The structure proposed for apteniol D is different from that of the compound obtained by total synthesis

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ABSTRACT

We describe the synthesis of 4,4'-oxyneolignan, the proposed structure for naturally occurring apteniol D. The diphenyl ether moiety in 4,4'-oxyneolignan was formed via classical Ullmann ether synthesis using excess copper powder in N,N-dimethylacetamide. The spectral data of synthesised apteniol D show differences compared to those of naturally occurring apteniol D.

1. Introduction

Apteniols A-G (1–7, Figure 1), isolated from A. cordifolia by DellaGreca in 2005 and 2007, are oxyneolignans, and are reported to be secondary metabolites that inhibit the germination of lettuce seeds (DellaGreca et al. 2005, 2007). Compounds proposed to have the same structures as apteniols A, B, C and G were synthesised in our laboratories (Nishikawa et al. 2014; Noshita et al. 2015), and a compound with the proposed structure of apteniol C was synthesized by Jung and Bräse (2009); however, the spectral data of the naturally occurring apteniols were not consistent with the spectral data of the synthesised versions. To confirm the structures of the remaining apteniols, we now report the synthesis of the compound with the structure proposed for apteniol D (4).
2. Results and discussion

The synthetic route for the synthesis of apteniol D is shown in Figure 2. Formation of the diphenyl ether, the key step in this synthesis, was performed via the Ullmann ether synthesis that was previously used in the preparation of apteniols A, B, C and G (Nishikawa et al. 2014; Noshita et al. 2015). First, coupling of vanilline and 4-bromo-3,5-dimethylbenzaldehyde via either the Ullmann ether synthesis or the Buchwald–Hartwig reaction was examined (data not shown). However, the reaction did not proceed under the conditions used for the Buchwald–Hartwig reaction. The desired diphenyl ether was also not obtained using the Ullmann ether synthesis; instead, 3,5-dimethoxybenzaldehyde, the product formed from reductive elimination of bromine, was obtained. In contrast, Ullmann etherification using syringaldehyde and 4-bromo-3-methoxybenzaldehyde in a sealed tube at 200 °C produced the desired compound 8 in 29% yield. Because about half of aldehyde without reacting are recovered, the yield of this reaction is low. In this step, 8 was obtained using the classical conditions, heating at 200 °C with excessive copper powder in N,N-dimethylacetamide (Shioe et al. 2013). Coupling of the phenol and aryl halide to form 8 is supported by the presence of two aldehyde carbon peaks at 189.5 and 190.9 ppm in the 13C NMR spectrum and detection of the pseudomolecular ion at m/z 317.1025 [M + H]+, consistent with the molecular formula of C17H16O6, using high-resolution fast atom bombardment mass spectrometry (HRFAB–MS). The formyl groups in 8 were then converted into the α,β-unsaturated diethyl ester 9 via the Horner–Wadsworth–Emmons reaction. Catalytic hydrogenation and subsequent hydrolysis of the ester groups afforded the desired dicarboxylic acid that corresponds to the proposed structure of apteniol D (4). The chemical structures of all the synthesised compounds were determined by 1H and 13C NMR spectroscopy and HR–MS analyses. The 1H and 13C NMR spectral data for 4 synthesized in this work and the data previously reported for 4 are shown in Table S1 (supplementary material).

The reported NMR data (see Table S1) for natural apteniol D (4) are similar to those of synthesised 4; however, differences exist between the data, even though they were obtained using the same solvent. Distinct differences are observed in the 1H NMR chemical shifts of H-5′, H-7′ and H-8′, which differ by 0.45, 0.19 and 0.22 ppm, respectively. In addition, the 1H NMR coupling patterns observed in the peaks for H-2′ and H-6′ for synthesised 4 (doublet and double-of-doublets, respectively) are different from naturally occurring 4 (singlet and doublet, respectively). In the 13C NMR spectra, the C-3, C-5, C-3′ and C-4′ carbon signals show more than 6 ppm differences when comparing synthesised 4 to naturally occurring 4.

Figure 1. Proposed structures of apteniols A–G.
At this stage, it is unclear if the correct structure was determined for naturally occurring 4; however, given the synthetic route that was used and the NMR and HRFAB–MS data, the structure determined for synthesised 4 is correct. The differences in the NMR data between synthesised and naturally occurring 4 suggest the possibility of an error in the interpretation of the data used for the structural determination of naturally occurring 4. Or this difference may come from rotational isomer. As shown in Figure 3, rotational isomers may exist because the diphenyl ether has two rotation axes (C-4-O and C-4′-O). Therefore, the difference in both spectra may come from these isomers (Mazzocchi et al. 1978; Feigel 1996; Duong et al. 2015).

3. Experimental

3.1. General experimental procedures

Melting points were measured using an MP-J3 (Yanaco, Kyoto, Japan), and are uncorrected. The IR spectra were obtained using a Nicolet iS10 FT-IR spectrometer (Thermo Fisher Scientific, Waltham, MA, U.S.A) with a diamond horizontal attenuated reflectance (ATR) accessory and co-addition of 16 interferograms. Calibration models were generated using OMNIC 9.2.98 software. The 1H and 13C NMR spectra were recorded using an Agilent 400-MR DD2 (Agilent, SantaClara CA, U.S.A) spectrometer with tetramethylsilane as the internal standard. Mass spectra were recorded using a JMS-700 (JEOL, Tokyo, Japan) mass spectrometer.
Column chromatography was performed on silica gel 60 N (100–210 mesh, Kanto Chemical Co. Tokyo, Japan). All chemicals were reagent grade and used as received, without further purification.

3.2. Synthesis of proposed apteniol D

3.2.1. Dialdehyde 8; 4-(4-formy-2-methoxyphenoxy)-3,5-dimethoxybenzaldehyde

A mixture of syringaldehyde (550 mg, 3.0 mmol), 4-bromo-3-methoxy-benzaldehyde (1610 mg, 7.5 mmol), copper powder (1140 mg, 18 mgatom, 99.5% purity) and \( N,N \)-dimethylacetamide (7.5 ml) was heated to 200 °C in a sealed tube under an \( \text{N}_2 \) atmosphere for 4 h with stirring. The cooled mixture was then filtrated; the filtrate was poured into water and extracted three times with \( \text{CH}_2\text{Cl}_2 \). The combined organic layers were washed with brine and extracted three times with \( \text{CH}_2\text{Cl}_2 \). The combined organic layers were washed with brine and dried over \( \text{Na}_2\text{SO}_4 \). Evaporation of the solvent and purification of the residue by column chromatography (\( \text{n}-\text{hexane/ethyl acetate} = 4:1 \)) on silica gel yielded dialdehyde 8 (280 mg, 0.87 mmol, 29%) as a pale yellow oil. \( \text{iR} \nu_{\max} \) (diamond ATR) \( \text{cm}^{-1} \): 2948, 1690, 1676, 1633, 1598, 1489, 1462, 1332, 1226 and 1026. \( \text{H NMR} \delta \) (400 MHz in CDCl\(_3\)): 3.82 (3H, s), 3.87 (6H, s), 6.58 (1H, d, \( J = 9.1 \text{ Hz} \)), 6.98 (1H, dd, \( J = 9.1, 3.2 \text{ Hz} \)), 7.21 (2H, s), 7.38 (1H, d, \( J = 3.2 \text{ Hz} \)), 9.95 (1H, s), 10.70 (1H, s). \( \text{C NMR} \delta \) (100 MHz in CDCl\(_3\)): 55.8, 56.4, 106.5, 109.5, 116.7, 123.3, 125.5, 133.7, 153.9, 154.9, 155.2, 189.5, 190.9. HRFAB–MS \( m/z \) [M + H\(^+\)]: Calcd. for C\(_{17}\)H\(_{17}\)O\(_6\): 317.1025, found: 317.1025.

3.2.2. Diester 9; Ethyl (E)-3-(4-(4-(E)-3-ethoxy-3-oxoprop-1-en-1-yl)-2,6-dimethoxyphenoxy)-3-methoxyphenyl) acrylate

Sodium hydride (60% dispersion in mineral oil, 120 mg, 3.0 mmol) was washed with dry \( \text{n}-\text{hexane} \) and suspended in 5 ml of dry tetrahydrofuran (THF). This suspension was added dropwise to a solution of triethyl phosphonoacetate (320 mg, 1.4 mmol) at ice-cooled temperature. The solution was then stirred at room temperature until gas evolution ceased. The resultant yellow solution was added dropwise to a solution of dialdehyde 8 (190 mg, 0.60 mmol) in dry THF at ice-cooled temperature. The reaction mixture was then stirred for 2 h at room temperature, poured into water and extracted three times with ethyl acetate. The combined organic layers were washed with brine and dried over CaCl\(_2\). Evaporation of the extract and purification of the residue by preparative thin layer chromatography (p-TLC) developed with \( \text{n}-\text{hexane/ethyl acetate} = 2:1 \) yielded diester 9 (104 mg, 0.23 mmol, 78%) as a pale yellow powder. Mp. 141.0–143.0 °C. IR \( \nu_{\max} \) (diamond ATR) \( \text{cm}^{-1} \): 2981, 1694, 1639, 1589, 1488, 1462, 1332, 1222, 1037 and 979. \( \text{H NMR} \delta \) (400 MHz in CDCl\(_3\)): 1.33 (3H, t, \( J = 7.1 \text{ Hz} \)), 1.35 (3H, t, \( J = 7.1 \text{ Hz} \)), 3.779 (3H, s), 3.784 (6H, s), 4.26 (2H, q, \( J = 7.1 \text{ Hz} \)), 4.28 (2H, q, \( J = 7.1 \text{ Hz} \)), 6.40 (1H, d, \( J = 15.9 \text{ Hz} \)), 6.49 (1H, d, \( J = 9.0 \text{ Hz} \)), 6.56 (1H, d, \( J = 16.1 \text{ Hz} \)), 6.74 (1H, dd, \( J = 9.0, 3.0 \text{ Hz} \)), 6.81 (2H, s), 7.07 (1H, d, \( J = 3.0 \text{ Hz} \)), 7.64 (1H, d, \( J = 15.9 \text{ Hz} \)), 8.20 (1H, d, \( J = 16.1 \text{ Hz} \)). \( \text{C NMR} \delta \) (100 MHz in CDCl\(_3\)): 14.3, 14.4, 55.7, 56.3, 60.4, 60.6, 105.3, 112.2, 115.3, 117.1, 118.3, 119.3, 124.2, 131.7, 134.9, 139.7, 143.3, 151.5, 153.6, 154.4, 166.8, 167.3. HRFAB–MS \( m/z \) [M + Na\(^+\)]: Calcd. for C\(_{25}\)H\(_{28}\)O\(_8\)Na: 479.1682, found: 479.1678.

3.2.3. Diester 10; Ethyl 3-(4-(4-(3-ethoxy-3-oxopropyl)-2,6-dimethoxyphenoxy)-3-methoxyphenyl)propanoate

A suspension of diester 9 (109 mg, 0.24 mmol) and 5% Pd/C (50 mg) in dry methanol was stirred at room temperature under hydrogen for 2 h. The reaction mixture was then filtered
and concentrated. The dry residue was purified by p-TLC (n-hexane-ethyl acetate = 2:1) to yield diester 10 (107 mg, 0.23 mmol, 97%) as a colourless oil. IR $\nu_{\text{max}}$ (diamond ATR) cm$^{-1}$: 2938, 1727, 1593, 1492, 1222 and 1039. $^1$H NMR $\delta$ (400 MHz in CDCl$_3$): 1.25 (3H, t, $J = 7.1$ Hz), 1.26 (3H, t, $J = 7.1$ Hz), 2.64 (2H, br-t, $J = 7.8$ Hz), 2.81 (2H, br-t, $J = 7.8$ Hz), 2.94 (2H, br-t, $J = 7.8$ Hz), 3.11 (2H, br-t, $J = 7.8$ Hz), 3.73 (3H, s), 3.74 (6H, s), 4.14 (2H, q, $J = 7.1$ Hz), 4.15 (2H, q, $J = 7.1$ Hz), 6.36 (1H, d, $J = 8.9$ Hz), 6.47 (2H, s), 6.53 (1H, dd, $J = 8.9$, 3.1 Hz), 6.77 (1H, d, $J = 3.1$ Hz). $^{13}$C NMR $\delta$ (100 MHz in CDCl$_3$): 14.4, 26.6, 31.6, 34.2, 36.2, 55.7, 56.2, 60.3, 60.6, 105.6, 111.8, 113.6, 115.9, 129.9, 131.7, 138.0, 150.9, 153.5, 154.1, 172.9, 173.7. HRFAB–MS $m/z$ [M + Na]$^+$: Calcd. for C$_{25}$H$_{32}$O$_8$Na: 483.1995, found: 483.1993.

3.2.4. Proposed apteniol D (4); 3-(4-(4-(2-carboxyethyl)-2,6-dimethoxyphenoxy)-3-methoxyphenyl)propanoic acid
A solution of diester 10 (107 mg, 0.23 mmol) and NaOH (100 mg, 2.5 mmol) in THF/H$_2$O (1:2, 10 mL) was stirred at room temperature for 12 h. The reaction mixture was then poured into 4% aqueous HCl and extracted three times with ethyl acetate. The combined organic layers were washed with brine and dried over CaCl$_2$. Evaporation of the extract and purification of the residue by p-TLC (CHCl$_3$–MeOH = 10:1) yielded 4 (43 mg, 0.11 mmol, 46%) as a pale yellow powder. Mp. 99.0–101.0 °C. IR $\nu_{\text{max}}$ (diamond ATR) cm$^{-1}$: 2935, 1693, 1596, 1424, 1290, 1195 and 1052. $^1$H NMR $\delta$ (400 MHz in CDCl$_3$): 2.72 (2H, t, $J = 7.2$ Hz), 2.89 (2H, t, $J = 7.8$ Hz), 2.95 (2H, t, $J = 7.2$ Hz), 3.09 (2H, t, $J = 7.8$ Hz), 3.72 (9H, s), 6.37 (1H, d, $J = 8.8$ Hz), 6.48 (2H, s), 6.54 (1H, dd, $J = 3.1$, 8.8 Hz), 6.76 (1H, d, $J = 3.1$ Hz). $^{13}$C NMR $\delta$ (100 MHz in CDCl$_3$): 26.3, 31.0, 33.6, 35.4, 56.1, 55.6, 105.5, 111.9, 113.5, 116.0, 129.3, 131.9, 137.4, 150.8, 153.4, 154.0, 178.2, 179.2. $^1$H and $^{13}$C NMR spectral data of naturally occurring apteniol D are shown in Table S1. HRFAB–MS $m/z$ [M + Na]$^+$: Calcd. for C$_{21}$H$_{24}$O$_8$Na: 427.1369, found: 427.1371.

4. Conclusion
Synthesis of apteniol D (4) was accomplished, based on the reported structure of naturally occurring 4. The $^1$H and $^{13}$C NMR data of synthesised 4 was similar but did not perfectly agree with the previously reported data from naturally occurring 4. The discrepancy in the NMR data has also been observed in the comparison of synthesised apteniols A, B, C and G in our previous works (Nishikawa et al. 2014; Noshita et al. 2015) to their reported natural product structures, suggesting that the actual structure of naturally obtained apteniols may be slightly different than their synthesised counterparts. However, it may be necessary to consider the presence of the rotational isomer. The biological activities of the synthesised apteniol D as well as compounds 8, 9 and 10 will be reported in the future.

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