Role of the Hydroxymethyl Group for the Inhibitory Activity of Penienone

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Penienone was isolated from the culture filtrate of Penicillium sp. No.13 and completely inhibited hypocotyl elongation and root growth of lettuce seedlings at a concentration of 300 mg/l. In order to elucidate the active site of penienone, three derivatives were prepared and tested for their inhibitory activity against the growth of lettuce seedlings. The results indicated that hydroxymethyl group in the molecule of penienone was essential to exhibit the remarkable inhibitory activity against the growth of lettuce seedlings.

Introduction

We have isolated and elucidated the structures of penienone (1) [1], penidienone [2] and penihydrone [1] from the culture filtrate of Penicillium sp. No.13 as the growth regulators of lettuce seedlings. 1 shows the inhibitory activity against the growth of lettuce seedlings. Penidienone shows weaker inhibitory activity than 1, while penihydrone does not show any remarkable inhibitory activity. To explore the structural units essential to the inhibitory activity of 1 in detail, three derivatives (Fig. 1) of 1 were prepared and tested for their inhibitory activity. Here we describe the biological activities of penienone and its derivatives on the growth of lettuce seedlings.

Results and Discussion

Penienone, penidienone and penihydrone were isolated from a culture filtrate of Penicillium sp. No.13 as described previously [1,2]. Acetylation of 1 with acetic anhydride in pyridine afforded a monoacetyl derivative (2) which was confirmed by the resonance at δ 2.01 (OAc) in the 1H NMR spectrum of 2. Oxidation of 1 with Jones' reagent [3] afforded a dehydroxymethyl derivative (3). The 13C NMR spectrum of 3 showed the absence of a signal for the hydroxymethyl group and the appearance of a new signal due to a methylene carbon at 43.9 ppm, adjacent to a carbonyl group. Catalytic hydrogenation over platinum oxide of 1 afforded a hexahydro-derivative (4). The 13C NMR spectrum of 4 showed six signals due to methane carbon atoms instead of six double-bonded methane carbon atoms in 1.

Penienone inhibited hypocotyl elongation and root growth of lettuce seedlings by 34% and 23% at a concentration of 30 mg/l, respectively. Com-
Complete inhibition was found at a concentration of 300 mg/l. The inhibitory activity of compound 2 (300 mg/l) on hypocotyl elongation and root growth was weaker than that of penienone at a concentration of 30 mg/l. Compound 3 did not show any remarkable inhibitory activity against hypocotyl elongation and root growth at a concentration of 300 mg/l. Compound 4 completely inhibited hypocotyl elongation and root growth at the same concentration, but showed no inhibitory activity at a concentration of 30 mg/l (Fig. 2). These results indicated that the hydroxymethyl group of 1 was essential to exhibit the remarkable inhibitory activity against the growth of lettuce seedlings.

**Experimental**

UV and IR spectra were recorded on a Hitachi 100–50 and a JASCO FT/IR-7000 spectrometers, respectively. $^1$H-NMR and $^{13}$C-NMR spectra were obtained on a JEOL JNM GX-270 spectrometer at 270.05 MHz and 67.80 MHz, respectively. MS spectra were taken on a INCOS 50 instrument.

**Preparation of (5S, 6R)-5-[(1E, 3E)-1,3-heptadienyl]-6-(acetoxymethyl)-2-cyclohexenone (2)**

Penienone (1) (20 mg) was acetylated with acetic anhydride (0.5 ml) and pyridine (1.0 ml) for 24 h at room temperature and worked up in the usual manner to yield 2 (18 mg) as a colorless oil.

**Compound 2**: UV (EtOH) nm (e): 230 (22,500); IR (KBr) cm$^{-1}$: 2964 (C= C), 1743 (O=C=O), 1680

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Fig. 2. Effect of penienone (1) and three derivatives (2–4) on the growth of lettuce seedlings. Concentration: (left) 30 mg/l, (right) 300 mg/l.
(C=O); MS: m/z (%) = 262 (M+20), 202 (38), 189 (47), 173 (17), 159 (100), 145 (63), 133 (57), 117 (34), 107 (43), 91 (73), 77 (49), 68 (65); 13C NMR (CDCl3/TMS): δ = 13.7 (C-13), 20.8 (CH3-20), 22.3 (C-12), 23.4 (C-4), 34.6 (C-11), 40.5 (C-5), 50.5 (C-6), 60.6 (CH3-OAc), 129.5 (C-9), 129.6 (C-2), 131.0 (C-7), 132.4 (C-8), 135.0 (C-10), 148.7 (C-3), 170.8 (CH=CO), 197.1 (C-1) ppm; 1H NMR (CDCl3/TMS): δ = 0.91 (t, 3H, J = 7.3, 13-H), 1.41 (tq, 2H, J = 7.3, 7.3, 12-H), 2.01 (s, 3H, OAc), 2.03 (dt, 2H, J = 7.3, 7.3, 11-H), 2.43 (m, 3H, 4-H, 6-H), 2.82 (m, 1H, 5-H), 4.20 (dd, 1H, J = 11.1, 3.9, CH2-OAc), 4.58 (dd, 1H, J = 14.7, 7.3, CH2-OAc), 5.46 (dd, 1H, J = 14.7, 8.8, 7-H), 5.65 (dt, 1H, J = 14.7, 7.3, 10-H), 5.96 (m, 1H, 9-H), 6.05 (m, 1H, 2-H), 6.08 (m, 1H, 8-H), 6.95 ppm (dd, 1H, J = 10.3, 5.5, 2.4, 3-H).

Preparation of (5S)-5-[(1E,3E)-1,3-heptadienyl]-2-cyclohexenone (3)

Penienone (1) (40 mg) was treated with Jones' reagent [3] in acetone (2.0 ml) for 12 h in an ice-cooled water bath and worked up in the usual manner to yield 3 (6 mg) as a colorless oil.

Compound 3: UV (EtOH) nm (ε): 225 (1,900); IR (KBr) cm⁻¹: 2930 (C=O), 1704 (C=O); MS: m/z (%) = 226 (M+13), 208 (28), 137 (22), 127 (73), 110 (77), 97 (100), 83 (88), 67 (73); 13C NMR (CDCl3/TMS): δ = 14.1 (C-13), 22.6 (C-7-12), 25.9 (C-4), 26.0 (C-5), 29.2 (C-7-12), 29.8 (C-7-12), 30.3 (C-7-12), 31.8 (C-7-12), 33.5 (C-7-12), 40.3 (C-3), 42.1 (C-6), 56.8 (C-2), 59.7 (C-14), 215.5 (C-1) ppm; 1H NMR (CDCl3/TMS): δ = 0.88 (t, 3H, J = 7.3, 13-H), 1.27 (br, s, 12H, 7-12-H), 1.45 (ddd, 1H, J = 13.7, 3.7, 2.0, 4-H), 1.62 (m, 1H, 5-H), 1.70 (m, 1H, 3-H), 1.97 (ddd, 1H, J = 13.7, 3.7, 2.0, 4-H), 2.09 (ddd, 1H, J = 14.7, 7.1, 3.7, 5-H), 2.25 (ddd, 1H, J = 11.5, 6.5, 2.8, 2-H), 2.32 (m, 1H, 6-H), 2.38 (m, 1H, 6-H), 2.70 (m, 1H, OH), 3.71 (m, 1H, 14-H), 3.85 ppm (m, 1H, 14-H).

Bioassay

The growth of lettuce seedlings treated with the compounds was examined by the established method [4]. The hypocotyl and root lengths of the seedlings after treatment were measured and compared with the values for an untreated control. Bioassay was performed three times and the mean values are shown in Fig. 2.

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References