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A SHORT, CONVENIENT SYNTHESIS OF 2'-DEOXYSHOWDOMYCIN

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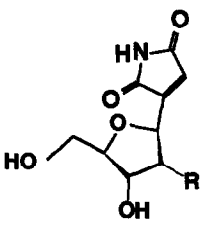
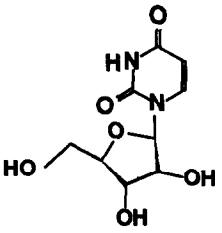
Abstract. A short, convenient synthesis of 2'-deoxyshowdomycin along with its 1'-epimer has been presented.

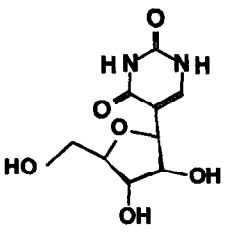
Our broad objective to explore steric and conformational constraints of formation of nucleic acid double helices has led us to the recent syntheses and biophysical investigations of a number of ring-expanded ("fat") nucleosides and nucleotides.[1] In this context, ring-contracted ("slim") nucleosides/-tides offer a logical extension to such studies, and showdomycin and its 2'-deoxy analogue serve as two simple protocols for initial explorations of this objective as they relate to RNA and DNA, respectively. Showdomycin is a naturally occurring antitumor antibiotic[2-5] first isolated from *Streptomyces showdensis* by Nishimura *et al* in 1964.[2] While the basis for its biological activity is unknown, one might speculate that showdomycin interferes with cellular processes involving the natural pyrimidine nucleoside uridine. Indeed, showdomycin may be considered as a ring-contracted or "slim" uridine or pseudouridine wherein a nitrogen atom from the six-membered pyrimidinedione nucleus has been eliminated to form a five-membered maleimide ring. The acid-base characteristic of showdomycin (pK_a = 9.29)[6] is also very similar to that of uridine (pK_a = 9.12)[7] and pseudouridine (pK_a = 9.1).[8] Furthermore, our molecular mechanics calculations based upon



R = OH; SHOWDOMYCIN
R= H; 2'-DEOXYSHOWDOMYCIN



URIDINE
(U)



PSEUDOURIDINE
(ψU)

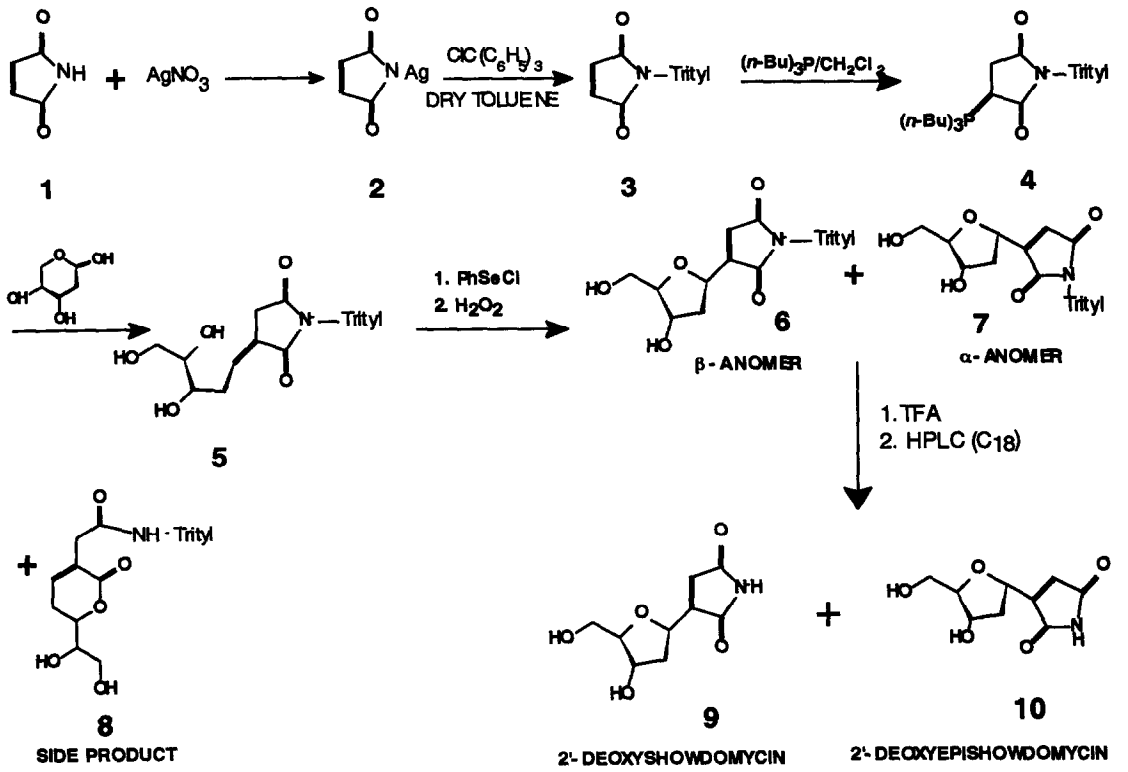
intermolecular interaction energies[9] revealed a comparable base-pairing binding energy for the showdomycin...A pair (ΔE_b = -10.59 kcal/mol) and the natural U...A pair (ΔE_b = -10.64 kcal/mol). An important question to be addressed, therefore, is whether showdomycin or 2'-deoxyshowdomycin could replace uridine or thymidine in RNA or DNA, and how would such a replacement affect base-pairing and stacking interactions as well as helical structure, stability, and conformation of nucleic acid double-helices? These intended studies have

necessitated the synthesis of both showdomycin and its 2'-deoxy analogue.

An examination of the literature revealed that while several syntheses of showdomycin have been reported,[10-19] there are only two full[20,21] and one partial[22] syntheses of 2'-deoxyshowdomycin known, and all of them are long, tedious, and low-yielding. Furthermore, while one of the full syntheses[21] reports the product as a mixture of β-D and β-L forms of 2'-deoxyshowdomycin, the other synthesis[20] gives neither the melting point nor the ^{1}H NMR data of the claimed β-D form that corresponds to natural showdomycin. The third partial synthesis[22] only describes the preparation of the necessary sugar precursor for the latter synthesis.[20] A fourth approach toward analogues of showdomycin, which employs an elegant ring-contraction method, however, gave the undesired α-anomer, 2'-deoxyepishowdomycin, exclusively.[23] Thus, the literature still lacks adequate information on physical and spectroscopic characteristics of 2'-deoxyshowdomycin.

Showdomycin is antiaromatic by the Hückel (4n + 2 π-electrons) rule. It is extremely base-labile,[2] and undergoes facile ring-opening as well as reduction reactions. In this context, the synthesis of 2'-deoxyshowdomycin was anticipated to be even more difficult since, in addition to the inherent base instability of the showdomycin aglycon, 2'-deoxynucleosides are generally more acid-sensitive and less stable than their oxy counterparts. Therefore, we deemed it necessary to protect the crucial imide NH of the maleimide ring so as to minimize probable side reactions. A trityl protecting group, which allows facile removal under mild acidic conditions, was used to successfully synthesize 2'-deoxyshowdomycin, along with its 1'-epimer 2'-deoxyepishowdomycin. The two anomers were also successfully separated by HPLC. The synthesis (**Scheme I**), which is modeled after the one reported for showdomycin by Barrett, *et al.*,[18] consisted of reacting *N*-tritylmaleimide (**3**), prepared from maleimide by the literature procedure,[24] with tri-*n*-butylphosphine to form *in situ* the ylide **4** which was immediately reacted with 2-deoxy-D-ribose to obtain the open-chain sugar precursor **5**, yield 68%, an off-white foam: ^{1}H NMR (DMSOd_6) δ 7.28 (m, 15H, three Ph), 6.66 (t, J = 15.3 Hz, 1H, olefinic CH), 4.74 (d, J = 6.0 Hz, ex. w/D_2O, OH), 4.63 (d, J = 5.4 Hz, ex. w/D_2O, OH), 4.38 (t, J = 11.1 Hz, ex. w/D_2O, OH), 3.48 (m, 2H, CH_2OH), 3.36 (s, 2H, ring CH_2), 3.23 (m, 2H, side-chain CH's), 2.40 + 2.22 (two m, 2H, allylic CH_2); ^{13}C NMR (DMSO-d_6) δ 174.56, 169.44 (C=O), 143.55 (Ph-C*i*), 136.18 (=CH), 129.03, 128.10, 127.02 (Ph-CH), 79.92 (qC), 75.29 (CH), 73.54 (qC), 71.26 (CH), 64.22 (CH_2OH), 34.26 (ring CH_2), 33.60 (allylic CH_2); *Anal.* C,H,N.[25] The use of tri-*n*-butylphosphine in place of triphenyphosphine afforded a more reactive ylide that could be reacted further without isolation with the sugar in a one-pot reaction. This modification has also enabled us to substantially reduce the reaction time (to 12 hours for the two steps combined as compared with 192 hours for the sugar-ylide condensation step alone as reported).[18] The separation of the two diastereomers of **5** by flash or rotating disc chromatography was hampered by the formation of a side product, mp 125 °C, whose spectroscopic and microanalytical data were consistent with the lactone structure **8**. This side product was gradually forming whenever the reaction solution was kept for a period of time before work-up. Formation of the side product was somewhat avoided by immediately working up the reaction, followed by chromatography. Ring closure of **5** was effected by sequential

Scheme I



treatments with phenylselenyl chloride and 3% hydrogen peroxide to produce *N*-trityl-2'-deoxyshowdomycin (**6**) and its 1'-epimer (**7**), in 39% yield, in a 60:40 ratio as determined by ^{1}H NMR ((DMSO-d_6): δ 7.28 (m, 15H, three Ph), 6.63, 6.54 (two s, 1H total, H-4, β:α = 60:40), 5.06, 5.00 (two d, 1H total, ex. w/D_2O, OH), 4.78 (m, 1H, H-1'), 4.67 (br s, 1H, ex. w/D_2O, OH), 4.15 (m, 1H, ribose CH), 3.80, 3.71 (two m, 1H total, ribose CH), 3.22 (m, 2H, collapses to d w/D_2O exchange, H-5'), 2.37, 2.04, 1.77 (three m, 2H total, H-2'). Deprotection of the maleimide N was carried out using TFA/H_2O (50:1). The resulting anomers **9** and **10** were separated from the mixture by HPLC using water as an eluent. The following are the physical and spectral data of the two anomers: Compound **9** (2'-deoxyshowdomycin): Recrystallized from a mixture of 2-propanol-$CHCl_3$-hexanes, mp 110-111 °C: ^{1}H NMR (DMSO-d_6) δ 10.81 (s, 1H, ex. w/D_2O, NH), 6.56 (s, 1H, H-4), 5.09 (s, 1H, ex. w/D_2O, 3'-OH), 4.82 (br t, J = 7.3 Hz, 1H, H-1'), 4.70 (t, J = 5.4 Hz, 1H, ex. w/D_2O, 5'-OH), 4.14 (br s, 1H, H-4'), 3.74 (br s, 1H, H-3'), 3.35 (m, 2H, collapses to a doublet w/D_2O exchange, 5'-CH_2), 2.08, 1.84 (two m, 2H total, diastereotopic H-2's); *Anal.* C,H,N.[25]; Compound **10** (2'-deoxyepishowdomycin): Recrystallized from 2-propanol-$CHCl_3$-hexanes, mp 110-111 °C (lit.[23] 112 °C): ^{1}H NMR (DMSO-d_6) δ 10.80 (s, 1H, ex. w/D_2O, NH), 6.46 (s, 1H, H-4), 4.99 (br s, 1H, ex. w/D_2O, 3'-OH), 4.82 (br t, 1H, H-1'), 4.72 (br s, 1H,

ex. w/D_2O, 5'-OH), 4.15 (m, 1H, H-4'), 3.82 (m, 1H, H-3'), 3.36 (m, 2H, collapses to a doublet w/D_2O exchange, 5'-CH_2), 2.40, 1.82 (two m, 2H total, diastereotopic H-2's); *Anal.* C,H,N.[25]

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