the biosynthesis of pyrimidines. *E. coli* cultures inhibited by azauracil have been observed to accumulate orotic acid and orotidylic acid[15] suggesting a block in the synthesis of the dihydroxypyrimidine nucleus rather than at sites of utilization of uracil, and, more specifically, at the decarboxylative conversion of orotidylic acid to uridylic acid.[16]

6-Oxadihydrouracil also inhibits the growth of *E. coli* Texas and its toxicity is competitively reversed by uracil over a 30-fold range of concentrations with an inhibition index of about 100. Further, uracil precursors and related compounds also did not reverse the inhibitory action of the analog in *E. coli*.

Neither 6-oxadihydrouracil nor 6-oxadihydrothymine were appreciably inhibitory to mammalian cells grown in tissue culture as indicated in Table V. Studies were carried out using HEp-2 human carcinoma, Jensen rat sarcoma, and WI-38 diploid human embryonic lung cells. Using HEp-2 cells, the per cent of control growth in the presence of 0.25, 2.5, and 5.0 μg/ml of the oxauracil derivative was 103, 88, and 101, respectively; the control growth was tenfold that of the initial inoculum. Subsequent assays at levels of inhibitor up to 50 μg/ml did not affect proliferation *in vitro* of these human cancer cells appreciably. In the same system, 6-oxadihydrothymine at levels of 5, 25, and 50 μg/ml gave values of 100, 114, and 88% that of control growth, respectively (with a sevenfold increase of cell growth over that of the inoculum). No striking differences in results were obtained with either of these analogs at these concentration levels using Jensen rat sarcoma and WI-38 lung cells in comparable tissue culture assays. In summary, neither of the oxa analogs proved to be appreciably inhibitory to growth of mammalian cell cultures even though they exhibited a relatively high toxicity to microbial growth.

TABLE V

EFFECT OF OXA ISOSTERES ON GROWTH OF MAMMALIAN CELLS *in Vitro*

Isostere, μg/ml	% of control proliferation[a] HEp-2	Jensen	WI-38
6-Oxadihydrouracil			
0.25	103	90	...
2.5	88	87	...
5.0	101	115	86
25	...	...	86
50	104	62	85
6-Oxadihydrothymine			
5	100	89	75
25	114	86	82
50	88	73	65

[a] Calculated by dividing the number of new cells produced in test compound cultures by those produced in nonsupplemented Medium 7a control cultures (× 100). Test compounds were introduced in the log phase of proliferation; for culture conditions see text. The HEp-2 and WI-38 cells are derived from human carcinoma and normal embryonic lung tissue, respectively; the Jensen cells were obtained from freshly excised Jensen sarcomas carried in Holtzman rats.

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Synthesis of Carbonate Analogs of Dinucleosides. 3′-Thymidinyl 5′-Thymidinyl Carbonate, 3′-Thymidinyl 5′-(5-Fluoro-2′-deoxyuridinyl) Carbonate, and 3′-(5-Fluoro-2′-deoxyuridinyl) 5′-Thymidinyl Carbonate[1]

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The synthesis of (3′→5′) carbonate analogs of dinucleosides is described. 3′-Thymidinyl 5′-thymidinyl carbonate (**10**), 3′-thymidinyl 5′-(5-fluoro-2′-deoxyuridinyl) carbonate (**15**), and 3′-(5-fluoro-2′-deoxyuridinyl) 5′-thymidinyl carbonate (**18**) have been synthesized. Thymidine was converted to 5′-O-tritylthymidine and treated with phosgene to give 3′-(5′-O-tritylthymidinyl) chloroformate (**6**). Subsequent treatment with thymidine and removal of the protective group afforded **10**. Compounds **15** and **18** were prepared by the same method. 3′-(5′-Phosphorylthymidinyl) 5′-thymidinyl carbonate (**14**) was prepared from **10** by reaction with diphenyl phosphorochloridate followed by hydrogenolysis of the protective groups. Compounds **10**, **14**, and **15** did not show significant inhibition of *Escherichia coli* growth or thymidylate synthetase.

In vitro inhibition of nucleic acid formation by nucleotides and their derivatives has been demonstrated for 5-fluoro-2′-deoxyuridine 5′-monophosphate (FUDRP) and 5-trifluoromethyl-2′-deoxyuridine 5′-monophosphate (F_3 TDRP).[2]

A potential site of inhibiting nucleic acid synthesis is the enzyme deoxyribonucleotidyltransferase (DNA polymerase).[3,4] Studies on the inhibitory action of nucleosides and nucleotides have demonstrated that the latter do not pass through cell membranes.[5] Recently Bloch and coworkers[6] have synthesized dinucleoside phosphates containing 5-fluorouracil; cellular permeability also limits the uptake of these compounds and the observed biological activity appears to be derived

(1) This work was supported by a Public Health Service predoctoral fellowship 5-F1-GM-29, 336 to E. A. C. and by Grants CA-5639 and 1K3-CA-10,739 from the National Cancer Institutes, National Institutes of Health. Presented at the XXIst IUPAC Meeting, Prague, Sept 1967.

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(4) (a) I. J. Slotnick, M. Dougherty, and D. H. James, Jr., *Cancer Res.*, **26**, 673 (1966); (b) N. Tanaka, K. Nagai, H. Yamaguchi, and H. Unezawa, *Biochem. Biophys. Res. Commun.*, **21**, 328 (1965); (c) N. S. Mizuno, *Biochim. Biophys. Acta*, **108**, 394 (1965); (d) N. C. Brown and R. E. Handschumacher, *J. Biol. Chem.*, **241**, 3083 (1966); (e) H. J. Rosenkranz and J. A. Levy, *Biochim. Biophys. Acta*, **96**, 181 (1965); (f) J. L. York and G. A. LePage, *Can. J. Biochem.*, **44**, 19 (1966); (g) E. C. Moore and S. S. Cohen, *J. Biol. Chem.*, **242**, 2116 (1967).

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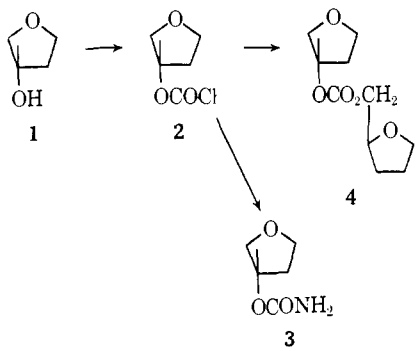
from the product of ester hydrolysis. Attempts to achieve activity by masking the phosphate moiety of nucleotides and preparing esters of nucleosides have been reported by Heidelberger and coworkers.[7]

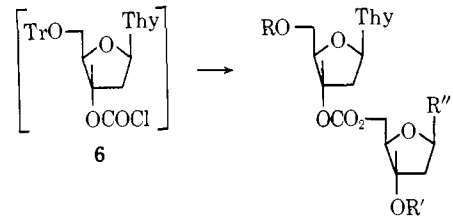
In an effort to overcome the barrier to permeability and to examine the effects on growth inhibition in a nucleotide analog the nonionic carbonate linkage was employed. Three dinucleoside carbonates have been prepared: 3′-thymidinyl 5′-thymidinyl carbonate (**10**), 3′-thymidinyl 5′-(5-fluoro-2′-deoxyuridinyl) carbonate (**15**), and 3′-(5-fluoro-2′-deoxyuridinyl) 5′-thymidinyl carbonate (**18**). The 5′-monophosphate of **10**, structure **14**, also was prepared.

Carbonate esters have been used extensively in the synthesis of carbohydrates; however, reports of carbonate analogs of nucleosides have appeared only recently. A bis-5′-nucleoside carbonate and a 2′,3′-cyclic carbonate have been reported by Hampton and Nichol.[8] Ogilvie and Letsinger[9] utilized the isobutyloxycarbonyl as a blocking group in nucleoside synthesis.

Methods leading to unsymmetrical dinucleoside carbonates were examined initially on model systems. Treatment of 3-hydroxytetrahydrofuran (**1**) with phosgene gave the intermediate chloroformate **2** characterized by nmr. Subsequent treatment with NH_4OH gave the carbamate **3** or with tetrahydrofurfuryl alcohol gave the unsymmetrical carbonate **4**, characterized by nmr.



Treatment of a THF solution of 5′-O-tritylthymidine[10] (**5**) with excess phosgene in the presence of 1 equiv of pyridine gave a solution of the unstable 3′-chloroformate ester of **5** (**6**). After excess phosgene was removed this solution was added to 3′-O-acetylthymidine[11] (**7**) in pyridine to yield the desired 3′-(5′-O-tritylthymidinyl) 5′-(3′-O-acetylthymidinyl) carbonate (**8**). The possibility that the chloroformate **6** would react with the 5′-hydroxyl of thymidine in preference to the 3′-hydroxyl was supported by the work of Baker and coworkers[12] who successfully obtained preferential reaction of phenyl chloroformate with thymidine to give the 5′-phenylcarbonylthymidine. The feasibility of utilizing unprotected thymidine in the final step would



8, R = trityl; R′ = OAc; R″ = thymine
9, R = trityl; R′ = H; R″ = thymine
10, R = R′ = H; R″ = thymine
11, R = $PO(OC_6H_5)_2$; R′ = H; R″ = thymine
12, R = R′ = $PO(OC_6H_5)_2$; R″ = thymine
13, R = $PO(OC_6H_5)_2$; R′ = OAc; R″ = thymine
14, R = PO_3H_2; R′ = H; R″ = thymine
15, R = R′ = H; R″ = 5-fluorouracil (FU)

then alleviate the necessity of removing the acetate, a potentially difficult reaction in the presence of the carbonate. The deacetylated product **9** was obtained directly by treating a pyridine solution of thymidine with **6** to give 3′-(5′-O-tritylthymidinyl) 5′-thymidinyl carbonate (**9**). Removal of the trityl group of **9** by refluxing in 80% HOAc gave 3′-thymidinyl 5′-thymidinyl carbonate (**10**).

The downfield shift in the nmr spectra of the 3′ and 5′ hydrogens caused by esterification of the OH groups at these positions was used to support the 3′→5′ carbonate linkage. The nmr of thymidine gives a peak for the 5′ protons at 3.64 ppm and a peak for the 3′ proton at 4.30 ppm. In the spectrum of 3′-O-acetylthymidine (**7**) the 5′ protons remain at 3.72 ppm while the 3′ proton now appears at 5.22 ppm, a downfield shift of 0.9 ppm due to the deshielding effect of the acetate ester. The 3′ proton of 5′-O-acetylthymidine remains at 4.34 ppm while the 5′ protons were shifted downfield by 0.6 ppm to 4.24 ppm. Finally, as expected, both the 3′ and 5′ protons of 3′,5′-di-O-acetylthymidine were shifted to downfield positions of 5.24 ppm for the 3′ proton and 4.25 ppm for the 5′ protons, shifts of 0.9 and 0.6 ppm, respectively. In the case of 3′-thymidinyl 5′-thymidinyl carbonate (**10**) it was impossible to observe the shift of the 5′ proton due to the complex absorption of the 3′, 4′, and 5′ protons in the region from 3.30 to 4.50 ppm; however, the 3′ proton at the carbonate moiety appears as a broad peak at 5.27 ppm a downfield shift of about 1.0 ppm which corresponds closely with the shift observed in the 3′-O-acetyl compounds.

Additional proof for the 3′→5′ linkage was obtained by tritylation of the unprotected carbonate **10** which afforded a small amount of 3′-(5′-O-tritylthymidinyl) 5′-thymidine carbonate (**9**) along with a large return of the carbonate starting material. The tritylation product **9** was shown to be identical with authentic trityl carbonate. This result demonstrates the presence of only one free primary OH. In order to show that the unprotected carbonate also contained one secondary OH, the trityl carbonate **9** was acetylated to give a product identical with an authentic sample of 3′-(5′-O-tritylthymidinyl) 5′-(3′-O-acetylthymidinyl) carbonate (**8**).

As a further demonstration of the 3′→5′ carbonate linkage, a sample of (5′-O-tritylthymidinyl) 5′-thymidinyl carbonate (**9**) was hydrolyzed (base) to give thymidine and 5′-O-tritylthymidine (**5**).

(7) (a) D. C. Remy, A. V. Sunthankar, and C. Heidelberger, *J. Org. Chem.*, **27**, 2491 (1962); (b) K. L. Mukherjee and C. Heidelberger, *Cancer Res.*, **22**, 815 (1962); (c) Y. Nishigawa, J. E. Casida, S. W. Anderson, and C. Heidelberger, *Biochem. Pharmacol.*, **14**, 1605 (1965).

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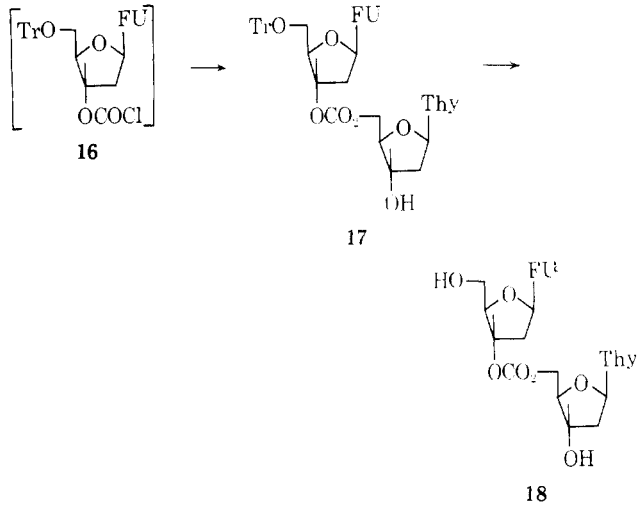

16 17 18

Using the procedure of Barnwell and coworkers[13] the phosphorylation of 3′-thymidinyl 5′-thymidinyl carbonate (**10**) was achieved by treatment with excess diphenyl phosphorochloridate at low temperatures to give both mono- (**11**) and diphosphorylated (**12**) products which were separated by fractional crystallization. Hydrogenolysis of 3′-(5′-diphenylphosphoryl-thymidinyl) 5′-thymidinyl carbonate (**11**) gave the crystalline phosphate **14**. Evidence of 5′-monophosphorylation as opposed to 3′-monophosphorylation was based on the nmr spectra of **11** and **12** and the finding that acetylation of **11** yields a monoacetylated product **13** in which the acetyl methyl appeared in the nmr spectrum at the same position as that of 3′-(5′-O-trityl-thymidinyl) 5′-(3′-O-acetylthymidinyl) carbonate (**8**) and integration for both 3′ protons at 5.30 ppm of **13** is observed.

The synthesis of 3′-thymidinyl 5′-(5-fluoro-2′-deoxyuridinyl) carbonate (**15**) was accomplished by treating 5-fluoro-2′-deoxyuridine[14] with a solution of **6** followed by removal of the trityl group to give **15**. The reverse dinucleoside carbonate, 3′-(5-fluoro-2′-deoxyuridinyl) 5′-thymidinyl carbonate (**18**) was prepared in a similar manner from 5′-O-trityl-5-fluorodeoxyuridine 3′-chloroformate (**16**).

Preliminary biological results[15] on growth inhibition of *Escherichia coli* B by compounds **10**, **15**, thymidine, and 5-fluoro-2′-deoxyuridine were obtained as previously described.[16] Compound **10** and thymidine showed no growth inhibition at 10^{-3} *M*. 5-Fluoro-2′-deoxyuridine as expected showed approximately 50% inhibition of growth at 10^{-5} *M*; however, the carbonate **15** showing approximately 50% inhibition at 10^{-3} *M* was much less active than 5-fluoro-2′-deoxyuridine. It is probable that the inhibition seen in **15** is derived from 5-fluoro-2′-deoxyuridine released by *in vivo* hydrolysis of the carbonate linkage. Studies on thymidylate synthetase inhibition[17] showed no inhibition at a ratio of (inhibitor)/(2′-deoxyuridine 5′-monophosphate) of 80 for **10** and 60 for **15**, and 15% inhibition at an [I]/[S] ratio of 50 for the phosphate **14**.

Experimental Section[18]

3-Hydroxytetrahydrofuran Carbamate (3).—A solution of 8.0 g (0.09 mole) of 3-hydroxytetrahydrofuran (**1**) in 150 ml of Et_2O was cooled to 10° while $COCl_2$ was passed into the solution for 2.5 hr. After purging with N_2 and standing 12 hr at room temperature the solvent was removed and the residue was distilled to give 11 g (80%) of **2**, bp 40–45° (0.6 mm). A positive $AgNO_3$ test, an intense 1780–1750-cm^{-1} ir band, and the following nmr spectrum (CCl_4) were obtained from **2**: 5.54 (quintet, methyne), 4.03 (m, 4,-CH_2O), 2.25 ppm (m, 2, CCH_2C).

The carbamate **3** was prepared by stirring a cold (5°) solution of 1.5 ml of **2** in NH_4OH for 30 min, extracting with Et_2O, drying ($MgSO_4$), and evaporating gave 2.5 g of solid, mp 78–80°. Recrystallization from C_6H_6–petroleum ether (30–60°) gave mp 81.5–82.5°. *Anal.* ($C_5H_9NO_3$) C, H, N.

3-Tetrahydrofuryl Tetrahydrofurfuryl Carbonate (4).—Tetrahydrofurfuryl alcohol (1.3 g, 0.013 mole) and dry pyridine (5 ml) were dissolved in 100 ml of dry Et_2O, cooled to 5°, and treated dropwise with a solution of 2.0 g (0.013 mole) of 3-hydroxytetrahydrofuran chloroformate (**2**) in 50 ml of dry Et_2O. After addition was complete the solution was stirred at 25° for 4.5 hr. The solid was filtered, 50 ml of C_6H_6 was added to the filtrate, and the latter was washed successively with 50-ml portions of H_2O, 3% HCl, 5% $NaHCO_3$, and H_2O. The solution was dried ($MgSO_4$) and evaporated to give 1.0 g of **4** as an oil: nmr (CCl_4), 5.2 (m, 1, C-3 methyne), 3.2–4.2 (m, 9, CH_2O), and 1.8–2.3 ppm (m, 6, CCH_2C). *Anal.* ($C_9H_{16}O_3$) H; C: calcd, 55.55; found, 56.01.

3′-(5′-O-Tritylthymidinyl) 5′-(3′-O-Acetylthymidinyl) Carbonate (8).—$COCl_2$ was passed for 1 hr into a cold (0–5°) solution of 2.5 g (5.2 mmoles) of 5′-O-tritylthymidine[18] in 100 ml of dry THF containing 0.42 g (5.2 mmoles) of pyridine. After stirring overnight at room temperature the mixture was filtered and concentrated (under vacuum) to 15 ml. This solution of **6** was added slowly to a cold (5°), stirred solution of 1.0 g of 3-O-acetylthymidine[11] in 40 ml of pyridine. After 3 hr at 25° the solution was evaporated to a yellow gum, dissolved in Me_2CO, and chromatographed on 20 g of silica gel. Elution with 2% MeOH–$CHCl_3$ gave 1.2 g (44%) of **8** as a solid, mp 105–120°. An nmr ($CDCl_3$) spectrum of **8** showed singlets at 1.45, 1.89, and 2.05 (3 H each, assigned to the three CH_3 groups), 7.55 (shoulder, C-6 protons), 7.29 (trityl), 6.40 (t, 2, C-1′ protons), 5.38 (m, 2, C-3′ methynes), and multiplets at 3.35–4.45 (C-4′, C-5′ protons) and 2.45 ppm (C-2′ protons). *Anal.* ($C_{42}H_{42}N_4O_{12}$) C, H; N: calcd, 7.05; found, 6.53.

3′-(5′-O-Tritylthymidinyl) 5′-Thymidinyl Carbonate (9).—A solution as described in the synthesis of **8** of the chloroformate **6** prepared from 2.75 g (5.7 mmoles) of 5′-O-tritylthymidine was added slowly to 1.10 g (4.5 mmoles) of thymidine in 50 ml of dry pyridine at 5°. After 6 hr at 25° the solution was poured into 200 ml of ice–H_2O; the yellow solid that formed was extracted with EtOAc–C_6H_6 and chromatographed on 20 g of silica gel. Starting materials were eluted with 1% MeOH–$CHCl_3$ and the product **9** with 5% MeOH–$CHCl_3$; recrystallization from $CHCl_3$ gave 0.84 g (25%): mp 185–188°; nmr ($CDCl_3$), 1.49 and 1.82 (s, 6, CH_3) 7.20–7.60 (m, 17, trityl and C-6 protons), and 5.35 ppm (m, 1, 3′-methyne adjacent to the carbonate); the remainder of the peaks matched those given for **8**. *Anal.* ($C_{39}H_{38}N_4O_{11}$) C, H, N.

Stirring 0.076 g of **9** in 50 ml of 5% NaOH–MeOH for 1 hr gave chromatographic evidence of hydrolysis (thymidine). After 26 hr the mixture was poured into 200 ml of H_2O, extracted with $CHCl_3$, dried ($MgSO_4$), and evaporated to yield 0.035 g of 5′-O-tritylthymidine.

Acetylation of **9** was accomplished by stirring 90 mg (0.12 mmoles) of **9** in 10 ml of dry pyridine containing 0.1 ml (1 mmole)

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(14) A generous gift of Dr. Harry Woods, Cancer Chemotherapy National Service Center, Bethesda, Md.

(15) The authors acknowledge Mrs. William Riggs for the biological results.

(16) M. P. Mertes, J. Zielinski, and C. Pilkor, *J. Med. Chem.*, **10**, 320 (1967).

(17) M. P. Mertes and N. R. Patel, *ibid.*, **9**, 868 (1966).

(18) Melting points were taken using a calibrated Thomas–Hoover unit. Spectral data were obtained using Varian A-60 and A-60-A nmr, Beckman IR-5, IR-8, and Cary 14 spectrometers. Chromatography was done using Brinkman silica gel (0.05–0.2 mm) chromatogram sheets or Whatman No. 1 paper. Elemental analyses were done by Midwest Microlabs, Indianapolis, Ind., or on an F & M 185 CNH analyzer. Where analyses are indicated only by symbols of the elements analytical results obtained for those elements were within 0.4% of the theoretical values.

of Ac_2O. After 2 days the solution was poured into ice–H_2O, and the solid was filtered, recrystallized from C_6H_6–cyclohexane, and chromatographed on 5 g of silica gel. Elution with EtOAc–$CHCl_3$ gave **8**, identified by nmr and chromatography.

3′-Thymidinyl 5′-Thymidinyl Carbonate (10).—An 80% HOAc (50 ml) solution of 2.0 g (2.7 mmoles) of **9** was refluxed for 10 min, poured into 200 ml of ice–H_2O, and filtered. Evaporation and recrystallization of the residue from MeOH gave 0.7 g (50%) of **10**, mp 205–210°, softens 137–150°; ascending chromatography in NH_4HCO_3 (16%) gave an R_f of 0.71. The nmr (DMSO-d_6) showed the expected resonance signals, similar to those found in **9**. *Anal.* ($C_{21}H_{26}N_4O_{11}$) C, H, N.

A sample (0.7 g, 1.37 mmoles) of **10** was heated in 50 ml of dry pyridine containing 0.7 g (2.75 mmoles) of trityl chloride for 2.5 hr. The solution was poured into 300 ml of ice–H_2O and filtered, and the solid was dissolved in $CHCl_3$, dried ($MgSO_4$), and chromatographed on 45 g of silica gel. Elution with 4% MeOH–$CHCl_3$ gave 0.22 g of a solid identical with **9** by melting point and ascending chromatography in *i*-PrOH–H_2O (4:6), R_f 0.78.

3′-(5′-Diphenylphosphorylthymidinyl) 5′-Thymidinyl Carbonate (11).—A cold (0°) solution of 100 mg (0.20 mmole) of **10** in 2 ml of dry pyridine was treated with 200 mg (0.75 mmole) of diphenyl phosphorochloridate[18] and maintained at 5° for 12 hr. The solvent was evaporated and the residue was dissolved in $CHCl_3$, washed with H_2O, dried, and chromatographed on 4 g of silica gel. Elution with CH_2Cl_2 containing increasing amounts of MeOH gave **12** and finally 0.027 g (25%) of **11** as glasses characterized by nmr.

It was found in subsequent reactions that the monophosphorylated product **11** could be separated from **12** by fractional crystallization from $CHCl_3$. *Anal.* (**11**, $C_{33}H_{35}N_4O_{14}P \cdot H_2O$) C, H, N, P.

Acetylation of 17 mg (0.023 mmole) of **11** was accomplished using 23 mg (0.23 mmole) of Ac_2O in 1.5 ml of dry pyridine. After 12 hr at 25° the solution was poured into ice–H_2O, extracted with $CHCl_3$, dried ($MgSO_4$), and evaporated to a glass. The nmr ($CDCl_3$) showed the acetyl methyl protons at 2.10 ppm and the two 3′-methynes at 5.30 ppm.

3′-(5′-Monophosphorylthymidinyl) 5′-Thymidinyl Carbonate (14).—An EtOH (5 ml) solution containing 100 mg (0.13 mmole) of **11** was added to prereduced PtO_2 (80 mg) in EtOH and reduced at atmospheric pressure for 18 hr; slightly more than the theoretical amount of H_2 was absorbed. Filtration and evaporation gave the product, mp 185–200°. *Anal.* ($C_{21}H_{27}N_4O_{14}P \cdot 2H_2O$) C, H; N: calcd, 8.95; found, 8.45.

3′-Thymidinyl 5′-(5-Fluoro-2′-deoxyuridinyl) Carbonate (15).—A C_6H_6 solution (75 ml) of the chloroformate **6** prepared from 1.5 g (3.1 mmoles) of 5′-O-tritylthymidine was added slowly (45 min) to a cold solution (0–5°) of 0.45 g (1.8 mmoles) of 5-fluoro-2′-deoxyuridine in 30 ml of dry pyridine. After stirring 12 hr, 50 ml of H_2O was added and the mixture was extracted with three 100-ml portions of $CHCl_3$. After drying and concentrating, the residue was chromatographed on 60 g of silica gel. Elution with $CHCl_3$ and 4% MeOH–$CHCl_3$ gave 0.56 g of the trityl product which was rechromatographed on 25 g of silica to remove minor impurities.

A solution of 0.447 g (0.59 mmole) of the tritylated compound was refluxed for 10 min in 20 ml of 80% HOAc, poured into 200 ml of ice–H_2O, and extracted with two 150-ml portions of C_6H_6. The residue of the aqueous solution was evaporated and washed with MeOH to give **15** as a solid; recrystallization from H_2O–MeOH gave 0.124 g (17%), mp 220–224°, uv, λ_{max}^{EtOH} 266 mμ. *Anal.* ($C_{20}H_{23}FN_4O_{11}$) C, H, F, N.

3′-(5′-Fluoro-2′-deoxyuridinyl) 5′-Thymidinyl Carbonate (18).—$COCl_2$ was passed for 45 min into a cold (0–10°) solution of 3.5 g (7.1 mmoles) of 5′-O-trityl-5-fluoro-2′-deoxyuridine[19] in 100 ml of dry THF containing 0.56 g (7 mmoles) of dry pyridine. After $COCl_2$ treatment, the mixture was filtered and concentrated (under vacuum) to 20 ml. This solution of **16** was added slowly to a cold (5°), stirred solution of 1.5 g (6.2 mmoles) of thymidine in 20 ml of dry pyridine. After 18 hr at room temperature the solution was poured into 300 ml of ice–H_2O. The aqueous solution was decanted from the heavy precipitate and the precipitate was dissolved in MeOH. Repeated dissolution and evaporation afforded 5.2 g of white semisolid which was chromatographed on 60 g of silica gel (Brinkman 0.20–0.05 mm). Elution with $CHCl_3$ and $CHCl_3$–2% MeOH removed starting materials and impurities. Elution with $CHCl_3$–4% MeOH afforded 3.21 g (68% based on thymidine) of 3′-(5′-O-trityl-5-fluoro-2′-deoxyuridinyl) 5′-thymidinyl carbonate (**17**). *Anal.* ($C_{39}H_{37}FN_4O_{11}$) C, H, F, N.

A solution of 0.95 g (1.2 mmoles) of the tritylated compound **17** was refluxed for 10 min in 20 ml of 80% HOAc, poured into 200 ml of ice–H_2O, and extracted with two 100-ml portions of C_6H_6. The aqueous solution was then evaporated *in vacuo*, washed with MeOH, and filtered to give 0.6 g (93%) of **18** as a white solid, mp 155–160°. *Anal.* ($C_{20}H_{23}FN_4O_{11}$) C, H, F, N.

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Experimental Tumor Inhibitors. Antitumor Activity of Esters of ω-Aryl-ψ-nitro-ψ-alken-1-ol and Related Compounds[1]

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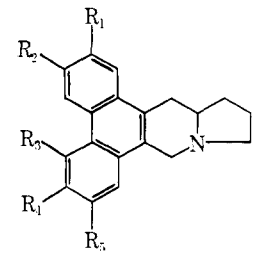
Preparation of a series of 5-substituted 4-nitro-4-penten-1-ol acetate and related analogs is described. Many compounds in this category demonstrated confirmed inhibitory activity against Walker carcinosarcoma 256 in preliminary biological testing. Structure–activity study indicated that the nitroalkene portion of the side chain is essential for the oncolytic property. The relative activity and toxicity of these compounds are dependent on the length of the aliphatic side chain and substitution at the terminal positions.

In connection with a structure–activity study of the alkaloids tylocrebrine (Ia) and tylophorine (Ib), which showed anticancer activity against leukemia L1210,[2] the phenanthro[9,10:6′,7′]indolizidine[3] nucleus (Ic) was prepared in this laboratory. Compound



Ia, R_1, R_2, R_3, R_4 = OCH_3; R_5 = H
b, R_1, R_2, R_4, R_5 = OCH_3; R_3 = H
c, R_1, R_2, R_3, R_4, R_5 = H

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