also agreed with those in the literature.[4] >99%ee (DAICEL chiral column AD-H, detected at 271 nm, 1 ml/min, *iso*-PrOH:hexane = 1:1, t_R 5.7 min).

*(7R,8S)-Morinol D (**4**).* The NMR data agreed with those for (7*S*,8*R*)-morinol D, 91% yield, $[\alpha]_D^{20} = -14$ (*c* 0.50, $CHCl_3$). ($[\alpha]_D^{20} = -22$ (*c* 0.40, $CHCl_3$) in the literature).[4] The NMR spectral data also agreed with those in the literature.[4] >99%ee (DAICEL chiral column AD-H, detected at 271 nm, 1 ml/min, *iso*-PrOH:hexane = 1:1, t_R 5.0 min).

Assay of cytotoxicity. The cytotoxicity of each compound was determined by a WST-8 assay (Kishida Reagents Chemicals, Osaka, Japan). Viable cells produce WST-8 formazan which is detectable by the absorbance at 450 nm. U266 cells were seeded in the culture medium containing various concentrations of each compound and cultured for 24 h. The WST-8 solution was added to the culture medium at 10% and incubated for 2 h, after which the absorbance at 450 nm was measured.

Organisms. Bacillus subtilis subsp. *subtiis* NBRC 13719^T, *Pseudomonas fluorescens* NBRC 14160^T, and *Staphylococcus aureus* subsp. *aureus* NBRC 14462 were purchased from the Biological Resource Center of National Institute of Technology and Evaluation (NITE), Biological Resource Center, Japan. *Escherichia coli* JCM 1649, *Listeria denitrificans* JCM 11481, *Salmonella choleraesuis* subsp. *choleraesuis* JCM 6977 and *Yersinia intermedia* JCM 7579 were obtained from RIKEN, Japan. The phytopathogenic fungi, *Colletotrichum lagenarium*, *Bipolaris oryzae*, *Fusarium solani*, and *Alternaria alternata*, had been isolated from a farm at Ehime University and were kindly presented by Dr. Ohguchi. Each fungus was cultured on potato dextrose agar (PDA: Sigma-Aldrich, Canada).

Antimicrobial assay. The antimicrobial assay was performed by the same method as that described in the literature.[12,15]

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References and Notes

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5) Method A: 4-methoxyphenylboronic acid, $Cu(OAc)_2$, LiOAc, and $Pd(OAc)_2$ in DMF. Method B: 1-iodo-4-methoxybenzene, $Pd(OAc)_2$, Et_3N in CH_3CN. Method C: 1-bromo-4-methylbenzene, $PdCl_2(PPh_3)_2$ and Et_3N in DMF. Method D: 1-iodo-4-methoxybenzene, $Pd(OAc)_2$ and K_2CO_3 in aqueous DMF. Method E: 4-methoxystyrene in the presence of 2nd Grubbs catalyst.
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