Notes

PD 116,779, A NEW ANTITUMOR ANTIBIOTIC OF THE BENZ[A]ANTHRAQUINONE CLASS

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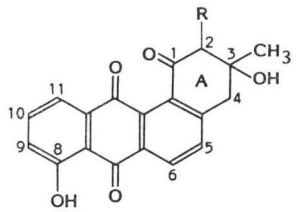
In the course of our anticancer drug discovery program, the fermentation broth of an unidentified actinomycete isolate (WP 3688) was found to exhibit *in vitro* activity against L1210 lymphocytic leukemia and HCT-8 human colon adenocarcinoma cell lines. Subsequent bioactivity-directed fractionation resulted in the isolation of a complex of antibiotics possessing the benz[a]anthracene skeleton. One of these compounds was identified as the known antibiotic, yoronomycin[1], on the basis of chemical and spectral data. A second component, PD 116,779 (**1**), proved to be a new antibiotic of the benz[a]anthraquinone class, closely related to tetrangomycin (**2**)[2]. This report describes the isolation and structure elucidation of PD 116,779, which was shown to be 2-hydroxytetrangomycin.

PD 116,779 was isolated from fermentation broths by extraction with EtOAc. Concentration of the extracts, followed by precipitation with petroleum ether, gave a crude product containing a mixture of several benz[a]anthraquinone antibiotics, as determined by TLC and UV analysis. Repeated silica gel chromatography, using $CHCl_3$ - MeOH mixtures as eluents, provided efficient separation of the antibiotic complex. Fractions containing PD 116,779 as the major component (by TLC analysis) were combined and concentrated to dryness. Crystallization of this product from $CHCl_3$ - MeOH yielded **1** as yellow needles: mp 205°C; UV λ_{max}^{MeOH} nm (ε) 266 (29,900), 398 (5,040); UV $\lambda_{max}^{MeOH+KOH}$ nm (ε) 257 (25,100), 312 (7,560), 408 (2,400), 488 (4,880); IR ν_{max}^{KBr} cm^{-1} 1702, 1669, 1637, 1591, 1280; ^1H NMR (DMSO-d_6) δ 1.17 (3H, s, CH_3), 3.00 and 3.20 (each 1H, d, J=17.6 Hz, H(4A) and H(4B)), 4.04 (1H, d, J=6.0 Hz, H(2)), 5.18 (1H, s, C(3)-OH), 6.11 (1H, d, J=6.0 Hz, C(2)-OH), 7.33 and 7.51 (each 1H, dd, J=8.4, 0.8, H(9) and H(11)), 7.74 (2H, m, H(10) and H(5)), 8.20 (1H, d, J=8.0 Hz, H(6)), 12.07 (1H, s, C(8)-OH); HR-MS m/z 338.0793 ($C_{19}H_{14}O_6$ requires 338.0790).

The UV spectra of PD 116,779 in MeOH and methanolic KOH closely resembles that reported for the benz[a]anthraquinone antibiotic, tetrangomycin (**2**)[2], including the expected bathochromic shift in base. High resolution mass spectrometry established $C_{19}H_{14}O_6$ as the molecular formula of PD 116,779 which differs from that of tetrangomycin by the presence of one additional oxygen atom. Strong absorptions in the IR spectrum at 1637, 1669, and 1702 cm^{-1} could be assigned to chelated and nonchelated quinone carbonyls and the expected C(1) ketone functionality, respectively, and provided additional evidence for the presence of the benz[a]anthraquinone nucleus.

The downfield region of the ^1H NMR spectrum of PD 116,779 exhibits a signal for a phenolic proton at 12.07 ppm as well as overlapping sets of aromatic signals belonging to an ABX and an AB system, consistent with the aromatic nucleus of **2**. The remaining ^1H NMR signals correspond to a pair of nonequivalent methylene protons, a tertiary methyl group, and >C*H*OH, as well as secondary and tertiary hydroxyl functionalities. Together with the data described above, these protons could be assigned to the A ring of the benz[a]anthraquinone skeleton. The lack of coupling between the >C*H*OH and CH_2 protons suggests that the tertiary methyl and hydroxyl groups are both attached to the C(3) position, as in tetrango-



PD 116,779 (**1**) R=OH
Tetrangomycin (**2**) R=H

mycin. The secondary hydroxyl functionality, therefore, must occupy one of the remaining C(2) or C(4) positions. The placement of the hydroxyl at C(2) was determined by analysis of the ^{1}H NMR homonuclear correlation spectrum (COSY), which clearly showed a correlation between one of the two aromatic protons (H(5)) in the AB system described above with both of the methylene protons, which necessarily must be placed on C(4). On the basis of these data, PD 116,779 can confidently be assigned structure **1**, differing from tetrangomycin only by the additional hydroxylation at C(2).

PD 116,779 demonstrates moderate cytotoxicity against the L1210 lymphocytic leukemia and HCT-8 human colon adenocarcinoma cell lines, with IC_{50} values of 3.55×10^{-7} and 4.08×10^{-7} M, respectively. Recently we reported the isolation and structure elucidation of PD 116,740, another antibiotic belonging to the benz[a]anthraquinone class, which demonstrated similar anticancer activity[3]. These findings, plus the reported antitumor properties of other benz[a]anthraquinone antibiotics such as aquayamycin[4] and saquayamycin[5], indicate that this interesting class of compounds may provide new models for the design of useful cancer chemotherapeutic agents.

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