

Yaequinolones, New Insecticidal Antibiotics Produced by *Penicillium* sp. FKI-2140

II. Structural Elucidation

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Abstract The structure and relative stereochemistry of yaequinolones, fungal insecticidal antibiotics, were elucidated by spectroscopic studies, including NMR spectral analyses. Yaequinolones possess a *p*-methoxyphenylquinolinone skeleton modified with different isoprenyl-derived side chains.

Keywords yaequinolone, insecticide, fungal metabolites, structural elucidation, *Penicillium* sp., *p*-methoxyphenylquinolinone

Introduction

Yaequinolones and structurally related compounds (**1**–**16**, Fig. 1) were isolated from the culture broth of *Penicillium* sp. FKI-2140 as insecticidal antibiotics against brine shrimp (*Artemia salina*). The fermentation, isolation and biological activity were described in the preceding paper [1]. In this study, we describe the elucidation of the structure of **1**–**7** and show that these compounds possess a quinolinone core with a variety of isoprenyl-derived side chains. In progress of this study, compounds **1** and **2** were identified as 3*R**,4*S**-dihydroxy-3,4-dihydro-4-(4'-methoxyphenyl)-2(1*H*)-quinolinone and 3*R**,4*R**-dihydroxy-3,4-dihydro-4-(4'-methoxyphenyl)-2(1*H*)-quinolinone [3], respectively. The structural elucidation of **8** and **9**, having a *p*-methoxyphenylquinolinone skeleton fused with an

isoprenyl pyran ring, was reported elsewhere [2].

Results

Structural Elucidation

The physico-chemical properties and ¹H and ¹³C NMR data of **1** to **7** are summarized in Tables 1, 2 and 3. ¹H and ¹³C NMR data of **13** and **14** are also shown for comparison. From the MS, ¹H and ¹³C NMR (data not shown), **10** to **16** were identified as 4-hydroxy-3,4-dihydro-3-methoxy-4-(4'-methoxyphenyl)-2(1*H*)-quinolinone (quinolinone A, **10**) [4], 4,5-dihydroxy-3,4-dihydro-3-methoxy-4-(4'-methoxyphenyl)-2(1*H*)-quinolinone (quinolinone B, **11**) [4], peniprequinolone (**12**) [5], penigequinolones A and B (**13**, **14**) [5, 6], 4'-methoxycyclopeptin (**15**) [5] and *trans*-dehydro-4'-methoxycyclopeptin (**16**) [7]. Compounds **1** and **2** were named yaequinolones A1 and A2 in this study, therefore these structural elucidations were described below.

Yaequinolones A1 (**1**) and A2 (**2**): The molecular formulas of **1** and **2** were revealed to be C₁₆H₁₅NO₄ by HR-FAB-MS, thus requiring 10 degrees of unsaturation. The similarity in the ¹³C NMR spectra (Table 3) of **1** and **2** strongly suggested that they have the same planar structure. The ¹³C NMR spectra of **1** and **2** showed 16 carbons, which were classified into 1 methyl, 8 *sp*² methine, 1 *sp*³ methine, and 6 quaternary carbons by analysis of the DEPT spectra. The connectivity of proton and carbon atoms was

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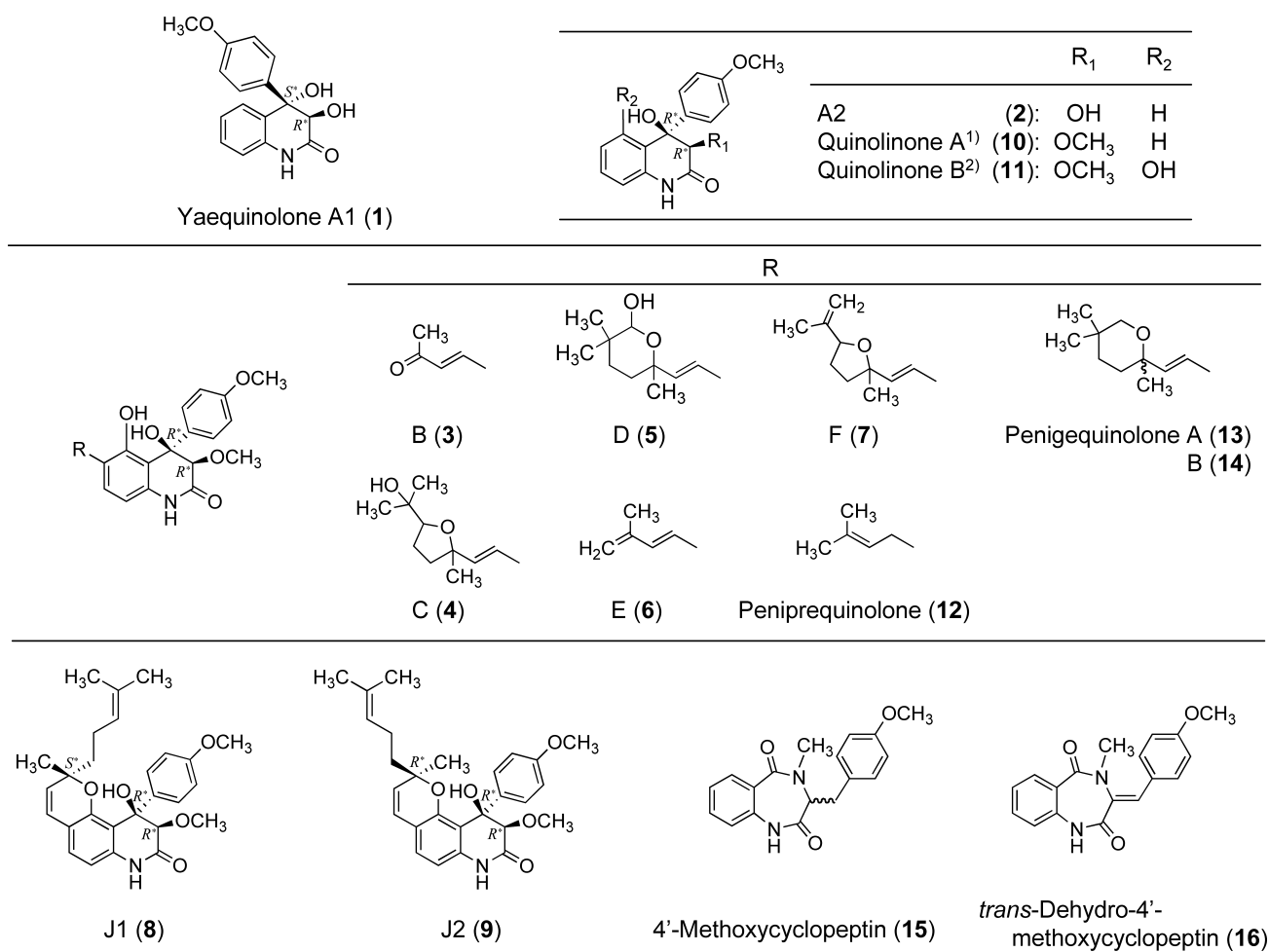


Fig. 1 Structures of yaequinolones and related compounds.

1) Quinolinone A: 4-Hydroxy-3,4-dihydro-3-methoxy-4-(4'-methoxyphenyl)-2(1*H*)-quinolinone. 2) Quinolinone B: 4,5-Dihydroxy-3,4-dihydro-3-methoxy-4-(4'-methoxyphenyl)-2(1*H*)-quinolinone.

established according to the HMQC spectra. As shown by the bold lines for **1** and **2** in Fig. 2, a partial structure I (–CH=CH–CH=CH–) and two partial structures II (–CH=CH–) become clear from the ¹H–¹H COSY spectra. The ¹³C–¹H long-range couplings of ²J and ³J in the HMBC spectra (Fig. 2) proved the following linkages. 1) The long-range couplings from H5 (δ 7.68) to C8a (δ 133.4) and from H8 (δ 6.85) to C4a (δ 130.3) suggested the presence of a 1,2-disubstituted benzene ring containing the partial structure I. 2) The long range couplings from H2'(H6') (δ 7.21) to C4' (δ 159.5) and from H3'(H5') (δ 6.73) to C1' (δ 131.6) suggested the presence of a 1,4-disubstituted benzene ring containing the two partial structures II. Furthermore, long-range coupling was observed from oxymethyl proton 4'-OCH₃ (δ 3.71) to C4', indicating that this benzene ring is a *p*-methoxyphenyl group. 3) The long-range couplings from H3 (δ 4.72) to C4 (δ 77.1), C4a, C2

(δ 170.7) and C1' and from H5 and H2'(H6') to C4 suggested that the *p*-methoxyphenyl group is attached to C4. 4) The presence of an amide group (δ_H 8.07, δ_C 170.7) was indicated by the NMR data. Considering the degree of unsaturation, it was thought that a quinolinone ring was formed from the disubstituted benzene ring and the amide group. Furthermore, considering the molecular formula, the two remaining protons should exist as two hydroxy groups at C3 and C4 of the quinolinone ring. Thus, identical planar structures were elucidated for **1** and **2**, as shown in Fig. 2.

Yaequinolone B (**3**): The molecular formula of **3** was determined to be C₂₁H₂₁NO₆ by HR-FAB-MS. The ¹H and ¹³C NMR spectra (Tables 2 and 3) resembled those of **2** except for the carbon signals of C3, C5 and C6. The differences are explained below. As shown by the bold lines for **3** in Fig. 2, the partial structure III (–CH=CH–) became clear from the ¹H–¹H COSY spectra, and the coupling

Table 1 Physico-chemical properties of yaequinolones **1** to **7**

| | 1 | 2 | 3 | 4 |
|---|---|--|---|---|
| Appearance | Pale yellow powder | Pale yellow powder | Pale yellow powder | Pale yellow powder |
| Molecular weight | 285 | 285 | 383 | 483 |
| Molecular formula | C ₁₆ H ₁₅ NO ₄ | C ₁₆ H ₁₅ NO ₄ | C ₂₁ H ₂₁ NO ₆ | C ₂₇ H ₃₃ NO ₇ |
| HRFAB-MS | | | | |
| calcd | 308.0899 (M+Na) ⁺ | 308.0899 (M+Na) ⁺ | 382.1291 (M-H) ⁻ | 483.2257 (M) ⁺ |
| found | 308.0888 (M+Na) ⁺ | 308.0884 (M+Na) ⁺ | 382.1306 (M-H) ⁻ | 483.2265 (M) ⁺ |
| UV λ _{max} ^{EtOH} nm (ε) | 209 (40,800), 226 (19,200), 250 (12,700), 280 (4,600) | 208 (43,000), 227 (18,900), 250 (11,300), 283 (5,100) | 228 (15,600), 334 (7,900), 357 (8,900) | 220 (25,900), 279 (13,800), 290 (10,400) 324 (12,900) |
| IR ν _{max} ^{KBr} cm ⁻¹ | 3426, 2910, 1683, 1606, 1508, 1301, 1247, 1178, 1137, 1027, 759 | 3253, 2925, 1689, 1602, 1509, 1380, 1253, 1174, 1081, 1033, 811, 769 | 3428, 2925, 1629, 1259, 1031, 806 | 3315, 2964, 1689, 1608, 1509, 1378, 1257, 1172, 1081, 1033, 806 |
| [α] _D ²³ (EtOH) | -32.2 (c 0.1) | -50.9 (c 0.1) | +41.2 (c 0.1) | +32.4 (c 0.1) |
| Solubility | | | | |
| soluble | CHCl ₃ , MeOH, Acetone, EtOAc, DMSO | CHCl ₃ , MeOH, Acetone, EtOAc, DMSO | CHCl ₃ , MeOH, Acetone, EtOAc, DMSO | CHCl ₃ , MeOH, Acetone, EtOAc, DMSO |
| insoluble | Hexane, H ₂ O | Hexane, H ₂ O | Hexane, H ₂ O | Hexane, H ₂ O |
| | 5 | 6 | 7 | |
| Appearance | Pale yellow powder | Pale yellow powder | Pale yellow powder | |
| Molecular weight | 483 | 381 | 465 | |
| Molecular formula | C ₂₇ H ₃₃ NO ₇ | C ₂₂ H ₂₃ NO ₅ | C ₂₇ H ₃₁ NO ₆ | |
| HRFAB-MS | | | | |
| calcd | 506.2155 (M+Na) ⁺ | 381.1576 (M) ⁺ | 465.2151 (M) ⁺ | |
| found | 506.2156 (M+Na) ⁺ | 381.1574 (M) ⁺ | 465.2151 (M) ⁺ | |
| UV λ _{max} ^{EtOH} nm (ε) | 221 (20,700), 279 (12,600), 288 (9,500), 324 (10,700) | 223 (18,800), 280 (8,400), 291 (8,300), 324 (6,800) | 219 (20,600), 279 (11,800), 287 (9,100), 324 (10,800) | |
| IR ν _{max} ^{KBr} cm ⁻¹ | 3428, 2937, 1687, 1610, 1511, 1378, 1257, 1176, 1072, 1033, 825 | 2937, 1687, 1600, 1255, 1174, 1076, 1037, 823 | 3426, 2925, 1689, 1608, 1461, 1261, 1093, 1031, 806 | |
| [α] _D ²³ (EtOH) | +56.1 (c 0.1) | +51.2 (c 0.1) | +76.9 (c 0.1) | |
| Solubility | | | | |
| soluble | CHCl ₃ , MeOH, Acetone, EtOAc, DMSO | CHCl ₃ , MeOH, Acetone, EtOAc, DMSO | CHCl ₃ , MeOH, Acetone, EtOAc, DMSO | |
| insoluble | Hexane, H ₂ O | Hexane, H ₂ O | Hexane, H ₂ O | |

constant (16.5 Hz) observed between H1'' (δ 7.80) and H2'' (δ 6.70) indicated that these olefin protons are oriented in the *trans* position. According to HMBC experiments, the cross peaks from H1'' and H₃4'' (δ 2.35) to C3'' (δ 199.4) showed the presence of an (*E*)-3-oxo-1-butenyl group containing the partial structure III. Furthermore, the cross peaks from H7 (δ 7.50) to C1'' (δ 138.1), from H1'' to C7 (δ 129.7) and from H2'' to C6 (δ 119.3) indicated that the (*E*)-3-oxo-1-butenyl group is attached to C6. The cross peaks from the methoxy protons of 3-OCH₃ (δ 3.62) to C3 (δ 84.0) and from H3 (δ 3.72) to the methoxy carbon (δ 59.7), indicated that the methoxy group is connected to C3.

Finally, the cross peaks from OH5 (δ 9.45) to C5 (δ 157.4) and C6 indicated the hydroxy group exists at C5. Thus, the planar structure for **3** was elucidated to be that shown in Fig. 2, which satisfied the molecular formula.

Yaequinolone C (**4**): The molecular formula of **4** was determined to be C₂₇H₃₃NO₇ by HR-FAB-MS, thus requiring 12 degrees of unsaturation. The ¹H and ¹³C NMR spectra (Tables 2 and 3) resembled those of **13** or **14** except for the carbon signals of the tetrahydropyran ring. As shown by the bold lines for **4** in Fig. 2, the partial structure IV (-CH₂-CH₂-CH-) became clear from the ¹H-¹H COSY spectrum. According to HMBC experiments, the cross

Table 2 ¹H NMR spectral data of yaequinolones **1** to **7**, **13** and **14**

| Position | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 13 | 14 |
|---------------------|-------------------|---------------------|----------------|----------------------------|----------------------------|----------------|---------------------|----------------------|----------------------|
| 3 | 4.72, s | 4.84, s | 3.72, s | 3.68, s | 3.71, d (1.1) | 3.70, d (1.0) | 3.68, d (1.1) | 3.69, s | 3.70, s |
| 5 | 7.68, br, d (7.7) | 6.78, dd (8.0, 1.1) | | | | | | | |
| 6 | 7.20, br, t (7.7) | 6.97, dt (8.0, 1.1) | | | | | | | |
| 7 | 7.30, br, t (7.7) | 7.29, dt (8.0, 1.1) | 7.50, d (8.1) | 7.35, d (8.4) | 7.35, d (8.3) | 7.45, d (8.4) | 7.35, d (8.1) | 7.38, d (8.3) | 7.39, d (8.3) |
| 8 | 6.85, d (7.7) | 6.86, dd (8.0, 1.1) | 6.40, d (8.1) | 6.32, d (8.4) | 6.35, d (8.3) | 6.34, d (8.4) | 6.31, d (8.1) | 6.36, d (8.3) | 6.36, d (8.3) |
| 2', 6' | 7.21, d (7.8) | 7.46, d (8.9) | 7.16, d (8.3) | 7.16, d (8.4) | 7.18, d (9.0) | 7.17, d (8.9) | 7.17, d (9.2) | 7.19, d (8.8) | 7.18, d (8.8) |
| 3', 5' | 6.73, d (7.8) | 6.98, d (8.9) | 6.84, d (8.3) | 6.81, d (8.4) | 6.82, d (9.0) | 6.82, d (8.9) | 6.82, d (9.2) | 6.82, d (8.8) | 6.81, d (8.8) |
| 1" | | | 7.80, d (16.5) | 6.77, d (15.7) | 6.69, d (16.7) | 6.80, d (16.5) | 6.84, d (16.0) | 6.73, d (16.6) | 6.73, d (16.6) |
| 2" | | | 6.70, d (16.5) | 6.24, d (15.7) | 6.14, d (16.7) | 6.84, d (16.5) | 6.33, d (16.0) | 6.14, d (16.6) | 6.13, d (16.6) |
| 4" | | | 2.35, s | 2.00, ddd (12.4, 7.5, 2.0) | 1.76, ddd (13.7, 4.2, 3.1) | 5.06, br, s | 2.03, m | 1.81, m | 1.81, m |
| | | | | 1.78, ddd (12.4, 7.2, 2.4) | 1.69, dt (13.7, 3.1) | 5.02, br, s | 1.87, m | 1.71, m | 1.71, m |
| 5" | | | | 1.91, ddd (15.0, 6.6, 2.0) | 1.52, dt (13.0, 3.1) | 1.95, s | 2.07, m | 1.45, m | 1.45, m |
| | | | | 1.86, ddd (15.0, 7.5, 2.4) | 1.36, m | | 1.79, m | 1.33, m | 1.33, m |
| 6" | | | | 3.89, dd (7.2, 6.6) | | | 4.46, dd (8.7, 6.4) | | |
| 7" | | | | 1.41, s | 4.60, s | | 1.42, s | 3.38, d (11.2) | 3.37, d (11.2) |
| 8" | | | | 1.34, s | 1.34, s | | 1.30, s | 3.23, dd (11.2, 2.0) | 3.22, dd (11.2, 2.0) |
| 9" | | | | 1.24, s | 0.87, s | | 5.08, br, s | 1.30, s | 1.30, s |
| | | | | | | | 4.82, br, s | 0.79, s | 0.79, s |
| 10" | | | | 1.13, s | 0.95, s | | 1.75, s | 1.00, s | 1.00, s |
| 3-OCH ₃ | | | | 3.60, s | 3.60, s | | 3.60, s | 3.61, s | 3.61, s |
| 4'-OCH ₃ | 3.71, s | 3.85, s | 3.62, s | 3.76, s | 3.76, s | 3.61, s | 3.76, s | 3.75, s | 3.75, s |
| NH | 8.07, br, s | 7.70, br, s | 7.50, br, s | 7.28, br, s | 7.75, br, s | 7.37, br, s | 7.44, br, s | 8.44, br, s | 8.44, br, s |
| 3-OH | ND | ND | | | | | | | |
| 4-OH | ND | ND | 4.61, br, s | 4.35, br, s | 4.56, br, s | 4.56, br, s | 4.53, br, s | 4.60, br, s | 4.60, br, s |
| 5-OH | | | 9.45, br, s | 9.16, br, s | 9.16, br, s | 9.17, br, s | 9.11, br, s | 9.12, br, s | 9.12, br, s |
| 7"-OH | | | | | ND | | | | |
| 8"-OH | | | | 2.18, br, s | | | | | |

Spectra taken at 600 MHz. Chemical shifts are shown with reference to CDCl₃ as 7.26 ppm. J values are given in Hz in parentheses.

Table 3 ^{13}C NMR spectral data of yaequinolones **1** to **7**, **13** and **14**

| Position | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 13 | 14 |
|---------------------|----------|----------|----------|----------|----------|----------|----------|-----------|-----------|
| 2 | 170.7 | 170.2 | 165.4 | 165.5 | 165.5 | 165.4 | 165.5 | 166.2 | 166.2 |
| 3 | 75.5 | 73.7 | 84.0 | 84.3 | 84.2 | 84.3 | 84.2 | 84.2 | 84.1 |
| 4 | 77.1 | 76.5 | 78.7 | 78.1 | 78.7 | 78.8 | 78.8 | 78.7 | 78.6 |
| 4a | 130.3 | 127.5 | 111.5 | 110.8 | 110.9 | 110.8 | 110.9 | 110.8 | 110.8 |
| 5 | 126.5 | 130.5 | 157.4 | 155.3 | 155.2 | 155.6 | 155.4 | 155.0 | 155.0 |
| 6 | 125.3 | 124.3 | 119.3 | 121.9 | 121.6 | 122.4 | 121.1 | 121.9 | 121.9 |
| 7 | 129.3 | 130.7 | 129.7 | 127.8 | 127.5 | 127.3 | 127.4 | 127.4 | 127.3 |
| 8 | 116.0 | 116.1 | 107.6 | 106.9 | 106.9 | 107.1 | 106.4 | 107.0 | 107.0 |
| 8a | 133.4 | 135.8 | 137.5 | 134.3 | 133.6 | 134.3 | 134.1 | 134.3 | 134.2 |
| 1' | 131.6 | 132.4 | 128.4 | 129.0 | 128.9 | 129.1 | 129.1 | 129.0 | 129.0 |
| 2', 6' | 128.4 | 127.9 | 127.8 | 127.9 | 127.8 | 127.9 | 127.7 | 127.4 | 127.3 |
| 3', 5' | 113.9 | 114.1 | 114.5 | 114.3 | 114.3 | 114.4 | 113.9 | 114.2 | 114.2 |
| 4' | 159.5 | 159.3 | 160.5 | 160.1 | 160.3 | 160.2 | 160.3 | 160.2 | 160.2 |
| 1'' | | | 138.1 | 121.1 | 123.4 | 122.6 | 120.4 | 123.2 | 123.1 |
| 2'' | | | 126.8 | 135.6 | 134.4 | 131.2 | 135.5 | 134.3 | 134.3 |
| 3'' | | | 199.4 | 83.3 | 77.1 | 142.9 | 83.2 | 74.4 | 74.3 |
| 4'' | | | 26.9 | 38.5 | 30.4 | 116.7 | 39.0 | 30.9 | 31.0 |
| 5'' | | | | 26.5 | 34.2 | 18.7 | 31.2 | 33.5 | 33.5 |
| 6'' | | | | 85.8 | 34.1 | | 82.3 | 29.6 | 29.6 |
| 7'' | | | | 26.7 | 97.8 | | 26.5 | 72.7 | 72.7 |
| 8'' | | | | 71.0 | 31.1 | | 145.7 | 29.2 | 29.1 |
| 9'' | | | | 27.5 | 26.2 | | 110.1 | 26.5 | 26.5 |
| 10'' | | | | 24.4 | 16.7 | | 18.5 | 24.0 | 24.0 |
| 3-OCH ₃ | | | 59.7 | 58.9 | 58.8 | 59.0 | 58.5 | 58.9 | 58.9 |
| 4'-OCH ₃ | 55.3 | 55.4 | 55.4 | 55.4 | 55.2 | 55.4 | 55.0 | 55.2 | 55.2 |

Spectra taken at 150 MHz. Chemical shifts are shown with reference to CDCl_3 as 77.0 ppm.

peaks from H5'' (δ 1.86) to C3'' (δ 83.3) and C8'' (δ 71.0), from H₃9'' (δ 1.24) to C6'' (δ 85.8), C8'' and C10'' (δ 24.4), from H₃10'' (δ 1.13) to C6'', C8'' and C9'' (δ 27.5) and from H₃7'' (δ 1.41) to C3'' and the olefinic carbon C2'' (δ 135.6) lead to a bigger partial structure V containing IV. The cross peak from H7 (δ 7.35) to C1'' (δ 121.1), from H1'' (δ 6.77) to C5 (δ 155.3) and from H2'' (δ 6.24) to C6 (δ 121.9) indicated that the partial structure V is attached to C6. Taking the remaining atoms (one hydrogen and two oxygens) and the chemical shifts of C3'', C6'' and C8'' (bound to an oxygen) into consideration, it was concluded that C3'' and C6'' should connect to the same oxygen to form a furan ring and a hydroxy group is attached to C8''. Thus, the structure of **4** was elucidated to be that shown in Fig. 2, which satisfied the molecular formula and the degree of unsaturation.

Yaequinolone D (**5**): The molecular formula of **5** was revealed to be $\text{C}_{27}\text{H}_{33}\text{NO}_7$ by HR-FAB-MS. The ^1H and ^{13}C NMR spectra (Tables 2 and 3) resembled those of **13** or **14** except for the carbon signal of C7''. The ^{13}C NMR chemical

shift of C7'' indicated the presence of the sp^3 dioxy quaternary carbon (δ 97.8), and we concluded that the hydroxy group was attached to C7''. ^1H - ^1H COSY and HMBC experiments supported the structure of **5** as shown in Fig. 2.

Yaequinolone E (**6**): The molecular formula of **6** was revealed to be $\text{C}_{22}\text{H}_{23}\text{NO}_5$ by HR-FAB-MS. The ^1H and ^{13}C NMR spectra (Tables 2 and 3) resembled those of **3** except for the carbon signal of C3''. In HMBC experiment examining **6** (Fig. 2), the cross peaks from H2'' (δ 6.84) and H₃5'' (δ 1.95) to C3'' (δ 142.9) and C4'' (δ 116.7) and from H₂4'' (δ 5.06, 5.02) to C2'' (δ 131.2) and C5'' (δ 18.7) indicated the presence of a methylene attached to C3'' instead of a ketone of **3**. Thus, the structure of **6** was elucidated to be that shown in Fig. 2.

Yaequinolone F (**7**): The molecular formula of **7** was revealed to be $\text{C}_{27}\text{H}_{31}\text{NO}_6$ by HR-FAB-MS. The ^1H and ^{13}C NMR spectra (Tables 2 and 3) resembled those of **4** except for the carbon signals of C8'' and C9''. In HMBC experiments, the cross peaks from H6'' (δ 4.46) to C9'' (δ

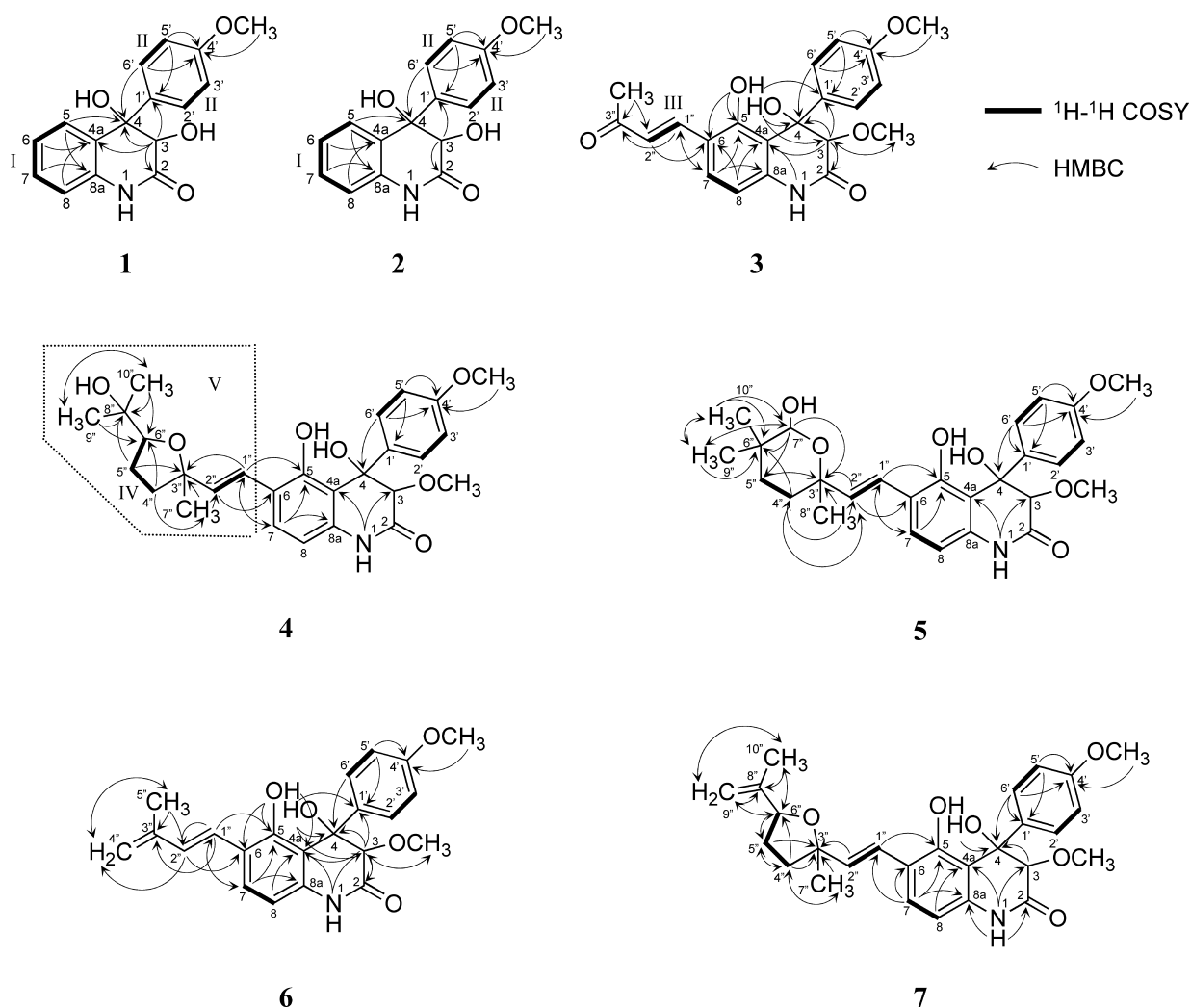


Fig. 2 ^1H - ^1H COSY and HMBC experiments of **1** to **7**.

110.1), $\text{H}_3^{10''}$ (δ 1.75) to $\text{C}8''$ (δ 145.7) and $\text{C}9''$ and from $\text{H}_2^{9''}$ (δ 5.08, 4.82) to $\text{C}6''$ (δ 82.3), $\text{C}8''$ and $\text{C}10''$ (δ 18.5) showed the presence of a methylene attached to $\text{C}8''$. Thus, the structure of **7** was elucidated to be that shown in Fig. 2.

Relative Stereochemistry of *p*-Methoxyphenyl Quinolinone Skeleton

Compounds **1** and **2** have two chiral carbons of $\text{C}3$ and $\text{C}4$ in the quinolinone ring. Their relative stereochemistry was elucidated by NOE experiments (Fig. 3). An NOE was observed between $\text{H}3$ and $\text{H}2'$ ($\text{H}6'$), but the intensity of NOE in **2** (7.1%) was much stronger than that in **1** (0.75%). These results indicated that the *p*-methoxyphenyl group and 3-OH in **1** are located on the same face of the quinolinone ring, while those in **2** are located on the opposite side. Therefore, the relative stereochemistry of **1** and **2** was determined to be $3R^*$, $4S^*$ and $3R^*$, $4R^*$, respectively.

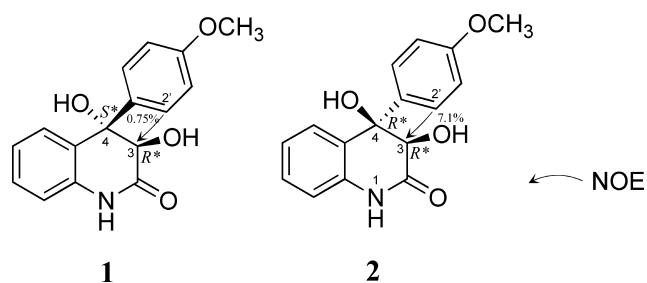


Fig. 3 NOE experiments of **1** and **2**.

NOE experiments for **3** to **7** showed similar results to those for **2**, indicating that the stereochemistry of their quinolinone skeleton is $3R^*$, $4R^*$.

Discussion

During this study we isolated 16 structurally related compounds from the culture broth of *Penicillium* sp. FKI-2140. Compounds **15** and **16** are diketodiazepins. Compounds **1**, **2**, **10** and **11** have the fundamental *p*-methoxyphenyl quinolinone skeleton, while the others have an additional isoprenyl derived (C5 or C10 unit) side chain attached to the fundamental skeleton.

Biosynthetic studies of this series of compounds have not been reported so far, but the biosynthetic pathway could be speculated to be as follows [5]. First, diketodiazepins **15** and **16** are biosynthesized from anthranilic acid and tyrosine, which are de- and re-cyclized to form the fundamental phenylquinolinones (**1**, **2**, **10** and **11**). Similar cyclization was reported in the biosynthesis of a phenylquinolinic ring in fungal viridicatin [7, 8]. Then, an isoprenyl (C5)- or geranyl (C10)-derived side chain are linked to the skeleton to form **3**, **6** and **12** or **4**, **5**, **7**, **13** and **14**, respectively. Finally, intramolecular dehydration occurs to produce **8** and **9**.

Although the relative stereochemistry of the side chains in yaequinolones could not be defined, that of the quinolinone skeleton was determined by NOE experiments. Compound **1** has the 3*R**, 4*S** stereochemistry, while **2** to **12** have 3*R**, 4*R**. In this study **13** and **14** were separated by using a chiral column, though they were originally isolated as a mixture. They were both found to have 3*R**, 4*R** stereochemistry. Thus, only **1** has different stereochemistry, probably due to racemization during cyclization from **15** or **16**.

Experimental

Spectroscopic Measurements

NMR spectra were recorded on a Varian Inova 600 spectrometer ($^2\text{-}^3J_{\text{CH}}=8$ Hz in HMBC). Chemical shifts are shown in δ values (ppm) relative to chloroform at 7.26 ppm for ^1H NMR and chloroform- d_1 at 77.0 ppm for ^{13}C NMR. FAB mass spectrometry was conducted on a JEOL JMS-AX505H spectrometer. UV and IR spectra were measured with a Beckman DU640 spectrophotometer and a Horiba FT-210 Fourier transform infrared spectrometer, respectively.

Optical rotations were recorded on a JASCO model DIP-181 polarimeter.

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